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Encontro de **F**ormação
da **O**rdem dos
Médicos **V**eterinários

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Animais de Companhia I

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Syncope: diagnostic reasoning and clinical management

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Introduction

Transient loss of consciousness (TLOC) is an apparent loss of consciousness with an abrupt onset, short duration, and spontaneous and complete recovery. The main forms of TLOC are: traumatic TLOC or concussion, non traumatic TLOC that are divided into three groups: syncope, epileptic seizures, and a miscellaneous group with rare disorders like cataplexy. Syncope is a T-LOC caused by a global cerebral hypoperfusion due to a fall of systemic blood pressure for a sudden decrease of vascular resistance and/or a drop in cardiac output.

Typical syncope is brief. Complete LOC in reflex syncope lasts no longer than 20 s in duration. However, syncope may rarely be longer, even as much as several minutes. In such cases, the differential diagnosis between syncope and other causes of LOC can be difficult. Recovery from syncope is usually accompanied by almost immediate restoration of appropriate behaviour and orientation. Sometimes the post-recovery period may be marked by fatigue. Pre-syncope is a term used to classify a state that resembles the prodrome of syncope but which is not followed by loss of consciousness. A sudden cessation of cerebral blood flow for a period as short as 6–8 s is sufficient to cause complete loss of consciousness. Systemic blood pressure is determined by cardiac output and total peripheral vascular resistance, and a fall in either can cause syncope, but a combination of both mechanisms is often present, even if their relative contributions vary considerably. A low or inadequate peripheral resistance can be due to inappropriate reflex activity causing vasodilatation and bradycardia and manifesting as vasodepressor, mixed, or cardioinhibitory reflex syncope. Other causes of a low or inadequate peripheral resistance are functional and structural impairments of the autonomic nervous system such in case of drug-induced dysautonomia or autonomic failure. In autonomic nervous failure, sympathetic vasomotor pathways are unable to increase total peripheral vascular resistance in response to the upright position. Gravitational stress, in combination with vasomotor failure, results in venous pooling of blood in the splanchnic and limb vessels, causing a decrease in venous return and consequently in cardiac output. The causes of transient low cardiac output are: a reflex causing bradycardia, known as cardioinhibitory type of reflex syncope; a cardiovascular causes, due to arrhythmia and structural disease including pulmonary embolism and hypertension; an inadequate venous return, due to volume depletion or venous pooling. According to the underlining mechabinsm syncope can be divided into three categories: reflex syncope, syncope due to orthostatic hypotension and cardiac syncope.

Reflex syncope

Reflex syncope traditionally refers to a heterogeneous group of conditions in which cardiovascular reflexes that are normally useful in controlling the circulation become intermittently inappropriate, in response to a trigger, resulting in vasodilatation and/or bradycardia and so in a fall in arterial blood pressure and global cerebral perfusion. Reflex syncope is usually classified based on the efferent pathway most involved, i.e. sympathethic or parasympathetic. The

term 'vasodepressor type' is commonly used if hypotension, due to a loss of upright vasoconstrictor tone, predominates. 'Cardioinhibitory tipe' is used when bradycardia or asystole predominate, and 'mixed' is used if both mechanisms are present. Vasovagal syncope is mediated by pain, emotion or by orthostatic stress. It is usually preceded by prodromal symptoms of autonomic activation (pallor, nausea). Situational syncope traditionally refers to reflex syncope associated with some specific circumstances (cough, micturation, defecation, vomiting, visceral pain, post-exercise syncope). Carotid sinus syncope is a rare spontaneous disorder that is triggered by mechanical manipulation of the carotid sinuses.

Orthostatic syncope

In contrast to reflex syncope, in case of dysautonomia, sympathetic efferent activity is chronically impaired so that vasoconstriction is deficient. Upon standing, blood pressure falls and syncope or pre syncope occurs. Orthostatic hypotension is defined as an abnormal decrease in systolic blood pressure upon standing. Orthostatic intolerance can induce syncope, but also dizziness, pre-syncope, weakness, fatigue and lethargy.

Classic orthostatic hypotension is characterized by a decrease in systolic blood pressure > 20 mmHg and in diastolic pressure > 10 mmHg within 3 minutes of standing.

Orthostatic hypotension can be induced by structural damages of the autonomic nervous system, drug induced autonomic failure and inadequate venous return due to volume depletion and venous pooling.

Cardiac Syncope

Structural cardiovascular diseases (valvular disease, ischemia, hypertrophic cardiomyopathy, cardiac masses, pericardial disease or tamponade), pulmonary embolus, or pulmonary hypertension can cause syncope when circulatory demands outweigh the impaired ability of the heart to increase its output. The basis for the syncope is inadequate blood flow due to mechanical obstruction. Nonetheless, in several cases, syncope is not solely the result of restricted cardiac output, but may be in part due to an inappropriate reflex or orthostatic hypertension.

Arrhythmias are the most common cardiac causes of syncope. They induce haemodynamic impairment, which can cause a critical decrease in cardiac output, and cerebral blood flow. Nonetheless, syncope often has multiple contributory factors, including heart rate, type of arrhythmia (supraventricular or ventricular), left ventricular function, posture, and adequacy of vascular compensation. The latter include baroreceptor neural reflexes as well as responses to orthostatic hypertension induced by the arrhythmia. Sick sinus syndrome, atrial standstill and the more severe forms of acquired atrioventricular block (advanced II degree AV block and complete AV block) are most closely related to syncope.

Diagnostic reasoning for syncope

Patient with frequent T-LOC can be assessed with echocardiography and prolonged heart rhythm recording such as Holter monitoring and cardiac event recorder. In current practice Electrocardiographic monitoring is usually undertaken with conventional 24–48 h, or even 7day event recorders. However, since in most of the patients symptoms do not recur during the monitoring period, the true yield of Holter in syncope may be low. Holter monitoring in syncope may be of more value if symptoms are very frequent. Daily single or multiple episodes of TLOC might increase the potential for symptom–electrocardiographic correlation. Seven day external loop recorders (R test) are external devices which

have a loop memory that continuously records and deletes electrocardiographic. When activated by the owners, typically after a symptom has occurred electrocardiographic is stored and can be retrieved for analysis. Implantable loop recorders (ILR) are implanted subcutaneously under local anaesthesia and have a battery life of up to 36 months. These devices have a solid-state loop memory that stores retrospective ECG recordings, when activated either by the owners after a syncope or automatically activated in the case of occurrence of predefined arrhythmias. Disadvantages include: the need for a minor surgical procedure, the fact that sometimes it can be difficult to differentiate between supraventricular or ventricular arrhythmias, the presence of under or oversensing due to the arrhythmia algorithm studied for the man and the cost of the device.

Clinical management of syncope

Arrhythmic and frequent reflex syncope are both at risk of sudden cardiac death. Clinical management for syncope provoked by intrinsic bradyarrhythmias such as sinus node dysfunction and atrioventricular blocks should be treated with a permanent pacemaker implantation. Same treatment can be used to reduce the clinical symptoms and the risk of sudden cardiac death in patient with frequent and therefore defined malignant reflex syncope. In this latter cases the clinical symptoms are just reduced since the vasodepressor component of the reflex syncope is not addressed. Supraventricular and ventricular arrhythmias according with the underlining mechanism and the eventual results of endocardial mapping can be treated with antiarrhythmic drug, such as sodium blockers, b-blockers, potassium blockers and calcium channel-blockers and in particular cases with radiofrequency catheter ablation.

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Viral Myocarditis – A very rare disease or just difficult to diagnose?

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Myocarditis is an inflammatory disease of the heart muscle, diagnosed by histology, immunology and immunohistochemistry. Myocarditis is the result of RNA and DNA virus infections (enterovirus, adenovirus, parvovirus B19, cytomegalovirus and herpes viruses) that induce a post-viral immune-mediated response. Myocarditis is considered a precursor of dilated cardiomyopathy with systolic dysfunction development in approximately 21% of patients over a period of about three years after the acute event. Myocarditis is also a cause of sudden death in approximately 12% of young adults. In dogs, there are only few case reports describing myocarditis caused by different viral agents, bacterial and protozoan. The clinical pictures of dogs suffering from myocarditis include myocardial disorders with hypokinetic-dilatation, acute heart failure, ventricular arrhythmias and transient atrioventricular block. The viral myocarditis has a triphasic pattern that includes an acute onset with viremia, a subacute phase with the host response and a chronic immune-mediated characterized by varying degrees of ventricular dysfunction. The acute phase is predominantly characterized by necrosis with loss of myocytes induced by virus, the cytotoxic effects of inflammatory mediators and oxidative stress products that induce endothelial dysfunction and ischemia. The viral activity induces, in addition, a complex immune response characterized by a remarkable infiltration of inflammatory cells such as natural killer cells and macrophages and the production of cytokinin. Neutralizing antibodies are not detectable until the fourth day after the viral infection begins when the virus titres are extremely high. The securities reached peak levels on the fourteenth day and correlate with viral elimination from cardiac tissues. The subacute phase lasts from day 4 to day 14 post-infection and is characterized by the infiltration of T lymphocytes. The biggest myocardial cell damage occurs during this period also begins when the infiltration of lymphocytes B. The humoral immune response plays an important role in the processes of myocardial damage with release of anti-myosin heavy chain antibodies. The chronic phase extending from days 15 to 90 days post-infection and is characterized by intense deposition of interstitial myocardial collagen, myocardial fibrosis and the progression of myocardial damage with cardiac dilatation, systolic dysfunction and congestive heart failure.

New studies have suggested that the progression to the stage of dilated cardiomyopathy is induced by the persistence of RNA viruses in myocytes beyond 90 days after the beginning of the event, with cell apoptosis and infiltrate of lymphocytes T. Myocarditis is preceded by flu-like symptoms, and gastrointestinal symptoms such as decreased appetite, nausea, vomiting and diarrhea. Cardiac manifestations of myocarditis appear a few hours to a few days after the first symptoms and are represented by heart failure, chest pain due to irritation pericardial, and arrhythmias such as the transient heart block (16% of patients) and ventricular arrhythmias. In the acute phase, it follows a monophasic clinical course, and the majority of patients recover spontaneously after a few days. On the other hand, some patients with acute myocarditis progressing rapidly in cardiogenic shock. Some patients progress to subacute or chronic forms, which port to chronic heart failure. Although certain blood markers such as troponin I can help in the suspected diagnosis of myocarditis, the gold standard for diagnosis remains the endomyocardial biopsy. The progressive increase of troponin in 24 hours with peak one or more days after the first upward may aid in differentiating myocarditis from acute coronary syndrome.

There are several clinical and histopathologic criteria for classifying myocarditis. According to the Dallas criteria, the acute myocarditis is characterized by lymphocytic infiltrates in association with myocyte necrosis, while the borderline myocarditis is characterized by inflammatory infiltrates without evidence of myocyte necrosis. The Dallas criteria are limited by the high variability of biopsy interpretation and why inflammatory processes non-cellular samples cannot be detected. Immunohistochemistry is gaining consensus in the diagnosis of myocarditis. The monoclonal antibodies allow the characterization and the localization of the infiltrating mononuclear cell: for example, CD3 for T cells, PGM1 (CD68) for activated macrophages, and human leukocyte antigen (HLA) - DR - can evaluate HLA class II. Another classifying method definable with endomyocardial biopsy involves the division of myocarditis in acute, fast and chronic progression. Acute forms are characterized by extensive necrosis with lymphoplasmacell infiltrate, the rapidly progressive forms are characterized by extensive cell damage and marked fibrosis, in chronic forms the cell damage is minimal and focal. One last classifying method combines the histological findings to clinical and divides in fulminant myocarditis, giant cell, eosinophilic, chronic, pediatric and neonatal.

Endomyocardial biopsy is a safe procedure is performed through the skin with a byoptome with predetermined curvature and is guided by fluoroscopy. Is usually done by passing through the jugular from the right ventricle, although some studies have shown that performing endomyocardial biopsies sinister may increase the diagnostic power especially if the right ventricular systolic dysfunction on echocardiography shows. In the acute phase, endomyocardial biopsy is usually not necessary, but virological diagnosis is possible using some techniques such as PCR and in situ hybridization. PCR techniques have positive results in 20% of patients with clinically suspected myocarditis or dilated cardiomyopathy. The frequency is much higher in the first phase of the disease. The in situ hybridization technique detects viral genome in 35% of affected patients. The sub-acute phase of the disease, where the immune activation occurs, it can be definitely diagnosed by endomyocardial biopsy. The diagnosis is safer when made a few days or a few weeks after the resolution of a viral infection, and in this phase many lymphocytic infiltration outbreaks can be detected with histology. In veterinary medicine there is only one study that evaluated the presence of viral genome histological specimens fixed with formalin in 18 dogs with dilated cardiomyopathy and 9 dogs with myocarditis. In this study only one dog with dilated cardiomyopathy showed the presence of an adenovirus type 1. The sensitivity of endomyocardial biopsy depends on the number of samples tested. The data coming from a large survey involving 4000 patients suggest that the frequency of positive biopsy in patients with myocarditis or dilated cardiomyopathy is low (10%), and increases with increasing the number of samples.

The risks of endomyocardial biopsy can be divided into immediate or delayed . The immediate risks include cardiac perforation with cardiac tamponade , ventricular or supraventricular arrhythmias , heart block , pneumothorax , puncture of central arteries , pulmonary embolism , hematoma at the venous access site , damage to the tricuspid valve , and the creation of a arteriovenous fistula within the heart. The use of an introducer along which runs through the tricuspid valve may decrease the risk of trauma induced by byoptome. Delayed complications include bleeding in the venous access site , damage to the tricuspid valve , pericardial tamponade , and deep vein thrombosis. In humans, the complication rate of endomyocardial biopsy varies from 1 to 1.5 % with very rare cases of death caused by myocardial perforation.

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Edward Hall

Diagnostic Approach to GI Signs

Vomiting and diarrhoea may be an indication of primary gastrointestinal (GI) disease, but can also be caused by a large number of extra-GI conditions. Concentration of investigations on the GI tract too early in the diagnostic approach may lead to unnecessary tests being performed and can result in an erroneous diagnosis with, at worst, fatal consequences. However, through careful history-taking and thorough physical examination, with particular emphasis on all body organ systems, the clinician should avoid such errors. Only after disease in other organ systems has been evaluated and ruled out is it safe to formulate a diagnostic plan for the investigation of GI disease.

Although vomiting and diarrhoea are the most easily recognised signs of GI disease, they may be caused by non-GI disease, and other less specific signs may be noted. Signs not specific to the GI tract are particularly important as they may provide clues that extra-GI disease is present.

Specific signs

- Vomition
- Haematemesis
- Diarrhoea
- Melaena
- Haematochezia
- Abdominal pain / discomfort
- Bloat
- Borborygmus
- Flatulence
- Dyschezia
- Tenesmus

Non-specific signs

- Anorexia
- Polyphagia ± pica
- Weight loss
- Fever
- Depression
- Polydipsia
- Coughing
- Behavioural changes
- Ascites and oedema
- Ptyalism
- Halitosis
- Pruritus
- Dehydration

Throughout investigation, attention must be paid to the patient's fluid and electrolyte status, and parenteral fluid therapy administered as necessary.

Withholding of food (and oral fluids) is generally appropriate in acute GI disease, but is pointless in chronic disease except if specifically indicated for any particular investigation as it merely leads to further malnutrition.

General Scheme for Diagnosis of GI Disease

Investigations

1. History
2. Physical examination
3. Minimum data base
4. Faecal examination
5. Specific laboratory tests
6. Imaging
7. Biopsy (endoscopy, laparotomy)
8. Empirical treatment (at some point)

Exclusion of non-GI (secondary) GI disease

Causes of secondary GI disease are excluded by a combination of history, physical examination, and a minimum data base of laboratory tests. A thorough systems review with questions investigating any potentially important signs such as polyuria/polydipsia, coughing, sneezing, dyspnoea, dysuria and neurological signs should be made.

Stage 1

- exclusion of dietary problems, parasitism, systemic disorders
- exclusion of non-GI disease
- anatomical localisation using history & physical exam (oesophagus, stomach, intestine, diffuse?)
- faecal exam and preliminary lab tests (minimum data base)
- many cases are resolved in this stage, and don't need in-depth investigation

Stage 2

Definitive aetiological/histopathological diagnosis via an extended data base:

- special tests (TLI, PLI, folate / B12)
- imaging
- anatomical localisation using history & physical exam (where to biopsy?)
- endoscopy or surgery with biopsy
- response to therapy

History

Good history taking is an acquired art, but a logical approach will produce the best results.

- Owners' primary complaint
  Although the clinician may recognise other problems as being more important, failure to address what the client perceives to be the major problem leads to dissatisfaction.
  Occasionally what is perceived to be diarrhoea by clients is either a normal response of the GI tract, or is not in fact even diarrhoea:
  - 'physiological' response
  - osmotic laxatives and cathartics
  - dietary indiscretion - excess fat, lactose, fibre
  - misdiagnosis
    - constipation with expulsion of scant amounts of liquid faeces
    - haematochezia without diarrhoea due to a rectal polyp frequently mistaken for colitis
    - tenesmus and/or dyschezia associated with prostatomegaly, perineal hernia/fistula
- Patient information
  Breed predisposition

Basenji	- lymphocytic/plasmacytic enteritis
Belgian shepherd	- gastric carcinoma
Boxer	- mastocytoma, histiocytic ulcerative colitis
Collies	- gastric carcinoma, exocrine pancreatic insufficiency
German shepherd	- EPI, SIBO, IBD (LPE, EGE)
Irish setter	- gluten-sensitive enteropathy
Lundehund	- lymphangiectasia, gastric carcinoma
Miniature Schnauzer	- hyperlipidaemia, pancreatitis, haemorrhagic gastroenteritis
Rottweiler	- lymphangiectasia, parvovirus (all black and tan breeds)
Shar pei	- hiatal hernia, inflammatory bowel disease, (IgA deficiency)
Siamese cat	- pyloric stenosis, inflammatory bowel disease, adenocarcinoma

Age & sex

The age of the patient influences the likelihood of different diseases. Dietary indiscretion, infectious gastroenteritis, and congenital pyloric stenosis are more common in younger dogs, whereas neoplasia is more common in mature animals. There are no reported sex predispositions in GI disease.

- Medical history
  - prior medical history should be established, especially:
    - vaccination and worming status
    - administration of NSAIDs
    - administration any drugs known to cause vomiting and/or diarrhoea
- Environmental & dietary history
  - whether the patient roams and perhaps scavenges (i.e. roams / eats garbage)
  - geographical history
  - possible exposure to infections
  - whether other pets or littermates or even the owner are ill
  - information on the previous day's intake may be all that is required in acute disease
  - investigation of chronic problems requires a complete dietary history which is often confused; rescheduling for a longer consultation may be helpful
- Body systems review
  A series of key 'closed' questions to ascertain whether other organ systems are involved, e.g.
  - change in drinking
  - cough, dyspnoea, tachypnoea
  - collapse, seizures
  - pruritus, etc., etc.
- Clinical signs
  - nature
  - frequency
  - localisation
  - severity
  - sequence
  - progression
  - modifications (inciting causes, alleviating factors)

Vomiting & diarrhoea

Cardinal signs of GI disease.

Abdominal discomfort or pain

Pain is produced by distension of a viscus and/or inflammation. Some cases of hypoadrenocorticism or lead poisoning may also show apparent pain on abdominal palpation. Stimulation of vomiting just by abdominal palpation may indicate generalised peritonitis or localised inflammation as in acute pancreatitis.

Appetite

An imprecise generalisation is that polyphagia in GI disease is a sign of malabsorption due to benign causes; anorexia tends to indicate surgical disease, malignancy, or disease of other organ systems. True polyphagia should be distinguished from hunger and repeated attempts to eat the same food in dysphagic animals. Polyphagia may be accompanied by coprophagy and other examples of pica. Coprophagy is most commonly seen in EPI and SIBO.

Ascites

Ascites and/or peripheral oedema occur when hypoproteinaemia is present, and may result from a protein-losing enteropathy (PLE). It is a marker of poorer prognosis. Any severe, generalised, and usually chronic intestinal disease, such as severe IBD, lymphosarcoma and lymphangiectasia may be a PLE. Parvovirus gastroenteritis is a PLE, but usually too acute to cause ascites.

Drinking

Apparent polydipsia may reflect dehydration because of GI fluid loss, and is often seen in acute vomiting. Checking urine specific gravity help confirm concurrent polyuria.

Gas

Excessive eructation, borborygmus and flatulence may merely indicate dietary indiscretion, but may indicate abnormal bacterial fermentation (e.g. in SIBO).

Halitosis

Halitosis is commonly related to oropharyngeal or perianal licking, and may be normal in animals fed a high protein diet. However, coprophagia and malabsorption may be responsible.

Weight loss

This may be due to a decreased intake, decreased absorption, or increased protein loss through the GI tract [protein-losing enteropathy (PLE)].

Physical examination

All body systems should be examined, but only those directly relevant to the GI tract are discussed.

Observation

If time is taken to observe the unrestrained patient an evaluation of its weight, respiratory rate, mentation and behaviour, and signs of hepatoencephalopathy can be made. The 'prayer position' is seen rarely, but if present reflects cranial abdominal pain associated with pancreatitis or deep gastric or duodenal ulcers. Offering food and water and observation is helpful in distinguishing dysphagia, regurgitation and vomiting. Nutritional status may reflect the severity and duration of the disease.

Oral examination

The mouth is examined, not only to assess hydration and cardiovascular status, but also to check for ulceration perhaps associated with uraemia. The base of the tongue should be checked for linear FB.

Abdominal palpation

Palpation is used to detect organ enlargement and/or displacement, abnormal masses or pain. Palpable masses may be related to the intestine (neoplasia, intussusception) or other abdominal organs. Pain is associated with GI distension & inflammation, pancreatic inflammation and peritonitis.

Rectal examination

The performance of a rectal examination is mandatory in all cases of vomiting and diarrhoea. It provides a sample of stool and may identify previously unnoticed diarrhoea or melaena, as well as rectal masses, hernias etc. A dry, tacky rectal mucosa may be found in cases of intestinal obstruction.

Minimum data base

- Routine haematology
- Serum biochemical screen
- Urinalysis
- Rectal cytology

Faecal examination

- Microscopy
  - undigested muscle fibres, fat droplets, starch granules - not useful
  - rectal cytology – to find inflammatory or neoplastic cells
- Parasitology

- Stool culture

  Microbiological culture of stool from a patient with diarrhoea is often undertaken in practice, perhaps because there is a belief that bacterial enteritis is common and/or because it is a simple test to perform. Although a culture is preferable to unjustified use of antibacterials, regrettably neither a positive nor a negative result is simple to interpret. A negative result may actually be a false negative, but if there is a "positive stool culture", a specific, known infectious agent has been isolated. Yet can it be assumed that the cause and potential treatment have also been identified? Whilst this is not a true false positive, as the organism is clearly present, the problem comes from knowing whether:
- the isolate is truly pathogenic?
- the isolate is the cause of the signs?
  - another, non-bacterial pathogen present?
  - another underlying disease?
- any treatment is necessary?
- any treatment is safe?

Salmonella spp.

Infection is quite rare in dogs, and treatment is probably unnecessary and potentially dangerous because of the development of resistance and/or the carrier state. Hygiene precautions are necessary as the organism is potentially zoonotic.

Campylobacter spp

Campylobacters are the most commonly isolated potential enteropathogens in dogs, and can cause acute and chronic disease. Acute enterocolitis typically results in diarrhoea that often contains blood and mucus; vomiting, inappetence, and abdominal pain with pyrexia are less common. However, most Campylobacter isolates in dogs are potentially the less pathogenic C. upsaliensis, and not C. jejuni. Thus although a tentative diagnosis may be made on the isolation of these organisms from an animal demonstrating these signs, isolation is not proof of causality.

Escherichia coli

Pathogenicity is based on the presence of virulence genes that encode for various toxins etc. However, identification of genes encoding virulence determinants by PCR does not necessarily indicate that the organism is responsible for the clinical signs.

Clostridium spp.

Clostridium perfringens and C. difficile may cause diarrhoea as a consequence of enterotoxin production under certain 'environmental conditions'. However, they are sometimes described as nosocomial infections as clostridial-associated diarrhoea is typically seen in hospitalized animals. Whilst infection with C. difficile may be a true nosocomial

event, C. perfringens is generally considered part of the normal resident microflora of dogs and cats that can cause disease through dysbiosis. 'Flare factors' that may trigger it to produce enterotoxin are believed to include hospitalization and sudden diet change, two events that frequently occur together.

Radiology

- to look for masses, complete or partial obstructions, etc.
  - loop of bowel > 2 x width of 10th rib, or > 1.5 height of body of L5 indicates obstruction
- unhelpful if significant ascites present
- barium studies useful for regurgitation (and vomiting?) but rarely diagnostic in malabsorption except when severe infiltrative bowel disease, and gut biopsy is still required
- Barium-impregnated polyethylene spheres (BIPS) to identify partial obstructions?

Ultrasound

- US is operator- and equipment- dependent
- to look for masses, complete or partial obstructions, intussusceptions, wall thickness and layering
- increased wall thickness is not a consistent feature of IBD, and may be due to oedema in a PLE
- loss of layering is a significant marker of potential malignancy
- mucosal striations may indicate lymphatic dilatation
- mesenteric lymphadenopathy may reflect inflammatory or neoplastic disease and FNAs may help

These two imaging techniques are complimentary, not mutually exclusive:

- general location of a mass is easier to appreciate on radiographs
- loops of gas-filled bowel aid radiographic interpretation but hinder US
- free abdominal fluid hinders radiographs but enhances US
- determination of organ of origin of a mass easier with US
- wall thickness and motility cannot be assessed on plain radiographs, but are evident on US

The use of intestinal biopsy is discussed later, but in acute GI disease, endoscopy and/or surgery with intestinal biopsy are usually not indicated for diagnostic purposes, and are only normally performed if there is a specific treatable condition diagnosed, e.g. foreign body, intestinal obstruction.

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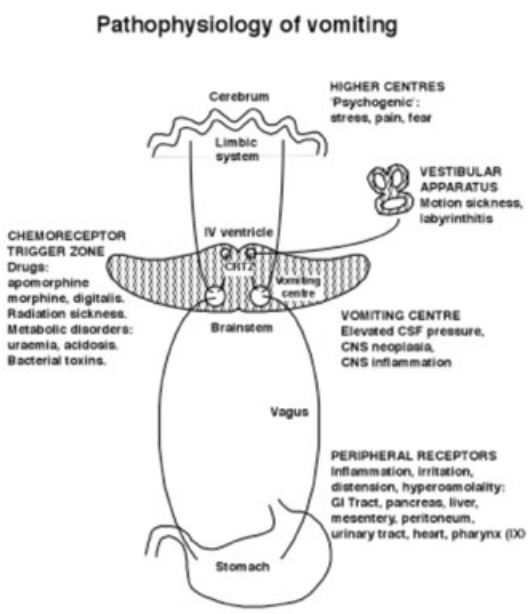
Vomiting and its control

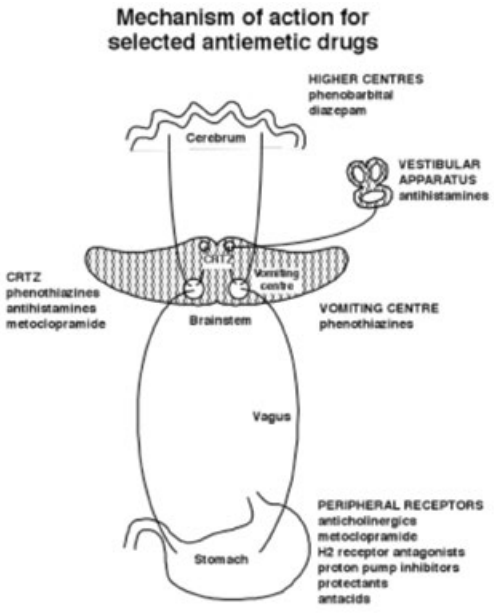
There are well over forty specific causes of vomiting recognised, and it is helpful to classify them by their mechanisms of action rather than listing them all. Primary vomiting is induced by primary GI disease, whereas secondary vomiting is a feature of disease in other organ systems. Further classification is related to the reflex nature of vomiting and the pathway stimulating the vomiting centre.

The efferent arm of the reflex from the vomiting centre is carried by cranial nerves IX and X, and co-ordinates the motor changes in the pharynx and GI tract with abdominal contractions. However, there are a number of afferents to the brainstem by which different diseases elicit vomiting.

Peripheral sensory receptors are located throughout the GI tract:

- majority in duodenum, but receptors in the stomach, jejunum, ileum and colon
- respond to distension, inflammation, irritation and hyperosmolality:
- intestinal inflammation causes vomiting more frequently than diarrhoea in cats
- chemoreceptors in the stomach that are stimulated by emetics such as ipecac
- receptors also in pancreas, liver, urogenital tract, mesentery and peritoneum
- receptors in pharynx transmit impulses via the glossopharyngeal
- no myocardial or coronary receptors in dogs, and vomiting is not a sign of heart disease;





The vomiting centre receives inputs from rest of the brain

- vestibular afferent input: inner ear disease and motion sickness may induce vomiting
- inflammatory CNS lesions, SOL's and increased intracranial pressure
- cerebral cortex: vomiting as a result of fear, stress and excitement is initiated here
  - psychogenic vomiting and so-called limbal epilepsy are rare, poorly defined causes
- the chemoreceptor trigger zone (CRTZ) on the floor of the fourth ventricle
  - stimulated by blood borne substances: apomorphine, morphine, uraemic toxins, digoxin, endotoxins, acidosis and some chemotherapy agents

Vomiting – historical clues

- Duration
- Frequency
- Timing
- Action
- Nature of vomitus

Anti-emetics

Maropitant (Cerenia)

Licensed for dogs and cats at a dose of 1.0 mg/kg q24h SC and 2 mg/kg q24h PO. It can be given slow IV, but the need for such rapid onset of action is rare, and adverse reactions have been recorded. The most common reaction when used by an approved route is a sting at the injection site, which is related to the temperature of the drug; inject cold.

Maropitant is an NK_1 antagonist, inhibiting neurotransmission by substance P at the final common pathway within the vomiting centre of the medulla. It is effective as a daily SC or IV injection (1 mg/kg), followed by oral tablets. The SC injection may sting, especially if the injection is warm. Maropitant's long duration of action is related to protein binding, and after five days treatment, a wash-out period of 2 days should be given before repeating the course if necessary. Maropitant is less effective against motion sickness as a dose of 8 mg/kg is needed, perhaps because it involves cortical activity in the sensation of nausea?

Metoclopramide (Vomend, Emeprid)

This is a well recognised and generally safe anti-emetic and prokinetic, and for a long time has been the first choice anti-emetic in dogs. Although it has no direct effect on healing of oesophagitis and gastritis, it helps by increasing lower oesophageal tone and emptying the stomach of toxic acid, pepsin and bile salts.

It has a central anti-D_2 dopaminergic effect, being active against emetics that stimulate the chemoreceptor trigger zone. It also has a peripheral cholinergic effect stimulating upper GI motility, ands is thus contra-indicated if there is a GI obstruction. In some patients, especially cats, it may cause bizarre behavioural changes ranging from lethargy to hyperactivity and even vestibular signs, although this is largely dose-related. It is excreted by the kidneys, and toxicosis is more likely in renal insufficiency. It is useful as a continuous intravenous infusion in puppies recovering from Parvovirus.

Ondansetron

Ondansetron (Zofran) is a 5-HT3 antagonist and is a very potent anti-emetic developed to give to humans receiving chemotherapy. It is also very effective in dogs but is very expensive. Side-effects are mild but include sedation and head-shaking. Related drugs (e.g. granisetron, tropisetron, dolasetron) are available.

Others

The phenothiazines (e.g. chlorpromazine) and antihistamines (e.g. diphenhydramine) remain effective anti-emetics in the right situation, but tend to cause drowsiness.

Prokinetics

Metoclopramide

As well as an anti-emetic, metoclopramide is an effective prokinetic in the stomach and duodenum. Its other actions, and that of ranitidine, are discussed elsewhere.

Cisapride

This was the most effective prokinetic available, but has been withdrawn over safety fears (prolonged QT interval, and human deaths). It acted by stimulating intestinal 5HT4 receptors, resulting in increased LES tone, decreased pyloric tone, and propulsive peristaltic waves in stomach, duodenum, jejunum and colon. It was indicated for gastro-oesophageal reflux and GI motility disorders including feline idiopathic megacolon. Prucalopride and tegaserod were developed but had similar effects and were not marketed. Cisapride is available in some EU countries, and mosapride in Japan.

Erythromycin

The emetic effect of antibacterial dosages (10 mg/kg) of erythromycin is well recognised. The mechanism is believed to be stimulation of motilin receptors in the GI tract. At lower doses, erythromycin stimulates more normal migrating motor complexes. Thus erythromycin, at doses of 1 2 mg/kg q 8h, can be used as an effective prokinetic.

Haematemesis

- occasional flecks of fresh blood in vomitus from an acutely vomiting dog are expected
- larger amounts of blood or recent onset in a chronically vomiting patient are significant
    - once a generalised bleeding problem or swallowing of blood has been excluded, haematemesis suggests gastric ulceration, or reflux of upper duodenal bleeding
    - fresh blood, clots or partially digested blood with the appearance of ‘coffee-grounds’ may be seen, and there is often consequent melaena
    - the blood usually originates from gastric mucosal ulceration, although not all dogs with gastric ulcers show macroscopic haematemesis

Gastric ulceration

Any mechanism that interferes with the gastric mucosal barrier is likely to cause gastritis and ulceration. Gastritis underlies many of the diseases that affect the stomach. If there is failure to heal after an acute gastritis, the response may be perpetuated as chronic gastritis or, if severe, may lead to gastric ulceration.

A combination of factors affecting different parts of the barrier is likely to be more devastating, as often happens with drug-induced gastric ulcers. Spontaneous ulcers in man were originally linked to ‘stress’ and hyperacidity, but are now known to be related to H. pylori infection. The role of similar spiral organisms in companion animals is controversial but the incidence of spontaneous ulcers seems low.

Causes of damage to the gastric mucosal barrier

Ischaemia

Any hypotensive event (dehydration, shock, general anaesthesia), and consequent reduction in gastric mucosal blood flow, has the potential to lead to damage to the gastric mucosal barrier, through lack of the normal protective microcirculation and acute energy deprivation of the mucosal cells. Whilst most 'sick' animals (e.g. pyometra) will have clinically insignificant gastric erosions, the potential for clinical significance when other factors, such as ulcerogenic drugs, are added must not be forgotten.

Metabolic diseases (hepatic, renal etc.)

Paraneoplastic syndromes

GI ulcers secondary to mast cell tumours secreting histamine and, more rarely, gastrinomas secreting excess gastrin are caused by gastric acid hypersecretion.

NSAIDs

The gastro-erosive effects of NSAIDs are well recognised although often under-diagnosed. In man there is also clear evidence of the development of distal SI inflammation and even stricture formation after prolonged NSAID use. The gastric effects are thought to related to non-selective inhibition of constitutive (and beneficial) cyclo-oxygenase 1 (COX-1) that normally produces cytoprotective PGs. Therapeutic benefits are linked to inhibition of induced, pro-inflammatory COX-2. Thus safety of NSAIDs has been linked to a high COX-2 : COX-1 ratio. Nevertheless no NSAID is effective and completely safe! Meloxicam is COX-2 selective and is considered relatively safe, but ulcers may develop even after months of successful treatment with no side-effects. Carprofen probably acts by other mechanisms, making them currently one of the safest NSAIDs in dogs.

There has been much active research to find specific COX-2. Deracoxib (Deramaxx), firacoxib (Previcox) and robenacoxib (Onsior) are now marketed for dogs; robenacoxib has preferred pharmacokinetics, with a short peak in serum but prolonged activity at the site of inflammation. Mavacoxib (Trocoxil) is plasma protein bound with a long half-life and is given once a month; adverse reactions are fortunately rare.

Other Drugs

Cytotoxic drugs may be ulcerogenic.

Corticosteroids are probably not normally ulcerogenic in normal animals except in very high doses. However, by inhibiting mucus production, cell renewal, and fibrosis, and thereby delaying wound healing, they have the ability to enhance any other ulcerogenic event. Indeed the combination of steroids and NSAIDs is potentially lethal.

Treatment of gastric ulceration

Mucosal protectants

Sucralfate

Sucralfate (Antepsin, Carafate) is a complex of sucrose octasulphate and aluminium hydroxide, available as tablets and suspension. It is simplistically believed to aid healing of oesophageal and gastric erosion/ulceration by precipitating and binding to the tissue, providing a barrier against the penetration of gastric acid and pepsin ("gastric Band-Aid"). Whilst this mechanism is important, it is now clear that sucralfate has other beneficial effects:

- inactivates pepsin

- binds refluxed bile acids
- cytoprotective through stimulation of endogenous PG synthesis
  i.e. increases mucus and bicarbonate secretion etc.
- binds and concentrates epidermal growth factor (EGF) where it stimulates cell proliferation etc.

The safety of sucralfate is well established, with occasional constipation being the only reported side-effect. However, its efficacy in treating GI ulceration and bleeding is not established.

The other debate about sucralfate is its drug interactions; it makes enrofloxacin unavailable for absorption. There is a theoretical argument that co-administration of H2 antagonists with sucralfate prevents their absorption; conversely the blockade of acid secretion prevents the binding of sucralfate. These arguments appear to have little basis in fact, and co administration is probably not a problem. Nevertheless most recommend these drugs are given at least an hour apart, although which should be given first is not clear! Considering its mechanism of action, it might actually make sense to give sucralfate both before and after feeding, with acid blockers being given with food.

Prostaglandins

The gastric mucosal barrier has been discussed earlier, but the role of prostaglandins in maintaining the barrier cannot be overlooked.

The cytoprotective effects of PGEs are related to:

- increased gastric mucus secretion
- increased gastric mucosal blood flow
- decreased gastric acid secretion
- stabilisation of histamine-containing ECL cells

Misoprostol (Cytotec) and enprostil are synthetic PGEs (E1 and E2 respectively). Misoprostol has been shown to be effective in dogs at protecting the gastric mucosa from aspirin-induced ulceration. It is not effective at healing pre-existing ulcers: equally, H_2 antagonists and sucralfate do not prevent the development of NSAID-induced damage, but are important in its treatment.

At the recommended dose of 2-5 µg/kg q8h, diarrhoea is a rare side-effect of misoprostol (cf. man) and is usually self-limiting. However, it will cause abortions and must not be used in pregnancy. There is insufficient evidence to recommend its use in cats.

Acid blockers

Acid blockade is an important part of healing oesophageal and gastric ulceration, and although effective drugs have been available for over 25 years, more recently more potent drugs needing just once a day treatment have become available.

Antacids (aluminium hydroxide, magnesium carbonate, calcium carbonate etc.)

These are safe and effective, but have fallen from favour because of the need to be given 4-6 times daily. However, as well as neutralising gastric acid they:

- decrease pepsin activity
- bind bile acids
- stimulate bicarbonate secretion
- possibly increase endogenous PG production

H_2 antagonists

Cimetidine (Tagamet) has been in use since the 70's; a veterinary licensed product (Zitac) is now available. It is an

effective H2 antagonist that can be given orally and parenterally, but it needs to be given at least three and probably four times daily. It also has significant drug interactions through its effects on hepatic cytochrome P_{450} activity and can cause steroid hormone-related side-effects.

Ranitidine (Zantac), nizatidine (Axid), and famotidine (Pepcid) do not have these side-effects, are more potent and need to be given only twice or even once daily. Increased potency (ranitidine and nizatidine 5x, famotidine 20-30 x) is not necessarily an advantage as equipotent dosages are used. However, reduction in dosage frequency to q 24h for famotidine is an advantage that may be offset by the increased cost! Nevertheless there is no data to prove that any one of these drugs is more effective at ulcer healing if used appropriately (i.e. correct dose and dosing interval). There is also evidence that ranitidine and nizatidine are prokinetic through stimulation of local acetylcholine receptors. This has led to recommendations for their use in gastroparesis and feline idiopathic megacolon. This prokinetic effect is probably a good reason to choose ranitidine over cimetidine to treat non-specific gastritis in addition to its preferable pharmacokinetic profile.

Proton pump inhibitors (PPi)

These drugs irreversibly bind the proton pump, i.e. the final step in acid secretion (H+K+-ATPase), and so are very potent inhibitors of acid secretion,, whereas H2 antagonists are competitive antagonists. It is debated whether twice daily administration is necessary initially, but after five doses, acid secretion is completely abolished. Because of their potency, PPis can cause reflex hypergastrinaemia, and this has been linked to induction of carcinoid tumours in rodents. A similar sequel has not been seen in other species, but the licensed use of these agents in man is limited because of the potential risks. No problems have been seen in dogs and cats, but their use has been limited until recently. Now, because of their once a day administration and only marginally greater cost, they are being used more frequently.

Omeprazole (Losec, Prilosec in USA) was the first proton pump inhibitor to be marketed, and is probably the only one needed in companion animal practice. It is available in convenient 10 and 20 mg capsules and an intravenous preparation.

Lansoprazole (Zoton), pantoprazole (Protium) and rabeprazole (Pariet, Aciphex) are now available but appear to have little benefit over omeprazole. Pantoprazole was the first to be marketed for intravenous use, but is reported to cause pulmonary oedema in dogs at high (7 mg/kg) doses. Intravenous Losec is thus safer, although both products come as 40 mg vials that have to be used immediately on reconstitution and so are expensive. Esomeprazole (Nexium) is the active isomer of omeprazole.

Omeprazole is indicated for severe reflux oesophagitis, gastritis and gastric ulcers, and in rare conditions such as gastrinomas (Zollinger-Ellison syndrome).

Roberto Santilli

Indications for the Holter exam. Its utility and limitations as a diagnostic test.

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Introduction

Due to the transient behaviour of cardiac arrhythmias several prolonged rhythm recording have been proposed: continuous ambulatory electrocardiography (Holter monitoring), external cardiac event recording and implantable loop-recorder (Reveal). These techniques have a significantly higher diagnostic yield when compared with routine electrocardiography (Holter 42-44%; external loop-recorder 84%; implantable loo-recorder 34-66 %), and are considered highly sensitive devices to assess need and efficacy of antiarrhythmic therapy.

24-hour Holter recording

Routine ambulatory electrocardiography is commonly performed in veterinary medicine to assess the presence and mechanism of cardiac arrhythmias due to the possibility to evaluate the behaviour of the rhythm disturbances and their correlation to the autonomic nervous system and the analyse proper function of the sinus node. Ambulatory electrocardiography has been invented in the late 1950's by Dr. Norman Holter, which built a system capable of long-term continuous electrocardiographic monitoring. To perform Holter monitoring, 3 channels of modified chest leads or an orthogonal system (X,Y,Z) are recorded using adhesive patch electrodes attached to a recorder. The patches, lead wires and recorder are then secured using a chest wrap. In most cases, Holter monitoring is performed over a continuous period of 24-48 hours during which the patient engages in routine activity noted on a specific diary. The exam is then downloaded on a software where instructed personal manually edit the recording. Most systems are capable of fully-automated analysis, although this usually results in a high degree of artefacts and incorrect classification of beats. Significant problems in the analysis of veterinary Holter recordings include the occurrence of sudden patient movement, which may be classified as ventricular ectopy and the presence of sinus arrhythmia, which is not accepted as normal by software algorithms designed for human medicine. Careful analysis of an exam may take as long as 2-3 hours when frequent arrhythmia or artefacts are present.

Holter monitoring is indicated in case of syncope and lethargy, familiar arrhythmias, familiar cardiomyopathy, to assess necessity and efficacy of antiarrhythmic treatment.

Patient with frequent transient loss of consciousness (T-LOC) can be assessed with echocardiography and prolonged heart rhythm recording such as Holter monitoring and cardiac event recorder. Holter monitoring in syncope may be of more value if symptoms are very frequent. Daily single or multiple episodes of TLOC might increase the potential for symptom–electrocardiographic correlation. In dog almost one fourth of animals evaluated presented a syncopal event during the recordings, with a correlation to the diagnosis in the 42% of the dog. An arrhythmia was ruled in as the cause of the syncope in the 30% of the cases, of these 20% was tachyarrhythmias and 10% bradyarrhythmias. In the 38% of the cases the Holter monitoring led to a therapeutic change.

The occurrence of ventricular ectopy, in the absence of non-cardiac disease, has been demonstrated to be a reliable marker for occult dilated cardiomyopathy in Doberman pinschers and significantly precedes any clinical signs of heart disease. Transient ventricular arrhythmias, often not detected with routine electrocardiography, have also been documented in Boxers and English Bulldog from several families affected with arrhythmogenic cardiomyopathy. In Doberman pinschers with no overt clinical signs of heart disease, Holter monitoring demonstrated the occurrence of over 100 ventricular premature complexes per 24 hour period in 81% of the cases that went on to develop overt dilated cardiomyopathy or sudden death. The presence of less than 50 ventricular ectopies in 24 hour period rule out dilated cardiomyopathy, border line for diagnosis are case in which ventricular ectopies are between 51 and 100, while dogs with more than 100 ventricular ectopies are considered affected.

Investigation of arrhythmogenic cardiomyopathy has demonstrated ventricular premature complexes in Boxers as young as 6 months old and ventricular ectopy has been demonstrated to precede clinical signs by 3-4 years. In the author's experience, similar results have been noted for Boxers and Engligh Bulldog screened for arrhythmogenic cardiomyopathy. The presence of less than 50 ventricular ectopies in 24 hour period rule out arrhythmogenic cardiomyopathy, border line for diagnosis are case in which ventricular ectopies are between 51 and 300, while dogs with more than 300 ventricular ectopies are considered affected. Holter monitoring has also been instrumental in characterizing a syndrome of sudden death in young German shepherds.

Holter monitoring has also been demonstrated to aid in the recognition of serious arrhythmias in dogs with non-cardiac disease. In a case series of 50 dogs with splenic disease a high prevalence (22 of 50) of rapid ventricular tachycardia was documented with Holter monitoring. Routine electrocardiography was normal in 29% of dogs with rapid ventricular tachycardia at > 3,000 ventricular ectopies (VE) per hour; 50% of dogs with rapid ventricular tachycardia at 1,000 to 3,000 VE/hr and 100% (6 of 6) of dogs with 10 to 300 VE/hr without rapid ventricular tachycardia. Periodic rhythm strips are therefore unreliable for detection of ventricular arrhythmias even when frequent ventricular arrhythmia were present.

Holter monitoring is also capable of quantifying supraventricular (SV) arrhythmias. Adjustment of prematurity indices minimum and supraventricular tachycardia rate are warranted for each exam analyzed. Atrial fibrillation is detected by most Holter systems today, although aberrant beats can be classified as ventricular ectopies. Parameters of supraventricular arrhythmia include total number of SV complexes, couplets and runs of SV tachycardia. Holter monitoring is an ideal means of assessing efficacy of antiarrhythmic therapy. This may involve determining the reduction of supraventricular or ventricular ectopy or the effect of ventricular response rate in cases with atrial fibrillation. In general, a 70% reduction in ectopy is required to distinguish normal variability from drug effect.

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Cardiac emergencies

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Introduction

Most common cardiac emergencies in dogs and cats include congestive heart failure (HF), cardiac tamponade, arrhythmogenic disease, and thromboembolic disease. Many of these diseases are life threatening and urgent treatment is needed to provide relief of symptoms and to avoid cardiac-related deaths.

Heart failure

Most dogs and cats presenting with HF are in severe respiratory distress, due to alveolar pulmonary oedema as final sequel of degenerative mitral valve disease (MVD), dilated cardiomyopathy (DCM) in dogs and hypertrophic cardiomyopathy (HCM) in cats. More rarely, congenital heart disease, heartworm and lungworm disease, or other conditions leading to pulmonary hypertension. The suspicious of HF is based on the analysis of historical, clinical and instrumental findings. The confirmation of the diagnosis of HF is also based on the furosemide-responsiveness.

Dogs and cats with HF typically exhibit cough (prevalently dog), dyspnea, lethargy, syncope or episodic weakness, abdominal distention (prevalently dog), and/or partial to complete anorexia. Precipitating factor of congestive HF, especially in cats include: corticosteroid or intravenous (IV) fluid administration, new onset tachyarrhythmia, chordae tendinae rupture, left atrial rupture, concurrent systemic disease, or a stressful event.

A careful physical examination can suggest the presence of the underlining disease: loud systolic murmurs and tachycardia in dogs with MVD, soft or barely audible systolic murmurs and tachyarrhythmias in dogs with DCM, presence of atrial fibrillation either in dogs and in cats is very suspicious of HF.

Thoracic radiography and echocardiography are usually helpful to confirm the diagnosis of HF. In the ECC department the use of focused echocardiography and thoracic ultrasound had improved timing and accurate diagnosis of either left or right HF. Focused echocardiogram are performed with patients in sternal recumbency while receiving oxygen supplementation typically after a low dose of sedation. The examination is brief and minimally stressful, less stressful than performing a thoracic radiograph. Focused echocardiography can be performed also by noncardiologist/nonradiologist veterinarians, which can easily identify pleural and pericardial effusions and left atrial enlargement. The left atrium is most commonly indexed to the aorta in a right short axis parasternal view at the base of the heart. A left atrium/aorta ratio of 2.0 or greater is very suggestive of HF (normal values: < 1.5 in cats and < 1.3 in dogs). In case echocardiography remains not available, thoracic radiography is still one of the highest-yield tests for the diagnosis of left and right HF. In most cases, the presence of venous distention, cardiomegaly (VHS more than 8.5 in cats and more than 10.5 in dogs), and pulmonary alveolar or interstitial pattern are suspicious of HF. In dogs, pulmonary oedema is typically a perihilar interstitial to alveolar pattern. The pulmonary pattern is commonly asymmetric in dogs

with MVD with an eccentric jet of mitral valve regurgitation or more symmetric in canine DCM. The distribution of radiographic pulmonary patterns is variable in cats with cardiogenic pulmonary oedema. In these patients concomitant pleural effusion and lung volume increase are visible.

Treatment of heart failure

The immediate goal of emergency therapy is to reduce abnormal fluid accumulations and to provide adequate or improved cardiac output. Cage rest and minimization of stress are mandatory and obtained using sedation, oxygen, furosemide, or thoracocentesis in patients with large pleural effusion. Sedation is generally administered if patients are dyspneic and anxious. Butorphanol is a very effective sedative for patients in respiratory distress (0.05 to 0.3 mg/kg IV, intramuscularly (IM), or 0.1 mg/kg (SQ). Oxygen therapy delivered by oxygen cage or nasal catheter is recommended to reduce the work of breathing. Furosemide, a loop diuretic, is the main drug to manage congestive HF using IV repetitive bolus doses or CRI. The initial dose of 1 mg/kg IV every hour untill the respiratory rate drops below 40 breaths per minutes is usually chosen. As alternative treatment option, a CRI of furosemide at 0,5 mg/kg/hour can be used.

In addition to furosemide, in patients severely dyspneic, nitroglycerin or nitroprusside are recommended due to their vasodilator actions. Nitroglycerin, a venodilator, is commonly used in a transdermal formulation, while nitroprusside, at a variable dose, starting at from 1 or 2 ug/kg/min in dogs and even lower in cats (0.5 mcg/kg/min) and titrating up based on blood pressure (mean of 70 mm Hg or systolic 90 to 100 mm Hg).

Amlodipine, a calcium channel blocker, can also be use in this setting since it acts primarily as an arterial vasodilator. Amlodipine can also be used in patients with refractory or recurrent congestive HF, especially in dogs with severe refractory HF caused by MVD.

The most important recent advancement in HF management is the addition of pimobendan (0,25 mg/kg IV), a calcium-sensitizing drug that improves contractility with minimal effects on myocardial oxygen consumption and a phosphodiesterase inhibitor, primarily leading to a balanced vasodilation. Although pimobendan is not licensed for cats is being used more frequently in the management of feline acute HF. Pimobendan at 0,15 mg/kg bid OS is typically added when a cat with HF has left ventricular systolic dysfunction identified echocardiographically, significant pleural effusion, renal insufficiency, or severe refractory pulmonary oedema. If patients with HF are unable to take oral medications and have signs of severe low cardiac output HF, dobutamine can be recommended (1 to 10 mcg/kg/min CRI). If large-volume pleural or abdominal effusion is present and causing patient discomfort, the most effective therapeutic option is the removal of the fluid by ultrasound guided centesis.

The treatment of the first episode of acutely decompensated HF is usually successful. A recent study showed an estimated 80% survival rate to discharge for dogs with acute HF. Once the diagnosis and initial urgent management of HF has been performed, a plan for continuing management and monitoring should be formulated usually using the triple therapy (furosemide 1-2 mg/kg bid os, ACE-I 0,5 mg/kg bid os, Pimobendan 0,25 mg/kg bid os). The importance to execute a prompt and efficacious acute treatment is important since survival times for most dogs or cats in HF with treatment vary from 6 months to 1 year, depending on the underlying cause of the heart disease and comorbidities.

Arrhythmias

Arrhythmias can occur with a wide range of cardiac and noncardiac diseases and the clinical picture varies greatly

from asymptomatic to syncopal episode, depending on the rate, type of arrhythmias, and the severity underlining cardiac disorder. Tachyarrhythmias can be suspected on physical examination in patients with signs of cardiogenic shock and on cardiac auscultation because of rapid tachycardia, generally at heart rates above 300 beats per minute. An electrocardiogram is necessary to further characterize the tachycardia type: sinus tachycardia, ventricular tachycardia (VT), supraventricular tachycardia (SVT). Supraventricular and ventricular tachycardias can be congenital and sustained inducing arrhythmic induced cardiomyopathy or can be secondary to cardiomyopathies in particular DCM, arrhythmogenic cardiomyopathy and myocarditis. Another very common electrical rhythm disturbance that can be seen in critically ill patients is accelerated idioventricular rhythm (AIVR), an ectopic ventricular rhythm with a rate of 66 to 180 beats per minute. AIVR is usually present for extracardiac causes, but rarely can be present in case of complete atrioventricular block or acute coronary syndrome. Bradyarrhythmias are identified with cardiac auscultation because of either a regularly slow heart rate or intermittent periods of sinus arrest. Bradycardia is generally defined as a heart rate of less than 60 beats per minute in dogs and less than 140 beats per minute in cats. An electrocardiogram is necessary to further characterize the bradycardia into sinus bradycardia, a sinus node dysfunction, atrial standstill, or AV block.

Electrocardiographic analysis is the cornerstone to define the type of tachycardia. Usually SVT are narrow QRS complex tachycardia (QRS duration < 70 ms) while VT are wide QRS complex tachycardia (QRS duration > 80 ms). Ventricular tachycardias usually present atrioventricular dissociation.

Treatment of cardiac arrhythmias

The need and urgency of treatment of SVT or VT depends on the hemodynamic status of patients, the rate of the arrhythmia, and underlying myocardial dysfunction. Most ventricular arrhythmias do not require specific treatment and benefit from supportive care (supplemental oxygen, fluid/electrolyte therapy, and treating the underlying condition). Some ventricular arrhythmias may instead require acute treatment with lidocaine, procainamide, amiodarone or sotalol or electrical cardioversion can be performed.

Lidocaine is the active principle more effective for the acute treatment of ventricular tachycardias with life-threatening or unstable. Start treatment with an intravenous bolus at a dose of 2.2 mg / kg to be carried out in 5 minutes under electrocardiographic monitoring. Repeat as needed to the same bolus dose with 5 minutes intervals up to a maximum dose of 8.8 mg / kg. In order to maintain adequate blood levels it is necessary to continue with a constant intravenous infusion. Since the lidocaine in continuous infusion requires 3-6 hours to achieve adequate plasma concentrations, it is possible that it is necessary to repeat bolus administration. The dosage of lidocaine in continuous infusion is 25-80 mg / kg / min.

Procainamide is the drug of second choice to use in case of refractory ventricular tachycardias to treatment with lidocaine. Procainamide is preferable to quinidine for its lower gastrointestinal effects, does not prolong the QT interval, it does not interact with digoxin and not cause hypotensive states. The dose of administration corresponds to 10-15 mg / kg bolus over 1-2 minutes. Following the administration should continue by continuous infusion at a dose of 25-50 mg / kg / min.

Nexterone administered intravenously for the treatment of ventricular tachycardia has recently been incorporated into clinical practice. The electrophysiological effects of of nextorerone are very similar to the amiodarone's ones. Nexterone administered intravenously has proven effective in ventricular tachycardias and atrial fibrillation the dose of 2 mg/kg IV bolus in 10 minutes followed by 0.8 mg/kg/hr for the first 6 hours and then 0.4 mg/kg/hr for the following 18 hours. The sotalol is administered intravenously in human medicine in a dosage of 0.3 mg / kg. In veterinary medicine we have been performed only preliminary studies on the administration of sotalol intravenously.

After acute control of ventricular arrhythmias, chronic treatment is needed to prevent recurrence. The most common drugs used alone or in association are: Mexiletine, amiodarone or sotalol.

Some clinicians use mexiletine as the drug of choice for chronic treatment of ventricular tachycardia. The electrophysiological properties are comparable to those of lidocaine. Mexiletine also has action of myocardial depression less than other antiarrhythmic drugs. To date, the treatment of malignant ventricular tachycardias is represented by the association between mexiletine (4-8 mg / kg PO q8h) and atenolol (0.5 mg / kg PO q12-24h).

In veterinary medicine there are still clinical trials defining the effectiveness of these Class III antiarrhythmic agents. The results available to date provide controversial results probably related to the different pathologies treated, the different doses used and an incomplete follow-up of patients. Experimentally, sotalol is effective in the inhibition of ventricular tachycardias catecholamine-dependent. In human medicine, amiodarone and sotalol are considered first-line drugs for the treatment of ventricular tachycardia. In the author experience most reentrant ventricular arrhythmia responded acutely to sotalol 2 – 3 mg/kg bid os.

The management of SVT is importantly different than a ventricular arrhythmia. SVTs are usually narrow QRS complex tachycardias that include atrial fibrillation, atrial flutter, atrial tachycardia, reentrant accessory pathway tachycardias. Atrial fibrillation is typically a sustained tachyarrhythmia, whereas regular SVTs are typically intermittent. In the emergency setting, most dogs and cats presenting with atrial fibrillation are in congestive HF and are typically managed with a combination of digoxin and diltiazem. The combination of these two drugs is superior to either drug alone at reducing the ventricular response rate of atrial fibrillation, which is the goal of therapy. Treatment of other supraventricular arrhythmias depends on the rate of the rhythm disturbance and the presence of any underlying myocardial dysfunction. A vagal maneuver (eg, ocular or carotid sinus massage) or chest thump may be successful in transiently breaking an SVT. However, medical treatment will usually be necessary to chronically control the SVT. For acute management of sustained, symptomatic SVT, an oral loading dose of 2-3 mg/kg orally is usually effective in breaking SVTs within 3 hours.

Most animals with clinically significant bradyarrhythmias present for syncope or weakness. When sinus bradycardia occurs with excessively long pauses, sinus node dysfunction is suspected. Sinus node dysfunction is an idiopathic dysfunction of the sinus node and commonly affects miniature schnauzers and West Highland white terriers. Most dogs with significant sinus node dysfunction disease will eventually require a pacemaker to resolve their symptoms. Most dogs with advanced or third degree AV block have either primary conduction system disorders or acute myocarditis. Most dogs present for syncope, weakness, and lethargy and dogs affected are at risk of sudden cardiac death. Based on a recent study, 24% will die within 1 month of presentation and 40% within 6 months. Cats with complete or third-degree AV block do not carry a similar poor prognosis since the median survival time is about 1 year. In most dogs with advanced or third degree AV block pacemaker implantation is recommended. Rarely acute atrioventricular block can require external pacing therapy before the implantation of permanent pacemakers.

Thromboembolic disorders

Aortic thromboembolism (ATE) is one of the most common complications associated with feline heart disease, mostly cardiomyopathies (in particular restrictive cardiomyopathy). The most common site of embolization is the aortic trifurcation. The diagnosis is based on clinical findings of acute onset paresis and pain associated with absent or diminished femoral pulses, cool extremities and lost of bleeding. Less commonly, a right front leg is affected. Tachypnea or respiratory distress is often present because almost 2/3 of cats present concurrent HF. At auscultation a murmur, gallop sound, and arrhythmia are very common. In two large retrospective clinical studies, approximately

half of cats were euthanized or died during the initial thromboembolic episode, while prognosis is usually better with partial or front leg embolism. About 15% of cases will have some degree of permanent musculoskeletal deficiency. If a cat survives an episode of ATE, expected long-term survival varies between a few months to one year.

Treatment of arterial thromboembolism

Acute management of ATE includes the use of analgesics, anticoagulants, antithrombotics, and treatment directed to manage HF, when present. Fentanyl or buprenorphine, are used as analgesics. In addition to heparin, an antiplatelet therapy, either aspirin or clopidogrel, is usually started. Clopidogrel, an ADP receptor inhibitor that decreases platelet aggregation, is most commonly used. Alternatively, new experts suggestion include the concomitant use of aspirin and clopidogrel. At some referral institutions, thrombolytic agents, such as tissue plasminogen activator, t-PA, are also used. Initially, these cats may have difficulty to urinate and may need to have their bladders compressed to prevent overdistention. Electrocardiographic monitoring is recommended because of possible hyperkalemic arrhythmias cause by ischemia-reperfusion injury. For chronic anticoagulation, combination therapy or monotherapy with aspirin and clopidogrel are recommended. Re-embolization is a common terminal sequel in cats with ATE.

Cardiac tamponade

Cardiac tamponade is a pathologic condition in which the pericardial effusion causes an increase of intrapericardial pressure resulting in impaired ventricular filling and drop in cardiac output. The most common cause of pericardial effusion in older dogs is neoplasia (atrial hemangiosarcoma and chemodectomas). Dogs with hemangiosarcoma have a very poor prognosis. Others common causes of cardiac tamponade is idiopathic pericarditis and left atrial rupture in dogs with MVD, while the most common cause of pericardial effusion in cats is HF.

Clinical signs typical of cardiac tamponade are syncope, lethargy, exercise intolerance, cough, polypnea, abdominal distention, and inappetence. Physical examination is characterized by muffled heart sounds and tachycardia with weak and paradoxus pulses. This type of pulse is recognized when an abnormally large decline in systemic arterial pressure is associated with inspiration. Other physical examination findings include slow capillary refill time, pale mucous membranes, jugular venous distention, ascites, and hepatosplenomegaly. Echocardiography is the most sensitive and specific modality used to diagnose pericardial effusion and cardiac tamponade. The echocardiogram allow to visualize the presence and the amount of pericardial fluid, the thickness of the pericardium, and presence of a cardiac tumor and the presence of cardiac tamponade with late diastolic collapse of right atrial wall and early systolic collapse of right ventricular free wall. The electrocardiographic abnormalities are: sinus tachycardia, low amplitude QRS with alternating QRS-T complex. At thoracic radiographs a classic globoid-shaped heart may be seen with a large amount pericardial effusion with distended caudal vena cava, pleural effusion, and a sometimes a mass lesion at the heart base or cranial thorax.

Treatment of cardiac tamponade

Pericardiocentesis is the only effective treatment of immediate treatment of cardiac tamponade. Placement of a peripheral IV catheter and administration of fluids to increase the cardiac output is suggested before performing pericardiocentesis. Mild sedation is sometimes needed. Patients are placed in left lateral recumbency. The pericardiocentesis site is prepared aseptically. Ultrasound is helpful to find the best site for the centesis. A modified seldinger technique with a 7F catheter is used to advance the introducer within the pericardial sac. A 14g needle is advanced in the pericardial cavity under echocardiographic guidance, than along the needle a guide-wire is placed and over

the wire the introducer is advanced. Once in the pericardial space an extension tubing, a 3-way stopcock and 60-mL syringe are attached, and drainage of the fluid started. Complications of pericardiocentesis include death resulting from a lethal arrhythmia or coronary artery laceration, transient arrhythmias, pneumothorax, or intracardiac puncture

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Edward Hall

Diet, antibiotics or steroids?

How to manage Inflammatory Bowel Disease (IBD)

Idiopathic inflammatory bowel disease (IBD) is a collective term used to describe disorders that are associated with persistent or recurrent gastrointestinal signs and characterised by histological evidence of intestinal inflammation. By definition idiopathic IBD is of unknown cause: but only after strict diagnostic criteria have been applied to exclude all known causes of intestinal inflammation, can it be deemed truly idiopathic in an individual case.

IBD is probably the most common cause of chronic vomiting and/or diarrhoea in dogs. In cats, vomiting without diarrhoea is considered the more common manifestation. Signs are variable and may wax and wane.

An individual patient may show some or all of the following signs depending on the site, severity and type of inflammation:

- vomiting
- haematemesis
- SI-type diarrhoea: large volume, watery, melaena
- LI-type diarrhoea: haematochezia, mucus, frequency and tenesmus
- abdominal discomfort / pain
- excessive borborygmi and flatus
- weight loss
- altered appetite: polyphagia or decreased appetite / anorexia
- hypoproteinaemia / ascites (protein-losing enteropathy)

Aetiology

A variety of histological types of IBD are recognised by pathologists:

- Lymphocytic-plasmacytic enteritis (LPE) or colitis (LPC)
- Eosinophilic (gastritis)-enteritis-(colitis) (EGE)

The most common form is LPE, but unfortunately none of these descriptions give the clinician any clue as to aetiology. It seems likely that predisposition to IBD is genetically determined, just as the immune response is, and there is probably some underlying immune defect, although experimental models of intestinal inflammation indicate that luminal antigens (i.e. commensal bacteria) drive the inflammation after loss of normal tolerance.

A study of steroid-responsive diarrhoea in dogs (idiopathic IBD) demonstrated a marked inflammatory response dominated by IgG expression and up-regulation of the expression of pro-inflammatory cytokine mRNA. Immunohistochemistry of biopsies from these patients showed increased CD4+ T cells and IgG plasma cells in lamina propria. Alterations in leukocyte subsets have also been demonstrated in LPC and HUC. Thus immunohistochemistry and molecular techniques have enhanced our ability to detect specific changes when routine histology merely shows inflammation.

Diagnosis

Ultimately, IBD is a histological diagnosis, but before intestinal biopsy is undertaken, laboratory tests and imaging examinations are performed. They are intended to rule out the known causes of intestinal inflammation. Endoscopy is the safest method of biopsy but has limitations, and in some cases exploratory laparotomy and full thickness biopsy is preferred, particularly to distinguish IBD from lymphoma and lymphangiectasia.

The problem

Historically, canine inflammatory bowel disease (IBD) was simply defined by histological evidence of mucosal inflammation on intestinal biopsy; lymphoplasmacytic and eosinophilic enteritis being considered cytological variants of the inflammation. Then, with the advent of flexible endoscopy and the ease by which intestinal biopsies could be obtained, the frequency by which a diagnosis of IBD was made increased greatly; indeed it is repeatedly stated that "IBD is the most common cause of canine chronic diarrhoea". However, it has become apparent that not only is the correct identification of intestinal inflammation problematic (especially with endoscopic duodenal biopsies), but also the reasons for the differences between the cytological types is uncertain and that, ultimately, intestinal inflammation is merely a final common pathological pathway that potentially can be caused by many things.

A diagnosis of 'intestinal inflammation' in dogs with diarrhoea is of no greater value than a diagnosis of 'dermatitis' in pruritic dogs; not all of them should be treated with steroids! The known, potential causes of intestinal inflammation (i.e. infection and food allergy) need to be excluded before a diagnosis of idiopathic IBD can be made. Extensive laboratory testing and imaging should be undertaken, just as skin scrapes, allergy testing etc are undertaken for pruritus. Yet no method is able to rule out all the known causes of intestinal inflammation completely, and so in order to help rule out infectious and food related causes of enteritis, patients often undergo empirical treatment trials; just as a selamectin trial may be used to rule in or rule out a diagnosis of sarcoptic mange. A positive response to antiparasiticides (e.g. fenbendazole), antibiotics or exclusion diets has been assumed to reflect a diagnosis of either occult parasitism, or antibiotic-responsive diarrhoea (i.e. a specific infection or the small intestinal bacterial overgrowth syndrome), or dietary sensitivity respectively. Idiopathic IBD has then been assumed to reflect steroid-responsive intestinal inflammation assuming the case has already failed the other empirical therapeutic trials.

Yet one potential flaw in reaching a diagnosis by sequential empirical trials is that the order in which treatments are trialled, e.g. fenbendazole – diet – antibiotics – steroids, may bias the outcome. It is recognized that certain antibacterials (e.g. metronidazole, oxytetracycline) have an anti-inflammatory as well as antibacterial effect, so is 'antibiotic-responsive diarrhoea' actually a mild form of IBD?

Confusingly, intestinal biopsies collected after exclusion diet trials have shown apparent persistence of intestinal inflammation despite clinical improvement. This outcome has been seen more frequently since the introduction of hydrolysed diets, with some clinicians claiming a positive response to diet alone in a majority of cases that typically would have been treated with steroids in the past. What is not yet clear is whether the persistence of histological changes despite clinical resolution is evidence that dietary antigens are a significant cause of intestinal inflammation or that histological remission simply requires a longer time period than the duration of studies conducted so far, or even whether hydrolysed diets merely provide an inflamed intestine with a more digestible diet that the diseased gut can process. So do cases that respond to a hydrolysed diet inevitably relapse when the diet is stopped? And even if hydrolysed diets are effective, is that because of changes in antigenic stimulation, or through modulation of the intestinal flora? If the latter, would antibiotics or probiotics be as effective?

Attempts to dissect the inflammatory response by examining biopsies by immunohistochemistry and by PCR for cytokine expression have so far only yielded conflicting results. Thus the distinctions between intestinal inflammatory conditions that respond to antibiotics and/or diet and/or steroids have become blurred, and so the term 'chronic enteropathy' is often now preferred. Although use of a broader term than 'idiopathic IBD' is probably more correct

since it seems likely that it encompasses a number of conditions, it does not give the practitioner guidance of how to diagnose and manage cases presented to them.

A practical approach

The rationale for the order of empirical treatment trials (i.e. fenbendazole – diet – antibiotics – steroids) was to use each modality in the order that was least likely to cause any harm to the patient; steroids was inevitably the last trial. This approach remains logical, and indeed many cases will respond before steroids are needed. However, empirical treatment trials remain a scientifically unsatisfactory although pragmatic approach.

More rewarding, assuming the dog's owner has the patience and finances, is to undertake a staged, investigative approach to try and reach a definitive diagnosis and treatment by:

1. Rule out non-GI disease as a cause of the clinical signs through the history, physical examination and a minimum laboratory database. This will help rule out diseases in other organ systems, including hypoadrenocorticism. An ACTH stimulation test should be performed if hypoadrenocorticism is considered possible.
2. Assuming primary GI disease, evaluate its effect on the minimum database; this has both some diagnostic and some prognostic value. There are no pathognomonic changes, but anaemia may indicate bleeding, panhypoproteinaemia may indicate a protein-losing enteropathy and a worse prognosis, and hypocholesterolaemia can indicate malabsorption. An inflammatory WBC response is not often seen in intestinal inflammation nor, if present, can it predict the type of inflammation, e.g. an eosinophilia can be seen with lymphoplasmacytic enteritis.
3. Evaluate pancreatic function by testing serum TLI, as pancreatic insufficiency clinically mimics intestinal causes of malabsorption.
4. Perform zinc sulphate faecal flotation to look for Giardia and endoparasitic ova, although empirical fenbendazole treatment may still be undertaken in case of occult parasitism. The value of stool culture is questionable; it is not likely to identify specific genotypes that can cause chronic disease, and routine microbiological techniques will not distinguish between Campylobacter jejuni and the more common and likely commensal isolate of C. upsaliensis.
5. Measure serum folate and cobalamin concentrations, both as markers of disease, but also as indicators for the need for supplementation.
6. Image the abdomen; radiographs and ultrasonography are complementary. Plain radiographs should identify larger masses, radio-dense foreign bodies, intestinal obstruction and displacement; contrast radiographs are of limited value, especially if ultrasonography is performed. Ultrasonography can identify radiolucent foreign bodies, and allows examination of the intestinal wall structure with measurement of intestinal wall thickness, which may be increased in neoplasia. Loss of layering is also indicative of neoplasia, whereas mucosal striations suggest lymphatic dilatation.
7. Perform intestinal biopsy either by endoscopy or laparotomy depending on where the lesion may be and what is suspected. This is to rule out non-inflammatory diseases.
8. If intestinal inflammation is diagnosed, then perform empirical trials as discussed.

A pragmatic approach

It can be argued that the empirical approach is acceptable if there is no suspicion of neoplastic disease, especially if diet is so often successful. Indeed it could be argued that a hydrolysed diet trial should always be performed before intestinal biopsy (assuming the patient is still eating), as a finding of intestinal inflammation still behoves the clinician to undertake a diet trial. Only if the disease is very severe are steroids the first choice, and even then they are usually combined with fenbendazole, diet and antibiotics with the hope that they can be subsequently withdrawn.

Furthermore, the identification of intestinal inflammation on biopsy is problematic; duodenal endoscopic biopsy is an insensitive method for a number of reasons: poor technique in collection, processing or interpretation, or sampling of

the wrong site; surgical biopsies may be more sensitive but carry significant risk of septic peritonitis. The WSAVA GI Standardization Group has tried to improve the quality of endoscopic biopsy technique and the reporting of intestinal inflammation, but problems persist. Furthermore, discordance between duodenal and ileal biopsies suggest the histological diagnosis can be missed. Thus it can be argued that even empirical steroids (and perhaps ciclosporin) are justified before biopsy, although empirical use of cytotoxic agents is probably too dangerous to contemplate. The risk is that neoplastic or surgical disease is overlooked, or that the patient has an unidentified infection or does not have inflammatory intestinal disease.

Treatment

The mainstay of treatment of idiopathic IBD has always been immunosuppression. Despite the concept that IBD is a loss of immunological tolerance to the normal intestinal flora, antibiotics are not effective alone. Furthermore, there may also be lack of tolerance to dietary antigens. Highly digestible, restricted fat 'intestinal' diets are helpful in the management of IBD. They provide less 'work' for the compromised intestine, and hopefully contain the optimum amounts of n3:n6 fatty acids, fibre and micronutrients etc. necessary for general intestinal health.

Corticosteroids

Prednisolone (and methylprednisolone) remains the first-choice immunosuppressive agent in dogs and cats. Dexamethasone has similar immunosuppressive effects, but deleterious effects on brush border enzyme activity and is not recommended. The aim of therapy is always to find the minimum effective dose.

Novel steroids

The general suppression of the HPA axis by prednisolone, and its side-effects have led to attempts to find safer steroids. Recently an enteric-coated formulation of budesonide has been developed for human IBD (Crohn's disease). The enteric coating protects it until it is released in the distal SI, and it is then almost completely degraded first-pass through the liver. In man it has 10% of the systemic HPA-axis suppression of equipotent doses of prednisolone. Budesonide has been given to dogs and cats with IBD with anecdotal success. However, the dosage has been extrapolated from man: steroid hepatopathy still seems to be a problem, and systemic effects can occur.

Azathioprine

It is an immunosuppressive agent, but its use is not as a first-line agent, but as a steroid-sparing drug in IBD patients suffering iatrogenic Cushings. As it takes several weeks to be fully effective it should probably be started at the same time as steroids in severe cases of IBD, and should not be withdrawn prematurely or relapse may occur.
Bone marrow toxicity is quite rare in dogs, but will occur quite rapidly (1-2 weeks) in some individuals. They probably lack the enzyme necessary to degrade 6 mercaptopurine, its active metabolite. This enzyme is commonly lacking in cats, which is why azathioprine cannot be recommended for cats.

Chlorambucil

In feline IBD, another immunosuppressive such as chlorambucil is a better choice, and may be better for dogs.

Cyclosporin (Ciclosporin)

Well known for its efficacy in keratoconjunctivitis sicca, this anti-rejection drug has been used to treat anal furunculosis successfully. Preliminary studies in idiopathic canine IBD have shown variable success.

5-Aminosalicylic acid (5-ASA) derivatives

Although many cases of colitis in dogs are secondary to SI disease or part of generalised IBD, isolated chronic colitis may occur. In such cases the use of sulphasalazine is appropriate. Within the drug a diazo bond binding a sulphamoiety to 5-ASA is cleaved by colonic bacteria, releasing free 5-ASA. This acts locally in high concentrations as an anti-inflammatory. Unfortunately its major side-effect, keratoconjunctivitis sicca, is well documented.

Kris Gommeren

Shock, clinical diagnosis and differentiation

*Although often used appropriately in the setting of emergency and critical care, 'shock' simultaneously is one of the most '**ab**used' words in emergency and critical care. This lecture will train you to go to the next level and properly define different types of shock.*

Shock is defined to a state during which tissue perfusion and/or oxygen delivery is insufficient to meet the cellular requirements. Unfortunately often veterinarians faced with emergencies rapidly jump to the conclusion that their patient is in shock without substantiating this claim, or taking a second to identify the underlying cause.

As perfusion is difficult to visualize in a clinical setting, arterial blood pressure is often used as a substitute for flow. A mean arterial blood pressure (MAP) below 60mmHg, or a systolic blood pressure (SAP) below 80mmHg is diagnosed as arterial hypotension and hence a state of shock. However, all animals with a blood pressure between 80 and 90mmHg should already be looked at carefully, as many would suspect these to be in shock.

A less frequently used, but possibly even more interesting marker of tissue perfusion are blood lactate concentrations. Lactate concentrations can increase due to anaerobic metabolism such as during physical exercise, local ischemia, or shock; but also due to decreased metabolism and clearance (hepatic or renal disease) or due to bacterial production. Hyperlactatemia due to anaerobic metabolism has been proven to be a more important prognostic marker than arterial blood pressure in canine critical care, therefore every general practitioner should ideally be able to assess these markers.

Classic types of shock are cardiac disease, hypovolemia, hypoxia or septic and anaphylactic shock. Regardless of the cause, the 2 main body responses are similar:

- Increased sympathetic tone: inducing peripheral vasoconstriction, tachycardia
- Activated RAAS system: water retention to increase vascular volume

This should result in pale mucous membranes, tachycardia, stronger pulses and shorter CRTs. In hypovolemic shock these mechanisms are ideal, however in cardiogenic shock they are inefficient (as cardiac energy consumption will increase), and in septic/anaphylactic shock these systems malfunction. The implications of these findings on treatment will be discussed in the next lecture.

Kris Gommeren

Shock, different causes, different treatment?

Patients in shock are in desperate need for rapid and appropriate support. Unfortunately different types of shock may benefit from other therapies. This lecture will help you to offer a more reflected and goal oriented approach to the initial stabilization of shock patients.

As explained in the previous lecture, there are different types of shock. The origin of the poor tissue oxygenation will impact the stabilization of the patient. In general, tissue oxygenation requires a decent respiratory function, cardiac function, hemoglobin content, volume status, vascular integrity and cellular function.

The different treatments that historically have been recommended for the stabilization of shock patients are consequently not indicated for all types of shock.

- Oxygen: administration of up to 100% oxygen for up to 24 hours is considered harmless, and can always be administered to shock patients, although the highest benefit should be expected in patients with respiratory or congestive cardiac disease.
- Large volume isotonic crystalloids and colloids: will be most beneficial to hypovolemic and relatively hypovolemic (i.e. septic and anaphylactic) shock patients. However in cardiac function these should be avoided.
- Blood products are indicated for severely anemic patients, and should be administered as soon as possible. In hemorrhagic patients initial stabilization with large volume isotonics whilst awaiting blood products can be indicated.
- High dose corticosteroids are only indicated in anaphylactic shock patients, but are contra-indicated in all other scenarios. Corticosteroids at physiological dosages remain a subject of controversy, and should be reserved for specialty services.
- Vasopressors: should theoretically only be reserved for patients suffering from inappropriate vasodilation (i.e. septic and anaphylactic shock). The inappropriate administration of vasopressors to hypovolemic patients will increase arterial blood pressure, but not tissue perfusion, and this should therefore be avoided.
- Positive inotropes: cardiogenic shock patients might benefit from positive inotropes. However, their increased oxygen consumption requires further investigation.

Dani McVety

The art of euthanasia & the science of death

Lap of Love Veterinary Hospice

The euthanasia appointment is unparalleled in emotion and sentiment. There are few things in veterinary medicine, or life moreover, that require as many outward displays of empathy, compassion, and commiseration from a doctor. The tone of voice, delivery of words, bedside manner with both patient and client, and the ability to honor moments special to the family become a delicate dance around death that the doctor and staff should carefully choreograph and continually improve. The client expects their pet to be saved in an emergency situation but helping the family and pet feel comfortable, understood, and secure in their most vulnerable moment, the death of their friend, truly transforms the professionals into heroes.

Overview of Important Points

1. Be comfortable with your sedation protocol. Use it frequently and be ready to manipulate it when circumstances require.
2. Rehearse your sedation and euthanasia explanation over and over. Do it in front of a mirror, record yourself saying it, ask your staff for feedback. This is your first performance of the last appointment and your best opportunity to convey your care for both the people and their pets.
3. Always stay calm, you are the "rock" in the room, the one that has all the answers and guides the appointment. Do not react to a client's emotions; be empathetic and compassionately confident.
4. Honor the silence, do not try to fill it.
5. Never underestimate the power of physical touch. Physical touch says more about your genuine care and concern than your words ever will.
6. Remind the family they are making the right decision. Always.
7. Compliment the pet; be affectionate and caring towards him/her.
8. Know your answer to "how do you do this." Make it optimistic; "This is an honor; I'm privileged to be part of this memory with you."

Euthanasia is not only a necessary, essential, and permanent part of our job, but also an art form that requires immense personal focus, unparalleled empathy, and a unique form of compassion for families in varying situations. As the only health care profession with the authority to end life, the veterinary staff has the duty to ensure that our clients fully understand each step we are taking and why. Euthanasia is truly an art form in which the human aspect plays just as much, if not more, of a role as the medical and technical skills. We will be the ones to change the face of human medicine; we are the experts, we are the leaders, and we should be the best.

Dani McVety

Convenience euthanasia

Lap of Love Veterinary Hospice

When it comes to ethical-border-line euthanasia requests, we have a very important decision to make as veterinarians, but we need to ask the right questions from the start. Instead of deciding whether or not you are comfortable euthanizing that pet, the question should be "what are the alternatives for this pet." By requesting euthanasia in the first place, the family is communicating to you that the human animal bond is broken. We can either help change the situation for them (remove the pet from their care via adoption or euthanasia), or do nothing by sending them home because "I just can't do it." And in my opinion, doing nothing is professional suicide; you've now ruined any rapport you had with that family, a small loss that does not create societal trust and respect for our profession. Helping a family, in whatever way, is far preferable than sending them home with a broken human-animal bond. Remember, medicine is not our product in the veterinary world, the human-animal bond is. Without that bond, they are not coming into our clinics. When euthanasia is requested, the family is telling us that there's something wrong with that bond and they care enough to tell you about it instead of letting the dog or cat go on the side of the road.

So what should be done in these extreme cases of uncomfortable euthanasia requests? Allow me to push the boundaries a bit; in my opinion, we must take responsibility for the pet in some way. As a house call hospice veterinarian, if I am at a home of a pet that I do not feel comfortable euthanizing, and with an owner that simply cannot go on, the pet will come home with me. Yes, it's happened. And have I euthanized animals that I may not have euthanized if they were mine? Absolutely. Have I euthanized animals that other veterinarians have refused to euthanize? Absolutely. Have I euthanized animals whose owners were completely at a lost, unable to go on for many reasons, and with tears in everyone's eyes (including mine), we knew it was a difficult but good decision? Absolutely. And when those families hug me, knowing that I did not judge them for that tough choice we made together, that I did not force an altruistic or idealistic view on them, and that I partnered with them in opting for the best alternative option for their pet, a new level of respect is earned.

Euthanasia Definitions

- Convenience euthanasia is a very subjective term. We use this phrase when euthanasia is requested for a pet that would otherwise be deemed adoptable under most circumstances and the family is unwilling to explore these options. For example, "my pet doesn't match the decor in my home any more" (yes, I've heard this). Personally, I do not offer convenience euthanasia in my practice, we offer support and resources to re-home these pets.
- Non-medical euthanasia is a term I use when describing a request that is not related to the medical stability of the pet. This is a broad term that includes behavior issues (such as aggression or improper elimination in the home), in addition to emotional or lifestyle changes of the family that precludes the pet from experiencing a quality of life.
- Non-imminent medical euthanasia is a term that describes situations like the 12-year-old cat. These conditions may be manageable or even curable under the right circumstances, but for whatever reason, those circumstances do not exist. This includes the parvo puppy that may survive with intensive care, the 5-year-old

intact female with a pyometra, or the young cat with a broken leg. Without the right resources and conditions (which may be too expensive), this pet would potentially suffer greatly. Rarely will I turn down this type of euthanasia request.

- Medical euthanasia describes most of the euthanasias that occur in our clinics; a choice that is made when the quality of life of the pet is deemed unsustainable by both the family and the veterinarian.

Non-Medical & Convenience Euthanasia Rules

- Do not euthanize a pet that you do not feel comfortable euthanizing. Period. (But say "no" carefully, keeping these other rules in mind.)
- Always help the family explore alternative options and think about how those options will effect the family and the pet down the road. Remember that a shelter is the deadliest place for a pet to be. Write them down, discuss them, think about what effect those alternatives have on OTHER animals in society.
- If you are comfortable euthanizing, even if you don't completely agree, you must help the family understand that although this is difficult for you (and them), you care greatly for the pet and the greater good.
- Do not get involved in cases if you don't plan to help, you will do more harm to our profession by judging and berating clients that if you simply hand them a number to a different veterinarian (preferable), or at least the local shelter or rescue organization.

Kris Gommeren

FAST, the value of ultrasonography in emergency and critical care

The rapid development of ultrasonography has led to the development of different techniques allowing general practitioners and intensivists to gain valuable information on their emergency patients rapidly, efficiently and with limited risk to their patients. Come and join the FAST-team!.

Over the past decades the use of echography to rapidly obtain information in ECC patients has been developed in human medicine. Fortunately, the last few years publications on the use of echography in ECC are appearing, and this tool receives continuously growing attention by ECC-clinicians.

FAST or **F**ocused **A**ssessment with **S**onography for **T**riage (originally Trauma) describes these short echographic procedures. A(bdominal)-FAST describes the rapid search for free abdominal fluid and the screening of the integrity of the gallbladder and urinary bladder. The probe is placed at four different sites, and detected free fluid is aspirated and analyzed. This technique can be helpful for the early diagnosis of hemo- and chyloabdomens, septic, biliary and urinary peritonitis, modified transudates and pure transudates. Repeated A-FAST examinations allow to detect ongoing blood loss, indicating the need to intervene surgically. At the level of the abdomen, techniques have also been described to assess the bladder volume to (although not extremely accurately) assess urine production in critical patients. At our institution we also apply A-FAST to evaluate gastric volume and motility in critical gastrointestinal patients.

T(horacic)-FAST and VetBlue techniques (Figure 2 and 3) are used to gain rapid information on cardiorespiratory function. The cardiac views of the T-FAST consists of transverse cardiac views, visualizing pericardial effusion; ventricular systolic function, thickness of the ventricular walls; signs of endocardiosis or endocarditis of the mitral valves and; the left atrium to aortic ratio indicating the risk of congestive heart failure and pulmonary edema as well as the volume status of these patients.

The VetBlue technique focusses on the presence of a glide sign or B-lines at the level of the lungs. Glide signs are normal, and the lack thereof indicate that the pleural sheets are no longer in contact (in essence a pneumothorax). B-lines instead are vertical hyperechoic lines indicative of the presence of liquid rather than air in the alveoli. This liquid can be a sign of congestive heart failure, pulmonary bleeding, a pneumonia or any other type of liquid. Although the B-lines themselves do not give any additional information on the nature of the liquid, the distribution of the lesions throughout the pulmonary field can be highly informative (e.g. aspiration pneumonia in the ventral lobes, congestive heart failure in the perihilar region etcetera).

Kris Gommeren

Diabetic ketoacidosis, targeting your therapy to save lives

Diabetic ketoacidosis and hyperosmolar syndrome are often devastating diagnoses both for owners and veterinarians. This lecture will provide a structured approach to these patients and will discuss the most common pitfalls in general practice.

Diabetic ketoacidosis (DKA) and hyperosmolar syndromes are rare complicated forms of diabetes mellitus (DM). DKA occurs whenever the cells can no longer rely on glucose to provide the necessary energy, and subsequently fatty acids are transformed into ketones to provide this required energy. As only minimal amounts of insulin are required to provide minimal energy requirements, DKA is usually seen in undiagnosed DM, or poorly controlled DM patients experiencing an additional triggering event (such as pancreatitis or Cushing's disease). Hyperosmolar syndromes are more often encountered in cats, where the osmotic diuresis of DM patients can result in severe dehydration, further increasing the hyperglycemia and provoking severe dullness, anorexia and adypsia in these patients.

The diagnosis of both syndromes is pretty straightforward, and based on glycemia, plus basic chemistry panel and a urinalysis.

Contrary to what was initially presumed, the therapy of DKA patients however is not an 'aggressive', but rather a 'controlled' approach. Glycemia should only be slowly corrected (decreasing not any more than 50-100mg/dL/h), and this is initially already achieved by rehydrating the patient. Meanwhile, I strongly encourage every general practitioner not providing 24 hour care or lacking the necessary infusion pumps and in house analyzers to transfer such a patient to a nearby larger facility.

Although many subcutaneous protocols have been described, constant rate infusions (CRIs) are much easier to control the glycemia and administer the small doses of insulin. During these initial hours the clinician should also screen for any underlying disease explaining the transition into DKA, and regular blood work will be required to screen for hypokalemia and hypophosphatemia. With a standardized approach and decent monitoring, the majority of your patients will however recover, and the diagnosis of DKA by no means indicates that such feline patients cannot prove to be reversible DM patients in the end.

Dani McVety

Veterinary hospice care: comfort beyond a cure - parts 1 & 2

Lap of Love Veterinary Hospice

Although medicine may not be able to cure a pet's terminal disease or old age, we can certainly help the owner keep their pet comfortable, clean, and happy, which is important not only for the welfare of the pet, but for the human-animal bond. With increasing number of positive experiences families have with human hospice coupled with the ever-increasing status of our pets in society, clients are requesting this care for their aging or terminally ill companion animals. Knowing what hospice care is, how to provide it in the clinic or home, and how to assist families in the mitigation of suffering will ensure you provide top-quality care at this delicate time.

The American Veterinary Medical Association views veterinary hospice as care that will allow a terminally ill animal to live comfortably at home or in a facility, and does not believe that such care precludes euthanasia. We define veterinary hospice as: A family-centered, medically supervised, and team-oriented service dedicated to maintaining comfort and quality of life for the terminally ill pet until a natural death occurs or the family elects euthanasia. It is important to note that a natural death is not the goal for veterinary hospice, it is simply a reality for many terminally ill pets whether they are in hospice care or not. The main purpose is comfort of the pet before death, whether from natural death or euthanasia. This care can take place in the clinic or home: the home is often preferred because it is where pets are most comfortable. However, education and medical direction begins at the clinic. Using the word 'hospice' to describe this care will help families realize that their pets are at the end of their lives and unable to be cured. Many times just the use of this word is a relief to pet owners! This terminology also illustrates that comfort and quality of life are the most important goals for these pets.

Veterinary hospice care is a unique case approach centered around education, preparation, and support for the pet and the client. Understanding what this service includes and what it does not include will help veterinary professionals solidify their role as animal advocators, strengthen the human animal bond, and maintain the highest level of client care. In this two-part lecture, we will learn how to discuss and offer hospice services to clients with terminally ill or severely geriatric companion animals and gain an understanding of the ways to evaluate quality of life to best serve our patients and their families.

Animais de Companhia II

Patrick Bourdeau

Canine Leishmaniosis: How to diagnose it? From suspicion to monitoring

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The diagnostic approach for Leishmania infection in dogs has two main purposes: the diagnosis of the disease in a sick dog and the detection of infection in a clinically healthy dog.

Other indications are the post-treatment evaluation during follow up and inclusion criteria for vaccination of really healthy dogs.

I How to diagnose a dog sick from Leishmaniosis ?

Diagnostic approach

The diagnosis initially requires the combination of Epidemiological compatibility (origin, place of life, risk of exposure, other dogs...)

1. Presence of clinical signs obtained from a thorough physical examination (the list of suggestive clinical signs is very long). The disease is said "proteiform" and sometimes only one sign is observed. Research systemically for eye, or joint signs and detect any lymphadenomegaly.
2. Clinico-pathological abnormalities are virtually always present if clinical signs. Routine diagnostic tests are performed, if not initially at least after confirmation of infection as they will be necessary for evaluation of the case (staging) : complete blood count (CBC), biochemical profile, urinalysis. Serum electrophoresis is an useful tool it is important to perform it at the very beginning to use it to evaluate the evolution of d-the disease during follow up.
3. The hypothesis of CanGL will be compared to other potential diseases that could be responsible of similar signs. A combination may exist. In many areas where CanGL is present other vectorial diseases may be present as well (monocytic Ehrlichiosis, Babesiosis, Hepatozoonosis, Dirofilariosis...) also some immuno-mediated disease may mimic CanGL (i.e. Pemphigus foliceus, systemic Lupus erythematosus) or neoplasia (lymphomas).

Diagnostic techniques

Some techniques are direct: Parasitology (cytology, histology, immunochemistry and culture of the organism in appropriate medium), or molecular (different PCR).

Cytology:

The most rapid by observation of amastigotes free or intracellular from: skin smears (or snap), lymph nodes, bone marrow, spleen. (Leishmania can be observed in other damaged tissue but rarely used for an initial diagnosis. The specificity is theoretically 100% (expertise is necessary to avoid confusion with artefacts like nuclear fragments, stains and even bacteria) Lymph nodes cytology is the most frequently used. Rapid staining techniques are performing enough. As the number of amastigotes is highly variable a long examination is necessary before to conclude to absence of parasites.

Histology:

Leishmania organisms can be detected within suggestive histopathological patterns like macrophagic, or lymphoplasmacytic infiltrates or reactive hyperplasia in lymphoid organs. Such observation should be confirmed by quantitative serology (necessary for grading). Immuno-histo-chemical staining techniques allow a specific identification with a better sensitivity than routine histopathology.

Molecular tools:

The PCR techniques applied to diagnosis have considerably changed and new techniques are developing. The effectiveness of PCR is highly influenced by:

- nature of samples: PCR on blood or urine samples are less sensitive than using, skin, bone marrow, lymph node, spleen or conjunctival swabs. There are also some recent studies using nasal swabs, hairs and even recently ear cerumen swabs. The performance of these last approaches needs more information before to be recommended.
- number of samples : a few years ago diagnosis were based on only one samples. Today it is asked to combine several samples from different nature due to the frequent false negative if only one sample is tasted
- repetition of samples : successive blood samples may alternate negative or positive results..
- techniques : those targeting on kinetoplast DNA are more sensitive than conventional PCR; Real time qPCR can detect low levels and can quantify the parasitic load with application for a better interpretation for diagnosis and for the follow-up of sick dogs.

A negative result do not exclude CanGL. A positive result doesn't necessarily correlate with clinical status but indicates the presence of Leishmania.

Many serological methods (qualitative and quantitative antibody tests) ah eve been developed. Specific cellular immunity tests in vivo (intradermal-Skin-Testing with leishmanin) or mainly in vitro others may indirectly evaluate the immune response of the host and (mainly used for research).

Serology:

Quantitative serology is recommended for the most accurate diagnosis, conclusive diagnosis of clinical leishmaniosis, and staging of the dog. There are 2 main techniques used in most countries.

- Immuno) fluorescence antibody test (IFAT) remains the official test. The cut-off for positive diagnosis of the disease is generally 1/80 or 1/100. The majority of clinical cases of CanGL have much higher titres.
- Enzyme-linked immuno-sorbent assay (ELISA) have been usefully developed. The performance of these techniques may be variable and "Units" differ also wit laboratories (O.D; %, units).
- Other tests developed are Direct Agglutination tests, Latex tests and Western Blot.
- For serological diagnosis it is necessary to make follow-up of fogs with the same laboratory to avoid any technique-linked variation.
- Rapid serological tests (immuno-chromatography) are only qualitative. Their specificity is considered good but sensitivity may be variable. Clinical cases are generally positive but should be followed by a quantitative test (staging and prognosis, then follow up).
- In vivo: Leishmanin Intradermal skin testing (IDST) was developed and used in humans for decades. In dogs some studies were made, mainly for epidemiology studies. This test reveals a specific cellular response in the host (delayed hypersensitivity). Thus leishmanin test could be a very simple and useful test for the detection of Leishmania in infected but clinically healthy dogs (resistant profile). These dogs are frequently seronegative but protective cellular immunity makes the IDST positive. In humans with VL the cure is characterized by a positivity of IDST not present during the clinical phase. IDST could be an interesting tool to evaluate the response of the dog to therapy or vaccination. Unfortunately no commercial leishmanin is available.

Specificity of diagnostic tests:
The question of diagnosis of Canine Leishmaniosis is complicated by the presence in some countries other species of Leishmania that may infect dogs (clinically or not). This is mainly important in south America. In Mediterranean basin at least two species (apart L. infantum) are now known to infect dogs with clinical disease (L. major, L. tropica). In south America Trypanosoma cruzi is widely prevalent in several countries. These protozoa share common antigens (Trypanosomatidae, genus Leishmania) and a cross-reaction may occur. It is an important question in dogs travelling or re-homing from these areas. However the titres obtained by cross reaction are much lower than the homologous test. A double serology allows identify the really offending pathogen.

The specificity of PCR techniques is higher than other techniques if really developed on the species of Leishmania. Most of PCR are not distinguishing between Leishmania species and have to be combined to restriction fragment length polymorphism (RFLP) or even sequencing.

It is important to say that serology (IFAT) does not cross-react with Babesia, Ehrlichia or Dirofilaria (commonly encountered in countries endemic for Leishmaniosis and possibly co-infecting the dogs).

II How to detect Leishmania in a clinically healthy dog ?

The question of diagnosis on healthy dogs is frequent: travelling dog, blood donor, vaccination. The quite recent knowledge on the vertical transmission could also raise the question for dogs used for breeding. The diagnosis techniques are the same as for a sick dog but their use and interpretation differ. Globally all the techniques are much less sensitive and specific in the conclusions.

Serology:
The first test to use. In case of positive result (quantitative or qualitative) the dog should be evaluated like a sick dog. Quantitative serology may reveal titres below the cut-off (Qualitative tests are negative at these levels). At these values the specificity is imperfect. However any level of antibodies is suspect and most of these dogs are probably infected. A follow-up is necessary or the combination with multiple samples for PCR.

Molecular:
the same sampling have to be done as for diagnosis. The conclusion is valuable only in case of positive results. A negative PCR never exclude the possibility of Leishmania infection. In practice PCR is not recommended but mainly used to confirm a borderline serology.

Blood donors: will be tested by serology then if negative by PCR on Blood. These dogs have to be treated with insecticides and rechecked every 6 months.

Follow up of infected dogs.
Serology is likely the best way to follow the evolution and quantitative PCR optional. A decrease of 2 titres is significant. The decrease of antibodies can be rapid (within 3 to 6 months) but generally requires a longer period. Dogs with CanGL could be evaluated every 6 months.

It is important to remember that treatment do not eradicate Leishmania. Thus conventional PCR may remain positive even if the dog is clinically healthy. Only Quantitative PCR may be of interest for the follow up.

Needs for future
Despite the development of new techniques and recommendation for diagnosis and management many questions

remain. For instance the use of vaccines complicates the interpretation of serology based diagnostic tools. New diagnostic techniques for the discrimination between natural infection and vaccination induced antibodies should be developed. Other needs are:

- New techniques for a better pre-vaccination screening to avoid to vaccinate dogs already infected.
- A better knowledge on the characteristics (level, duration) of cross-reacting antibodies elicited by vaccination
- Cellular immunity tests to distinguish between responses to vaccination and evaluate the response to treatment of infected dogs to natural infection.
- Research on biological markers significant for accurate prognosis and both for the evaluation (prognosis) and the follow up (evolution) of the infection post treatment or post vaccination.

Information available on the Leishvet website : http://www.leishvet.org/
and on: https://parasitesandvectors.biomedcentral.com/articles/10.1186/1756-3305

Patrick Bourdeau

Management of dogs infected by Leishmania and prevention: From apparently healthy to sick dogs

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Leishmania infantum (Protozoa Trypanosomatidae) is responsible in dogs in Europe of a systemic infection. This infection may result in different situations in dogs from clinically healthy to a very severe disease called Canine Generalized Leishmaniosis (CanGL). The dogs participate to the reservoir of *Leishmania* with potential zoonotic consequences. In endemic, but also non-endemic, areas this results in different approaches to manage the evolution of infection in infected dogs or the protection of naïve non-infected dogs. These aspects will be presented and focused on individual measures.

I Leishmania in infected and sick dogs : the treatment

Canine Leishmaniosis is considered as a « disease » as soon as the dogs have clinical signs and or clinico-pathological signs caused by Leishmania organims. It is important to remember that many signs in CanGL are not specific: an infected dog can be also simultaneously infected by other agents (*Babesia, Ehrlichia….*). It is to verify in a Leishmania infected dog that the clinical signs are not due to these agents. The objectives of the treatment are the control of the disease, the decrease of the parasitic load, the control of the role of source of infected dogs for other animals or humans.

What is the current specific therapy?

It is based on the use of 3 molecules with scientific evidence efficacy. They can be combined in different protocols.

- Meglumine Antimoniate (Glucantime® Merial) and miltefosine (Milteforan® Virbac) are leishmanicidals. The first acts mainly on enzymes essential to the metabolism of fatty acids of the parasites, the second modify the cell membrane synthesis resulting in apoptosis of Leishmania and may be by modulating TCell activity toward a higher activity of macrophages (oxydative burst).
- Allopurinol, a hypoxanthin compound, is metabolized by *Leishmania* producing a dysfunction in RNA transcription and making impossible the multiplication of the parasite (Leishmaniostatic).

Many protocols have been proposed. The current recommended treatment is: one month of 1) Glucantime (75-100mg/kg once or 40-75mg/kg BID SC) or Mltefosin (2mg/kg/Day orally) 2) combined with allopurinol (10-15 mg/g BID orally). The choice of the therapeutic approach is modulated by the grading stage. Recommendations for the choice of treatment have been proposed by different groups i.e. Leishvet monitoring with 4 stages of the disease from I (mild) to IV (very severe). Relapses may occur and it is recommended to maintain allopurinol for at least the first 6 months (see below).

Clinical efficacy:

Grade I (treatment optional or allopurinol alone) and grade II have a good prognosis because generally respond well to treatment and the disease has no or limited irreversible effect. The evolution of grade III is more unpredictable and

grade IV has a very poor prognosis with irreversible lesions (immuno-pathological lesions, kidney lesions...).

Post treatment allopurinol is stopped when 1) clinical cure is obtained, 2) clinico-pathological abnormalities are corrected 3) ideally quantitative serology turns negative (<1/80 titer IFAT). In some dogs allopurinol has to be maintained for years.

Antiparasitic efficacy:

Clinical cure doesn't mean parasitological cure. The treatment may temporarily abolish infectiousness to sand flies then decreases the role of source of the dog. The dogs remains however infected.

Tolerance

Side effects of glucantime are common: pain at injection sites, anorexia, vomiting and diarrhoea. Nephrotoxicity has been confirmed experimentally even in healthy dogs. An increased and frequent control of kidney function is recommended if used in Grade III and even II.

Miltefosin may induce benign vomiting in more or less 10% of dogs (more rarely diarrhoea) without nephrotoxocity. Allopurinol is globally very well tolerated. However, logically, it can be responsible of the production of xanhinuria and urolithiasis (up to 15% of dogs?). Detection of cristalluria during the follow-up is recommended.

Other molecules?

Many molecules have shown activity on Leishmania in vitro, much less in laboratory animals (mice models), even less in humans, very few in dogs.

- Amphotericin B is active but not recommended. It has to be used intravenously and is highy toxic for kidneys. The most important is: its use should be kept to treat humans (visceral leishmaniasis).
- Pentamidin, buparvaquone, enrofloxacin, marbofloxacin, metronidazole, spiramycin, ketoconazole ....do not afford enough evidence of strong effects in sick dogs to be recommended as first line treatment. However metronidazole 25mg/kg/D + spiramycin 150 000 U/D 3months, or marbofloxacin 2mg/kg/D one month could be considered as a second choice as optional treaement in low grade I dogs.
- Aminosidin is known to be active on Leishmania. It is nephrotoxic at high doses but could be tolerated at doses less than 15mg/kg/D. More studies are needed.

Other aspects of management of infected sick dogs?

Non specific therapy?

CAnGL is a disease induced by the parasite but developed by the host. Most of the pathogenic effects are secondary to host response toward the parasite. The control of the parasite itself is not enough to manage the disease mainly in stages IV and III but even II.

The symptomatic treatment, sign the by sign, is needed. In this aspect the use of corticosteroids, theoretically prohibited for such a disease, can be useful (i.e. eye lesions). In most of dogs kidney lesions are a major concern. They have to be managed following IRIS recommendations.

It is still not clear if combined diseases are promoted by Leishmania : some associations are common, the most frequent being bacterial pyoderma that has to be detected and treated.

Follow up?

Follow up visits have to be organized in sick dogs; ideally 2-3 times/year (at least once). The clinical evaluation is completed (≥ 1/year) by quantitative serology, haematological parameters and biochemistry.

Ectoparasiticides?

Every dog diagnosed with Leishmania infection becomes also a recognized source for other dogs (or cats) and humans. The use of ectoparasiticidal drugs (in order to avoid the infection of vectors) is part of the therapeutic protocol of every infected dog. The protection should be permanent by using the same products as for prevention (see corresponding chapter)

Other aspects of management

It is now clearly established that non-vectorial transmission is not exceptional. Every infected dog should be discarded for life to be a blood donor, to be used for breeding (venereal transmission form males to females, vertical transmission during pregnancy).

Is there an indication for Immuno-modulation or immunotherapy?

Another approach is to act on immune status of infected but healthy animals (no clinical signs, no clinico-pathological signs) to produce/maintain a resistant profile (Th1 targeted). The question is also the use for treatment in sick dogs.

Non specific:

Domperidone has been shown to stimulate a Th1 cell response, favourable to resistance to infection. One study indicates efficacy (1mg/kg BID orally one month). After 3 months a clinical improvement was seen in most dogs with, after one year, a reduction of antibody titres in 75% of those with few clinical signs initially, and one third of those with initial severe disease. Another study suggests a preventive effect (reduction factor 7 of CanGL in treated dogs). The molecule is well known in human medicine (Motilium®) and has been launched in veterinary medicine in some Mediterranean countries (Leishgard® 0,5 mg/kg/day one month every 4 months) both for prevention or therapy. It appears important to have more studies, independent and better designed before to conclude on efficacy and lack of side effects. This molecule could be used in the mnagament of grad I or be rather proposed as a second choice for prevention in cases when vaccine option is not chosen. It is difficult to maintain this recommendation in the future in the absence of new independent and robust studies.

Specific: vaccines as a therapy?

The use of a vaccine as a part of therapeuty in sick animals in order to reorientate the immune response toward a Th1 effective response is a temptation. One initial study with an experimental vaccine resulted in a positive effect. Two studies have been made with Leishmune®. The first with experimentally infected then vaccinated dogs showed a better evolution in the vaccinated dogs. The second was done on spontaneously infected sick dogs that were either vaccinated either vaccinated and treated. Positive effects were noticed in both groups with a better efficacy in vaccinated/treated dogs. There is no study with the European vaccine Canileish®. However it has been shown that vaccination of infected dogs with this vaccine reduces the infectiousness of dogs to sand flies.

In Canine clinical leishmaniosis there is an exhaustion of the immune system (lymphocytes) with likely makes difficult the response of the immne system to stimulation.

II Leishmania and healthy dogs: Prevention

In a collective approach the preventative measures include also the management of infected dogs (see previous paragraph). We will focus only on individual prevention in non-infected dogs.

The prevention is based first on the use of repellent insecticidals. This aspect is mandatory in every dog. The use of vaccine is only a second line of defence and never proposed without a preliminary prescription of insecticides. It has

to be explained to the dog owners that vaccine never replace insecticides but may be combined, giving a chance to the dog not to develop the disease if infected. Vaccine do not intend to avoid the infection but to reduce the risk (and severity) of the disease by inducing an appropriate immunity..

What repellent insecticidals to use?

Leishmania infection is biologically transmitted only by some species of phlebotomine and the prevention of the bites can be obtained only with tested molecule that demonstrated an efficacy to prevent the bite. As the bite occurs very quickly and Leishmania promastigotes instantaneously transmitted, it is important to use insecticides able to repel sand flies. Today only pyrethroids may have potentially these properties. They act by contact on sand flies (contact repellent).

Products available:

- Two collars are currently launched with studies demonstrating efficacy in prevention of CanGL.
  A deltamethrin containing collar (Scalibor® MSD) claims an activity of 6-8 months. The efficacy has been shown both in experimental conditions on sand flies and in natural condition in the prevention of infection in dogs, at least when used simultaneously in many dogs for a long period. It could even reduce the incidence of human seroconversion.
  A flumethrin (combined to imidacloprid) containing collar (Seresto® Bayer) has been tested in southern Italy in kennel dogs with very good results. This collar claims an antiparasitic activity of 7-8 months on fleas but no information is available on the repellent effect on sand flies.
  Collars are very long acting, easy to fit and visible. Their residual activity is hampered by the use of multiple baths and water. The time for effective protective diffusion in the hircoat may be quite long (up to one week) which can be an issue in dogs that have to be regularly shampooed.
- Topical preparations applied as spot-on contain permetrhin.. They have been tested against sand flies in experimental condition and provide the regulatory repellent effect for 2 to 4 weeks depending of the product. Permethrin in spot-on is alone (Ex-spot®) or combined to other insecticidal and/or acaricidal molecules in many products (imidacloprid in Advantix®, fipronil in Effitix® and Frontline Triact®, Dinetofuran in Vectra 3D®, Indoxacarb in Activyl plus®). One study showed that population-based application of spot-ons to dogs is able to reduce Leishmania infection incidence
  Spot-on have a variable residual effect. This defines the frequency of application. It has been shown that dog owners often do not apply well the spot-on, reducing the residual effects against fleas. Thus it is important to insist on the quality and frequency of application. It is generally said that it needs 2 days for insecticidal protection in the haircoat with spot-on.
- Many sprays contain pyrethroids (sometimes other pyrethroids that permethrin, the only one tested). With permethrin the efficacy is immediate but residual effect is lower. A weekly application is recommended. It is difficult to base prevention on very frequent use of sprays in endemic areas. In return they are very useful for short period of exposure. They are the only formulation able to manage the "windows of efficacy" produced by shampoos or bath on activity of both collars and spot on. The combination of spot-on or collar and strategic spray applications is recommended by the author (high-risk areas or periods). Natural compounds have no demonstrated value for prevention of canine leishmaniosis (effect on sand flies unknown). Most are designed to repel mosquitoes in humans, do not have residual effect and should be applied (at least?) daily.

When to apply insecticides?

In endemic areas the insecticidal control covers the entire sand fly season: April to October. The activity of sand flies is highly dependant of local climate, thus depending of the year and the place this period can be longer or shorter.

Other methods for avoidance of sand flies bites ?

These methods can be occasionally useful. (owner involved in a protective global approach for his animal and himself).

- Remove favourable sand flies breeding places (e.g., compost, woodpiles, stones...)
- Use domestic insecticides in houses (i.e: plugs; also protect humans; also effective against mosquitoes)
- Keep dogs indoors from dusk to dawn
- Pyrethroids impregnated mosquito nets can be useful on windows of buildings were the dogs sleep.

Vaccines?

Many research have been done since the 1980's to develop vaccines against Leishmania infantum, initially in specific mice models. The concept of theses vaccine is not to produce antibodies (humoral response of limited efficacy if any) but induce a protective cell-mediated immune response (T helper1 with production of IL-12, IFNγ , IL-2 and TNFγ), in order to mount a recall response at the time of exposure to parasites then to prevent establishment of an initial infection or control its progression. Apart individual protection the presence of dog free of or with very low number of parasites is important to manage and limit the indirect role of dogs as reservoir to other dogs and humans.

(Leishmune®) was the first vaccine marketed in Brazil (2004) (but recently withdrawn). It contained a glycoprotein GP63 of Leishmania FML (fructose-mannose ligand) combined to Saponin as adjuvant. Several studies showed the performance in: prevention of severity of CanGL; protection (80%?); therapeutic effect (reduction of severity of clinical and clinicopathological signs); reduction of infectivity to sand flies. When used massively in an endemic area it was observed a reduction in incidence of CanGL and Human cases. (LeishTec®) was more recently used in Brazil. It contains a recombinant antigen (A2) combined with saponin as adjuvant.

CaniLeish® is the first vaccine developped and launched in Europe since 2011. It contains highly purified antigens (ES) obtained from specific technique of culture of Leishmania infantum called (LiESAp). The concept vaccine tested experimentally used as adjuvant MDP (muramyl dipeptide). The commercial vaccine replaced the adjuvant by (QA-21) an adjuvant from Quillaja saponaria used also in other vaccines.
This is still the only vaccine currently available in Europe. It is used as a primo-vaccination with 3 injections on dogs older than 6 months with a preliminary verified negative serology. This is followed by annual booster injections. Several publications are available on the immunological performances with demonstration of Th1 amplification. The vaccine reduces the infectivity of dogs to sand flies as compared to non-vaccinated dogs. The vaccine would afford a protection rate of more than 92% (efficacy 68.4%).

(Letifend®) is the second vaccine in Europe very soon available (registered February 2016). It is based on a recombinant protein composed from 5 different antigenic determinants = Protein Q. Its second originality is the claim of activity in the absence of any adjuvant. The protocol of vaccination is one injection on dogs above 6 months of age with a negative quantitative serology. Immunity is said to be acquired one month later and booster injections are annual. Apart the document of European Medical Agency, there are still few publications on this vaccine. The vaccine would reduce by 5 the risk of development of the disease. As this vaccine is new ther is no information on performances in the fields apart the information available in official documents.

Some controversial points

A common point of these vaccines is the limited number of studies or publications on their performances. This is due to the high difficulty to experimentally reproduce the disease (artificial conditions) and define the markers of resistance, and the difficulty to make well designed studies in the fields (long term studies, interference of many parameters, annual variation of incidence etc).

Tolerance?

Major side effects of vaccines are cutaneous reactions at injection sites induced mainly by the adjuvant. They were

quite frequent with Leishmune (concentration of saponin). Reactions have been also documented with Canileish likely in relation with the QA-21 effect. As it contains saponin, Leishtec is also probably associated to some local reactions. Some systemic reactions have been also observed. The frequency of these side effects would not be significantly more frequent with Canileish than with other vaccines in dogs. Letifend could be considered at low risk as it has no adjuvant. Only very transient scratching at injection site is mentioned.

Interference with diagnostic tools?

The consequence of vaccination of a dog with some Leishmania vaccines (Leishmune, Canileish) is the production of antivaccinal antibodies that cross react with usual IFAT or ELISA. It is then impossible to use them to differenciate a vaccinated dog (protected) and an infected dog. For this reason a test was developed Speed-leishK® which is positive with natural antigens (infection) only and negative in Canileish vaccinated dogs. Leishtec induced antibodies are said not to cross-react with serology and Letifend vaccine antibodies do not cross react with IFAT.

Vaccination of healthy but already infected dogs?

The efficacy of a vaccine in already infected dogs is unknown. Such dogs can develop later the disease that could be misinterpreted as a failure of protection. The use of "in house tests" (Rapid immunochromatography) give specific results but sensitivity is variable. Even a negative PCR never exclude the presence of the parasite. Many vaccinated dogs in endemic areas are likely already infected.

The final decision to vaccinate or not should be based upon consideration of individual benefit/risk (like all vaccines), age (old dogs?), breed (susceptibility), habitat (rural, outdoors versus apartement?).
Current published studies sufficiently support both from experimentally infected dogs, cytokine profiles or results in the field trials the use of vaccination, with available vaccines. New studies and long term evaluation will be necessary.

References:

Most important references of this text will be presented during the conference.

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Leishmaniosis in other species: The feline and horse situation

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I Feline Leishmaniosis

Importance

Leishmania infantum infection in dogs is well documented. In return, the knowledge on Feline leishmaniosis (FeGL) is very limited and remained controversial and neglected for decades although infection was described since 1912. Until a recent period the cat was even considered as virtually refractory to infection, based on the information from the few available experimental studies. However the techniques used were unlikely able to detect subclinical infection. The development of feline medicine and more sensitive diagnostic techniques (serological, molecular) resulted in the description of an increased number of cases. Less than 60 documented cases have been published today. This limited number masks the reality of the circulation of the parasite in cats and likely signifies a lack of diagnostic.

Parasites and transmission

Several species of *Leishmania* have been described in cats, mostly in South America including *L. infantum* (= *L. chagasi*). Only *L. infantum* will be discussed here. The prevalence amongst studies of *L. infantum* infection in cat populations ranges from 0 to 68.5%. The number is highly influenced by the characteristics of the study (distribution of cats, cat population, techniques used and cut-off values for serology). It seems clear that the cat population is infected in any area where CanGL is endemic.

It is clear that sand flies may bite cats and consequently may transmit *Leishmania*. Vectorial transmission by bites of infectious phlebotomine is likely the most important way for transmission as cat may easily serve as host for sand flies and infectiousness from cat to sand flies was shown (also demonstration of the hability to feed properly the vector). Other natural routes of transmission known in dogs (venereal, dog to dog, vertical) have not been described in cats.

Clinical signs

The most common clinical signs reported in FeGL are skin (or muco-cutaneous) lesions and lymph node enlargement in more than 50% of the cases. These signs can be observed or not in combination with systemic signs.

Skin lesions can be generalized, multifocal, localized, symmetrical or not and include ulcerations, nodules, occasional exfoliative dermatitis. Cutaneous and muco-cutaneous nodules are more frequent on the head, (eyelids, nose, lips) or extremities. They are usually small (less than 1 cm), non pruritic, ulcerated or not.

The dermatological contributioncan be difficult to assess when FeGL is associated to other diseases like : alopecia and demodicosis, pruritus and flea bite hypersentitivity or feline eosinophilic granuloma complex, neoplasia (squamous cell carcinoma) or even pemphigus foliaceus. In thelesions the organisms can be found in variable abundance by histopathology, immunohistchemistry or qPCR techniques. Proliferative and ulcerative chronic inflammation of the oral mucosa is also a common finfing associated with FeGL.

The most frequent haematological abnormality reported is a mild to severe normocytic normochromic non regenerative anemia but also variable pancytopenia. Hyperproteinemia with hypergamma globulinemia is a common finding and hypoalbuminemia is occasional. Similarily to dogs, renal proteinuria and increased serum creatinine are not rare. Lymphocytosis and an increase in serum ALT activity are also reported in seropositive cats.

Diagnosis

Differential:

As lymph node enlargement is the most common sign, apart from skin and muco-cutaneous lesions, FeGL should be included in the differential the long list of diseases with solitary or generalized lymphadenomegaly (FeLV, Feline Infectious Peritonitis Virus (FIPV), *Bartonella, Mycobacteria, Toxoplasma, Cryptococcus* or other systemic mycoses), lymphoma or neoplasia.

As FeGL may induce ophthalmologic disease (recurring or chronic), a differential with similar clinical conditions (FIV, FeLV, FIPV, *Bartonella, Toxoplasma*, neoplasia or paraneoplastic syndrome) is necessary.
Feline chronic gingivostomatitis (FCGS) syndrome is frequent and considered multifactorial in cats and FeGL shoud be considered as a differential (or a component).

Parasitology: Amastigotes can be found from same samples as in dogs, also occasionally from blood smears and cytology from nasal exudate or cornea.

Molecular biology: The same techniques as used in dogs have been applied for the diagnosis of FeGL.

Serology:

The most common test used is IFAT using a recommended cut off value of 1:80. The titre range is wide from low to high in clinical cses. Quantitative ELISA is also employed and could to be more sensitive. Other techniques hav been les employed. Direct agglutination test was found less sensitive than IFAT or ELISA [23] and Western Blot more sensitive than IFAT.

Cross reactions could exist between feline antibodies to other flagellates: other *Leishmania, Trypanosoma* species (potential problem in cats living in, or rehoming from, south or central America) but not sporozoa like *Toxoplasma gondii*.

Treatment and monitoring

Treatment protocol of FeGL is still not validated due to the limited number of published cases and lack of comparative studies. Allopurinol is well tolerated at dosages 5-10 mg/kg BID (or 20 SID) may afford improvement within weeks to months. Clinical cure may be followed by relapse after discontinuation of treatment. Meglumine antimoniate may produce clinical cure and was generally obtained in the few cats treated but long-term follow is not available from these cases.

There are no studies on the pharmacokinetic and pharmacodynamic characteristics of these drugs in cats. A strict follow up is necessary like in dogs.

The prognosis appears to be variable from good to poor. Some cats are affected by chronic renal failure or hepatic disease. As treatment procedures are not still validated the prognosis in cats is particularly guarded. Visceral dissemination of Leishmania is frequently confirmed after Euthanasia.

Both treated and untreated cats may survive for years and the death is mainly due to renal or heart disease related or not to *L.infantum* infection. Like in dogs recommended therapy in case of renal disease would follow the IRIS (International Renal Interest Society) recommendations.

Prevention

There is little information on prevention in cats. An healthy cat can be tested in endemic area through antibody testing and blood PCR. Sick cats with disease requiring immunosuppressive therapies should be preliminarily tested in endemic areas. Most topical insecticidal containing pyrethroids are toxic to cats. The recent launch of a flumethrin collar seems to be the only option as it can be used in cats and have shown prophylactic effect in dogs. However no data is available for cats. Up to now there is no information on immunoprophylaxis in cat.

Portuguese publications on Feline Leishmaniosis: Maia *et al.* 2008, Marcos *et al.* 2009, Cardoso *et al.* 2010, Maia *et al.* 2010, Vilhena *et al.* 2013, Maia *et al.* 2014, Maia *et al.* 2015.

II Equine Leishmaniosis

Importance

Horse has only quite recently recognized as a species that may develop clinical leishmaniosis. Cases have also been reported in donkeys. Several studies have shown that equids had contact with the parasite by using a serological approach (in Brazil in endemic area 14.5% to 40% were positive in IFAT ≥ 40 or ELISA, in Venezuela 10%, in Italy 9%). In the north of Portugal one recent study (2013) showed antibodies to *L. infantum* in 4.0% of horses by using direct agglutination test (DAT titer 200 (n=5), 800 (n=1) and ≥1600 (n=1).

Species of Leishmania infecting horses

L. infantum and L. braziliensis have been characterized in the new world. *L. infantum* is involved in Europe but recently another original species, close or similar to a newly described asiatic agent of visceral human leishmaniasis in Thailand, *L. siamensis*, has been identified in horses through molecular analysis.

Clinical signs

The first 3 cases of Equine leishmaniosis described in Europe were reported in Spain and the first case in Portugal in 2005. Cases have also been observed in other European countries and described in Germany Switzerland, France (personal observation).

The clinical description in horses is limited: Typical lesions are nodular, sometimes crusty or ulcerated, generally localized to the pinna, adjacent neck, top of the head, cheeks. They may be solitary or multiple. The lesions may heal spontaneously but sometimes relapse. Scrotal lesions were also reported in donkeys.

Diagnosis:

The diagnostic of Equine leishmaniosis is most often a discovery from histopathology performed after surgery on nodules. Biopsy of the lesions or even cytology of the nodules could be also diagnostic. The dermal inflammatory infiltrate is lymphohistiocytic, pyogranulomatous with sometimes tuberculoid granulomas and contains amastigotes in variable abundance. In horses antibodies may be present (low titer in IFAT, transient?) and it is considered that leishmaniosis could be naturally (most often?) cleared by host response.

Treatment and prevention

There is no validated protocol for treatment. Some cases have been treated more or less successfully by surgery or neglected. There is no study on preventive methods. The transmission to horse could occur from sand flies bites but the question on the potential role of mechanical vector (horse flies, stable flies...) could be raised. The use of pyrethroids containing products both on the infected horse and horses living around could be an option for prevention.

Portuguese publications on Equine Leishmaniosis: Rolao *et al.* (2005), Lopez *et al.* (2013), Gama *et al.* (2014).

III Leishmania infection and wildlife

More and more information is now available, suggesting or demonstrating the wide circulation and distribution of Leishmania infantum within a variety of species other than dogs (or cat).

The demonstration of the role of a species as part of the reservoir needs the evidence of the presence of *Leishmania* in a significant number of animals, a long term maintain of infection and the capacity to infect sand flies (xenodiagnosis). An increasing number of mammal species have been shown to be (likely) infected by using molecular tools. In return few have been tested on xenodiagnosis. Rare cases of clinical leishmaniosis are detected in nature but some were observed in zoo animals.

Wild carnivores and insectivores

High rates of *Leishmania* infection (10–40 %) have been found in red foxes (*Vulpes vulpes*) in different areas of Europe where the disease is endemic but good xenodiagnostic studies are still lacking. The infection was also not surprisingly detected in Wolves (*Canis lupus*) (Italy, Portugal, Spain).

Clinical disease has been observed in Foxes, Wolfes, Lion.

PCR studies have shown the presence in Iberian lynx, mongoose, stone marten, pole-cat, mink, wild cat, genet, badger and even recently hedgehog.

Leporids

The Iberian hare (*Lepus granatensis*) is abundant in Iberian Peninsula. This species has been clearly shown, to be involved (reservoir) in the oubreak of Human Visceral Leishmaniasis in Fuenlabrada, Madrid. Healthy, naturally infected hares were infectious to *P. perniciosus*, a competent vector for *L. infantum* and blood meal analysis of sand flies captured showed a feeding preference for hares. Similarily wild rabbits (*Oryctolagus cuniculus*) have been recently shown to be frequently infected and demonstrated also infective for sand flies (*P. perniciosus*). Rabbits are an important source of blood meals for *P. perniciosus*.

Rodents

Rat (*Rattus rattus*) is an abundant rodent species in Europe. High rates of *Leishmania* infection (15–45%) have been found in Italy using PCR and also Infectiousness has been proven. Infection was also detected in domestic mouse (*Mus musculus*) and rat *R. norvegicus*

Leishmania infection seems to be typically asymptomatic in wildlife.
It appears clearly that the concept of reservoir of *Leishmania infantum* has to be deeply revised and includes many species that can play different epidemiological role in the different countries or ecosystems.

Portuguese publications on *Leishmania* in wild fauna: Abranches *et al.* 1983, Abranches *et al.* 1984, Seioao Santos *et al.* 1996, Helhazar *et al.* 2013, Diaz Saez *et al.* 2014, Cardoso *et al* 2015.

The author strongly recommend the lecture of the review paper on feline leishmaniosis.
LeishVet update and recommendations on feline leishmaniosis : Pennisi *et al.* Parasites & Vectors (2015) 8:302.

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Veterinary Dermatology: Old and new challenges, new products

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The evolution of Veterinary Dermatology during the last decade produced a large amount of new knowledge on more and more complex diseases. In the same time the prevalence of most important classical and well known dermatoses didn't decrease but some disease changed their clinical aspects.

The evolution of environment and mode of life of companion animals resulted in a convergence towards more allergic/hypersensitivity situations like in human medicine. In parallel new concepts of medicine appeared and some very new groups of molecules were launched. Three major domains in dermatology will be commented: Ectoparasitic diseases, Immuno-allergology and skin infections.

I Ectoparasiticides the two approaches

Topical the new challenges
Topical products work by contact and protect against external parasites.

Sprays
are known for their quick onset of action. The theoretical distribution of the active ingredient after a spray administration is immediate. This rapid onset of action include knock-down 1h post-treatment but also anti-feeding effect but can lack persistency and inaccurate dosing may happen.

Spot-on pipettes
are designed for easy administration but the solution must be applied at skin level and run-off can happen if large volumes are applied in single spots. Active ingredients from spot-on solutions must distribute from the administration area and cover the body surface to reach the effective concentrations. Their onset of efficacy could be 6h against fleas: 24h against mosquitoes and ticks. Parameters such as the hair length, the skin lipid layer (possible removal by shampoos) and effects of frequent water immersion, weekly shampoos may decrease the performance.

New definitions.
These products may provide reliable and unique weapon against ectoparasites of companion animals. Only some of the contact-based topicals can indeed provide repellency. The increasing warning about not only bite induced hypersensitivity but mainly vectorial transmitted disease has driven the development of new definitions.

Speed of kill is the earliest time at which "complete" efficacy is achieved. For topical ectoparasiticides the term "preventative efficacy" may be used while systemic products may use the term "persistent killing effect". Repellency efficacy is defined as 95% reduction within 24 hours post infestation in the number of ticks (dead or alive) or fleas that have taken a blood meal (improperly but regulatory termed anti-feeding effects). This value is 80% at 1h00 for

mosquitoes or sand flies.

Only molecules in the pyrethroids group have these properties. This is the reason for which permethrin was (re)introduced in most spot-on (Activyl-plus™, Advantix™, Duowin™, Effitix™, Exspot™, Frontline Triact™, Vectra 3 D™) with a protection of 2 to 4 weeks depending of the insect (mosquitoes or sand flies) and the product. The 2 other molecules used are in long lasting collars Deltamethrin (Scalibor™)(dogs) and Flumethrin (Seresto™)(dogs and cats)

Systemics: the new molecules

Systemic products have been available for a long time. Newer products have improved efficacy and safety. The advantage is their wide and excellent distribution to all parts of skin. The disadvantage is their activity is conditioned first by the bite from the arthropod.

Macrocyclic lactones:

Selamectin (Revolution™, Stronghold™) (used as a systemic spot-on rapidly absorbed through the skin, and distributed via the blood) has an activity against a variety of internal and external parasites including Fleas, Sarcoptes and Otodectes. Selamectin is also active on ticks but this characteristic do not appear in the claim in Europe as the action is too slow for the prevention of vector borne diseases. The other macrocyclic lactones have no activity against fleas at usual dosage and variable activities against other ectoparasites.

Neonicotinoids:

Nitenpyram (Capstar™) is administered PO in pill form to kill fleas in both dogs and cats. It is absorbed rapidly, with maximal blood concentrations reached within 1.2 hr in dogs and 0.6 hr in cats. Fleas begin to die within 20–30 min of administration, with 100% flea mortality within 3–4 hr. The compound is rapidly eliminated within 24–48 hr, primarily as unchanged nitenpyram. Other neonicotinoids are not used as systemics (imidacloprid, dinetofuran). Neonicotinoids are insecticidal and have no effect on mites and ticks.

Recent products

Spinosyns:

A recent family of insecticides derived from the fermentation of the actinomycete, Saccharopolyspora spinosa. Spinosad (Comfortis™) is administered PO as a chewable tablet and has activity against fleas (and lice) for dogs and cats. The compound is extremely rapidly absorbed after oral administration. Spinosyns are also acaricidal to a less extent (although not often mentioned). Spinosad has a strong and rapid activity on ticks but this activity doesn't cover the usual 4 weeks as claimed for most of ixodicidal products for dogs bur rather 10 days. Thus it can be used for curative effect or short term protection. (Spinetoram is a second molecule in the spinosyn group but is used topically without systemic diffusion).

Isoxazolines

The most recently discovered group of compounds that have both potent insecticidal and acaricidal activities. Isoxazolines have a novel mode of action and specifically block arthropod ligand-gated chloride channels. They are eliminated through the biliary system. Afoxolaner, fluralaner and sarolaner (NexGard™, Bravecto™ and Simparica™) are currently the 3 compounds approved for use in veterinary medicine in dogs and are given as chewable tablets (and recently fluralaner in cats as a spot-on). The compounds are readily absorbed after oral administration and provide 4 weeks (Afoxolaner, Sarolaner) to (8) 12 weeks (Fluralaner) of insecticide and acaricidal activity. The 3 products are registered for the control of fleas (1 Afo. -1Saro. -3 Flu months) the ticks (1Afo.-1Saro. month and 8 weeks Rhipicephalus Flu., 12 weeks Dermacentor, Ixodes Flu.) and treatment of Sarcoptes acariosis (Sarolaner). These products have all very high initial concentrations and an extended time above the effective dose their length of action is dose dependent.

As a consequence the real spectrum of these products is much wider than registered. All 3 are active on Sarcoptes, Otodectes and even Demodex (communication, publication in press...). It appears that the treatment of canine demodicosis could be much easier with such products. In return these molecules dugs do not have repellent effect and are unable to prevent the transmission of quickly transmitted agents like Ehrlichia.

II Management of atopic dermatitis

Canine Atopic Dermatitis is an inflammatory and pruritic allergic skin disease developing on genetically predisposed dogs.

The mechanisms of the dermatitis are complex and change with time. This is typically a multifactorial disease including : interaction of genetic factors, intrinsically and acquired epidermal barrier dysfunction and immunologic factors. With time the IgE mediated reaction (HS1) changes toward mechanisms closer to a contact allergic dermatitis. Cutaneous microbiome is modified and participates to the evolution of the dermatitis.

The diagnosis is based on well-known criteria, the currently use being Favrot's criteria. To be effective these criteria have to be evaluated on dogs on which other primary pruritic dermatitis have been ruled out.

The treatment of CAD is based on the control of bacterial/fungal complications (shampoos), the control of any pruritic stimulus (fleas) and, ideally, the eviction of offending allergens.

In practice it is important to control the pruritus in many dogs.

Antipruritic drugs are corticosteroids – antihistamines – Cyclosporine – and ,secondarily, slower or less active compouds.

Oclacitinib (Apoquel™) a Janus kinase inhibitor.
Janus kinases (JAKs) are enzymes linked to T-cells that induces cytokine production. Oclacitinib inhibits Janus kinase1-dependent cytokines involved in allergy and inflammation and IL-31 which trigger pruritus perception. It is rapidly absorbed orally , active and has a short residual effect. It acts as rapidly as Corticosteroids) (experimentally reduction of pruritus in 1-3h). A study in dogs with CAD showed a strong effect (versus placebo) or similar efficacy as corticosteroids, a similar but faster activity as compared to cyclosporin.

The indication is the control of pruritus associated with allergic dermatitis and clinical manifestations of CAD at a dose 0,4-0,6 mg/kg BID 14 days then SID.

Adverse effects are rare, mild, transient and spontaneously regressive most of the time: diarrhoea and vomiting, sometimes anorexia, and lethargy

The question of risk of exacerbation of pre-existing disease was raised: malignant neoplasia (after 21 and 60 days) generalized demodicosis (after 28 days). This subject is still controversial without clear demonstration of risk. Nevertheless the control of pruritus in a dog do not interfere with the evolution of a causal underlying disease and the use of Oclacitinib, may artificially mask it for a time.

Short-term treatment with oclacitinib does not significantly interfere with intra-dermal skin testing.
There are still few studies in cats and results are controversial.

Proactive Therapy

The concept of Proactive therapy was developed in human medicine in atopic dermatitis. This proactive approach starts with an intensive topical anti-inflammatory therapy until all lesions have mostly cleared, followed by a long-term, low-dose, intermittent application of anti-inflammatory therapy to the previously affected skin, together with an emollient treatment of unaffected skin.

Similar skin barrier dysfunctions have been observed in canine atopic skin as in humans but only one study was conducted until now. The idea was to use hydrocortisone aceponate spray (Cortavance™) applied 2 consecutives days per week after dealing with clinical manifestations of atopic dermatitis in dogs: The time before relapse was much longer than in the control placebo group.

III Fighting against bacterial resistances

In human medicine there is an increasing prevalence of infectious diseases due to drug or multidrug-resistant pathogenic bacteria.

The most important are *Staphylococcus aureus* Meticillin Res (SARM), *Pseudomonas aeruginosa*, ESBL (Extended-β-lactamase-producing) *E. coli* and *Acinetobacter baumanii*.

The evolution of contacts between humans and animals increases the risk of exchange of resistant bacteria in a world with slow development of new antimicrobial drugs

Antibiotherapy

Some clues for a rational use of topical and systemic antimicrobials can be based on:

- The evidence of non self-limiting bacterial infection must be proven
- The identification of underlying predisposing conditions (Hypersensitivities, ectoparasites)
- Antimicrobial susceptibility testing.
- In case of uncomplicated and local infection use topical therapy alone in first line (antiseptic +/- topical antibiotic (ex : fusidic acid) before systemic therapy

Choice of first line drugs: (evolutive and suggested list)

- A spectrum as narrow as possible
- Critically important antimicrobials, not allowed for veterinary use, should be only the « last chance » treatment (Check country legislation).
- The preparation of the treatment
- Antimicrobial therapy must not be a susbstitute for good hygiene practices by the veterinarian:
- Environmental hygiene, Good medical and surgical practices in case of suspicion of MDR or contagious disease; special consulting ; confinement ; special care
- Client education, compliance assessment - A good follow up is necessary- monitoring are key of success.

The choice of the drug :

1st choice: use well-tolerated narrow Spectrum Antibiotics with antistaphylococcal activity
Empirical treatment is possible: cefalexin, amox/ac clav, clindamycin, fusidic acid (topical).
2nd choice: use only after bacteriology and when antibiogram shows that 1st choice is inefficient. Newer broad spectrum antibiotics with medical importance in human and veterinary practice cefovecin, enrofloxacin, marbofloxacin.
3rd choice: Use only for treatment on demonstrated MDr strains or when 1st and 2nd choice inefficient and topical therapy not possible ; According to culture test and country rules : chloramphenicol, florphenicol.

Duration :

It is important to never underdose and the length of treatment has to be long enough.

- Superficial pyoderma : minimum 2 to 3 weeks + 1 week after clinical and cytological cure
- Deep pyoderma : minimum 4 to 6 weeks + 2 weeks after clinical and cytological cure
- Never stop treatment before cure

Dosage:

- Amox/Ac clav : 12.5 to 25 mg/kg bid PO
- Cefalexin : 22 to 30 mg/kg bid, or 30 to 40 mg/kg sid
- Clindamycin : 11 mg/kg bid PO
- Enrofloxacin: 5 to 20 mg/kg sid PO
- Marbofloxacin: 2.5 to 5 mg/kg sid PO

In case of bacterial otitis it is important to keep in mind that 1) there is no effect of systemic treatment 2) the concentration of antibiotics is 100-1000 times above in vitro susceptibility testing concentrations
The treatment should focus first on adequate cleaning agent and topical antibiotics.

Antiseptics and other antibacterial compounds.

Chlorhexidine

Remains the antiseptic of choice for the treatment of superficial infections. It is bactericidal against gram+ and gram- bacteria

Concentrations 1 to 4% are effective against Staphylococcus spp. and other bacteria (faster at higher conc.). It can be associated to tris-EDTA for otitis externa.

tris-EDTA seems to enhance the activity of chlorhexidin or topical antibiotics in ears.

Chlorhexidine may have a residual antistaphylococcal effect (up to 10 days) when used on the haircoat alone or combined (miconazole, Triz-EDTA and phytosphingosine)

Other antibacterial compounds.

There is a recent interest for old but neglected or newly discovered antimicrobials :
Silver compounds, DMSO, N-acetylcystein, Medical grade honey, Propolis, Antimicrobial peptides, Bacteriophages, Incomplete iron salt of polyacrylic acid.

N-acetylcysteine

A non-antibiotic drug with antibacterial properties (Inhibition of growth of gram+ and gram- bacteria (S. aureus, P. aeruginosa) at concentration 2-5%. It has synergistic action against P. aeruginosa with some antibiotics (carbenicillin, ticarcilin, ciprofloxacin), decreases biofilm formation and could be used as a potential agent synergistic with antibiotics by enhancing permeability and facilitation of access to bacteria. It has a proven efficacy and safety profile.

Micronized silver (silver ions).

Silver has been used for disease prevention for many centuries. It is biocidal to many different types of microorganism at doses 0.3 to 2.0 ppm. Compared to silver ions, nanoparticles have longer lasting biocidal properties and are easier to incorporate into matrix materials. Bacterial (and probably fungal) sensitivity to silver is genetically determined (plasmides).

Silver ions have different targets, moreover they increase membrane permeability and increase efficacy of antibiotics (synergy with enrofloxacine).

Nanosilver is safely used in dermal wound care since decades; ointments and bandages are used routinely in hospitals.

Pet care nano-products, as shampoos composed of silver nanoparticles, are already approved for use in dogs and cats : silver colloidal nanoparticles associated with hyaluronic acid are found in shampoos: Phlox®

Antimicrobial creams could have a broad sprectrum (from gram+/- bacteria C. albicans and M. sympodialis and dermatophyte?) and be effective at concentrations as low as 0,1%.

Dimethyl sulfoxide (DMSO)

Hygroscopic organic solvent miscible with lipids, and water, excellent vehicle able to cross skin barrier and microbial membranes. It has many properties including antimicrobial activity against bacteria (S. pseudintermedius), fungi and viruses (concentration dependant).

Antimicrobial peptides

Host defense peptides (HDPs) are central effector molecules of innate immunity. Their synthesis is stimulated by different infectious and noninfectious molecular signals (danger associated molecular patterns (DAMPs). They are produced by inflammatory and epithelial cells (including keratinocytes)

In vitro, natural canine AMPs (β-defensins and cathelicidins) are rapidly effective against meticillin-resistant or susceptible strains of *Staphylococcus* and *P. aeruginosa*, with killing effects within 2hours. Some recent and promising studies have been conducted on the antibacterial effect of topically applied AMPs on bacteria isolated from dogs with pyoderma. These molecules could be used in the antibacterial treatment in a near future.

Marc Dhumeaux

Uroliths in small animals; why does that happen?

Brief description of lecture:

Uroliths are a common cause of urinary tract disease in dogs and cats. This lecture reviews the mechanisms of formation of the three most common types of uroliths in dogs and cats: struvite, calcium oxalate and ammonium urate.

Abstract:

Urinary stones, or uroliths, are a frequent cause of urinary tract disease in dogs and cats. Clinical signs frequently associated with stones include haematuria, abdominal pain and recurring urinary tract infections. Stones lodged in the urethra may be responsible for complete urinary obstruction, which can have severe consequences if the obstruction is not removed in a timely fashion. The stones formed in the renal pelvis may cause pyelic or ureteral obstruction. Unilateral obstructions of the upper urinary tract are not always immediately associated with clinical signs in the animal, but they can nevertheless lead to irreversible renal lesions.

Detailed knowledge of the factors predisposing to stone formation, as well as their physical and chemical characteristics, allow the clinician to design the best therapeutic plan for their elimination and the prevention of recurrence.

Mechanism of formation for uroliths

Urine is naturally an aqueous environment intended to dispose of metabolic waste products in a dissolved form. In some conditions, certain waste products, minerals in particular, may precipitate and form crystals. If these crystals persist for enough time in the urine, they can aggregate and form bladder stones. We sometimes define urinary crystals suspended in the urine as microliths, and bladder stones as macroliths.

The prerequisite for the formation of urinary crystals is a urine supersaturated with components of these crystals. A "stable solution" is one in which the physicochemical conditions of the urine do not allow the formation of crystals, while a "metastable solution" is one in which the formation of crystals is possible, however, aggregation is not sufficient to result in the formation of bladder stones. Lastly, a solution that allows the formation of stones is termed an "unstable solution".

Numerous physical and chemical factors predisposing the patient to the formation of urinary stones have been identified. These factors differ depending on the type of crystals. One factor common to the formation of all types of stones is the urinary concentration. The less concentrated the urine, the less it is saturated in elements responsible for formation of crystals, significantly reducing the chance of stone formation. Thus, increasing the consumption of water in order to promote diuresis will be common to all strategies for prevention of stones, regardless of their type.

The majority of foods designed for the prevention or dissolution of urinary stones have an increased level of sodium, in order to promote water intake and diuresis.

The concept of relative supersaturation (RSS), developed in human urology in the 1960s and adapted for companion animals, has allowed improvements in the design of foods intended for the prevention and dissolution of urinary stones. RSS is an in vitro method based on the determination of the urinary pH and of the urinary concentrations of different analytes involved in the formation of stones. These include calcium, oxalate, sodium, potassium, magnesium, urate, ammonium, citrate, phosphate and pyrophosphate. All these data are analysed by a computer programme that calculates the concentration of a great number of complexes formed by the interaction of different ions present in the urine at a given pH. A RSS is specific to one type of urinary crystal, and RSS values have been determined under which the prevention or even the dissolution of urinary stones is possible. For example, $RSS_{struvite} < 1$ corresponds to an undersaturated urine, and is compatible with the prevention and even the dissolution of this type of stone.

1. Struvite stones

Struvites, also known as magnesium ammonium phosphate uroliths, are one of the two types of stones most frequently diagnosed in dogs and cats (the other one being calcium oxalate). As their name implies, they result from the crystallisation of ammonium, phosphate and magnesium ions. The components of these stones are present in normal urine, but the formation of crystals depends on factors able to alter the concentration of these different ions and the urinary pH.

The mechanism for formation of struvite crystals differs between dogs and cats. In dogs, the great majority of struvites are formed secondary to a urinary tract infection with urease producing bacteria, whilst in cats, they develop in a sterile urine in most cases. Urine pH plays an important role in the solubility of struvite crystals. Indeed, it has been shown that an acidic pH (< 6.5) allows the dissolution of struvite crystals whilst an alkaline pH (> 7) promotes their formation (Lulich *et al*. 2011).

In dogs as well as in cats, the proportion of struvite stones submitted for spectrophotometric analysis has markedly decreased over the last three decades. The decline was especially observed in the 1980s and beginning of the 1990s. At the beginning of the 1980s, almost 80% of stones submitted for analysis were struvites. The reduction in the number of struvites occured simultaneously with the increase in calcium oxalate stones in both species. Since the 2000s, the proportion of struvite stones amongst all stones submitted for analysis has varied between 40% and 50% in both dogs and cats (Osborne et al. 2009). The development and improvement of foods designed for the prevention and dissolution of struvite stones has probably played a role in this phenomenon.

Struvite stones are radio-opaque. A 2% risk of false-negative results was reported using radiography without contrast. This can be explained by the fact that stones measuring less than 3 mm in diameter can be undetected by conventional radiography. Generally, ultrasound or double contrast cystography are more sensitive than conventional radiography for diagnosing urinary stones (Feeney et al. 1999). A study in dogs showed that conventional radiography was useful for predicting the type of stone present in the urinary tract. Stones with pyramidal shape, larger than 10 mm, with an ovoid shape and having smooth contours had positive predictive values of 90%, 100%, 80% and 75% for a diagnosis of struvite (Feeney *et al*. 1999).

a. Struvites in dogs

In dogs, despite physical and chemical parameters favouring the formation of struvite crystals, the development of this type of stone is unlikely without the presence in the urine of the urease enzyme, produced by some bacteria.

The bacteria most commonly reported in association with struvite stones are *Staphylococcus pseudointermedius* and *Proteus* spp. Bacteria which sometimes produce urease and often found in association with struvite stones in dogs include *Escherichia coli*, *Pseudomonas* spp. and *Klebsiella* spp (Palma *et al.* 2013). The role of urease is to convert urea into ammonia. The ammonia produced during this reaction acts as a buffer for the protons present in the urine causing production of ammonium ions. The latter may then react with magnesium and phosphate ions allowing the formation of struvite crystals. The buffering effect of ammonia is also responsible for an increase in urinary pH, promoting crystallogenesis.

Cases of struvite stones associated with bacteria not producing the urease enzyme and cases of sterile struvites have rarely been reported in dogs. The mechanism of formation in these cases is not well understood and may be similar to the one in cats.

In dogs, struvite stones are more frequently diagnosed in females than in males. This can be explained by a higher risk of urinary infection in female dogs.

In dogs, around 95% of struvite stones are discovered in the lower urinary tract, and only 5% in the upper urinary tract (renal pelvis and ureters). Around one third of stones in the upper urinary tract in dogs are struvites.

b. Struvites in cats

Unlike in dogs, struvite stones in cats are formed in sterile urine in approximately 95% of cases. Thus, in the large majority of cases, the formation of struvite stones is only influenced by the urine's physicochemical factors such as the urinary pH, urinary concentration and the concentration of calculogenic minerals such as magnesium and phosphorus (Hostutler *et al.* 2005). Increased fibre consumption may also play a role in the formation of struvite stones in cats (Lekcharoensuk *et al.* 2001).
A sexual predisposition has not been shown in cats. One epidemiological study showed a higher risk (odds ratio > 2) for several breeds such as Chartreux, Ragdoll, Himalayan and the Domestic Shorthair and a lower risk (odds ratio < 0.5) for Abyssinian, Birman, Russian Blue, Rex, Siamese and cross-bred cats (Thumchai *et al.* 1996).
Cats of all ages may be affected. One study, however, showed a higher incidence in cats between 4 and 7 years, with a median age for affected cats of 5.75 years. The rare cases of struvites associated with a urinary infection were more frequent in cats younger than 1 year or older than 10 years (Thumchai *et al.* 1996).

2. Calcium oxalate stones

a. Epidemiological data

Today, calcium oxalate stones are encountered with a frequency similar to that of struvite stones, and represent 40% to 50% of stones submitted for analysis in both dogs and cats (Osborne *et al.* 2009).

These stones are more frequently found in males than in females, with a ratio of 2:1 in dogs and 1.5:1 in cats (Gisselman *et al.* 2009). This sexual predisposition is also present in humans. A protective role of oestrogens, through reduction in urinary oxalate excretion and increase in urinary citrate excretion, has been suggested. A higher risk of developing these kinds of stones has likewise been identified in sterilised individuals and obese individuals (Lekcharoensuk *et al.* 2000; Houston *et al.* 2003). Certain canine and feline breeds are predisposed to calcium oxalate stones.

Calcium oxalate stones are mainly found in the lower urinary tract in dogs and cats, and only 2% to 3% are located in the renal pelvis or ureter (Gisselman *et al.* 2009). Kidney and ureteral stones are composed of calcium oxalate in

about one-third of cases in dogs and in almost all cases in cats (Ross et al. 1999). A large number of cats diagnosed with calcium oxalate stones in the upper urinary tract are also diagnosed with chronic kidney disease. It is likely that repeated, bilateral renal obstructions or sub-obstructions are the cause for renal damage in these individuals.

Similarly to struvite stones, calcium oxalate stones are systematically radio-opaque, making their detection possible by conventional radiography.

b. Pathophysiology

As their name implies, they are formed through crystallisation of calcium and oxalate. The reason why certain animals are prone to developing calcium oxalate stones is still not well understood. However, hypercalciuria, hyperoxaluria and a persistent high urinary concentration are considered as risk factors.

Hypercalciuria may result from an increase in intestinal absorption of calcium, from an increase in its bone resorption or from decreased renal reabsorption. A digestive hyperabsorption has been described in miniature Schnauzers suffering from calcium oxalate stones (Lulich *et al.* 1991). Primary hyperparathyroidism is a cause of hypercalcemia and hypercalciuria through increased bone resorption of calcium and is frequently associated with the formation of calcium oxalates in dogs. Hyperadrenocorticism may likewise contribute to the formation of calcium oxalate through an increase in calciuresis (Feldman and Nelson 2004).

The development of calcium oxalate stones may also result from a deficit in substances which normally inhibit their formation.

Pyrophosphate excreted in the urine is a natural inhibitor of precipitation of calcium oxalate. Pyrophosphate is derived from phosphate and a restriction in phosphate, sometimes considered to limit the risk of struvite formation, is thought to be a risk factor for the formation of calcium oxalate stones.

Excessive restriction in magnesium may likewise play a role in the formation of oxalate stones. This mineral may effectively complex with oxalate, reducing the latter's capacity to react with calcium for the formation of crystals. Lastly, nephrocalcin is a substance naturally present in urine that inhibits the formation of calcium oxalate crystals. Its activity may be decreased in patients predisposed to these types of stones (Carvalho *et al.* 2006).

Contrary to struvites, the solubility of calcium oxalate crystals is not influenced by the urinary pH. Acidosis and aciduria, however, may contribute to formation of this type of stone through an increase in calciuresis and a reduction in urinary excretion of citrate, a competitive inhibitor of the formation of calcium oxalate crystals.

3. Ammonium urate stones

Ammonium urate stones are the third most frequent type of stone encountered in dogs and cats, representing 5 to 10% of stones submitted for analysis in both species. This frequency has not changed over the past decades, unlike that for struvite and calcium oxalate.

Urate is a product of the degradation of purines coming from the metabolism of proteins and nucleic acids. Under normal conditions, uric acid is converted into allantoin in the liver under the action of the enzyme uricase, and only a small quantity of uric acid is excreted in the urine. Allantoin is very soluble in the urine, whilst uric acid may complex with different cations, especially ammonium ions, to form ammonium urate crystals.

The large majority of urate stones are found in the lower urinary tract. These stones are radiolucent or only slightly radiopaque, making the use of ultrasound or double contrast radiography necessary for their detection.

a. Dalmatians

A recessive autosomal genetic mutation exists in Dalmatians responsible for a strong predisposition to urate stones. All Dalmatians are homozygotes for this mutation (Adams and Syme 2010).

These dogs produce hepatic uricase normally, but cannot ensure the transport of uric acid into the hepatocytes for its conversion into allantoin and into the renal proximal tubule cells for its reabsorption. This results in hyperuricosuria, causing formation of urate crystals.

In more than 90% of cases, the urate stones submitted for analysis are from male individuals. It is likely that the smaller diameter of the urethra in male dogs make them more prone to urinary obstructions than females, justifying the ablation of stones and their submission for analysis more frequently in males than in females.

The prevalence of urate stones in male Dalmatians is estimated at approximately 30% (Bannasch *et al*. 2004).

The genetic mutation responsible for the formation of urate stones in Dalmatians has also been identified in English Bulldogs and Black Russian Terriers. The frequency of the mutation is, however, much lower in these two breeds than in Dalmatians.

b. Hepatic dysfunction

Hepatic dysfunction predisposes to urate stones through decreased hepatic conversion of uric acid into allantoin, causing hyperuricosuria. The hepatic dysfunction may likewise be responsible for reduction in the conversion of urea into ammonia, causing hyperammoniuria. Even if, theoretically, all forms of hepatic insufficiency should predispose patients to urate stones, the latter are most often encountered in association with a portosystemic shunt.

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Marc Dhumeaux

Uroliths in small animals; How to get rid of them for good?that happen?

Brief description of lecture:

Treatment options for dissolution and ablation of the three most common types of uroliths (struvite, calcium oxalate and urate) are explained and recommendations to prevent recurrence in predisposed patients are given.

Abstract:

1. Treatment of struvite stones

Treatment options for struvite stones include dissolution and ablation. In all cases, once the stones are eliminated, preventative measures will need to be taken in order to prevent recurrence.

a. Techniques for ablation of urinary stones

Different techniques, more or less invasive, for ablation of urinary stones have been described. These techniques are valid regardless of the nature of the urinary stone.

Voiding urohydropropulsion may be considered in dogs and cats for elimination of stones sufficiently small in size. Small struvite stones are particularly amenable to this technique because their contours are generally smooth reducing the chance of injuring the urethra during their passage. As a general rule, this technique may be attempted for stones less than 5 mm in diameter in male or female dogs weighing over 8 kg, those less than 3 mm in small dogs or female cats and those less than 1 mm in male cats. In order to obtain maximal relaxation of the urethra, deep sedation or general anaesthesia are recommended.

Laser lithotripsy is used in some institutions for non-invasive ablation of bladder stones. For this, an endoscope brings the fibre laser into contact with the stone in order to fragment it into pieces sufficiently small to be evacuated by urination or using an endoscopic basket.

Extracorporeal shockwave lithotripsy is often used for the elimination of kidney stones in human medicine, but at the moment, it is used very infrequently in veterinary medicine due to equipment availability.

Surgical techniques for stone removal include cystotomy and antepubic cystoscopy in the case of bladder stones, and ureterotomy, nephrotomy and nephroscopy in the case of stones located in the upper urinary tract.

b. Dissolution of struvite stones

The dissolution therapy differs between cats and dogs. Indeed, in the latter, in which stones are due to a urinary tract infection in the majority of cases, elimination of the infection through appropriate antibiotic therapy is the most important part of the dissolution process. The choice of antibiotic should be based on urine culture and sensitivity, and should be continued for approximately one month after the elimination of the stones in order to avoid recurrence. As they dissolve, the stones can release bacteria. Urine cultures every four weeks during the dissolution process and approximately one to two weeks after the end of the antibiotic treatment are recommended to ensure the absence of bacterial resistance and to confirm complete elimination of the infection.

In both species, a food specifically designed for the dissolution of struvite stones is recommended. The goal of calculolytic food is to reduce urinary pH, reduce the intake of calculogenic minerals, reduce the urine concentration and, for dogs only, reduce production of urea as a substrate of bacterial urease.

Acidification of the urine increases solubility of struvite crystals. Achieving an acidic urine pH is considered a key element in the success of treatment. In dogs, if a sufficiently acidic urinary pH (< 6.5) is not obtained using a specific diet only, the administration of DL-methionine may be considered at 100 mg/kg twice daily. This medication must be used cautiously in cats due to the risk of haemolytic anaemia.

In dogs, a diet reduced in protein ensures a reduction in the production of urea available as substrate for the bacterial urease, which can decrease the production of ammonia.

Foods designed for the dissolution of struvite stones are likewise low in magnesium, the intake of this mineral in excessive quantities in the diet having been clearly identified as a factor predisposing to struvite stones in cats (Lekcharoensuk *et al*. 2001).

Increased levels of sodium in these types of foods stimulates water intake, increasing diuresis and reducing urinary concentration. The result is a reduction in urine saturation of calculogenic minerals.

In dogs, the average duration of the dissolution process is 3 months (Adams and Syme 2010). In cats, it has recently been shown that a diet formulated with $RSS_{struvite} < 1$ allowed a dissolution of stones within an average time of 18 days (10 – 55) (Houston *et al*. 2011).

The duration of treatment depends in large part on the initial size of the stone and owners' compliance.

Imaging of the urinary tract (radiographs or ultrasound) is recommended at least every 4 weeks during the dissolution protocol to ensure its efficacy. When radiography is used as a follow-up technique, it is recommended to continue the medical treatment for at least one month after confirmation of the absence of mineralized material in the urinary tract as stones cannot be detected accurately using this imaging modality when their size is smaller than 3 mm.

After a successful dissolution protocol for struvite stones, a diet designed for the prevention of relapse is recommended. In dogs, it is also very important to monitor carefully for signs of relapse of urinary tract infection which, without early recognition and treatment could rapidly lead to recurrence of struvites in predisposed individuals.

2. Treatment and prevention of calcium oxalate stones

Calcium oxalate stones cannot be dissolved, and the only treatment option consists of their ablation using the above-mentioned techniques.

It was shown that the recurrence rate in the three years following ablation of calcium oxalate stones was around 50% (Lulich *et al*. 1999). The implementation of preventative measures in order to reduce the risk of relapse is thus vital for this type of stones.

Diets rich in fat, phosphorus, potassium and magnesium have shown a benefit for the prevention of calcium oxalate stones. A reduction in carbohydrate intake has shown a benefit in dogs but not in cats (Lekcharoensuk et al. 2002a; Lekcharoensuk et al. 2002b). The key element in the prevention of calcium oxalate stones is increasing hydration in order to reduce urine concentration. This can be achieved by using a wet food or by adding water to dry foods. It is recommended to obtain a urinary specific gravity lower than 1.035 to 1,040 in cats and 1.020 in dogs in order to reduce the risk of relapse. Repeating urinalysis regularly allows confirmation that this objective is achieved. Regular urinalyses are also important to confirm the absence of calcium oxalate crystalluria.

When dietary measures are not sufficient to ensure adequate dilution of the urine, the administration of a thiazide diuretic should be considered. This type of diuretics works by inhibition of sodium reabsorption in the distal tubule, which has a consequence to increase the renal reabsorption of calcium. As a result, the urine becomes less concentrated and the urine calcium concentration is reduced. In dogs, a significant reduction in urinary concentration of calcium has been shown with the use of hydrochlorothiazide at 2 mg/kg BW every 12 hours (Lulich et al. 2001). Furosemide and other loop diuretics are contraindicated for the prevention of calcium oxalate stones because they increase calciuresis.

Utrasound or radiographs of the urinary tract are recommended at 2 to 4 weeks, 3 months and then at least twice a year after the ablation of the calcium oxalate stones. The goal of these regular rechecks is to detect stones when they are small enough to be extracted by voiding urohydropropulsion rather than with the use of more invasive or expensive techniques.

3. Treatment and prevention of urate stones

Ammonium urate stones may be dissolved with adapted therapy. The treatment consists of a diet low in protein, alkalisation of the urine and administration of a xanthine oxydase inhibitor. The purpose of restricting protein is to reduce the quantity of purines causing the formation of uric acid.

The solubility of the urate crystals is lower at an acidic pH. Diets designed for the dissolution or prevention of urate stones aim to obtain a slightly alkaline urinary pH: between 7 and 7.5. If a slightly alkaline pH is not obtained with the use of a diet alone, potassium citrate may be considered as an alkalinising agent, at an initial dose of 50 mg/kg BW twice a day. Producing a urinary pH higher than 7.5 is not recommended, due to the risk of formation of calcium phosphate stones.

Allopurinol, an inhibitor of the enzyme xanthine oxydase is used at a dose of 15 mg/kg BW twice daily during the dissolution process in order to decrease the production of uric acid. It is important to use allopurinol only in combination with a low-purine diet as an excess of purine precursors in this situation could increase the risk of xanthine stone formation.

Allopurinol is metabolised by the liver and therefore, is not recommended in patients suffering from a portosystemic shunt because prolonged elimination can significantly increase the risk of xanthine stone formation.

On average, urate stones are dissolved in 3.5 months using a combination of dietary modification, alkalisation of the urinary pH and treatment with a xanthine oxidase inhibitor (Bartges et al. 1999). Follow-up by ultrasonography or

double contrast radiography is recommended every 4 weeks in order to ensure the efficacy of the dissolution treatment. If, after 8 weeks of treatment, the size of the stones has not decreased, the diagnosis should be re-evaluated and an alternative therapeutic strategy implemented.

In patients suffering from a portosystemic shunt, correction of the shunt and restoration of adequate hepatic function may be associated with spontaneous dissolution of urate stones (McCue *et al*. 2009).

Once the dissolution (or ablation) of urate stones has been done, it is important to reduce the risk of relapse by feeding a diet reduced in purine and promoting a slightly alkaline urinary pH. In the event of relapse despite the modified diet, long-term therapy with allopurinol should be considered. Regular follow-up on the urinary tract by ultrasonography or double contrast cystography is then recommended due to the risk of xanthine stone formation. As for all types of stones, increasing consumption of water in order to obtain less concentrated urine is a key element of the preventative strategy.

At this time, strategies for dissolution of urate stones have not been established in cats, and surgical ablation of the stones is the option most commonly used.

As for dogs, a low-protein diet promoting a slightly alkaline pH, as well as increased water consumption are recommended to prevent recurrence. Diets developed for management of chronic renal failure, preferably in wet form, are generally adequate for preventing relapse of urate stones in cats.

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Marc Dhumeaux

Management chronic kidney disease in dogs and cats ; what is the evidence?

Brief description of lecture:

Whenever possible veterinary medicine should be based on scientific evidence. In the past three decades a large amount of work has been done focusing on management of chronic kidney disease in small animals. We have now reached a stage where the recommendations made to improve the outcome of canine and feline patients with CKD are evidence-based. In this lecture, we will review the evidence that led the current standards for management of chronic kidney disease in dogs and cats.

Abstract:

Chronic kidney disease (CKD) is a frequent cause of morbidity and mortality in dogs and cats. With the exception of renal transplantation, which is not readily available for small animals, there is no cure. CKD is a self-perpetuating progressive disease process, which at a late stage results in clinical signs of uremia. Our goals when managing CKD in small animals include delaying the progression of the disease, decreasing the risk of uremic crisis, maintaining quality of life and increasing survival. These goals are achievable by following evidence-based recommendations coming from the last three decades of research.

1. IRIS classification of chronic kidney disease

The international renal interest society (IRIS) designed a classification in four stages of dogs or cats with CKD based on their fasting serum creatinine concentration while being normally hydrated (Table 1). This classification is helpful to establish a prognosis for animals with CKD (it is accepted that a higher IRIS stage is associated with a worse prognosis). The classification allows the clinician to make specific recommendations for patient management based on the stage of the disease. It is also useful in clinical research where affected animals can be placed in different study groups based on their IRIS stage. Depending on the outcomes in the various groups, it may be possible to reveal specific features of a given stage, ultimately leading to stage-specific recommendations for patient management.
Because proteinuria and hypertension can worsen the progression of CKD, the IRIS classification takes the level of proteinuria and blood pressure into consideration. The presence and degree of proteinuria and hypertension are both presented as substages in the IRIS classification (Table 2 and 3).

Recently the serum (or plasma) symmetric dimethylarginine (SDMA) was introduced as a useful indicator of early renal insufficiency in dogs and cats. Since the SDMA becomes elevated before the creatinine, it can play a role in identifying patients with CKD before the onset of azotemia (IRIS stage 1). Since 2015, the SDMA was added to the IRIS guidelines for management of CKD in dogs and cats.

Based on the IRIS staging (and substaging) of a patient with CKD, evidence-based guidelines for treatment and monitoring can easily be followed by the practitioner.

	Serum creatinine µmol/L **mg/dL**	
IRIS stage	Dogs	Cats
1	<125 **<1.4**	<140 **<1.6**
2	125 – 180 **1.4 – 2**	140 – 250 **1.6 – 2.8**
3	181-440 **2.1 – 5.0**	251 – 440 **2.9 – 5.0**
4	>440 **>5.0**	>440 **>5.0**

Table 1. IRIS stages of CKD based on serum creatinine concentration in dogs and cats

Urine protein:creatinine ratio		
Dogs	Cats	Substage
<0.2	<0.2	Non-proteinuric
0.2 to 0.5	0.2 to 0.4	Borderline proteinuric
>0.5	>0.4	Proteinuric

Table 2. IRIS substages based on proteinuria in dogs and cats

Systolic blood pressure mmHg	Blood pressure substage	Risk of target organ damage
<150	Normotensive	Minimal
150 – 159	Borderline hypertensive	Low
160 – 179	Hypertensive	Moderate
≥180	Severely hypertensive	High

Table 3. IRIS substages based on systolic blood pressure in dogs and cats

2. Evidence-based recommendations for management of CKD

a. Feeding a renal diet

Compared to maintenance diets, the diets designed for management of CKD in dogs and cats are typically markedly reduced in phosphorus, mildly to markedly reduced in protein, mildly reduced in sodium and supplemented with omega 3 fatty acids and antioxidants.

There is now strong evidence that this type of diet can prolong survival and decrease the risk of uremic crisis when compared to maintenance diets in both dogs and cats with CKD (Elliott *et al.* 2000, Jacob *et al.* 2002, Plantinga *et*

al. 2005, Ross, Osborne *et al*. 2006). It has been shown that while adequate quality of life is maintained, the survival time of canine and feline patients fed a renal diet is on average 2 to 3 times longer than the one of patients fed a maintenance diet.

At this point, we cannot be certain of the role of each component of the diet on the overall benefit. For example, even if it seems widely accepted that phosphorus restriction is beneficial in patients with CKD, there is a controversy about the role of protein restriction. There are concerns particularly in cats about the ability to maintain adequate nutritional status with a diet significantly reduced in protein. However based on the available studies comparing renal diets to maintenance diets in client owned cats, no obvious signs of malnutrition secondary to protein restriction were observed.

When it is not possible to maintain the serum phosphorus within the normal range, despite feeding a diet reduced in phosphorus or when the patient refuses to eat a low phosphorus diet, using a phosphate binder in combination with the diet should be considered. Even if this approach seems very sensible, there is no strong evidence of efficacy of phosphate binders to improve the hyperphosphatemia, decrease the risk of uremic crisis or prolong survival in dogs and cats with CKD.

b. Monitoring and treating proteinuria

In both dogs and cats, proteinuria is known as a negative factor for progression of CKD (Jacob *et al*. 2005, Chakrabarti *et al*. 2012). This is due to the structural damage to the kidney caused by an excessive amount of protein reaching the renal tubule, resulting in further renal dysfunction (Chakrabarti *et al*. 2013, McLeland *et al*. 2015).

The current recommendation is to treat the proteinuria when the urine protein:creatinine ratio is above 0.5 in dogs and 0.4 in cats. ACE inhibitors are most commonly used for management of proteinuria in dogs and cats. More recently the angiotensin receptor blocker telmisartan (Semintra®) was licensed as an alternative (or adjuvant medication) for management of proteinuria in cats.

There is good evidence that treatment with an ACE inhibitor can reduce the proteinuria and delay the progression of CKD in dogs with glomerular disease (Grodecki *et al*. 1997, Grauer *et al*. 2000). In cats, randomized blinded controlled clinical trials using benazepril and telmisartan also showed that treatment could reduce the proteinuria (King *et al*. 2006, Sent *et al*. 2015).

c. Monitoring and treating hypertension

In people with CKD, hypertension is recognized as a negative factor for disease progression and treating the hypertension is of upmost importance as part of the management of a patient with CKD. The rational for treatment of systemic hypertension is to protect the sick kidneys from glomerular hypertension, which can lead to glomerular damage and proteinuria.

Systemic hypertension is common in association with CKD in dogs and cats with 20 to 60% of patients affected in both species depending on the studies.

One study in dogs with CKD showed that hypertension was associated with negative outcomes including uremic crisis and death (Jacob *et al*. 2003). In that study, the dogs with hypertension and CKD were also significantly more proteinuric than the normotensive ones, which could be a confounding factor.

At this point, an association between hypertension and negative outcome of CKD has not been clearly demonstrat-

ed in cats. However, one study in hypertensive cats with or without azotemia showed that treatment of hypertension with amlodipine resulted in a significant decline of proteinuria and increased survival (Jepson *et al*. 2007). These results entertain the belief that there is a strong link between hypertension and proteinuria and that both of them should be managed concurrently.

It is recommended for systemic blood pressure to be monitored in all canine and feline patients with CKD and for patients with persistent hypertension to be treated.

Typical medications for treatment of hypertension in dogs and cats include ACE inhibitors and the calcium channel blocker amlodipine. It is accepted that the effect of ACE inhibitors on blood pressure is minimal but it is the drug of choice for proteinuria, which is frequently associated with sustained hypertension. Amlodipine is much more effective at reducing blood pressure than ACE inhibitors. The dose often needs to be titrated up to achieve a satisfactory control of blood pressure (ideally systolic blood pressure < 150 mmHg).

d. Treating severe anemia

As CKD progresses to end-stage, there is a risk for the patients to become significantly anemic. The anemia of CKD is at least partially due to decreased functional renal mass resulting in decreased production of erythropoietin (EPO) as a stimulus for red blood cell production. Beside the clinical signs associated with profound chronic anemia (lethargy, exercise intolerance, inappetence), the reduced oxygen carrying capacity from anemia may play a significant role in the progression of kidney disease through renal tissue hypoxia.

Treatment with exogenous EPO can be considered in canine and feline patients with CKD when the anemia is severe enough to be responsible for clinical signs due to decreased oxygen tissue delivery. These typically occur when the hematocrit drops below 20 to 22%. The goal with EPO therapy is to reach a hematocrit of at least 25%, which should be associated with a resolution of clinical signs of anemia.

One older study showed that recombinant human EPO (Epogen®) was effective at increasing the hematocrit in dogs and cats with severe anemia secondary to CKD (Cowgill *et al*. 1998). Unfortunately, using this drug in these species appears to be associated with an unacceptably high incidence of anti-EPO antibodies production resulting in pure red cell aplasia. These antibodies would not only react with the exogenous EPO prescribed but would also cross-react with the patient's endogenous EPO, resulting in severe refractory anemia that could only be managed with repeated blood transfusions.

More recently the recombinant human EPO analogue darbepoetin (Aranesp®) has been used with seemingly more success in canine and feline CKD patients with severe anemia. One retrospective study in cats showed significant increase of the hematocrit in more than half of the patients treated (Chalhoub *et al*. 2012). There are anecdotal reports of the use of darbepoetin in dogs and results are also encouraging.

A typical darbepoetin protocol in dogs and cats consists of weekly injections at a dose of 1 µg/kg SC until the target hematocrit (at least 25%) is reached. This usually takes 2 to 4 weeks. Thereafter the dosing interval can be reduced (typically to every 2 to 4 weeks) while ensuring that the hematocrit remains above 25%.

It is commonly reported that darbepoetin is less immunogenic and less likely to cause irreversible pure red cell aplasia than standard EPO. However, to our knowledge, the incidence of antibody production and refractory anemia as a potential side effect of darbepoetin has yet to be evaluated in small animals.

Another potential side effect of EPO therapy is hypertension. Due to the deleterious effects of hypertension in

patients with CKD, close monitoring of systemic blood pressure in patients supplemented with EPO is strongly recommended.

It has been shown that without iron supplementation, human patients treated with EPO could exhaust their iron stores resulting in ineffective erythropoiesis. This is due to the high demand in iron associated with stimulated erythropoiesis. For this reason, and even if iron deficiency is uncommon in dogs and cats with CKD, iron supplementation is recommended when EPO is prescribed. At least an injection of iron dextran should be administered when initiating EPO therapy and further injections should be considered throughout the treatment (usually no more than one monthly) while monitoring the patient's serum iron concentration.

e. Management of appetite and nausea associated with uremia

Common complications of uremia include anorexia and vomiting. Two recent studies in cats with CKD showed encouraging results to improve to appetite and decrease the incidence of vomiting when using the drugs mirtazapine and maropitant (Cerenia®), as an appetite stimulant and an anti-emetic medication, respectively.

The study evaluating the effect of mirtazapine on the appetite of cats with CKD showed that this medication given at a dose of 1.88 mg every other day orally resulted in a statistically significant increase in appetite compared to a placebo. Cats treated with mirtazapine also gained significant weight compared to cats receiving the placebo. Interestingly a statistically significant decrease in vomiting was also observed in the group treated with mirtazapine in comparison to placebo (Quimby and Lunn 2013).

The study evaluating the effects of maropitant on prevention of vomiting was performed in a population of feline CKD patients with a complaint of chronic vomiting. In that study, the group of cats receiving maropitant at a dose of 4 mg orally once daily had significantly less vomiting episodes than the cats receiving a placebo. This study did not show any beneficial effect of maropitant on the appetite (Quimby *et al*. 2015).

Studies of the benefits of the above two drugs in canine patients with CKD are so far not available to our knowledge.

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Mónica Augusto

Adrenal pathologies in everyday small animal practice

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Hyperadrenocorticism (Cushing's disease) and hypoadrenocorticism (Addison's disease) are the most frequent canine adrenal pathologies in small animal practice. The basic pathophysiology, diagnostic approach and current treatment options will be reviewed. Moreover, differences between feline and canine patients will be highlighted.

1. Introduction

Hypothalamic-pituitary-adrenal (HPA) axis: The hypothalamus, by secreting corticotropin-releasing hormone (CRH), has a control over the secretion of adrenocorticotropic hormone (ACTH) by the anterior section of the pituitary gland. In turn, ACTH stimulates adrenocortical secretion of cortisol. Cortisol closes the cycle by inhibiting secretion of hypothalamic and pituitary hormones – negative feedback mechanism. The main hormones secreted by the adrenal cortex are cortisol, corticosterone and aldosterone, whilst the adrenal medulla is responsible for the production of catecholamines. Both CRH and ACTH are secreted in pulsatile and episodic manner. ACTH primary function is to stimulate glucocorticoid secretion from the adrenal cortex. Whilst stimulation of adrenocortical mineralocorticoids is mediated primarily by renin-angiotensin-aldosterone-system (RAAS) and serum potassium concentration[20].

Histologically, the adrenal cortex is composed by three zones: zona glomerulosa, zona fasciculata e zona reticularis. All the zones can synthetize corticosterone but due to enzymatic differences between the zona glomerulosa and the other two inner layers, the adrenal gland functions as distinct units: zona glomerulosa produces mainly aldosterone, whereas the zona fasciculata secrets mostly glucocorticoids and the zona reticularis sex hormones (androgens and oestrogens)[20].

2. Pathophysiology and pathogenesis

"Cushing's disease" applies to cases in which hypercortisolism occurs secondary to inappropriate and excessive secretion of ACTH by the pituitary gland, and subsequently causes bilateral adrenal gland hyperplasia, i.e. pituitary-dependent hyperadrenocorticism (PDH). More than 90% of patients with PDH have a pituitary tumour (either micro or macroadenoma)[20].

Additional pathophysiologic classification of hyperadrenocorticism (HAC) include: autonomous secretion of cortisol by functional adrenocortical tumour (adenoma or carcinoma); iatrogenic HAC due to exogenous glucocorticoid administration; secretion of ACTH from ectopic tissue; food-dependent cortisol secretion and in humans also pituitary hyperplasia caused by excessive CRH secretion due to a hypothalamic disorder (not yet reported in dogs or cats)[20].

Primary functional adrenocortical tumours (FAT) secrete excessive amounts of cortisol, and occasionally corticos-

terone and desoxycorticosterone, independent of pituitary control and in an intermittent and random way. Because hypothalamic CRH and circulating ACTH concentrations are suppressed, the contralateral adrenal gland is usually atrophied[20].

Cats have fewer glucocorticoid receptors in their skin and liver and lower binding affinity compared to dogs, which decreases their susceptibility to adverse glucocorticoid effects and development of iatrogenic HAC. However, long-term use of glucocorticoids causes insulin resistance, and approximately 80% of cats are presented with concomitant, poorly regulated diabetes mellitus[14]. The effect of excessive glucocorticoids on the skin is more severe in cats, who are normally seen with skin fragile syndrome. Naturally occurring HAC is an infrequent diagnosis in cats, but similarly to dogs, 80% of cats have PDH, while the remaining 20% have FAT, of which one-third is malignant[14].

On the opposite spectrum of adrenal gland pathologies, there is typical "Addison's disease" or hypoadrenocorticism, which refers to the lack of adrenocortical hormones (glucocorticoids and mineralocorticoids). However, up to 30% of dogs only have glucocorticoid deficiency (hypocortisolism or "atypical' hypoadrenocorticism") and these dogs have no electrolyte abnormalities (i.e. aldosterone concentration is unaltered)[25].

Hypoadrenocorticism is believed to have a genetic basis in Bearded Collie, Portuguese Water Dog, Standard Poodles, Nova Scotia duck tolling retrievers, soft-coated wheaten terriers and Pomeranians and suspected familial predisposition in Leonbergers, Labrador Retrievers, and other breeds[8,9,10,13,22,28].

The most common cause of primary hypoadrenocorticism is an immune-mediated adrenalitis, frequently lymphoplasmacytic inflammation, which leads to adrenocortical atrophy[26]. Rapid withdrawal of exogenous corticosteroids (any dose and/or length of treatment that suppressed HPA axis) can also lead to secondary adrenal atrophy, but in this case, it's a reversible process[25].

Other causes of primary hypoadrenocorticism include granulomatous inflammation due to fungal disease, amyloidosis, haemorrhagic infarction, iatrogenic disease (e.g. mitotane or trilostane administration) and metastatic neoplasia[17,25,26]. Reduced secretion of CRH by the hypothalamus and/or ACTH by the pituitary gland also leads to a decrease in synthesis of adrenocorticoidal hormones, particularly glucocorticoids but is encountered only rarely (secondary hypoadrenocorticism)[25,26].

Cortisol deficiency affects cellular energy metabolism, vascular sensitivity to catecholamines, glycaemic control (gluconeogenesis and glycogenolysis), leading also to dullness and weakness. On the other hand, aldosterone deficiency triggers sodium, chloride and water loss by the kidneys, plus potassium and hydrogen retention. In turn, progressive hyponatraemia instigates a decrease in circulating blood volume, cardiac output, perfusion and impaired renal concentration ability[25].

Hypoadrenocorticism is uncommon in cats but most reported cases have immune-mediated primary adrenal failure. Cases of bilateral adrenal infiltration due to lymphoma, and adrenal failure secondary to trauma have also been documented[24,25].

3. Diagnostic approach

The primary indication for performing diagnostic tests in patients with suspected adrenal disease is the presence of one or more clinical signs and/or physical examination findings consistent with hyper- or hypofunction of the adrenal gland[5]. Though recently, patients are also screened for FAT because an adrenal mass has been found incidentally on abdominal imaging.

Low Na: K ratio and lack of a stress leucogram in a sick dog (especially a normal lymphocyte count and eosinophilia) should raise the index of suspicion for hypoadrenocorticism. However, patients with only hypocortisolism (atypical hypoadrenocorticism) don't have the classical electrolyte abnormalities (hyponatraemia and/or hyperkalaemia). Because the development of "clinical syndrome" requires at least 90% loss of the adrenocortical function, it is usually a gradual process, and often in the beginning signs are only perceptible in times of stress (e.g. surgery, infection, trauma etc.)[25,26]. Other patients are presented for the first time in a metabolic crisis without a triggering factor (Addisonian crisis). It is therefore important to consider hypoadrenocorticism as possible differential for any patients with vague clinical signs[26].

3.1. Hyperadrenocorticism

In the authors opinion, HAC is likely over-diagnosed because of the high number of false-positive results due to non-adrenal illness. Therefore, endocrine screening tests for HAC should not be performed in patients that just have an increase in ALKP on routine biochemistry without other compatible clinical signs or patients that are systemically unwell.

Screening tests for hyperadrenocorticism include urine cortisol: creatinine ratio (UCCR), low-dose dexamethasone suppression test (LDDST) and/or adrenocorticotropic hormone (ACTH) stimulation tests.

UCCR: Urine sample should be collected at home, ideally in the morning, and either before or at least two days after the visit to the clinic. UCCR has a very high sensitivity (99-100%) but very low specificity (20-25%, which increases with pool of 2-3 samples)[5]. UCCR is also affected by non-adrenal illness, so a positive UCCR should be followed with another screening test.

LDDST is the screening test of choice for HAC, apart when iatrogenic HAC is suspected. Serum or plasma cortisol concentrations are determined before, 4 hours and 8 hours after administration of dexamethasone, at dose of 0.01-0.015 mg/kg IV. Lack of suppression on LDDST is consistent with diagnosis HAC (cortisol level at 8h ≥ lab-cut-off). LDSST has sensitivity 85-100% and specificity 44-73%[5].

In cats, higher dose of dexamethasone is usually required to suppress cortisol levels so dose of 0.1 mg/kg dexamethasone IV is given but interpretation of the test results is similar to dogs[14].

ACTH Stimulation Test has lower sensitivity compared to LDDST but it is quicker to perform and is less affected by non-adrenal illness. It remains the gold standard for diagnosis of iatrogenic HAC. Serum or plasma cortisol concentrations are determined before and 1 hour after administration of 0.5 ug/kg of synthetic ACTH. Exaggerated cortisol production is consistent with diagnosis of HAC. ACTH stimulation test has sensitivity 57-95% and specificity 59-93%[5].

Tests to differentiate adrenal-dependent from pituitary-dependent disease include LDDST, High-Dose Dexamethasone Suppression Test (HDDST), plasma endogenous ACTH (e-ACTH) and imaging (abdominal ultrasonography, pituitary CT or MRI).

LDDST: PDH is suspected when cortisol level at 4h < lab-cut-off and/or 4h and/or 8h cortisol level <50% baseline but >lab cut-off. Patients who fail to meet any of the suppression criteria (i.e. cortisol is high at all three times) could either have PDH or adrenal-dependent disease6. The dexamethasone suppression test is recommended over ACTH stimulation for initial screening of cats with suspected HAC[14].

HDDST: Cortisol suppression after administration of 0.1 mg/kg of dexamethasone is consistent with diagnosis of

PDH (cortisol level at 4h or 8h < lab cut-off and/or <50% baseline). However, like LDDST lack of suppression could be either PDH or adrenal-dependent disease[5,6,20].

e-ACTH: Samples need to be collected into chilled silicon-coated glass or plastic tube with EDTA, and centrifuged within 15 minutes, transferred to plastic tube and frozen immediately (plasma proteases degrade e ACTH). Patients with PDH have normal to elevated ACTH levels, while patients with adrenal-dependent disease have low to undetectable ACTH levels[6].

Diagnostic Imaging: Routine adrenal gland ultrasonography, CT or MRI can be used to differentiate PDH from adrenal dependent disease.

3.2. Hypoadrenocorticism

Aldosterone deficiency leads to hyponatremia, hypochloraemia and hyperkalaemia. So, the Na:K ratio is typically lower than the normal (<28). In addition, the lack of stress leucogram, particularly normal lymphocyte count, in a sick dog with consistent clinical signs should raise the suspicion for hypoadrenocorticism.

Endocrine tests for hypoadrenocorticism include basal cortisol concentration, ACTH stimulation test, measurement of endogenous ACTH (e-ACTH) and plasma aldosterone concentration (PAC).

Basal cortisol concentration ≤ 55 (2ug/dL) has sensitivity of 100%, which makes this a very useful test to rule out hypoadrenocorticism[15,18]. However, a low basal cortisol should always be followed by ACTH stimulation test.

ACTH stimulation test is the gold standard to diagnose hypoadrenocorticism. Most dogs have values < 27.6 nmol/L (1ug/dL) on both the pre- and post- ACTH samples[25,26].

e-ACTH: Patients with primary hypoadrenocorticism have markedly increased ACTH levels (>100 pg/mL) due to the lack of negative feedback; whilst patients with secondary hypoadrenocorticism usually have undetectable e-ACTH (<20 pg/mL)[26].

PAC: pre- and post- ACTH stimulation test PAC measurement should be performed in animals with hypocortisolism and normal electrolytes ("atypical hypoadrenocorticism"). Normal aldosterone rules out mineralocorticoid deficiency but an abnormal result is inconclusive (optimal dose of ACTH and peak secretion has not been determined yet)[6,27].

4. Current treatment options

4.1. Hyperadrenocorticism

Apart from symptomatic supportive treatment, specific therapy for HAC include medical therapy or surgery (treatment of choice for FAT). Transsphenoidal hypophysectomy has been successfully performed in dogs and cats[21], and in cases of macroadenomas, radiotherapy would be another treatment option.

Trilostane is a synthetic, competitive inhibitor of the 3β-hydroxysteroid dehydrogenase, blocking primarily cortisol production[23]. Current recommended starting dose of trilostane range from 1-2.5 mg/kg PO q 24 hrs, and adjusted per clinical signs and ACTH stimulation test results. Several studies suggest that administration of trilostane twice daily at low doses (0.5-1 mg/kg PO q 12 hrs) may provide superior clinical control and cause fewer side effects compared to higher, once daily dosing[2,3].

There is limited veterinary literature in regards to proven efficacy of trilostane therapy in cats [7,21,23] but surgical therapy appears to be the best option for cats with FAT[12].

4.2. Hypoadrenocorticism

Patients with hypoadrenocorticism need lifelong therapy, which can be either:

Desoxycorticosterone pivalate (DOCP) is a long acting ester of desoxycorticosterone acetate (DOCA). Current recommended starting dose (2.2mg/kg SC every 25 days) which usually can be reduced in most dogs. DOCP has also been successfully for the management of hypoadrenocorticism in the cat[30]. To adjust the dose, electrolytes should be monitored on day 10-12 day. If K is high, the next dose is increased by 10%. To adjust frequency, electrolytes should be rechecked just before the next injection. If K is low, or normal mid-range, increase the next dose interval by 2 days. If K is high, decrease the dose interval by 2 days. To avoid weekend injections, it's easier to administer DOCP every 28 days. Careful monitoring of electrolytes is advised, especially in the first 2-3 months.

Physiological dose of prednisolone (0.1-0.2mg/kg/day) should be given together with the DOCP injections, and tapered to the lowest dose possible. It is important to avoid excess prednisolone supplementation as medication is life-long and often dogs develop signs of hyperadrenocorticism later in life. However, during periods of stress (i.e. boarding, disease, elective surgery, etc.), this dose may have to be increased.

Fludrocortisone acetate is an oral synthetic mineralocorticoid that also has an intrinsic glucocorticoid activity, therefore additional prednisolone is not necessary, apart from during periods of stress. Fludrocortisone should be started on the low recommended dose and titrated upwards, depending on clinical signs and Na and K values (15-20 µg/kg P0 q 24 hr or 0.1-0.5 mg/dog q12-24h).

In cats with hypoadrenocorticism, fludrocortisone acetate at 0.1 mg/day or a starting dose of DOCP (12.5 mg/once a month) have been prescribed but clinical signs such as anorexia, lethargy, and weakness may sometimes take longer to resolve than in dogs[26,30].

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Rare adrenal gland pathologies in small animal practice

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Phaeochromocytoma and feline primary hyperaldosteronism (Cohn's Syndrome) are rather infrequent diseases in small animal practice involving the adrenal glands, which might be underdiagnosed. A basic understanding of these pathologies will therefore prompt the clinician to recognize potential cases and advise on available treatment options.

1. Introduction

Phaeochromocytoma is a rare catecholamine-secreting neuroendocrine tumour. Secretion of excessive amounts of catecholamines is usually sporadic and produce a variety of non-specific clinical findings including systemic arterial hypertension, weakness, arrhythmias, collapse or even sudden death in cases of acute, sustained release of catecholamines. Because clinical signs can be vague and episodic, the diagnosis should also be considered in animals with a unilateral or bilateral adrenal mass even without overt clinical signs[1]. Phaeochromocytoma is rare in cats with only a few case reports published in the veterinary literature[4,6,12,22].

Conversely, primary hyperaldosteronism (Conn's syndrome), which is an adrenocortical disorder characterized by an autonomous hypersecretion of aldosterone, is seen more frequently in cats than dogs. Hyperaldosteronism causes sodium and water retention and increases potassium excretion by the kidneys, leading to systemic arterial hypertension and significant hypokalaemia[24].

2. Pathophysiology and pathogenesis

2.1. Phaeochromocytoma

The adrenal medulla is composed of postganglionic neurons (chromaffin cells) which are part of the sympathetic nervous system and produce, store, and secrete a variety of neurotransmitters and neuropeptides[9]. The primary secretory products are catecholamines - dopamine, norepinephrine and epinephrine.

Catecholamines are synthesized from the amino acid tyrosine by a series of enzyme modifications. One important rate-limiting step in this pathway is the initial conversion of L-tyrosine to L-DOPA by tyrosine hydroxylase, because in normal dogs, high cytoplasmic concentrations of norepinephrine inhibit the activity of tyrosine hydroxylase, controlling catecholamine production – negative feeding back mechanism.[14] In normal dogs, there is also a high concentrations of a methylating enzyme (phenylethanoalamine-N-methyltransferase) PNMT), which converts norepinephrine to epinephrine and can be induced by cortisol. Finally, two main hepatic enzyme systems, catecholamine-O-methyltransferase (COMT) and monoamine oxidase (MAO), mediate the catabolism of catecholamines to metanephrine, normetanephrine, and vanillylmandelic acid, which are excreted in the urine. In cases of pheochromo-

cytomas, the negative feeding back mechanism is lost so the metabolism of catecholamines may be increased[14].

Catecholamine secretion increases in response to physiologic stressors (such as hypotension, hypoglycaemia, hypoxia) but secretion is variable and unpredictable, and may be induced by other factors including certain drugs (e.g. metoclopramide and steroids) in cases of phaeochromocytoma. Tissue response to catecholamines depends on the number and type of catecholamine receptors on the cell membrane. However, α-1 adrenergic receptors predominate when concentration of plasma catecholamines are increased, resulting in vasoconstriction and hypertension[14].

2.2 Hyperaldosteronism

Aldosterone is a potent mineralocorticoid produced by the zona glomerulosa of the adrenal cortex. Its secretion is regulated by the renin-angiotensin-aldosterone system (RAAS), potassium concentration, and to a much lesser extent by adrenocorticotropic hormone (ACTH)[24]. In the kidneys, aldosterone causes transfer of sodium in exchange for potassium and hydrogen in the distal tubule.

Renin-angiotensin-aldosterone system: RAAS maintains extracellular fluid volume, circulatory pressure and electrolyte homeostasis. The release of renin by the juxtaglomerular cells is mediated by different mechanisms including: baroreceptors in the afferent arteriole of the renal glomeruli to decreases in perfusion pressure; sodium content in the glomerular filtrate and activity of sympathetic neurons linked to cardiac baroreceptors. Renin in circulation cleaves angiotensinogen to angiotensin-I, which in turn is converted to angiotensin-II. Angiotensin II mediates vasoconstriction, promotes renal tubular reabsorption of sodium and stimulates the release of aldosterone from the adrenal gland (secondary hyperaldosteronism). RAAS is usually activated as compensatory response to heart failure, gastrointestinal disease, water deprivation or other causes of hypovolaemia[24].

Primary hyperaldosteronism (PHA) is typically caused by a unilateral adrenocortical tumour (adenoma or carcinoma) although idiopathic bilateral micronodular hyperplasia has also been reported[15,24]. Cats with aldosterone-secreting adrenal tumours can also have concurrent production of other corticosteroids such as excessive amounts of progesterone and signs of HAC[2]. Aldosterone has also been associated with progression of kidney disease and given that chronic renal disease is relatively common in older cats, it should be considered as a differential[16].

3. Diagnostic approach

3.1. Phaeochromocytoma

Because the clinical presentation of phaeochromocytoma depends on the type, amount and frequency of catecholamine secretion, patients can be asymptomatic or arrive with life-threatening hypertensive crisis. Animals are considered hypertensive when systolic pressure is persistently >160 mm Hg, diastolic pressure >100 mm Hg, or both. Even so, failure to document systemic hypertension in a dog with appropriate clinical signs and/or adrenal mass does not rule out the diagnosis[14,25].

Many abnormalities in routine blood and urine tests are caused by concurrent disorders that are commonly existent in dogs with phaeochromocytoma. Proteinuria, due to hypertensive glomerulopathy or concurrent illness, is found in approximately 20% of dogs[10]. Hyposthenuria and isosthenuria is usually identified due to the inhibitory effect of circulating catecholamines on vasopressin secretion and activity[14]. Abdominal imaging is fundamental in the diagnosis of pheochromocytoma. The most common finding is an adrenal nodule or mass, with a normal-sized contralateral adrenal gland. Abdominal ultrasonography or computed tomography (CT) provides also important information regarding local tumour invasion (commonly to caudal vena cava, phrenicoabdominal vein or kidney) and thoracic im-

aging (three-viewed inflated thoracic radiographs or ideally CT) helps to assess the presence of distant metastatic disease[27].

Endocrine testing for phaeochromocytoma include measurement of plasma or urinary catecholamines and their metabolites. These tests have become more widely available, increasing the number of cases diagnosed ante-mortem. However, its interpretation is not always simple. Dogs with phaeochromocytoma have significantly higher concentration of plasma metanephrines and normetanephrines and higher concentration of urinary epinephrine, norepinephrine, metanephrines and normetanephrine-to-creatinine ratio than healthy dogs[11,18,19,23,26]. However, some of those results may overlap between dogs with phaeochromocytoma, hyperadrenocorticism and non-adrenal disease. Based on current published studies, urinary normetanephrine concentration and urinary normethanephrine to creatinine ratio appear the most useful endocrine diagnostic tests. In neutered dogs with adrenal tumors an undetectable serum concentration inhibin would also support a diagnosis of phaeochromocytoma[3]. In cats, plasma normetanephrine was significantly higher in sick cats with non-adrenal disease compared to healthy cats and markedly higher in the cat with a suspected phaeochromocytoma[28] but available literature is limited.

A definitive diagnosis of phaeochromocytoma requires histopathology and immunohistochemistry (positive chromogranin A staining) but differentiation between benign and malignant tumors remains difficult and might have to be based on the tumour biological behaviour instead[14,25].

3.2 Hyperaldosteronism

Primary hyperaldosteronism should be suspected in any cat presented with systemic arterial hypertension, hypokalaemia and/or chronic kidney disease (which would be also the main differential diagnosis), especially if phosphate concentration is low-normal[8,16,21]. Cats are typically presented with muscle weakness due to hypokalaemic polymyopathy (commonly seen as cervical ventroflexion and hind limb ataxia) and/or complications of systemic hypertension (e.g. sudden onset of blindness due to retinal detachment or intraocular haemorrhage). Other less common signs include polyuria, polydipsia, polyphagia and presence of systolic heart murmur or left ventricular hypertrophy[24].

Ideal screening test for primary hyperaldosteronism include ratio between plasma aldosterone concentration (PAC) and plasma renin activity (PRA), i.e. aldosterone: renin ratio (ARR). The combination of a high-normal or elevated PAC and a low PRA is consistent with increased aldosterone secretion, given the low or no RAAS stimulation[15,16]. Measurement of PAC alone must be interpreted carefully to avoid false positive results due to an appropriate physiological response to renin, and in conjugation with concurrent documented hypokalaemia. Even so, interpretation of results can be difficult as low PAC can sometimes be seen in cats with primary hyperaldosteronism, in particular those with bilateral micronodular hyperplasia; additionally, because of expected fluctuations in secretion of both hormones, a single ARR value within normal range does not exclude the diagnosis either[16].

An alternative would be to measure aldosterone in the urine which reflects aldosterone secretion over a longer period of time, and bypasses the need to measure renin concentration simultaneously[7]. However, urinary aldosterone: creatinine ratio might be within normal limits even in cats with hyperaldosteronism and therefore a suppression test with fludrocortisone would be necessary to prove independent secretion of aldosterone[8]. Moreover, medications that might affect RAAS need to be discontinued before preforming the test (e.g. spirolactone, ACE inhibitors, angiotensin-receptor blockers such as telmisartan, diltiazem, amlodipine etc.). Unfortunately, most cats with primary hyperaldosteronism have severe systemic arterial hypertension and discontinuing some of these anti-hypertensive medications could be detrimental and not indicated for diagnostic purpose alone[8,17]. From a practical point of view, often only baseline aldosterone concentration is measured. Demonstration of high aldosterone (>1000 pmol) in a cat with hypertension, hypokalaemic and presence of an adrenal mass would be consistent with diagnosis PHA[24]. However, in cats with idiopathic micronodular hyperplasia (who have normal adrenal glands) ARR would be preferred.

Lastly, if primary hyperaldosteronism is suspected or confirmed, abdominal and thoracic imaging should be pursued, either by means of abdominal ultrasonography and three-viewed inflated thoracic radiographs or computed tomography. Imaging helps with surgical decision making as it provides important information about the affected adrenal gland, possible extension into caudal vena cava but also metastatic disease.

4. Current treatment options

Adrenalectomy is the treatment of choice for non-metastatic unilateral adrenocortical tumours, despite the high risk of complications and mortality rates. In general, all patients should be stabilized medically prior to surgery by controlling systemic arterial hypertension and in cases on PHA also the existing hypokalaemia.

In cases of phaeochromocytoma, adrenalectomy is associated with high risk of hypertensive and hypotensive crises, cardiac arrhythmias and haemorrhage. In the dog with normal adrenal function, induction of anaesthesia may lead to changes in blood pressure that in turn stimulate catecholamine secretion. In a dog with a phaeochromocytoma, this response is often exaggerated, causing severe hypertension and life-threatening tachyarrhythmias[13,20]. Excessive catecholamine secretion also occurs during manipulation of the tumor during surgery therefore γ -adrenergic blockade through administration of phenoxybenzamine (starting dose of 0.25-0.5 mg/kg BID PO and slowly increased to 1-2 mg/kg for approximately two weeks prior to surgery) should be instigated to reverse vasoconstriction, hypovolaemia and control fluctuations of blood pressure and heart rate during general anaesthesia and to decrease perioperative mortality[13,14,25].

For cats with PHA without a visible adrenocortical tumour (e.g. cats with idiopathic bilateral micronodular hyperplasia), non-resectable tumours, evidence of metastatic disease or when surgery is not an option for any other reason, medical therapy should be continued long-term with a combination of spirolactone (competitive aldosterone receptor antagonist), potassium supplementation and anti-hypertensive medication such as amlodipine besylate[24]. The prognosis for cats with PHA that undergo adrenalectomy, and survive the immediate peri- and postoperative period, is typically good with no need for ongoing therapy[21]. In cases where surgery was not possible, the prognosis is more variable and depends on response to medical management. In some cases, hypertension becomes refractory to medical management and cats more commonly die from progressive kidney disease or thromboembolic event[24].

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Nikola Katic

Acute Abdomen – When to Cut?

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"Good surgeons know how to operate, better ones when to operate and the best when NOT to operate!"

Acute abdomen is a collective term for a number of conditions that are presented with clinical signs of mild to marked abdominal discomfort, abdominal distension, generalised gastrointestinal signs and/or shock. Some of these may necessitate an immediate surgical correction, however, in others one is better off with an appropriate conservative treatment or postponed surgical intervention.

Generally, one or more organs or systems can be involved in the etiology of acute abdomen and the list of differentials is rather long. Due to plethora of possible causes management of cases with acute abdomen is often very challenging. Proper history, physical examination, collection of free abdominal fluid, ultrasonography and radiology are minimal diagnostic setup for such cases.

History

It should be taken very carefully and placed in the frame of the patient (sex, age, breed...). Specific set of questions should be posed: everyday's habits of the animal, unusual events (trauma, fight...), contact with other animals, any medical or surgical treatments (e.g. previous surgery), presence of vomitus, diarrhoea, melena, polyuria, stranguria...

Physical examination

Nothing can substitute a thorough physical examination and it must not be omitted. Manual palpation of the abdomen is inevitable when acute abdomen is suspected. Superficial palpation should be performed before deep abdominal palpation and can help differentiate causes such as abdominal herniations. Deep palpation is useful in localising pain within the abdomen (e.g. right cranial quadrant is painful in cases with pancreatitis). In some cases deep palpation may not be possible due to splinting of the muscles of the abdominal wall which is a consequence of a reflex rigidity due to peritoneal irritation and pain. In cases of severe abdominal effusion a "wave" is produced on abdominal ballottement. Apart from palpation visual inspection (e.g. Cullen sign - red discoloration around the navel in haemoabdomen), auscultation (presence of peristalsis...), percussion and rectal examination should be performed.

Pain in acute abdomen cases can have different manifestation. Seriousness of the condition usually cannot be judged solely upon it. Some patients are extremely painful while other might express not more than an abdominal discomfort or tensing on palpation of affected area even in the presence of a septic peritonitis. This is even more true for cats.

Abdominocentesis and Diagnostic Peritoneal Lavage

It is a quick and highly specific test. As less as 5ml/kg of free fluid in abdomen are sufficient for adequate sample collection. In smaller amounts a peritoneal lavage or ultrasound-guided abdominocentesis might be indicated. To increase the chance of retrieving sufficient sample four-quadrant abdominal tapping is used. One should keep in mind that a small amount of free air might cause an artefact on plain abdominal radiography if performed after abdominocentesis.

The fluid should be analysed (grossly, cytologically, biochemically and microbiologically) after the collection and can help immediately determine whether or not surgical intervention is indicated:

- PCV that is as high/low as peripheral and follows it on the succeeding tap is a sign of ongoing haemorrhage. Immediate transfusion is started and if these attempts fail to stabilize the patient surgery is indicated. Pressure bandage around abdomen might help control intraabdominal bleeding. It is essential to differentiate hemoabdomen from inadvertent puncture of a vessel or spleen. The main difference is that intraperitoneal blood does not clot!
- Presence of bacteria (intra/extracellular), foreign material, or degenerated neutrophils which are the predominate population of leukocytes are clear indications for exploratory coeliotomy due to suspected septic peritonitis
- If septic peritonitis is suspected, but bacteria are not seen on the smear an exploratory coeliotomy is warranted if:
    - Glucose < 50mg/dL in the tap may indicate septic peritonitis (false positive if samples are not immediately processed)
    - Difference between glucose in the tap and peripheral blood of > 20mg/dL will rise a suspicion of a septic peritonitis; however, false negatives are possible in gastric perforation, very acute peritonitis or localised infection
    - Lactate concentration > 2,5 mmol/L is a good indicator for septic peritonitis in dogs, however this is not useful in cats
    - Blood to fluid lactate below -2,0 mmol/L (sensitive in dogs but not in cats)
- If creatinine in the tap as 2 times and/or potassium 1,4 times greater than in the peripheral blood the diagnosis of uroabdomen is made. However, this is still not indication for an immediate surgery. Urodiversion can be done via indwelling urinary catheter or minimal invasively placed prepubic cystostomy catheter. There is a good chance that smaller leaks will heal on their own. An emergency surgery is hardly ever needed. Furthermore, it is contraindicated in hyperkalaemic and uremic patients; a previous stabilisation is obligatory. Urine is usually sterile and if needed surgery is done in stable patient. If urinary infection is present a septic peritonitis is concerned and should be treated as such.
- Leakage from the biliary system will cause a bile peritonitis. Concentration of bilirubin in the abdominal tap higher than in the peripheral blood is a valuable indicator. Presence of bile crystals will confirm the diagnosis. Bile peritonitis will take some time to develop, however, not so in cases with infected bile peritonitis. Surgical correction of bile leakage is obligatory in any case, however, patients should be stabilised before.

Laboratory findings

Once a patient with acute abdomen is stabilised further evaluation is performed. Full chemistry panel, complete blood cell count, electrolyte, blood gas and urine analysis, coagulation profile, and upon need, special tests (e.g. pancreatic amylase and lipase in suspicion of a pancreatitis) are done. Bacteriological sampling should not be omitted.

Imaging

Abdominal radiography is a standard tool for evaluation of most patients with acute abdomen. It is easily and quickly performed and can give valuable information whether surgical intervention is warranted. GDV, organomegaly, masses, obstruction, foreign body, free gas, effusion calculi, herniations can be recognised on plain radiographs. As spinal pain may mimic abdominal pain plain radiography sometimes help differentiate between the two (e.g. IVDD, diskospondilytis can be recognised on survey radiographs). Contrast radiography can also add some information in certain cases. If a GIT perforation is suspected use of organic iodine contrast is indicated as it is less irritating than free barium in the peritoneal cavity. Pneumoperitoneum can be seen on radiographs in case of perforation of the abdominal cavity (e.g. bite wounds, penetration wounds), perforation of the GIT or if gas producing bacteria are present within the abdominal cavity. Pneumoperitoneum is a clear indication for exploratory coeliotomy, however, one should keep in mind that free air might be present in the abdomen up to 2 weeks after previous surgery, or if abdominal tap is performed prior to abdominal radiography, thus this finding should be interpreted with caution. Thoracic radiographs as a standard part of the assessment of trauma or oncologic patients may reveal other conditions that need to be addressed surgically (e.g. diaphragmatic hernia) or may otherwise influence prognosis and treatment (e.g. severe lung contusion or presence of metastasis).

Abdominal ultrasonography is a very useful tool and may often lead to a definitive diagnosis. It is noninvasive, can help characterize intraabdominal organs, facilitate collection of free abdominal fluid and guide fine needle aspiration of affected organs. It is, however, operator dependant. FAST (Focused Assessment with Sonography of Trauma) has become standard of care in human emergency units. This portable ultrasound test is also applied in veterinary medicine for triage of trauma patients and has proven useful for detection of free abdominal fluid, however with some limitations and need of centesis for definitive confirmation. Serial examinations are recommended to increase the sensitivity of this test.

Advanced imaging (CT, MRI, Scintigraphy…) have already found their application in veterinary medicine, however, they are not as routinely used in assessment of the acute abdomen in small animal surgery as it is the case in human medicine.

This overview has not exhausted all possible scenarios and should only serve as an exemplary guide. Apart from few clear indications for immediate exploratory or corrective abdominal surgery making decision when to operate in case of an acute abdomen is rather a process. One should aim at assessing as many factors as possible within as short time as possible while processing the available information in the frame of the patient/owner. While doing so it is prudent to rely on available medical evidence that is relevant for that particular case. Not for every patient shall surgery be beneficial, however, "it takes wisdom, experience, strength and courage not to intervene. The minute that a surgeon cuts the skin harm is done. The benefit of a treatment will have to exceed that harm before the doctor is doing good"*.

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Nikola Katic

Evidence-based medicine - how to search for scientific evidence

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In the most ideal setting every patient would obtain the most optimal treatment for its condition(s) and eventually re-establish the health as it used to be before the problem(s) occured. Although the ultimate intent of every physician is to achieve this goal many issues are on their way while creating an appropriate treatment in a clinical setting. Establishing the"rightest treatment" for the patient is a process. At the beginning of the twentieth century Sir William Osler, a human doctor, put this into the following thoughts:

'Medicine is practice of an art which consists largely in balancing possibilities... It is a science of uncertainty and an art of probability ... Absolute diagnoses are unsafe and made at the expense of conscience'

Indeed, several strategies are at our hand for tailoring the most ideal plan for our patients. The simplest strategy relies on our previous experiences. It is so called "the art of medicine" which is intuitive and often unconcious. The decision making process is "expert-like", however many decisions made this way are not much more than an educated guess and though this may be sufficient in a number of cases some patients will eventually become inadequate treatment. This should never be used as the only principle and it should at best be combined with the data from basic and clinical research at disposal for the particular condition.

The term evidence-based medicine emerged in the 80's and indicated an urge to apply medical research into clinical decision making. It suposes that the best available evidence should be consciously incorportated into decision making process while heading through diagnostics towards the final treatment. This combination of personal experience/expertise with external best evidence from scientific research, and values/expectiations of client/patient is known as *triad of evidence based practice*. *Five steps* have to be followed while applying this into the daily clinical reasoning:

1. *Ask* focused clinical question
2. *Acquire* evidence
3. *Appraise* evidence
4. *Apply* evidence (through the triad of evidence based practice)
5. *Assess* evidence application

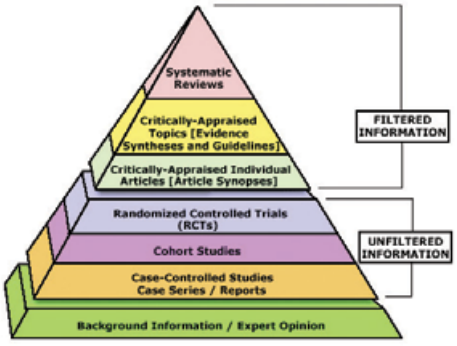
It soon became clear that the evidence at physician's disposal was not of the same quality so that different systems to classify it have been created. Most commonly used system is a graphical representation of *evidence* in form of a *pyramid* (see figure). The information with the weakest evidence is found in the sources that are placed at the base the pyramid. The evidence becomes stronger towards the top of it. Information of the least quality are expert opinions, ideas and in vitro research ("tube tests"). Validation of these concepts are rather vague and may even mislead a clinician. These are followed by case reports and case series which usually provide some information which though relevant for that particular patient or patients might not prove applicable to another cases. Retrospective studies are usually mastered by means of inclusion or exclusion criteria, still the valuability of this data is at best limited.

Structured reviews are next on the scale and might represent a valuable source of information provided that the author critically analyses data based on well known guidelines for that type of research. Prospective clinical trials, that are double/triple blinded, randomised and include control group (placebo) are certainly of a decently high value when integrated into clinican's decision making proces. At the very top of the evidence based pyramide are meta-analyses which are structured evaluations of the available evidence that are conducted according to the very strict guidelines.

Unfortunately, most of the data in veterinary medicine does not achieve the highest level of evidence, still this must not be used as an excuse for not integrating it into the clinical reasoning. It is certain that for a fairly big number of conditions in veterinary medicine the evidence at one's disposal is rather low, however although not perfect it is ***the best scientific evidence there is***. One should be able to differentiate between different source of information and critically evaluate evidence and its application for the presented case. After the evaluation of the quality of research (bias, trial design, population size, generalizability...) it is obligatory to classify the evidence upon the ***risk/benefit assessment*** for an individual patient or group of patients as:

- ***Level A:*** Good scientific evidence suggests that the benefits of the clinical service substantially outweigh the potential risks. Clinicians should discuss the service with eligible client/patients.
- ***Level B:*** At least fair scientific evidence suggests that the benefits of the clinical service outweighs the potential risks. Clinicians should discuss the service with eligible client/patients.
- ***Level C:*** At least fair scientific evidence suggests that there are benefits provided by the clinical service, but the balance between benefits and risks are too close for making general recommendations. Clinicians need not offer it unless there are individual considerations.
- ***Level D:*** At least fair scientific evidence suggests that the risks of the clinical service outweighs potential benefits. Clinicians should not routinely offer the service to asymptomatic patients.
- ***Level I:*** Scientific evidence is lacking, of poor quality, or conflicting, such that the risk versus benefit balance cannot be assessed. Clinicians should help patients understand the uncertainty.

As suggested in guidlines of the Good Clinical Practice one should not omit to document relevant patient's data and by doing so evaluate application of evidence in the clinical setting.
Many tools are found on the web to help physicians apply evidence-based medicine principle into their daily routine. A good starting point is the Centre for Evidence-Based Medicine of the University of Oxford http://www.cebm.net. It is essentially constructed as a guide through the 5 steps of application of the Evidence-Based Medicine and can be used at all level of experience. However, many other sources can be used.
Medicine has been developing and prospering together with the human race. A huge progress has been achieved and the trend is unstopable. The amount of produced information by modern medical research is immense and one can hardly keep up with it. This may lead to creation of gaps in knowledge of medical personell which further on may lead to unsatisfactory treatments for patients. It remains personal responsibility of each of us to provide the highest standards of care to our clients/patients while actively searching for the best existing scientifing evidence and implementing it in the clinical decision making process. Tailoring the treatment for our patients while relying on the prinicples of the evidence-based practice is simply second to none.



Nikola Katic

Selected conditions of shoulder and elbow

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Introduction

Different disorders of the shoulder and elbow can cause lameness of different grades of the thoracic limb. Trauma (acute or chronic (repetitive) injuries), congenital or degenerative causes can be in the background of these disorders. While younger animals tend to suffer more often from acute and developmental conditions, older patients are more prone to chronic, degenerative diseases. As both, shoulder and elbow, represent rather complex functional systems mastering their anatomy is of the utmost importance when it comes to diagnoses and treatment of disorders of these joints. Systematic collection of information by means of clinical, orthopaedic and/or imaging exam is inevitable tool in order to diagnose and treat those.

"Anatomia fundamentum medicinae est"

Anatomy

Shoulder

This joint is the most mobile in all quadrupeds. Its primary motion are flexion and extension, however, adduction, abduction and rotation cannot be fully neglected. Its stability depends on:

- Cohesive forces of the synovial fluid within the joint space
- Joint capsule and its reinforcements in terms of medial and lateral gleno-humeral ligaments
- Tendons and muscles that surround it (rotator cuff)

Elbow

Distal humeral condyle is in close relation with the head of the radius by means of *cochlea humeri* and with the ulna by means of *trochlea humeri*. Weight load is equally shared by proximal joint surfaces of radius and ulna and there are some indications that a change of this relationship may be the cause of certain pathologic conditions of the elbow. Normal range of motion of the elbow in dogs is approximately 130° and its stability depends on:

- Anconeal, and medial and lateral coronoid process in extension
- Lateral, medial, annular ligament as well as intraosseus ligament between radius and ulna
- Flexors and extensors of the elbow.

Clinical exam

Beside a general clinical and orthopaedic/neurologic exam for the detection of the lameness some specific tests are often used for specific determination of joint pathology of the shoulder and elbow. Just as an illustration, pain on full extension of the elbow may indicate (but not exclusively) pathology of the anconeal process (ununited anconeal process), while pain on digital palpation while pronating the elbow may be indicative of processes of the medial coronoid process. Similarly, pain on palpation of the biceps tension in full flexion of the shoulder may be an indicator of the pathology of the origin of the biceps tendon. All these tests should not be omitted while examining these joints.

Imaging

Imaging plays an important role in detection of orthopaedic disorders.

Radiographic evaluation

It is used as a first imaging modality for various conditions of shoulder and elbow. For some disorders such as OCD of shoulder and elbow, degenerative joint disease and fractures this modality is sufficient to pose radiographic diagnosis. In other cases, additional advanced imaging may be needed.

Ultrasound

This modality is used mostly in determination of injuries to ligaments and tendons. It is very useful for evaluation of integrity of the biceps tendon, especially of its intra-articular portion.

Computer tomography

This diagnostic tool is becoming a common place in veterinary medicine. Its versatility and ability to reconstruct anatomic structures (especially bones) is commonly used for evaluation of structures of shoulder and elbow. It is especially well applicable for detection and evaluation of the fragmented coronoid process as well as incomplete ossification of humeral condyle. However, it is limited when it comes to evaluation of soft tissues and specifically of changes of the joint cartilage.

Other imaging modalities

Magnetic resonance imaging, scintigraphy and other tools are less commonly used for evaluation of shoulder and elbow. However, they may prove useful for clarification of differentials.

Arthroscopy

This is a very useful clinical tool to assess intra-articular changes of shoulder and elbow. Moreover, it can be used for therapeutical purposes in the same session. Illustratively, it is very often used for removal of OCD flaps in the shoulder as well as for removal of FCP fragments and debridement of changed subchondral bone in the elbow. With increased proficiency of the operator more advanced procedures can be performed.

Common disorders of elbow

Elbow dysplasia – ED

It is a complex of disorders defined by International Elbow Working Group (IEWG) and consists of:

- Ununited anconeal process
- Fragmented coronoid process
- OCD of humeral condyle
- Elbow incongruence

There seem to be a genetic background of elbow dysplasia so that the IEWG recommends screening on this condition in dog breeds prone to it. Clinically, this condition is manifested by forelimb lameness of different grades, which may be intermittent or increased after strenuous exercise, pain on joint manipulation and eventually positive coronoid test... Treatment is selected based on which of the joint components are affected and a perfect understanding of causative relationships is necessary in order to appropriately choose it.

Degenerative joint disease of Elbow (Cubarthrosis)

This condition is a consequence of ED and represents a sequel of both treated and untreated dysplastic elbows. This condition can be highly debilitating so that either a joint arthrodesis or even total elbow replacement may be necessary in affected animals. However, most of animals with this condition will have a positive response to conservative treatment in terms of sufficient pain management and physical therapy.

Common disorders of shoulder

OCD of humeral head

This condition is commonly seen in young large breed dogs (affected age 6-10 Months) and it represents a developmental disorder of endochondral ossification. Animals with this condition are usually presented to a vet with a sudden onset of forelimb lameness (usually unilateral, although both sides might be affected). Overly fast growth, feeding food high in protein and energy are just some of suspected causes for this disease. In the initial phase the joint cartilage is solely thickened and with the time a cartilage flap is created (osteochondritits disecans). The disease becomes manifest in this phase. Usually a radiographic exam will provide sufficient information. An OCD lesion is commonly seen as a subchondral defect of the caudal pole of the humeral head. Therapy is based on removal of the cartilage flap (this is commonly done arthroscopically) and curettage of the affected subchondral bone. This should facilitate creation of fibrocartilage that is supposed to cover the OCD lesion. Prognosis after such treatment is ususaly fair to good.

Biceps tendinopathy

According to some authors this is the most common shoulder related cause of lameness in older animals. It could be primary, due to repetitive trauma to the tendon, or secondary, as a consequence of other conditions of the shoulder. It is usually diagnosed by combination of diagnostic methods: clinically, biceps tendon test or cranial drawer, ultrasonographic exam or magnetic resonance. Eventually, the diagnosis is confirmed on arthroscopy, which at the same can be used for treatment (tenotomy, eventually combined with tenodesis). Prognosis after such treatment is usually good.

Medial instability of the shoulder

This is one of underdiagnosed causes of lameness linked to the shoulder. Although exact cause remains unknown repetitive microtrauma is suspected in the background of this disorder. Affected animals will be presented with lameness of various intensity which could be constant or intermittent. Therapy with non-steroidal anti-inflammatory drugs is commonly unsuccessful. Therapy is based on surgical reconstruction of the medial gleno-humeral ligament followed by used of Velpeau-sling and shoulder hobbles for 4-6 weeks. Prognosis is fair to good and could be improved by use of physical therapy.

Conclusion

List of disorders of elbow and shoulder is herewith not exhausted. This should, however, offer a short insight into complexity of pathology of these two joints and motivate the reader to search for further information in modern veterinary literature.

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Animais de Produção

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How far can we expect to improve the improvements of the fertility of cattle?

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Abstract

In the last decades, low fertility has been linked to numerous factors in high producing dairy herds. The detection of oestrus continues to present difficulties and, although progress has been made in regards to oestrus synchronization and artificial insemination, the reproductive performance of dairy cows has not improved substantially. However, higher fertility is often registered in high-producing dairy herds when compared with lower producers. Further, our attention not only should be directed to postpartum reproductive disorders and subsequent fertility, but it is also of great interest to understand the effects of pregnancy loss once a cow is pregnant. Following a positive pregnancy diagnosis, late embryonic/early foetal loss is becoming the most common complication of gestation in high producing dairy herds. This presentation expresses our views on factors of a non-infectious nature that affect the fertility of lactating dairy cows following AI. Special attention is paid to factors related to the cow and its environment and to some approaches to increase reproductive efficiency such as the control of anoestrous cows and confirmation of oestrus at insemination. Pregnancy maintenance during the early foetal period, involving the problem of the twin pregnancies, is discussed also as a critical step.

Introduction

In the past decades, fertility declining besides increased milk production has been linked to numerous factors in high producing dairy herds (Beam and Butler, 1999). Late embryonic/early foetal loss is becoming furthermore the most common complication of gestation in high producing dairy herds (López-Gatius, 2012). However, management practices related to the well-being of cows can improve both milk production and fertility of dairy cows. In effect, there is a tendency towards a higher level of management in high producing compared to lower producing herds (Calus *et al.*, 2005). Despite of this, the reproductive success is a multifactorial and complex process aggravated in warm countries in which summer heat stress is a major factor impairing fertility. This presentation expresses our views on factors affecting fertility in high producing dairy herds. Special attention is paid to factors related to the cow and its environment and to some approaches to increase reproductive efficiency such as the control of anoestrous cows and confirmation of oestrus at insemination. Pregnancy maintenance during the early foetal period, involving the problem of the twin pregnancies, is discussed also as a critical step.

Management and environmental factors

Low fertility has been often associated with high milk production, but this problem cannot be solely attributed to milk production. In three extensive studies including 24,366 AI we could not detect a negative effect of milk production on fertility (López-Gatius *et al.*, 2005a, b; Garcia-Ispierto *et al.*, 2007b). However, extensive studies have made possible

to link high milk production in individual cows to high fertility (Lucy, 2001; López-Gatius *et al*., 2006). For example, early fertile cows (cows that become pregnant before 90 days postpartum) were those who produce more milk at day 50 postpartum (López-Gatius *et al*., 2006). Good management practices perhaps allow expression of genetic potential of early fertile cows, whereas lower producer receive inadequate care. The findings of studies that have identified factors promoting fertility (early fertile cows) should be incorporated in routine checks conducted on herds. Data derived from these types of study are often more interesting than those of studies examining factors related to fertility failure.

The bull and AI technician effects on fertility have been extensively reported. In several studies performed in our area we could determine that some bulls decreased fertility at an odds ratio range of 0.31 to 0.44 (López-Gatius *et al*., 2005a; García-Ispierto *et al*., 2007b), whereas one single bull increased fertility by a factor of 4.7 (López-Gatius *et al*., 2005b). However, we should consider whether the decline on fertility of high producing dairy herds can be attributed sometimes to the male (DeJarnette *et al*., 2004), many environmental and herd management factors will affect fertility estimates of an inseminating bull (Foote, 2003). Therefore, a continuous control of seminal doses entering in a herd should be useful to promptly locate a negative bull. In the same way, re-training and continuous control of inseminators should be considered.

Management and environmental factors such as the use of a bull in the herd; inseminator, poor nutrition or the loss of the body reserves (negative energy balance); and housing elements (concrete slatted or dirty floors) can affect fertility. However, most studies report the seasonal effect as a major environmental factor affecting fertility. High temperatures have been strongly linked to low fertility (see review by López-Gatius, 2012). For example, average conception rates to first AI in an extensive study performed in North-Eastern Spain were 43 and 22% for the cool and warm periods, respectively (López-Gatius, 2003). During warm period, any stressor such as high temperatures, could compromise the benefits of milk production. Metabolic demands due to high milk production under heat stress conditions can compromise the reproductive functions of cows (Labèrnia *et al*., 1998; De Rensis *et al*., 2015). The use of the temperature-humidity index (THI) or temperature alone to control a farm environment would depend on the individual farm and on each environmental situation to adopt cooling measures (Garcia-Ispierto *et al*., 2007a).

Control of anoestrous cows

Oestrous cyclicity disappears when the cows are under different types of stress (anoestrous or non-cyclic animals). Anoestrus is a broad term that indicates the lack of expression of oestrus (or absence of oestrous signs), despite an efficient oestrus detection approach. A true anoestrous condition is primarily characterized by anovulation (Peter *et al*., 2009). The incidence of anoestrous cows can reach figures close 40% in the herd, probably reflecting inadequate management practices (Peter *et al*., 2009). It is very important to identify the different types of anoestrus to apply adequate synchronisation protocols at herd level. This subject will be expanded in the next presentation.

Confirmation of oestrus

Detection of oestrus remains a major problem in the XXI century (Roelofs *et al*., 2010). Cows are often falsely identified as being in oestrus and inseminated when conception cannot occur (López-Gatius, 2000, 2012). Oestrous signs in pregnant cows at all stages of pregnancy make the situation even more difficult (Thomas and Dobson, 1989). From 19% (Sturman *et al*., 2000) to 40% (Nebel *et al*., 1987) of AI were incorrectly performed in pregnant cows, inseminations which can cause embryonic mortality or abortion (Vandemark *et al*., 1952). Through rectal examination of the bovine reproductive tract either by hand or by ultrasonography, an animal can be correctly diagnosed as being ready for service (Roelofs *et al*., 2010).

Clinical aspects of gestation control programmes

Early foetal loss, peaking between days 40 and 50 of gestation is becoming the most common complication of gestation in high producing dairy cows, with more than 90% of pregnancy losses following pregnancy diagnosis occurring usually before day 90 (López-Gatius and Garcia-Ispierto, 2010; López-Gatius, 2012). An early foetal loss of 10-12% is a commonly accepted figure, reaching until 40% in some Factors strongly affecting early fetal loss are parity (cows versus heifers), semen-providing bull, warm season, and twin pregnancies, whereas the presence of an additional corpus luteum has been identified as a strong positive factor favoring pregnancy. Cows bearing twins have a pregnancy loss of 3 to 7 times higher than single pregnancies, and losses are from five to nine times higher for unilateral than for bilateral twins (López-Gatius and Garcia-Ispierto, 2010; López-Gatius, 2012).

The first problem in diagnosing twin pregnancies is that the two embryos must be clearly located. Ultrasound exams are accurate and common practice for twin pregnancy diagnosis in dairy cattle. Carrying twins has been extensively described as an emerging principal non-infectious factor jeopardizing pregnancy maintenance and reducing the lifespan of dairy cows (Andreu-Vázquez *et al*., 2012a). Probably genetics and improvements at the farm level related to increased milk productivity have diminished the risk of embryo loss in twin pregnancies and thus raised the twinning rate. Breeding synchronization protocols for fixed-time artificial insemination are becoming standard components of the current breeding management of lactating cows, and some of them can increase the twin pregnancy rate (Andreu-Vázquez *et al*., 2012b). It is therefore foreseeable that over the years to come, the twinning rate will continue to increase along with milk productivity. Obviously, the incidence of twin births is closely related to the twin pregnancy rate, exceeding 18% the twin pregnancy rate in some herds (Andreu-Vázquez *et al*., 2012b). Therapeutic approaches for the problem of twin pregnancies include GnRH (Bech-Sàbat *et al*., 2010) treatment or induced embryo reduction (López-Gatius, 2012). With similar results, manual rupture of the amniotic vesicle or transvaginal ultrasound-guided aspiration of allanto-amniotic fluid have been proposed as methods of choice to perform twin reduction in cows on Day 28-41 of gestation. However, benefits and risks of induced twin reduction should be quantified.

Spontaneous embryo reduction has been described in natural twin pregnancies in dairy cattle, with incidences from 11.2% to 28.4% (López-Gatius, 2012). Corpora lutea and embryos are vulnerable to the effects of stress factors such as heat stress in cows maintaining their pregnancies (López-Gatius *et al*., 2010). The presence of a dead co-twin at the time of pregnancy diagnosis is highly related to pregnancy loss thereafter, which occurs in more than 60% of the cases. However, the remaining cows stay pregnant as single pregnancies. Interestingly, most cases of single embryo mortality in twins occurs around Days 28-40 of gestation and rarely occurs after Day 60 (López-Gatius and Hunter, 2005; López-Gatius *et al*., 2010). This involves a very important practical aspect. The fate of pregnancies with live twins on Day 60 is either twin delivery or abortion. Therefore, the assessment of a normal twin pregnancy on Day should be a component of the management policy of a herd. Since the gestation length in cows carrying twins is several days less than the time for single births, the dry-off period can be advanced for non-aborting cows carrying twins. Adequate nutrition during the dry-off period and additional care at parturition can further increase neonatal survival of the twin calves.

Concluding remarks

From a, integral management stand point, one should pay attention to cows becoming pregnant within 90 days, the training and re-training of the AI technicians, be careful not to inseminate pregnant animals and improve oestrus detection. Semen providing bull should be frequently monitored; and THI and/or temperature measurements should allow us to decide to establish better cooling systems in the herds.

Assessment of normal development of gestation on day 60 after insemination should be included in the routine control of gestation in the herd. The dry-off period can be advanced several days for non-aborting cows carrying twins and additional care of these cows before and at parturition can further increase neonatal survival of the twin calves. Administration of GnRH on the day of pregnancy diagnosis in all cows with twins included as a routine treatment in the herds may reduce dramatically the incidence of pregnancy loss in twin pregnancies.

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Paratuberculose em Portugal

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Introdução

A produção leiteira mundial enfrenta um preocupante cenário de distribuição mundial da Paratuberculose (PTB); esta preocupação resulta da produção de evidências do impacto negativo na produção aliado ao potencial risco zoonótico do agente *Micobacterium avium spp tuberculosis* (***Diabettes mellitus*** tipo I, doença de Crohn, Aterosclerose, Sindrome de Blau, "Hashimoto thyroiditis" e esclerose múltipla (Naser et al, 2013; Sechi e Dow, 2015) que pode agravar a emergente perda de confiança dos consumidores.

As prevalências Europeias são difíceis de aferir quer pela diversidade e heterogeneidade de resultados publicados, quer da heterogeneidade de definição de caso e de abordagens à sua quantificação. As prevalências reportadas são geralmente baixas ao nível do animal, mas elevadas ao nível da exploração. Na Europa foram estimadas prevalências próximas de 20%, ou no mínimo de 3-5% ao nível individual e > 50% ao nível das explorações; nenhum país dispõe ainda de informação suficiente para poder considerar-se livre da doença (Nielsen & Nils Toft, 2008).

Portugal não dispõe de programas oficiais de controlo de MAP. Apesar de existirem alguns programas voluntários que incluem a monitorização da doença, não apresentam medidas de controlo de infeção definidas. Foi reportada uma prevalência na região Norte do país de 45,9% ao nível das explorações e de 2,3% ao nível das vacas (Correia-Gomes, 2010).

A fisiopatogenia particular da PTB a par das características dos testes de diagnósticos disponíveis, colocam um desafio quer ao diagnóstico individual precoce, quer à caracterização das explorações face à presença da infeção e consequentemente à eficácia dos programas de controlo do tipo "teste-refugo" (serão discutidos estes aspetos). Como consequência, os programas voluntários de controlo de doença ou de redução da presença de MAP no leite destinado a consumo humano dependem da implementação de uma estratégia concertada de prevenção de novas infeções, da identificação de animais adultos potencialmente excretores aliada a refugo seletivo de animais baseada no risco de excreção para o ambiente. Este tipo de programas existe em diversos países (não em Portugal) e tem suporte em esquemas de testagem estruturada e repetida no tempo que são dependentes da prevalência esperada ou real nas explorações ao longo do tempo, e onde são utilizados com frequência mais do que um tipo de teste, quer a indivíduos quer a pools amostrais de indivíduos, leite de tanque ou ambiente.

No presente trabalho foi determinada a proporção de explorações positivas tendo por base 222 explorações abrangidas pelo contraste leiteiro, em 48 distritos. Posteriormente foi investigado o impacto da positividade frente a MAP, avaliada por Enzime Linked Imunossorbant Essay (ELISA), sobre a produção leiteira corrigida aos 305 dias (305EMP) e as contagens de células somáticas (SCC).

Resultados

Das 1263 explorações registadas em contraste leiteiro, 222 foram classificadas relativamente à presença de MAP, com base na testagem sobre amostras de sangue de todos os animais da exploração com mais de 30 meses, utilizando um ELISA bifásico indireto (NEG: todos os animais testados e com resultado negativo; POS: se diferente do anterior). Depois de georreferenciadas a totalidade das explorações em contraste utilizando o programa ArcGis Vs10, foi determinada a fração amostral e calculadas as proporções de explorações infetadas. Os 48 distritos estudados englobaram 1074 (85%) do total das explorações em contraste no ano de referência. A proporção global de explorações positivas foi de 58% (CL 95%: +-3). A prevalência de explorações POS por distritos variou de 0 a 100%. Serão apresentados mapas georreferenciados ilustrando a proporção de explorações POS ao nível dos distritos.

A partir de uma amostra de 329 explorações, um total de 15.300 vacas (6,45% da população nacional e 20% das explorações registadas em contraste leiteiro) e dados de 45.000 lactações, foram investigados os efeitos da positividade frente a MAP na Milk305 e SCC. As vacas foram classificadas em POS, NEG ou DUB (resultado ELISA positivo, negativo ou duvidoso respetivamente); as explorações foram classificadas em função da classificação da totalidade dos animais presentes com mais de 30 meses em fortemente negativas (SNEG) negativas (NEG) não-negativas (NNEG) e positivas (POS). Foi posteriormente atribuída uma classificação a cada vaca e respetivas lactações, atendendo à classificação individual e à classificação da exploração de produção em NEG_SNEG, NEG_NEG, NEG_NNEG, NEG_POS, DUB_NNEG, DUB_POS, POS_NNEG e POS-POS, considerando "Classificação Individual"_"Classificação de Exploração".

Do total das vacas observadas, 4,5% foram POS. Relativamente às explorações, apenas 14,6% foram classificadas como S_NEG. 39.0% Foram classificadas como NEG, 18,2% como N_NEG e 29,2% como POS. As explorações POS e N_NEG totalizaram 47,4% das explorações e apenas 17,2% das vacas pertenciam a explorações potencialmente livres de MAP. 58,3% Das vacas eram animais NEG em explorações POS. Independentemente do estatuto das vacas, 62,2% dos partos e lactações ocorreram em explorações POS, sugerindo um elevado risco de infeção e dispersão do agente nas gerações de reposição se não forem tomadas medidas de controlo.

A variação da 305MEP e das SCC em função dos estatutos frente à infeção por MAP foram investigados através de regressão hierárquica multinível, considerando os níveis "número de lactação" (LactNr), "vaca" (Cow) e "exploração" (Farm). Foram consideradas no modelo as interações LacNr*Estatuto Individual (LacNr*Cow_MAP) e LactNr*Farm_MAP, que se revelaram significativas (p>0,05); o efeito quadrático do nº de lactação permitiu incluir no modelo a variação fisiológica da produção dos animais em lactações sucessivas.

Em média as 305MEP foram superiores nas explorações POS que nas explorações SNEG NEG e NNEG.

Foram concebidos modelos diferentes para diferentes questões colocadas. Em primeiro lugar foram construídos os modelos incluindo todas as explorações (logo todos os animais e lactações) na análise (1). Seguidamente foram apenas retidos para análise os dados das explorações positivas (todos os animais e lactações observadas nas explorações POS) (2). Foi ainda realizada a análise e o modelo, considerando apenas os animais (e consequentemente lactações) negativos, pertencentes a todas as explorações (3).

(1) Quando consideradas todas as explorações na análise, observou-se que:

As 305MEP das 1ªs lactações de vacas POS foram superiores às das 1ªs lactações de vacas não negativas (POS+NNEG), mas as vacas POS vão produzindo progressivamente menos leite que as não negativas, nas lactações seguintes, resultando numa redução acumulada à quinta lactação de 1168.6 Kg.

A 305MEP foi superior nas explorações POS que nas não positivas; mas as vacas NEG das explorações POS produziram mais leite que as vacas NEG de explorações NEG (3), o que parece indicar um maior nível produtivo por parte dos animais residentes em explorações POS.

As SCC foram superiores nas vacas POS, sofrendo incrementos progressivos com o aumento do nº de lactação.

As SCC foram mais elevadas nas explorações POS que nas não POS e aumentaram igualmente com o aumento da lactação. As lactações POS nas explorações POS foram as que apresentaram as SCC mais elevadas (3).

(2) Quando consideradas para análise apenas as explorações positivas e correspondentes vacas e lactações observou-se que:

Apesar de que a 305MEP das 1ªs e 2ªs lactações foi superior (diferença de 205,5 e 31,7kg respetivamente) para as vacas POS relativamente às não positivas, a produção corrigida acumulada na vida (até à 5º lactação) foi mais baixa em 869,4 kg que a das vacas não positivas nas mesmas explorações POS. Os resultados parecem sugerir uma maior suscetibilidade à infeção por MAP por parte das vacas com maior capacidade produtiva.

As SCS observadas foram superiores nas vacas POS e a diferença foi aumentando ao longo das lactações, relativamente às SCC das vacas não positivas. Quando analisamos apenas vacas negativas (de todas as explorações), as vacas NEG de explorações POS tiveram SCC superiores às das vacas NEG em explorações NEG (3). Poderemos questionar se o efeito do aumento das SCC resulta da infeção por MAP, ou se resulta do aparente maior potencial produtivo de vacas em explorações positivas, que é um reconhecido fator de risco para infeções da glândula mamária.

Importa relembrar que em fases precoces da infeção, o sistema imunitário produz uma resposta defensiva de tipo celular frente ao MAP, mantendo-o sobre controlo. Nesta fase a resposta imunitária humoral, que se traduz pela produção de anticorpos (Ac) específicos é ténue, comprometendo a sensibilidade dos diagnósticos baseados na deteção de Ac circulantes, nomeadamente o ELISA. Também a especificidade do diagnóstico é afetada pela prevalência da infeção; quanto menor a prevalência real, maior a possibilidade de ocorrência de falsos positivos ao diagnóstico.

Discussão

De acordo com a investigação mais recente, os animais com infeções latentes (low-path) poderão não sofrer perdas produtivas de longo curso (R. L. Smith et al., 2015). Por esta razão as infeções latentes podem funcionar como confundidor nas avaliações de impacto da PTB nas explorações; não é geralmente possível incluir o estado de progressão da doença nos modelos de análise, sendo os animais com infeção latente e ativa frequentemente considerados conjuntamente.

Existe uma considerável heterogeneidade dos resultados reportados sobre a influência do MAP na produção leiteira sendo estes em alguns casos antagónicos (Conor G. McAloon et al, 2016). Numa metanálise recente foi possível observar que a redução de produção atribuível à paratuberculose é real mas a sua magnitude é modesta; na mesma concluiu-se ainda que quando o efeito é medido com base nos resultados de cultura fecal, a redução é de 1,87kg vaca/dia (IC 95% = 2,34 -1,40), equivalente a uma redução de 5,9% na produção global (Conor G. McAloon et al, 2016). Os resultados apresentados que reportam uma fase inicial de maior produção dos animais POS, e uma segunda fase onde a produção é inferior à dos animais NEG está de acordo com resultados mais recentes, que observaram posteriormente a uma fase inicial de maior produção, uma redução da produção em aparente função da intensidade

clínica da doença (R. L. Smith et al., 2015). Consideramos que o estádio de infeção é um modificador do efeito da infeção e que a inclusão da interação entre o estatuto individual frente a infeção por MAP com o nº da lactação, foi capaz de diferenciar pelo menos parcialmente este de outros fatores de variação das observações.

Existe tal como se reporta, uma elevada proporção de explorações positivas em Portugal, que se traduz por uma pequena fração de animais detetáveis / infetados; O curso da doença nos animais afetados é muito variável e os impactos individuais são difíceis de assessorar. Não obstante, fica demonstrado que existem efeitos negativos na produção e na saúde do úbere; que existe um elevado risco de infeção para as novas gerações e que o potencial de exposição do leite de tanque não deve ser desconsiderado.

Conclusão

O presente estudo confirma a elevada prevalência da infeção por MAP nas explorações leiteiras nacionais e um intenso potencial de exposição de vacas e vitelos, dado que mais de 60% dos partos ocorre em explorações infetadas. O risco de exposição do leite recolhido é considerável e não deve ser negligenciado, à luz do princípio da precaução e das dúvidas que persistem na comunidade científica sobre a sua participação em doenças humanas como ***Diabettes mellitus*** tipo I, doença de Crohn e Aterosclerose, Sindrome de Blau, "Hashimoto thyroiditis" e esclerose múltipla (Naser et al, 2013; Sechi e Dow, 2015)

De igual modo os efeitos observados sobre a produção leiteira e saúde do úbere dão suporte à necessidade de implementar medidas de controlo ao nível das explorações. Estas deverão assentar primordialmente na limitação da exposição dos animais jovens à contaminação feco-oral, principal via de infeção com MAP.

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Present and future utilization of hormone therapy in reproductive cattle management

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Abstract

Synchronization of oestrus and ovulation in groups of animals remain an indispensable part of reproductive control management in dairy herds. The most current used methods include the combination of gonadotrophin releasing hormone (GnRH) and prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) seven days later with or without progesterone (P4). Five-day P4-based protocols however have provided results that compare favourably with those observed for 7-day protocols. This presentation revises present and future prospects of hormone treatments for synchronizing oestrus and ovulation for fixed-time insemination (FTAI) in dairy cattle.

Introduction

The health and fertility of dairy cows improve fpllowing management practices that improve animal well-being (Windig *et al.*, 2005). This has meant a higher level of management in high producing compared to lower producing herds (Calus *et al.*, 2005). However, both the high incidence of cows suffering anoestrus (Peter *et al.*, 2009) and the poor detection of oestrus (Roelofs *et al.*, 2010) remain a major concern in dairy herds.

Oestrous cyclicity disappears in cows are under different types of stress (anoestrous or non-cyclic animals). Anoestrus is a broad term that indicates the lack of expression of oestrus (or absence of oestrous signs), despite an efficient oestrus detection approach. A true anoestrous condition is primarily characterized by anovulation (Peter *et al.*, 2009). The incidence of anoestrous cows can reach figures close 40% in the herd, probably reflecting inadequate management practices (Peter *et al.*, 2009).

Examples of effects from both non-detection and erroneous detection of oestrus:

- Inadequate detection of oestrus has been related to an annual cost greater than US$ 300 million by the USA dairy industry (Senger, 1994).
- Pregnant cows can show oestrus signs that are indistinguishable from those of true oestrus in non-pregnant animals (Thomas and Dobson, 1989).
- More than 40% of cows were inseminated at a time of high milk P4 levels (Nebel *et al.*, 1987).
- About 19% of the inseminations were performed in pregnant cows (Sturman *et al.*, 2000).
- In the Netherlands, 4% of the calves born were from an insemination before the last one (Dijkhuizen and van Eerdenburg, 1997).

These are more than cogent reasons why breeding synchronization protocols for fixed-time insemination (FTAI) have become routine components in the current reproductive management of lactating cows. Synchronizing oestrous cycles of the cow depends on control of the functional lifespan of the corpus luteum (CL). There are two ways to facilitate control of the CL that result subsequently in oestrus and ovulation. The first method involves long term administration of a progesterone (P4) with subsequent regression of the CL during the time the P4 is administered. Oestrus and ovulation occur within 2 to 8 days after P4 withdrawal. The second method involves the administration of a luteolytic agent that shortens the normal life span of the CL. This is accompanied generally with oestrus and ovulation within 48 to 120 hours after treatment. The most recently developed synchronization treatments combine different hormones that control oestrous cycle length and follicular dynamics in order to achieve a precise onset of oestrus and normal fertility after a single FTAI, independent of the status of the animal. This presentation revises present and future prospects of hormone treatments for synchronizing oestrus and ovulation for FTAI in lactating dairy cows.

Synchronizing oestrus and ovulation for fixed-timed artificial insemination (FTAI)

The optimal protocol for FTAI would be the one that increases the oestrous rate and reduce the negative effect of different types of anoestrus. To achieve that purpose, several combinations of hormones have been tested world-wide. Such treatments may control oestrous cycle length and follicular dynamics in order to achieve a precise onset of oestrus and normal fertility after single insemination, independent of the status of the animal. For example, the prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$)-based ovulation synchronisation protocol denoted "Ovsynch", is extensively applied for FTAI of lactating dairy cows (Rabiee *et al.*, 2005) and P4-based FTAI synchronisation protocols are used in lactating dairy cows and heifers (Yániz *et al.*, 2004; Macmillan, 2010).

Ovsynch-based protocols

The Ovsynch method consists of gonadotrophin releasing hormone (GnRH) treatment given at random stages of the oestrous cycle (synchronises a follicular wave - FW) followed by $PGF_{2\alpha}$ or synthetic analogue 7 d later (luteolytic effect on a CL). A second dose of GnRH is administered 36 h after $PGF_{2\alpha}$ treatment (in order to synchronise ovulation) and the cows are inseminated 16 to 20 h later without detection of oestrus (Pursley *et al.*, 1995, 1997). This triple treatment (GnRH- $PGF_{2\alpha}$ -GnRH) is the reason why the Ovsynch method has been termed "GPG".

Numerous modifications derived from Ovsynch or GPG. Some referenced examples:

"Select-Synch" – the second GnRH treatment is removed and cows inseminated at oestrus (Lemaster *et al.*, 2001).

"Co-Synch" – the second dose of GnRH plus AI are applied 48 or 72 h after $PGF_{2\alpha}$ treatment (Lemaster *et al.*, 2001; Brusveen *et al.*, 2008).

"Heatsynch" – the second dose of GnRH is changed to oestradiol benzoate. (Barros *et al.*, 2000; Kasimanickmam *et al.*, 2005).

"Double-Synch" – two Ovsynch protocols applied 7 d apart. (Giordano *et al.*, 2012).

"Presynch" – two $PGF_{2\alpha}$ doses applied 14 d apart, 14 d before Ovsynch. (El-Zarkouny *et al.*, 2004).

"G6G" - $PGF_{2\alpha}$ and GnRH 2 d later applied 6 d before Ovsynch. (Bello *et al.*, 2006).

However, FTAI following Ovsynch seems to have no beneficial effects in heifers, due to an inconsistent FW pattern and thus an insufficient response to the first GnRH injection, and in anoestrous cows, given their lack of prostaglandin responsive CL. P4-based protocols seem to provide solution to this problem and to the fact that higher milk producers have an elevated hepatic clearance of steroids hormones (Sangsritavong *et al*, 2002). Moreover, the positive effect of P4 protocols has been tested in both, cycling and non-cycling (anoestrous) animals, and in lactating cows and heifers.

Progesterone-based protocols

Ovulation and oestrus can be induced by P4 treatment (for 7-9 days) in combination with GnRH or an analogue, and $PGF_{2\alpha}$ or an analogue (Roche *et al.*, 1992; Yániz *et al.*, 2004; Macmillan, 2010). Progesterone-based protocols allow for effective FTAI in lactating dairy cows, regardless of whether cows are cyclic or non-cyclic (anoestrous cows) and also without the need to detect oestrus (López-Gatius *et al.*, 2001, 2004, 2008). Five-day P4-based protocols have provided results that compare favourably with those observed for longer protocols either in heifers as in lactating cows (Rabaglino *et al.*, 2010; Lima *et al.*, 2011; Ribeiro *et al.*, 2012; Garcia-Ispierto *et al.*, 2013; Garcia-Ispierto and López-Gatius, 2014; Colazo and Ambrose, 2015; López-Gatius *et al.*, 2015).

The success of five-day P4-based protocols is probably due to the conversion of younger, healthier oocytes into follicles with a shorter dominance life than longer protocols (Cerri *et al.*, 2009). These shorter oestrus synchronization protocols make use of different combinations of hormones such as GnRH, equine chorionic gonadotropin (eCG), and/or $PGF_{2\alpha}$ given as two luteolytic doses 24 h apart upon P4-device removal. The ovarian response and fertility resulting from each treatment were due more to the ovarian structures present at the time of treatment than to the different combinations of hormones investigated (Garcia-Ispierto and López-Gatius, 2014).

In dairy herds with a high incidence of anoestrus the eCG has been added to the TAI protocols (reviewed by De Rensis and Lopez-Gatius, 2014), improving reproductive performance of anoestrous cows (Garcia Ispierto *et al.*, 2012). In this regard, cows with no oestrus signs over 21 days subjected to a shortened P4 protocol that included eCG treatment resulted in P/AI similar to that obtained in cows that were AI after spontaneous oestrus (Garcia Ispierto *et al.*, 2013). In addition to the increased ovulation rate of healthier follicles in acyclic cows, eCG could also enhance oestrous behaviour increasing the service rate in those herds that utilize oestrus detection (Sa Filho *et al.*, 2010).

Implications

Although specific synchronization to different types of anoestrus may give better results than Ovsynch alone (López-Gatius *et al.*, 2004, 2008), efforts have been performed during the last decades to find one single method for all cows finishing the waiting period. Short P4-based protocols (5 days) for FTAI are showing acceptable results. Equine chorionic gonadotrophin added to P4-based protocols can improve reproductive performance of anoestrous cows. Progesterone seems to withstand the tests of time.

Under our work conditions, we are applying one single 5-day P4-based protocol for all cows with acceptable results:

Cows are treated with a progesterone-releasing intravaginal device (PRID) plus GnRH upon PRID insertion. The PRID is left for 5 d, and these animals receive $PGF_{2\alpha}$ or analogue on PRID removal. Twenty-four hours later, the cows received a second $PGF_{2\alpha}$ dose, a second GnRH dose 24 h later and they are inseminated either at oestrus (cows show-

ing oestrus) or 60 h after PRID removal (FTAI: cows with no oestrous signs).

In some herds, however, cows showing a mature CL during the weekly reproductive visit receive a $PGF_{2\alpha}$ dose. These cows either are inseminated if they show oestrous signs within 3 d, or receive a GnRH dose to enter in a G6G protocol.

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Fernando López-Gatius

Impact and control of *Neospora caninum* and *Coxiella burnetii* in cattle fertility

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Abstract

Neospora caninum is a protozoan parasite with a preference for cattle and dogs as hosts. When *N. caninum* infection occurs in cattle it induces abortion, bovine neosporosis being a main cause of abortion worldwide. However, not all infected cows abort and it is not yet understood why this occurs. At present there is no effective treatment or vaccine. Q fever is a zoonosis produced by *Coxiella burnetii*, a bacterium that is widely distributed worldwide. Domestic ruminants are the most important source of *C. burnetii* for human infection. However, the symptoms described in cattle remain inconsistent. Drawing mostly from results derived from our research on these topics, some epidemiologic aspects and clinical suggestions on bovine neosporosis and Q-fever are highlighted here. Emphasis is placed on gestation control programs, concerning *Neospora*-seropositive cows for management of the disease at herd level, and on the scarcity of data available and possible control actions on Q fever.

Introduction

Neospora caninum is an obligate protozoan parasite which infects a wide range of warm-blooded animals but with a preference for cattle and dogs. Bovine neosporosis is a disease of international concern as it is among the main causes of abortion in cattle worldwide (Dubey and Lindsay, 1996; Dubey and Schares, 2011). *Neospora caninum* is one of the most efficiently trans-placental transmitted organisms in cattle. Up to 95% of calves are born infected (Dubey *et al*., 2007). The majority of calves born from infected mothers are clinically normal, but they are infected for life and *N. caninum* infection can be maintained over several generations by vertical transmission (Pabón *et al*., 2007).

Coxiella burnetii is an intracellular bacterium spread worldwide that causes Q fever in animals and also in humans. Domestic ruminants such as cattle, goats and sheep are considered to be the primary reservoir species for exposure of humans (Arricau-Bouvery and Rodolakis, 2005). Bacteria have been found in placenta or aborted fetuses (Parisi *et al*., 2006). With an important controversy (Ortega-Mora, 2012; Agerholm, 2013; Garcia-Ispierto *et al*., 2014), the main clinical manifestations in cattle are late abortion (Woldehiwet, 2004), infertility (To *et al*., 1998) and metritis and placenta retention (López-Gatius *et al*., 2012). Interactions throughout gestation between *Neospora*- and *Coxiella*-infection have been furthermore described in cattle. Both the *N. caninum* and *C. burnetii* infection modify endocrine patterns throughout gestation. Cows seropositive to both, *N. caninum* and *C. burnetii*, show higher plasma progesterone levels (Garcia-Ispierto *et al*., 2010).

Drawing mostly from results emerging from our research on these topics, some epidemiologic aspects and clinical suggestions on bovine neosporosis and Q-fever are highlighted here. Emphasis is placed on gestation control programs.

Bovine neosporosis: from research to clinic

Abortion is the main clinical manifestation of bovine neosporosis with most abortions occurring at 5-7 months of gestation (Dubey *et al*., 2007). For example, in our geographical area of study, based on the odds ratio, the risk of abortion was found to be 12-19 times higher in *Neospora*-seropositive dairy cows than in seronegative cows, ranging from 30 to 44% in seropositive animals (López-Gatius *et al*., 2004a, b), and maintaining a similar risk of abortion over several years (Pabón *et al*., 2007; Mazuz *et al*., 2014). In fact, in commercial dairy herds routinely examined by us in a reproductive management program, 0% is a common figure for the abortion rate during the second and third terms of gestation for *Neospora*-seronegative animals (Almería and López-Gatius, 2013; López-Gatius *et al*., 2016). However, chronic *N. caninum infection* prior to pregnancy seems to be not responsible for abortion before day 90 of gestation (López-Gatius *et al*., 2004b).

Our studies have also revealed that *Neospora*-infection does not affect the fertility of animals chronically infected prior to pregnancy (López-Gatius *et al*., 2005a). Interestingly, *Neospora*-infected cows suffering abortion had a high likelihood of good fertility within the first month of abortion compared to the case of abortions due to different types of stress (Santolaria *et al*., 2009). Crossbreed pregnancies dramatically reduce the abortion risk in Neospora-infected dairy cows, especially if Limousin semen is used (López-Gatius *et al*., 2005b; Almería *et al*., 2009). In the latter study, if cows inseminated with Limousin semen showed a low antibody titer, these results were even more striking and abortion rates similar to those recorded in seronegative animals were obtained.

To date, neither serological tests nor markers for serodiagnosis have served to attribute abortion to *N. caninum* in an individual cow (Almería and López-Gatius, 2015). Despite of this, however, yearly whole-herd serological screening for antibodies against *N. caninum* using ELISA tests is a very effective method both to detect *N. caninum* infection and to estimate the risk of abortion at the herd level. Numerous studies have related a raised risk of abortion to elevated antibody titres in cows, but not in heifers (Dubey *et al*., 2007; Almería and López-Gatius, 2015).

There are data consistent with a sylvatic cycle, including domestic and wild canids and ruminants, probably important in the biology of *N. caninum*. For example, in Spain, antibodies against this protozoan have been observed in wild carnivores (Sobrino *et al*., 2008), wild ruminants (Almería *et al*., 2007) and wild birds (Molina-López *et al*. 2012; Darwich *et al*., 2012). These findings could have important implications for domestic cycles, since the sylvatic cycle may affect the infection prevalence in cattle herds in a given area (Almería and López-Gatius, 2013; López-Gatius *et al*., 2016).

Does *Coxiella burnetii* affect reproduction in cattle?

Q fever has been related to stillbirth and aborted foetuses (Arricau-Bouvery and Rodolakis, 2005) and the presence of the bacterium in placental and foetal tissues has been extensively described (Garcia-Ispierto *et al*., 2014). A question that is still under debate is whether a rise in bulk tank milk antibodies may be linked to an increased risk of abortion. Although there is positive correlation between serology and shedding, the presence of seronegative animals that shed *C. burnetii* and seropositive animals that do not, questions the significance of serology in dairy cows (Garcia-Ispierto *et al*., 2014). However, whole-herd serological screening for antibodies against *C. burnetii* using ELISA tests may help to decide vaccination or not in the herds in which reproductive disorders increase.

Abortion, stillbirth and placenta retention have been related to subsequent postpartum diseases (LeBlanc, 2008). However, in *C. burnetii* infection there is no experimental evidence pointing to a direct link between *C. burnetii* infection and postpartum disease in dairy cows (Agerholm, 2013; Garcia-Ispierto *et al*., 2014). Although *C. burnetii* may be present in the genital tract of healthy animals (Garcia-Ispierto *et al*., 2013; Tutusaus *et al*., 2013), its presence could

not be related to the subsequent postpartum disorders. However, we may assume that the real clinical impacts of *C. burnetii* infection on conception rate in dairy herds have not been adequately addressed in the different studies. In addition, the high costs of tests (for example, PCR for all routes of shedding such as milk, faeces and vaginal fluid during the postpartum and insemination periods) question the real need for these kinds of study.

Today, measures to control *C. burnetii* infection on a farm consist of antibiotherapy or vaccination. Antibiotherapy, although demonstrated to reduce shedding in cattle does not reduce abortion or prevent shedding (Angelakis and Raoult, 2010) and is not economically viable in dairy herds. Milk obtained following antibiotic treatment has to be removed from the food chain in Europe. Thus, vaccination against *C. burnetii* is the only possible solution to prevent bacterial shedding in a herd.

Two vaccine types against *C. burnetii* have been developed: phase I and phase II. The phase I vaccine seems the most protective. In dairy cattle, a Th2 immune response and reduced shedding has been observed following a phase I C. burnetii inactivated vaccine, only in non-pregnant animals that are seronegative and/or PCR-negative (Guatteo *et al.*, 2008). When used in infected animals during the peri-insemination period, vaccination does not prevent C. burnetii shedding (Guatteo *et al.*, 2008) questioning vaccination in adults. Recently, two studies have examined the use of this vaccine during the dry period. Although it did not reduce shedding during postpartum period (Tutusaus *et al.*, 2014), the vaccine was able to improve the subsequent fertility of the herd, especially when applied to *C. burnetii* seronegative animals (López-Helguera *et al.*, 2013).

Concluding remarks

The use of more than one technique is recommended to increase the sensitivity of a positive diagnosis of *N. caninum*-associated abortion. Antibody titres determined using ELISA tests need to be quantitatively considered rather than just classifying seropositivity versus seronegativity. The use of semen from beef breeds in *Neospora*-infected dairy cows not only reduces numbers of *Neospora*-infected heifers as replacement animals, but also diminishes the risk of abortion.

Q fever has been scarcely reported in cattle, probably because of its difficult diagnosis at the farm level. The presence of *C. burnetii* in dairy herds has been not yet clearly demonstrated to negatively affect reproductive performance. Specific genotypes of the bacterium could explain the current controversy over this issue (Jado *et al.*, 2012).

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Carla Mendonça

Revisão de alguns procedimentos cirúrgicos em espécies pecuárias

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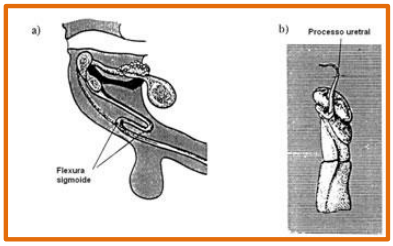
Obstrução urinária em ruminantes

- Causas
- Fatores predisponentes
- Tratamento médico
- Tratamento cirúrgico



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- **Na obstrução uretral total e rotura da uretra o tratamento cirúrgico é urgente, com risco de não ser possível a recuperação do animal após se instalar urémia ou consequente rotura da bexiga.**
- **O procedimento cirúrgico é simples , económico e exequível em regime ambulatório, permitindo a recuperação do animal.**

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Técnica cirúrgica

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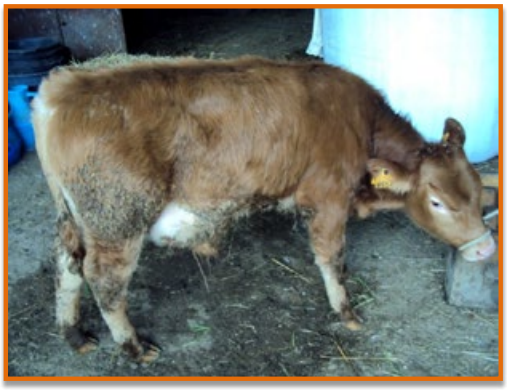
caso clinico

- Vitelo de 3 meses de idade da raça Minhota.
- Anorexia, prostração; relutância em permanecer em estação ; tumefação na região abdominal ventral caudal.



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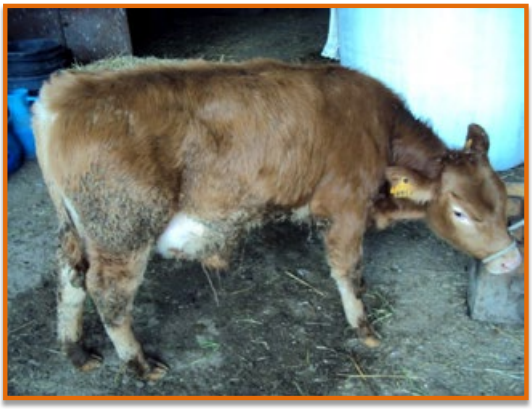
Exame clínico:

- Temperatura corporal 38,5ºC.,
- Desidratação ligeira (7%),
- Taquicardia,
- Frequência respiratória normal,
- Sem sinais sistémicos de urémia



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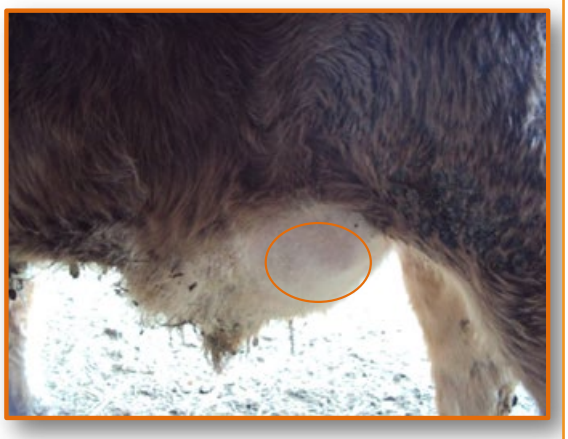
Exame clínico:

- Anúria (pêlos do prepúcio secos),
- Tumefação no abdómen ventral caudal, envolvendo pénis e escroto, já com sinais de isquémia a nível cutâneo



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•**Exame clínico:**
•Palpação da tumefação: compatível com edema (frio ao toque, sem dor, ficando impressa a pressão exercida com os dedos) .
•Punção com agulha de 18G: verificou-se presença de liquido sero-sanguinolento, com forte odor urémico.

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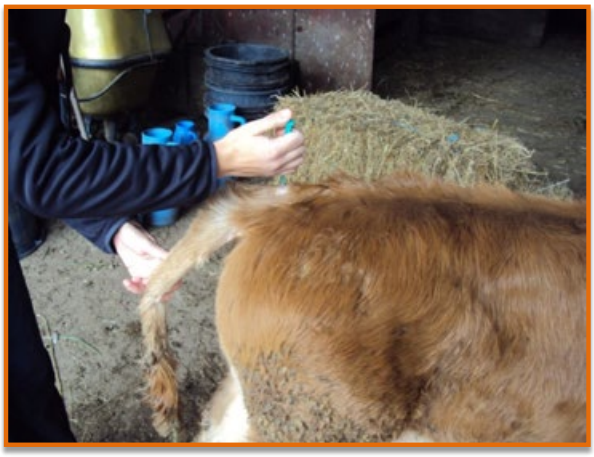

Anestesia: **manter o animal em estação**

- **Sedação ligeira (0,5 ml de xilazina a 2%, IM)**
- **Epidural baixa (2ml de lidocaína a 2%).**

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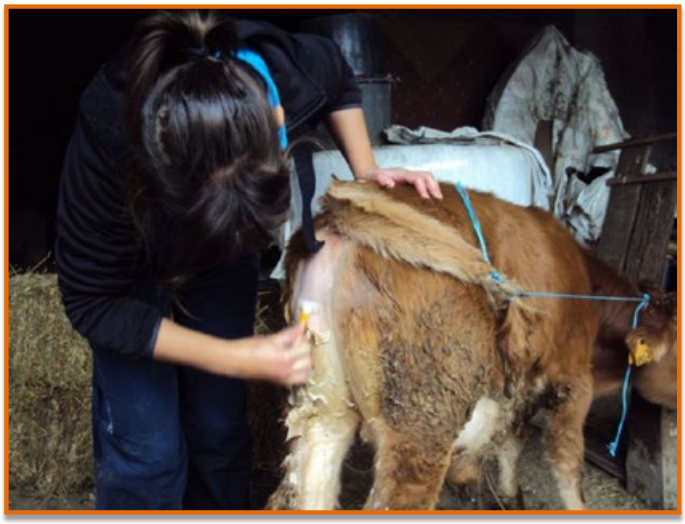

Tricotomia da região do períneo (do ânus à base do escroto, e 10 cm para cada lado da linha média do períneo).

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Assépsia cirúrgica: região do períneo, pré-lavagem (Povidona Iodada solução em espuma) e Anti-sépsia (solução dérmica de Povidona Iodada, intercalada com álcool a 96% vol).

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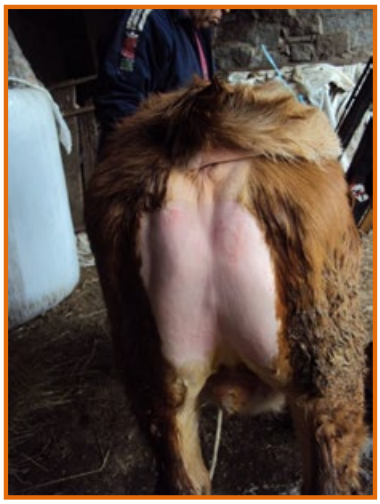
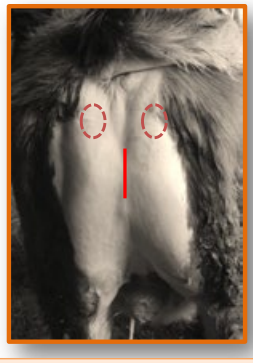
4 **Anestesia local e incisão cirúrgica:** Identificação do local de incisão e anestesia local, em linha com lidocaína 2%. / procaína.





A incisão realizou-se na linha média do períneo,
15 a 20 cm ventral ao ísquion.

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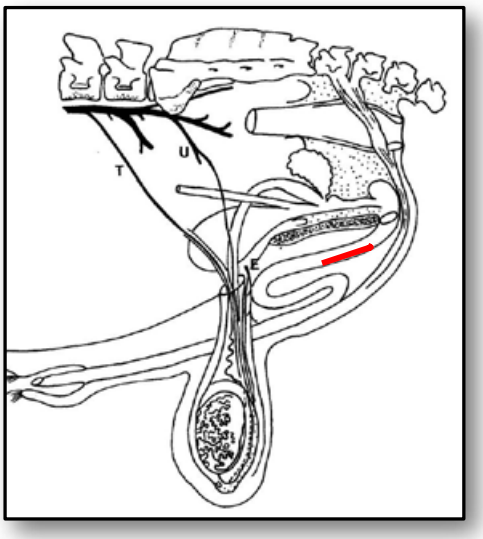

- Representação anatómica do aparelho reprodutor masculino: indicado a vermelho o local da uretrostomia, (2).

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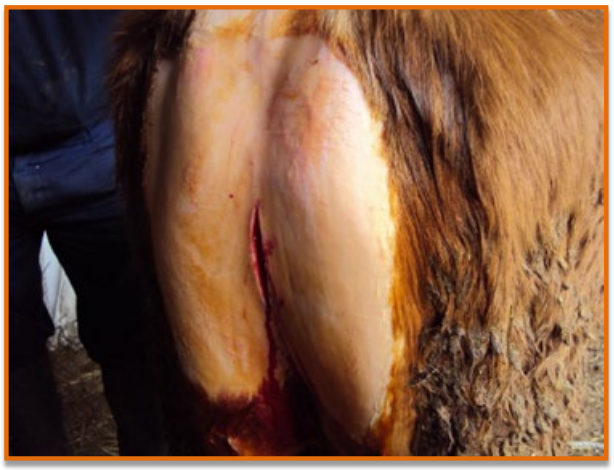

Incisão da pele , com 8-10 cm de comprimento.

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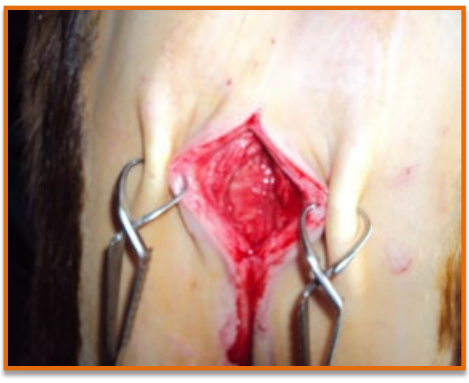

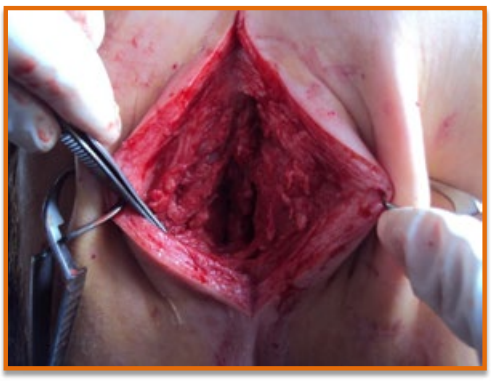

Dissecção romba dos tecidos subcutâneos, em profundidade (aprofundar a disseção 3-4 cm) , até identificação dos músculos retratores do pénis.

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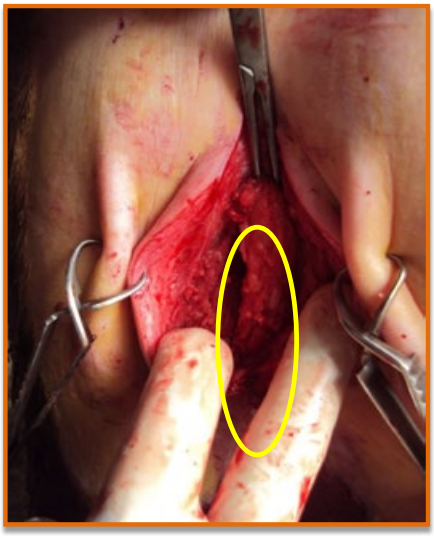

- Os músculos surgem como duas estruturas tubulares a envolver o pénis.
- É necessária a sua secção romba para permitir o fácil acesso e exteriorização do pénis .

Identificação e secção dos músculos retratores do pénis.

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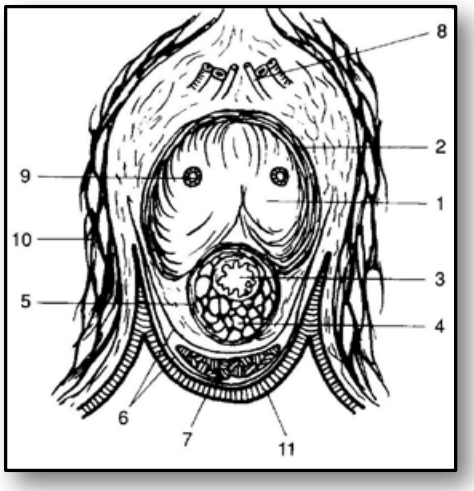

- Secção horizontal da região perineal, indicando as estruturas implicadas na uretrostomia: 1- corpo cavernoso; **2- túnica albugínea; 3- uretra; 4- corpo espongiforme; 5- túnica albugínea do corpo espongiforme;** 6- fáscia perineal ; 7- musculo retrator do pénis; 8- artérias, veias e nervos do pénis; 9- artérias profundas do pénis; 10 - músculos mediais da coxa; 11- pele .

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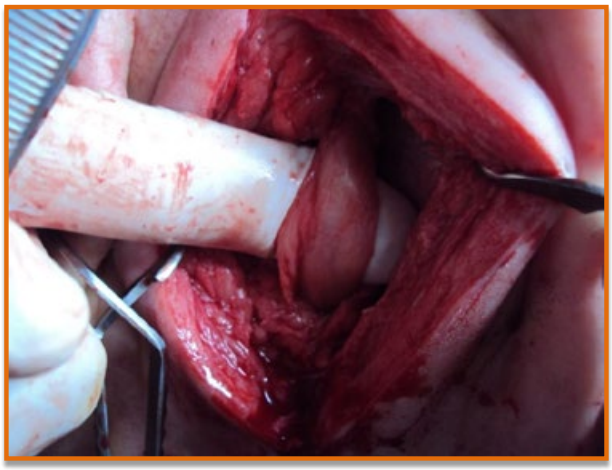

Identificação e individualização do pénis:

- De salientar que este localiza-se a uma profundidade de 6– 7 cm da superfície da pele, sendo necessário exercer bastante tensão para o exteriorizar.

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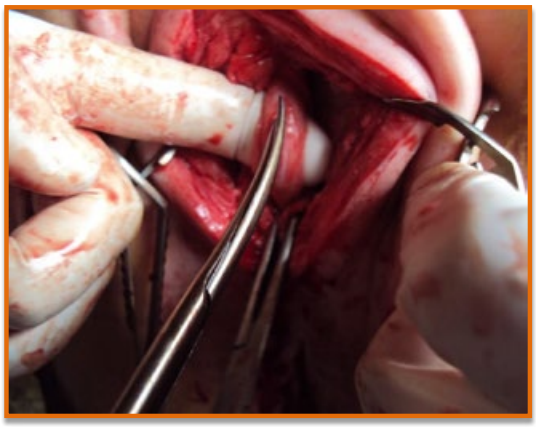
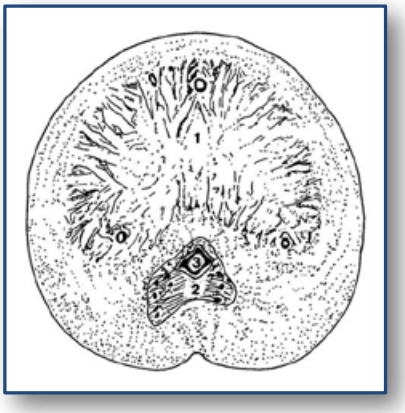


Acesso à uretra :

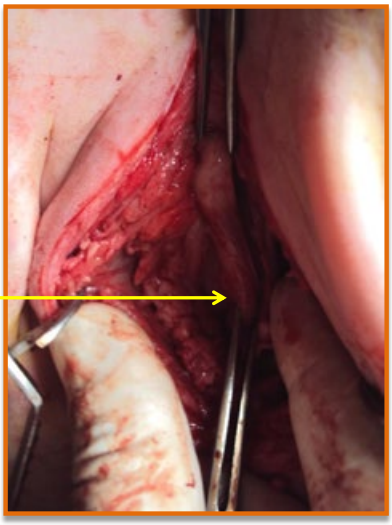
- Incisão longitudinal do pénis, precisamente na sua linha média .
- Incisão dos músculos cavernosos com 0,5-1 cm de profundidade, para aceder ao lúmen da uretra.
- Hemorragia será notória assim que se atinge o lúmen uretral.
- Prolongar a incisão da uretra distalmente em 5-6 cm.



Corte transversal do pénis a nível caudal:

- 1- corpo esponjoso;
- 2- corpo cavernoso;
- 3- uretra.

10



Penectomia : Necessidade de secção transversal do pénis, de modo a diminuir a tensão da sutura da mucosa uretral à pele .

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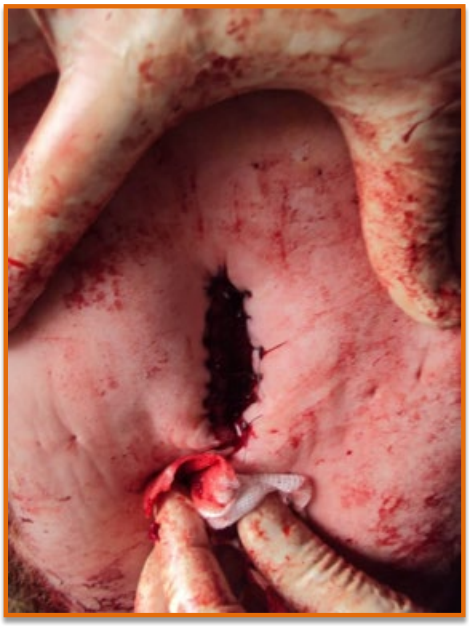
Transposição peniana perineal

- Incisão da uretra (5 cm de comprimento)
- Sutura da uretra à pele com material monofilamentar - Gliconato (Monosyn , Braun).
- Sutura inicial com 2 pontos, um cranial e outro caudal à linha de incisão.



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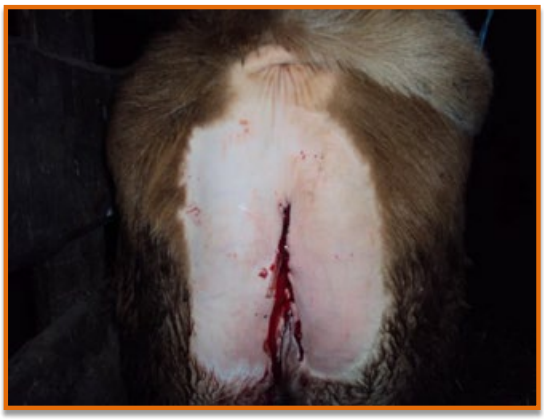

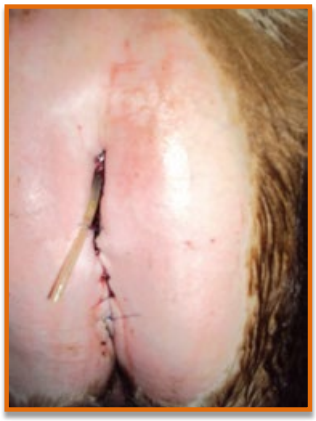

Colocação de cateter urinário de silicone .

- Fixação à pele e manutenção por 3 dias.
- A limpeza diária da sutura foi feita com solução de lavagem com Povidona Iodada de uso ginecológico.

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Incisões no tecido conjuntivo edemaciado no abdómen ventral caudal

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Terapêutica pós-operatória:

- Fluidoterapia: Nacl 0,9%
- Diazepan/ Butilescopolamina
- AINE (sem Insuf. Renal!!)
- Enrofloxacina

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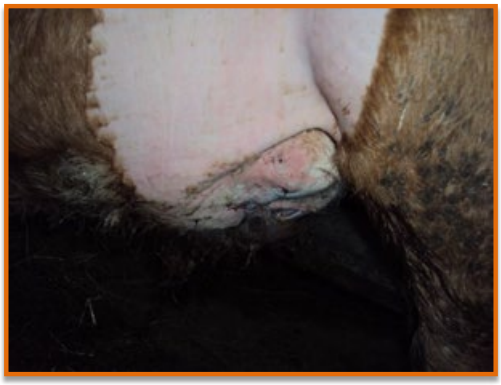
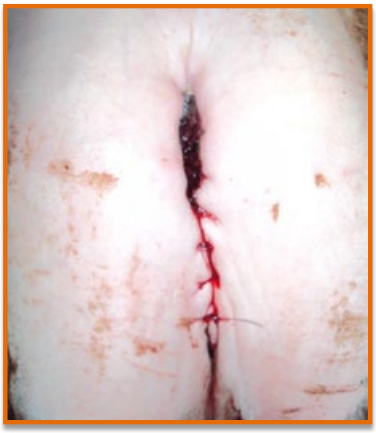
15 Aos 3 dias após a cirurgia:





Redução do edema abdominal ventral, sem necrose dos tecidos
Remoção do cateter urinário.

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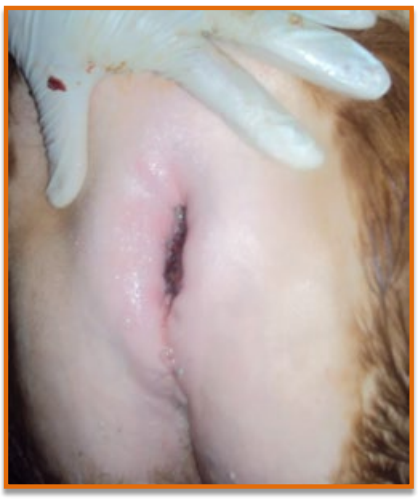
16 Aos 10 dias após a cirurgia:



- **Boa cicatrização,**
- **A sutura da pele foi removida**
- **Mantendo-se a sutura da mucosa uretral à pele .**

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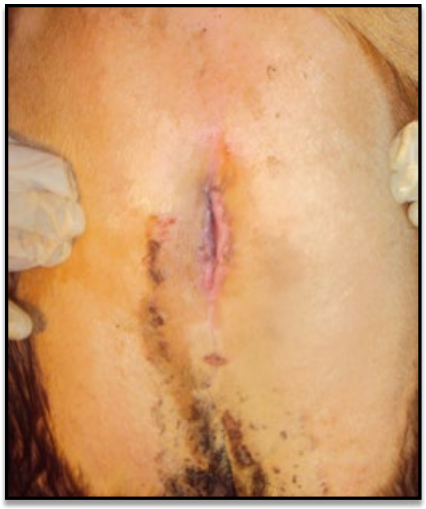
Aos 15 dias após a cirurgia:



Retirou-se finalmente a sutura da mucosa uretral.

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3 meses após a cirurgia:

O animal foi encaminhado para o abate com boa condição corporal.

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Urolitíase em pequenos ruminantes
Técnica cirúrgica

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Cirurgia Urogenital em Pequenos ruminantes

- Remoção divertículo uretral
- Uretrostomia perineal
- Cistotomia/ cistostomia

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I. Remoção divertículo uretral:

- **Efetivo em 50% dos casos**
- **Recidivas em 80% casos em algumas horas/dias**

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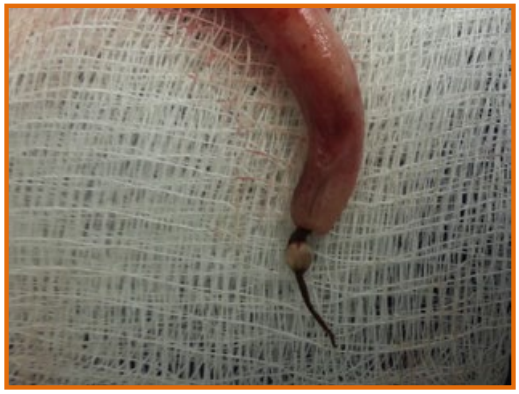
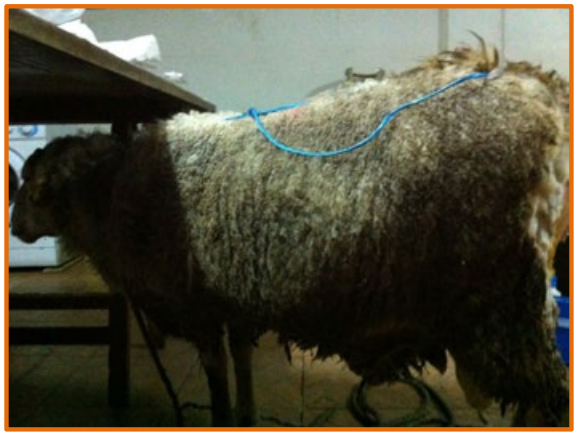


Remoção divertículo uretral:

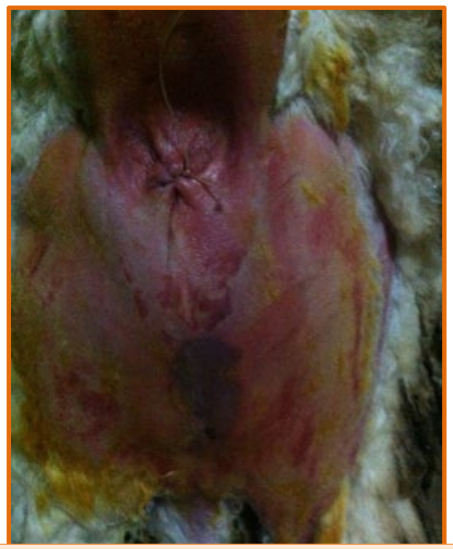
Exteriorização do pénis: lidocaina 2% no prepúcio
Remoção do processo uretral, obliquamente com recuso a tesoura

II. Uretrostomia perineal:

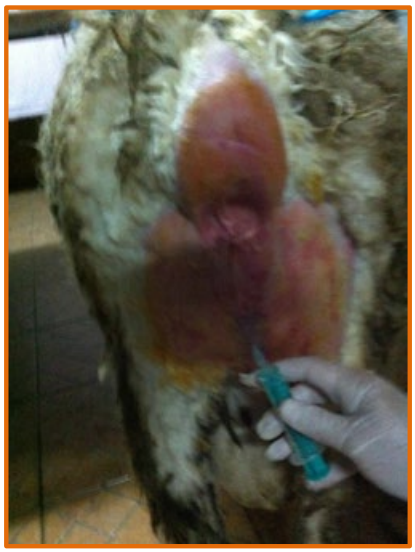
- Resposta a rotura uretral
- Cirurgia de emergência
- Risco de recidiva por estenose uretral



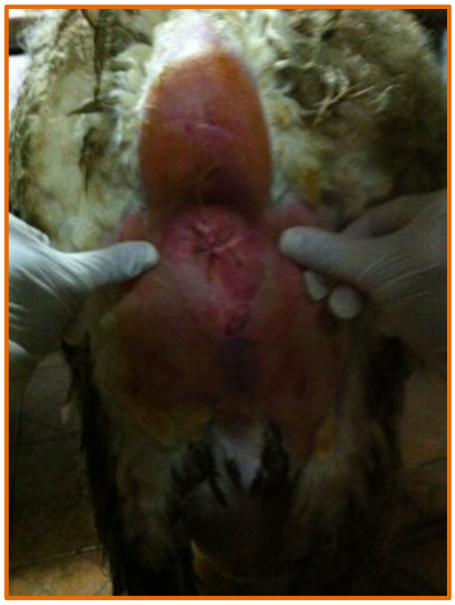
Animal em estação
Realização de anestesia epidural com lidocaína 2% (2,5mg/kg) / Procaína.

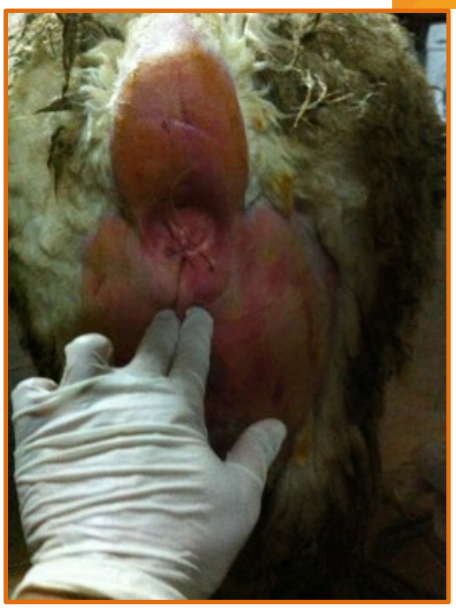


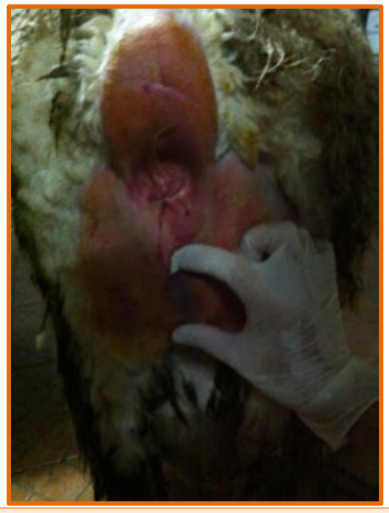
- Sutura em bolsa de tabaco no anûs
- Tricotomia e antissepsia da região perineal

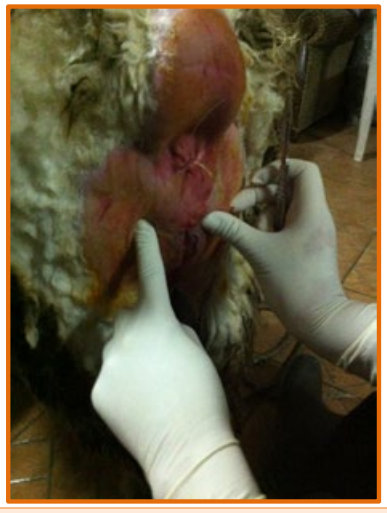


- Anestesia local em linha, lidocaína 2%/ Procaína

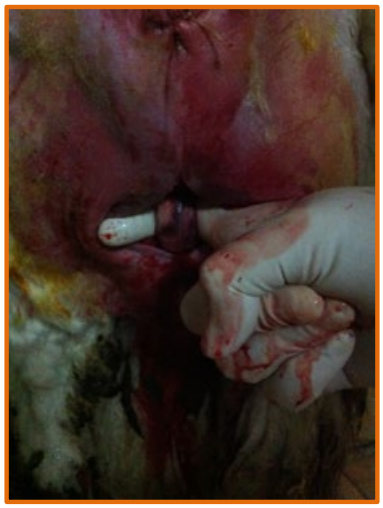


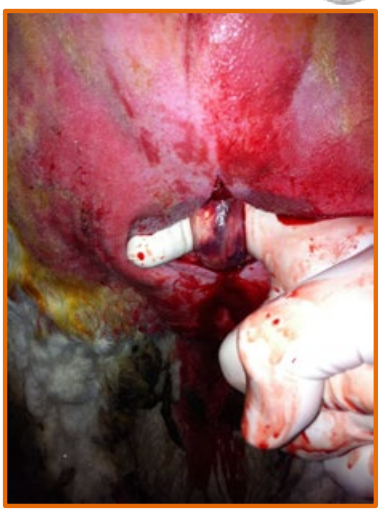






Realizada incisão longitudinal de 5 cm na pele da região ventral do períneo e disseção romba do tecido subcutâneo para exteriorizar o pénis.





Após secção transversal, o pênis é fixado à pele com dois pontos com fio absorvível sintético
Incisão longitudinal do pénis até atingir a uretra. Sutura da uretra á pele, sutura simples descontínua

III. Cistomia/cistostomia:

Dispendiosa

Pós-cirúrgico mais longo(2-3 semanas) em ambiente hospitalar

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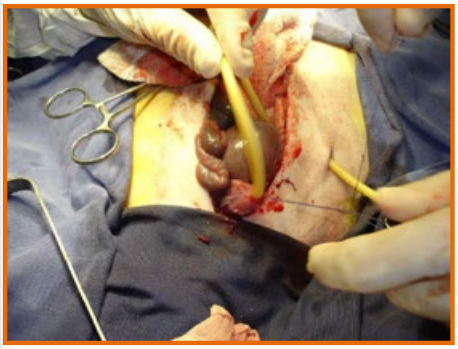
Cistotomia:

•Laparotomia, incisão de pele retro-umbilical paramediana direita, 2-3 cm paralelamente á linha média, estendendo-se cranialmente, aproximadamente 6 cm a partir da glândula mamária .

• Visualização da bexiga. Cistotomia na região dorsal da bexiga, onde se colocaram 2 suturas de apoio, proximal e distal à incisão.

• A bexiga é esvaziada com auxílio de material de sucção, lavada com solução fisiológica

•Deve ser realizada hidropropulsão retrógrada da uretra com cateter uretral



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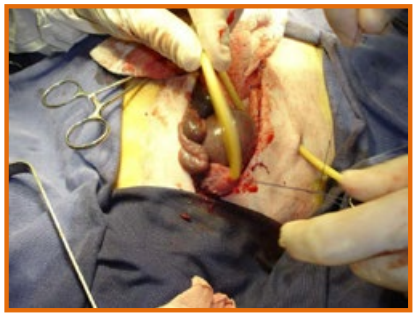
Cistostomia:

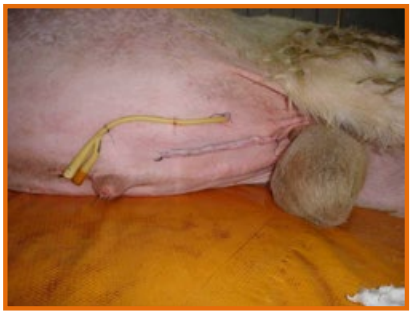
•Pequena incisão (1 a 2cm) de pele e musculatura, lateral à incisão paramediana, insere-se um cateter de Foley (16 a 24 French) até a bexiga.

•Fixar o cateter com uma sutura em bolsa de tabaco na parede.

• Extremidade distal do cateter de Foley que possui o balão foi inserida no interior da bexiga

•Vesicopexia, à parede abdominal, em vários pontos.





Cirurgia Urogenital em Ruminantes

Resolução cirúrgica de abcesso do canal do úraco em um novilho

Centro de Congressos de Lisboa, 26 e 27 novembro 2016

Canal do úraco persistente

A infeção dos remanescentes umbilicais com envolvimento intra-abdominal pode envolver a **veia umbilical** (onfaloflebite – Fig. 1.A), as **artérias umbilicais** (onfaloarteríte – Fig.1.B) e/ou o **canal do úraco** (infeção e abcesso – Fig. 1.C).

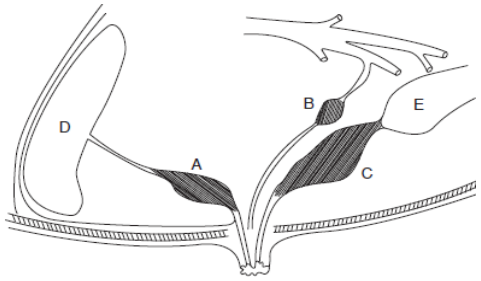


Figura 1: A- Veia umbilical; B- Artéria umbilical; C- **Canal do ùraco**; D- Figado; E- Bexiga.

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Abcesso do canal do úraco

Clinicamente patente em vitelos com 1-2 semanas de idade
Com exteriorização de uma secreção purulenta do umbigo
Sinais clínicos: disúria, polaquiúria, piúria e cistite.

Infeção persistente em vitelos com mais idade é indicativa para tratamento cirúrgico, podendo culminar com cistite cronica.

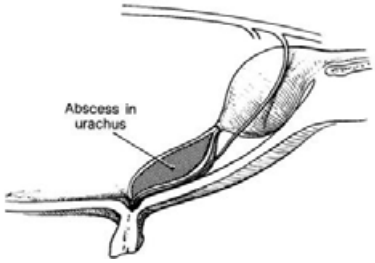

Figura 2: Imagem esquemática de abcesso do canal do úraco

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Caso clínico

- Vitelo de **6 meses** de idade da raça Holstein-Frísia.
- Diminuição progressiva de apetite e perda de condição corporal (há cerca de 2 semanas)
- Sinais de cólica intermitente
- Região umbilical tumefacta com a presença de um corrimento purulento
- Hipertermia

Figura 3. Secreção purulenta exteriorizada

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Resolução cirúrgica de abcesso do canal do úraco

1

ANESTESIA:

Decúbito dorsal, sob fluidoterapia contínua (catéter de 23G na veia marginal da orelha), Anestesia: Xilazina IM + Quetamina IV +Butorfanol.

•

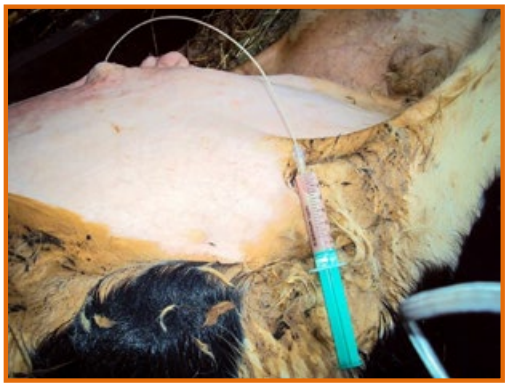

Figura 4: Aspiração da secreção purulenta, através da fístula umbilical.

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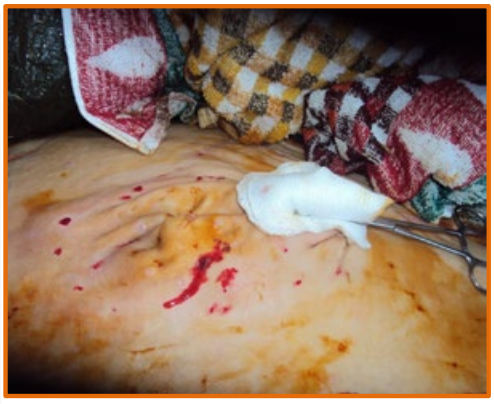
Resolução cirúrgica de abcesso do canal do úraco

2

Assepsia e antissépsia:

assepsia cirúrgica da região do umbigo (Solução em espuma de povidona iodada) e Antissepsia (solução dérmica de povidona iodada, intercalada com álcool a 96%).



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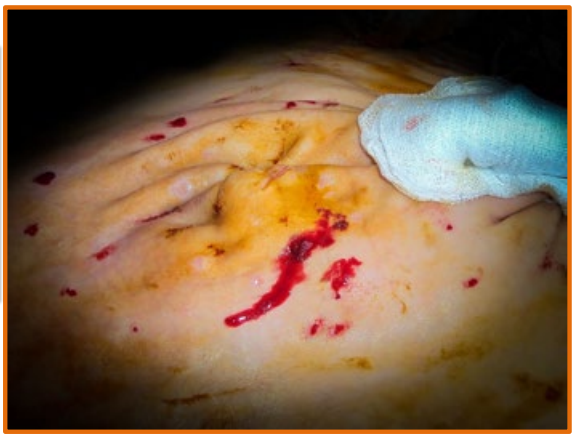
3

Colocação pinça de Allis no prepúcio, de forma a salvaguardar a assépsia do campo operatório.
Anestesia local em linha (Lidocaína a 2%).



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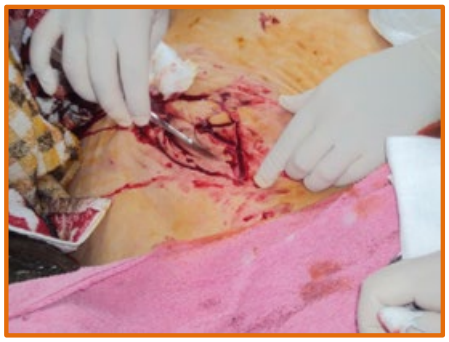

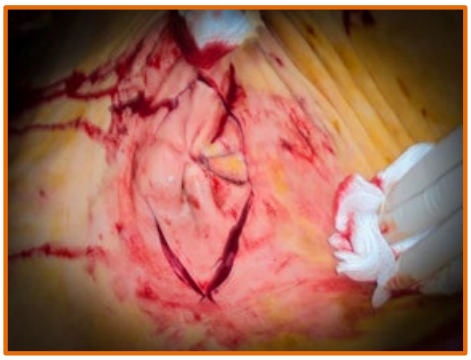

Incisão elíptica a envolver cicatriz umbilical, estendendo-se caudal e paralelamente ao prepúcio.

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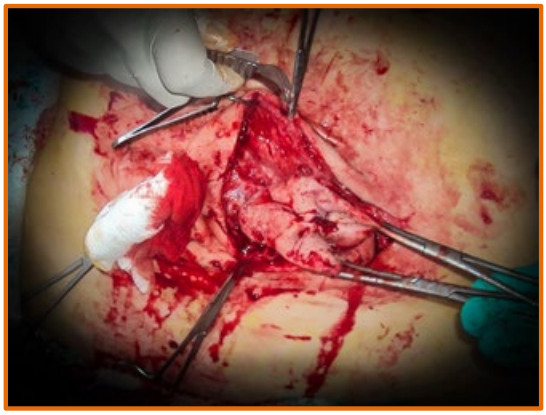

Desbridar o tecido subcutâneo, mantendo a ligação entre a pele e os remanescentes do cordão umbilical.

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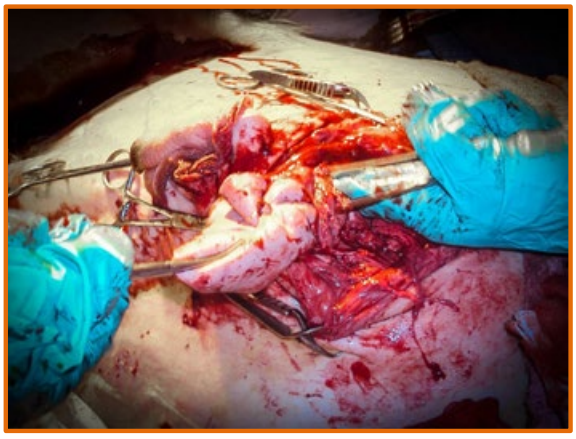

Incisão das camadas musculares com acesso à cavidade abdominal pela linha alba.

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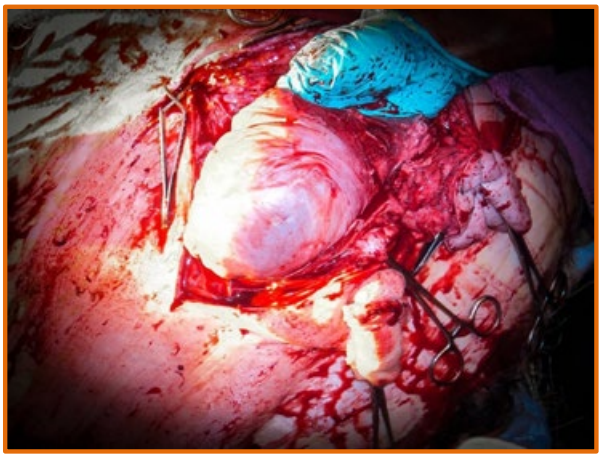

Ao realizar a laparotomia exploratória, infeção e abcesso do canal do úraco.
Procedeu-se à disseção romba do canal do úraco desde a cicatriz umbilical até á sua inserção na parede vesical.

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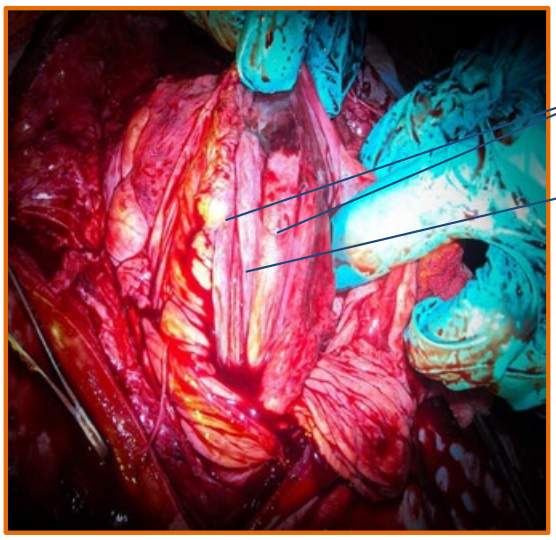

Identificação das estruturas

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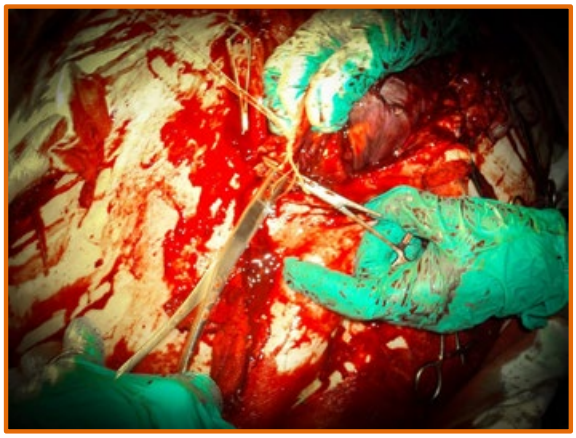

As artérias foram laqueadas com sutura de transfixação.

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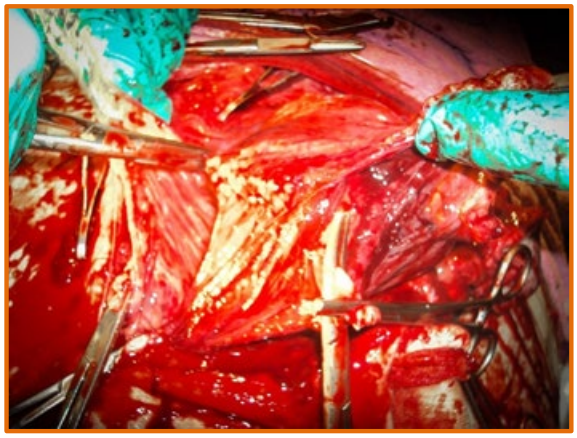

Devido à comunicação entre o abcesso e o lumen vesical, foi necessário a excisão do ápex da bexiga, de forma a garantir a remoção total das estruturas envolvidas e a reduzir o risco de infeção peritoneal.

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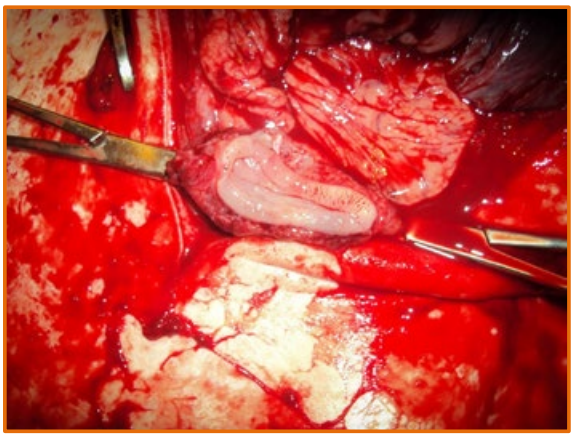

Procedeu-se cistectomia

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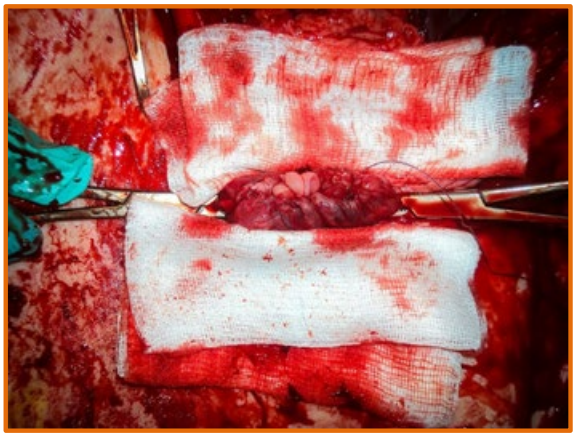

Procedeu-se a sutura invaginante, dupla, não perfurante da parede vesical (Sutura de Cushing) com gliconato, 1USP),

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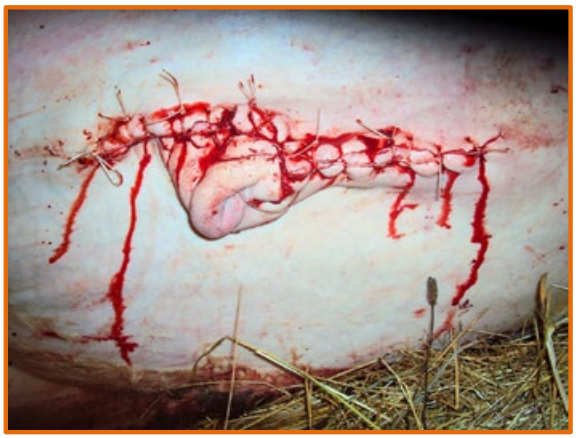

Sutura simples interrompida das camadas musculares e sutura de aposição do tecido conjuntivo com Catgut crómico, 3USP, Sutura simples descontinua da pele, com Caprolactam , 2 USP.

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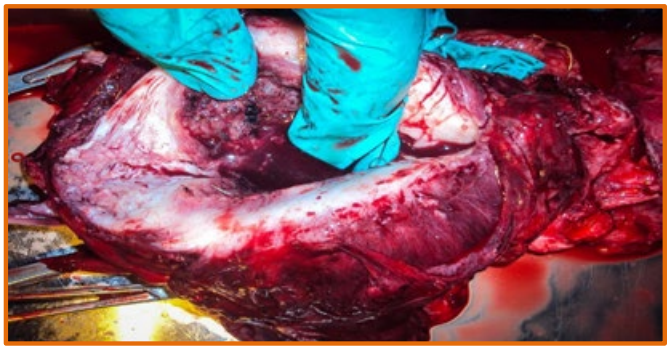
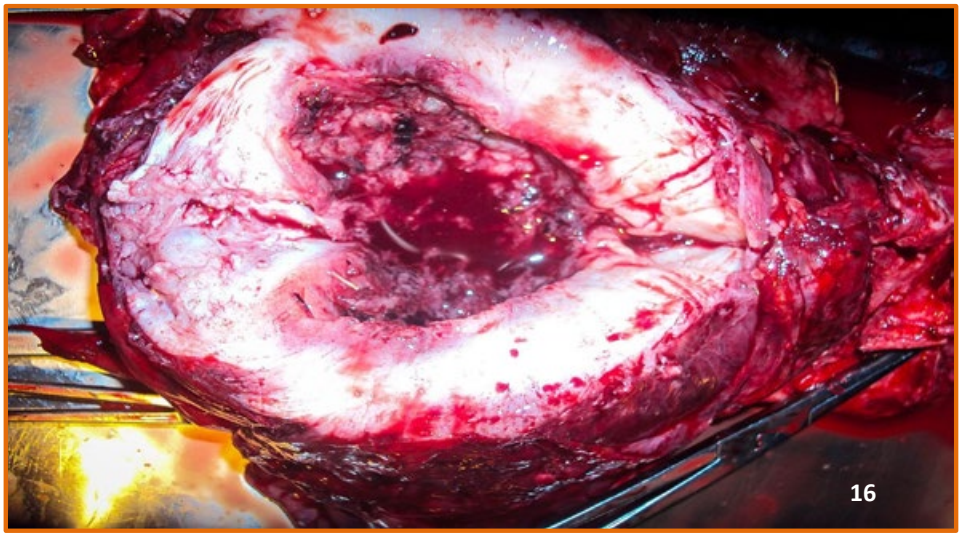
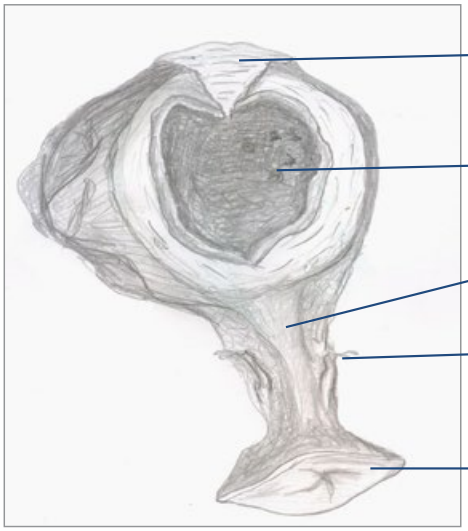
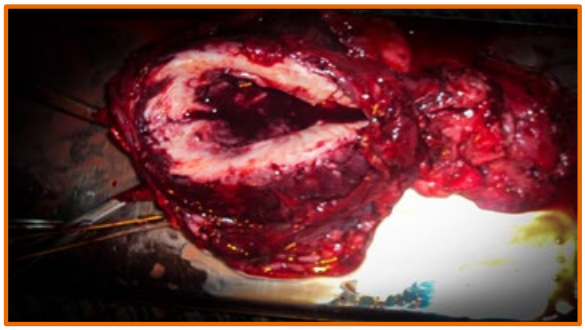
Acompanhamento pós-operatório:
Antibioticoterapia sistémica com enrofloxacina , SID durante 15 dias ; carprofeno, SID, durante 5 dias, para controlo da dor.



A dissecção da massa extraída cirurgicamente : absesso (conteúdo pio sanguinolento) de cerca de 12 cm de díâmetro e parede capsular com cerca de 4 cm de espessura . Esta estrutura envolvia não só o úraco, como também a parede vesical







O abcesso do canal do úraco evidencia-se clinicamente em vitelos mais jovens (1-2 semanas de vida) sendo muito raro a sua exibição clínica em vitelos com idade tão avançada, o que tornou todo o procedimento cirúrgico mais moroso e complexo. Em contrapartida, o risco de ruptura do abcesso e consequente peritonite , neste caso foi reduzido devido à espessa cápsula fibrosa que o circunscrevia.

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Cirurgia Urogenital em Ruminantes

Histerectomia decorrente a prolapso uterino irredutível: Espécies pecuárias

Carla Mendonça, Marisa Bernardino, José Ferreira Neves

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Tratamento

I. REDUÇÃO

II. AMPUTAÇÃO

III. EUTANÁSIA

Quando considerar a Histerectomia em espécies pecuárias?

- Prolapso uterino irreduzível.
- Hemorragia grave
- Torção uterina com isquémia do órgão.
- Trauma e lacerações extensas da parede uterina.

Vantagens

- Permite a sobrevivência do animal,
- Procedimento relativamente fácil :
  - mas não descurar os riscos inerentes
  - Ter em atenção Choque Hipovolémico e tratá-lo.
- Material necessário: acessível e económico

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Material

- Solução anti-séptica
- Bisturi, tesoura,
- Agulha grande redonda atraumática
- Material Sutura não absorvível
- Lidocaina

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Material

- Elástico contenção / Câmara de ar

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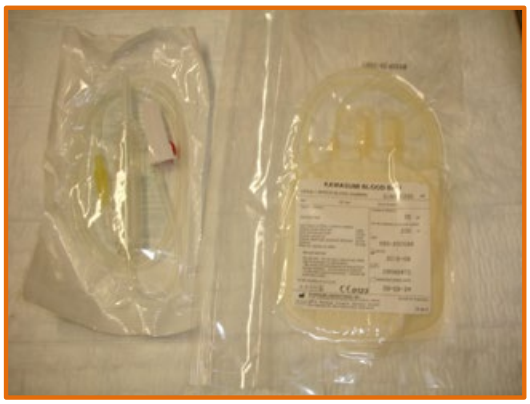
Material

- Kit de transfusão:



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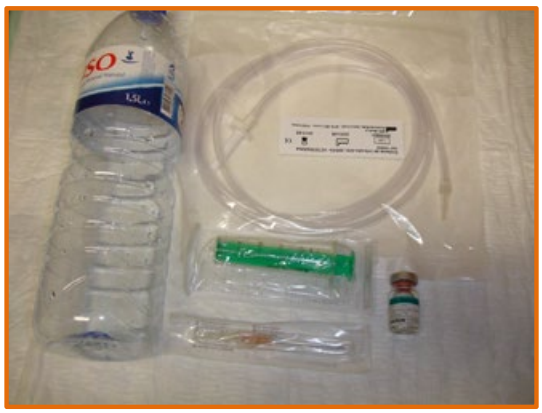
Material

- Na falta de Kit Transfusão



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INDICAÇÕES:

1. **Trauma extenso da parede uterina**
2. **Involução do cervix**
3. **Edema e congestão pronunciada do órgão**
4. **Prolapso tardio (até 1 semana após o parto)**
5. **Risco de morte por hemorragia – ruptura artéria uterina**

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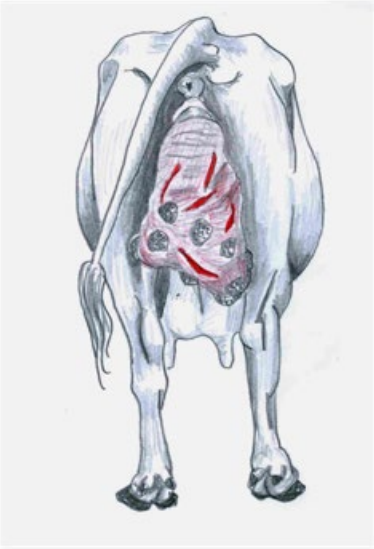
1. Trauma extenso da parede uterina:

A manipulação decorrente da redução do prolapso, irá ampliar a extensão das lesões



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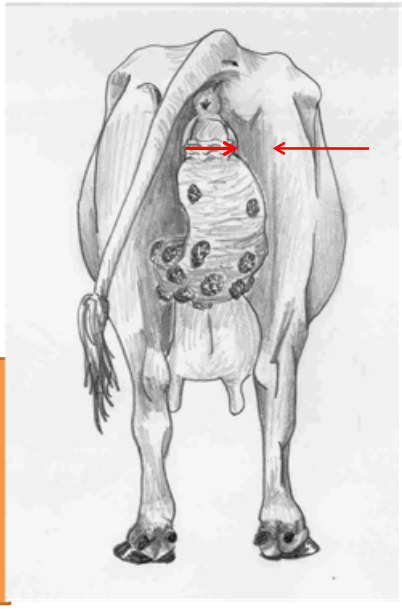
2. Involução do cervix



- **O que impossibilita recolocação do órgão na cavidade abdominal, especialmente se o prolapso ocorreu há algum tempo e se o edema for acentuado.**

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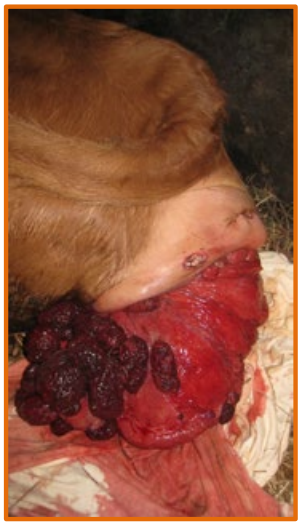
3. Edema e congestão pronunciada do órgão



- Tornam o endométrio mais friável e ao toque aumenta o risco de trauma e hemorragia
- Grande volume do órgão dificulta a redução
- Pouca maleabilidade da parede uterina dificulta a redução
- Desvitalização do endométrio

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4.Risco de morte por hemorragia

- Ruptura da artéria uterina em animais nervosos que se debatem durante a imobilização
- Animal encontra-se moribundo, qualquer tentativa de redução uterina pode resultar em morte, **amputação imediata** pode salvar!!

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CONSIDERAÇÕES

- Risco de ruptura da artéria uterina é superior em animais nervosos, de difícil contenção e em estação
- Versão de vísceras abdominais (Bexiga, intestinos)
- Choque hipovolémico/ Hemorrágico

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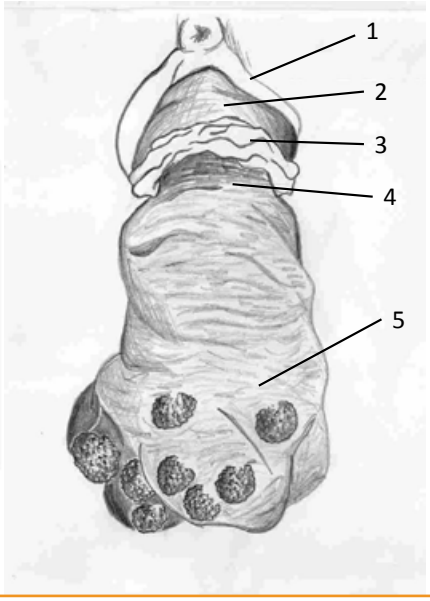
Assepsia Cirúrgica

1. Lavar útero e períneo com solução anti-séptica diluída.

2. Se persistir a suspeita de versão da bexiga e se a sua recolocação na cavidade abdominal não foi possível:



1 – Vulva; 2 – parede vaginal; 3 – cervix; 4 – corpo uterino; 5 – corno uterino

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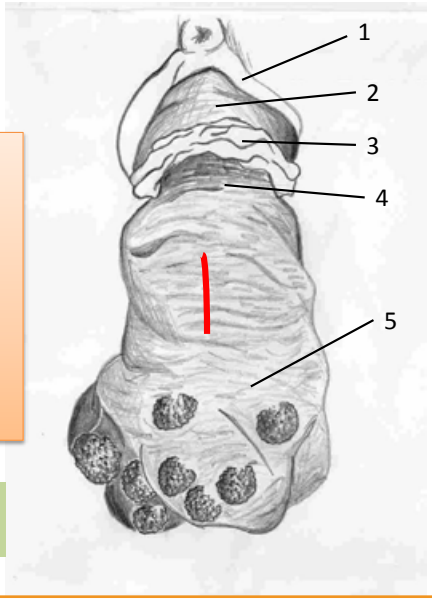
3. Se o grau de repleção da bexiga não permitir a sua recolocação na cavidade abdominal, promover esvaziamento da bexiga:
   - Cateterismo
   - Cistocentese



1 – Vulva; 2 – parede vaginal; 3 – cervix; 4 – corpo uterino; 5 – corno uterino

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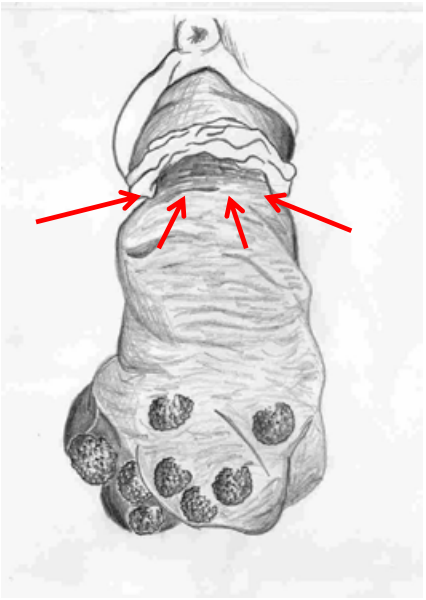
4. Colocar Garrote:

Local de colocação de garrote :

- Posteriormente ao cervix
- Anterior as carúnculas



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4. Colocar Garrote:

- A fita deve ser puxada até permitir uma redução de **50% do diâmetro original.**

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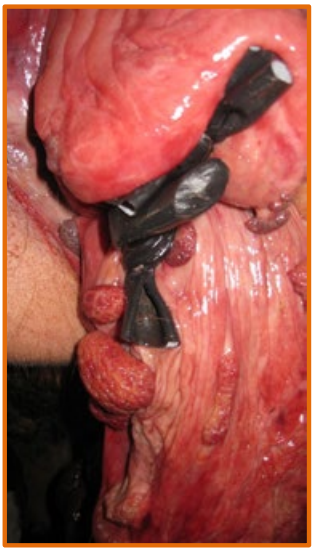
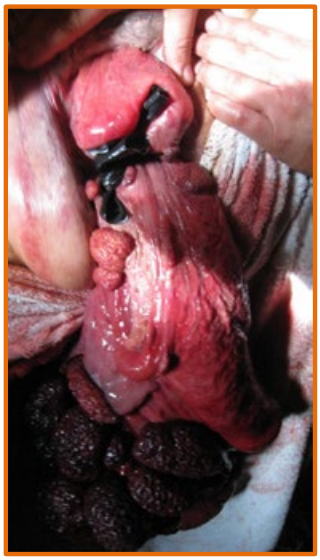
4. Colocar Garrote:





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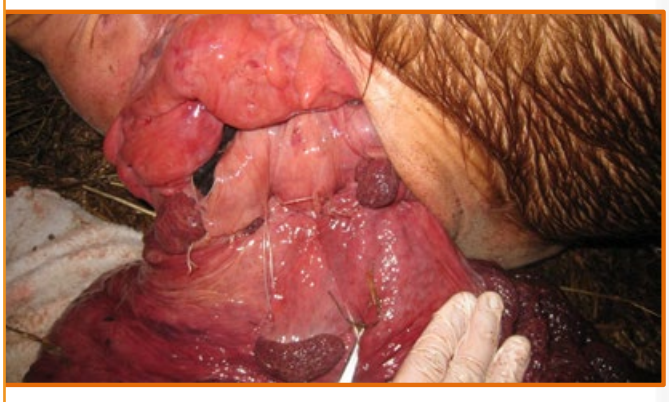
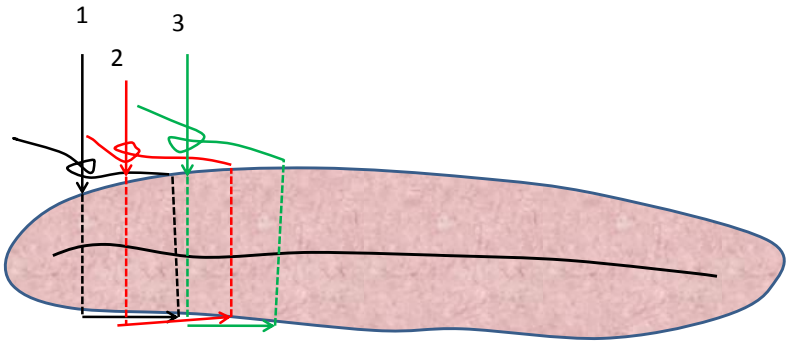
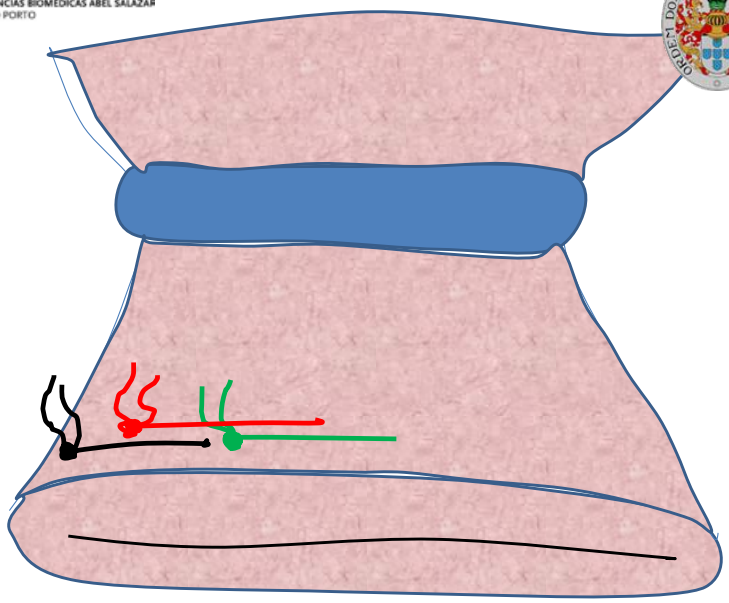
5. Acrescentar vários pontos de tensão abaixo do garrote, intercalados e proceder à excisão parcial e alternada com a aplicação da sutura.





Pontos de tensão , intercalados, atravessam totalmente a parede uterina; em que o ponto seguinte inicia-se a metade do ponto anterior.



Excisão 5 a 10 cm abaixo da sutura de tensão

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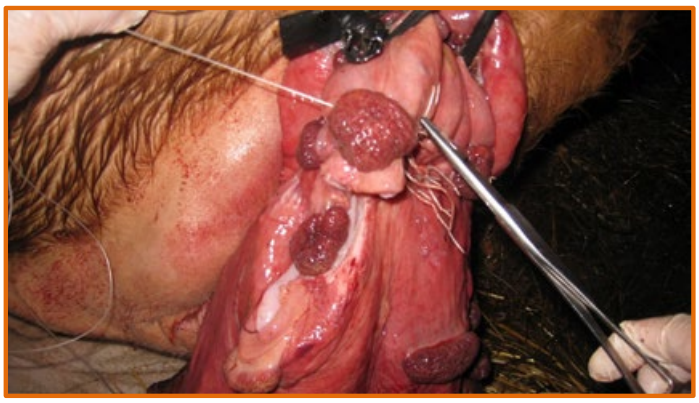
6. Proceder à excisão parcial alternadamente com a aplicação da sutura.



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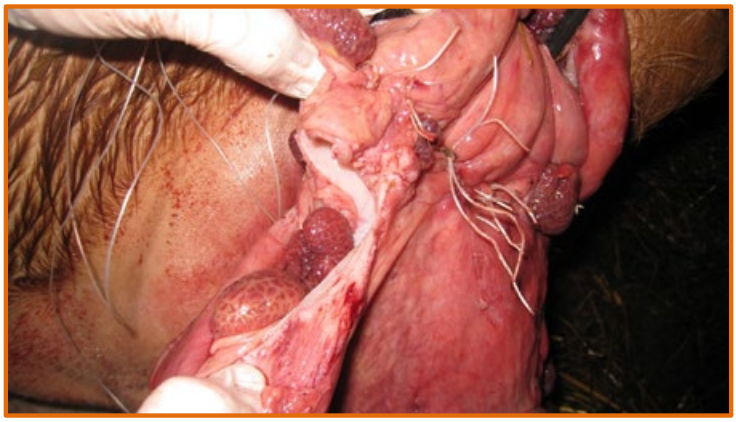
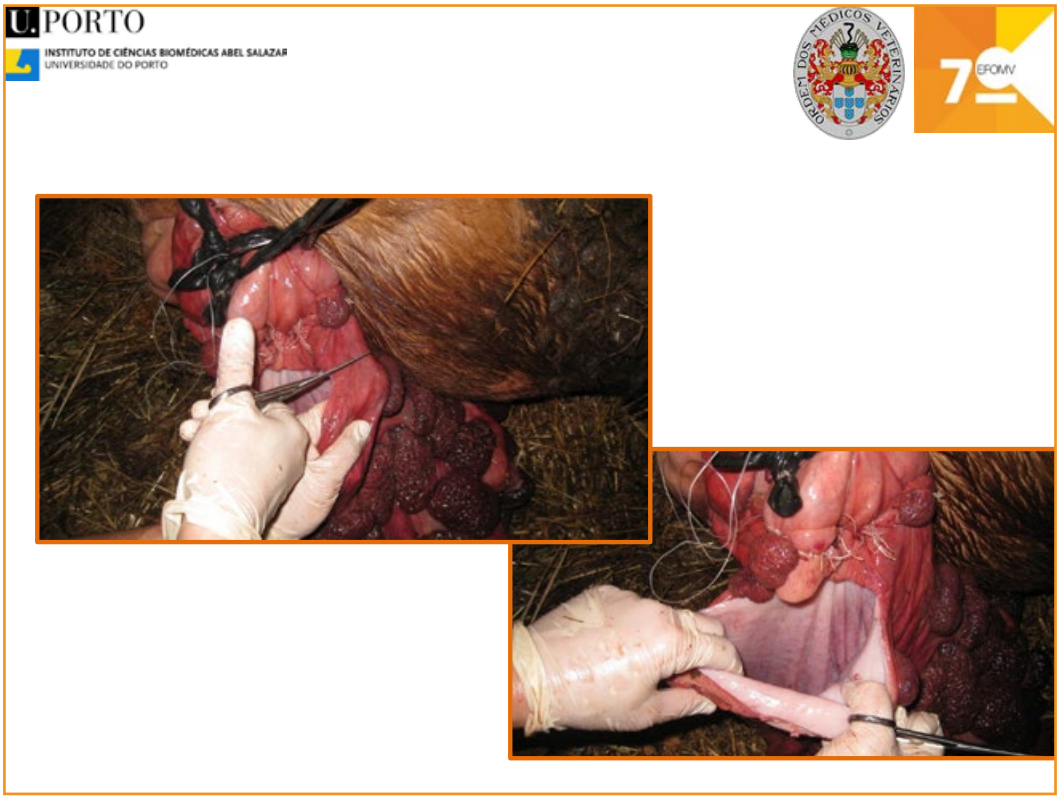
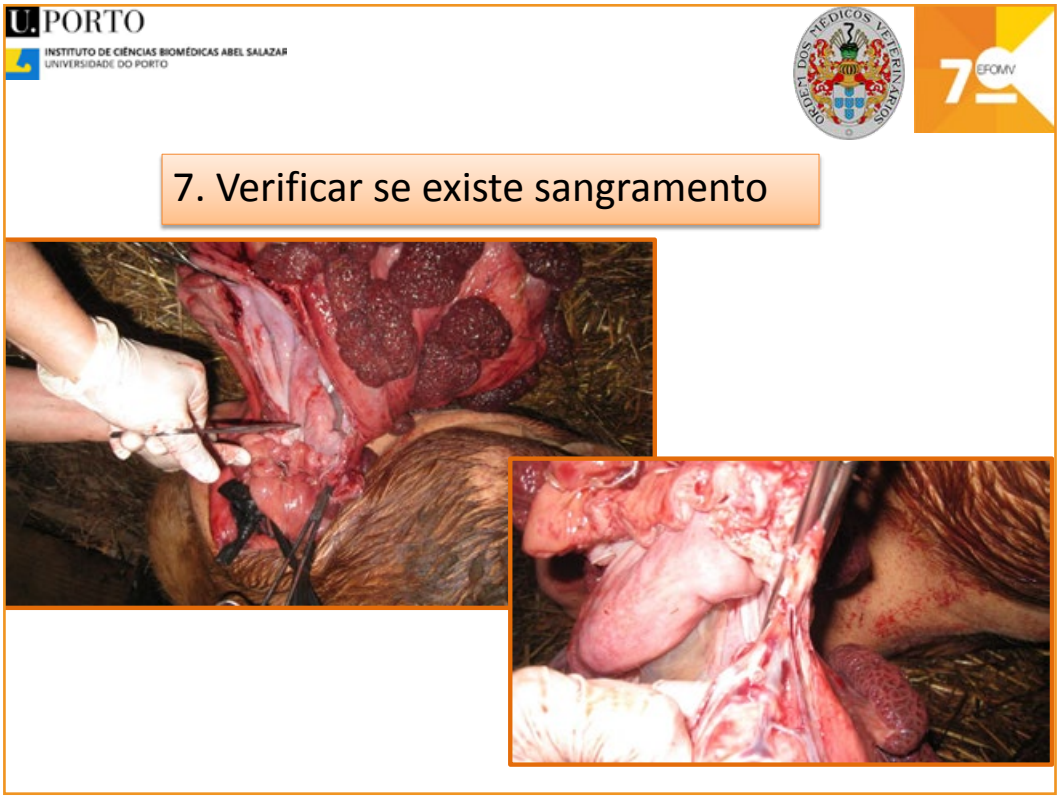
6. Permite laquear rapidamente vasos de grande calibre que não tenham ficado laqueados pela sutura de tensão.






7. Verificar se existe sangramento

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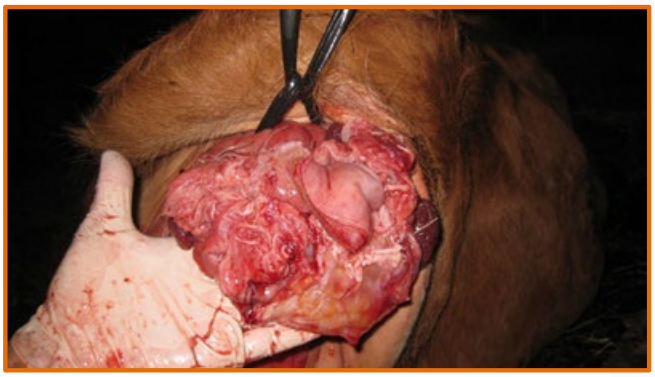
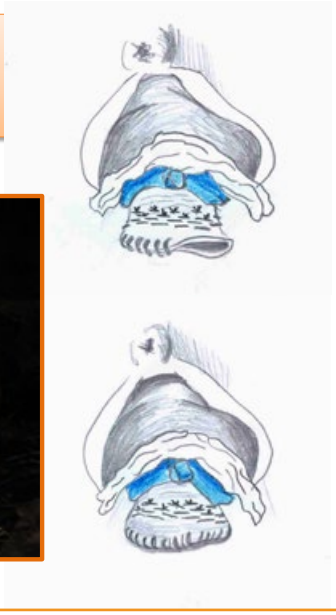
8. Aproximação dos bordos da parede uterina





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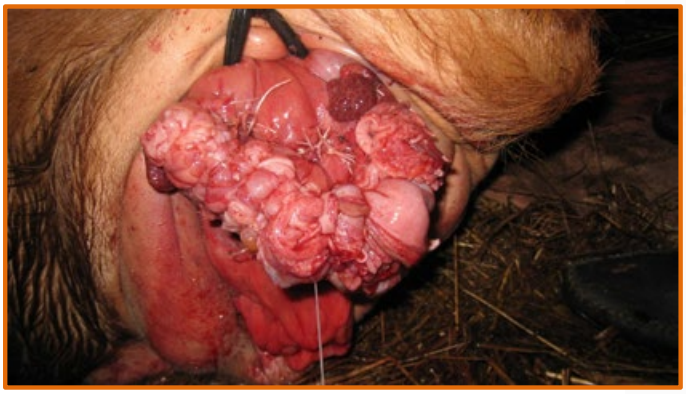
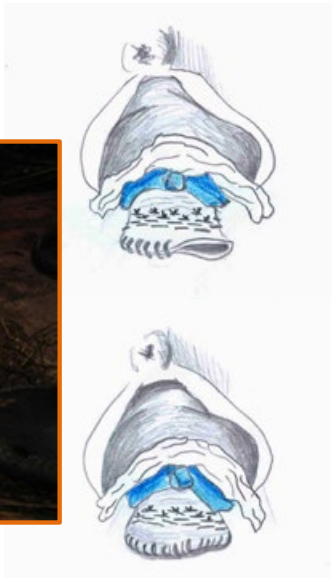
9. Reduzir o prolapso vaginal





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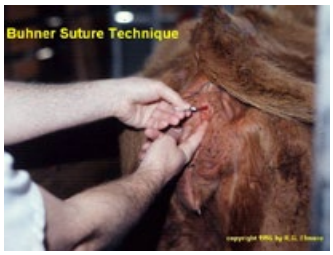
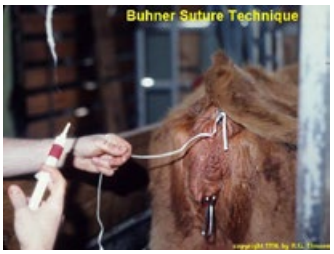
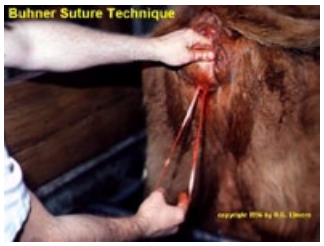
10. Sutura de contenção eventualmente







Sutura de contenção: Sutura Buhner

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Outros cuidados

- Antibioticoterapia
- **Corrigir hipovolémia:**
  - Fluidoterapia
  - Transfusão sanguínea

	Bovinos Leite	Bovinos carne	Pequenos ruminantes	Suínos
Numero casos	6*	4	7	4
Transfusão	5	0	0	0
Sobrevivência	6	3	5	2

Histerectomia- pequenos ruminantes

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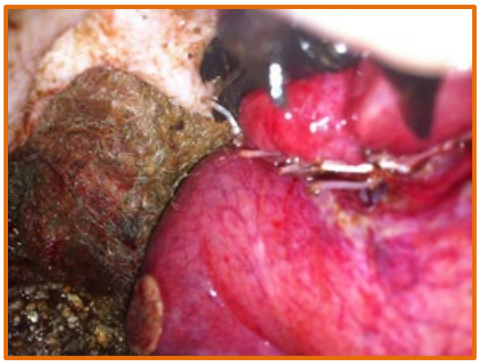

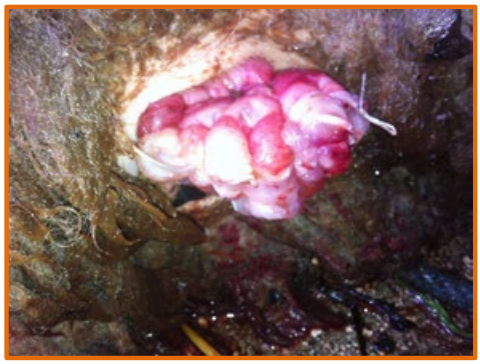

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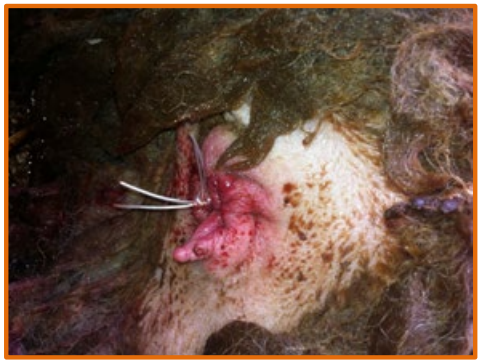
Sutura de contenção: ponto colchoeiro vertical



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Histerectomia- suínos

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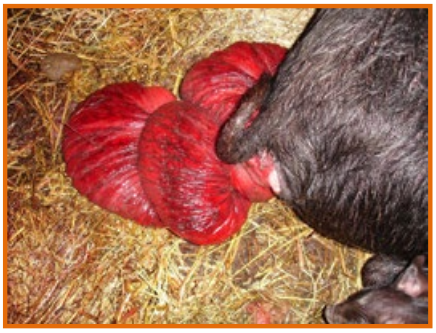
Prognostico:

Muito Reservado



Na grande maioria dos casos a morte ocorre devido a choque hipovolémico (hemorragia e congestão do órgão)

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Procedimentos cirúrgicos:

•**Fluidoterapia** (veia marginal da orelha):

✓Soro fisiologico isotonico ou Lactato de Ringer

✓Permite combater o choque hipovolémico,

✓Permite administração do anestésico (Ketamina)

✓Permite manter uma via aberta para medicação de urgência

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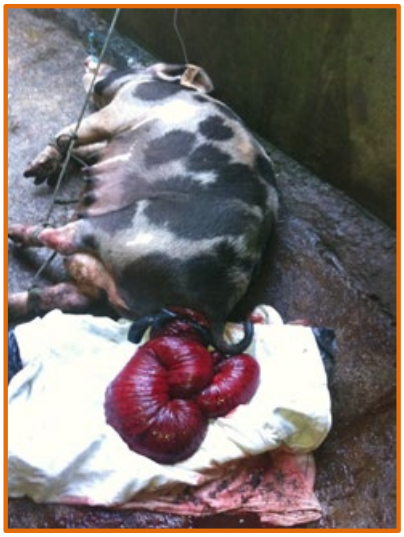
•Imobilização física da paciente; Limpeza do útero



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OBRIGADA !

Luís Lopes da Costa

Controlo da Campilobacteriose Genital Bovina (CGB) na exploração aleitante e feedlot

CIISA – Centro de Investigação Interdisciplinar em Sanidade Animal
Faculdade de Medicina Veterinária, Universidade de Lisboa

Introdução

A Campilobacteriose Genital Bovina (CGB) é uma doença venérea, sexualmente transmitida pelo coito e/ou sémen de touros infectados, portadores assintomáticos e disseminadores da doença na população susceptível. A doença tem distribuição mundial e a sua prevalência pode ser elevada nos sistemas de produção extensiva com recurso à monta natural.

A bactéria Gram negativa causal, *Campylobacter fetus subs. venerealis* (Cfv) é exclusivamente transmitida pela via venérea (ou através de sémen infectado utilizado em inseminação artificial (IA)) e ocasiona infertilidade devida a endometrite, morte embrio-fetal e aborto. A espécie *Campylobacter fetus*, inclui ainda a subespécie *Campylobacter fetus subs. fetus* (Cff), que pode ocasionar aborto esporádico na espécie bovina e aborto enzoótico na espécie ovina, por transmissão oral, ocorrendo a infecção da placenta por via sistémica.

No touro, a Cfv aloja-se sobretudo nas pregas prepuciais, mas também no corpo e glande peniana, infectando-se o sémen na sua trajectória ao longo da uretra. Os touros mais velhos (idade > 5 anos) apresentam maior desenvolvimento das pregas prepuciais, tendo a potencialidade de albergar uma maior população da bactéria, o que aumenta a sua probalidade de induzir a infecção em fêmeas susceptíveis. Na vaca ou novilha, a Cfv invade a vagina e coloniza o útero originando endometrite. Em caso de gestação, a infecção do embrião, ou do feto e placenta, induz mortalidade embrio-fetal e aborto.

Diagnóstico: Binómio Doença e Bactéria

O conhecimento sobre a patogénese da doença apresenta ainda algumas lacunas relevantes. A relação entre observação clínica da CGB e diagnóstico laboratorial da Cfv pode não ser facilmente perceptível. Nem todos os casos de diagnóstico laboratorial positivo são acompanhados de diminuição muito significativa da fertilidade na vacada. Os efeitos observados nesta dependem de múltiplos factores como a proporção de touros infectados, a sua idade e grau de infecciosidade, a existência de fêmeas susceptíveis sem imunidade adquirida de contágios anteriores, a presença de produtos de aborto de elevada infecciosidade, assim como aspectos de maneio entre os quais o encabeçamento da exploração, a relação macho: fêmea e a condição corporal. A interligação com a ocorrência de outras doenças infecciosas com impacto na reprodução como o IBR ou o BVD é desconhecida.

Neste contexto, o diagnóstico deve considerar o binómio doença e bactéria causal. A introdução de um touro ou mais touros infectados numa vacada indemne provoca a infecção das fêmeas (pode atingir 100% da vacada) e a doença surge sob a forma epizoocótica, pois as fêmeas recém-infectadas podem infectar os touros sãos existentes (que ficam persistentemente infectados). Isto origina uma diminuição significativa da fertilidade da exploração (ob-

servámos valores de 20%-50%) e o aumento do intervalo entre partos, assim como a dispersão acentuada da distribuição de partos. Nas fêmeas, a infecção pode ser depurada após 3 a 6 ciclos éstricos (2 a 5 meses) subsequentes à infecção primária (se não houver re-infecção), tornando-se as fêmeas mais resistentes à re-infecção em épocas reprodutivas ulteriores. Na ausência de introdução de animais (touros e fêmeas) novos na vacada, isto pode conduzir a uma situação de endemia, havendo alguma recuperação da fertilidade (eventualmente até 60%-80%). No entanto, a persistência da Cfv nos touros e em algumas fêmeas constitui um foco de contágio para novos animais e constitui um entrave à venda de reprodutores.

O diagnóstico da doença assenta pois na avaliação dos indicadores reprodutivos da exploração (o que pressupõe a existência de registos fidedignos) e na avaliação dos seus efeitos clínicos - mortalidade embrio-fetal e aborto no primeiro e menos frequentemente segundo trimestre de gestação. A observação de repetições nas montas é um sinal clínico relevante, porém na prática difícil de concretizar na exploração em extensivo. A detecção da Cfv é sobretudo realizada nos touros e nos produtos de aborto.

A técnica original "*gold standard*" de diagnóstico da Cfv é o seu isolamento do smegma prepucial e/ou sémen do touro, do muco cérvico-vaginal e conteúdo uterino da fêmea ou dos produtos de aborto (placenta, pulmão, fígado e abomaso do feto) (OIE, 2009). Esta técnica pressupõe uma colheita do material em condições de assépsia, o seu envio rápido (no próprio dia) ao laboratório em meio apropriado e a temperatura de refrigeração (5-8ºC). A Cfv é de crescimento fastidioso e é rapidamente ultrapassada pelo crescimento de contaminantes. Estes condicionantes podem determinar uma elevada taxa de falsos-negativos, assim como uma demora no resultado laboratorial (2-3 semanas).

Outras técnicas foram descritas (van Bergen e col., 2005), incluindo a pesquisa de anticorpos (IgA) no muco cérvico vaginal, anticorpos (IgG) circulantes e imunofluorescência directa. Estas técnicas são inespecíficas e não permitem diferenciar as subespécies de *Campylobacter fetus*. Mais recentemente foram descritas várias técnicas de *polymerase chain reaction* (PCR) que detectam sequências de ADN específicas das duas subespécies. Na FMV-UL foi desenvolvida uma técnica de PCR em tempo real (quantitativa; qRT-PCR) que tem sido utilizada para o diagnóstico de Cfv em amostras de raspagem e lavagem prepucial de touros. A colheita da amostra é realizada com higiene básica, em solução salina ou PBS (Phosphate Buffer Saline) e o resultado pode ficar disponível em 2 dias (Duarte e col., 2014).

Situação em Portugal

Até 2014 não foi comunicada a existência de focos de doença ou o diagnóstico da bactéria causal no país. Nesse ano, na sequência de um projecto desenvolvido pela FMV-UL foi diagnosticada a CGB e a Cfv em explorações aleitantes do Alentejo (Duarte e col., 2014). Este trabalho tem sido alargado a todo o Alentejo e Ribatejo. No presente momento, foram testados 252 touros de 54 explorações aleitantes, tendo-se revelado positivos 110 (43,7%) deles, pertencentes a 32 explorações (59,3%). Estes resultados não podem ainda espelhar a prevalência da Cfv pois o diagnóstico (da CGB e da Cfv) ocorreu em parte em explorações que o solicitaram devido à detecção de baixa fertilidade.

Apesar do exposto, a CGB parece representar um factor com impacto relevante na fertilidade das explorações aleitantes em sistema extensivo do Alentejo e Ribatejo. O grau de impacto não é fácil de aferir devido à simultânea detecção de outras doenças (IBR, BVD) e de práticas de maneio reprodutivo incorrectas. A competitividade do sector aleitante, a rentabilidade das empresas, a certificação de explorações e animais e o actual sistema de subsídio à produção indexado à fertilidade, tornam necessária o controlo da CGB e da disseminação da Cfv.

Recentemente (2015), no âmbito de um programa de controlo voluntário das doenças infecciosas que afectam a reprodução bovina (VITINDEMNE), associado aos ADS de Estremoz e Monforte, estão a ser implementadas medidas

integradas que visam o controlo e eventual erradicação da CGB e do Cfv nas explorações aderentes, potenciando a comercialização e exportação de animais, incluindo reprodutores.

Controlo na exploração aleitante

É baseado na biosegurança, diagnóstico e subsequentes medidas em caso de positividade. O controlo da entrada de animais na exploração é uma premissa fundamental. Os touros adquiridos devem ser previamente testados, assim como os residentes o devem ser anualmente antes da época reprodutiva. Idealmente todos os touros positivos a Cfv devem ser refugados. A ausência de touros positivos é um bom indicador de ausência da Cfv na vacada. No caso das fêmeas, é muito oneroso fazer a sua testagem completa. As novilhas virgens (não colocadas à reprodução), as vacas paridas recentes (antes de entrar à reprodução) e as vacas gestantes no segundo e terceiro trimestres de gestação podem ser consideradas "limpas". As vacas no primeiro trimestre de gestação estão num estatuto intermédio, enquanto as vacas alfeiras devem ser consideradas potencialmente infectadas.

Em presença de um efectivo infectado com evidência clínica de CGB deve ser realizado o diagnóstico de gestação para constituir subgrupos com maneio distinto. As vacas "limpas" podem ser colocadas à reprodução com touros indemnes ou submetidas a IA. As vacas com gestações recentes devem ser acompanhadas e, se parirem, transitar para o subgrupo das "limpas". As vacas vazias podem idealmente ser refugadas ou submetidas a IA, para não infectarem touros indemnes ou serem infectadas por touros positivos (caso estivessem não infectadas).

No caso de uma vacada endemicamente infectada, já com recuperação parcial da fertilidade, em que a totalidade ou grande proporção dos touros estão infectados, uma alternativa é recorrer à vacinação. Infelizmente não existe vacina disponível em Portugal ou União Europeia, embora existam várias vacinas (bacterinas) no mercado Americano. A produção de uma autovacina pode neste caso ser ponderada, sendo vacinados todos os animais, fêmeas e touros. O tratamento dos touros mais velhos é pouco (ou nada) eficiente. Para os touros mais jovens, com menor carga bacteriana nas pregas prepuciais, pode ser tentado o tratamento sistémico e local, sendo considerados curados os animais com dois testes negativos subsequentes.

Em qualquer dos casos, a fertilidade da exploração deve ser continuamente monitorizada através da análise dos registos reprodutivos e da vigilância das ocorrências clínicas (repetições de cobrições, abortos).

Controlo no *feedlot*

Um dos achados possíveis é a detecção de touros positivos jovens, antes da entrada à reprodução. Este é um dos aspectos da patogénese da doença ainda nebulosos. O contágio pode provir da cohabitação com touros mais velhos, por sodomia, ou eventualmente por infecção ascendente a partir do local de estabulação ou durante a amamentação, a partir de produtos de aborto ou corrimentos cérvico-vaginais (situações que carecem de investigação). De facto, a infecção prepucial e peniana é possível antes da puberdade, entre vitelos contemporâneos de exploração. Isto parece indiciar que nestas explorações a CGB é endémica.

Em feedlots do Alentejo e Ribatejo detectámos até ao momento 12% de vitelos (6-8 meses de idade) positivos a Cfv. O controlo dentro do feedlot é difícil, mas deve basear-se sobretudo na biosegurança, adquirindo apenas animais de explorações certificadas. Uma vez detectada a Cfv no *feedlot*, pode realizar-se o vazio sanitário e implementar medidas de higiene entre lotes. O impacto destas medidas sobre a permanência da bactéria é dificíl de antever. A presença de Cfv em vitelos pode constituir um entrave à sua comercialização, e em particular à sua exportação.

Conclusão

A CGB foi diagnosticada em Portugal em 2014 pela primeira vez. A doença pode afectar de maneira diferenciada a fertilidade da exploração aleitante, consoante a forma epidémica ou endémica em que se encontra. Em qualquer dos casos afecta a fertilidade, intervalo entre partos e distribuição de partos da vacada. A contribuição para o grau de ineficiência reprodutiva e de prejuízo económico nem sempre é de fácil aferição por poderem estar presentes outros agentes de doença (IBR, BVD) e/ou maneio reprodutivo inadequado. A taxa de detecção em explorações e touros até agora verificada, sugere que é um problema potencialmente relevante da exploração bovina aleitante e do feedlot no Alentejo e Ribatejo.

O controlo da doença deve ser realizado de forma integrada, considerando a biosegurança, a monitorização da eficiência reprodutiva através da análise de registos e da vigilância de casos clínicos, o diagnóstico etiológico e o estabelecimento de práticas como o refugo de animais positivos, o diagnóstico de gestação ou a vacinação.

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Diarreias neonatais em explorações leiteiras

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Resumo

As diarreias neonatais são das principais causas de morte no primeiro mês de vida dos vitelos. A sua etiologia é multifatorial e a sua incidência numa dada exploração depende essencialmente da atenção dada a alguns pontos críticos no maneio destes animais. Um bom programa de maneio de vitelos deverá ser baseado em medidas de prevenção. Esta baseia-se essencialmente em dois fatores: aumentar a resistência dos animais (essencialmente através de um bom maneio ótimo de colostro) e diminuir a pressão de infeção a partir do ambiente. Evitar que surjam diarreias neonatais deverá ser sempre objetivo, no entanto, caso surjam, o sucesso está dependente de uma deteção precoce e da existência de protocolos de tratamento adequados na exploração.

Introdução

As diarreias neonatais são muito frequentes e constituem uma queixa comum e uma causa de perdas económicas elevadas para produtores de leite em todo o mundo. As diarreias neonatais são responsáveis por cerca de metade das mortes antes do desmame[1–3]. Podem ter etiologia infeciosa e não infeciosa. Vários agentes entéricos estão envolvidos no desenvolvimento desta patologia e, apesar de um único agente primário poder estar na origem da doença, a co-infecção em vitelos com diarreia é comum[4]. A etiologia multifatorial das diarreias em vitelos dificulta o seu controlo. Por outro lado, tem havido grandes melhorias no sentido de diminuir taxas de morbilidade e de mortalidade, nomeadamente no que diz respeito ás diarreias em vitelos antes do desmame, altura onde estas são mais elevadas[2].

A saúde dos vitelos e a incidência maior ou menor de doença entérica depende essencialmente de dois fatores: (1) resistência do animal à doença; (2) pressão de infeção no ambiente em que este se encontra. No sentido de diminuir morbilidades e de mortalidades é essencial que se foque a atenção nestes dois fatores.

Este manuscrito tem como objetivo abordar brevemente os principais agentes etiológicos das diarreias neonatais e rever as práticas de maneio mais importantes com vista a diminuir a incidência de diarreias no período pré-desmame, altura crítica na vida destes animais. Serão revistas também, de forma sucinta, as possíveis abordagens terapêuticas aos animais doentes.

Agentes Etiológicos das diarreias neonatais

Os agentes patogénicos mais comuns das diarreias neonatais são a *Escherichia coli* K99/F5, rotavirus e coronavírus e o *Cryptosporidium spp* (≥85% *C. parvum*), sendo o rotavírus e o *C. parvum* os mais frequentemente isolados a partir

de fezes de vitelos[5]. *E. coli* K99/F5 é tipicamente responsável pelas diarreias em vitelos entre o 1º e o 4º dia de vida e os outro três causam, geralmente, diarreias a animais entre 1 e 3 semanas de idade. Os agentes mais comuns associados a surtos de diarreias neonatais são também encontrados em fezes de animais saudáveis, o que significa que a presença do patógeno não é necessariamente causal[6]. Outros agentes menos comuns de diarreia neonatal são a *Salmonella spp*, outras E. coli (como a enteropatogénica, a enterohemorrágica e a produtora de toxina Shiga), *Clostridium perfringens* e torovirus[4,6].

Pontos chave no maneio dos vitelos

Os valores de mortalidade e morbilidade variam grandemente de exploração para exploração. Não existem, à data, muitos dados nacionais, no entanto em Espanha, por exemplo, os valores reportados andam entre os 2,2% e os 12%[7]. Esta variação depende das medidas de maneio, que diferem de exploração para exploração, e é elucidativa quanto às oportunidades de melhoria ainda existentes em muitas vacarias. No sentido de estabelecer um bom programa de maneio de vitelos o foco deverá basear-se em maximizar a resistência/imunidade dos animais e minimizar a exposição dos mesmos a agentes infeciosos.

Maximizar a resistência/imunidade do animal

1. Encolostramento

Devido á estrutura da placenta bovina, o vitelo nasce sem imunoglobulinas e depende inteiramente da sua absorção a partir do colostro para obter imunidade e proteção adequada contra organismos patogénicos até que o seu sistema imune esteja desenvolvido, o que ocorre entre as 3 e 4 semanas de vida[8]. A transferência de imunidade passiva adequada é um dos fatores mais importantes para a saúde futura dos vitelos. Considera-se que a transferência de imunidade passiva foi adequada quando a concentração de IgG no soro do vitelo 24-48h após o nascimento é superior a 10 mg/ml[9]. O sucesso da transferência da imunidade passiva depende da qualidade do colostro, do volume de colostro ingerido e da altura após o nascimento em que é ingerido.

A qualidade do colostro é considerada adequada se este tiver uma concentração de IgGs superior a 50g/L. Esta poderá ser avaliada com o recurso a um densímetro ou a um refratómetro com escala de brix[10]. A qualidade do colostro depende de fatores independentes do maneio, como a raça11,12 e o número de lactações[11]; e de fatores dependentes do maneio entre eles a nutrição e duração do período seco e programa vacinal durante a secagem.

Taxas elevadas de falha de transferência de imunidade passiva estão associadas á prática de deixar o vitelo a mamar diretamente da mãe[13,14]. No sentido de atingir valores de 10mgl/ml de IgG no soro, um vitelo deverá consumir no mínimo 100 g de IgG. Atualmente recomenda-se a administração de 8,5%-10% do peso vivo de colostro (4 L)[15]. A rapidez após nascimento com que o colostro é administrado é também fundamental. Assim que o vitelo nasce o epitélio intestinal inicia um processo de maturação que leva á total impermeabilização ("fecho") do mesmo para as imunoglobulinas. A capacidade de absorção é máxima às 2h e o "fecho" completo dá-se 24 h após o nascimento[9]. Assim a primeira administração de colostro deverá ser feita, com recurso a sonda esofágica se necessário, de modo a administrar 4 L nas primeiras 6 horas de vida[8].

A pasteurização e criação de um "banco" de colostro está cada vez a tornar-se prática mais comum em explorações de média e grande dimensão. A pasteurização de colostro com qualidade previamente testada (600C durante 60 min) não provoca a diminuição do nível de IgG, ao mesmo tempo que diminui significativamente a contaminação bacteriana do mesmo facilitando a absorção de imunoglobulinas e diminuindo o risco de tratamento para diarreias neonatais e outras patologias[16].

2. Estratégia vacinal

Existem algumas vacinas no mercado de administração durante a secagem que protegem contra rotavírus, coronavírus e *E. coli*. Estas vacinas podem ajudar a aumentar a imunidade dos vitelos recém-nascidos. No entanto é importante salientar que a sua eficácia depende do encolostramento adequado do vitelo neonato.

3. Minimização do stress

O stress inibe o sistema imune, tornando os vitelos mais suscetíveis a doenças. Fatores causadores de stress são, por exemplo: stress térmico, acesso inadequado a comida e água, superpovoamento, má ventilação, dor, alterações súbitas de alimentação, mudanças de grupo, transporte e desmame. Deste modo é importante diminuir o efeito cumulativo dos vários agentes de stress, nomeadamente na altura do desmame.

4. Nutrição

A interação entre o estado nutricional e estado imunitário está bem estabelecida. O maneio nutricional dos vitelos tem um enorme impacto na sua saúde e resistência a doenças. Uma nutrição inadequada, nomeadamente uma restrição proteica/calórica pode causar um estado de imunodepressão e predispor os vitelos a doenças. É essencial que os animais tenham sempre acesso a comida de qualidade e água fresca. A qualidade e quantidade de leite consumido é fundamental para que as necessidades nutricionais dos vitelos sejam respeitadas para que estes possam fazer face ás suas necessidades metabólicas de manutenção e para que possam ter um ganho de peso médio diário adequado. Tradicionalmente os vitelos eram alimentados antes do desmame com leite de substituição a 10% do peso vivo (4 L/dia em vitelos Holstein). Vitelos a mamar diretamente da mãe atingem 20% do peso vivo de ingestões diárias. Esta prática de restrição da quantidade de leite de substituição fornecido permite apenas suprir as necessidades de manutenção e tem como objetivo incentivar o vitelo a ingerir concentrado para que o seu desmame possa ser feito o mais precocemente possível reduzindo o potencial para o aparecimento de diarreias e reduzindo os custos de alimentação e de maneio. No entanto a estratégia adotada no passado para diminuir a ingestão de uma alimentação liquida e aumentar a ingestão de uma ração sólida, promovendo o desenvolvimento ruminal, não conseguiu reduzir nenhuma dessas variáveis[17]. Vitelos a que é permitido um aumento da ingestão de leite para 15% do peso corporal, e atrasado o desmame até às 10-12 semanas, apresentam maiores ganhos médios diários. Vitelos desmamados mais tarde apresentam menos comportamentos de fome, maior tempo a ruminar e maior tempo deitados, evidenciando maior bem-estar animal[18,19].

Minimizar a exposição do animal a agentes infeciosos

Desde o nascimento, com a chegada do vitelo à maternidade e o contacto do mesmo com o ambiente e possivelmente com outros animais, que este é exposto a vários agentes infeciosos. É fundamental que se diminua ao máximo a pressão de infeção, no sentido de reduzir a probabilidade de doença. A maternidade é onde tudo começa e deverá estar limpa e seca com material de cama suficiente. A maternidade deverá ser apenas isso, estando livre de outros animais especialmente se doentes. No sentido de evitar a exposição a gentes infeciosos existentes no ambiente onde a mãe se encontra, estes deverão ser separados o mais rapidamente possível. Os vitelos recém-nascidos não deverão ser misturados com outros animais mais velhos. Preferencialmente deverão ser colocados numa cama individual (ex. iglo) previamente limpa e desinfetada. Os utensílios utilizados para administração de leite, concentrado e água deverão ser adequadamente higienizados.

Abordagem terapêutica do vitelo com diarreia

Preferencialmente boas práticas de maneio deverão prevenir as diarreias neonatais. No entanto se surgirem é importante que sejam diagnosticadas precocemente no sentido de tratar atempadamente a evitando sequelas para o futuro do animal ou mesmo a morte.

As diarreias neonatais provocam desidratação, acidose metabólica, desequilíbrio eletrolítico e hipoglicémia. Assim, uma parte fundamental do tratamento de animais acometidos, passa por fluidoterapia adequada. A reidratação oral deverá ser iniciada assim que inicia um episódio de diarreia e continuar enquanto esta persistir. Um bom reidratante oral deverá conter[20]:

- 90-130 mmol/l de sódio;
- 40-80 mEq/l de cloro;
- 10-30 mmol/l de potássio;
- Glucose na proporção de 1:1 a 3:1 de sódio para facilitar a sua absorção;
- Capacidade alcalinizante de 60-80 mmol/l.

Animais com desidratação superior a 8%, deverão receber fluidoterapia intravenosa (IV). Escolhas potenciais são solução isotónica de Cloreto de sódio (0,9%), bicarbonato de sódio a 8,4%, lactato de ringer e dextrose 5%. Os fluidos administrados por via IV deverão ser sempre previamente aquecidos.

A antibioterapia rotineira não é recomendada devido ao aumento de resistências a antibióticos verificadas nos últimos anos. No entanto, vitelos com sinais sistémicos de doença (depressão, febre e anorexia), provavelmente sofrem de septicémia por *E. coli* e deverão ser tratados com AB como a amoxicilina, ampicilina, cefalosporinas e fluorquinolonas. ABs usados em medicina humana em último recurso, como as fluorquinolonas e cefalosporinas de 3ª e 4ª geração só deverão ser utilizados se outros falharem. Para o tratamento do *Cryptosporidium spp*. a Paromomicina é relativamente eficaz na diminuição de sintomas e excreção de oocistos[21], no entanto e seu custo é bastante elevado e no futuro os aminoglicosídeos deverão ser bastante limitados para uso em animais de produção. A halofuginona poderá ser uma alternativa para a profilaxia do *Cryptospuridium spp*.

As diarreias neonatais são geralmente acompanhadas de dor e desconforto abdominal, pelo que o uso de anti-inflamatórios não esteroides (AINES) é aconselhado.

Conclusão

As diarreias neonatais têm etiologia multifatorial e uma natureza complexa. As taxas de mortalidade e morbilidade variam bastante de exploração para exploração. Tal deve-se essencialmente a diferenças no maneio dos vitelos e à atenção prestada a "pormenores" por parte do produtor. O envolvimento permanente do produtor, trabalhadores, veterinário e nutricionista da exploração é essencial no sucesso de um bom programa de maneio dos vitelos. Protocolos de maneio do vitelo recém-nascido que incluem um protocolo de colostro e protocolos de tratamento para as principais patologias neonatais são essenciais ao bom funcionamento e sucesso da recria. Estes protocolos deverão ser simples de ler, conhecidos e compreendidos por todos e revistos frequentemente.

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Toxémia de gestação em pequenos ruminantes

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Resumo

A toxemia de toxemia é um problema muito comum em pequenos ruminantes no final da gestação. Nesta apresentação serão discutidos a fisiopatologia, sinais clínicos, diagnóstico, prognóstico e medidas terapêuticas e preventivas desta doença. São realçados as principais diferenças entre ovinos e caprinos relativas a esta afecção.

Introdução, Resultados, Discussão

A toxemia de toxemia é um problema muito comum em pequenos ruminantes no final da gestação, com especial incidência em animais com gestações múltiplas, muito gordos ou muito magros. Anorexia, edema dos membros (cabras) prostração e sinais nervosos como "star gazing" e cegueira (ovelhas) são os sinais clínicos mais comuns. O diagnóstico baseia-se nos sinais clínicos e nalgumas alterações na urina (cetonúria e acidúria) e no sangue (BHBA aumentado e pH, K+ e pCO2 diminuidos). A glicémia geralmente está diminuída mas em caso da morte intra-uterina dos fetos pode estar aumentada. Alguns dos parametros acima referidos (sinais clínicos e valores sanguíneos) podem ser usados como indicadores de prognóstico. A condição corporal como potencial factor de risco não é tão eficaz em cabras como em ovelhas. As diversas opções terapêuticas usadas apenas são eficazes na fase inicial da afecção. É possível que algumas cabras com toxemia de gestação possam ser intolerantes à glucose que pode estar associada a fígado gordo. Em virtude da alta taxa de mortalidade observada em animais com toxemia de gestação, a implementação de medidas preventivas é essencial. São realçados as principais diferenças entre ovinos e caprinos relativas a esta afecção.

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Maria José Pinto

Oportunidades e condicionalismos para a exportação de pequenos ruminantes vivos

Os efeitos da globalização e da dificuldade em escoar os animais no mercado nacional e da U.E, a preços concorrenciais, levaram a que a produção nacional dirigisse a sua atenção para mercados inexplorados ou até mesmo em certa medida desconhecidos para Portugal.

A abertura de novos mercados para exportação de animais vivos e produtos animais, está sujeita à demonstração, por parte do país candidato à exportação, do cumprimento das exigências para importação impostas pelo país importador. A demonstração da segurança do sistema de produção, constitui um exercício de natureza técnica, incluindo a descrição detalhada dos controlos efetuados, do estatuto sanitário do território nacional relativo às doenças animais e das medidas de biossegurança implementadas.

Compete à Direção-Geral de Alimentação e Veterinária (DGAV), organismo responsável pelos controlos oficiais tendo em vista a garantia da defesa da saúde e bem-estar animal e da segurança alimentar, a condução das negociações técnicas com as autoridades congéneres dos países terceiros tendo em vista a criação das condições de exportação, e consequente abertura do mercado. No entanto, o processo de abertura dos mercados, nas suas múltiplas complexidades, não se esgota nas relações a nível exclusivamente técnico, sendo muitas vezes essencial um acompanhamento ao nível diplomático e político, por forma a forçar o desenvolvimento das negociações e evitar a falta de progresso dos dossiers submetidos aos países de destino.

A fim de alcançar o sucesso da abertura de mercado, entre outros países que concorrem ao acesso a estes mercados globalmente apetecíveis, este esforço deverá ser desenvolvido pela DGAV, com o apoio das Embaixadas de Portugal nos países de destino, mas também e essencialmente pelos produtores/operadores do setor que se candidatam à exportação.

Os resultados recentemente obtidos são indicador de que os animais portugueses têm procura e são bem aceites nos novos mercados internacionais.

Regra geral, a habilitação para exportação é um processo constituído por vários passos tratando-se de processos morosos e dispendiosos e que por vezes acabam por acarretar prejuízo económico mutuo para o importador e exportador que acabam por perder, à data, a oportunidade de negócio.

Para além da complexidade do processo de habilitação, as condições exigidas para efetivação das mesmas, são frequentemente complexas e o seu cumprimento vai além das condições exigidas para produção e colocação no mercado nacional e da UE. Realce-se que estas exigências são imposição do país terceiro de destino, e a possibilidade de exportação está dependente do seu cumprimento.

Sendo certo de que o Princípio Fundamental que norteia todos estes processos é o da transparência, em que o país potencial importador deverá indicar de forma clara as sua exigências e o país potencial exportador não poderá omitir informação, muitas das vezes verifica-se que o grau de exigência que é feito acaba por funcionar como um mecanismo protecionista e uma verdadeira barreira à exportação e os processos de negociação arrastam-se por tempo quase indeterminado.

Nuno Carolino

Seleção de reprodutores em pequenos ruminantes para produção de carne

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A seleção é sem dúvida uma ferramenta poderosíssima e imprescindível para que o Homem possa utilizar os animais para diferentes fins. Este processo consiste na reprodução diferencial dos indivíduos da população, ou seja, na escolha dos animais que irão produzir descendentes e, desta forma, originar novos indivíduos que possam expressar características desejáveis para determinadas funções.

Pretende-se através da seleção (escolha de futuros reprodutores machos ou fêmeas) aumentar ou diminuir a frequência de genes relacionados com a expressão de algumas características desejáveis ou indesejáveis, respetivamente. Desta forma, tanto em animais de produção, como em animais de companhia, anualmente os criadores deparam-se com a tarefa de selecionar os animais (machos e/ou fêmeas) que irão utilizar como reprodutores. Ou seja, a partir dos animais disponíveis nos seus efetivos ou mediante a aquisição/utilização de animais de outras explorações, sémen ou outro material genético, os criadores tentam escolher para reprodutores, indivíduos que possam transmitir determinadas características aos descendentes. Interessa assim aos criadores a possibilidade de identificar animais geneticamente superiores para estas características.

No caso particular de um criador estar integrado num programa de melhoramento de uma determinada raça, deverá selecionar os animais de acordo com os objetivos de melhoramento já definidos (características que se pretendem melhorar) para essa população, o que exigirá um enorme rigor nesta atividade. No caso dos criadores cujo objetivos é produzir animais para determinado fim, seja carne, leite ou outro produto ou função, deverá selecionar para reprodutores os animais que possam transmitir aos seus descendentes características que lhes permitam atingir esses fins.

Os sistemas de produção de ovinos de carne apresentam distintas características em função do grau de intensificação. Em Portugal predominam os sistemas extensivos com tipos de pastoreio muito variáveis, consoante as características das explorações, mas utiliza-se bastante o sistema de pastoreio rotacional, com alguns ajustamentos da carga animal. Sendo explorados maioritariamente região sul do país (cerca de 50% no Alentejo), utilizam zonas marginais, através do pastoreio de zonas incultas, pastagens semeadas, resíduos de colheitas e áreas florestais.

Poderemos considerar três parâmetros fundamentais para o sucesso das explorações de ovinos de carne:

- Produtividade numérica – nº de animais comercializados por ovelha e por ano
- Produtividade ponderal – kg's comercializados por ovelha e por ano
- Qualidade do produto – conformação, gordura, qualidade da carne

Enquanto a produtividade numérica é essencialmente determinada por características relacionadas com a fêmea reprodutora, entre outras, características maternais, prolificidade e longevidade, a produtividade ponderal depende adicionalmente de fatores relacionados com os filhos, designadamente, potencial de crescimento, eficiência alimentar, conformação, rendimento da carcaça, etc.

A qualidade do produto é, sem dúvida, o parâmetro mais subjetivo e atualmente é sobretudo determinado por questões diversas de mercado, como sejam a idade dos animais, o peso ao abate, o rendimento e conformação da carcaça que têm impacto no valor de comercialização dos animais/carcaça, mas é questionável a sua relação com os atributos qualitativos da carne, como por exemplo, a composição em ácidos gordos ou parâmetros físicos (pH, cor, perda de peso pro cozimento, capacidade de retenção de água, tenrura, etc.)

De um modo geral, as características produtivas dependem de muitos genes e a sua expressão, o fenótipo, é normalmente influenciado por causas ambientais (p.e., alimentação) o que dificulta todo o processo de seleção.

O valor fenotípico de um animal para determinado registo (e.g., o peso ao desmame ou a prolificidade) pode ser um indicador do seu valor genético, mas também reflete as condições ambientais (criador, ano, época de nascimento, idade, alimentação, etc.) a que o mesmo foi sujeito. Desta forma, a informação fenotípica de um indivíduo, por si só, poderá ser um indicador pouco preciso do seu mérito genético.

Como um reprodutor transmite à descendência apenas parte dos seus genes e não as condições ambientais a que foi sujeito, interessa ao criador conhecer o valor genético dos animais, ou seja, qual o valor de um animal num programa de seleção ou o que o animal poderá transmitir à descendência.

O valor genético de um animal representa o seu valor como reprodutor ou a soma dos efeitos de cada um dos alelos que afetam a característica, e pode ser interpretado como a sua superioridade ou inferioridade genética para o caracter em causa, cuja metade será transmitida à descendência.

O sucesso da seleção e, consequentemente dos programas de melhoramento genético, dependem de vários parâmetros (precisão e intensidade de seleção, variabilidade genética, intervalo de gerações) que, por sua vez, dependem da quantidade e qualidade da informação disponível individual e familiar, da forma como esta é utilizada ou combinada, sendo essencial o método de seleção dos animais.

Existem diversos métodos de seleção dos animais, tais como seleção individual, seleção Familiar - seleção pela ascendência e teste de descendência, seleção combinada ou índices de seleção, BLUP – Modelo Animal, seleção assistida por marcadores genéticos, seleção genómica, etc. Na seleção individual os animais são selecionados apenas com base nas suas próprias performances, sendo, por isso mesmo, a metodologia mais simples e apresenta grandes limitações. A seleção assistida por marcadores tem apresentado avanços substanciais nos últimos anos, particularmente em algumas características, tais como a resistência a doenças (p.e., scrapie), quantidade e qualidade de músculo (p.e., GDF-8 e CAST) e prolificidade (p.e., inverdale e booroola), mas apesar do seu interesse, a sua utilização a curto prazo dificilmente se poderá generalizar a todos os tipos de características com interesse para os criadores. A utilização da seleção genómica em ovinos de carne até à data ainda é muito escassa, devido, entre outros fatores, à necessidade de grande quantidade informação produtiva e genealógica acumulada ao longo de gerações, pela reduzida utilização da inseminação artificial e pelos elevados custos dos Chips de SNP's (polimorfismos de nucleotídeos simples) embora com a capacidade de avaliar milhares nucleotídeos espalhados por todo o genoma.

Alguns países, em que a produção ovina tem grande importância, como é caso da Austrália e da Nova Zelândia, já iniciaram programas de seleção para características de produção de carne e lã, que incluem informação genómica baseada em Chips de alta densidade (LAMBPLAN E MERINOSELECT). Apesar das vantagens e boas perspetivas desta nova técnica de seleção a longo prazo, é necessário demonstrar a sua aplicabilidade e rentabilidade nos ovinos, tanto de carne como de leite.

A seleção genómica não impede as atividades necessárias aos atuais programas de seleção considerados nos dias de hoje "clássicos", sendo fundamental manter o controlo dos registos produtivos e genealógicos.

Assim, atualmente, a nível internacional e em diversas espécies pecuárias (ovinos, bovinos, suínos, aves, caprinos, equinos, etc.) o recurso ao BLUP - Modelo Animal para a avaliação genética está generalizado. Quando comparado com a seleção fenotípica, apresenta diversas vantagens que, em termos práticos, significam que o valor genético de um indivíduo predito pela metodologia BLUP - Modelo Animal considera:

- O mérito genético de todos os seus parentes mais ou menos distantes pela inclusão da matriz de parentescos – relação de parentesco entre todos os animais.
- O valor genético dos participantes nos diferentes acasalamentos. Isto é, um macho não será prejudicado por ser acasalado com fêmeas de mérito inferior ou vice-versa.
- Todos os registos produtivos disponíveis (registos repetidos no mesmo indivíduo ou nos seus parentes, etc.).
- Os efeitos ambientais a que um registo produtivo é sujeito (e.g., diferentes ambientes/explorações, épocas de nascimento, sexo, idade, etc.).

Através da avaliação genética com o BLUP - Modelo Animal, pretende-se estimar com a maior precisão possível o valor genético de cada animal para as diversas características de interesse para cada raça ou população, com base na informação produtiva disponível (própria e de parentes) e levando em consideração efeitos ambientais que possam influenciar a expressão do potencial genético (ano e mês de nascimento, sexo, idade do animal, etc.).

Nos últimos anos, em diversas espécies pecuárias, a seleção dos futuros reprodutores tem sido um elemento chave para o sucesso das explorações e para o progresso genético registado.

Independentemente das características das explorações de ovinos de carne e da idade mais ou menos precoce em que se irão selecionar os futuros reprodutores, para além do estado higio-sanitário dos animais e de aspetos morfo-funcionais, é essencial que os animais tenham mérito genético superior (valor genético) para as características economicamente mais importantes para essa exploração. No caso, de se tratar de um criador com animais de um determinada raça é ainda fundamental a sua adequação ao respetivo a padrão racial.

José Alfaro Cardoso

Fisiologia da Reprodução

São vários os factores relacionados com a genética, nutrição, sanidade ou biossegurança e de meio ambiente ou bem-estar animal, que influenciam as performances reprodutivas de qualquer espécie animal.

Existem fronteiras muito ténues entre estes factores, estando eles sempre interrelacionados, de tal forma que se torna por vezes muito difícil avaliar qual deles terá a maior quota de responsabilidade no desempenho reprodutivo.

A inter-relação entre estes diversos factores, está no entanto, sempre condicionada à forma como interagimos com eles. A esta interacção dá-se vulgarmente o nome de maneio.

O factor humano é portanto, através de um bom ou mau maneio, o grande responsável pelo bom ou mau desempenho reprodutivo.

Um mau maneio pode conduzir a patologias clínicas evidentes com o seu agente etiológico bem definido, ou simplesmente originar o que eu apelido de "Pseudopatologia Reprodutiva", onde há total ausência de agente infeccioso ou patológico, mas nem por isso coexistindo menos transtornos reprodutivos.

É esta "Pseudopatologia", que em larga percentagem afecta as explorações suinícolas.

Para entendermos melhor as falhas reprodutivas na espécie porcina em particular, devemos incidir a nossa maior atenção no período compreendido entre o início do estro, com as manifestações de cio a ele associados e os 24 dias após a IA/CN.

Este é o período crítico do ciclo reprodutivo da porca, com reflexos na sua rodutividade e durante o qual a influência da intervenção humana mais se evidencia.

Desde uma correcta detecção de cio e um bom maneio do varrasco/semén, até à completa fusão endométrio-embrião, variadíssimos aspectos anatomo-fisiológicos interferem e se conjugam para que haja uma eficaz nidação e manutenção de embriões de boa qualidade e em quantidade suficiente, não só para a mautenção da gestação, mas principalmente para a rentabilidade produtiva da porca.

Efeito do plasma seminal na actividade uterina

Os estrogénios presentes no plasma seminal, para além de terem um efeito regulador do início da ovulação em relação à IA/CN actuando sobre LH e a PGF2α folicular, estimulam o endométrio induzindo a libertação imediata de PGF2α, com consequente aumento da contractibilidade do miométrio durante algumas horas. Este aumento da contractibilidade uterina tem um efeito de enorme importância, reduzindo o refluxo seminal, mas principalmente conduzindo os espermatozoides até aos oviductos, e auxiliando ainda o transporte no seu interior.

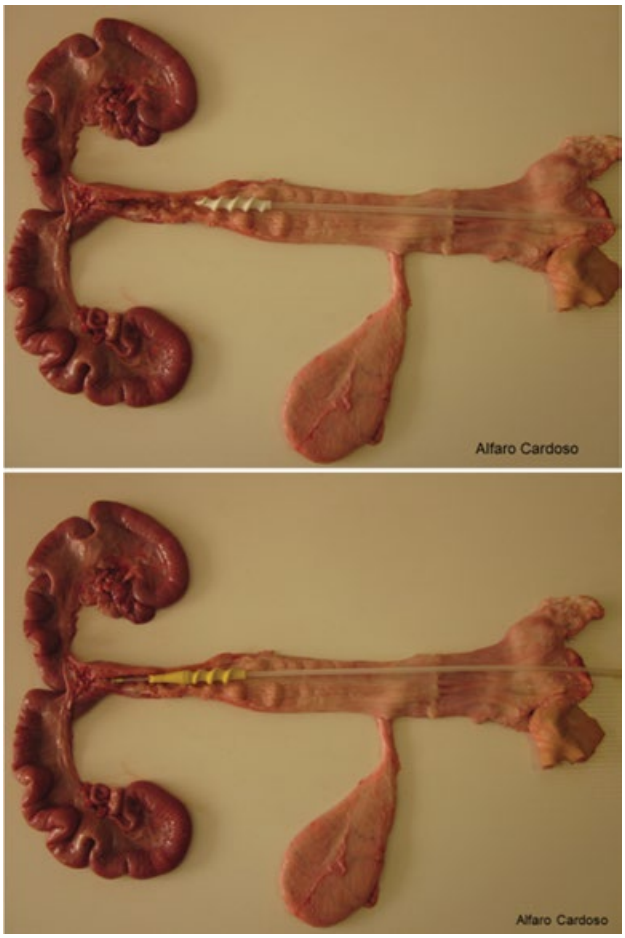
O plasma seminal tem ainda a capacidade de reduzir a actividade fagocitária espermatozoide-neutrófilo e de inibir a ligação dos espermatozoides às células epitelias uterinas (CEU)

Imunologia reprodutiva do tracto genital feminino

Assim que o sémen contacta com o tracto genital feminino, imediatamente a porca responde com uma reacção imunológica com resposta inflamatória consequente, caracterizada por elevada e rápida fagocitose espermática.

Segundo alguns autores esta fagocitose é mais elevada na zona do corpo uterino. Teóricamente, o semén depositado mais próximo da bifurcação dos cornos uterinos não estará tão sujeito a este ataque.

Esta teoria é defendida pelos adeptos da IA pós cervical.



Parece lógica esta resposta imunitária, dado que o sistema imunitário da mucosa uterina tem como função a defesa de agentes agressores do tracto genital.

No entanto e neste caso particular, a resposta imunitária é selectiva.

A porca rapidamente terá que distinguir agentes agressores tais como bactérias comensais, bactérias ou vírus transmitidos sexualmente ou veículados durante a IA, dos constituintes do sémen e mais tarde dos constituintes fetais.

Esta reacção imunológica tem acção directa na selecção espermática, na indução e manutenção da tolerância por parte da mãe em relação a antigéneos paternos, na restruturação do endométrio, determinante para que haja uma boa implantação e placentação, e no suporte imunológico aos tecidos fetais durante a gestação.

As moléculas activas no plasma seminal e associadas aos espermatozoides interagem com as CEU após a IA/CN induzindo a síntese de citoquinas próinflamatórias. Estas citoquinas provocam o recrutamento de células inflamatórias no endométrio, tais como macrófagos, células dentríticas e granulócitos. Os macrófagos e as células dentríticas participam na remodelação do tecido endometrial e na activação da tolerância imunitária maternal à gestação. Os granulócitos, nomeadamente os neutrófilos, atravessam o epitélio endometrial para o lúmen e asseguram essencialmente a manutenção da esterilidade uterina. As citoquinas inflamatórias activadas pelo plasma seminal são também secretadas para o fluido luminal, onde exercem acções tróficas no embrião durante a préimplantação. Para além destes aspectos, o plasma seminal exerce uma função moduladora importante na interacção espermatozoide-neutrófilo ao reduzir as actividades fagocitárias dos neutrófilos. Além disso, inibe a capacidade de ligação dos espermatozoides às CEU. A relevância destas descobertas deve ser considerada na IA, quando são usadas concentrações baixas de espermatozoides e volumes reduzidos de plasma seminal.

A reacção imunológica Influencia também o processo ovulatório. Waberski (1995,1997) inseminou marrãs com um dos cornos uterinos laqueado e verificou que o sémen acelerou a ovulação apenas no ovário correspondente ao corno não laqueado.

Aspectos anatomo-fisiológicos

O oviducto tem cerca de 20 cm e é constituído por três porções a partir do ovário: infundibulo, ampola e istmo.

Os espermatozoides só se encontram a salvo do brutal ataque fagocitário, daí a importância da contractibilidade uterina, quando entram no istmo, também chamado reservatório espermático, podendo sobreviver aqui cerca de 30 horas.

O istmo actua como um filtro que selecciona e permite a passagem dos espermatozoides mais aptos, unindo-os a moléculas de carbohidratos e glicoproteínas, aumentando a capacidade de penetração dos espermatozoides, inibindo a polispermia e favorecendo o desenvolvimento embrionário.

A polispermia acontece quando há falta de bloqueio da membrana do oócito à passagem de mais de um espermatozoide.

É também durante a sua permanência no istmo que os espermatozoides sofrem o processo de capacitação.

A fase final da espermatogénese, a maturação, tem lugar no epidídimo. No entanto, embora maduro, o espermato-

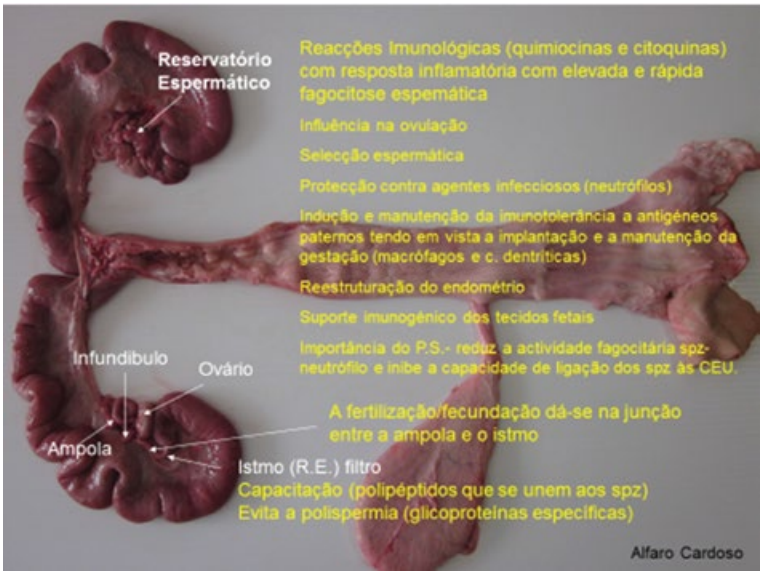
zoide não apresenta capacidade para fertilizar o óvulo. Para isso é necessário que lhe sejam retiradas ou alteradas substâncias, tal como o colesterol que ele absorveu ao nível da membrana plasmática durante a maturação, e que simultaneamente se una a polipéptidos específicos. Estas alterações que levam cerca de 6 horas constituem o processo de capacitação.

No entanto, o espermatozoide capacitado, embora consiga penetrar na camada celular do *cumulos oophorus*, não tem ainda capacidade para penetrar na corona radiata e muito menos atingir a zona pelúcida.

Para que tal seja possível, tem que sofrer ainda a reacção acrossómica que consiste na fusão do acrossoma com a sua membrana plasmática, formando poros que permitam a saída de enzimas (hialuronidase e acrosina).

Só assim o espermatozoide consegue atravessar a *corona radiata*, lisar a zona pelúcida e fertilizar o óvulo.

Esta reacção do acrossoma ocorre perto da zona pelúcida, embora para alguns autores e consoante as espécies, possa ter o seu início na ampola do oviducto.



Fertilização e desenvolvimento embrionário

O local da fertilização é na zona istmo-ampolar do oviducto. Os espermatozoides filtrados e capacitados no istmo encontram-se com os óvulos que começam a ser libertados 24-36 horas depois do início do cio (ovulação) e que vão chegando ao oviduto ao longo de 6-18 horas (duração da ovulação). Permanecem viáveis na ampola do oviducto durante 8-12 horas.

Logo após a fertilização, a estrutura gerada (zigoto) sofre o processo denominado clivagem cerca de 30 horas após a fertilização com a formação dos blastómeros.

45 horas após a fertilização o zigoto já originou 4 blastómeros.

Ao 4º dia, começam a ser libertados para os cornos uterinos aumentando para 12 a 16 blastómeros e formando uma massa compacta de células (mórula).

Passados cerca de 4 dias, a mórula começa a receber fluidos uterinos começando a surgir pequenos espaços cheios de líquido. Há um rearranjo de células, formando um aglomerado em forma de botão contendo uma cavidade cheia de líquido (blastocisto).

O conjunto de células periféricas do blastocisto irá originar o trofoblasto, o botão polar origina o embrioblasto, e a cavidade repleta de líquido dá origem ao blastocele.

O trofoblasto contribuirá para a formação de parte da placenta e o embrioblasto irá originar o embrião.

Aos 12-14 dias os blastocistos encontram-se distribuidos pelos cornos uterinos evidenciando-se já áreas de união ectodermo-epitélio endometrial e começam a produzir estrogénios, que actuam sobre o endométrio, inibindo a passagem da PGF2α até ao corpo lúteo através da veia uterina, reorientando-a para o lumen uterino. Desta forma, o corpo lúteo continua a manter a sua produção de progesterona (P4), essencial para a manutenção da gestação (reconhecimento materno de gestação-1º sinal).

Aos 24 dias a fusão endometrio-embrião está completa.

RECONHECIMENTO DA GESTAÇÃO

Reconhecimento da gestação: é bifásico e depende dos embriões

- Primeiro sinal (dia 12), E2 embrionários reorientam a PGF2α para o lume uterino
- Segundo sinal (dia 18), prolongamento da reorientação da PGF2α

Intervalos de 25-30 dias entre estros = 2º sinal falha ou é inadequado

O período entre o 18º-22º dias é o mais crítico para a implantação embrionária.

Corpo Lúteo dependência

Ao contrário de outras espécies, em que grande parte da gestação é mantida pela progesterona produzida pela placenta, a gestação na espécie porcina é mantida até ao parto, exclusivamente pela progesterona produzida nos corpos lúteos.

O nível de progesterona é crucial e para isso são necessários C. lúteos em quantidade e bem desenvlvidos.

Para isso, há que ter em conta todos os cuidados de maneio, principalmente até ao 24º dia de gestação, em que as situações mais diversas de stress possam conduzir à luteiólise total ou parcial ou a C. lúteos pouco desenvolvidos.

Mas o papel da progesterona não se limita apenas à manutenção da gestação.

O útero da porca, sob o efeito da acção da progesterona, segrega uma enorme quantidade de várias proteínas de enorme importância para a nutrição e desenvolvimento do embrião/feto.

Ainda mais importante se torna na espécie porcina, porque o trofoblasto não invade o epitélio uterino, ligando-se de uma forma muito superficial.

A falta de progesterona, condicionando a produção das referidas proteínas, tem reflexos no peso dos leitões ao nascimento, na mortalidade ao parto, a más formações (carência de vit.A) podendo levar até à morte o concepto.

Apenas dois exemplos que ilustram a importância das proteínas específicas produzidas no endométrio:

A glicoproteína uteroferrin, que transporta o ferro tão necessário ao concepto e a RPB (retinol-binding protein) e a RABP (retinoic acid-binding protein) que transportam a vit.A essencial para o desenvolvimento do concepto.

Demonstrativo da enorme importância que a presença destas duas proteínas, principalmente da RPB, tem para a boa manutenção reprodutiva e embrio-fetal, é o facto de que o próprio blastocisto aos 10 dias de gestação, já produzir RBP bem como a produção no endométrio de RBP durante a fase luteínica do ciclo éstrico da porca devido ao aumento do nível de progesterona.

Em conclusão, a fase crítica, muito assente em práticas de maneio, situase entre o dia em que a porca manifesta o cio e os primeiros 24 dias de gestação com maior relevo para os dias 18º e 22º.

A chave do sucesso reprodutivo está em larga medida ligada à manutenção em qualidade e quantidade de Corpos lúteos, associada a um bom aproveitamento dos óvulos que a porca põe à nossa disposição, através da forma como encaramos a detecção do cio, a IA/CN com todas as condicionantes relativas à qualidade do sémen ou à utilização do varrasco e a uma alimentação adequada e bom alojamento.

É fundamental portanto, aliar boas técnicas de maneio reprodutivo, a um bom maneio alimentar suportado por um bom programa nutricional adaptado à genética utilizada e às condições ambientais e de bem-estar, nunca esquecendo as elementares normas de biossegurança e sanidade, evitando situações de patologia ou de Pseudopatologia, e tendo sempre em conta de que não estamos a lidar com máquinas, mas simples animais limitados, porque sujeitos às Leis da Natureza.

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Vasco Simões

Patologia reprodutiva em suínos: abordagem diagnóstica

As falhas reprodutivas em suinicultura representam uma perda económica importante para o produtor que é com alguma frequência desvalorizada. Entre os problemas mais frequentes podemos destacar os abortos, retornos ao cio, anestro, aumento do número de fetos mumificados, aumento do número de nados-mortos e diminuição da prolificidade. Corrégé *et al.* (2013) estimou uma perda de 618€ por cada porca detetada vazia à ecografia e 724€ para um aborto a 85 dias de gestação.

O diagnóstico de um problema reprodutivo é com frequência complexo e laborioso. Em termos gerais estes podem ser classificados de origem infeciosa ou não infeciosa, agudos ou crónicos. Uma das dificuldades de diagnóstico reside no facto de estes serem muitas vezes de origem multifatorial. Entre as causas não infeciosas destaca-se, de uma forma não exaustiva, as causas ligadas ao varrasco (qualidade do sémen) e às porcas (idade, condição corporal), a alimentação (plano alimentar, equilíbrio nutricional, micotoxinas), maneio (deteção do estro, técnica e protocolo de inseminação) e fatores ambientais (infertilidade de Outono, qualidade do alojamento, temperatura das salas). No que diz respeito às causas infeciosas destacam-se patologias de origem viral (PCV2, PRRS, doença de Aujeszky, Parvovirose e Influenza) e bacteriana (Leptospirose). No entanto, a generalização dos programas vacinais e desenvolvimento de protocolos de biossegurança tem vindo, segundo alguns autores, a limitar a incidência destes agentes nos problemas de reprodução (Filha *et al.*, 2006).

A primeira etapa na abordagem a um problema reprodutivo passa pela realização de uma anamnese completa do problema. Além das informações transmitidas pelo produtor é importante proceder a uma análise exaustiva dos resultados GTTT (Gestion Technique des Troupeaux de truies – Gestão técnica do efetivo de reprodutoras) ou, na ausência destes, dos registos da exploração. A aplicação do princípio 5W1H (what, who, when, where, why, how much), permite obter uma boa descrição do quadro clinico. Segue-se a observação dos animais, exame clinico e identificação de fatores de risco que possam não ter sido referidos na anamnese. Este é um passo importante na abordagem diagnóstica uma vez que vai permitir descartar algumas das hipóteses inicialmente colocadas e orientar eventuais exames complementares para as hipóteses mais prováveis.

O recurso a exames complementares constitui a última etapa no processo de diagnóstico e não deve sobrepor-se às etapas precedentes, mas servir como ferramenta para confirmar um diagnóstico provável. Um conhecimento aprofundado da patologia suspeita é necessário no momento de decidir quais os exames mais adequados, o tipo e o número de amostras a enviar e as condições de conservação destas. A realização de autopsias *in situ* pode fornecer informações essenciais na orientação do diagnóstico, no entanto precauções especiais devem ser tomadas em termos de assepsia caso se pretenda fazer recolha de amostras para exames laboratoriais. Devido à fragilidade de certos vírus como o PRRSv, pode ser preferível enviar os animais ainda vivos ao laboratório ou, caso tal não seja possível, refrigerados. Para concluir, o número de amostras a enviar deve igualmente ser tido em conta: a título de exemplo, para um diagnóstico de parvovirose, o envio de um único feto mumificado raramente permite a identificação do agente.

Principais patologias com impacto reprodutivo em suínos

O parvovirus suino (PVS) tem sido uma das principais causas de falhas reprodutivas em porcas, ainda que a sua ocorrência tenha vindo a diminuir com a aplicação de programas vacinais nos efetivos reprodutores (Zimmerman, 2012). A infeção ocorre sobretudo por contato oro-nasal com fezes e secreções contaminadas, fetos e anexos embrionários. A infeção pode igualmente ocorrer por inseminação com sémen contaminado (Bican *et al*., 2002). As marrãs são particularmente suscetíveis, uma vez que os anticorpos maternais, que persistem entre 4 e 6 meses, encontram-se já demasiado baixos durante a primeira gestação (Paul *et al*. 1982).

Os sintomas, de índole reprodutiva, dependem essencialmente do momento ao qual se dá a infeção. Um dos sinais indicativos de parvovirose suína é a presença de fetos mumificados de diferentes tamanhos juntamente com leitões aparentemente saudáveis à nascença. Outras consequências da infeção incluem morte embrionária com retorno ao cio, pseudo-gestação e ninhadas pequenas (1-4 leitões) (Zimmerman, 2012).

A deteção do vírus realiza-se em fetos de menos de 14 centímetros (distância crown-rump lenght) por imunofluorescência ou PCR, preferencialmente a partir do pulmão e coração (Gava *et al*., 2009). Fetos com mais de 16 centímetros (correspondendo a 70 dias de gestação) são já imunocompetentes e desenvolvem uma resposta imunitária na sequência da infeção. A deteção de anticorpos faz-se recorrendo ao teste de Inibição da Hemaglutinação ou ELISA (Mieli, 2005). A utilização de testes serológicos em porcas é de difícil interpretação uma vez que a maioria dos efetivos é vacinada ou tiveram um contacto precedente com o PVS. Recomenda-se por isso, na suspeita de uma infeção recente, a realização de duas colheitas de sangue a 3 semanas de intervalo de forma a demonstrar uma seroconversão ativa.

PRRS (Porcine reproductive and respiratory syndrome) é uma patologia de suínos domésticos e selvagens provocada por PRRSv. É excretado através das secreções nasais, saliva, fezes, urina e, de forma momentânea, no sémen. A transmissão pode ocorrer por via intranasal, intramuscular (Yoon *et al*., 1999), oral, intrauterina, vaginal e transplacentaria (Bøtner *et al*., 1994). Em porcas, é responsável por uma diminuição da fertilidade com retornos ao cio regulares e irregulares, febre e anorexia, abortos tardios e partos precoces (110-113 dias), aumento do número de nados-mortos e de fetos mumificados, sobretudo no último mês de gestação.

O diagnóstico pode ser feito por deteção do vírus ou de anticorpos. Para a deteção viral recomenda-se a recolha de sangue, uma vez que o período de virémia é relativamente longo, sobretudo na sequência de um primeiro contacto (Wills *et al*., 2003). A sua identificação é igualmente possível a partir do pulmão, linfonodos, baço e amígdalas de fetos mumificados ou nados-mortos. RT-PCR é a técnica que apresenta melhores resultados. Além das amostras já referidas, esta técnica permite igualmente a análise de sémen (Benson *et al*., 2002) e de saliva (Kittawornrat *et al*., 2010). No entanto, não permite a distinção entre antigénio viral e vacinal. A imuno-histoquímica pode ser igualmente utilizada mas apresenta sensibilidade inferior (67%) (Halbur *et al*., 1996). Para a deteção de anticorpos recorre-se frequentemente ao teste ELISA devido à sua elevada sensibilidade e especificidade (Seo *et al*., 2016) e possibilidade de testar em simultâneo os dois genótipos. No entanto, devido à inexistência de vacinas DIVA (differentiating infected from vaccinated animals) este não permite a diferenciação entre anticorpos de animais vacinados e infetados.
PCV2 (Porcine Circovirus type 2), agente ubiquitário presente na maioria das explorações a nível mundial, é responsável por diferentes quadros clínicos podendo afetar porcos em crescimento e reprodutores. Está presente nas secreções nasais, saliva, fezes, urina, leite e sémen. As manifestações clinicas dependem do momento da infeção, destacando-se os retornos ao cio regulares e irregulares, aumento do número de fetos mumificados e nados-mortos, abortos e partos prematuros. Estes sintomas ocorrem com maior frequência em explorações recentes, com uma proporção elevada de marrãs, e cujo efetivo nunca tenho sido exposto ao PCV2 (Togashi *et al*., 2011).

O diagnóstico pode ser complexo uma vez que os sintomas reprodutivos não são específicos do PCV2 e os animais podem encontrar-se seropositivos sem apresentarem doença. Na presença de abortos, nados-mortos ou fetos mumificados privilegia-se a deteção viral a partir do miocárdio através de imuno-histoquímica ou PCR (Madson *et al.*, 2009). O exame histológico revela uma miocardite com focos de necrose e fibrose (Segalés *et al.*, 2005). A serologia em porcas com recurso ao teste ELISA é de difícil interpretação, pelo que se recomenda a realização de colheitas de sangue seriadas para identificar uma eventual seroconversão.

A doença de Aujeszky, também conhecida por pseudoraiva, é provocada por um Herpesvirus porcino tipo 1 (SHV). Afeta preferencialmente suínos domésticos e selvagens, sendo que estes últimos desempenham um papel importante enquanto reservatório do vírus. A transmissão entre animais faz-se essencialmente por contacto oro-nasal, ainda que o vírus se possa encontrar igualmente na urina, sémen e leite (Prodanov-Radulovіγ *et al*, 2011). Em animais gestantes o SHV atravessa a placenta provocando morte embrionária, retornos ao cio, abortos, aumento do número de fetos mumificados e de nados-mortos. As porcas podem igualmente apresentar febre e sintomas respiratórios. Os nados-mortos apresentam petéquias e focos e necrose a nível cutâneo e hepático, sinais fortemente indicativos de uma infeção a herpesvírus. Os leitões que sobrevivem à infeção in utero apresentam tremores musculares, convulsões e elevada percentagem de mortalidade 3 a 5 dias após o nascimento.

O diagnóstico é possível através da identificação do agente a partir de leitões com sintomatologia nervosa e nados-mortos ou, alternativamente, através de testes serológicos às porcas. A identificação viral faz-se a partir de swabs nasais e amostras de órgãos (cérebro, amígdalas, pulmão) recorrendo à PCR em tempo real (Fonseca Júnior *et al.*, 2013). Esta técnica permite igualmente a distinção entre antigénios virais e vacinais (Ma *et al.*, 2008). Para a deteção de anticorpos a OIE recomenda a utilização da seroneutralização ou dos testes ELISA (OIE, 2012). No entanto estes podem dar origem a falsos-negativos caso a infeção se encontre em fase latente (McCaw *et al.*, 1997).

A gripe suína, provocada pelo vírus Influenza tipo A, é responsável por importantes perdas económicas devido aos quadros respiratórios e reprodutivos que provoca. Os principais subtipos que afetam os suínos são H1N1, H1N2 e H3N2, com diferentes linhagens genéticas presentes em resultado de constantes mutações e recombinações genéticas (Lewis *et al.*, 2016). Após uma fase de incubação de 24 a 36 horas, o vírus é excretado durante um período relativamente curto (5 a 7 dias) nas secreções oro-nasais (Choi *et al.*, 2005). Os sintomas mais frequentes são febre, anorexia, prostração, corrimento nasal, tosse e dispneia. A nível reprodutivo têm sido descritos a presença de abortos, retornos ao cio e aumento do número de nados-mortos (Zimmerman, 2012). Estes sintomas parecem ser consequência da febre elevada e da ação de citocinas pró-inflamatórias, uma vez que o vírus parece não ter uma ação direta sobre os fetos (Meijer *et al.*, 2015).

À necropsia, os animais em fase clinica apresentam uma pneumonia intersticial, com lesões em "mosaico", acompanhada de rinite, traqueíte e bronquite hemorrágica. A análise histológica revela a existência de um infiltrado celular ao nível dos brônquios e bronquíolos composto essencialmente por neutrófilos e linfócitos (Watanabe *et al.*, 2012). A presença do vírus pode ser posteriormente confirmada através de imuno-histoquímica e imunofluorescência.
A deteção viral através da RT-PCR em tempo real é cada vez mais utilizada. A análise pode ser feita a partir de swabs nasais, pulmão ou saliva e permite a identificação do subtipo e linhagem genética (Simon *et al.*, 2014). Deve ter-se no entanto em consideração que o período de excreção viral pode ser relativamente curto o que poderá resultar em falsos-negativos. Os testes serológicos podem ser uteis na avaliação do estatuto imunitário do efetivo ou caso os animais já se encontrem na fase de convalescença. O teste de inibição de hemaglutinação é ainda hoje o método de referência mas tem vindo a ser substituído progressivamente pelos testes ELISA (Detmer *et al.*, 2013). Recomenda-se a realização de duas colheitas de sangue com 3 a 4 semanas de intervalo. Um aumento de quatro vezes do título de anticorpos é fortemente indicativo de uma infeção recente (Janke, 2000).

Leptospira, espiroqueta que afeta a maioria dos mamíferos, é um importante agente de infertilidade em suínos. São

conhecidos mais de 260 serovares patogénicos mas apenas um número limitado afeta os suínos. Entre eles destacam-se os serovares pomona e bratislava, pela frequência com que são identificados e pelo seu potencial patogénico em suínos (Rocha, 1998). A infeção ocorre por contacto entre as mucosas e excreções contaminados como urina, anexos embrionários ou sémen. É responsável infertilidade, abortos tardios, aumento do número de nados-mortos e fetos mumificados, ninhadas pequenas e por vezes corrimento vaginal mucopurulento.

O diagnóstico de leptospirose é extremamente complexo. Foram documentadas reações cruzadas entre serovares patogénicos e não-patogénicos nos testes serológicos, o que dificulta a interpretação dos resultados (Bolin, 1994). Certos serovares como bratislava provocam uma resposta imunitária discreta, que pode traduzir-se por um aumento passageiro e pouco pronunciado do título de anticorpos.

O teste serológico mais utilizado é o Teste de Aglutinação Microscópica (MAT). Recomenda-se a realização de colheitas de sangue seriadas de pelo menos 15 porcas de diferentes idades, imediatamente após a ocorrência de abortos e três semanas depois. Não existe no entanto consenso no que diz respeito à interpretação do título de anticorpos. Segundo Bolin (1994), uma infeção recente traduz-se por um titulo superior a 800, enquanto que para o serovar bratislava é comum estes situarem-se entre 50 e 200. O título de anticorpos vacinais tende a ser mais baixo que num caso de infeção natural. A PCR pode ser utilizada para deteção de leptospiras na urina, corrimentos vaginais e macerado de órgãos (placenta, rim, fígado e pulmão de fetos) (Miraglia *et al*., 2008).

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Rui Cordeiro

Avaliação dos dados zootécnicos para avaliação das falhas reprodutivas

Os dados zootécnicos contêm informações de máxima importância para a análise reprodutiva do efectivo reprodutor de uma exploração. Através deles conseguimos, de uma forma rápida e concisa, detectar, inespecificamente, falhas de maneio e até problemas sanitários.

Dados zootécnicos traduzem a principal ferramenta de avaliação de desempenho. Eles reflectem, de forma numérica, o desempenho de diversos parâmetros de uma exploração pecuária. Os dados zootécnicos mais utilizados são: a Taxa de Partos (Tx partos), a Taxa de fertilidade (Tx de Fertilidade), o Intervalo Desmame Cobrição (IDC), o Índice de Partos (IP), a % ou nº de Nascidos Totais (NT), a % ou nº Fetos Mumificados (FM), a % de abortos e a % de retornos. Qualquer ocorrência que impeça o óptimo desenrolar da reprodução é uma falha reprodutiva. As falhas reprodutivas mais comuns são: o Aborto, o Retorno ao Estro, o aparecimento de Fetos Mumificados e existência de porcas Vazias ao Parto. Naturalmente que todas estas falhas podem ter vários padrões. Deveremos analisar o tempo de gestação a que ocorreu o aborto, as características dos fetos, qual a "idade" do retorno ao cio, se cíclico ou acíclico... Deveremos ter em atenção os tamanhos dos fetos mumificados, a sua homogeneidade, se nasceram ou não juntamente com nascidos vivos, etc.

Há muitos recursos para analisar os dados zootécnicos. Há uma enormidade de tabelas e gráficos que nos ajudam a comparar parâmetros e, caso queiramos, a aprofundar o diagnóstico. Cada clínico deverá eleger os que mais lhe interessam por forma a se familiarizar com eles, tornando assim mais fácil a sua análise de rotina.

Existem uma série de factores, não directamente ligados à análise de dados mas sim à sua preparação, que são de extrema importância para o sucesso de qualquer tipo de diagnóstico. Dividem-se em factores humanos e não humanos. Nos humanos temos desde falhas voluntárias e não voluntárias das pessoas que recolhem os dados, erros das pessoas que os registam (quer no escritório, quer no campo) e falhas das pessoas que os analisam, quer pela sua capacidade de diagnóstico, quer pelo tempo de dedicação à análise e, muitas vezes, da sua capacidade para executar as conclusões daí retiradas. Os factores não humanos têm relevância quando comparamos dados zootécnicos retirados de diferentes programas informáticos. As fórmulas de cálculo nem sempre são consensuais e quando nos apercebemos estamos a comparar o mesmo índice calculado de maneira diferente. Por exemplo, numa determinada situação, 28,60 leitões desmamados/fêmea/ano no "PigChamp" corresponde a 29,95 leitões desmamados/fêmea/ano no "Farm".

Sendo nós clínicos, naturalmente somos levados a encontrar uma explicação "sanitária" para tudo. Na minha opinião a grande maioria nas falhas reprodutivas têm que ver com problemas de maneio. É claro que muitas destas falhas de maneio culminam com um problema sanitário mas, não vamos conseguir resolver as falhas reprodutivas, resolvendo por si só as questões sanitárias. É necessário ir ao âmago da questão, é necessário corrigir maneio, é necessário ter a certeza que o maneio não tem falhas para pensarmos na sanidade como causa dos problemas reprodutivos. Os problemas de maneio passam por questões fundamentais como Biossegurança (interna e externa), manutenção da

Condição Corporal da porca, óptima detecção de cios, exemplar acto de inseminação, boa adaptação da reposição, bom registo de dados, administração adequada de imunogénicos, etc. As patologias que, comummente causam problemas reprodutivos são: PRRS, Influenza suína, Parvovirose, Circovirose, Doença de Aujeszky, Leptospirose, Clamidiose e Brucelose.

Podemos concluir que a análise dos dados zootécnicos é uma preciosa ajuda na gestão de uma exploração, revelando-se especialmente importante na detecção de problemas reprodutivos. Há que seguir algumas regras na recolha e organização dos dados e, claramente, há que não facilitar na aplicação das conclusões retiradas. De notar que o maneio desempenha um papel fundamental no êxito reprodutivo de uma exploração. Geralmente os problemas sanitários são secundários a erros de maneio.

Gestão Veterinária

Mark Opperman, Sheila Grosdidier

Price is Only an Issue in the Absence of Value: Learn how to improve upon your practice's perception of value

Veterinary Management Consultation, Inc, Evergreen, Colorado, USA

How To Determine Your Practice's *"Perception of Value"*

What Is "Perception of Value?"

_____

_____

_____

Price is only an issue in the absence of value!

1. The "Three Minute Syndrome"
2. Internet Reviews Online - Is there a review posted about your practice?
3. How is "Perception of Value" Measured?
4. What Effect Does It Have on Your Practice?

How Do Clients Determine a Practice's "Perception of Value?"

$$\text{Value} = \frac{\text{Benefit}}{\text{Price}}$$

***Let's Put Ourselves in our Clients' Shoes and Try to Determine our Practice's "Perception Of Value"**

The Initial Contact

1. Telephone
   - Message on hold
   - Telephone etiquette
2. In person

Automated answering services can cause you to lose up to 50% of your clients!

The Initial Impression

1. Signage

2. Outside Physical Environment

- Building appearance
  - Your entrance-way
- Parking lot
- Landscaping

3. Marriott versus *Motel 6*

4. Entering the Reception Area

- Appearance of reception room
- Cleanliness, odor control
- Comfort of seating
- Distractions - ease of managing one's pet
- Background music
- Condition and age of magazines and client education material
- Posters and informational wall hangings affixed to the wall

5. The Receptionist: The first and most important contact

- How was the client greeted?
  - Smile; Use of pet's name; Knowledge of what services the client needs

The Receptionist: The first and most important contact at your practice!

- How well do our receptionists present themselves?
  - Professional, Knowledgeable & Informative, Uniforms & Name Badges, Organized or Disorganized, Handling of stressful situations
- I can't find the medical record!
  - Paper Files vs. Electronic Files
  - Writing Tablets

6. The Wait: constructive or destructive time?

- How long is too long? _____
  - Communication with the client
- Appointments kept on schedule
  - Ten Minute Flex Scheduling
  - "E" Slots
  - Discharge Appointments
- Effectively utilizing this time
  - Educational CDs, DVDs or video tapes
  - Client handouts
  - Exam Room Technicians

Technician Appointments	10 Minute	20 Minute	30 Minute	40 Minute
Nail Trims	Recheck Appointments Seen Within Past 30 Days	Canine/Feline Annual Visits —Under 6 Years Old—	Canine/Feline Annual Visits —6 Years or Older—	New Puppy or Kitten Visit
Blood Draws Only	Nail Trims with Doctor	Check Paw, Minor Medical Problem, Etc.	New Pet Visit —1 Year or Older—	ALL Exotics Visits
SQ Fluids	Anal Gland Expression with Doctor	Health Certificate / Exam	Check Ears or Check Skin Allergies (Itching)	Skin Lesions, Ear & Skin Problems, Bad Allergies
Suture Removals	Microchipping Only with Doctor	Soft Paws Applications	Check limping or Check lump	
Anal Gland Expression	2nd Bordetella Vaccination	6 Month Exam	ADR or Vomitting, Diarrhea, Not Eating, Etc.	
Microchipping Only	2nd Lyme Vaccination		Most Medical Problems	
			Animals Starting Allergens	

The Office Visit

1. Escorted to the exam room

2. Greeted by doctor or exam room technician

3. Appearance of exam room
   - Distractions

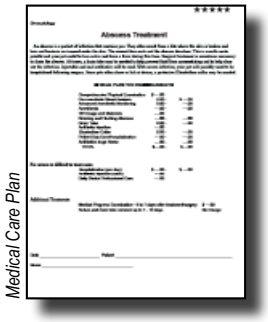
4. Appearance of doctor and staff

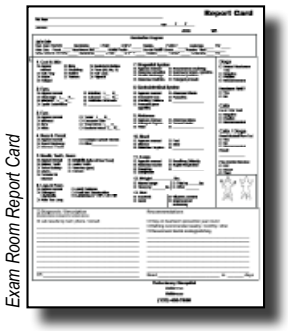
5. The "ideal" out-patient office visit

- Greeting
- Overview of visit
- Special touches
- What else needs to be handled?
- Client needs communicated to doctor
- Physical exam
- Review & recommendations


Pre-Exam Checklists


Dental Report Card


Medical Care Plan


Exam Room Report Card

6. Doctor's "bed side manner"

- Body language
- Quality of time spent
- Handling of patient
- Communicative ability
- Treatment of patient
- Use of paraprofessional staff
- To treat or not treat in the exam room
- Concluding the visit

Have you considered video surveillance to ensure office visit consistency?

What do You Consider Your Practice's "Perception of Value" to be?

7. Eight steps to a successful exam room visit
    - Introduction
    - Talk to and touch the pet
    - Do something
    - Say something
    - Show something
    - Give them something
    - Listen
    - End on a positive note

The Exit

1. Processing paper work
2. Filling prescriptions
3. Handling of payment
4. Answering questions

Mark Opperman, Sheila Grosdidier

Financial Management: What to do and what to look for to improve the bottom line of your practice

Veterinary Management Consultation, Inc, Evergreen, Colorado, USA

Financial Management: What to Look for and What to Do About It

I. What Are Your Practice's Pulse Points? — Scoreboard Your Practice

A. Revenue Overall Hospital
   1. Monthly Sales
   2. New Clients
   3. Number of Invoices
   4. Average per invoice (ACT)

B. Develop Doctor Production Report
   1. Monthly Sales
   2. Number of invoices generated
   3. Average client charge (ACT)

C. All information should be benchmarked
   1. This month / Last month
   2. This year / Last year

D. All information should be compared with industry standards
   1. Industry standards per full-time equivalent DVM for overall hospital invoices
      a. Monthly sales - $54,016 ($648,200/year)
      b. Number of invoices – 442 per month or 5,300 per year
      c. ACT - $122.30
   2. Industry standards per full-time equivalent DVM for DVM professional services
      a. Monthly sales - $48,500 ($582,000/year)
      b. Number of invoices – 242 per month or 2,900 per year
      c. ACT - $200.69
   4. Average number of new clients per full-time equivalent doctor
      a. 219 per year
      b. 18.25 per month
   5. Average number of new patients per full-time equivalent doctor
      a. 33.5 per month or 402 per year
   6. Average number of active clients per full-time equivalent doctor
      a. 931
   7. Average number of active patients per full-time equivalent doctor
      a. 1,383 per year

E. How do you increase the average invoice?
   1. Charge for what you do

2. Institute controls to make sure all charges are billed
3. Review pricing structures and product mark-ups
4. Review discount policies
5. Excel at client education to market existing and new services

II. Look for Strengths and Weaknesses Within the Income Areas of Your Practice

A. Comprehensive Physical Exam

B. Wellness Comprehensive Physical Exam

C. Items to Consider

1. Hospital Services
   a. Are surgeries and anesthesia procedures timed and billed appropriately?
   b. Are medical care plans being used?
2. Current Trends in Revenues of Most Hospitals

MEDICAL SERVICES	**% of Total Revenue**
Examinations/Consultations	14.8%
Professional Services + Diagnostic Services	4.6%
Diagnostic Imaging	4.8%
Vaccinations	7.5%
Laboratory	17.6%
Surgery	5.3%
Anesthesia	3.6%
Hospitalization	1.8%
Treatments	3.3%
Dentistry	2.5%
Integrative Therapies	0.4%
Other	.9%
Discounts	2.1%
Total	***69.2%***

MEDICAL PRODUCTS	**% of Total Revenue**
Medicine Dispensed	12.5%
Flea, Tick, Heartworm Products	9.3%
Online Pharmacy	.2%
Total	***22%***

NON-MEDICAL REVENUE	**% of Total Revenue**
Over-the-Counter Products	.9%
Diets (therapeutic & retail)	4.3%
Bathing & Grooming	1.4%
Boarding	2.2%
Total	***8.8%***

3. Consider Additional Services - Ancillary

Puppy Training Classes	> 20% of practices	House Calls	> 35% of practices
Adoption Programs	> 20% of practices	Behavior Counseling	> 45% of practices
Pet Transportation	> 20% of practices	Nutrition Counseling	> 50% of practices

4. Consider Additional Services – Medical
   a. Ultrasound
   b. Acupuncture
   c. Endoscopy
   d. Ophthalmology
   e. Laser Surgery

Invest in new equipment - Today's hospital budget is 2 to 3% of gross revenue

Enter your practice data in the light blue cells.

Clinic Name Here

End of Month Report

Month: March
Year: 2016

	Last Year:			Last Month:	
INCOME	**Mar 2016**	**Mar 2015**	**Difference**	**Feb 2016**	**Difference**
Total Income					
Number of Business Days					
Transactions					
Average per Transaction					
Number of New Clients					
Total Accts Receivable					

INCOME ACCOUNTS					
Professional Services			Boarding		
Dentistry			Grooming		
Surgery			Diet Products		
In-house Lab			Pharmacy		
Outside Lab			Flea Products		
Radiology			General Inventory		
Anesthesia			Other Income		
Clinical Trial			Discounts		

Forecasted Income		More than last year
Forecasted Reached?		

Accounts Payable	
Previous Balance:	
+ Current A/P*:	
- Payments Made:	
Accounts Payable Balance:	

EOM Checking Account Balance

* Current A/P is <= 30 days, incurred since last report

STAFF SALARIES			
	Industry Avg	Dollars	% to Gross
Receptionist	6% - 8%		
Technicians/Assistants	9% - 10%		
Animal Handlers	4%		
Administrative	3% - 4%		
Staff Veterinarians	14%		
Total			

Clinic Name Here

End of Month Report

Month: March
Year: 2016

Cost of Goods Sold	$0.00	% of Gross Rev		$0.00	% of Gross Rev
Dentistry			Pharmacy		
Surgery			Flea Products		
In-house Lab			General Inventory		
Radiology			Grooming		
Anesthesia			Other		
Dietary Products			Outside Lab		

SALES BY PROVIDER

		Last Year		Last Month	
Dr. One	Mar 2016	Mar 2015	Difference	Feb 2016	Difference
Total Income					
Transactions					
Average per Transaction					

Dr. Two	Mar 2016	Mar 2015	Difference	Feb 2016	Difference
Total Income					
Transactions					
Average per Transaction					

Dr. Three	Mar 2016	Mar 2015	Difference	Feb 2016	Difference
Total Income					
Transactions					
Average per Transaction					

Dr. Four	Mar 2016	Mar 2015	Difference	Feb 2016	Difference
Total Income					
Transactions					
Average per Transaction					

III. Some Points to Consider

A. Our ability to drive service revenue is a factor of time and efficiency

B. A doctor's time is a finite and scarce commodity

1. "There are only twenty-four hours in a day."

C. More Transactions

1. Less doctor time per transaction

D. Less Transactions

1. More doctor time

E. Quality of Personal vs. Professional Life

All practitioners must find a balance of time and efficiency that is perceived to be fair and equitable to the client.

IV. How Can We Increase Our Income and Improve Upon Our Quality of Life?

A. Delegate and leverage your health care team.

1. Make your staff a resource, not a cost

B. Ask Yourself: What tasks or activities currently performed by the doctor can be delegated to members of the veterinary health care team?

C. Develop passive income areas of the practice by utilizing your veterinary health care team.

D. Look to those services that don't require direct, full-time involvement of the doctor.

E. What are the most common passive income areas?

1. Dental Cleanings
2. Radiograph Procedures
3. Lab Work
4. Nail Trims
5. Ear Cleanings
6. Bandage Changes
7. Anesthesia Induction
8. Boarding
9. Grooming

V. Characteristics of Successful Practices

A. Will average 38% more visits per week
B. Doctors will not work an excessive number of hours
C. Will produce 51% more personal gross revenue
D. Will receive more than 100% greater personal compensation
E. Will utilize 1 additional non-DVM staff per DVM on practice than others
F. Will average total expenses 2% greater than other practices
G. Will have an average client transaction $7-$10 higher than others

VI. Invest in Continuing Education

A. Budget .6-1% of gross revenue and include entire health care team in training

VII. Industry Trends

A. Limited DVM availability
B. How do we leverage our doctors' ability to enhance revenue?

VIII. Can We Achieve a "10" Quality of Personal and Professional Life?

A. Dream or Reality?

IX. Variable Expenses as a % of Total Revenue

Drugs and medical supplies	10.1%
Heartworm/flea/tick products	4.5%
Laboratory	4.1%
Cremation/care of remains	0.6%
Diets (therapeutic and retail)	3.1%
Items purchased for OTC sale	0.4%
Practice vehicle expense (gas, repairs, Ins.)	0.2%
Collection expense	0.0%
Bad debt/returned checks	0.1%
Credit card/merchant fees	1.6%
Refunds to clients	0.0%
Medical waste disposal/radiation badge monitoring	0.1%
Online pharmacy drug cost	0.1%
Online store food cost (therapeutic & retail)	0.1%
Online store items purchased for OTC retail	0.0%
Sales and use tax	0.7%
TOTAL	**25.7%**

X. Fixed Expenses as a % of Total Revenue

Advertising and promotion	0.8%
Professional/business dues and subscriptions	0.3%
Office and computer supplies	0.9%
Postage/freight and delivery	0.3%
Printing	0.1%
Insurance (health and liability)	1.9%
Insurance (workers' comp./industrial insurance)	0.6%
Telephone, answering service, Internet	0.5%
Continuing education, meetings and travel	0.6%
Business meetings	0.1%
Repairs and maintenance - equipment	0.5%
Business consultation services	0.4%
Accounting services	0.3%
Legal services	0.1%
Payroll service/retirement administration cost	0.2%
Laundry and uniforms	0.1%
Business gifts and flowers	0.1%
Charitable contributions	0.1%
Entertainment	0.1%
Bank charges	0.1%
Other/miscellaneous	0.2%
Technical (IT) support contracts	0.4%
Recruitment and relocation	0.0%
Licenses and permits	0.2%
Personal property tax	0.2%
Franchise tax and other taxes	0.1%
TOTAL	**9.2%**

Doctor Production Report

Dr. 1Name	March					April					May				
	2016	% Gross	2015	% Gross	% Diff	2016	% Gross	2015	% Gross	% Diff	2016	% Gross	2015	% Gross	% Diff
Diagnostic Income	$7,943.42	22.34%	$7,387.55	24.96%	-0.03	$7,533.00	24.21%	$7,865.85	27.41%	-0.03	$10,887.93	27.34%	$6,373.61	22.21%	0.05
Pharmacy Income	$8,291.88	23.32%	$5,318.68	17.97%	0.05	$6,030.13	19.38%	$5,839.84	20.35%	-0.01	$8,490.51	21.32%	$4,984.67	17.37%	0.04
Professional Vlue Inc	$17,696.69	49.77%	$13,031.80	44.03%	0.06	$14,558.82	46.79%	$13,662.65	47.61%	-0.01	$18,227.52	45.77%	$13,192.02	45.97%	0.00
Miscellaneous Income	$1,624.95	4.57%	$3,859.52	13.04%	-0.08	$2,993.29	9.62%	$1,328.67	4.63%	0.05	$2,218.21	5.57%	$4,146.72	14.45%	-0.09
TOTAL INCOME	$35,556.94	100.00%	$29,597.55	100.00%		$31,115.24	100.00%	$28,697.01	100.00%		$39,824.17	100.00%	$28,697.02	100.00%	
Average Invoice	$139.99		$130.39			$142.08		$133.47			$145.34		$120.07		
Number of Invoices	254		227			219		215			274		239		

Dr. 2Name	March					April					May				
	2016	% Gross	2015	% Gross	% Diff	2016	% Gross	2015	% Gross	% Diff	2016	% Gross	2015	% Gross	% Diff
Diagnostic Income	$9,652.18	27.41%	$7,508.77	23.34%	0.04	$6,991.76	20.18%	$6,927.83	22.21%	-0.02	$11,144.35	30.41%	$7,280.31	23.34%	0.07
Pharmacy Income	$7,166.06	20.35%	$6,820.30	21.20%	-0.01	$6,049.37	17.46%	$5,418.12	17.37%	0.00	$7,091.19	19.35%	$6,612.79	21.20%	-0.02
Professional Vlue Inc	$16,765.42	47.61%	$13,663.13	42.47%	0.05	$18,196.60	52.52%	$14,339.15	45.97%	0.07	$16,714.70	45.61%	$13,247.41	42.47%	0.03
Miscellaneous Income	$1,630.41	4.63%	$4,179.04	12.99%	-0.08	$3,409.26	9.84%	$4,507.30	14.45%	-0.05	$1,696.76	4.63%	$4,051.89	12.99%	-0.08
TOTAL INCOME	$35,214.07	100.00%	$32,171.24	100.00%		$34,646.99	100.00%	$31,192.40	100.00%		$36,647.00	100.00%	$31,192.40	100.00%	
Average Invoice	$147.34		$134.61			$146.19		$135.62			$145.42		$136.81		
Number of Invoices	239		239			237		230			252		228		

Dr. 3Name	March					April					May				
	2016	% Gross	2015	% Gross	% Diff	2016	% Gross	2015	% Gross	% Diff	2016	% Gross	2015	% Gross	% Diff
Diagnostic Income	$5,544.22	23.17%	$5,277.37	27.34%	-0.04	$4,503.18	20.48%	$4,181.03	22.34%	-0.02	$5,558.07	23.17%	$4,671.37	24.96%	-0.02
Pharmacy Income	$4,816.79	20.13%	$4,115.34	21.32%	-0.01	$4,355.86	19.81%	$4,364.44	23.32%	-0.04	$4,828.82	20.13%	$3,363.16	17.97%	0.02
Professional Vlue Inc	$12,538.50	52.40%	$8,834.86	45.77%	0.07	$11,090.85	50.44%	$9,314.67	49.77%	0.01	$12,569.82	52.40%	$8,240.41	44.03%	0.08
Miscellaneous Income	$1,028.92	4.30%	$1,075.16	5.57%	-0.01	$2,038.31	9.27%	$855.30	4.57%	0.05	$1,031.49	4.30%	$2,440.49	13.04%	-0.09
TOTAL INCOME	$23,928.43	100.00%	$19,302.73	100.00%		$21,988.20	100.00%	$18,715.44	100.00%		$23,988.20	100.00%	$18,715.43	100.00%	
Average Invoice	$104.49		$88.14			$100.40		$84.69			$103.85		$84.69		
Number of Invoices	229		219			219		221			231		221		

Medical Care Plan

Dermatology

ABSCESS

An abscess is a pocket of infection that contains pus. They often result from a bite where the skin is broken and hair and bacteria are trapped under the skin. The wound then seals and the abscess develops. This is usually quite painful and your pet could be less active and have a fever during this time. Surgical treatment is sometimes necessary to drain the abscess, then your pet will be placed on antibiotics. With severe infection, your pet will possibly need to be hospitalized following surgery.

ESTIMATE OF COSTS FOR DRAINING ABSCESS

Examination	$ 0.00		
Draining and Flushing Abscess	0.00	-	0.00
Drain Tube	0.00	-	0.00
OR Usage and Materials	0.00		
Anesthesia	0.00	-	$ 0.00
Patient Day Care/Hospitalization	0.00		
Antibiotic Injection	0.00		
Elizabethan Collar	0.00	-	0.00
Antibiotics to go Home	0.00	-	0.00
TOTAL	$ 0.00	-	$ 0.00

For severe or difficult to treat cases:

Hospitalization (per day)	$ 0.00
Antibiotic Injection (each)	0.00
Daily Doctor Professional Care	0.00

Additional Treatments:

Recheck exam recommended 3-7 days after treatment/surgery	$ 0.00
Suture and drain tube removal up to 7-10 days	Included

Signed _____ Date:_____
(owner)

Mark Opperman, Sheila Grosdidier

Team Building: What do you need to do to develop and maintain “10” employees?

Veterinary Management Consultation, Inc, Evergreen, Colorado, USA

How to Develop a Team of "10" Health Care Team Members

- What is a "10" employee?

Creating a Resource Pool of Applicants

- In house advertising
- Employee referral program
- Okay, steal them
- Print
- Internet
- Employment agencies
- Employee Referral Program (found online)
- Other - Veterinary or Technician schools, Associations

Three Step Interview Process

- Initial Interview
  - Atmosphere
  - Overall impression
  - Match candidate to position – qualifications
  - First impressions
- Second interview
  - In-depth
  - Interview Report form - page 43
  - Tour of practice – *observe responses*
- Observational interview
  - Purpose
  - Observational Interview Release form - page 45

Reference checking

- Script for Checking references - page 47
- Pre-Employment Questions to Avoid - page 49
- Issues with standard references
- Online references
- Options
- Pre-employment testing

Training Employees

- How to Recognize a Potentially Great Health Care Team Member - page 51
- Job Descriptions
  - New Trends
  - Primary Functions/Essential Tasks
  - Dynamic
  - Standard Job Descriptions - page 53
  - Tiered Job Descriptions - page 57

Orienting new team members

- Phase Training Programs - page 67
  - Develop a step wise plan for all positions
  - Create a time-line
  - Consistency
  - Match the job description
  - Assign a training nurse or supervisor/mentor
  - Slowly expand a team member's responsibilities
- Recommendations for training new team members

1.

2.

3.

4.

5.

Electronic Background Screening

- Why?
  - Cost
  - Liability
  - Relevance

Options Include:

- Criminal History - County, State, and Federal
- Positive / Negative Credit - Caution
- Sex Offender Registry
- Social Security Verification
- Employment Verification
- Education Verification - licenses and degrees
- Driving Violations
- Financial Industry
- Workman's Compensation - Caution
- National Theft Database
- Drug and Alcohol Database

BACKGROUND SERVICES
www.hireright.com
www.intellicorp.com
www.PSIbackgroundcheck.com

Payroll Companies
ADP
Paychex

Drug Screening

- Over 70% of illegal drug users are employed, as are heavy and binge alcohol users
- These individuals -
  - Have absenteeism
  - Are less productive
  - Change jobs frequently (when screened)
  - Have high numbers of accidents
- Many insurance companies offer a discount on workman's compensation insurance
- Avoid liability

Medical Marijuana

- At this time, testing for marijuana by employers for "medical or compassionate" users is allowed
- Most states that allow marijuana use view the law as a criminal law and not a workplace law
- The courts are testing all laws, but it appears that safety in the workplace is preferential
- Know that ADA rules may apply in certain circumstances
- Check your state laws regularly

Pre-Employment Testing - Other Types

- Competency - Job Knowledge
- Integrity
- Personality
- Physical Exam
- Personality

INTERVIEW - FIRST IMPRESSION

Name of Applicant: _____

Date: _____ Evaluated by: _____

Rate each category: 1=Poor, 3=Average, 5=Great!
Circle rating and check appropriate boxes.

1. APPEARANCE **1 2 3 4 5**

- ❑ Neat ❑ Sloppy
- ❑ Well groomed ❑ Lack of Grooming
- ❑ Appropriate Attire ❑ Dressed for: ____

2. ATTITUDE **1 2 3 4 5**

- ❑ Enthused ❑ Lacking Spirit
- ❑ Interested ❑ Lack of Interest
- ❑ Inquisitive ❑ Dull

3. PERSONAILITY **1 2 3 4 5**

- ❑ Assertive ❑ Arrogant
- ❑ Confident ❑ Presumptuous
- ❑ Reserved ❑ Shy, Timid
- ❑ Good eye contact ❑ Poor eye contact

Should we schedule an interview?

- ❑ **Yes** ❑ **No**

INTERVIEW REPORT

Name _____ Date _____

Position Desired _____

Check the appropriate box in each category, then make additional comments below.

APPEARANCE	BEARING	EXPRESSION	JOB KNOWLEDGE	MOTIVATION	PERSONALITY
❑ Indifference tc attire & grooming sloppy, unkempt	❑ No bearing, lacks confidence slovenly posture	❑ Uncommuni-cative, confused thoughts, poor vocabulary	❑ None as pertains to this position	❑ None, apathetic, indifferent, disinterested	❑ Unpleasant
❑ Careless in attire, poor grooming	❑ Often appears uncertain, poor posture	❑ Poor speaker, hazy thoughts, ideas	❑ Will need considerable training	❑ Doubtful interest in position	❑ Slightly objectionable
❑ Functional attire, neatly groomed	❑ Holds self well, seems confident	❑ Speaks well, expressed ideas adequately	❑ Basic, but will learn on the job	❑ Sincere desire to work	❑ Likeable
❑ Well groomed	❑ Sure of self, reflects confidence	❑ Speaks, thinks clearly, with confidence	❑ Well versed in position, little training needed	❑ Strong interest in position asks questions	❑ Pleasing
❑ Immaculate attire and grooming	❑ Highly confident, inspires others asserts presence	❑ Exceptional, speaks clearly, concisely with confidence, ideas well thought out	❑ Extremely well versed, able tc work without further training	❑ Highly motivated, eager to work, asks many questions	❑ Extremely pleasing, charming individual

Overall Impression:

- ❑ Unsatisfactory
- ❑ Marginal
- ❑ Satisfactory
- ❑ Very Good
- ❑ Excellent

ADDITIONAL COMMENTS: _____

Should we interview further?

- ❑ Yes
- ❑ No

_____
(Interviewer)

_____
(Date)

Observational Interview Release Form

(Date)

TO: Hospital Name
Address
Address

It is understood and agreed that my observation at (Hospital Name) is strictly on a non-work basis with no remuneration associated with the activity. I am neither an employee nor an agent, nor am I associated with the (Hospital Name) in any capacity other than as an observational interview for employment and therefore, hold the (Hospital Name) and its associates harmless and free of any and all claims which may arise as a result of my observational interview.

Witnessed this _____ day of _____, 20_____

Witness	**Applicant**
_____	_____
(signature)	**(signature)**
_____	_____
(print name)	**(print name)**

EMPLOYMENT REFERENCE PHONE SCRIPT

[*Applicant*] has applied for a position with our business and you were listed as a former employer. [*Applicant*] has signed a release that authorizes you to give us the following information. [*Give identifying information they request to help them locate the applicant's file. You may have to offer to send a copy of the applicant's release form to the former employer in order to obtain the information.*]

Can you verify that [*Applicant*] worked for your company from [*date*] to [*date*]?

What was [*Applicant's*] job title?

Could you give me a brief description of the duties [*Applicant*] performed?

Please verify that [*Applicant's*] final rate of pay was $[*amount*] hourly / weekly / biweekly / monthly / annually.

Was [*Applicant*] reliable?

Was the work [*Applicant*] performed satisfactory?

Did [*Applicant*] get along with coworkers and supervisors?

If relevant, did [*Applicant*] get along with customers or clients?

What reason did [*Applicant*] give for leaving your employ?

Would you rehire [*Applicant*]?

Would you recommend [*Applicant*] for a position as [*position you are trying to fill*]?

Thank you for taking the time to speak with me. Is there anything else that you think I might find helpful in making a hiring decision with respect to [*Applicant*]?

Checklist of Pre-Employment Questions to Avoid

The employer should avoid any of the following questions either on the application form or during the interview at the risk of providing evidence of unlawful discrimination.

1. Marital Status, or "Mr./Miss/Mrs."
2. Prior Married Name/Maiden Name (another way of inquiring about gender and marital status)
3. Spouse's Name/Spouse's Work
4. Age/Date of Birth (except may ask if at least 18 if that is a requirement of the job)/Dates of Education or Work
5. Sex- only acceptable where Bona Fide Occupational Qualification (BFOQ) exists
6. Past Salaries or Expected Earnings
   a) Lowest Salary Willing to Accept
   b) Salary Desired
7. Height and Weight – only acceptable where BFOQ exists
8. Race/Color/Color of Eyes/Hair/Skin Complexion
9. National Origin/Fluency Requirements
10. Citizenship (except may ask if legally permitted to work in U.S.)
11. Number of or Age of Children/Day Care Plans/Plans for Having Children
12. Garnishments
13. Credit References/Indebtedness (absent job relatedness and business necessity)
14. Arrest Record (Compare conviction record, but conviction may not be absolute bar to employment, must consider recency and job-relatedness) - convictions, not arrests
15. Certain Questions Regarding Military Service
    a) Current Military Status
    b) Branch of Service
    c) Type of Discharge
16. Health/Disability/Physical or Mental Impairments/Smoker/AIDS
17. Person to Contact in Case of Emergency

18. Religion/Creed (including association/memberships without qualifying statement).

19. Availability for Saturday or Sunday work (may state that the veterinary practice is open in the evening and Saturday and give the hours the practice is open. Then ask if there are any hours that applicant is not available to work)

20. Nepotism/Relatives Working for Company

21. Sexual Orientation/Affectional Preference. See also local ordinances

22. Status with regard to Public Assistance

23. Union Membership or Sentiments

24. Social Security Number

25. Owns Own Home

26. Transportation Arrangements

27. Past Civil Lawsuits/Judgments Against Applicant

28. Denied Fidelity/Surety Bond or Government Clearance

29. Past Worker's Compensation Claims/Injury History

30. Driver's License - Unless Part of Job

31. Addictions

32. High School Diploma

33. Photographs

How to Recognize Potentially Great Health Care Team Members Before You Hire Them…

- **They put family first**, before work.
- Money is important, but **job satisfaction is their number one concern**. They tend to work long hours and take pride in their work.
- **They have a well-defined sense of their role**. This can be detected in the manner in which they dress for the interview. They have a lot of energy that can be directed in a positive way.
- **They are emotionally mature**. This is evidenced in their concern for others, financial judgement, and length of time previously employed.
- **They are compatible**. Remember, you'll have to work long hours with these individuals, so trust your feelings about getting along.
- **They are motivated based on family destiny**. Ask about father's and mother's occupation, type of education planned for family, and how the candidate relates to other members of the family.
- **They can channel hostilities**. Hire someone with enthusiasm, not a rebel willing to jump on any bandwagon.
- **They want a boss they can respect** as being competent.
- **They consider accuracy a crucial element** of the job.
- **They have a need to finish a task** once it's started.

– JOB DESCRIPTION –

RECEPTIONIST

INTRODUCTION

The purpose of this position is to serve as receptionist at [Practice Name], to perform record keeping duties, to perform clerical duties related to patient care and treatment, and to provide miscellaneous support to the veterinary practice manager and health care team. These service functions include, but are not limited to, reception (visitor and telephone), maintenance of veterinary medical records, accounts maintenance, cash processing, data entry, word processing and mail service. This position requires a practical knowledge of hospital organization and services, the basic rules and regulations governing visitors and animal patient treatment, data transcribing, word processing, and a practical knowledge of the standard procedures, veterinary records and terminology used in the hospital.

PRIMARY JOB RESPONSIBILITIES

- Provide friendly, quality client care to the patients and clients of [Practice Name].
- Receive incoming calls, screen those that are handled by other health care team members and take care of routine calls. The routine calls include those seeking information about veterinary services ("telephone shoppers"). Provide knowledgeable sub-professional advice concerning the care and treatment of animals.
- Follow established hospital policies and procedures in referring clients for immediate treatment of their pets when requests are accompanied by complaints of acute symptoms. Determine nature of injury/illness and attempt to reassure distressed pet owners. Determine whether immunizations and/or tests are current. Recommend update of necessary immunizations and/or tests to clients when applicable.
- Schedule appointments, obtaining all necessary data concerning the patient and owner. Prepare all required forms in advance when possible.
- Prepare to receive appointments by retrieving client records, preparing needed forms in advance of clients' arrival. Complete required forms such as new client form, patient visit form, client report, consent forms, estimates, payment agreements, etc and obtain all necessary information.
- Check clients in - Greet clients in a professional, friendly, hospitable manner.
- Discharge patients. Review charts of patients being discharged from the clinic for completeness of information, make new appointments or note changes in patient status as necessary. Enter charges and set up future reminders in system. Present clients with medications, instruction.
- Assure that all financial obligations are met by owners. Collect client fees, make change, process credit card transactions and assist in making count of cash drawer, run end of day transactions.
- Perform over-the-counter selling of specialty merchandise comprised of pet grooming aids and sundry veterinary items. Exercise technical knowledge of products sold and demonstrate salesmanship abilities. Explain and demonstrate products, answer questions concerning products purchase/ use.

- Fill veterinary prescriptions with appropriate medication; provide routine instructions to owners concerning prescriptions for medications.
- Collect lab specimens from pet owners, match patient record to the sample and submit samples to veterinary technician or nurse.
- Assist in the updating of client files; prepare and mail thank you cards and "welcome aboard" cards, reminders. Follow-up with clients when clinic records indicate no recent visits.
- As required, enter data into the computer system, retrieve and modify computerized records. The practice management software includes, but is not limited to, such areas as reminder list of patients for periodic notifications, receipt and/or invoicing to update medical/financial records; accounting to include the general ledger, accounts payable, accounts receivable, billing and aging of accounts, income distribution, inventory control, client records, pet records, medical records, payroll; word processing to produce letters for general correspondence and special mailings to clients, etc.
- Perform a variety of clerical duties, receiving, sorting, distributing mail, sending out mailings, cleaning, organizing reception area, type memos, correspondence, reports and other documents. Assist in the ordering, receiving, stocking and distribution of supplies.
- Work well with all employees and ensure that your actions support the hospital, the doctors, and the practice philosophy.
- Perform other duties as assigned.

CONTROLS OVER WORK

The receptionist works under the direct supervision of the receptionist team leader/office manager and/or veterinary practice manager, who will indicate general assignments, limitations and priorities. Recurring assignments are performed independently. Deviations or unfamiliar situations are referred to the supervisor. Completed work is reviewed for technical accuracy and compliance with established procedures.

SKILLS AND KNOWLEDGE

- Possession of strong organizational skills.
- Excellent verbal and written communication skills. Possess exceptional interpersonal communication skills.
- Knowledge of hospital procedures and operating instructions for making appointments, assembling patient medical records, recording test results, relaying information regarding patient's condition, and compiling and submitting data on patients treated.
- Knowledge of the spelling and meaning of commonly used terminology of veterinary medicine to accurately record results of tests and file veterinary medical reports according to alpha, numeric or subject matter headings.
- Requires strong client service skills. Personal contacts are with pet owners affected by a variety of problems, visitors and other healthcare team members. Considerable tact and diplomacy is required. Must accurately relay owner's account of the medical complaint(s) of the pet(s) involved to the healthcare team member who will be involved in treating the patient(s).

- Knowledge of the structure and content of the English language including the meaning and spelling of words, rules of composition, and grammar.
- Ability to work independently on assigned tasks as well as to accept direction on given assignments.
- Knowledge of computers and relevant software applications including MS Office (Word).

PHYSICAL DEMANDS

The physical demands described here are representative of those that must be met by an employee to successfully perform the essential functions of this job. Reasonable accommodations may be made to enable individuals with disabilities to perform the essential functions.

Amount of Time Spent on Task

Task	None	Less than 1/3	1/3 to 2/3	More than 2/3
Stand				**X**
Walk				**X**
Sit			**X**	
Use hands to finger, handle, or feel				**X**
Climb or balance		**X**		
Stoop, kneel, crouch, or crawl		**X**		
Talk or hear				**X**
Taste or smell			**X**	

The job requires the following lifting requirements and/or exerted force be performed on the job.

Amount of Time Spent on Lifting Amounts

Lifting Amount	None	Less than 1/3	1/3 to 2/3	More than 2/3
Up to 10 pounds			**X**	
Up to 25 pounds			**X**	
Up to 50 pounds			**X**	
Up to 100 pounds		**X (with assistance)**		
More than 100 pounds		**X (with assistance)**		

Specific vision abilities required by this job include close vision, distance vision, color vision, peripheral vision, depth perception and ability to adjust to focus.

WORK ENVIRONMENT

While performing the duties of this job, the employee is exposed to hazards associated with aggressive patients; hazards associated with infected animals and controlled substances; exposure to unpleasant odors and noises; exposure to bites, scratches and animal wastes; possible exposure to contagious diseases.

Note: When duties and responsibilities change, job description will be reviewed and subject to changes of business necessity.

ESSENTIAL FUNCTIONS:

- Professionally administer all phone calls - answering client inquiries in a prompt and friendly manner, scheduling appointments, recording messages.
- Requires strong communication and client service skills. Considerable tact and diplomacy is required. Ability to greet clients in a professional, friendly, hospitable manner - check clients in, discharge patients.
- Collect client fees, post and record payments, make change, process credit card transactions and run end of day transactions.
- Input data into computer software system.
- Open and close practice.
- Perform a variety of clerical duties, mailings, cleaning, organizing reception area, type memos, correspondence, reports and other documents.
- Ability to multi-task.
- Regular attendance and timeliness are an essential function in order to fulfill the requirements of this position.
- Perform general physical activities that require bending, standing, stooping, moving from room to room, sit, talk, and listen; may be required to walk or stand for long periods of time; will use hands to manipulate, handle, or feel; will reach with hands and arms.

– JOB DESCRIPTION –

RECEPTIONIST – TIER III

INTRODUCTION

The purpose of this position is to serve as receptionist at the [Practice Name], to perform record keeping duties, to perform clerical duties related to animal patient care and treatment, and to provide miscellaneous support to the veterinary practice manager and healthcare team. These service functions include, but are not limited to, reception (visitor and telephone), maintenance of veterinary medical records, accounts maintenance, cash processing, data entry, word processing and mail service. This position requires a practical knowledge of hospital organization and services, the basic rules and regulations governing visitors and animal patient treatment and a practical knowledge of the standard procedures, veterinary records and terminology used in the hospital.

PRIMARY JOB RESPONSIBILITIES

- Direct Receptionist I and Receptionist II in their performance of a variety of clerical and public contact duties that facilitate the work of the practitioners, veterinary technicians, kennel assistants and the veterinary practice manager who directly or indirectly provides patient care. Oversee the screening and assembly of veterinary records and files for active use, storage or disposal in accordance with the established records control schedules.
- Open the practice and set up for the morning as directed.
- Close the practice for the evening as directed.
- Clean and straighten the public areas of the practice including the front desk, reception area, waiting area, office, public bathroom and exam rooms.
- Welcome clients and patients to the practice with a warm and friendly demeanor and provide for their comfort while they are in the practice. This includes greeting clients, offering coffee, showing them to waiting area, etc. Maintain an up-to-date magazine selection in the waiting area.
- Answer incoming telephone calls utilizing proper telephone etiquette. Screen those calls that are handled by other healthcare team members and take care of routine calls. Routine calls include those seeking information about veterinary services. Offer to send a hospital brochure to any telephone shopper calling the hospital. Provide knowledgeable sub-professional advice concerning the care and treatment of animals including questions regarding hospital services, fees, animal care and treatment in accordance with hospital policies. Appropriately direct other questions and communication to a veterinarian, practice manager or other health care team member.
- Prepare to receive appointments by retrieving client records and preparing needed forms in advance of clients' arrival. Complete required forms such as new client form, patient visit form, client report, consent forms, estimates, payment agreements, etc and obtain all necessary information.
- Handle emergency situations by following established clinic policies and procedures in referring clients for immediate treatment of their animals when requests are

accompanied by complaints of acute symptoms. Determine nature of injury/illness and attempt to reassure distressed pet owners.

- Follow hospital policies regarding patient admittance. Determine whether immunizations/tests are current. Recommend update of necessary immunizations/tests to clients.
- Notify doctors of patient arrival. Relay all necessary information to the doctors and technicians.
- Discharge patients which includes entering all charges into the computer, reviewing the discharge instructions and medications. Ensure that future reminders are set up in the computer system for the patient.
- Present clients with medications, instructions, new client kits and any other take home items.
- Review the services that were rendered to the pet (verbally itemize the client receipt) and inform client of the total amount due. Assure that owners meet all financial obligations or that acceptable arrangements have been made.
- Accept payments from the client. Accurately process cash, checks, charge card payments and credit account payments.
- Schedule appointments for the clinic after obtaining all necessary data concerning the animal and owner. Prepare all required forms such as animal clinical records, health certificates, immunization certificates, lab reports, release forms and euthanasia certificates in advance, if possible.
- Dispense medications including providing routine instructions to owners concerning prescribed medications.
- Perform over the counter selling of pet foods and supplies. Exercise a technical knowledge of products sold.
- Assist in the updating of client/patient files as needed including name, address, telephone numbers and vaccination and heartworm history
- Retrieve and re-file medical records accurately and promptly.
- Perform an end-of-day procedure each evening. This would include reconciling invoices and balancing the cash drawer, running end-of day computer reports, preparing the bank deposit and presenting the reports and deposit information to the practice manager or owner.
- Enter data into the computer system as required. Retrieve and modify stored records. Maintain health certificate and rabies certificate files, including sending copies to appropriate government agencies. Maintain and purge medical records as directed.
- Prepare and send client correspondence such as reminder cards and letters, thank you letters, sympathy cards and welcome-to-the practice cards. Prepare miscellaneous correspondence as needed.
- Send reminder notices to clients for periodic notifications. Make recalls to clients on a timely basis from a call back list.
- Perform a back up of the computer system on a regular basis as directed.
- Perform/oversee the performance of posting daily business, posting hospital invoices, mailing statements, taking care of collection accounts, maintaining accounts receivable

file, posting accounts payable invoices, paying accounts payable, inventory control, filing posted invoices and performing related tasks.

- Work well with all team members and ensure that your actions support the hospital, the doctors, and the practice philosophy.
- Performs other duties as assigned.

CONTROLS OVER WORK

The Receptionist III works under the direct supervision of the veterinary practice manager who will indicate general assignments, limitations and priorities. Recurring assignments are performed independently. Deviations or unfamiliar situations are referred to the supervisor. Completed work is reviewed for technical accuracy and compliance with established procedures.

SKILLS AND KNOWLEDGE

- Possession of strong organizational skills.
- Excellent verbal and written communication skills. Possess exceptional interpersonal communication skills.
- Knowledge of hospital procedures and operating instructions for making appointments, assembling patient medical records, recording test results, relaying information regarding patient's condition, and compiling and submitting data on patients treated.
- Knowledge of the spelling and meaning of commonly used terminology of veterinary medicine to accurately record results of tests and file veterinary medical reports according to alpha, numeric or subject matter headings.
- Knowledge of the structure and content of the English language including the meaning and spelling of words, rules of composition, and grammar.
- Ability to work independently on assigned tasks as well as to accept direction on given assignments.
- Requires strong client service skills. Personal contacts are with animal owners affected by a variety of problems, visitors and other healthcare team members. Considerable tact and diplomacy is required. Must accurately relay owner's account of the medical complaint(s) for the animal(s) involved to the healthcare team members who will be involved in treating the patient(s).

PHYSICAL DEMANDS

The physical demands described here are representative of those that must be met by an employee to successfully perform the essential functions of this job. Reasonable accommodations may be made to enable individuals with disabilities to perform the essential functions.

Amount of Time Spent on Task

Task	None	Less than 1/3	1/3 to 2/3	More than 2/3
Stand				**X**
Walk				**X**
Sit			**X**	
Use hands to finger, handle, or feel				**X**
Climb or balance		**X**		
Stoop, kneel, crouch, or crawl		**X**		
Talk or hear				**X**
Taste or smell			**X**	

The job requires the following lifting requirements and/or exerted force be performed on the job.

Amount of Time Spent on Lifting Amounts

Lifting Amount	None	Less than 1/3	1/3 to 2/3	More than 2/3
Up to 10 pounds			**X**	
Up to 25 pounds			**X**	
Up to 50 pounds			**X**	
Up to 100 pounds		**X (with assistance)**		
More than 100 pounds		**X (with assistance)**		

Specific vision abilities required by this job include close vision, distance vision, color vision, peripheral vision, depth perception and ability to adjust to focus.

WORK ENVIRONMENT

While performing the duties of this job, the employee is exposed to hazards associated with aggressive patients; hazards associated with infected animals and controlled substances; exposure to unpleasant odors and noises; exposure to bites, scratches and animal wastes; possible exposure to contagious diseases.

ESSENTIAL FUNCTIONS:

- Professionally administer all phone calls - answering client inquiries in a prompt and friendly manner, scheduling appointments, recording messages.
- Requires strong communication and client service skills. Considerable tact and diplomacy is required. Ability to greet clients in a professional, friendly, hospitable manner - check clients in, discharge patients.
- Collect client fees, post and record payments, make change, process credit card transactions and run end of day transactions.
- Input data into computer software system.
- Open and close practice.
- Perform a variety of clerical duties, mailings, cleaning, organizing reception area, type memos, correspondence, reports and other documents.
- Ability to multi-task.
- Regular attendance and timeliness are an essential function in order to fulfill the requirements of this position.
- Perform general physical activities that require bending, standing, stooping, moving from room to room, sit, talk, and listen; may be required to walk or stand for long periods of time; will use hands to manipulate, handle, or feel; will reach with hands and arms.
- The employee must be able to occasionally / frequently lift and/or move up to 50 pounds. [Select the appropriate terms depending on the size of the practice and other team members available to assist with lifting]

– JOB DESCRIPTION –

RECEPTIONIST TEAM LEADER

INTRODUCTION

The Receptionist Team Leader trains, supervises, and assists the receptionists, plans and coordinates a variety of service functions that are related to the front desk operations and the healthcare team at the [Practice Name]. This position also encompasses all of the duties of the receptionists. These functions include, but are not limited to, reception (client and phone), maintenance of veterinary medical records, accounts maintenance, cash processing, ordering of administrative supplies and equipment, data entry and retailing of veterinary sundry items. The Receptionist Team Leader is under the direct guidance and supervision of the Practice Manager.

PRIMARY JOB RESPONSIBILITIES

- Train, supervise and assist the receptionists in their performance of a variety of administrative and public relations and client education duties which facilitate the work of the practitioners, technicians, kennel assistants, groomer(s) and the veterinary practice manager who directly or indirectly provide patient care. Ensure adherence to quality standards, deadlines, and proper procedures, correcting errors or problems.
- Oversee the screening and assembly of veterinary records and files for active use, storage or disposal in accordance with the established records control schedules.
- Oversee auditing of charts for completeness of information. Refer to the veterinary practice manager for questions concerning charges and/or treatment.
- Oversees the inventory and purchase of office supplies and forms storeroom, reorders supplies/informs the veterinary practice manager of the need for supplies to maintain pre-determined stock levels.
- Place routine service calls for maintenance of office equipment using predetermined vendors.
- Create employee schedules that align with doctor, team and surgery and outpatient needs while monitoring overtime and payroll costs.
- Provide guidance and direction to subordinates, including setting performance standards and monitoring performance. Direct on the job training. Conduct timely performance evaluations, mediate interpersonal problems and address concerns from Technicians and Exam Room Assistants. Make recommendations to the practice manager concerning personnel matters and assist with follow through of disciplinary actions.
- Conduct monthly departmental meetings and schedule in-service meetings as deemed appropriate and necessary.
- Motivate and inspire the receptionist team to provide high quality care to the patients and clients of [Practice Name].
- Work well with all team members and ensure that your actions support the hospital, the doctors, and the practice philosophy.
- Maintain effective employee-management communication. Periodically review front desk operations for efficiency and accuracy. Make recommendations to the veterinary practice manager as to ways in which the front desk can be enhanced.

- Knowledgeable regarding related federal and state animal health laws and regulations including OSHA. Ensure that the hospital and the healthcare team are in compliance with regulations. Inform the veterinary practice manager of any regulatory issues.

Receptionist Responsibilities

- Provide friendly, quality client care to the patients and clients of [Practice Name].
- Receive incoming calls, screen those that are handled by other health care team members and take care of routine calls. The routine calls include those seeking information about veterinary services ("telephone shoppers"). Provide knowledgeable sub-professional advice concerning the care and treatment of animals.
- Follow established hospital policies and procedures in referring clients for immediate treatment of their pets when requests are accompanied by complaints of acute symptoms. Determine nature of injury/illness and attempt to reassure distressed pet owners. Determine whether immunizations and/or tests are current. Recommend update of necessary immunizations and/or tests to clients when applicable.
- Schedule appointments, obtaining all necessary data concerning the patient and owner. Prepare all required forms in advance when possible.
- Prepare to receive appointments by retrieving client records, preparing needed forms in advance of clients' arrival. Complete required forms such as new client form, patient visit form, client report, consent forms, estimates, payment agreements, etc and obtain all necessary information.
- Check clients in - greet clients in a professional, friendly, hospitable manner.
- Discharge patients. Review charts of patients being discharged from the clinic for completeness of information, make new appointments or note changes in patient status as necessary. Enter charges and set up future reminders in system. Present clients with medications, instruction.
- Assure that all financial obligations are met by owners. Collect client fees, make change, process credit card transactions and assist in making count of cash drawer, run end of day transactions.
- Oversee and/or perform over-the-counter selling of specialty merchandise comprised of pet grooming aids and sundry veterinary items. Exercise technical knowledge of products sold and demonstrate salesmanship abilities. Explain and demonstrate products, answer questions concerning products purchase/use.
- Fill veterinary prescriptions with appropriate medication; provide routine instructions to owners concerning prescription for medications.
- Collect lab specimens from pet owners, match patient record to the sample and submit samples to veterinary technician or nurse.
- Assist in the updating of client files; prepare and mail thank you cards and "welcome aboard" cards, reminders. Follow-up with clients when clinic records indicate no recent visits.
- As required, enter data into the computer system, retrieve and modify computerized records. The practice management software includes, but is not limited to, such areas as reminder list of patients for periodic notifications, receipt and/or invoicing to update

file, posting accounts payable invoices, paying accounts payable, inventory control, filing posted invoices and performing related tasks.

- Work well with all team members and ensure that your actions support the hospital, the doctors, and the practice philosophy.
- Performs other duties as assigned.

CONTROLS OVER WORK

The Receptionist III works under the direct supervision of the veterinary practice manager who will indicate general assignments, limitations and priorities. Recurring assignments are performed independently. Deviations or unfamiliar situations are referred to the supervisor. Completed work is reviewed for technical accuracy and compliance with established procedures.

SKILLS AND KNOWLEDGE

- Possession of strong organizational skills.
- Excellent verbal and written communication skills. Possess exceptional interpersonal communication skills.
- Knowledge of hospital procedures and operating instructions for making appointments, assembling patient medical records, recording test results, relaying information regarding patient's condition, and compiling and submitting data on patients treated.
- Knowledge of the spelling and meaning of commonly used terminology of veterinary medicine to accurately record results of tests and file veterinary medical reports according to alpha, numeric or subject matter headings.
- Knowledge of the structure and content of the English language including the meaning and spelling of words, rules of composition, and grammar.
- Ability to work independently on assigned tasks as well as to accept direction on given assignments.
- Requires strong client service skills. Personal contacts are with animal owners affected by a variety of problems, visitors and other healthcare team members. Considerable tact and diplomacy is required. Must accurately relay owner's account of the medical complaint(s) for the animal(s) involved to the healthcare team members who will be involved in treating the patient(s).

PHYSICAL DEMANDS

The physical demands described here are representative of those that must be met by an employee to successfully perform the essential functions of this job. Reasonable accommodations may be made to enable individuals with disabilities to perform the essential functions.

Amount of Time Spent on Task

Task	None	Less than 1/3	1/3 to 2/3	More than 2/3
Stand				X
Walk				X
Sit			X	
Use hands to finger, handle, or feel				X
Climb or balance		X		
Stoop, kneel, crouch, or crawl		X		
Talk or hear				X
Taste or smell			X	

The job requires the following lifting requirements and/or exerted force be performed on the job.

Amount of Time Spent on Lifting Amounts

Lifting Amount	None	Less than 1/3	1/3 to 2/3	More than 2/3
Up to 10 pounds			X	
Up to 25 pounds			X	
Up to 50 pounds			X	
Up to 100 pounds		X (with assistance)		
More than 100 pounds		X (with assistance)		

Specific vision abilities required by this job include close vision, distance vision, color vision, peripheral vision, depth perception and ability to adjust to focus.

WORK ENVIRONMENT

While performing the duties of this job, the employee is exposed to hazards associated with aggressive patients; hazards associated with infected animals and controlled substances; exposure to unpleasant odors and noises; exposure to bites, scratches and animal wastes; possible exposure to contagious diseases.

Up to 100 pounds		**X (with assistance)**		
More than 100 pounds		**X (with assistance)**		

Specific vision abilities required by this job include close vision, distance vision, color vision, peripheral vision, depth perception and ability to adjust to focus.

WORK ENVIRONMENT

While performing the duties of this job, the employee is exposed to hazards associated with aggressive patients; hazards associated with infected animals and controlled substances; exposure to unpleasant odors and noises; exposure to bites, scratches and animal wastes; possible exposure to contagious diseases.

Note: When duties and responsibilities change, job description will be reviewed and subject to changes of business necessity

ESSENTIAL FUNCTIONS:

- Provide guidance and direction to subordinates, including setting performance standards and monitoring performance. Direct on the job training. Conduct timely performance evaluations, mediate interpersonal problems and address concerns. Make recommendations to the practice manager concerning personnel matters and assist with follow through of disciplinary actions.
- Oversee the screening and assembly of veterinary records and files for use.
- Professionally administer all phone calls - answering client inquiries in a prompt and friendly manner, scheduling appointments, recording messages.
- Requires strong communication and client service skills. Considerable tact and diplomacy is required. Ability to greet clients in a professional, friendly, hospitable manner - check clients in, discharge patients.
- Open and close practice.
- Ability to multi-task.
- Collect client fees, accurately post and record payments, make change, process credit card transactions and run end of day transactions.
- Perform a variety of clerical duties, accurately input data into computer software system, mailings, cleaning, organizing reception area, type memos, correspondence, reports and other documents.
- Regular attendance and timeliness are an essential function in order to fulfill the requirements of this position.
- The employee must be able to occasionally / frequently lift and/or move up to 50 pounds. [Select the appropriate terms depending on the size of the practice and other team members available to assist with lifting].

RECEPTIONIST
PHASED TRAINING PROGRAM

Employee (Trainee) Name _____ Hire Date _____

***Purpose**: The purpose of this program is to introduce the Receptionist to the practice and bring them into the hospital's philosophy of care and service. Through this program, the new Receptionist will become familiar with the day-to-day operations, management, and standards of care within our hospital*

Although a probable duration is stated for each phase of training, these are meant only as a guide and neither the trainer nor the trainee should sign off on a phase until they feel that they fully understand and are comfortable performing all the job tasks listed.

Phase I - Welcome to Our Practice!

Probable Duration: One Day

Skill/ Knowledge	Trainer	Description	Date Training Complete	Trainer's Initials
Parking		Show employee parking area.		
Personal Storage		Provide employee with personal storage space. Discuss protection of personal property at work		
Hospital Orientation and Tour		Orientation - Provide a detailed hospital tour which points out emergency exits, eye wash station, employee restrooms and employee break room. Identify the exam rooms, kennel, surgery/treatment area, pharmacy, radiology, etc. and what each area is used for.		
Introductions		Introduce employee to doctors and other healthcare team members. Identify trainee's immediate supervisor.		
Required Forms		**Complete Required Forms** ❑ W-2 form ❑ I-9 form ❑ Verify Social Security card & driver's license as required by I-9 ❑ Complete all required new-hire forms ❑ Other _____ *(Note: All forms are to be kept in confidential personnel file, under lock and key. All current I-9 forms should be kept in a separate file under lock and key).*		
Notebook		Give new team member an empty notebook for training notes.		

Skill/ Knowledge	Trainer	Description	Date Training Complete	Trainer's Initials
Job Description		❑ Present employee with Receptionist job description. ❑ Review general expectations for the position, as well as protocol for annual review. ❑ Present employee with a blank performance evaluation form ❑ Review the hospital's management structure (i.e. hierarchy of authority) ❑ Review the receptionist duties to be completed daily		
Hospital Procedures Manual		Present employee with hospital procedures manual. Make sure the At-Will Employment acknowledgement and acknowledgement that manual has been reviewed and read forms are signed by the employee and placed in their personnel file.		
Benefits		Review benefits and effective dates.		
Phased Training Program		Present employee with a copy of the phased training program. Explain protocol *(trainee to sign off on each phase, trainee to ask if has questions)*.		
Time clock and Employee Schedules		❑ Demonstrate operation of time clock. Explain procedure for clocking in/out. ❑ Discuss timelines and attendance expectations ❑ Show employee the proper protocol for submitting a request for days off form and how work schedules are presented and posted.		
OSHA Training		Conduct OSHA training. Explain OSHA standards, MSDS sheets, etc. Give employee handout regarding safety and complete OSHA test. Inform team member what they are to do if an OSHA officer shows up and ask for a tour of the practice. Make sure they know the practice OSHA safety officer's/coordinator's name		
Uniforms		Present team member with uniform. Review hospital dress standards.		
Observe Position		Trainee to observe (senior) receptionist. (1 hour)		

Telephone Procedures		Show proper way to: ❑ Answer phone, ❑ Take messages ❑ Place callers on hold ❑ Route messages to doctors and other team members Watch LifeLearn training CD "Enhancing your Telephone Skills." Note to practice: This CD can be purchased at www.lifelearn.com		
Basic Animal Handling		Learn basic animal handling principles. Before signing off, trainee must demonstrate proper animal handling with at least two patients.		
Conclusion of Phase I		Review of Phase I of training program. Trainee is asked if he/she has any questions or needs further training on any part of Phase I. Trainee signs off on Phase I.		

Phase I of Training Complete

My signature below signifies that I have completed Phase I of the Receptionist Phased Training Program and that I fully understand all concepts covered and I am comfortable in my knowledge and ability to perform the procedures introduced in Phase I of this program.

_____ _____

Employee (Trainee) *Date*

Phase II

Probable Duration: One Week

Skill/ Knowledge	Trainer	Description	Date Training Completed	Trainer's Initials
Reference Materials		Present trainee with materials to review. ❑ Present Trainee with the Common Medical Terminology handout. ❑ Other client education materials ❑ Other _____		
Review Materials		Review: ❑ Review above presented materials with trainee ❑ Other _____		
Scheduling		Explain: ❑ Basic appointment scheduling procedures. ❑ Scheduling guidelines and special circumstances (heartworm season, etc.). ❑ Fecal test		
Logging On/Off		Demonstrate how to log on and off the computer properly. ❑ Review company policy regarding computer use and password maintenance/usage.		
Software		Complete veterinary software training module		
Greeting Clients		Explain the proper way clients and their pets are to be greeted and treated when they come to the practice.		
Obtain client information		Review obtaining all necessary data from clients to prepare forms i.e. new clients, consent forms, medical care plans (ie, estimates).		
Obtaining a Weight		Demonstrate how to obtain a weight on a pet.		
Wait time		Demonstrate how to handle situations where there is an extended wait		
Alert Assistant About Visit		Explain outpatient protocol -- the assistant is to be alerted that the client and patient are ready.		
Controlling Odors		Explain procedure for controlling odors and maintaining a neat and tidy front desk. Discuss danger in using bleach and that bleach should NEVER be mixed with ammonia.		

Skill/ Knowledge	Trainer	Description	Date Training Completed	Trainer's Initials
Noise Pollution		Explain procedure for minimizing noise pollution. (e.g. barking dogs are escorted to a private area or an exam room) Explain proper use of ear plugs.		
Pulling Forms		Show how to retrieve forms & the filing/computer system. Before signing off, trainee must demonstrate the ability to properly handle.		
Checklist		Demonstrate how to use and/or create a checklist.		
Messages		Review the proper way to answer the phone and take messages.		
Confirmation Calls		Explain procedure of calling clients the day before their appointments to confirm their appointment.		
Surgery Quotes		Explain the proper procedure for quoting surgery prices.		
Medical Care Plan Book		Demonstrate how to use the Medical Care Plan Book and the appropriate way to go over a medical care plan.		
Fax, Copier, Phone System		Demonstrate the use of necessary office equipment.		
Mail		Explain how to take out and pick up the mail.		
Vaccination Due Dates		Explain how to check vaccination due dates. Before signing off, trainee must demonstrate the ability to handle this task properly.		
Conclusion of Phase II		Review of Phase II of training program. Trainee is asked if he or she has any questions or needs further training on any part of Phase II. Trainee signs off on Phase II.		

Trainee Comments - Phase II

Use this area for any comments you have concerning this phase of your training. This will help us to improve our training systems and ensure that adequate training is provided to you. Your comments will be read by the management of the practice and kept in your confidential employee file.

Phase II of Training Complete

My signature below signifies that I have completed Phase II of the Receptionist Phased Training Program and that I fully understand all concepts covered and I am comfortable in my knowledge and ability to perform the procedures introduced in Phase II of this program.

Employee (Trainee) *Date*

Phase III

Probable Duration: One Week

Skill/ Knowledge	Trainer	Description	Date Training Completed	Trainer's Initials
New Client Adding		Demonstrate how to add a new client.		
Entering Charges		Demonstrate the correct procedure for entering charges into the computer. Before signing off, trainee must demonstrate the ability to correctly enter charges.		
Payments from Clients		Explain the process of accepting payment from clients ❑ Credit cards ❑ Cash ❑ Check ❑ Care Credit		
Charge List		Demonstrate how to print a list of charges.		
Team Meetings		Review recent team meeting minutes and the protocol for reviewing minutes if employee is unable to attend a meeting.		
Hospital Tours		Explain protocol for client tours or when clients are allowed to visit patients in boarding or the hospital.		
Treatment Board		Demonstrate how to properly use the treatment board.		
Contagious Soak		Demonstrate the procedures followed for a contagious soak.		
Vaccine Protocol		Demonstrate a working knowledge of vaccine protocol.		
Appointment Scheduling		Demonstrate basic appointment scheduling.		
Surgery Appointment Scheduling		Demonstrate the ability to schedule surgery appointments.		
Hospital Organization		Explain the organization of the hospital and workflow.		
Surgery Forms		Demonstrate how to correctly fill out surgery forms.		
Collect laboratory specimen		Collect laboratory specimens from pet owners: ❑ Match patient record to the sample ❑ Submit the samples to veterinary technician or nurse ❑ Present clients with medications and routine instructions		

Assign Bloodwork (Outside Lab)		Demonstrate the proper way to assign bloodwork to an outside laboratory. The trainee must demonstrate the ability to handle this task properly.		
Assign Bloodwork (In-house)		Demonstrate the proper way to assign bloodwork within the practice. Before signing off, trainee must demonstrate the ability to handle this task properly.		
Outside Labs		Explain the procedure for calling outside laboratories.		
Communicate with Clients		Learn hospital guidelines for communicating with clients in different types of situations such as general queries, scheduling appointments, routine and non-routine medical questions, patient emergencies, prescription refills		
Medical Recalls		Demonstrate the procedure to follow when recalling clients. Before signing off, trainee must demonstrate the ability to handle this task properly.		
Cleaning Exam Rooms		Explain how to properly clean and disinfect an examination room.		
Boarding Slips		Show how to complete a boarding slip.		
Boarding Reservation		Explain how to make a boarding reservation.		
Cancel Boarding Reservations		Demonstrate the ability to properly cancel a boarding reservation.		
Admitting Boarders		Demonstrate the correct procedure to follow when admitting boarders. Before signing off, trainee must demonstrate the ability to handle this task properly.		
End of Life Appointments		Explain how end of life appointments are scheduled and how greeter should anticipate and prepare for these types of appointments.		
Receipting Out		Demonstrate how to check-out a client ❑ Review charts for completeness ❑ Make new appointments ❑ Note changes in patient status ❑ Enter future reminders		
Marketing		Discuss marketing to clients ❑ Discuss how to promote the practices products, programs and services. ❑ Explain the use of passive marketing ❑ Ensure that employee gains a technical knowledge of products sold		
Conclusion of Phase III		Review of Phase III of training program. Trainee is asked if he or she has any questions or needs further training on any part of Phase III. Trainee signs off on Phase III.		

Trainee Comments - Phase III

Use this area for any comments you have concerning this phase of your training. This will help us to improve our training systems and ensure that adequate training is provided to you. Your comments will be read by the management of the practice and kept in your confidential employee file.

_____

_____

_____

_____

_____

Phase III of Training Complete

My signature below signifies that I have completed Phase III of the Receptionist Phased Training Program and that I fully understand all concepts covered and I am comfortable in my knowledge and ability to perform the procedures introduced in Phase III of this program.

_____ _____

Employee (Trainee) *Date*

Phase IV

Probable Duration: One Week

Skill/ Knowledge	Trainer	Description	Date Training Completed	Trainer's Initials
Opening		Demonstrate the procedure for opening the hospital		
Closing		Demonstrate the procedure for closing the hospital		
Surgical Charges		Demonstrate how to check surgical charges. Review the travel sheet. All services rendered should be highlighted.		
Price Quotes		Explain how and when the Trainee is to quote prices.		
Client Transaction Reports		Demonstrate how to prepare a Client Transaction Report.		
Vaccination Protocol Handout		Present trainee with vaccination protocol handout and explain how to use.		
Correspondence		Demonstrate how to communicate with clients regarding medical status, medical instruction, itemize and review the client statement, inform clients about hospital policies, payment and credit policies		
Print Correspondence		Demonstrate how to print client correspondence i.e. reminders, thank you notes, new client letters		
Bank Deposits		Explain how to prepare the bank deposit and complete the deposit slip.		
Credit Cards		Explain the correct procedure to follow when batching credit cards.		
End of Day		Explain the End of Day procedures. Before signing off, trainee must demonstrate the understanding of this task.		
Returning Products		Demonstrate the correct procedure to handle returns.		
Coupons		Explain how to handle coupons.		
Bounced Checks		Explain the procedure to follow when a check bounces.		
Accounts Receivable		Explain the procedure for handling accounts receivable issues.		

Skill/ Knowledge	Trainer	Description	Date Training Completed	Trainer's Initials
After Hours ER Fee		Explain the after hour's emergency fees.		
Prescription Filing		Demonstrate how to correctly fill a prescription and the expectation that all prescriptions should be proofed.		
Controlled Substances		Demonstrate the correct procedure used when dispensing controlled substances.		
Recognizing an Emergency		Discuss referring clients for immediate treatment of their pets when the requests are accompanies by complaints of acute symptoms		
Heartworm Testing & Prevention		Explain the practice's philosophy and established protocol for heartworm testing and prevention.		
Flea Prevention 101		Explain basic flea prevention protocol.		
Client Complaints		Explain the procedure for handling client complaints.		
Displays and Retail		Explain how to restock and arrange the retail and point of purchase display areas		
Refreshment Area		Demonstrate how to restock and maintain the refreshment area		
Office Supplies		Explain the protocol for ordering inventory and office supplies		
Clean Front Area		Demonstrate how the front desk and printer should be cleaned.		
When In Doubt		Assure the employee that whenever he or she is in doubt or needs help, they are expected to seek assistance and guidance.		
Conclusion of Phase IV		Review of Phase IV of training program. Trainee is asked if he or she has any questions or needs further training on any part of Phase IV. Trainee signs off on Phase IV.		

Trainee Comments - Phase IV

Use this area for any comments you have concerning this phase of your training. This will help us to improve our training systems and ensure that adequate training is provided to you. Your comments will be read by the management of the practice and kept in your confidential employee file.

_____

_____

Phase IV of Training Complete

My signature below signifies that I have completed Phase IV of the Receptionist Phased Training Program and that I fully understand all concepts covered and I am comfortable in my knowledge and ability to perform the procedures introduced in Phase IV of this program.

_____ _____

Employee (Trainee) *Date*

THE 60 DAY NEW EMPLOYEE SURVEY

One of the best sources of knowledge and innovation is new team members. We want to know what you've learned about our practice and how you think it can be improved. Please use extra paper where needed.

1. Background

Name: _____ Position Title: _____

Date of Hire: _____ Name of Current Department _____

2. Job Description

Please describe in your own words the three most important things you do in your job:

1. _____
2. _____
3. _____

Do you feel that your job title is properly named: Yes No *(Circle One)*

If "No," what should it be? _____

_____

Name the three ***most*** enjoyable aspects of your job:

1. _____
2. _____
3. _____

Name the three ***least*** enjoyable aspects of your job:

1. _____
2. _____
3. _____

3. Hiring Process

Tell us about the hiring process.

A. The job described to me was an accurate assessment of the job.

Yes No *(Circle One)*

If "no," what was ***not*** accurate: _____

_____

B. The time it took from submitting my application to being hired was reasonable.

Circle all that apply:

a. The time it took was too long.
b. The hiring process was confusing, didn't know how long the process would take.
c. I agree. The time was reasonable.
d. If I could change one thing about the hiring process, it would be:

_____

C. What improvements can be made in the practice's hiring process so that we can hire better team members?

_____

_____

D. What improvements can be made in the practice's hiring process so that we can hire better team members?

_____

_____

4. What did you like best about the training?

_____

_____

How can the practice improve the orientation process which introduces new team members to the practice's operations, personnel, products and services?

_____

_____

What can the practice do to provide you with skills training so that you can excel at your job?

_____

_____

Would you be interested in cross training in another position?

Yes No *(Circle One)*

If so, please state job position you would be interested in training for: _____

_____

How would you rate the quality of the training you have received?

1 2 3 4 5 6 7 8 9 10

Poor Fair Excellent

(Circle One)

Comments: _____

_____

5. Wage and Hour Issues

Are you unclear about any wage or hour issues (pay, overtime, vacation, missed time from work, etc.)?

Yes No *(Circle One)*

If so, please indicate any questions that you have.

_____

Did you have your wage and how raises are calculated explained to you?

Yes No *(Circle One)*

6. Practice Policies and Procedures

Are you unclear about any practice policies or procedures as set forth in the employee manual or by your supervisor? Yes No *(Circle One)*

If so, please indicate any questions you may have: _____

_____

7. Comments

What advice would you give to new employees about this practice?

_____

_____

If you are aware of any possible improvements to the way we run our practice, please give us your comments or suggestions:

_____

_____

Thank you!

Signature: _____ Date:

Reviewed with team member by: _____ Date: _____

Mark Opperman, Sheila Grosdidier

How to Make Social Media Your Friend Instead of Your Enemy

Veterinary Management Consultation, Inc, Evergreen, Colorado, USA

Make Friends With Social Media

Sheila Grosdidier, BS, RVT, MCP, PHR

Is it possible to have a hardware headache? To breakout in social media hives every time someone says Facebook? Or get a paralyzing vision of Alfred Hitchcock's The Birds *when the word Twitter is spoken?*

The realm of what is available on the Internet and through technology can be overwhelming. Successful practices know that today's business environment calls for utilizing every expense dollar for maximum return and growing revenue with limited budgets. The practice of increasing practice development while decreasing the marketing budget has become the norm.

However, when practices hesitate to purchase technology to integrate into their businesses or embrace the latest in social media, it affects the full development of the practice and ultimately influences the value of the business over time.

Here are 12 ways to leverage each aspect of technology available in today's world. Identify which ones you are currently utilizing and those you should be integrating into your practice.

1. **Websites**

   Most practices have a website, but the questions are:

   - Does it reflect who you are?
   - Is it updated?
   - Do you create a reason for clients and potential clients to return?

   Chances are you spent a significant amount on the sign in front of your practice. Now, consider that more potential clients go by your website than drive by your practice each day. The Bayer/Brakke Study identified that over 37% of owners use the Internet to evaluate veterinarians prior to their visit.[1] Remember, keeping your website information current (**Table 1**) is one of the ways that search engines rank websites (see **Google Analytics**).

2. **Email**

   While many marketing budgets have been drastically cut in small businesses, email marketing is one area that has actually increased from 2009 to date.[2] The reason is obvious: because it works. Clients want to hear from you; they seek information to make decisions about the needs of their pets.

   Most veterinary software programs have email address fields. What percentage of your active client base has an email address entered? Set a goal with the practice team to begin collecting client email addresses when clients come in for an appointment. Your goal should be to exceed 80%. When asking for an address, remind clients that you want to save paper as well as send them important information quickly (**Table 2**).

3. **Facebook**
"Small businesses that use online technology grow twice as quickly, bring in twice the revenue, and hire twice the employees as those small businesses that do not," according to David Fischer, vice president of Facebook.[3] While small businesses have not engaged in social media as fervently as large companies, more emphasis is being placed on helping small businesses take advantage of getting their message out to potential clients.

Facebook enables you to create a community of clients and potential clients, to impart information, and to open up a dialogue with pet owners. In January 2012, Facebook will launch a program that provides up to 10 million dollars in free advertising online to small businesses (go to facebook.com/marketing?sk=app_244881505558365).[4] See **Table 3** to view the benefits of Facebook according to other businesses and **Table 4** for 5 things you need on your Facebook Page.

4. **LinkedIn**
Seventy million users know that LinkedIn is more than just a site to post a resume and look for a job. It is an ideal site to make contacts with experts in a variety of fields. Looking for information on a marketing consultant? Need to know more about a public official? Want to know if there's a potential new associate looking for work in your area? Use this tool to emphasize your experience, knowledge, and education. Your clients are checking for you there.

5. **Twitter**
Is it possible to build your business 140 characters at a time? Twitter gives you the opportunity to create interest, supply information, and implement an effective marketing strategy. Want to share pet information, remind users about a pet charity event coming up, or impart your thoughts on that latest animal issue in your area? This is the tool to get you out there.

Many clinics re-tweet (take information that they have received from other tweeters) and send it to their user list. Sending these ultra short messages takes some creativity and needs some thought, but it's worth it. Make a list of tweets you want to send in a month as you find them from other sources, such as websites, blogs, books, or other tweeters. Then, when you want to send one out, you have it ready. Check out twitter.com/#!/FindAVet or wefollow.com/twitter/veterinary.

6. **Google Analytics & HootSuite**
Each day, we are committed to documenting and evaluating the progression of health issues and successful treatment of pets. Google Analytics (google.com/analytics) is a free program that assesses the metrics of your web presence. It will provide information about your social presence, such as the amount of website traffic, who is returning, and whether you are bringing in potential clients (**Table 5**). HootSuite is a social media dashboard that tracks all social media sites (Facebook, Twitter, LinkedIn, blogs, etc) from one program.

7. Texting

Eighty-seven percent of texts are read within 30 minutes of delivery[5] so it's never been so easy to maintain close contact with clients.

- Ask clients for permission to text them, as some plans charge extra for texting.
- Designate a cell phone for the practice that allows clients with patients in the hospital to directly call or text that hotline with questions about their pets.
- A veterinary technician on your team can cover the calls to provide expedited service to clients with the most medically fragile pets.
- You can text your clients pictures of their pets in recovery or when they are ready to go home, reinforcing to clients how much you care about their pets and peace of mind.

In addition, many veterinary software programs can verify approval for texting with a client and several companies can set up texting for appointment reminders and other pertinent information.

8. Blogs

Blogs provide information and content that is updated on a fairly regular (sometimes daily) basis. It gives you the ability to keep your name at the forefront with both clients and potential clients, share a more intimate view of the practice, discuss issues in veterinary medicine, and build a relationship with your clients (**Table 6**). Blogs are also an excellent tool to introduce new diagnostics and procedures, reassuring clients that their pets are receiving top-of-the-line care.

9. iPad

The iPad has rocketed up the "must have" list of technology items, not only for leisure time but for savvy, fast moving businesses as well. Why should you consider it for your practice? Its mobility, depth of applications, and ease of use combined easily provide the needed return on investment.[6]

The less than 2-pound iPad has 10 hours of battery life and thousands of applications, including the ability to calculate blood transfusion rates and volumes, let you know a client just sent an email, and type while you talk. See **Table 7** for the top 15 iPad business applications.

10. Veterinary Management Software

Ask any of the major software creators and they will tell you the biggest challenge they face is that their software features are underused. Having a veterinary software program and merely using it for invoicing and basic inventory function is like having a Ferrari and never going over 30 MPH. Review the list of amazing things your veterinary management software should do for your practice in **Table 8**.

11. Online Reputation

Like the majority of Internet users, you most likely peruse business or product reviews to determine who receives your business.[7] The question is—who is writing these reviews? For decades, veterinary practices have based client growth on personal recommendation and many of those recommendations have migrated to the Internet. It is essential to track reviews of your practice quickly and efficiently to ensure you good reputation does not suffer (**Table 9**). However, now Internet companies have begun to offer services to "control" the online review process<Author: What does this mean?>.

12. Mobile Devices

Humans have become super computer beings with the power of their smart phones. At the simplest, smart phones combine your phone, voicemail, email, address book, appointment calendar, and web browser into one device; however, you can also access information from a veterinary database, calculate a drug dose, send a radiograph for review, instantly know what's happening on your social media sites, or receive a reminder that an airline ticket has been reduced in price. There are literally 1000s of applications available to you (**Table 10**).

Now that we've reviewed the various aspects that make up modern day technology for small businesses, in a future article we'll explore how to prepare a plan that engages technology in your practice. This plan will focus on making good financial decisions; getting the best returns on your technology investments; and harnessing the potential of what the future holds in communication, organization, and business development.

Table 1: ***Six Ways to Improve Your Website Now***

1. Do the clutter test:	Can you navigate easily around your website? Users must be able to find what they need easily and quickly or they will give up.
2. Position yourself as a credible resource:	A potential client needs to feel confident in you and your staff's abilities in order to visit your practice. Communicate to website visitors what sets your practice apart from other practices.
3. Make it personal:	Add client testimonials, comments, and pictures. Introduce yourself and your practice team and what you each specialize in.
4. It's time to go visual:	Include short video clips on pet care, pictures of the staff and practice, and handouts on important health information.
5. Tell your story:	Share success stories of pets that are living better lives because of the treatment they received at your practice. Describe what your practice does for charity. Include interviews with staff members.
6. Go virtual:	Present a virtual tour of your practice and the procedures that take place, or provide an invite to come visit and tour the practice.

Table 2: ***Improve Your Email Contact with Clients***

Fine Tuning Your Emails	Why & How To Do It
Have a clever and personalized subject line.	This avoids ending up in the spam folder and encourages opening the message.
Make reading an email feel like time well spent.	Email isn't free—it costs your clients time to read it. Dial up the value and dial down the selling .
Allow clients to opt out of receiving emails.	They may love you, but may not have the time right now to receive additional email.
Keep emails short and to the point.	Provide key points and concise details with links to your website or Facebook page for further information.
Don't be afraid to try something fun.	How about a pet picture or story contest? Or a charity fundraiser at the clinic. These activities encourage interaction and build relationships.
Utilize surveys and offer a prize, such as a discount on a veterinary service, to a winning respondent.	Online surveys are readily available and make it easy to obtain essential feedback: Ask clients their likes and dislikes about your services. Find out if you are exceeding their expectations or have some work to do in that area.

Table 3: ***Why Businesses Have a Facebook Presence***

85%	Share basic information about the business
62.4%	Share content, such as videos and pictures
46%	Have conversations with customers
27.2%	Customer support
23%	Contests

Results from Buzzom.com's survey, Infographic: Why Small Businesses Should Have a Facebook Page[3]

Table 4: ***Your Facebook Page - Three Critical Points***

1. **Have it everywhere:** Include the link to your Facebook page on all electronic and hard copy communication.
2. **Consider a unique name:** Naming your page Your Pets Vet is easier to remember than Binder River Valley Veterinary Center and Boarding.

3. **Consistency is key:** Develop a plan of what information you want to post and how often you would like to do so - don't let it stagnate!

Table 5: ***Google Analytics***

What You Can Track

- How many clicks on a page (what's getting the most attention)
- Users who find you through a search engine
- The amount of time users surf your site

Additional Applications

- Alerts sent directly to your phone or email
- Ability to set up client surveys

Table 6: ***Eight Ways to Make Your Blog Great***

1. **Determine your goal.**	Your blog should have a focus that is easily recognized by the reader.
2. **Make it visually appealing.**	You only have once chance at a first impression. Present a professional, attractive blog that encourages users to return often.
3. **Make it easy to read.**	Write in a way that is easy to follow and use a font that is easy to read.
4. **Make it interesting.**	Use pictures coupled with good pet care information and fun/inspiring stories (ala James Harriott).
5. **Make it searchable.**	Decide upon a meta-title to make it easy for a user to easily find your blog.
6. **Tag it.**	Another way to find your blog is through index words. Be sure to identify them so users can find you.
7. **Attract traffic.**	Include the URL as part of the practice's website to allow both websites to come up when the practice name is searched.
8. **Don't go it alone.**	A team of people can rotate updating the blog to keep it fresh and take the pressure off a single person.

Table 7: ***Twelve Business Applications for Your iPad***

1. **Square Up** https://squareup.com	Take credit card payments from any place, at anytime.
2. **MightyMeeting** mightymeeting.com	Store demo videos and presentations to share in online meetings from the web, tablet, or smart phone.
3. **iDisplay** www.shapeservices.com	Turn your iPad into a second monitor for your computer.
4. **Drop Box** dropbox.com/ipad	File all your photos, documents, and videos in an online folder for 24/7 access.
5. **Dragon Dictation** nuancemobilelife.com	Using your voice, you can text or email, update Facebook, send notes and reminders to yourself, or tweet to the world.
6. **iThoughtsHD** ithoughtshd.com	Create mind maps for your ideas or plans.
7. **Evernote** evernote.com	Take notes, create to-do lists, search through images, and share your memories
8. **Google Analytics** www.google.com/analytics/index.html	Web analytics solution that provides insight into website traffic and marketing effectiveness.
9. **Whiteboard** http://itunes.apple.com/us/app/whiteboard	Provides the ideal environment for writing notes, sketching charts, and recording brainstorming sessions
10. **Veterinary Differential Diagnosis Application** ellenstechnologies.com	This app systematically walks you through a differential diagnosis—from clinical signs to possible causes to a possible diagnosis.
11. **dvm360 Application** dvm360.com/ipad	News, medicine, business, team training…all in one place
12. **DVM Calc Application** http://itunes.apple.com/us/app/dvm-calc	Contains 26 practical, 15 constant rate infusion, and 10 toxicity calculators.

Table 8: ***Management Software - What Else It Should Do for You***

1. Allow you to access your database from a mobile application.
2. Target specific groups of clients for marketing, updates on medications, changes in medical management communication.
3. Capture pictures to track medical cases, add personal touches to communication, and document progression of disease (such as a picture of the mouth that shows how dental disease has progressed over time).
4. Provide access to training that is timely, cost effective, and meets the needs of all team members.
5. Have an online community that allows you to easily interact with other users to ask questions, get ideas, and share files.
6. Enable technology to work in harmony (laboratory equipment, online test results, accounting database) and provide access for clients to check on their pets' preventive health care services and products.
7. Enhance practice workflow while avoiding obstacles or a hindrances.
8. Provide valuable data to make solid business decisions.
9. Allow for individuality and leverage what is unique to your business.
10. Adapt to the changing world of veterinary medicine, practice needs, client service, and patient care.

Table 9: ***Monitor Your Online Presence***

Google Alerts googlealerts.com	Sets up an alert that emails you anytime your business name, personal name, or selected topic(s) are posted to the Internet. (Reviews, video, social media, websites, etc. can all be tracked.)
Technorati technorati.com	Tracks all blog postings for key words you identify. Want to know if someone has posted a link to your site or is mentioning you in a blog or discussion? This is your tool.
TweetDeck tweetdeck.com	Even if you aren't a tweeter, this service will let you know what's being said about you on Twitter.
BoardTracker boardtracker.com	Follow any mention of your name or business on discussion boards or forums.
Social Mention socialmention.com or **Topsy** topsy.com	This service tracks you or your business in a broad variety of social media—Facebook, Twitter, Digg, Google +, etc.

Table 10: ***Mobile Applications You Have to Try***

Apple (iPhone)	**Android**
Quickoffice: Update Microsoft Word or Excel files	**Google Voice**: Want visual voice mail, voice mail turned into text, or free conference calling? Here you go.
Iterminal: Accept credit card payments from wherever you are	**Intuit GoPayment**: Accept credit cards anywhere
Dropbox: Access files from your computer on your phone	**Dropbox**: Access files from your computer on your phone
Evernote: Store files, video, and pictures online for access anywhere	**Evernote**: Store files, video, and pictures online and access anywhere
Find more at itunes.com	*Find more at androidapps.com*

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Mark Opperman, Sheila Grosdidier

How to Deterimine Your Client Fees: If you and your team are comfortable with your fee schedule, everyone will be more compliant.

Veterinary Management Consultation, Inc, Evergreen, Colorado, USA

How To Determine Your Client Fees Fairly & Accurately

I. "Shopped" and "Exposed" Services

II. Calculate Your Fees Based On:

A. Overhead costs per minute

B. Direct costs at percent (%) mark-up

C. Return on time to the doctor

III. In Hospital Service Fees - <u>Overhead costs per minute</u>

A. Calculate overhead costs/minute/DVM

B. Formula: ALL expenses – compensation to doctors (both owners & associates) including related costs – inventory costs ÷ number of hours the doctors are scheduled (both office hours & surgery) ÷ 60

IV. In Hospital Fees - <u>Direct costs</u>

A. Direct costs definition: Inventory costs, costs of materials used in the procedure

B. Formula: 2x cost due to costs of ordering

IV. In Hospital Fees - <u>Return on time to the Doctor</u>

A. Return on time to the Doctor definition: Per minute cost of the doctor's time spent on a procedure

B. Formula: Cost of DVM's time/minute x # of minutes spent on procedure

C. In hospital procedures

D Surgery

1. General/Soft Tissue

2. Orthopedic

V. How to Determine Pharmacy Fees

A. Set % mark-up
B. Minimum per pill charge
C. Pharmacy preparation charge
D. Minimum Rx charge

VI. Outside Laboratory

A. 2 x cost plus $5.00, or
B. 2.5 x cost

Equinos

Tim Greet

Surgical Philosophy

Rossdales Equine Hospital, Newmarket - UK

Surgical "philosophy"

- all aspects surrounding surgery
- accurate diagnosis
- realistic evaluation of prognosis
- honest client communication
- facilities for general anaesthesia or standing sedation and surgery
- expertise of surgeon and team
- facilities for post op management

Is the patient a suitable candidate for surgical treatment?

Potential surgical candidates

- has an accurate diagnosis been made?
- have all reasonable options been discussed with the client? ie informed consent - this can sometimes be difficult if one is working through a third party
- if the horse is insured have insurers been informed (including of a ga if required)?
- where is the procedure to be performed and by whom?

Surgical scenarios

- emergency potentially life-saving surgery
- corrective surgery following accurate diagnosis
- surgery as part of diagnostic process
- more speculative surgery but with informed consent of owner (ship going down with all guns blazing??)

surgical urgency

- many patients can be considered as genuine surgical emergencies (eg most colic, synovial sepsis and some fracture patients)
- in others longer evaluation time may afford a more accurate pre-op. diagnosis or prognosis
- in some cases there may be owner or horse trainer pressure for prompt action (eg stage of season or insurance aspects?)
- priorities must be welfare considerations and providing enough information for client to allow informed consent

Some surgical procedures are by definition carried out in order to enhance the diagnosis

- exploratory laparotomy or laparoscopy
- diagnostic arthroscopy, tenoscopy, or bursoscopy
- exploratory paranasal sinus surgery
- exploratory surgery on a possible cryptorchid
- clearly there are other methods of investigating these types of patient
- exploratory surgery may offer the possibility of treatment

Ancillary considerations before deciding to operate on a horse

- presumption that the rationale for the procedure has been fully resolved
- on standing patient or under ga?
- presumption that informed consent has been obtained from all relevant parties
- surgical plan a in place (plus alternate plans b and c)
- surgical competence and facilities (will be discussed later)

Surgery under a ga

- provided facilities are good - an effective way to restrain the patient for most procedures
- a controlled surgical environment
- necessary for many procedures
- less effective for others
- 1.6% risk of death within 7d (johnston et al 2005) cf human fatalities ~0.0001%

Current surgical repertoire in standing equine patients

- castration (open)
- wound repair
- paranasal sinus surgery
- tooth removal
- facial fracture repair
- eye enucleation
- upper airway laser surgery
- thoracic surgery
- laparoscopy (laparotomy)
- umbilical hernia repair
- urogenital surgery
- ligament and tendon surgery
- resection of dsps
- foot surgery
- synovial lavage (arthroscopy!)
- limb fracture repair

Reasons for operating on standing patients

- avoid the risks of a ga
- more effective sedative agents available
- more convenient for some procedures
- reduction in surgical time (some procedures)
- reduction in haemorrhage (nasal and sinus surgery)
- better access (ovariectomy)

The correct approach to surgery

- there is no such thing as one way to do a procedure
- there may be an infinitely preferable way of doing something eg endoscopic rather than open surgery
- do not succumb to "fashion"
- stick within your comfort zone

Basic decisions - synovial surgery

- endoscopic approach almost always indicated
- lateral or dorsal recumbency?
- esmarch and tourniquet?
- additional imaging required (fluoro or radiography)?

Basic decisions - fracture repair

- nature of injury eg open or closed fracture, comminuted etc
- age, size and temperament of patient?
- future function and client expectation?
- screws?
- dcp or locking plate?
- standing or under a ga

Repair of limb fractures in standing patients

- non-displaced lateral condylar fractures of mc/ mt 3
- medial parasagittal fractures of mc/mt 3
- proximal phalangeal fractures
- nowadays we repair most of our cannon and pastern fractures by this method

Basic decisions - colic

- is the owner contactable during the procedure?
- survival at all costs or judgment-based euthanasia if complicated?
- routine postoperative management or massive support involving expensive medication?
- are there resources to ensure effective postoperative care?

Things to remember

- decompress intestine whenever possible
- if in doubt cut it out (necrotic intestine)
- perform end-to-end anastomosis of small intestine wherever possible
- foal mesentery is very friable
- often the difference between life and death depends heavily upon the postoperative support team
- prompt interference always affords the best prognosis

Requirements for successful laparoscopy

- equipment
- technique and experience
- patient temperament (standing)
- patient size (no?)
- empty gi tract
- appropriate reason for the use of a laparoscopic approach

Procedures amenable to laparoscopy

- gonadectomy and in particular ovariectomy has been revolutionised
- adhesiolysis (only exceptional cases)
- visceral biopsy
- major gastrointestinal surgery not possible

Surgical preparation

- starvation ~48 hrs (empty gi tract)
- manual examination per rectum (preferably by surgeon)
- pre-op medication (acepromazine 30 mins prior)
- quiet environment
- stocks and non-slip floor
- detomidine and butorphanol bolus plus detomidine drip (12.5mg in 0.5L 0.9% normal saline)
- morphine if difficult patient or xylazine "top up" if ataxic
- analgesia - flunixin meglumine

Basic decisions - upper airway

- has a reliable diagnosis been reached?
- if sinus surgery- have the teeth been adequately evaluated?
- if treating an abnormal respiratory noise at exercise- has dynamic endoscopy been carried out?

Lessons learned

- careful thorough desensitisation of larynx and nasopharynx before standing surgery (esp. when using a hook knife)
- hook knives are sharp (use guarded blade)
- less is more in laser surgery
- careful handling of pharynx during laryngoplasty reduces postoperative coughing
- there are few lasting "cures" for horses with dynamic upper airway problems

Basic decisions - sinus surgery

- standing or under ga
- most standing as good access and reduced haemorrhage
- dental involvement? (oral endoscopy; gamma scintigraphy, ct or convincing radiological evidence?)
- simple approach with topical local or maxillary block?

Modern imaging

- has facilitated the more difficult diagnoses
- ct offers fantastic images of sinus structure and dental apices
- oral endoscopy allows comprehensive examination of the whole mouth
- scintigraphy helps to identify dental problems if no structural change detected

Prepare for the unexpected

- always try to anticipate likely problems
- a flexible approach and alternative plans are necessary
- always warn owners carefully of potential complications
- it is better that a client takes a patient home than that expectations exceed the likelihood of delivering a satisfactory result

Should I operate on this patient?

Surgery is part of my skill set?

- until the development of the ECVS surgical training in the UK was on an entirely ad hoc basis
- the majority of equine surgery was carried out by vets with no specific surgical training
- "I operate on dogs and cows; horses are surely no different"
- "fancy referral veterinary clinics would charge my client an arm and a leg!"

Considerations about operating on a patient

- am I competent to do the job?
- is the condition an emergency?
- can the horse be moved safely?
- how close is the nearest referral centre?
- what facilities do I have available?
- client's wishes?
- value of animal?
- insurance aspects?

Emergencies which cannot travel or be referred

- the rcvs specifies that we should only work within our own competence level
- it is invidious to speculate on the competence level of individuals
- genuine emergencies like gi catastrophes or emergency c. sections are sometimes better tackled urgently than referred
- increasingly litigious horse-ownership, but some clients cannot afford referral fees and are very happy to accept the first opinion vet's surgical expertise and experience level if explained honestly

Some emergencies can be travelled safely

- colic patients which are stable
- fracture patients in which the fractured limb has been adequately supported
- will depend largely on clients wishes and local veterinary expertise or experience
- many equine surgical procedures can be tackled very effectively with minimal equipment and facilities
- some emergencies (colic, c.section, some fractures, synovial sepsis)
- some elective orthopaedics (check lig and ann lig des; splint removal)
- head and neck surgery (sinuses; some throats)
- wounds

Facilities

- safe anaesthesia (including recovery)
- operating theatre
- equipment
- personnel

Safe facilities for standing surgery

- a quiet area outside the busy areas of the hospital is the best place
- good non-slip flooring
- good lighting and drainage
- "adaptable" stocks
- head restraint
- overhead electric sockets
- screen
- equipment and drugs near at hand
- scrub facilities
- security of area (laser surgery)

Top quality support staff

- good facilities are all very well ..... but
- good well-trained personnel are more important
- equine veterinary nurses and other support staff are essential to developing surgery in practice and improving the service provided

Preparation of the patient

- an accurate diagnosis
- an honest prognosis
- informed consent (client ± insurers)
- appropriate medication
- pre-anaesthetic check
- mouth wash and feet wrap (shoes?)
- jugular catheter inserted aseptically
- clip and prepare surgical site

Perioperative medication

- pre-anaesthetic sedation
- antibiotics (depends on procedure)
- nsaids (in foals + gastric protectant)

Perioperative antibiotics

- nature of the procedure, temperament of patient?
- racehorse - doping?
- implants (plates, screws, tie-back or -forward, hernia mesh)
- enterotomy or enterectomy
- synovial sepsis or bone infection
- clean orthopaedic (arthroscopy, check ligament)
- standing hook knife or laser throat surgery

General anaesthesia

- endotracheal tube and inhalational anaesthesia
- monitoring (expertise of personnel?)
- ventilator?
- fluids and catheterisation
- recovery

Surgery in standing patient

- commonly performed these days
- control of patient is fundamental
- some patients unsuitable
- training of support staff is critical
- careful case selection

Scrub-up and patient prep

- trolley preparation in advance
- do as much as you can with the patient standing
- patient and surgeon scrub should be simultaneous
- theatre etiquette is necessary no matter in what kind of theatre!

Aseptic technique

- philosophy of "aseptic technique" = we are taking every care of the patient
- basic rules to be followed under all circumstances
- trained theatre staff are vital in policing this
- when designing an operating theatre thought needs to be given to the flow of people and horses and to the "clean - sterile - dirty" cycle

"Four section system"- traffic light code

- anaesthetic area
- service area (prep area to allow support of anaesthesia, surgery, lab kit and office)
- theatres
- cleaning and sterilising and changing room area

What if the patient represents a risk?

- Streptococcus equi equi
- Salmonella sp
- Clostridium difficile
- avoid patient if not emergency surgery
- if emergency surgery essential hospital must have a biosecurity policy
- if horse must be operated it is vital to have appropriate biosecurity management measures in place

Colic surgery

- does not require particularly high tech equipment or facilities
- surgical familiarity with the abdomen is very helpful!
- equally important is a team to look after the patient postoperatively

Caesarean section

- always try cvd first
- c. section can be performed with very limited facilities
- incise over identifiable limb
- haemorrhage often profuse
- crush suture of uterus
- placental removal?

Orthopaedic surgery

- angular and flexural deformities
- annular ligament desmotomy
- splint removal
- simple fracture repair
- septic synovitis

Septic synovitis

- "wound awareness"
- always treat as urgent
- synoviocentesis and synovial distension
- radiography (especially foals)
- endoscopic (ingress/egress lavage?)
- monitor by synoviocentesis
- beware the draining synovial cavity

Standing facial flap surgery

- no high tech facilities (stocks useful)
- small oscillating saw helpful
- some horses may resent the noise (ear plugs)
- block skin
- remove bone flap?
- reduced haemorrhage
- can operate on almost all types of sinus lesion

Postoperative management

- intensive care for colic patients
- round the clock monitoring?
- regular checking of iv catheters and heparin saline lavage
- careful monitoring of supportive casts and bandages
- careful monitoring of horses with synovial sepsis
- neonatal intensive care is the most expensive to deliver

Don't forget

- check postoperative faecal production
- if in doubt tube with electrolytes until normal output
- tpr and observation of horse's behaviour (urine production)
- routine monitoring of feet

Take home messages

- there is no mystique to equine surgery
- knowledge of anatomy and basic common sense will effectively underpin many of the commonly performed procedures
- cadaver surgery is really useful practice (remember the apocryphal story about Gary Player and bunkers)
- good nursing and support staff will make up for lack of sophisticated facilities
- keep the client well-informed
- never be afraid to ask or to refer
- know your limitations!

Andy Durham

A clinical approach to liver disease in the horse

BSc.BVSc.CertEP.DEIM.DipECEIM.MRCVS,
RCVS and European Specialist in Equine Internal Medicine

The clinical investigation of horses suspected to have liver disease essentially comprises 4 components: clinical examination; examination of blood samples; diagnostic ultrasonography; and liver biopsy
These 4 components have their strengths and weaknesses of which the practitioner should be aware and will be discussed further below.

Clinical examination

There are three fundamental aims of the investigation of cases of suspected liver disease: firstly, to differentiate subjects genuinely suffering from liver disease from those which are not; secondly, to determine the type of liver disease affecting the subject; and thirdly, to differentiate those subjects which are likely to survive from those which are not.

Many clinical signs are widely recognised to be associated with liver disease in horses (table 1, figure 1). However, the fundamental distinction between liver disease and liver failure (or insufficiency) is an important basic principle in this respect. Many cases of liver disease are subclinical and it has been suggested that at least 60% of liver function must be lost before there is functional liver failure and consequent precipitation of clinical signs1 implying that liver disease is of much greater prevalence than liver failure. This has two important consequences which affect the diagnostic and prognostic value of clinical signs in suspected hepatopathy cases. Firstly, clinically normal horses may be suffering from liver disease; secondly, the presence of any overt signs of hepatic dysfunction implies a considerable degree of liver pathology which may therefore have prognostic relevance. Amongst a group of horses investigated for liver disease only 17/61 (28%) horses with biopsy confirmed liver disease were showing typical clinical signs2. However, when clinical signs of liver disease were present, only about 50% of these cases survived3.

- depression
- weight loss
- anorexia
- abdominal pain
- photosensitisation
- diarrhoea
- neurologic dysfunction*
- jaundice
- oedema
- pruritus
- coagulopathy (epistaxis)
- polydipsia/polyuria
- coronitis

Table 1. Clinical signs associated with failure of the liver to perform its usual physiologic functions (*marked depression, apparent confusion and disorientation, ataxia, blindness, headpressing, circling, persistent yawning, bilateral laryngeal paralysis, foot stamping and apparently motiveless wandering).

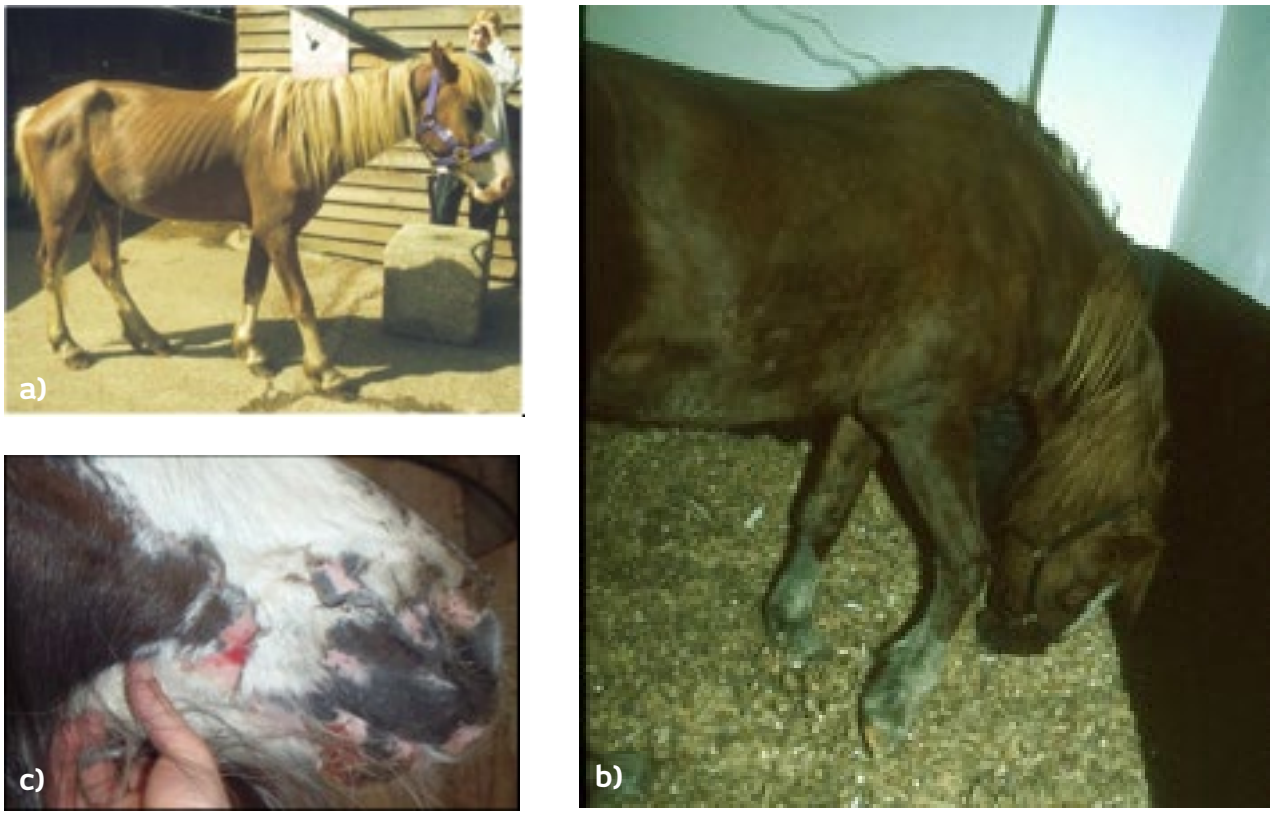

Figure 1. Images showing a) weight loss; b) severe neurologic disturbance (hepatic encephalopathy); and c) photosensitisation in horses with hepatic insufficiency.

When faced with an apparently clinically healthy horse with serum biochemical evidence of hepatic disease a dilemma arises whether or not to pursue investigation and treatment at that time. However, not to do so risks further progression and deterioration of the subclinical disease to a point where prognosis is worse and treatment is more difficult. It is this author's experience that liver disease is most commonly seen as an "outbreak" in horses. However, the recognition of such outbreaks is hampered by the subclinical nature of many cases as described above. When a case of liver disease (subclinical or clinical) is identified in an individual horse then it is good practice to perform more widespread testing of herdmates as, in most instances, many more horses are seen with a subclinical affliction.

Blood testing in liver disease

Given the relatively poor prognosis attributed to cases of clinically overt liver failure there may be considerable advantages in the identification and treatment of subclinical hepatopathy cases. The most common initial diagnostic method in subclinical hepatopathy cases is analysis of blood samples. Biochemical substances measured in the blood of suspected equine hepatopathy cases can be subdivided into substances reflecting damage to liver cells and substances reflecting impaired liver function. Intuitively, elevated serum concentrations of liver-derived enzymes (e.g. AST, GGT, AP, GLDH, SDH) arise primarily from damaged liver cells which may or may not recover (population 2, figure 2). Such analytes might therefore be reflective of ongoing hepatic insult but bear no direct relation to the remaining mass of healthy liver cells (population 1, figure 2) which serve to maintain hepatic function. It is therefore not surprising that previous investigations have not consistently found a strong association between liver-derived enzymes and severity of liver failure[1,3,4] (table 2).

Analyte	survivors		non-survivors		P
	n	mean value	n	mean value	
globulins (g/L)	70	34.6	23	47	<0.001
albumin (g/L)	70	34.7	24	30.7	<0.001
bile acids (umol/L) ((u((mmol/l)	46	10.3	15	31.4	0.001
GGT (iu/L)	76	175	23	403	0.001
AP (iu/L)	60	460	20	922	0.004

Table 2. Comparison of serum biochemical data in horses with liver disease found to be significantly different between surviving or not surviving cases for 6 months post diagnosis[3].

There are several biochemical analytes which might theoretically be more closely associated with the remaining mass of functional liver cells (population 1, figure 2) and therefore better reflect remaining hepatic function. These are primarily endogenous substances which accumulate in the blood as a result of failure of extractive and processing functions normally performed by the liver and other substances whose serum concentrations are normally maintained by hepatic synthesis. These include bilirubin, bile acids, clotting factors, clotting times (PT and APTT), albumin, globulin, fibrinogen, NH3, urea, glucose and various amino acids. Abnormalities of these functional parameters might therefore be more useful in the differentiation of hepatic failure from cases of adequately compensated hepatic disease and consequently have both diagnostic and prognostic value (table 2).

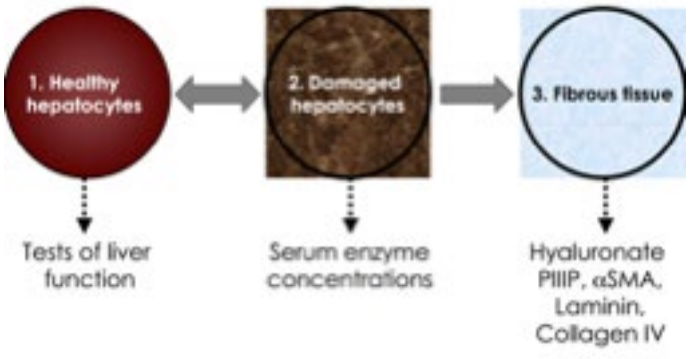

Figure 2. Schematic representation of liver cell populations and their primary influences on clinicopathological tests (for explanation see text).

Although undoubtedly useful, the diagnostic and prognostic values of serum enzymes are generally disappointing and should be interpreted with caution[2,3]. In contrast to a general high regard for GGT as the most clinically useful liver-derived enzyme in common usage one study indicated only 14% specificity for liver disease although it was unusual to see cases with liver disease without an increased GGT[2]. Only GGT concentrations > 399 iu/L had significant prognostic value[3]. Mild to moderate elevations of serum GGT may often be misinterpreted. Previous equine studies have suggested good diagnostic and prognostic value of serum concentrations of AST[5] and GLDH[6] although these conclusions were not supported by other studies[2,3].

Several authors have suggested that relative differences in serum concentrations of liver derived enzymes may infer the underlying nature of the liver disease with a primarily biliary source for GGT and AP and hepatocellular sources for ALT, AST, GLDH and IDH[1,7,8]. However, the association between enzyme patterns and histopathology is by no means absolute and should not be over-interpreted.

Functional indicators of hepatopathy offer greater diagnostic and prognostic value than serum enzymes although will not generally be increased unless hepatic insufficiency is present and, as previously mentioned, we should aim to intervene with diagnostic tests and treatment before this stage has been reached. Serum globulins were found to be most clinically useful prognostic blood analyte in one study[3] with globulins > 45 g/L signifying a poorer prognosis. Hyperglobulinaemia in association with liver disease is probably due to systemic immunostimulation by intestinal-derived antigenic material following loss of the protective barrier of Kupffer cells. Several reports have indicated that raised bile acids are a useful indicator of liver insufficiency and also a poor prognosis when > 20 micromol/L[3,6,9,10]. Bile acids, like other functional markers, tend to be normal in the early stages of liver disease. Although albumin is synthesised by the liver, its relatively long serum half-life in the horse perhaps explains the rarity of marked hypoalbuminaemia in equine liver disease[11]. Nevertheless, liver disease cases with hypoalbuminaemia have generally had poor outcomes[3,11].

Erythrocytosis, leucocytosis and neutrophilia were found to be indicators of a poor prognosis in horses with suspected liver disease[3]. The association between erythrocytosis and a poor prognosis most probably reflects dehydration and/or catecholamine release in those cases with a worse clinical status, although absolute erythrocytosis is a further possible explanation perhaps worth investigating. Absolute erythrocytosis is recognised as a paraneoplastic syndrome in cases of hepatoblastoma and hepatocellular carcinoma in horses[12,13]. Neutrophilic leucocytosis may have resulted from a systemic inflammatory response following loss of Kupffer cells as suggested for hyperglobulinaemia or, perhaps, simply a stress response.

Ultrasonography of the liver

Although ideally 3.5 – 6.0 MHz curvilinear probes are used, remarkably useful images of the liver can be obtained with standard 5.0 – 10.0 linear rectal or tendon probes, especially in thin animals. The liver is superficial and easily imaged using ultrasound immediately below the ventral margin of the lung although much of the dorso-cranial liver is not imageable owing to overlying lung. Most liver is situated on the right side of the horse and is typically imageable via at least 6 or 7 intercostal spaces (ICSs) somewhere between the 6th and 15th ICS, although some normal horses might only have imageable liver via as few as 1 or 2 ICSs. The normal hepatic image from the right side is approximately triangular in shape being imaged superficially to the bright echogenic arc of the colon (Figure 3). Typically 10-12 cm of liver projects caudoventrally from the expiratory border of the right lung in the 13th intercostal space. A smaller area of liver is imageable from the left side immediately caudal to the heart and cranial to the spleen and stomach somewhere between 6th and 9th ICSs (Figure 4). When imaged from the left side the liver should not be confused with the caudomedially adjacent spleen which has similar ultrasonographic architecture but is significantly more hyperechoic than the liver and has fewer blood vessels (Figure 4).

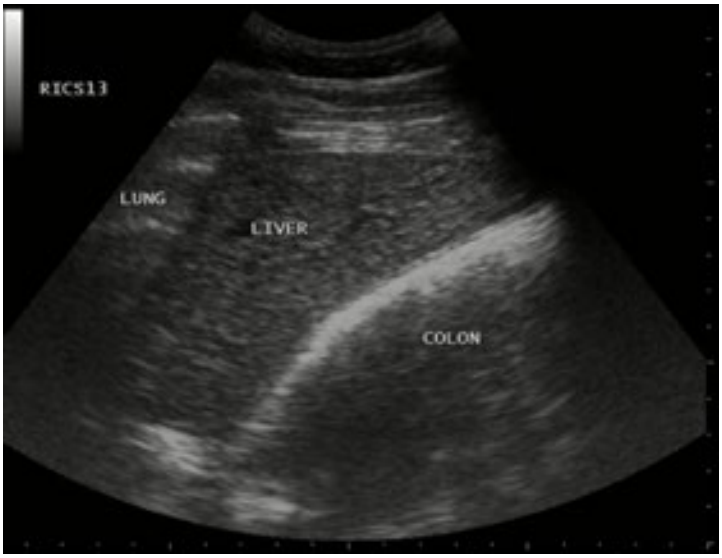

Figure 3.
Typical image of liver via right 13th intercostal space (dorsal left).

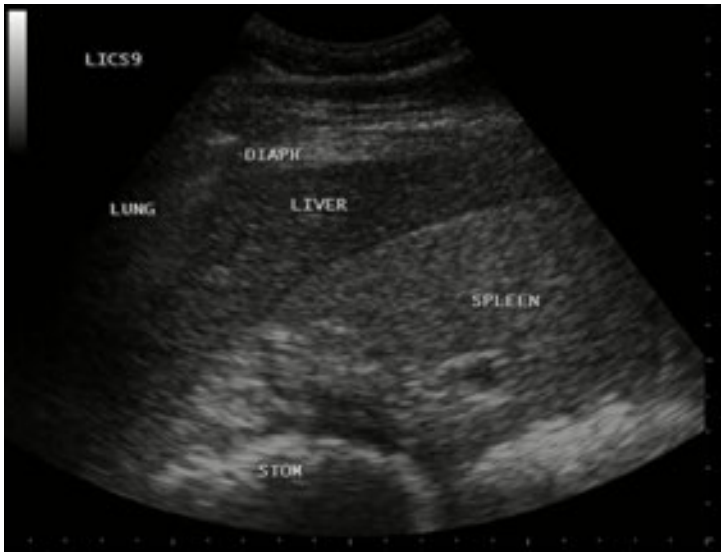

Figure 4.
Typical image showing liver next to spleen and stomach via the left 9th intercostal space.

The normal liver has a relatively hypoechoic homogeneity interrupted by anechoic blood vessels. Blood vessels in the readily-imageable superficial regions are typically <9 mm in diameter although larger vessels are sometimes seen in ponies due to the ability to image relatively deeper hepatic tissue than in horses. The large central caudal vena cava is often imageable from the right side in ponies and should not be confused with a cystic structure. Bile ducts cannot normally be visualised and vascular structures should generally be assumed to be blood vessels. In larger blood vessels flow can often be visualised in real time with standard B-mode ultrasonography (using a 3.5 to 5 MHz transducers) although Doppler can be used to establish active blood flow where doubt exists over the identity of dilated vessels. Fibrosis, haemosiderosis and lipidosis may all increase the echogenicity of hepatic tissue.

In this author's view, the primary usefulness of hepatic transabdominal ultrasonography is to guide liver biopsy and a site is usually chosen based on thickness of imaged hepatic tissue, absence of large blood vessels and, occasionally, focal presence of hepatic tissue with an abnormal ultrasonographic appearance. The widespread availability of diagnostic ultrasound makes the ongoing use of unguided biopsy techniques questionable.

Although the majority of cases of hepatopathy do not have discernable ultrasonographic abnormalities, images classified as abnormal have a high specificity for the presence of significant liver disease and are associated with poorer outcomes. Definition of ultrasonographic abnormalities is frequently subjective with the commonest classifications being changes in general echogenicity and size although focally abnormal tissue (eg. hydatid cysts, hepatoliths, neoplasia), excessively dilated vessels and rounded liver margins are among the more objective abnormalities occasionally imaged. In one study, horses with evidence of moderate fibrosis, severe haemosiderosis or moderate to severe biliary hyperplasia were significantly more likely to have ultrasonographic abnormalities detected and other authors have also proposed that fibrosis and lipidosis may increase the echogenicity of hepatic tissue.

Liver biopsy

If liver disease is suspected on the basis of preliminary non-invasive tests then liver biopsy remains the 'gold-standard' technique by which to confirm the presence of liver disease, establish prognosis and guide the choice of appropriate specific therapy.

It is a commonly stated dogma that liver disease in horses is almost always diffuse and therefore the site of biopsy is rarely of any great importance. It is generally assumed that collection of a biopsy from any site is likely to be representative of the underlying hepatopathy. However, work currently underway at Liphook involving multi-site biopsy

from clinical cases has unexpectedly revealed some variability between biopsy specimens from different sites of the same liver. However, further data are still being gathered and further analysis is required before firm conclusions can be made. Nevertheless on the basis of this author's experience of comparison of multi-site biopsies then I would generally recommend that more than one biopsy is collected.

The main theoretical adverse effect of liver biopsy in the horse is haemorrhage (figure 5), although in the author's experience this is exceedingly rare, especially when the technique is performed under ultrasonographic guidance, and the requirement for pre-biopsy coagulation assessment is questionable14 and never performed by this author. Nevertheless when collecting a liver biopsy from a horse with known coagulopathy then pre-treatment with 2-10 L of freshly sedimented plasma or whole blood might be wise. A quarter of human patients report some degree of abdominal pain following liver biopsy and therefore routine systemic analgesia should also be administered in horses.

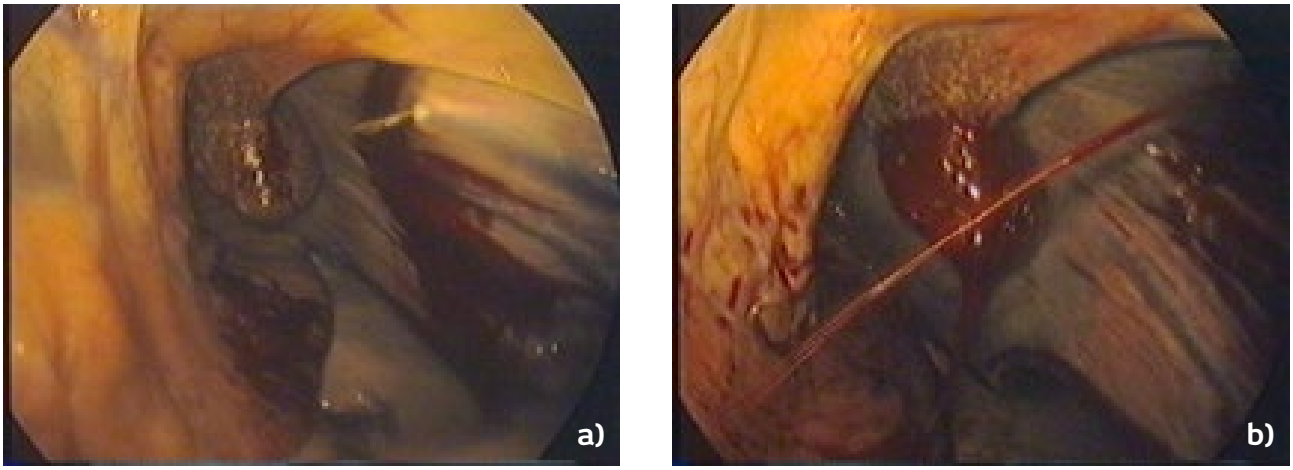

Figure 5 a.b. Laparoscopic images of a biopsy procedure from a horse with prolonged clotting times and resulting in apparent marked haemorrhage although no adverse clinical consequences arose.

a. With ultrasonographic guidance

Guides are available for many transducers which fix the biopsy needle in the plane of the ultrasonographic image and may facilitate the procedure although some clinicians choose to operate the transducer and biopsy needle separately from each other.

1. The subject is sedated.
2. Biopsy site and transducer are prepared for a sterile procedure.
3. 5 mL local anaesthetic is infiltrated subcutaneously and through the intercostal muscles to the parietal peritoneum.
4. A small stab incision is made through the skin.
5. A small amount of sterile coupling gel or alcohol is applied to the skin/transducer.
6. A 14 gauge 16cm biopsy needle is advanced perpendicularly to the skin into liver and the biopsy is collected. Slight angling cranially is helpful to increase the hepatic target.
7. The procedure is repeated if a suitable biopsy specimen is not obtained (2 or 3 attempts sometimes required).
8. Biopsy specimens are placed in 10% neutral buffered formalin for histopathologic examination and/or plain sterile containers for bacteriologic culture.
9. Topical antiseptic spray is applied to the skin incision.
10. A single dose of 2 mg/kg phenylbutazone iv is administered.

b. Without ultrasonographic guidance

The right 13th intercostal space midway between two lines drawn between the point of the shoulder and the tuber coxae and the point of the elbow and the tuber coxae is preferred but will not always be a suitable site.

1. to 4. as above.

5. A 14 gauge 16cm biopsy needle is inserted slowly perpendicularly to skin (or aimed slightly cranial) until rhythmic movement of diaphragm is felt (typically about 5 cm from skin surface).

6. The needle is further advanced by about 2-5 cm and the biopsy is collected.

7. The procedure is repeated if a suitable biopsy specimen is not obtained (more than 5 or 6 attempts are not recommended).

8. to 10. as above.

Interpretation of liver biopsies

Marked periportal and bridging fibrosis have frequently been regarded as poor prognostic indicators although long term survival of cases with severe periportal and bridging fibrosis has been reported suggesting other factors are also important15. In one study a scoring system was developed in order to attempt to attribute a prognostically useful broad comparative index of histopathologic severity8 (table 3). Biopsy scores from 73 cases showed a strong and statistically significant association with survival and survival times. Essentially horses with scores 0-2 tend to do well and have a good prognosis. Horses with scores between 8-14 have a poor prognosis for survival. Those horses with scores between 3 and 7 have a guarded prognosis and deserve reasonably aggressive therapy and close further monitoring.

Variable	absent	mild	moderate	severe
Fibrosis	-	-	2	4
Necrosis or megalocytosis	-	1	2	2
Inflammatory infiltrate	-	-	1	2
Haemosiderin accumulation	-	-	-	2
Biliary hyperplasia	-	-	2	4

Table 3. Biopsy scoring system8. Minimum score = 0, maximum score = 14.

In addition to establishing the presence of liver disease and determining prognosis, biopsy might potentially help establish the precise type of liver disease. Minor hepatopathic changes including mild portal lymphocytic infiltrates, mild to moderate haemosiderin accumulation and cloudy swelling or granularity of hepatocytes are frequently encountered in horses without liver disease and this should be considered when interpreting liver biopsy specimens.

Several specific types and causes of liver disease in adult horses are described in many recent equine medicine texts and include hepatotoxicosis (e.g. mycotoxins, pyrrolizidine alkaloids, iron), acute idiopathic hepatitis/necrosis (Theiler's disease), infectious (clostridial) necrosis, chronic active hepatitis, hepatic lipidosis, cholelithiasis, cholangiohepatitis, helminthiasis, amyloidosis, haemochromatosis, neoplasia, abscessation, hypoxaemia, septicaemia and endotoxaemia. However, in the author's experience such clear diagnostic categorisation is somewhat idealised and impossible in most clinical cases seen in practice. For example, common biopsy findings include increased portal lymphocytic infiltration, perhaps with some biliary hyperplasia and fibroplasia – not definitive for any particular type of liver disease. Furthermore, attempted culture of biopsies from cases of suppurative hepatitis is usually unsuccessful. Although sometimes possible, determination of aetiology should not be an expected result of liver biopsy in the

horse. Nevertheless, certain histopathologic findings may serve to guide therapy even in the absence of a specific diagnosis. For example, marked lymphoplasmacytic infiltrates may indicate glucocorticoid therapy; neutrophilic infiltrates may indicate antibacterial therapy; haemosiderosis may require phlebotomy and careful dietary advice to limit further iron ingestion; fibrosis may require anti-inflammatory drugs (e.g. prednisolone, pentoxyfylline, azathioprine) and antioxidants (Vitamin E).

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Tim Greet

Surgery in standing horses

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Historical perspective

- until the availability of safer general anaesthetic agents in the late 1960s equine surgical procedures were carried out on standing patients or on horses cast under some form of sedation or narcosis
- more effective sedatives became available for equine use in the1980s
- publication of the initial results of the CEPEF survey indicated the unacceptably high risks (1.6%) of routine equine general anaesthesia (johnston et al 1995)

Reasons for operating on standing patients

- avoid the risks of GA
- more effective sedative agents available
- more convenient for some procedures
- reduction in surgical time (some procedures)
- reduction in haemorrhage (nasal and sinus surgery)
- better access (ovariectomy)

current surgical repertoire in standing equine patients

- castration (open)
- wound repair
- paranasal sinus surgery
- tooth removal
- facial fracture repair
- upper airway laser surgery
- thoracic surgery
- laparoscopy (laparotomy)
- umbilical hernia repair
- urogenital surgery
- ligament and tendon surgery
- resection of DSPs
- foot surgery
- synovial lavage (arthroscopy!)
- limb fracture repair

open castration in standing patient

- benefits - avoid GA; reasonable access in well handled horse; time saved (especially for multiple patients); less expensive
- disadvantages / risks - surgeon safety; high testicle/ cryptorchid; unsuspected hernias potentially more problematic; no ligatures; tunic not closed

laparoscopic cryptorchidectomy

- benefits - avoids GA; technically straightforward; easy evaluation of "testicular status"; safe in sensible patient
- disadvantages - inguinal testicles; contralateral scrotal testicle; even mini-laparotomy delays return to exercise; cost of equipment and expertise required
- wounds
- benefits - avoids GA (especially useful if complicated by organ damage or a fracture); many are accessible; avoids complications of anaesthetic recovery for wounds where no bandage or cast possible eg head, trunk
- disadvantages - site may be inaccessible or awkward; more difficult evaluation (eg tendon injury); certain limb casts are easier to apply under GA; if multiple lesions or complex reconstruction required (eg eyelids); difficult patient temperament

paranasal sinus surgery

- standing sedated horse
- subcutaneous local around site of incision
- maxillary nerve block or lavage sinus with local anaesthetic
- open flap or trephine

maxillary nerve block

- useful for sinus surgery or extraction of maxillary cheek teeth
- local anaesthetic injected into the maxillary foramen in the pterygopalatine fossa

alternative simple approach

- block skin at site of incision
- sedate horse with detomidine and butorphanal
- may need additional topical local anaesthetic within sinus for some procedures
- plug ears with cotton wool as horse may be sensitive to noise of oscillating saw

lesions which can be treated

- chronic sinus empyema
- progressive ethmoidal haematoma
- maxillary cyst
- other sinus mass

facial flap surgery in standing patient

- benefits - avoids GA; reduced haemorrhage; improved access and orientation
- disadvantages - some patients intolerant (ear plugs?); if no maxillary block must be careful of infraorbital canal; radical fenestration into nasal passages more difficult

removal of cheek teeth from standing horse

- advantages - avoids GA; in most cases is relatively easy; dramatically reduces risk of oronasal fistula with maxillary teeth
- disadvantages - can be tricky in some horses because of temperament or very small mouth; can be difficult to pull fractured teeth or some of those with advanced disease; not as easy to deal with cement bodies or apical deformities; long reserve crowns in small mouths
- ocular enucleation in standing patients
- inject local anaesthetic around eyelids and deeply in eye socket
- routine transpalpebral dissection
- obviates vagal cardiovascuar effects
-

facial fracture repair

- usually limited in equine patients
- typically retrieving bone fragments from a depressed fracture into sinus
- usually comminuted and often minimally stable
- remove fragments which are likely to sequestrate
- wire remainder and repair wound
- minimal soft tissue cover
- sinus lavage

repair of rostral maxillary and mandibular fractures

- can be done under a short GA or in sedated standing horse
- tension band wires inserted into holes created with a 3.2mm drill or steinmann pin
- more complex fractures or difficult patients are best operated under a general anaesthetic
- facial and mandibular fractures
- advantages - avoids GA; many can be repaired simply with wires; recovery from GA can compromise facial repair
- disadvantages - complex fractures requiring major reduction or many drill holes; difficult patients; some horses resent the sound of the drill (ear plugs?)

upper airway laser surgery

- ventriculocordectomy
- epiglottal entrapment (not my preferred method)
- treatment of DDSP
- aryepiglottal fold ablation
- remove subepiglottal cyst (not my preferred method)

upper airway laser surgery in standing patient

- benefits- surgery easily performed in standing patient; sites usually more accessable cf traditional surgical approach; easy endoscopic access to most parts of the airway and far harder to evaluate anatomy under a GA
- disadvantages- collateral damage can be disastrous, probably even more likely under GA! rarely truculent patients can be a problem; no genuine disadvantage!

other minimally invasive airway surgery in standing horses

- hook knife treatment of epiglottal entrapment
- injecting formalin into progressive ethmoidal and other sinus "haematomas"
- laryngoplasty! (Fabrice Rossignol)

other minimally invasive techniques

- these techniques are less commonly performed under a GA because there is usually no need to do so
- the disadvantages of their use all relate to the nature of the treatment eg palatal injruy with the hook knife and the risk of encephalitis with large volume injection of formalin
- some opt to use open hook via mouth either standing or under a GA

thoracic surgery

- pleuroscopy
- rib resection for thoracotomy
- investigation of thoracic mass (biopsy?)
- treatment of severe fibrinous pleurisy
-

drainage of septic pleuritis in standing patient

- simple chest drain approach in acute cases
- thoracotomy in chronic or very septic cases especially if necrotic tissue necrotic tissue
- may use pleuroscopy via thoracotomy to improve view

pros and cons of thoracic surgery in standing patient

- advantages- patients often have severe respiratory compromise; accessible and achievable; reduced morbidity and mortality
- disadvantages- limited control of "panic-stricken patient" but even then is probably still safer; major surgery might be more problematic but very rarely attempted

laparotomy in standing patient

- collection of ileal biopsy in grass sickness
- other intestinal biopsy
- correction of large colon displacement
- correction of uterine torsion

pros and cons of standing laparotomy

- pros- avoids a GA; less expensive; can correct some intestinal and uterine problems more easily than under GA; ileal biopsy relatively straight forward
- cons- in difficult patients might result in catastrophe; not easy in horses with abdominal pain and many lesions can not be corrected; only limited ability to investigate GI tract; haemorrhage and difficulties in flank closure

laparoscopy in standing patient

- diagnostic purposes
- abdominal gonadectomy
- visceral biopsy
- closure of nephrosplenic space
- repair inguinal hernia
- vascular ablation in hysterectomy
- vessel and ureter sealing in nephrectomy

preparation is important

- starvation (empty gi tract)
- manual examination per rectum by surgeon
- pre-op sedation with acp
- quiet environment
- detomidine and butorphanol bolus
- detomidine slow infusion
- morphine if difficult or xylazine "top up" if ataxic
- flunixin pre-operatively

closure of the nephrosplenic space

- recurrent nse is an uncommon problem but we have had a few cases
- closure should be carried out electively after horse has recovered from nse
- requires intra-abdominal suture placement
-

laparoscopic surgery in standing horse

- pros- avoids GA; many techniques are more straightforward this way (eg ovariectomy); some techniques can only be performed this way (nephrosplenic space closure); postoperative morbidity reduced; hospitalisation times are reduced
- cons- very limited value for "colic patient"; very large ovaries can be problematic; gonad removal requires a flank laparotomy which can reduce its value in cryptorchids cf minimally inavsive approach under GA (time to return to exercise)

umbilical hernia repair

- application of a rubber ring to standing foal has largely replaced surgical closure under a ga
- care must be observed when twisting sac to eliminate intestine
- occasionally will become badly infected requiring surgical revision under a ga
-

urogenital surgery in standing mare

- repair of perineal lacerations
- urethral extension procedures
- much easier under epidural (9 mls 10% xylazine solution) in standing patient
- orientation of soft tissues infinitely easier to configure

urogenital surgery in standing mares

- pros- the best way to perform surgery as the anatomical relationships are very clear; performing it under a ga would be much more difficult
- cons- the rare mare which can not be restrained by chemistry; the rare unfortunate response to epidural anaesthesia

tendon and ligament surgery in the standing horse

- patellar desmotomy or split
- ddf tenotomy
- lat dig ext tenectomy

patellar ligament desmotomy

- careful case selection
- standard approach under sedation and local analgesia
- patellar splitting less invasive but effectiveness still being evaluated

a word of warning!

- using a disposable scalpel for medial patellar ligament desmotomy is contra-indicated
- even using it for a skin incision can have its problems if the patient is sufficiently awkward!

ddf tenotomy in severe laminitis

- severe distal phalangeal displacement
- last resort?
- standing patient under local analgesia
- pros- usually straightforward; easy orientation for surgical approach; avoids ga risk
- cons- retrieving lat digital extensor tendon can be quite hard; severely laminitic horse may have trouble standing on three legs; temperamental horse may be tricky

foot surgery in standing horse

- distal phalangeal sequestrum
- keratoma
- palmar digital neurectomy
- applying a foot cast (forelimb only)

distal phalangeal sequestrum

- sedated horse
- pd nerve block
- apply tourniquet (vetwrap)
- good sharp hoof knife and curette
- hospital plate (bar shoe)

keratoma removal in standing horse

- set up as for sequestrum removal
- cut two parallel lines in hoof corresponding to lesion
- attempt to "peel off" keratoma with segment of hoof wall
- careful inspection of proximal extent of lesion

foot casting in standing horse

- can be applied effectively to most horses for a front foot
- more difficult to apply a good cast to a hind foot
-

foot surgery in standing horse

- benefits- avoids ga; very practical for sequestrum removal; some keratoma cases can also be managed this way
- disadvantages- I much prefer performing pd neurectomy and keratoma removal under ga; tricky patients may only be manageable under ga, especially if hind limb involved; if horse has multiple limb problem may be easier under ga; applying a good foot cast to a hind foot is very much more difficult in standing horse

resecting dorsal spinous processes

- diagnosis!
- perkins, schumacher, kelly, pollock and harty 2005 vet surg
- sedation and copious local analgesic
- oscillating saw or hammer and chisel ± rongeurs using standard approach
- quiet room

dsp resection

- benefits- eliminates ga; easier access; improved haemostasis
- disadvantages- may be difficult in a small number of patients because of temperamental problems!

synovial lavage in standing horse

- synovial wounds or sepsis cases
- capacious joints like tarsocrural
- some tendon sheaths (digital, tarsal)
- some bursae (calcaneal and subcut. at point of hock)
- economic reasons
- concurrent fracture or other injury which precludes safe ga
- use egress cannulae and arthroscopic fluid pump

synovial lavage in standing horse

- benefits- cheaper; avoids ga risk; successful particularly in acute cases; large cavities preferred
- disadvantages- distension is painful; less forgiving especially if use needles rather than cannulae; patient tolerance often limited even under sedation; debridement limited; foreign material may remain; no visual assessment of intrasynovial structures; endoscopic lavage and debridement must be considered the gold standard

repair of limb fractures in standing patients

- non-displaced lateral condylar fractures of mc/ mt 3
- medial parasagittal fractures of mc/mt 3
- proximal phalangeal fractures

fracture repair in standing horse

- pros- no ga and therefore reduced risk of catastrophic failure of repair; not difficult to perform; most horses tolerate well (tourniquet)
- cons- perhaps technically not as satisfactory?; temperament may make very difficult in a few horses; fracture reduction less good (limb weight bearing)??? arthroscopic monitoring not possible so not suitable if fracture displaced or fragment to be removed

safe facilities for standing surgery

- a quiet area outside the busy areas of the hospital
- good non-slip flooring
- good lighting and drainage
- "adaptable" stocks
- head restraint
- overhead electric sockets
- screen
- equipment and drugs near at hand
- scrub facilities
- security of area (laser surgery)

Tim Greet

How reliable is endoscopy at rest for respiratory evaluation during a pre-purchase examination? The Vet's Perspective.

Rossdales Equine Hospital, Newmarket - UK

Pre-purchase examination

- assessment of the suitability of a horse for a purchaser's specific needs
- the examiner must find out in detail the requirements and in an ideal world will know the purchaser or try to obtain a little background information about them

assessment of upper airway function

- is function adequate during resting respiration?
- is dynamic function adequate?
- this will depend entirely on the intended use of the horse
- respiratory requirements for a "sprinter" are quite different than for a "stayer" and different again from a show jumper or a show horse

clinician's approach to evaluating dynamic upper airway function

- respiratory noise at exercise?
- exercise intolerance?
- endoscopy at rest or after exercise
- dynamic endoscopy
- any assessment during exercise will only be useful if the test is able to mimic the horse's intended future use

obstruction during quiet respiration

- conditions obstructing quiet respiration inevitably cause obstruction at fast exercise
- usually readily identified by endoscopy during resting respiration or by using other imaging modalities
- this presentation will consider those conditions causing obstruction only at exercise

airway obstruction only at exercise

- 4 bad
- epiglottal entrapment
- recurrent laryngeal neuropathy
- arytenoid chondritis
- pharyngeal cyst
- laryngeal web
- nasopharyngeal collapse *
- intermittent ddsp *
- aryepiglottal fold instability *
- vocal cord collapse *
- cricotracheal ligament collapse *
- epiglottal retroversion *
- ventromedial corniculate luxation *

can endoscopy at rest provide information about upper airway function during exercise?

- some causes of major functional disturbance are obvious at rest
- some "clues" to dynamic dysfunction may be noticed (eg subepiglottal ulcer)
- stimulating swallowing encourages laryngeal motility and may help to identify dysfunction
- respiratory effort can be stimulated by administering chemicals (doxapram) or by nasal occlusion
- unless future exercise effort can be mimicked the test will be significantly less informative than dynamic endoscopy

conditions causing obstruction only at fast exercise

- historically such conditions presented a problem to clinicians carrying out a PPE even if resting endoscopy was available for use (eg ddsp)
- nowadays remote dynamic endoscopy is available for more accurate diagnosis
- many of these conditions are common only in the racehorse
- most but not all seem to be performance-limiting
- mismatch between noise at exercise and endoscopic findings at rest?
- post-exercise endoscopy can provide information
- however often fails to correlate to findings during fast exercise
- dynamic endoscopy is best practice under such circumstances

what can dynamic endoscopy reveal about the urt during fast exercise?

- intermittent ddsp
- intermittent epiglottal entrapment
- medial deviation of the aryepiglottal fold(s)
- vocal cord collapse
- nasopharyngeal collapse
- cricotracheal ligament collapse
- epiglottal retroversion
- dynamic laryngeal collapse (rln)

intermittent dorsal displacement of the soft palate during galloping

- easy to recognise if full ddsp
- prodromal "billowing" (instability) is a more subjective diagnosis
- some horses seem to displace after a gallop
- if exercise inadequate or no racing stress horse may not displace
- ddsp during resting endoscopy of very debatable significance (? Barakzai and Dixon evj 2011?)
-

intermittent epiglottal entrapment

- may be noted during resting endoscopy (ulcer?)
- typically triggered by swallowing reflex
- may not occur during every exercise period
- may be very transient
- probably just as likely to be identified during endoscopy at rest (stimulate swallowing)

vocal cord collapse

- can not be diagnosed without dynamic endoscopy
- typically a consequence of recurrent laryngeal neuropathy (or cricothyroid muscle dysfunction? Holcombe et al 2006)
- of greatest significance in absence of major collapse of arytenoid
- represents more of a therapeutic dilemma than a diagnostic problem

nasopharyngeal collapse

- can not be diagnosed without dynamic endoscopy
- may be dorsal, dorsolateral or lateral impingement
- may be associated with other URT problems
- typically racehorse or show horse
- pharyngeal lymphoid hyperplasia has no effect on function at exercise and no value as a predictor of collapse

instability and medial deviation of the aryepiglottal folds

- can not be diagnosed by endoscopy at rest
- typically a problem in racing thoroughbreds
- may be associated with other airway problems

epiglottal retroversion

- can only be diagnosed by dynamic endoscopy
- seen in many types of horse
- loud "pig-like" inspiratory noise
- usually affects performance

cricotracheal ligament collapse

- only possible to diagnose using dynamic endoscopy
- significance uncertain
- in many cases seems asymptomatic

…… but what about recurrent laryngeal neuropathy ?

- traditionally regarded as a straight-forward diagnosis using endoscopy at rest
- grades of "normal" and "abnormal" laryngeal movement (different grading systems)
- how does the appearance of the resting larynx compare with that at exercise?

horse 1

- multiphasic movement of left side of larynx
- capable of symmetrical abduction during quiet respiration
- full symmetrical abduction throughout fast exercise

horse 2

- reduced left adduction (compensatory right over-adduction)
- slightly delayed and marginally reduced left abduction at rest
- normal symmetrical abduction at exercise

horse 3

- at rest basically a symmetrical larynx
- at exercise severe dynamic left-sided laryngeal collapse

horse 4

- moderately reduced left-sided movement with slight asymmetry at rest (certainly more marked than horse 3)
- symmetrical fixed abduction at exercise

horse 5

- essentially symmetrical larynx with normal movement at rest and even with nasal occlusion
- bilateral vocal cord collapse and right aryepiglottal fold impingement
-

how reliable is endoscopy at rest in a ppe?

- it cannot identify the many recently recognised causes of upper airway obstruction at exercise
- it is unreliable in diagnosing less advanced cases of RLN and intermittent DDSP
- it is reliable at identifying advanced cases of RLN and physical causes of airway obstruction present during quiet respiration

managing purchasers' expectations

- insist that the horse must undergo an exercise test equivalent to that which the horse will be required to perform in future
- if endoscopy is requested point out the limitations of resting and post-exercise endoscopic examination
- dynamic endoscopy is the gold standard

if dynamic endoscopy is unavailable what is going to help protect you from litigation?

- careful pre-examination discussion with purchaser (managing expectations)
- appropriate exercise test to evaluate respiratory sounds
- video-recording of resting or post exercise endoscopy ???
- laryngeal palpation
- laryngeal ultrasound (CAL muscle)

Andy Durham

Treating diarrhoea in mature horses

BSc.BVSc.CertEP.DEIM.DipECEIM.MRCVS,
RCVS and European Specialist in Equine Internal Medicine

Diarrhoea is a very common problem in adult horses. The majority of cases remain idiopathic although recent diagnostic aids have dramatically improved the diagnostic rate and are well worth employing.

Unlike in foals, small intestinal disease alone is very unlikely to cause diarrhoea in adult horses – there will almost invariably be large intestinal disease (alone or with small intestinal disease). There are 4 general classes of diarrhoea in adult horses that present to us in practice and it is helpful to categorise cases prior to deciding the therapeutic approach:

1. Most commonly, an innocuous mild diarrhoea in horses that are systemically well with good haematological and serum biochemical results and are not significantly threatened by the effects of the diarrhoea
2. Subacute/chronic protein losing enteropathies
3. Acute severe colitis cases in which fluid, electrolyte, acid-base and protein disturbances along with endotoxaemia are acutely life-threatening
4. Cases are occasionally encountered that produce fairly normally formed faeces along with a separate "water phase". Although frequently innocuous for the horse, this does bother owners considerably.

After categorising the diarrhoea case as above, certain empirical therapies may be applied pending the possibility of a specific diagnosis:

- mild innocuous diarrhoeas should receive larvicides, codeine phosphate and also psyllium might be employed if in a sandy area. Attention to dietary quality may also be useful. Some cases might benefit from a short course of glucocorticoids additionally if the above does not help.
- Subacute/chronic protein losing enteropathies are usually treated with prolonged courses of glucocorticoids and possibly azathioprine or metronidazole.
- Severe acute colitis cases will require fluids, electrolytes and frequently colloid therapy. They may also merit empirical treatment with larvicides and adsorbents. Probiotics, antimicrobials, NSAIDs and glucocorticoids might be used in some cases.

These various treatments are described below:

Larvicides

The correct choice of anthelmintic for killing larval cyathostomes is hard to define with several contrasting research findings. Broadly the 2 main licensed choices are fenbendazole at 7.5-10 mg/kg po sid for 5 days (Panacur Equine Guard 10%: 40-50 ml daily per 500kg) or moxidectin at 0.4 mg/kg po (Equest gel). However, given the widespread benzimidazole resistance in cyathostome populations worldwide, it is increasingly hard to recommend fenbendazole. One-off treatment is advisable as an empirical approach to mild diarrhoeas. If larval cyathostominosis is suspected

then repeat treatments every 4 weeks for 4 or 5 cycles is advisable owing to probably poor kill rate of early stage 3 larvae (L3s). Additional therapy with codeine and corticosteroids is usually required if severe diarrhoea and/or weight loss is present and close attention to restoring plasma oncotic pressure is mandatory.

Codeine phosphate

This is very effective as empirical treatment of mild non-specific diarrhoeas and for reducing fluid losses in more severe colitis cases although its use in the latter is controversial. Codeine phosphate may be used in mild cases at an initial dose of around 0.5-1 mg/kg bid (e.g. 5 x 60mg tablets per 500kg) or up to 3 mg/kg tid (25 x 60 mg tablets per 500 kg) in severe cases. Whatever the initial dose, therapy should be gradually withdrawn according to response as abrupt cessation often leads to recurrence. For example, a good response to 5 tablets bid could be followed by reducing by one tablet each day. Higher dosages require more rapid but still gradual reductions - a good response to 25 tablets tid could be followed by reducing to 15 bid-tid on the next day and then to 10, 8, 6, 4 etc...if there is still control of the diarrhoea. Problems with impaction are occasionally seen but are usually manageable if detected early. Loperamide (Imodium, 0.1-0.2 mg/kg bid-tid) can be used as an alternative but is more expensive.

Psyllium

Psyllium is often fed to horses considered to be affected by, or at risk from, sand enteropathy with the intention of increasing sand removal from the colon. Not all studies have supported its usefulness but it has a reasonable evidence basis when used at 0.5 to 1 g/kg perhaps combined with mineral oil. Longer term use of psyllium as a preventative can be used at the same dose rate but is very expensive at this rate.

Anti-inflammatory agents

Glucocorticoids

A low dose of dexamethasone (0.05 mg/kg) or prednisolone (1 mg/kg) may be indicated for a few days in persistent mild diarrhoea if codeine phosphate has not been effective alone.

Chronic diarrhoea due to chronic inflammatory bowel disease carries a guarded prognosis but a long course of prednisolone (1 mg/kg sid) or dexamethasone (0.1-0.05 mg/kg) may benefit some cases. Intramuscular injections might be worthwhile at the start of the course due to concerns over malabsorption. Treatment can be reviewed every 2-3 months according to clinical response and serum albumin concentrations and may have to continue for several months, or occasionally years.

Judicious doses of dexamethasone (0.05 mg/kg iv) may sometimes be helpful in more severe colitis cases in an attempt to reduce bowel oedema and inflammation. Laminitis risk should be considered in such cases.

All cases of larval cyathostominosis should be treated with dexamethasone at 0.05-0.10 mg/kg im daily along with larvicides. Prednisolone at 1 mg/kg orally sid may be a safer option and might be required for several weeks in combination with a reducing dose of codeine phosphate in some cases.

Azathioprine

Azathioprine may be used as an anti-inflammatory agent in horses where glucoocorticoids have been unsuccessful or are undesirable due to concerns regarding adverse effects. Azathioprine appears safe to use in horses with very rarely reported adverse effects even after several weeks of use although monitoring leucocyte numbers might be wise during prolonged therapy (suppression of leucocyte synthesis is the most likely adverse effect). Although there are questions over the bioavailability of the drug it is widely accepted as efficacious in horses at a dose of 3 mg/kg once daily by mouth.

NSAIDs
Much recent evidence suggests significant impairment of mucosal recovery by NSAIDs so they should only be used if necessary in diarrhoea cases, and preferably not until rehydration has been achieved due to the possibility of renal injury. Nevertheless NSAIDs are indicated to control pyrexia and some of the effects of endotoxaemia and may improve appetite.

Firocoxib (0.1 mg/kg sid-bid iv or po) may be argued to be the preferred choice due to high COX-2 specificity. Although specific inhibition of COX-2 may be less harmful to a normal gastrointestinal mucosa, there is no evidence to suggest that it creates less interference with recovery of damaged/diseased GI mucosa. The harmful effects of NSAIDs might possibly be countered by the PGE analogue misoprostol @ 2 ug/kg qid (5 x 200 ug 'Cytotec' capsules) and this can be tried in suspected NSAID-toxicity cases.

Oxypentafylline
This is another anti-inflammatory agent without the harmful effects of NSAIDs or glucocorticoids that can reduce several inflammatory cytokines and can be used at 8 mg/kg bid po. Not many reports exist in cases of colitis although there is some evidence to support its use in endotoxaemic cases.

Lidocaine
Although better known as a prokinetic for postoperative colics, it is well recognised that lidocaine offers both analgesia and also potent anti-inflammatory actions on the intestine. This author frequently uses this drug in colitis cases by constant rate infusion of 3 mg/kg/hr (15 mL 2% solution per 100 kg per hour) after an initial "loading bolus dose" of 1.3 mg/kg iv (6.5 mL 2% solution per 100 kg).

Antimicrobials

Use of antimicrobials in cases of diarrhoea is controversial but there are clear indications in some cases. Systemically sick or pyrexic colitis cases should receive broad spectrum antibiosis (eg. gentamicin/penicillin) to counter probable bacterial translocation through a compromised mucosa. Also cases of confirmed bacterial enteritis should receive antimicrobials depending on in vitro sensitivity patterns or intuitively predicted susceptibility. Generally oral enrofloxacin (5 mg/kg sid iv or 7.5 mg/kg sid per os) is the drug of choice for aeromoniasis and salmonellosis, and oral metronidazole (15 mg/kg loading dose followed by 7.5 mg/kg q 6 hours) for enteric clostridiosis. Metronidazole might also be indicated in chronic IBD cases owing partially to the suspected aetiology involving immune dyregulation and reaction against enteric microbes and also due to anti-inflammatory properties of this drug.

Intestinal adsorbents

Di-tri-octahedral (DTO) smectite (Biosponge; Platinum performance, California)
This is perhaps the best evidence-based product and has been shown to neutralise endotoxin and clostridial exotoxins and can be used in acute severe colitis cases (3g/kg initially followed by 1 g/kg tid-qid) and also mild diarrhoeas. There is evidence of neutralisation of clostridial toxins and endotoxin in vivo at doses used in vivo. A more potent toxin-neutralisation effect when compared with bismuth subsalicylate has been demontrated.

Bismuth subsalicylate is an alternative (Pepto-bismol) @ 1 ml/kg bid per os bid or activated charcoal at 0.5 -1 g/kg sid-bid.

Pre/probiotics

Many prebiotic and probiotic products are marketed for horses with gastrointestinal diseases and may have a reasonable evidence basis in some other species. The quality of commercially available probiotic products was scrutinised and questioned in one study which found that most products contained few, if any, viable or potentially beneficial organisms and sometimes potential pathogens were encountered. This author has also found a pure growth of Enterococcus gallinarum, an occasional cause of antibiotic-resistant nosocomial infections in humans, in a commercially available equine probiotic product claiming to contain Lactobacilli. An evidence-based approach to the design of appropriate equine bacterial probiotic products has thus far been unsuccessful. In contrast, the investigation of yeast-containing probiotics including Saccharomyces cerevisiae and Saccharomyces boulardii have generally provided better quality supportive evidence in horses with gastrointestinal disease and have been shown to protect against the adverse effects of starch overload and also to significantly reduce the duration of diarrhoea in clinical enterocolitis cases. Evidence of benefit does not exist in chronic diarrhoea cases however. Addition of short chain fructooligosaccharides (scFOS) (up to 30 g daily) has also been shown to protect the colon from dysfermentative effects of carbohydrate overload.

Omeprazole

There is a high incidence of gastric ulceration in diarrhoea cases –most likely as a result, rather than cause, of the diarrhoea. Nevertheless, if significant ulceration is present then omeprazole is indicated.

Fluid therapy

This is a key part of management of acute and severe diarrhoea cases. A vital point worth emphasising at the outset is that fluid therapy in the absence of colloid to retain the fluid intravascularly is potentially very harmful in hypoproteinaemic horses. Therefore intravenous hetastarch (or plasma) is a key initial part of the management of protein losing enteropathies.

Quantity of fluid

The estimated fluid requirements are the sum of:

pre-existing deficit (D) + maintenance (M) + ongoing losses (L)

- The pre-existing deficit (D) in a dehydrated horse will typically range from 5% BWT (any less will probably not be clinically detectable) to 12% BWT (any more will be probably be associated with collapse and death).
- Maintenance requirements (M) are somewhat dependent of environmental temperature and ongoing dietary intake but approximate:

  Adult horses: 4-5% BWT/day = 40-50 mL/kg/day = 2 mL/kg/hr

  Foals: 10% BWT/day = 100 mL/kg/day = 4 mL/kg/hr
- Ongoing losses (L) can only be guesstimated

The plan for fluid therapy should be quickly written down as this is not a difficult task. The plan should be provide for M and L and to "catch up" D over 6-12 hours.

e.g. 500 kg horse, estimated 8% dehydration, losing 2 L/h in diarrhoea

D = 0.08 x 500 = 40 L. Replacement over 8 hours will require 5 L/h

M= 500 x 2 mL/kg/hr = 1 L/hr

L = 2 L/h

Therefore the plan is for 8 L/hr for first 8 hours followed by 1 L/hr (M) plus any ongoing losses.

Type of fluid

Colloid

This is required as a first step if there are signs of oedema or if estimated colloidal osmotic pressure is <15 mmHg (where COP= ([alb]x0.55)+([glob]x0.25)-4.4). Hydroxyethyl starch 6% (e.g. Voluven, Baxters) can be given at 10-15 ml/kg iv over >45 mins. This is the easiest approach due to ready availability of large volumes although plasma could be given instead if available. Hetastarch therapy can be repeated daily for at least 3 days depending on progress.

Plasma transfusion tends to be associated with superior clinical effects in this author's experience. The main limitation is availability. Donors are best selected as young geldings as these are unlikely to be previously sensitised to foreign red cells (occurs mainly during previous transfusion or pregnancy). Up to approximately 2% of BWT as blood can be collected from a healthy donor (e.g. 500 kg donor - 10 litres blood). This will probably result in harvest of around 55-60% of the collected blood as plasma (e.g. 500 kg donor - 6 litres plasma). Thus collection of 6 L plasma from a single horse may represent around 200 g albumin (6 L x 35 g/L). If transfused into a hypoalbuminaemic 500 kg recipient (estimated plasma volume 35 L), then serum albumin will be increased by approximately 6 g/L at most. Hence at least 2 or 3 donors are preferred if hypoalbuminaemia is marked (eg < 15 g/L).

Electrolytes

It is unlikely that sodium and chloride will require specific supplementation in most cases as these ions are plentiful in iv fluids. However, problems may arise with deficits in K, Ca and Mg as well as acid-base abnormalities.

Potassium

Potassium may be estimated as the "exchangable cation deficit" from the degree of dehydration and [Na+] although it is probably easiest and acceptable simply to guesstimate a K+ replacement plan from the plasma concentration of K+ and the administered fluid rate.

In cases of hypokalaemia in adult horses it is generally safe to supplement Hartmann's solution (contains 4 mmol/L K+) with an additional 20 mmol/L K+. This can be done by adding 10 mL 15% KCl per litre of Hartmann's. The maximum administration rate of K+ should be < 0.5 mmol/kg/hour. Thus the supplemented Hartmann's (24 mmol/L K+) should not be given faster than 10 L/hour for a 500 kg horse (infusion rates as fast as this are actually hard to achieve). Safe adminstration rate may become an issue in foals and ponies with this supplemented solution and the safe infusion rate should be calculated (e.g. 50 kg foal – no more than 25 mmol/hour = 1 L/hour; 250 kg pony – no more than 125 mmol/L = 5 L/hour).

If hypokalaemia persists despite supplementing as above then either further K can be added (e.g. 40 to 60 mmol/L – subject again to max rate of 0.5 mmol/kg/hr) or plasma Mg2+ should be checked as hypomagnesaemia may impair restoration of plasma potassium. Alternatively oral supplementation with KCl can be given.

Calcium

Hypocalcaemia is common in colitis cases and can be replaced if very low (< 2 mmol/L total Ca; or <1.0 mmol/L ionised Ca2+). There are formulae available for calculation of supposed calcium deficits although in this author's experience they are rarely helpful or accurate. Most clinical calcium deficits in horses are correctable with 100 mL 40% calcium borogluconate and this can be administered slowly intravenously (in fluid bag) or subcutaneously. Should this not resolve the hypocalcaemia then the dose can be repeated.

Routine supplementation of Hartmann's (contains 2 mmol/L Ca2+), can be provided with 10 mmol/L calcium (10-15 mL/L 40% or 20-25 ml/L 23% calcium borogluconate).

Magnesium
As Mg2+ is primarily an intracellular cation, deficits cannot be calculated practically. There is no magnesium in Hartmann's solution and it can be safely supplemented with 1 to 2 mL 25% MgS04 per litre.

Hypomagnesaemia can be treated with empirical MgSO4 supplementation at 2 mg/kg/min up to a maximum of 50 mg/kg:

eg 500 kg horse; TMg = 0.4 mmol/L
MgSO4 supplementation 2 x 500 = 1,000 mg/min up to max of 50 x 500 = 25,000 mg
25% MgSO4.7H20 contains 122 mg/mL MgSO4 (=1 mmol/mL) so rate required is 1000/122 = 8 ml/min up to max of 25,000/122 = 200 ml:

ie 200 ml over 30 mins. This can be repeated after a few hours if test results indicate persistent hypomagnesaemia.

Diet

Starch and fructan ingestion should be eliminated or restricted to ensure complete small intestinal digestion and absorption so that the large bowel is not disturbed by rapidly fermented non-structural carbohydrates. To this end temporary abstinence from grazing and exclusion of concentrate feeds (or at least limitation of concentrate feeds to no more than 1 g starch per kg bodyweight per meal (e.g. ≤ 200 to 300 g concentrate feed per 100 kg bodyweight per meal) is advisable although such feeds could be given every 4 to 6 hours. Additionally a source of easily fermentable fibre such as non-molassed sugar beet pulp, psyllium or soya hulls may be offered along with free access to a good quality grass or alfalfa hay. Oil should ideally be avoided until diarrhoea has resolved and then might be gradually introduced starting at 0.1 ml/kg bodyweight per day and increasing over 2 to 3 weeks to 1.0 ml/kg bodyweight per day if weight gain is considered important.

Cases with normally formed faeces along with a separate "water phase" are likely to be from either failure of hindgut water absorption and/or loss of the association between water and the faecal material in the rectum. The latter may be promoted by highly fibrous and indigestible diets – such as poor quality forage and/or poor mastication due to dental disease. Thus, attention to dentition in association with an increase in dietary fibre quality is frequently successful – eg good quality hay, alfalfa, increased grazing, sugar beet pulp. Occasional cases may require low dose codeine and prednisolone treatment but still others will fail to respond to all of the above.

Maria Pimenta

Transferência de embriões em éguas. Como e porquê. Aspetos práticos do dia a dia e alguns protocolos utilizados

A transferência de embriões em éguas é realizada quando se pretende obter mais de um descendente por ano de uma égua ou para a obtenção de produtos de animais demasiado jovens (2 anos) ou que estejam em competição ou, ainda, que tenham alguma patologia reprodutiva ou clínica que lhes impossibilite manter uma gestação a termo. Podem também ser realizadas apenas para evitar o risco do parto em éguas valiosas.

Hoje em dia, a técnica mais utilizada é a não cirúrgica em que os embriões são recolhidos por lavagem uterina transcervical e são transferidos também por via transcervical.

As éguas dadoras são inseminadas ou cobertas por monta natural, sendo realizados todos os tratamentos necessários, da mesma maneira, como se destinassem simplesmente a emprenhar. Devem ser monitorizadas diariamente quando estão em estro para a detecção exata do dia da ovulação. Por vezes, é necessária a utilização de agentes indutores da ovulação para uma melhor sincronização com a receptora.

As éguas receptoras, destinadas a um programa de transferência de embriões, deverão ser selecionadas consoante a sua saúde reprodutiva, idade (ideal dos 4 aos 8 anos), comportamento e tamanho, que deve ser semelhante ao da dadora. É de extrema importância o seu acompanhamento durante todo o seu ciclo reprodutivo para descartar qualquer problema que lhes possa diminuir a probabilidade de emprenhar após a transferência de um embrião.

A recolha de embrião é realizada geralmente entre o 7º (D7) e 9º dia (D9) após a detecção da ovulação da dadora. O embrião chega ao útero cerca de 6,5 dias após a ovulação. Em éguas velhas (>18 anos), o transporte dos embriões pelo oviducto e a sua migração para o útero pode ser mais tardia e por essa razão a recolha não deverá ser realizada antes do 8º dia.

Quando é utilizado sémen congelado, a recolha deve ser feita cerca de meio dia mais tarde, pois a fecundação não se realiza no momento da ovulação.

Uma vez no útero, o crescimento embrionário é muito rápido pelo que os embriões D9 (éguas jovens) ou D10 já podem ser demasiado grandes para transferir.

Para a recolha de embrião, o animal deverá estar contido num tronco, podendo ser sedado, com a cauda atada e a zona perivulvar lavada. A recolha é feita por lavagens uterinas em que é introduzido um cateter de Foley (24 a 32 FR) estéril pela cérvix e o cuff é insuflado. São realizadas lavagens sucessivas, geralmente entre 4 a 6 lavagens, com um volume entre 500 ml a 2 l, em que o fluido (Lactato de Ringer) é recuperado, por gravidade e massagem uterina, e filtrado. Pode ser administrada oxitocina para facilitar a recuperação de fluido.

Existem diversos sistemas de recolha: sistema aberto ou fechado - de uma ou duas vias; e existem diferentes tipos de filtros. Em certos filtros a pesquisa de embrião na lupa pode ser feita diretamente nos mesmos e noutros é necessário passar o seu conteúdo para uma ou mais placas de Petri.

Uma vez terminada a recolha, o filtro ou placa de Petri é então observado à lupa. Deve ser feita uma procura pormenorizada e quando visualizado um embrião, este é transferido para uma placa de manipulação com meio especial (Holding media) onde é lavado por "diluições sucessivas". Toda a manipulação do embrião deve ser realizada com a menor contaminação possível.

As duplas ovulações devem ser registadas para que, no dia da recolha, se tenha em atenção a pesquisa de dois embriões.

Os embriões recolhidos podem ser mórulas ou blastocistos e podem ser graduados consoante a sua morfologia. Esta graduação está diretamente relacionada com a taxa de prenhez após a transferência e inversamente relacionada com a perda embrionária entre o 16º e 50º dia.

Podem também ser recolhidos oócitos não fertilizados (UFOs) que, por vezes, são dificilmente diferenciados das mórulas. Os UFOs são geralmente de pequena dimensão, de forma redonda ou oval, mas são achatados e não "rolam" quando são manipulados.

Consoante o seu tamanho, o embrião é carregado numa palhinha de 0,25 ou 0,5 ml e transferido com auxílio da Cassou gun ou pode ser carregado diretamente numa pipeta de inseminação. São feitas três colunas de meio, intercaladas por colunas de ar. O embrião é colocado na segunda coluna de meio. A primeira coluna de meio deve ser a maior com a finalidade de "empurrar" o embrião.

Entretanto, é preparada a receptora para a transferência. Geralmente, são utilizadas éguas 4 a 8 dias após a sua ovulação. Ou seja, a receptora deve ovular no mesmo dia ou até 4 dias após a dadora (isto se a recolha for feita no D8).

A receptora deve ser examinada, por palpação e ecografia, no dia da transferência, isto é, previamente a esta operação. É importante verificar o tónus uterino e cervical, a presença do corpo lúteo e a ausência de líquido no útero.

A transferência é feita pelo método transcervical em que o embrião é colocado no corpo ou corno uterino, tendo o cuidado de não manipular excessivamente a cérvix. Pode ser feito por meio manual, semelhante à inseminação, ou com auxílio de uma pinça especial (Wilsher forceps). No método manual devem ser utilizadas uma ou duas luvas de palpação estéreis ou viradas do avesso. Visando uma menor contaminação, podem ser utilizadas duas luvas em que é cortada a mão da segunda luva, servindo de uma primeira barreira da vulva para o vestíbulo e é utilizada uma bainha sanitária que é rompida já dentro da cérvix. A "injeção" do embrião deverá ser feita lentamente, principalmente em embriões de grandes dimensões, pois estes são muito sensíveis e podem danificar-se contra a parede uterina. Após a inovulação com auxílio da Cassou gun deverá verificar-se se o embrião não ficou retido entre os orifícios laterais da bainha. Se tal acontecer, procede-se a uma nova transferência.

A receptora pode receber suplementação hormonal (Altrenogest) após, ou mesmo uns dias antes, da transferência. Se tal for o caso e a receptora emprenhar, deverá manter-se a suplementação até no mínimo 100 dias de prenhez (120 dias).

Por vezes são administrados anti-inflamatórios não esteroides (Flunixine Meglumine) o que pode fazer parte do pro-

tocolo de rotina ou aplicar-se apenas no caso de manipulação excessiva da cérvix. A administração de antibióticos no dia da transferência também pode fazer parte da rotina ou apenas em caso de suspeita de contaminação da receptora pelo embrião. As receptoras devem ser mantidas nas mesmas condições anteriores à transferência de modo a não provocar situações de stress.

É possível utilizar éguas em anestro como receptoras. Estas não deverão apresentar nenhum folículo de dimensão superior a 20 mm. Utiliza-se um protocolo em que é administrado estradiol, em dois dias consecutivos, e progesterona no terceiro (limitação na Europa). O dia da aplicação de progesterona será contado como dia 0 (correspondente ao dia de ovulação). Nestes casos, todas as éguas prenhas terão de ser suplementadas com progesterona até a placenta começar a produzir esta hormona, pois não terão a fonte primaria endógena de produção por ausência de corpo lúteo.

O sucesso de um programa de transferência de embriões depende de inúmeros fatores e deve ter-se em atenção todos os pormenores. A esterilidade e limpeza são de extrema importância para que a contaminação seja a menor possível.

É importante manter bons registos de todas as éguas, principalmente, quando se trabalha com um número elevado de animais.

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Equine Metabolic Syndrome

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Both human and equine metabolic syndromes are primarily created and maintained by chronically excessive caloric intake and physical inactivity with further important and related susceptibility factors also being involved. The major defining clinical consequence of EMS is laminitis although it is possible that further pathogenetic endpoints may become better recognised in the future. Thus, for the time being at least, EMS might be regarded as "A clinical syndrome associated with an increased risk of laminitis that includes insulin dysregulation and any combination of increased generalised or regional adiposit y , weight loss resistance, dyslipidemia, and altered adipokine concentrations".

Laminitis

When a middle-aged, obese, sedentary native pony is grazed simultaneously on the same pasture as a fit, lean, young Thoroughbred, then only the former is likely to develop laminitis; implying differing physiologic/metabolic/endocrine responses to the ingested nutrients. Recent studies have suggested that endocrinopathic mechanisms may be important predisposing factors in approximately 90% of clinical laminitis cases, a figure that at first glance might seem surprisingly high. However, for far longer than the recent discussions of endocrinopathic laminitis, veterinarians and horse owners have recognised that many laminitis-prone individuals share similar characteristics such as breed, age, obesity, a rich-diet and low-workload (table 1). Conversely they would also recognise that laminitis arising in the absence of these factors (e.g. a young, lean, fit, Thoroughbred) is relatively unusual (i.e. less than 10% of cases!).

Chronicity and recurrence are frequent features of EMS-related laminitis. Fundamentally there are probably 2 reasons why cases of laminitis might recur: firstly, that damage caused by previous disease episodes makes the foot inherently more susceptible to further laminitis attacks; and secondly, that the endogenous and exogenous risk factors associated with the previous laminitis episode (e.g. IR, obesity, excessive grazing) are still present. Indeed, a common question from the owner of a de novo laminitic animal is "Will this mean that my pony will always continue to suffer from laminitis?". This question can be answered by the famous quotation from Henry Ford: Q "What do you get if you always do what you have always done?"; A "What you always got!". It is obvious that recurrence should be expected in those cases where the fundamental causal factors are not subsequently managed and controlled effectively. It is therefore vital that laminitis cases encountered in practice are not simply treated and managed for laminitis in an isolated fashion, but that epidemiology is considered thoroughly so that identification and control of significant risk factors can be instigated. This comprises investigation of historical, clinical and clinicopathologic factors.

Obesity

Obesity has long been associated with laminitis-susceptibility and is a prominent epidemiologic feature of EMS and human metabolic syndrome. The precise links between obesity and laminitis are not fully understood but a causal association with IR is a likely candidate.

It has been estimated that around half of the horses in the UK are overweight. Obesity is broadly defined in a medical context as "excessive adiposity to an extent where health is negatively affected". Obesity is therefore primarily a functional and metabolic status, rather than simply a morphological and physical status, and may not always directly correlate with the size of adipose deposits. In contrast, in general colloquial parlance obesity may be defined entirely according to visual preconceptions that obese individuals will represent the extreme of excess bodily condition manifesting as a severe degree of adipose coverage over the entire body. Indeed, laminitis-prone individuals are often overtly and unequivocally morphologically obese with an absence of palpable ribs, large fatty deposits behind the shoulders and tail-head and a large firm crest. However, others may have palpable or even visible ribs leading some to describe them as "lean", although retaining a large cresty neck or perhaps other hidden fat deposits. The point at which an individual's health becomes compromised by the presence of adipose deposits (i.e. when obesity develops) is variable and dependent on the amount, the site and the metabolic activity of adipose tissue in that individual, emphasising the importance of "regional obesity". In human patients there is substantial evidence indicating that fat distribution is a better predictor of cardiovascular disease than overall fat volume. Uneven development and disappearance of regional adipose deposits in horses and ponies is also well described. Two published studies have specifically proposed clusters of risk factors defining EMS and both included markers of regional adiposity and also laboratory markers of the pathophysiologic consequences of adipose dyregulation (Table 1).

At least 3 of the following4:

- BCS ≥7 with localized fat deposits on neck and tailhead
- Reverse inverse square of insulin < 0.32 [mU/L]−0.5
- Modified insulin response to glucose > 5.6 mU insulin2/[10•L•mg glucose]
- Triglyceride concentration > 57.0 mg/dL

At least 3 of the following5:

- BCS ≥7
- CNS ≥4
- Insulin > 32 mU/L
- Leptin > 7.3 ng/mL

Table 1. Proposed working definitions of EMS. Laminitis susceptibility is defined when at least 3 of the 4 listed factors are identified4,5. BCS= Body Condition Score; CNS=Cresty Neck Score.

0	No visual appearance of a crest. No palpable crest
1	No visual appearance of a crest, but slight filling felt with palpation
2	Noticeable appearance of a crest, but fat deposited fairly evenly from poll to withers. Crest easily cupped in one hand and bent from side to side
3	Crest enlarged and thickened, so fat is deposited more heavily in middle of the neck than toward poll and withers, giving a mounded appearance. Crest fills cupped hand and begins losing side to side flexibility
4	Crest grossly enlarged and thickened, and can no longer be cupped in one hand or easily bent from side to side. Crest may have wrinkles/creases perpendicular to topline
5	Crest is so large it permanently droops to one side

Table 2. Cresty neck score.

Unfortunately the definition of excessive adiposity in an individual when seen by a practitioner is usually based on a subjective opinion, a situation that does not facilitate the conversion of sceptical horse owners who do not believe that their animal is obese. Body mass index (body mass/height2) has been described in horses as a possible objective index of adiposity, although has received little attention. The lack of widespread availability of weighbridges does not, in any case, encourage the use of any index relating to body mass.

Body condition scoring (BCS) is perhaps the most frequently used semi-objective index of obesity although generally still lacks widespread acceptance and usage in practice and may require tailoring for different breeds, ages, genders and activities. Lack of clarity and user-friendliness is not aided by confusing and poorly defined descriptive terminology that is hard to apply, requiring the user to distinguish fat deposits that feel "spongy", "soft", "very soft", "patchy" or "bulging". Nevertheless BCS has been shown to correlate well with markers of IR and laminitis susceptibility and one study found that BCS >6.8 was a good indication that total body fat content exceeded 20%. The "cresty-neck score" (CNS) is a semi-objective means of describing regional obesity with respect to the apparently particular associations between crest fat, IR and laminitis (Table 2). However, again breed, gender and perhaps age effects are also important with interpretation as one study found that associations between both BCS and CNS with hyperinsulinaemia was only significant for ponies and not for horses.

Practical and objective morphometric measures of both abdominal/trunk fat (girth:height ratio, waist:height ratio) and crest fat (crest height, neck circumference, neck circumference:height ratio) have been described by several studies and related to other markers of obesity, IR and laminitis. However, it has been found that these objective morphometric measures are generally less strongly correlated with body fat percentage and serum biochemical indices of IR than are the semi-objective BCS and CNS. Nevertheless, given the obvious attraction of objectivity, it has been suggested that certain cut-offs of morphometric measures could be used in ponies and horses to indicate overweight, obesity and crestiness (Table 4). Indeed in one large study of a particular pony herd, laminitis was predicted with reasonable accuracy in ponies with a girth:height ratio >1.30 and neck circumference:height ratio > 0.71

	Pony	Horse
Overweight	Girth:height ≥1.33	Girth:height ≥1.26
Obese	Girth:height ≥1.38	Girth:height ≥1.29
Cresty	0.5NC:height ≥0.68	0.5NC:height ≥0.63

Table 4. Suggested objective morphometric measurements indicative of excessive adiposity in horses and ponies (0.5NC = mid-neck circumference)

Dietary control of obesity and laminitis risk

Dietary control of the processes of EMS and increased laminitis risk is important in two broad fashions. Firstly, excessive adiposity can only be controlled by creating a relative dietary calorie deficit whereby energy expenditure exceeds intake. Secondly, restriction of dietary non-structural carbohydrates (NSC: simple sugars, starches and fructans) limits post-prandial hyperinsulinaemia which is recognised as a key trigger factor for laminitis. Dietary sugars and starches are likely to derive from concentrate feeds whereas grazing (in temperate climates at least) supplies primarily fructans and simple sugars. Tropical grasses can be starch-rich.

Thus dietary control for EMS and laminitis may be summarised by the restriction of both dietary calories and NSC, whilst ensuring continued provision of other dietary components such as proteins and micronutrients, and should perhaps be implemented gradually especially in hyperlipaemia-susceptible breeds. Efforts should be made to extend the feeding period as long as possible by using narrow-weave, double or triple haynets, centrally hanging rather than wall-hanging hay nets, etc...

Caloric requirements of individual obese animals cannot be predicted with any reasonable degree of accuracy due largely to inter-individual variability and the recently demonstrated spectrum of "weight-loss-sensitive" and "weight-loss-resistant" individuals which is not entirely breed dependent. In that study, the proportionate weight loss varied by more than 3 fold (0.16-0.55% body mass per week) amongst a group of horses despite identical dietary management. Furthermore it is unusual to be able to accurately calculate the exact digestible energy and more detailed nutrient analysis of an entire equine ration in practice. Thus an estimate of total required intake must inevitably be made and then followed by a monitoring protocol so that further changes and adjustments to the diet can be made as required.

Undoubtedly the most effective general means of achieving weight loss is when the carer has total control of exactly what and how much the horse or pony is eating. This is impossible when grazing is allowed, even for short periods and even with a grazing muzzle. Ponies are notorious in their ability to rapidly consume surprisingly large quantities of grass.

A diet with restricted calories, low NSC, high fibre and adequate protein and micronutrients is required. At its simplest, the diet must comprise 2 components:

1. The staple of the diet will be preserved forage, preferably hay. In native ponies and donkeys then a small amount of straw might be mixed with the hay although this should be done gradually and carefully to minimise the risk of impaction colic. Forage should be weighed dry and then soaked for at least an hour in warm water before feeding in order to remove some of the water soluble carbohydrates (WSC: simple sugars and fructans) from the forage and therefore reduce the glycaemic and insulinaemic effects of the diet.
2. Achievement of requirements with respect to protein and micronutrients is also essential. Providing an additional supplementary feed in addition to forage has the dual advantage of providing a vehicle for delivery of oral drugs and also allows balancing of the diet with respect to protein and micronutrients. Such additional feed can be formulated from several commercial sources by estimating protein and micronutrient requirements and mixing appropriate amounts of chaff-based feeds and feed-balancers (NB. proprietary label recommendations are not necessarily appropriate during dietary restriction programmes).

Evidence from recent studies suggests that in order to achieve weight loss in good-doing types within a reasonable time-frame, total daily dry matter intake will need to be restricted to between 1.0 and 1.5% bodyweight daily (approximately 1.2-1.7% weighed "as fed"). This typically represents approximately one half of voluntary feed intake and therefore represents, necessarily, quite a harsh diet.

Laboratory testing

Study of EMS cases has revealed several measurable plasma analytes that serve as markers for the dysmetabolism that culminates in increased laminitis susceptibility and predominantly reflect dysregulation of glucose and lipid metabolism. Although the exact pathophysiologic pathways leading to laminitis have not been elucidated, it is likely that some of these analytes may have direct pathologic relevance whilst others simply act as indirect markers of abnormal metabolic responsiveness. Several measurable plasma analytes associated with EMS are described further below.

a. Tests for dysregulation of glucose and insulin metabolism

As previously discussed insulin resistance (IR) and a tendency towards excessive postprandial hyperinsulinaemia are both frequent features of EMS. Hyperinsulinaemia and IR are clearly related to one another but are not synonymous. One or both may have logical evolutionary origins and may augment one another. IR may stimulate hyperinsulinaemia as part of a compensatory mechanism to overcome ineffectiveness of insulin as well as promoting a longer plasma half-life of secreted insulin. Conversely hyperinsulinaemia may promote IR via its anabolic effects on adiposity and possible receptor downregulation in the face of persistent high levels of stimulation. Whatever the precise interrelationship of IR and hyperinsulinaemia, it is clear that both indicate increased risk of laminitis. Hyperglycaemia, although relatively common in humans with metabolic syndrome, is relatively rare in EMS and its pathologic importance and relevance have been questioned.

Pathophysiologic studies of endocrinopathic laminitis suggest a more direct relevance for hyperinsulinaemia as it is clear that laminitis can be predictably triggered by hyperinsulinaemia in the absence of IR. However, the importance of IR may simply be due to indirect predisposition to hyperinsulinaemia or perhaps further undefined mechanisms. As obesity is probably the main predisposing factor for IR in horses, tests for IR may also act as useful laboratory markers for obesity.

Thus, in summary, tests for hyperinsulinaemia and tests for IR are probably both helpful in the monitoring of treatment success in cases of EMS. However, there is a difference in meaning and direct relevance of both types of test with hyperinsulinaemia possibly more directly relating to laminitis risk and IR being of indirect help in monitoring causal factors such as obesity.

i. Tests for hyperinsulinaemia

Perhaps the simplest useful test for laminitis susceptibility is measurement of insulin concentration. In addition to its simplicity, this test may be most pathogenically relevant given that it is likely to be hyperinsulinaemia rather than IR or hyperglycaemia that most probably triggers laminitis.

As discussed above, an important defining feature of EMS is that certain individuals, and not others, are likely to suffer laminitis following pasture ingestion. Several studies have indicated that laminitis-prone individuals characteristically demonstrate an excessive hyperinsulinaemic response to forage and/or orally adminstered NSCs, which may well represent a fundamental and crucially relevant metabolic/endocrine difference between normal horses and those predisposed to laminitis. In this respect the use of tests that examine the insulinaemic response to oral carbohydrate ingestion might appear even more attractive. In the USA, the "oral sugar test" (OST) has proved popular, whereas in the United Kingdom the "in-feed glucose test" is commonly used (tables 5 and 6). These two similar tests challenge the horse with corn syrup and glucose respectively and compare the insulinaemic response of the tested individual to that expected in a normal animal. It is assumed that an excessive insulinaemic response to the ingested carbohydrate represents a risk factor for laminitis. Comparison of 21 normal and 199 laminitis-prone individuals indeed revealed significantly greater insulin concentrations in the latter group at 2 hours following in-feed glucose (Figure 2).

Oral Sugar Test

- Fast overnight (allow 1 flake of hay)
- Dose with 15 mL Karo Light corn syrup per 100 kg BWT
- Measure serum insulin at 60-90 minutes post dosing
- Normal response < 60 mU/L insulin

Table 5. Outline procedure of the oral sugar test.

In-feed Glucose Test

- Overnight fast
- Give 0.5 or 1.0 g/kg BWT glucose or dextrose powder in a non-glycaemic feed (e.g. chaff)
- Measure serum insulin and plasma glucose after 2 hours
- Normal response:
  - 0.5 g/kg dose: 2 hour insulin < 57 mU/L
  - 1.0 g/kg dose: 2 hour insulin < 87 mU/L

Table 6. Outline procedure of the in-feed glucose test (AE Durham, unpublished data)

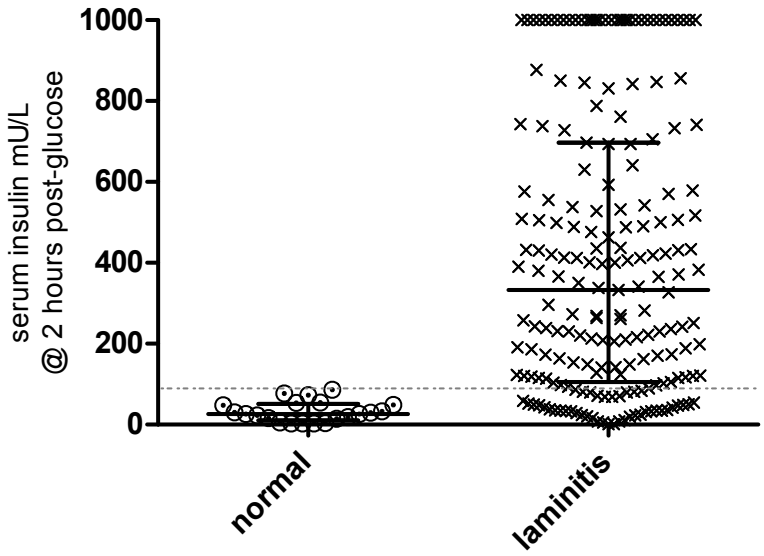

Figure 2. Serum insulin concentrations from 21 normal and 199 laminitis-prone horses and ponies subject to the in-feed glucose test 2 hours following administration of 1.0 g/kg dextrose. Dotted line represents cutoff for normal response. Median (interquartile ranges) are 26.3 (10.4-51.1) and 333 (105-697) mU/L respectively (P<0.001). Using a cutoff of 81 mU/L, 78% of laminitis-prone individuals demonstrated hyperinsulinaemia.

i.Tests for insulin resistance

The combined glucose insulin test (CGIT) (table 7) has been well described in the equine literature with the inference that, following intravenous administration of exogenous glucose and insulin, IR individuals demonstrate poorer correction of induced hyperinsulinaemia and hyperglycaemia (Figure 3).

- glucose is administered as 150mg/kg 50% dextrose iv,
- followed by insulin (0.1 U/kg) iv
- blood samples collected @ 1, 5, 15, 25, 35, 45, 60, 75, 90, 105, 120, 135, and 150 mins

Table 7. Protocol for combined glucose insulin test

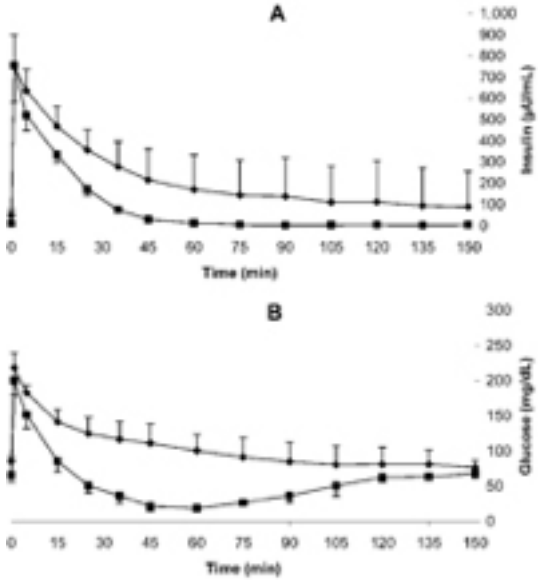

Figure 3. A. Insulin, and B. Glucose responses during the CGIT in normal (squares) and IR (circles) horses

In order to make the CGIT more acceptable in practice, a shortened test has been advocated comprising measurement of plasma glucose at 45 minutes (normal response ≤ baseline) and insulin at 75 minutes (normal response < 100 mU/L).

More recently a slightly simpler test for IR, the insulin response test (IRT), has been described comprising intravenous insulin challenge only and dispensing with the need for exogenous glucose in the CGIT (Table 8, figure 4).

- 0.1 U/kg insulin iv
- blood samples collected @ 0, 20, 40, 60, 90, 120, 180, and 240 min

Table 8. Protocol for the insulin response test

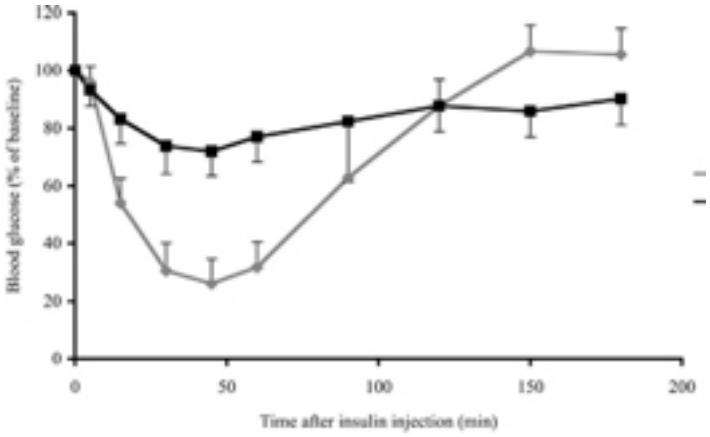

Figure 4. Glucose responses during the IRT in normal (circles) and IR (squares) horses

Again the IRT has a curtailed format comprising testing at baseline and 30 minutes only. A normal response is expected to demonstrate a >50% decrease in baseline glucose at 30 minutes.

a. Tests for dysregulation of lipid metabolism

Obese, insulin resistant and laminitis-prone horses have been shown to have higher than normal plasma triglyceride concentrations, NEFAs, VLDL and HDL-cholesterol.

Recent interest is growing in the pathophysiologic relevance and diagnostic usefulness of adipokines in obese horses. Adipokines are a large and diverse family of proteins important in the pathophysiology of conditions associated with obesity and are synthesised and secreted by adipocytes, macrophages and other cells present in adipose tissue. Several adipokines have been identified and studied in other species including leptin, adiponectin, resistin, visfatin, apelin, chimerin, retinol binding protein 4 as well as inflammatory cytokines released both by adipocytes and inflammatory cells within the fat such as TNF-alpha, IL-6, MCP1 (CCP-2) and IL-1.

Leptin acts to suppress appetite centrally and improve insulin sensitivity peripherally. Obesity is paradoxically associated with hyperleptinaemia as this is a result of selective leptin resistance. Several studies in horses have revealed leptin to be greater in obese horses and also to be predictive of laminitis when > 7.3 ng/mL. Adiponectin is a further adipokine peptide which is one of the most highly expressed products in adipose tissue. It is found as a trimer, hexamer, or high molecular weight (HMW) multimer, with the latter being most biologically active. Compared to other adipokines, adiponectin appears to have beneficial metabolic, vascular, insulin-sensitising and anti-inflammatory effects and it is decreased in both obesity and IR in horses. It appears that although leptin might reflect obesity, adiponectin might be a better predictor of the functional effects of obesity (i.e. insulin resistance). Obese, insulin resistant horses have abnormally low plasma levels of adiponectin and this might add more information to the simple observation of body condition score or other morphometrics.

Medical aids in EMS management

Pharmacologic treatment of EMS might also be considered but should never be regarded as a substitute for managemental countermeasures. There is a danger that, given the practical difficulties of achieving weight loss in extremely metabolically efficient individuals, that some owners might regard pharmacologic assistance as an easier alternative to dietary and exercise control. This suggestion should be firmly repudiated in preliminary discussions of

the proposed EMS-management plan as sole reliance on pharmaceutical interventions is highly likely to fail.
Thus far the only pharmacologic agents have received significant attention in EMS cases comprise L-thyroxine and metformin hydrochloride.

L-Thyroxine (levothyroxine)

Early mistaken suspicions that obese, lethargic, laminitis-prone individuals were clinically hypothyroid, first led to the use of L-thyroxine (e.g. Thyro L, Lloyd Inc, Shenandoah, Iowa; Soloxine, Virbac Ltd, Bury St Edmunds, Suffolk) for the intention of replacement therapy in such cases. Although it is now accepted that hypothyroidism is an extremely rare occurrence in horses that does not play a role in laminitis-susceptibility, evidence suggests that exogenous thyroid hormone may nevertheless have beneficial effects in obese laminitis-prone subjects. One study found that mares typically lost approximately 0.5% bodyweight weekly when treated with L-thyroxine on an increasing dosage regime from 0.05-0.20 mg/kg daily over 8 weeks. In a further more prolonged study by the same group, daily administration of 0.10 mg/kg L-thyroxine was associated with approximately 0.6% bodyweight loss per week over the course of 16 weeks although weight loss did not continue with longer term treatment for up to 48 weeks. It was suggested that L-thyroxine leads to lipolysis and mobilisation of adipose stores due to an increased basal metabolic rate. In association with the weight loss, serum insulin concentrations were seen to decrease slightly and insulin sensitivity approximately doubled during treatment. Significant adverse health effects of exogenous L-thyroxine were not seen although minor cardiac changes are described. Use of L-thyroxine in EMS cases is recommended alongside dietary restriction, both to augment the effect on weight loss and also to guard against the possible adverse effects of increased appetite during treatment. Typically a dose of approximately 0.1 mg/kg orally q 24 hours is recommended for between 3 and 6 months depending on response in body condition. At the end of the treatment period the drug should be gradually withdrawn over 2-4 weeks to allow reestablishment of normal responsiveness of the pituitary-thyroid axis. Outside of the USA, use of L-thyroxine is greatly limited by cost.

Metformin hydrochloride

A study of laminitis-prone horses and ponies reported a significant decrease in plasma glucose and insulin within 2 weeks of beginning metformin therapy at 15 mg/kg q 12 hours. However, subsequent studies indicated very poor oral bioavailability of metformin in horses of only 7% following fasting and 4% in the fed state, compared with 50-60% in humans and rats. Further studies found no detectable effect of metformin on peripheral insulin sensitivity in both normal horses and insulin resistant ponies at 15 mg/kg, casting serious doubt on the existence of any real clinical benefits of the drug in EMS cases.

However, all of the findings in the above equine studies may be reconciled by hypothesising that metformin in horses may have negligible systemic effects due to poor bioavailability, but may well maintain significant direct enteric effects as described in other species. A further study has recently confirmed significant moderation of hyperglycaemia and hyperinsulinaemia in horses when metformin was administered at a dose of 30 mg/kg prior to oral glucose dosing also consistent with the findings in other species (figure 5). Thus there is accumulating evidence that metformin decreases enteric glucose absorption and subsequent insulinaemia in horses as well as other species.

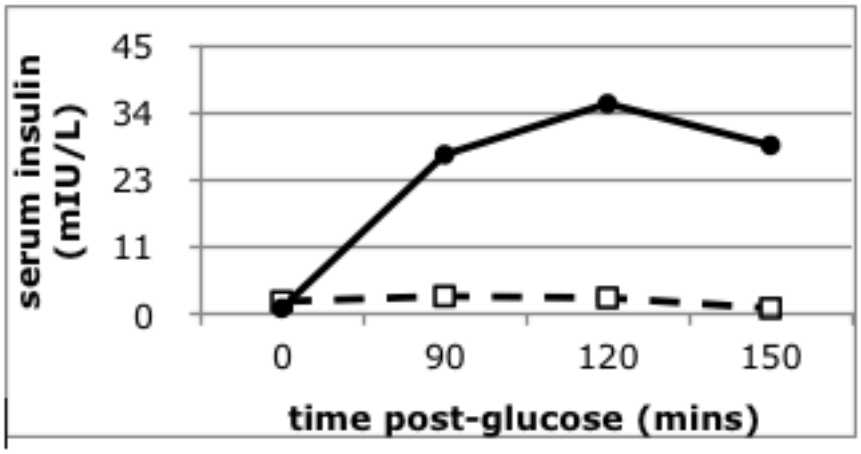

Figure 5. Insulin responses to an in feed glucose dose with (dashed lines) and without (solid lines) pre-treatment with metformin

It is relevant that current suspicions of the apparent pathogenetic links between insulin resistance and laminitis focus on diet-induced hyperinsulinaemia rather than tissue insulin insensitivity per se. In this respect, the lack of any detectable insulin sensitising effect of metformin in horses, yet the demonstration of moderation of hyperglycaemia and hyperinsulinaemia serves to maintain clinical interest in the drug. However, if the effect of metformin is to decrease post-prandial hyperglycaemia and hyperinsulinaemia then its use in subjects where diet is controlled as per recommendations (i.e. low NSC and mildly glycaemic/insulinaemic) is questionable. The drug might be best applied in insulin resistant cases that are still ingesting significant NSC from grazing, for example PPID cases or EMS cases that are being allowed some limited grazing following on from an effective weight loss and exercise control programme. The moderation of glucose absorption might also be expected to assist weight control in such individuals. Rather than straightforward twice-daily dosing, it might be wise to target drug administration pre-turnout to limit the subsequent glycaemic and insulinaemic effect of grazing. It should also be stated however, that any putative clinical benefits of metformin in horses are still based largely in anecdote although one study commented that laminitis was improved in 14/18 (78%) treated subjects. Further studies are clearly required.

Tim Greet

The emergency management of acute trauma in competition horses

Rossdales Equine Hospital, Newmarket - UK

acute trauma in competition horses

- fractures and dislocations
- other severe orthopaedic injuries
- wounds (open synovial structures)
- facial and respiratory trauma
- recumbency
- cranial trauma with neurological complications

fractures and dislocations

- acute management of these cases is vital
- protection and support of the injured limb and other legs
- effective and safe transport of the case to a suitable place for more detailed assessment
- careful evaluation of the nature of the injury (radiography, ultrasound, scintigraphy, ct, mri)
- repair (if possible and desirable)

acute assessment of the patient

- first look
- first aid
- move from site of accident
- more detailed evaluation
- decision making regarding treatment

initial considerations

- what is the most humane approach regarding treatment or euthanasia?
- informed consent of the owner?
- is the horse insured and what type of policy?
- what are the economic constraints?
- is moving the horse really in its best interests?
- to where can it be safely moved?
- can it be moved in the best condition to ensure the optimal outcome?

"first look"

- immediate euthanasia?
- clear cut decision?
- may still be preferable to move from site of incident to allow a more accurate evaluation and prognosis

euthanasia more appropriate?

- a few injuries are so clear cut that immediate euthanasia is the only humane course of action to prevent unnecessary suffering
- better to move horse from scene of accident to allow more objective assessment of injury

stabilise patient before loading

- support limb(s)?
- sedation?
- analgesics (nsaids)?
- antibiotics?
- someone to travel with the horse?

first aid (injured limb support)

- robert jones dressing - use of splints (Walmsley 1993 EVE)
- proprietary splint
- whatever comes to hand!

applying a robert jones bandage

- "soffban"
- cotton wool
- conforming bandage
- multiple layers
- "cylinder" leg (at least 3 times size of normal limb)
- "elastoplast" for tension
- incorporate splint (duct tape)

additional measures

- analgesics (judicious- remember pain is protective)
- sedation?
- support other limbs
- antibiotics (open fracture?)
- transport to assessment centre

if the horse is transported a long distance

- extra importance of protection and support of injured limb and the other limbs
- greater value in having someone travelling with horse
- need for additional analgesic or sedative medication?
- need for antibiotic cover?
- hydration status? (Barbaro)

receiving the emergency patient (in the hospital)

- receiving "team" alerted and prepared (out of hours)
- specialised ramp for unloading patient ?
- horse unloaded directly at the examination area?
- radiography team prepared
- theatre and surgical team prepared?

fracture evaluation

- multiple radiographic images
- special projections?
- fracture patient limb "wobble"
- keep exposure time short

magnetic resonance imaging of fractures

computed tomographic imaging of fractures

- probably more useful for imaging complex fractures
- our system requires a ga for the limbs of adult horses
- ability to reconstruct impressive 3 d images
- could be of great value for pre-operative planning for a repair

common injuries

(non-racehorse)

- tend to be due to direct trauma rather than "repetitive strain" injuries commonly seen in racehorses
- stifle injuries common but almost all bones or joints can sustain damage
- many are responsive to surgical treatment

joint luxation

common severe injuries in racehorse

- sagittal fractures (mc3; p1)
- major carpal fractures
- major long bone fracture
- pelvic fracture
- luxation SDFT
- superficial digital flexor tendon rupture

severe wounds

- trunk and abdomen
- proximal and distal limb
- involving synovial structures
- eyelids, nostrils and lips
- other facial sites

wound assessment

- wound may be the least important component of the injury
- always examine the whole animal
- attend to critical issues first airway, breathing, circulation)
- consider wound only when patient stable

endoscopic debridement and copious synovial lavage

- gold standard for managing synovial infections
- allows a thorough inspection of the intrasynovial environment and identification of otherwsie undetectable injuries
- easy to remove inflammatory debris and wide bore cannula allows copious lavage
- removal of any fracture fragments or foreign material

eye injuries

- tend to be very painful
- if global rupture - enucleation
- corneal injuries may be repaired
- subpalpebral catheterisation can be very useful

severe head lacerations

- bleed profusely
- often look frightening to owners or riders
- almost always heal very well (good blood supply)

respiratory trauma

- dyspnoeic horses must be evaluated rapidly
- oxygen support?
- emergency tracheotomy?
- endoscopy and radiography valuable

chest injuries

- is the horse dyspnoeic?
- is there a pneumothorax?
- thoracic ulatrsonography and radiography (rib fracture?)
- emergency pleural decompression (bilateral?)
- intrapleural haemorrhage? (pleural drainage?)

severe facial injury

- respiratory obstruction?
- emergency tracheotomy?
- epistaxis?
- facial fracture / are other structures involved eg tmj, nasal septum, hard palate?
- open sinus?
- facial stability

recumbent horse

- always a very stressful situation!
- basically two sorts of recumbent horses
- those that recover with time and support
- those that end up dead or being euthanasia

major cranial trauma with neurological complications

- usually falling over a fence
- severe cases usually recumbent
- bleeding from ears ± nose
- careful clinical and neurological assessment
- if horse can stand, can it be moved safely?
- if horse can be moved radiographic evaluation of skull
- most cases euthanased

Novas Áreas da Medicina Veterinária

Anabela Moreira

Animais de Companhia Suspeita de mau trato! E agora

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No exercício da sua actividade profissional os médicos veterinários poderão ser confrontados com casos declarados ou suspeitos de mau trato ou de abandono. No quadro normativo português, é dado actualmente aos animais de companhia um particular destaque, através de um capítulo específico a eles dedicado no Código Penal. O novo enquadramento jurídico, desejado e necessário, carreia novos desafios a todos os agentes envolvidos e em particular aos Médicos Veterinários, uma vez que apenas a eles poderá ser atribuída, oficialmente e com valor jurídico, a avaliação médico-legal do animal sujeito a ou suspeito de mau trato.

Face a este panorama, que não é inédito apenas em Portugal, e na ausência de uma estrutura e procedimentos padrão similares aos existentes no ramo humano da Medicina Legal, é necessário que a comunidade médico veterinária adquira conhecimento de como actuar em situações que sejam ou possam ser do foro criminal, de modo a contribuir de forma relevante para a correcta elaboração dos processos. É ainda desejável que, apesar da ausência da supra citada estrutura que funcione como entidade responsável pela elaboração de manuais de procedimentos, estes possam ser de alguma forma normalizados de modo a evitar assimetrias, nomeadamente no que respeita à actuação em casos suspeitos de crime.

Além de todas as naturais dúvidas e incertezas que o novo enquadramento legal suscita, nomeadamente no que respeita aos conceitos de animal de companhia e de mau trato, acrescem ainda outras questões relacionadas quer com as funções do Médico Veterinário quer com a natureza dos próprios eventos. Assim o Médico Veterinário com funções de autoridade sanitária poderá ser colocado perante eventos que menos frequentemente acontecem na prática privada. Por outro lado não é raro que um evento sinalizado como suspeito de crime de maus tratos envolva um número de animais bastante elevado que, em muitos casos, ultrapassa a capacidade de resposta local em recursos humanos, técnicos, logísticos e financeiros.

Independentemente do número de animais envolvidos, que nem sempre são da mesma espécie, é necessário ter sempre presente que cada animal é uma vítima e deve ser considerado como um caso isolado para efeitos de investigação devendo a sua individualidade ser respeitada, nunca esquecendo que também é uma prova da prática do ilícito criminal. No entanto esta é uma "prova" que, felizmente em muitos dos casos, está viva e que naturalmente será diferente a cada momento que seja observada, pelo que se impõem a fixação de todas as informações não só no momento da primeira intervenção como no seguimento. A colheita exaustiva de informações, a tomada de imagens estáticas ou vídeo e a elaboração de um relatório médico transformam-se assim numa ferramenta essencial para que não só um processo seja correctamente instruído como que seja facilmente percebido pelas autoridades os factos e implicações do evento, no que respeita os aspectos médico veterinários.

Se a suspeição de mau trato de um animal ocorrer durante uma consulta num CAMV a equipa deverá estar familiarizada com os procedimentos legais de modo a que, independentemente do curso do caso, possa, se necessário, prestar auxílio às autoridades. Árvores de decisão, conhecimento dos pontos que são aceites internacionalmente

como sinalizadores de maus tratos, existência de formulários de modo a poderem ser realizados registos minuciosos são alguns aspectos que, se disponíveis, podem ser de grande ajuda no momento em que o Médico Veterinário é colocado perante um caso suspeito.

Quando em presença de um evento onde o número de animais e/ou espécies seja apreciável, muitas vezes há a necessidade de reunir condições para uma intervenção, sendo estes casos mais frequentemente apresentados aos Médicos Veterinários que desempenham funções oficiais. A existência prévia de planos de contingência, que muitas vezes requerem logísticas semelhantes às actuações em situações de desastres e catástrofes, é desejável diminuindo consideravelmente falhas que muitas vezes apenas são detectadas quando da elaboração final do inquérito judicial.

Infelizmente nem sempre as vítimas são encontradas vivas ou conseguem sobreviver aos danos e nesse caso o animal, ainda que cadáver, continua a ser uma prova e como tal deve ser submetido a uma avaliação, que neste caso se consubstancia na necrópsia forense.
Como nota final importa referir que a articulação entre as várias entidades envolvidas, nomeadamente Médico Veterinário, Autoridades e Ministério Público é não só desejável como imprescindível para que todo o processo possa atingir os seus objectivos.

Anabela Moreira

"Traduzindo da Medicina para o Direito: importância e particularidades de um relatório médico-veterinário em contexto forense"

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Harmonizando as várias definições de Medicina Legal, é aceite o conceito generalista de que ela é a utilização e aplicação dos conhecimentos da Medicina em questões de Direito. Assim, aos Médicos Veterinários envolvidos em casos, declarados ou suspeitos, de mau trato ou abandono será inevitavelmente pedido a produção de um relatório médico de avaliação do estado actual ou uma prova pericial, que contribua para o esclarecimento dos factos. Ainda que não prescindindo da linguagem médica, técnica e científica, o relatório médico-veterinário ou a prova pericial deverá ser estruturada e elaborada de modo não só a ser percebida pelas autoridades judiciais como a suportar, ou não, os factos ou indícios constantes do processo.

O relatório médico veterinário é de extraordinária importância, pois desejavelmente será a peça fundamental para que o Ministério Público prossiga para além da fase de inquérito, sendo muitas vezes também a base para estipular a pena.

Embora genericamente se fale em relatório Médico Legal, este pode ter diversas abordagens de realização: pode ser um Relatório Clínico ou um Relatório de Necrópsia, para animais vivos ou cadáveres respectivamente, podendo no mesmo processo e para o mesmo animal coexistir ambos se o animal sucumbiu posteriormente aos danos infligidos ou foi realizada eutanásia. Pode ainda ser considerado como um relatório preliminar, quando a informação veiculada é importante para uma tomada urgente de decisão embora haja ainda informação não disponível como por exemplo resultados de exames complementares, ou já ser um relatório final onde todas as informações estão plasmadas, considerando-se que um relatório preliminar dará origem sempre a um final. Pode ainda o Médico Veterinário, ser instado a pronunciar-se acerca de factos e informação existente, recolhidos ou não por si, fornecendo a sua opinião. Em qualquer dos casos o documento resultante deve ser estruturado logicamente e ser completo.

Não existe um modelo único para a elaboração de relatórios, até porque cada caso poderá apresentar particularidades diferentes e raras vezes são exactamente iguais. Para facilitar a estruturação lógica de um relatório a existência de diferentes secções pode ser um valioso auxílio, mas não é indispensável que seja apresentado desse modo, desde que a informação pertinente seja transmitida.

A identificação do caso, a entidade requisitante do exame e a data da sua realização, a identificação completa do Médico Veterinário bem como a identificação/caracterização pormenorizada do animal são imprescindíveis. Detalhes do exame externo e/ou interno, no caso de necrópsia, evidências de lesão bem como a lista de todos os procedimentos médicos, cirúrgicos, amostras colhidas e resultados têm que estar incluídos no relatório, onde também deve constar, se disponível, a história clínica anterior. Quando a informação disponível é vasta é aconselhável fazer um sumário após a descrição detalhada. No caso de cadáveres uma estimativa do tempo de sobrevivência após lesão, do intervalo post mortem, do mecanismo, causa e etiologia da morte e concausas, se as houver, são também

informações da maior importância. Para além da descrição detalhada e de um eventual sumário, o relatório deve terminar com uma conclusão acerca da avaliação efectuada interpretando e dando opinião sobre os factos médicos abstendo-se de qualquer juízos de valor sobre o caso. Este trecho acabará por ser da maior relevância para o processo judicial, devendo ser elaborado numa linguagem facilmente percebida por profissionais não médicos.

Um aspecto transversal a ter sempre presente na elaboração de um relatório médico-veterinário num contexto forense é que a ausência de uma determinada evidência pode ter exactamente a mesma importância que a sua presença, pelo que deve ficar registado não apenas o que se encontra mas também o que está ausente.

Mariana Portugal

Medidas de controlo de doenças em ambiente de CRO

A gestão e o controlo de doenças infeciosas em cães e gatos continua a ser um dos maiores desafios enfrentados pelos centros de recolha. Considerando que num centro de recolha oficial, temos um ambiente onde ficam confinados muitos animais com antecedentes médicos e históricos de exposição desconhecidos, o controlo e a prevenção de doenças infeciosas que "entram" num centro de recolha é difícil.

Embora o risco de introdução da doença não possa ser erradicado, existem algumas estratégias sistemáticas para a minimização da transmissão de infeções. Entre elas, podemos falar na redução da gestão da população e na redução do stresse; no controlo profilático de todos os cães e gatos, na limpeza e desinfeção eficaz, na separação de jovens e adultos; no diagnóstico atempado e na segregação de animais infetados e vigilância de animais expostos. Ainda assim, o desenvolvimento destas estratégias é dependente do conhecimento do possível agente patogénico em causa e dos fatores que contribuem para o risco de transmissão. A interação sinérgica entre o hospedeiro, o microrganismo e os fatores de transmissão, determina o risco de infeção.

Assim, para o controlo de infeções, importa atuar em 3 frentes diferentes: na fonte de infeção, na via de transmissão e no hospedeiro susceptível. A prevenção de infeções em estabelecimentos onde se prestam cuidados de saúde deve ser direccionada primariamente para a interrupção da fonte até ao hospedeiro, visto que os factores relacionados com o agente e com o hospedeiro são mais difíceis de controlar.

As respostas à doença, em geral, e à doença infeciosa, em particular, são uma parte integral e indissociável de qualquer programa de saúde de um centro de recolha. Um plano de resposta deve incluir medidas para minimizar a transmissão para animais não afetados e pessoas, assegurando também o cuidado apropriado aos animais afetados. Devido à ampla variedade de agentes patogénicos, aos diferentes modos de transmissão, e às especificidades de cada centro de recolha, não existe uma abordagem única para todas as circunstâncias.

No geral, a avaliação de agentes patogénicos específicos, em centros de recolha oficiais, é uma ferramenta de difícil aplicação, devido aos fracos recursos, humanos, monetários e técnicos. Assim, os métodos de prevenção de doenças são muitas vezes inespecíficos, incluindo a desinfeção profilática do ambiente e isolamento, tratamento ou eutanásia dos animais doentes.

Instalações e superfícies inadequadas ou projetos de CRO mal concebidos podem contribuir para a transmissão de agentes patogénicos. Quanto maior o número de áreas distintas, maiores possibilidades de isolamento de animais e consequentemente minimizar a transmissão de doenças por contacto direto.

A utilização adequada do verdadeiro isolamento é crítica para limitar a propagação da doença. Idealmente, uma área de isolamento deve ser usada apenas para essa finalidade, de acesso restrito e deveria ser localizada de modo a que a movimentação destes animais seja feita sem expor os animais saudáveis.

Considerando que na maioria dos centros de recolha, o número de trabalhadores é limitado e que os mesmos terão que executar outras funções para além do isolamento, é fundamental que o pessoal entenda a natureza de uma doença infeciosa e tenha instruções claras, de modo a minimizar o potencial de transmissão destas doenças.
A observação diária dos animais alojados é outra prática vital, a fim de reconhecer as alterações na condição de saúde dos animais que requeiram atenção ou resposta imediata.

Os programas de controlo de doenças que focam exclusivamente a melhoria do sistema imunitário dos animais, através de beneficiações a nível da nutrição, vacinação, desparasitação e tratamento, acabam por falhar se todas as vias de transmissão da doença não são salvaguardadas. Concretamente, os médicos veterinários que praticam "medicina de abrigos" devem prestar toda a atenção aos detalhes, nomeadamente à localização e manutenção da ventilação, seja natural ou artificial, à limpeza e desinfeção, à desratização e desinfestação, ao armazenamento da alimentação, à higienização do equipamento de proteção individual, entre muitas outros fatores que podem interferir, direta ou indiretamente, no controlo da transmissão de doenças.

No contexto de um centro de recolha de animais, a minimização do stresse tem o potencial de melhorar consideravelmente o bem-estar animal, diminuir as taxas de infeções e transmissão de doenças, e consequentemente aumentar a "adotabilidade" dos animais.

O alojamento em centros de recolha apresenta múltiplos fatores stressantes, que podem incluir: captura, transporte, sobrelotação, isolamento, doença, alterações na temperatura ambiental, alterações no fotoperíodo e na ventilação, odores estranhos, ruído, outros animais, alterações alimentares, maneio e contenção, diferentes rotinas diárias, diferentes tratadores, manipulações diárias inesperadas, ausência de contacto humano familiar e presença de contacto humano desconhecido. Concretamente, qualquer situação ou elemento não familiar pode provocar apreensão e acionar uma resposta ao stresse.

Com o aumento da estadia, torna-se progressivamente mais importante providenciar espaço para a estimulação mental e física, pelo que deve haver alternativas ao alojamento primário. Nos alojamentos de longa duração, o ambiente físico deve incluir oportunidades de esconderijos, jogos e brincadeiras, alimentação, micção e defecação. Nos gatos, também deverão existir locais ou estruturas para arranhar, subir/trepar, e empoleirar-se. Os espaços exteriores devem ser protegidos das condições climatéricas adversas, do vandalismo e da possibilidade de fuga ou de atitudes predatórias. A estadia prolongada de um animal é um fator de risco para o aparecimento de doenças infeciosas e comportamentais, tornando-o menos "viável" para adoção.

Provavelmente, o mais eficaz enriquecimento ambiental é quando o pessoal gosta de trabalhar com animais e quer, e pode, passar tempo de qualidade interagindo diariamente com os animais, assegurando contacto social e docilidade.

Uma nutrição adequada e o exercício têm profundas implicações no bem-estar. Não só são essenciais para a manutenção do peso e da condição corporal adequados, como uma boa nutrição desempenha um importante papel no suporte da função imune e o exercício regular está associado à saúde e bem-estar comportamental. Uma nutrição deficiente, especialmente em animais cuja saúde está já comprometida, pode contribuir para aumentar a suscetibilidade às doenças e a disseminação das mesmas.

A gestão eficaz da população é a estratégia mais importante para a redução da transmissão de doenças infeciosas em centros de recolha de animais, que afeta a eficácia de todas as outras, incluindo a redução do stresse, a resposta à vacinação, alojamentos separados, limpeza e desinfeção, o reconhecimento imediato dos animais doentes, e espaço para isolamento e quarentena.

A sobrelotação é, sem dúvida, o grande fator de risco para surtos de doenças infeciosas. O aumento da densidade populacional leva a um maior risco de introdução da doença, taxas de contato mais elevadas para a transmissão da doença, aumento das doses infetantes no ambiente, ventilação reduzida e fraca qualidade do ar.

O objetivo da gestão da população passa por limitar ou adequar o número de cães e gatos em relação à respetiva capacidade de alojamento e as decisões devem ser tomadas no sentido de controlar a sobrepopulação o mais rapidamente possível, incluindo a suspensão da entrada de animais, ou eventualmente promovendo a transferência para outros locais de acolhimento. No caso de sobrelotações frequentes, torna-se necessário rever e alterar as políticas, reduzir as admissões e/ou alargar as instalações.

Infelizmente, o número de lares que procuram um animal de companhia, sem olhar à raça, à beleza, ao porte, é insuficiente para o número de animais que aguardam um novo detentor. Todavia, todos devem ser respeitados e neste prisma, os médicos veterinários que exercem funções em centros de recolha de animais, no âmbito da gestão da saúde animal, física e mental, têm um papel preponderante, não só pela sua atuação direta e pelo exemplo que representam, como também pela possibilidade que têm em poder transmitir os seus conhecimentos, de modo a que também o pessoal, que com eles trabalha, possa enveredar pelo mesmo caminho.

As diversas competências de um médico veterinário municipal ou de um médico veterinário ao serviço da autarquia, extravasam todo o trabalho desenvolvido num centro de recolha, e não raras vezes, impedem a sua constante permanência e supervisão, sendo esta também uma importante razão para que a formação do pessoal seja feita regularmente.

As limitações inerentes a um centro de recolha são, por norma, exclusivas e devem ser compreendidas de modo a estabelecer objetivos realistas. O influxo constante de animais, de desconhecidas proveniências e estatutos sanitários, a constante movimentação de visitantes e as políticas de controlo de animais errantes e políticas legislativas que nem sempre têm como primeiro objetivo a prevenção da transmissão de doenças, tornam o assunto complicado de gerir, principalmente quando está em causa o surto de uma doença. Adicionalmente, as restrições orçamentais, frequentemente as instalações mal projetadas e a análise dos meios de comunicação também são pontos a ter em conta nas tomadas de decisão.

Alguns centros de recolha enfrentam estes desafios melhor do que os outros. Em instalações onde os cuidados com os animais não são os ideais, muitas vezes o problema é a formação de pessoal, por vezes inadequada, insuficiente ou limitada pela elevada rotatividade. Para um atendimento de qualidade é preciso ter em conta os diversos detalhes que requerem uma atenção cuidadosa, bom treino e boa formação, uma equipa com experiência e motivação e procedimentos padrão operacionais adequados e praticáveis.

A "medicina de abrigos" ou "shelter medicine" é uma área de especialidade que tem crescido no âmbito da medicina veterinária. A gestão geral da saúde de explorações de animais de espécie pecuária é tema tradicionalmente abordado em faculdades de medicina veterinária, no entanto, raramente é discutido, por rotina, em ambiente académico, a gestão da saúde de uma população (confinada) de animais de companhia, concretamente de cães e gatos.
A medicina veterinária praticada num centro de recolha não é simplesmente a prática da medicina veterinária de pequenos animais num centro de recolha, isto é, nos centros de recolha, o objetivo é prevenir a doença, em vez de a tratar. Os CRO não são hospitais: o tratamento é demorado, habitualmente dispendioso e muitas vezes resulta em dor e sofrimento prolongado do animal. O maneio de uma população de cães e gatos prevalece sempre relativamente ao maneio de um animal em particular e a prevenção é menos onerosa, mais fácil e melhor para os animais.

Mariana Portugal

Estratégias de higienização e desinfeção de instalações de CRO

A frequência da ocorrência de doenças e a sua gravidade estão diretamente relacionadas com o nível de contaminação ambiental e esse, por sua vez, depende do sistema de maneio e do plano de limpeza e desinfeção (PLD) das instalações.

A limpeza e a desinfeção são uma importante parte de qualquer plano de controlo de infeções. Quando os planos são implementados apropriadamente, a introdução ou a disseminação de organismos patogénicos podem, na maior parte dos casos, ser minimizadas ou mesmo prevenidas.

Distingue-se claramente a limpeza e a desinfeção como dois processos distintos, em que a limpeza envolve a remoção de matéria orgânica visível com sabão ou detergente, enquanto que a desinfeção envolve a aplicação de um químico, ou outro procedimento, com o objetivo de eliminar os microorganismos remanescentes que não podem ser adequadamente removidos através da limpeza, a quel é essencial porque o tempo de sobrevivência de muitos agentes infecciosos é prolongado pela presença de matéria orgânica, e a matéria orgânica também diminui a eficácia dos desinfetantes. Dependendo do nível de desinfeção usada, a desinfeção elimina ou previne o crescimento de muitos ou da maioria dos patogénios.

Apesar de serem habitualmente realizadas desinfeções num centro de recolha oficial, são demasiadas vezes efetuadas com químicos ou métodos que não revelam a sua ótima eficácia, o que acaba por criar uma falsa sensação de segurança e pode mesmo traduzir-se na continuação da disseminação de agentes patogénicos, não obstante os esforços realizados para os controlar.

Assim torna-se necessário um PLD eficaz, seguro e prático, que deve incluir os métodos específicos e os agentes para atingir os objetivos da limpeza e desinfeção.

Independentemente do tipo de construções, o objetivo de um PLD é manter uma concentração baixa de agentes patogénicos, diminuindo, desta forma, a probabilidade de infeção.

Mas não há um programa padrão de desinfeção para uso por todos os CROs, porque no seu planeamento, há muitos fatores a ponderar, designadamente o tipo de superfícies e volumes a limpar, os utensílios necessários e os existentes, as condições estruturais, as características da população animal alojada, os agentes infecciosos mais frequentes, o custo dos detergentes e desinfetantes e orçamento disponível, a seleção dos desinfetantes, a diluição e tempo de atuação dos desinfetantes seleccionados, a frequência de desinfeção, o treino e a formação dos pessoal responsável pelas higienizações, a segurança do pessoal e dos animais, os recursos humanos disponíveis para executar o PLD e a capacidade para cumprir o plano.

Como estratégias de permitam uma boa implementação, segurança e eficácia do PLD, os materiais e equipamentos

de limpeza, os detergentes e os desinfetantes, as dosagens e as recomendações de uso, assim como os equipamentos de proteção individual, devem estar prontamente disponíveis, facilmente acessíveis e bem compreendidos pelo pessoal.

A formação do pessoal responsável pelas higienizações sobre os métodos adequados, salvaguardando a saúde e o bem-estar animal, deve abranger todas as áreas do CRO, nomeadamente canis com recinto exterior ou canis interiores ou canis bicompartimentados com ou sem porta de separação, gatis (método limpeza seca, método passar-o-gato, método "gato-na-caixa"), veículos de transporte de animais, equipamento móvel, recintos ou parques exteriores, enfermarias, salas de higienização de animais, postos de profilaxia, salas de campanhas de esterilização (quando existentes) ou outras zonas existentes. Realça-se que a atenção que cada assistente operacional do centro de recolha oficial dá aos pormenores, acaba por se expressar em diferenças abismais na "saúde" do centro de recolha.

A seleção adequada do desinfetante é também um ponto fundamental para o sucesso do PLD. Neste âmbito, há variados aspetos a ter-se em conta, designadamente o espetro de atividade, os agentes patogénicos potenciais, a possivel inativação pela matéria orgânica, a toxicidade para pessoal e animais, a corrosividade, o tempo de contacto requerido, a atividade residual, os efeitos ambientais, o custo. Também são fatores a considerar, na escolha do mais conveniente, as superfícies a desinfetar, o ambiente envolvente, a casuística e as práticas de rotina em uso.

Um desinfetante ideal deve possuir um amplo espectro, não sofrer alterações na sua ação na presença de matéria orgânica, água dura ou detergente, luz, ter atividade potente, ter ação rápida e duradoura e com poder remanescente, persistindo quando diluído, ser económico, ser inodoro, ter uma capacidade de conservação ilimitada, não ser corrosivo para superfícies metálicas, não atacar ou manchar roupa e matérias de uso corrente, não causar deterioração de borrachas, plásticos, ser compatível com sabões, detergentes e outros produtos químicos, ser atóxico e não irritar a pele de pessoas e animais, ser biodegradável, ser solúvel e estável na água, não causar o aparecimento de estirpes resistentes à ação germicida, ter elevado poder de penetração nas superfícies dos materiais, ser estável nas concentrações originais e nas diluídas e ser de fácil aplicação nas superfícies com poucas ferramentas especializadas

Nenhum desinfetante satisfaz todos estes critérios, pelo que a seleção do produto deve equilibrar estes fatores de modo a escolher o melhor desinfetante para um uso específico. O conhecimento das características de cada tipo de desinfetante, é essencial para que o médico veterinário possa selecionar o produto mais eficaz e mais eficiente do ponto de vista custo-benefício, sendo que a utilização da concentração apropriada permite alcançar os melhores resultados para cada situação.

Conhecendo as doenças infecciosas que podem complicar o dia-a-dia num CRO, nomeadamente provocando surtos com uma morbilidade e/ou mortalidade elevadas, dão-se exemplos práticos (ex: Cão com parvovirose – Que desinfetante utilizar? Qual a concentração? Em todas as superfícies? Como controlar as infeções nosocomiais?) que permitem ao médico veterinário do CRO entender a importância do tema.

É importante salientar que a elaboração e implantação de um plano de limpeza e desinfeção é muito específico e somente um médico veterinário terá o conhecimento técnico necessário para a estruturação de um PLD eficaz. Idealmente, um plano para centros de recolha de animais deve ser desenvolvido e orientado, e periodicamente revisto, por um médico veterinário com experiência em centros de recolha.

Enquanto que algumas informações sobre limpeza e desinfeção podem ser extrapoladas de várias fontes, os planos devem ser baseados no conhecimento básico e recomendações desenvolvidas especialmente para centros de recolha oficiais, tendo em conta os agentes quase permanentes e habituais (calicivírus, parvovírus, giardia, fungos...),

a população muito vulnerável (animais não vacinados, muito jovens, desnutridos, com co-infeções, em stresse), o constante movimento de entrada e saída de animais, e frequentemente a elevada rotatividade do pessoal.

A limpeza e desinfeção é essencial para minimizar a transmissão de doenças em todas as circunstâncias, mas se para o centro de recolha é impraticável a execução de um bom programa veterinário de saúde animal, se tiver um fraco sistema de ventilação ou instalações deficientes, ou for incapaz de isolar eficazmente todos os animais doentes dos animais saudáveis, uma correta higienização torna-se ainda mais crítica.

Uma adequada limpeza e desinfeção ajuda a reduzir a transmissão de doenças infeciosas para pessoas e animais e resulta num ambiente mais limpo e saudável. Um centro de recolha oficial limpo tem também o benefício adicional de aumentar o nível de conforto dos animais e do staff e representa uma imagem positiva para a população.

Medicina Veterinária na Aquicultura

David Sutherland

Opportunities and Challenges for Veterinarians in aquaculture

Aquaculture – the growing of fish, shellfish and algae in water – has been going a long time. The Chinese started growing carp in about 3000 BC. Monks in Europe grew carp from the middle ages, and the first trout and flatfish hatcheries started in the 19th century. However, modern aquaculture only really started to take off with first experiments with salmon farming in 1960s.

In 2013, aquaculture produced 97.2 million tonnes of finfish, crustaceans, molluscs, seaweeds and other animals, worth an estimated US$157 billion, made up of 575 aquatic species and species groups. 70.2 million tonnes of this was animals, and 27 million aquatic plants. This made up 43% of all fish produced in the world. 70.2 million tonnes of this was animals, and 27 million aquatic plants. Asia was the biggest producer (62.5 million tonnes) with China alone producing 59% of the world's aquaculture products. Europe produced 4% (FAO). This is increasing annually.

The biggest producer in Europe in 2014 was Norway (1.4 million tonnes, mainly salmon) followed by Turkey and United Kingdom. Portugal produced 5,760 tonnes in 2014.

Aquaculture has expanded a lot in the last fifty years, and so has the disease problems it has encountered. This has lead to a greater veterinary involvement in fish. WAVMA (World Aquaculture Veterinary Association) estimate that there are around 10,000 aquaculture veterinarians in the world, although this includes those involved in ornamental fish. Fish Veterinary Society in UK has about 100 members.

Getting training is often a bit of a challenge in the first place for an aquatic veterinarian. I graduated when fish farming was still in its infancy, so we did not get much instruction on fish. Even now, some undergraduates not get much instruction on fish. However, there are many sources of training and information.

The first problem an aquatic veterinarian encounters in the field is that his/her patients are in another environment, which we do not share. Oxygen is much less available in water than in air – only 9.08 mg/litre at 20°C in freshwater at 100% saturation, and it is less for seawater. Water can also contain other substances which can harm fish – ammonia, nitrite, suspended solids, and heavy metals. The aquatic veterinarian must inform himself about water chemistry, and how it affects fish.

If this is not enough, the fish is assailed by as wide a variety of infectious agents in water as on land – viruses, bacteria, protozoa, fungi, as well as roundworms, flukes, and various kinds of crustaceans, including sea lice.

How to examine a sick fish? They do have facial expressions, so assessment of stress is more difficult than for a dog, for example.As for a terrestrial animal, a sick fish separates itself from the "herd" (or shoal), and moves less quickly than its fellows. The skin is often darker than normal, and it swims in a strange manner. It can turn momentarily on its side (or "flash") swim on its side, or upside down. Lesions, discolouration and damage may be apparent. An increased

respiration rate may be apparent. The stock keeper may report a reduction in appetite, or complain about an increase in mortality. So the veterinarian can adapt the skills he learned for terrestrial animals.
However, in most cases, fish must be withdrawn from the water for examination. Live, healthy fish can be anaesthetized and examined for gill condition (E.g. for AGD) or parasite numbers (e.g. lice numbers). Sick fish or recently dead fish can be examined for eye damage, gill condition, external damage, and internal pathology.

The changes seen in different diseases are often similar, however, and the fish veterinarian must rely more on diagnostic tests than terrestrial vets to find or confirm their initial diagnosis. Bacteriology is used extensively, and histology is routine. Many of the diseases we encounter are viral, and PCR tests are used more in fish than in terrestrial animals. Water may have to be tested for substances, or for algae which may adversely affect the fish.

Once a diagnosis has been made, the veterinarian has less of a choice of licensed treatments than is available for terrestrial animals. For instance, only three antibiotics are licensed for fish in the UK (Oxytetracycline, Amoxycillin and Florfenicol) with another (Oxolinic Acid) available under import license. Only one anaesthetic (Tricaine) is licensed. There may well be cases where products not licensed for fish have to be sought. For Pharmaceutical companies, aquaculture is not a big market, and more data has to be generated before a product can be licensed for fish. This is especially true for bath treatments for sea lice. Not only that, but there is often the extra legislative barrier of environmental laws, as aquaculture products are released directly into the environment. In Scotland, SEPA (Scottish Environment Protection Agency) places restrictions on how much each farm site can release of medicines and treatments.

As if this was not bad enough, there may not be any product available to treat a condition, such as many of the protozoan diseases. Freshwater hatcheries have traditionally used substances which were unlicensed, but one (Malachite Green) was banned under environmental legislation, and another (formalin) is under threat from the EU.

However, there have been great advances in fish health. In the early days of salmon farming, a bacterial disease called furunculosis was a major problem, and antibiotic therapy had often to be administered, sometimes two or three times in the life of a fish. In the early nineties, effective vaccines became available, and the amount of antibiotic used in salmon farming plummeted. Norway, Scotland and Faroe Islands use very little antibiotic per kilo of salmon produced. Great strides have also been made in diagnosing fish diseases.

Another aspect of fish disease is that it can often move significant distances through the water, or indeed by through wild fish. For instance, farms within five kilometers of another infected with Infectious Salmon Anaemia (ISA) are reckoned to have a greater chance of infection. The same holds for Pancreas Disease in salmon. Fish farms have therefore moved to system where all the sites in an area are stocked at the same time, and fallowed together. Even lice treatments are co-ordinated.

Conclusion

Despite all the problems and challenges that aquaculture throws to a veterinarian, it is still a growing field, both in quantity of fish to look after, new species being cultured, and new diseases emerging. So, it is certainly worth considering if you are looking for a new challenge.

David Sutherland

Some Diseases of fish farmed in Portugal - Sea Bass & Sea Bream

Pasteurellosis

Agent – *Photobacterium damsel ssp. piscicida*
Affects sea bream, sea bass.

Sea bream – causes anorexia, focal necrosis on the gills.
Sea bass – causes a haemorrhagic septicaemia

- Haemorrhages on the gut and gills
- June – October
- Renal and heart necrosis
- Chronic form – nodules in the spleen.

Diagnosis – growth on TSA (Tryptone Soya Agar) Tryptone Soy broth or Brain Heart infusion 25C for 24 hours

- very small smooth convex grey-white colonies

Treatment – antibiotics

Control – vaccination of broodstock & juveniles

- Immunostimulant & vitamins
- Good hygiene
- Disinfection of water supply

Vibriosis

Sea bream – *Photobacterium damselae ssp. Damselae*
Dark skin, lethargy, distended abdomen.

Control – good hygiene,

- Antibiotics

Vibrio alginolyticus
Haemorrhages
Dark skin
Skin lesions

Vibrio anguillarum biotypes I & II; *Vibrio ordalli* biotype II
Sea bream & Sea bass

Sea bream – lethargy, anorexia, head down
Sea bass – dark fish

- Pale gills
- Haemorrhages around the mouth fin edges and on the skin.
- Petechiae on the intestine
- Renal and heart necrosis
- Spleen enlarged

Spring & autumn
Rapid temperature fluctuations
Grows on TSA or TSBS agar (25C for 24 hours).
Good hygiene
Antibiotics
Vaccination – Ichthiovac PD (Hipra) immersion for 60 seconds

Winter Disease

Sea bream
Pseudomonas anguilliseptica
Belly up syndrome, haemorrhages

Control – disinfection and drying out

- Adopt feeding regime

Lymphocystis

Sea bream
Iridovirus
White "pseudotumour"
Control – reduce feeding rate

- Reduce biomass
- Avoid stress
- Low pathogenicity

No treatment

Aquareovirus

Low pathogenicity
No treatment

Viral encephalopathy & retinopathy (Nodavirus)

Sea bass & Sea bream
Caused by Betanodavirus (Nodaviradae)
Most commonly occurs in larval or juvenile fish (but can occur in sub-adult or even adult fish)
Mortality can be 15 – 100%, lasting from 48 hours to several weeks.

Signs – abnormal swimming behaviour, hyperactivity, sporadically pop their heads above the surface of the water

- Colour change
- Blindness
- Abrasions
- Emaciation
- Abnormal hyperinflation of the swim bladder, but often little gross signs.

Histology – vacuolation of the grey matter of the brain & spinal cord, as well as the granular

layers of the retina – Intracytoplasmic inclusion bodies

Diagnosis – PCR

- Horizontal & vertical transmission

Distended Gut Syndrome

Sea bream
Virus
Spinning motion, stay still with the head down.
Control – UV treatment of incoming water

Monogenea

Microcotyle (Sea bass)
Treat with formalin (200 PPM for one hour, 300 PPM for 30 minutes)
Organic iron 2000 PPM for 15 – 20 days
Diplectanum aequans
Ceratothoa oestroides

Lernathropus kroyeri (Sea bass)
Copepod,
Occurs on the gills, causing erosion, desquamation and necrosis of secondary lamellae.
Treat with emamectin (50ug/kg for seven days).

Protozoa

Parasitic enteritis (Sea bream) *Myxidium leei* – Lethargy,

- Hyperpigmentation
- Abdominal distension

Avoid stress

Crytocaryon (marine white spot)
White spot
Ceratomyxa
Trichodina – wheel shaped parasite

Treatment - formalin

Epitheliocystis
Small lesions on the gills.
Not usually very pathogenic.

Vaccination strategy

Sea Bass (Vibriosis, Pasteurellosis)

- 1 – 2g nursery (immersion)
- 5 – 10g nursery/cages (oral/immersion
- Cages (injection/oral)

Aquavac Vibrio & Vibrio Oral

- 1 – 2g immersion (30 seconds, 100 kg/litre vaccine)
- Oral booster 3 – 4 months later
- 2nd oral boost or injection 3 – 6 months after 1st boost (at least 25g for injection

Sea bream (Pasteurellosis)

- 1 – 2g nursery (immersion)
- 5 – 10g oral/ immersion

Turbot

Bacteria

Vibriosis (*Vibrio anguillarum*)

- Darkened skin
- lethargy
- frayed skin
- popeye (exophthalmia)
- skin ulcers

Antibiotics
Vaccine (Ichthiovac, Hipra)

Flexibacter (*Tenacibaculum maritinum*)

- Grey patches on the dorsal fin
- Lesions on head and mouth
- Sometimes gill rot
- Affected tissue sometimes pale yellow

Bacteria in skin smears
Vaccines
Antibiotic

Furunculosis (*Aeromonas salmonicida ssp. Salmonicida*)

- Boil like lesions

Antibiotics
Vaccines available

Other Vibrios – *V. splendidus, V. Pelagius, V. harveyi*

Pseudomonas (*Pseudomonas anguilliseptica*)

- Abdominal distension, pale areas

Edwardsiella (*E. tarda*)

- Exophthalmia
- Haemorrhages
- Ascites
- Purulent fluid

Streptococcosis (*Strepococcus paraubreris*)

- Haemorrhages on the fins, skin and serosal surfaces
- Distended abdomen
- exophthalmia
- Ulcers
- Sometimes purulent exudate around eyes and base of the fins

Vaccine – Ichthiovac STR (Hipra) 0.1 ml/fish 30 – 40g
Antibiotics
Immunostimulants

Parasites

Amoebic Gill Disease – *Neoparamoeba perurans*.

Fish go off their feed, go ventral side up.
Excess mucus on the gills.
Amoeba present in gill smears

Histology

- gill filaments clubbed, lamellae fuse, amoebae present
-

Treatment

- freshwater baths – two hours + < 3 parts per thousand
- Hydrogen peroxide (800 – 1400 PPM for 20 minutes)

Scutocilliosis (*Philasterides dicentrarchi*)

- Skin ulcers
- Darkened skin
- Behaviour changes
- Popeye
- Abdominal distension

Density reduction

Myxosporidiosis (*Enterozoon scophthalami*)

- Cysts on skin and gills

Density reduction, disinfection

Microsporidiosis (*Tetramicra brevifilum*)

Endo parasite
Density reduction

Trichodina

- Flashing
- Skin darkening
- Lethargy

Disinfectant bath

Enteromyxon scophthalmi (gut parasite)

- Anorexia
- Sunken eyes
- Starvation
- Mortality can be up to 100%

Ichthyobodo **(Costia)**
Grey patches on the skin & gills
Present in gill smears, skin scrapes
Treatment - Formalin

Summary

Farmed fish are susceptible to a variety of bacterial, viral and parasitic agents. Whilst there are effective treatments and vaccines for some conditions, they are lacking for some diseases, and the only option is to carry out husbandry measures to alleviate the situation.

Novas Espécies de Companhia

Xavier Valls

Parasitosis comunes en Reptiles: ¿Cómo diagnosticar y como tratar?

Clínica Veterinaria Exotics, Barcelona

Resumen:

Los parásitos son muy comunes en los reptiles en estado salvaje y cuando se mantienen en cautividad. Tenemos parásitos externos que afectan a la piel, orejas, narinas y fosetas termoreceptoras y parásitos internos que encontramos en el aparato digestivo, respiratorio, renal y circulatorio. Una correcta anamnesis y un estudio al microscopio óptico de los parásitos externos que visualizamos y de las secreciones bucales, orina y heces nos permitirá identificar fácilmente a los parásitos más comunes y elaborar un buen plan terapéutico para eliminarlos.
No solemos recomendar realizar desparasitaciones sistemáticas sin una previa exploración del reptil ni sin un análisis de orina y heces.
Hemos recopilado los casos de parasitosis de los saurios, serpientes y tortugas que durante las últimas dos décadas han sido visitados en nuestro centro.

Animales explorados:

Iguánidos (*Iguana iguana, Basiliscus spp, Sceloporus spp, Sauromalus spp*., otros)
Agámidos (*Uromastix spp, Pogona vitticeps, Agama spp*., etc.)
Gecónidos (*Eublepharis macularius, Geco geco, Phelsuma spp*, etc.)
Camaleónidos (*Chamaeleo calyptratus, Furcifer pardalis, Chamaeleo senegalensis*, etc.)
Varánidos (*Varanus exantematicus, Varanus niloticus y Varanus spp*).
Bóidos (*Boa constrictor, Phyton regius, Phyton reticulatus, Phyton molurus, Morelia viridis*, etc.)
Colúbridos (*Elaphe guttata, Lampropeltis spp*)
Tortugas (*Testudo graeca, Centrochelys sulcata, Centrochelys pardalis, Geochelone elegans, Agrionemys horsfieldii, Astrochelys radiata, Geochelone elegans, Trachemys scripta, Mauremys leprosa, Graptemys spp. Pseudemys spp*. etc.)

La metodología que seguimos en todos los casos es la misma:

1. Exploración física general del animal
2. Anamnesis completa, observación macroscópica de las heces/orina/secreciones
3. Análisis directo al microscopio de la orina y de las heces (a poder ser frescas o recién hechas)
4. Estudio bajo lupa de los parásitos externos en caso de detectar la presencia de éstos Cuando hay secreciones en cavidad oral, realizamos el estudio de éstas mediante observación directa al microscopio óptico y/o realizando citología y tinción con Diff Quick y observación al microscopio.

Parásitos comunes en saurios: los parásitos en los saurios mantenidos como mascota son frecuentes. En la naturaleza, la mayor parte de los animales tienen un cierto grado de parásitos intestinales y/o externos, sin que éstos influyan es su crecimiento, absorción de nutrientes, etc.
Una buena higiene y profilaxis que incluya una desparasitación sistemática minimiza el riesgo de contraer parásitos intestinales en los saurios. Por otra parte, en grupos extensos de animales, se recomienda una desparasitación basada en lo observación de los parásitos que encontramos en los análisis directos. Se debe evitar introducir animales nuevos en colecciones correctamente desparasitadas sin pasar por una cuarentena y desparasitación preventiva. Los casos de parasitosis externas en grupos de saurios al igual que en grupos de ofidios, deben ser tratados a consciencia, actuando sobre los animales y sobre el medio donde viven.

Enumeramos a continuación los parásitos más comunes que vemos en saurios:

1. Parásitos externos: *Hirstiella trombiidiformis* en Iguánidos, ácaro rojo de las Pogonas.
2. Parásitos intestinales: nematodos (oxiúridos, estrongílidos y áscaris), platelmintos (tenias), coccidios (*Eimeria spp y Isospora spp*), protozoos flagelados (trichomonas) y amebas.
3. Parásitos en las vías urinarias: protozoos flagelados (*Hexamita spp*)
4. Parásitos sanguíneos: nematodos (filarias) y protozoos (plasmodium, hemogregarinas, etc.)

Parásitos comunes en tortugas: los parásitos en las tortugas de tierra mantenidas en cautividad son muy frecuentes. En la naturaleza, la mayor parte de los animales tienen un cierto grado de parásitos intestinales, y generalmente las tortugas que provienen de su país de origen tienen parásitos externos (garrapatas). Las tortugas de agua, suelen tener protozoos intestinales, sin embargo es poco común según nuestra experiencia, ver metazoos en el intestino de éstas.

En algunos animales detectamos heces más blandas de lo normal, en ocasiones malolientes e incluso con comida sin digerir. Los parásitos intestinales de mayor tamaño son los áscaris, y éstos son visibles a simple vista (normalmente el propietario te dice que la tortuga "saca lombrices"). La mayoría sin embargo son de reducido tamaño y requieren ser analizados mediante estudio de las heces bajo microscopio.

En general, los animales intensamente parasitados suelen estar delgados (pesan poco) y algunos tienen poco apetito

El diagnóstico diferencial en los casos de deposiciones blandas y/o malolientes:

- Enteritis o gastroenteritis por gusanos redondos/planos (nematodos/cestodos)
- Enteritis o gastroenteritis por protozoos
- Enteritis o gastroenteritis bacteriana
- Enteritis o gastroenteritis por microsporidios (coccidios, criptosporidios)
- Enteritis o gastroenteritis fúngica
- Enteritis o gastroenteritis por ingestión de cuerpo extraño

Enumeramos a continuación los parásitos más comunes que vemos en tortugas:

1. Parásitos externos: garrapatas en tortugas de tierra (familias *Ambliomma, Aponomma, Hyalomma, Ixoides*)
2. Parásitos intestinales: nematodos (*oxiúridos y áscaris*), protozoos flagelados y amebas.
3. Parásitos en las vías urinarias: protozoos flagelados (*Hexamita spp*)
4. Parásitos sanguíneos: protozoos (*Plasmodium principalmente*)

Parásitos comunes en serpientes: los parásitos en las serpientes mantenidas como mascota individual son poco frecuentes. En cambio, colecciones extensas de ofidios (serpentarios o criadores con cierto volumen de animales), suelen tener bastantes problemas con los parásitos externos, sobre todo con *Ophionyssus natricis*, y en ocasiones también garrapatas que se localizan entre las escamas, en las fosetas termoreceptoras o en el ojo entre el espéculo y las escamas que envuelven al ojo. También son frecuentes los parásitos pulmonares en serpientes, *Rhabdia spp* es el parásito más comúnmente observado en las vías respiratorias de los ofidios (pricipalmente en colúbridos: *Elaphe spp., Lampropeltis spp, Pituophys spp*).
Las serpientes son en su inmensa mayoría animales que se alimentan de pequeños mamíferos y de aves (carnívoros estrictos), por tanto no deben tener nunca protozoos flagelados ni ciliados en los excrementos o en la orina. La presencia de un solo protozoo en las heces de estas especies indica que hay un problema patológico. **¡No confundir** los parásitos de los reptiles con los parásitos y microorganismos que se observan en los intestinos de sus presas! (ácaros de la piel de los conejos/ratas/ratones, presencia de parásitos intestinales tipo coccidios, *Passalurus spp* de los conejos, *Sacharomyces*, tricomonas de las aves, etc.).
También encontramos en ocasiones presencia de metazoos en serpientes: nematodos (estrongiloides) y platelmintos (tenias).
Los parásitos hemáticos los vemos en ciertas ocasiones, sobre todo en animales que son importados de su país de origen. No suelen ser parásitos que tratemos ya que no suelen dar ninguna sintomatología al animal.

¿Cómo diagnostico?

En el 99% de los casos mediante una observación directa de las heces, saliva, muestra de sangre y de orina al microscopio óptico. Podemos teñir las muestras recogidas con una tinción rápida de Diff Quick® para resaltar ciertos microorganismos o con tinciones especiales tipo Ziehl neelsen (nosotros usamos la tinción en frío de Kinyoun) para detectar gérmenes ácido alcohol resistentes (Micobacterias, Nocardia o Criptosporidios por ejemplo).
Los parásitos externos suelen identificarse a simple vista o con ayuda de una tira de papel adhesivo transparente y observando en la lupa.

¿Cómo Trato los parásitos?

El tratamiento variará en función del parásito detectado y de la condición física en la que se encuentra el reptil en el momento de ser explorada.

Nosotros solemos usar los siguientes protocolos de desparasitación interna:

1. Metazoos redondos: áscaris, estrongilus y oxiúridos. Fenbendazol (Panacur®) a dosis de 50 mg/g vía oral mediante sonda rígida y repetimos a los 15 días. Al mes volvemos a analizar las heces y si aún detectamos parásitos o huevos hacemos otra tanda de desparasitación. NO USAMOS NUNCA VERMECTINAS EN QUELONIOS por su toxicidad a estas moléculas.

2. Metazoos planos: tenias y trematodos. Praziquantel (Droncit®) a dosis de 8 mg/kg vía oral mediante sonda rígida y repetimos a los 15 días.

3 Protozoos flagelados: tricomonas. Metronidazol (Flagyl®) a dosis de 100 mg/kg vía oral mediante sonda rígida y repetimos a los 15 días. El tratamiento de las tricomonas es muy cuestionado en las tortugas de tierra, ya que se consideran saprófitas y ayudan a la digestión de los nutrientes. Sin embargo, nuestra experiencia, en caso de animales con gran número de estos parásitos en heces de tortugas con diarreas profusas o muy malolientes, recomendamos tratarlas para disminuir la concentración de protozos en el intestino de la tortuga.

4. Protozoos flagelados: tricomonas. Cuando no nos funciona el tratamiento con metronidazol, utilizamos para el control de tricomonas la paromomicina (Humatin®) a dosis de 35 a 100 mg/kg vía oral cada 24 horas durante 10 días seguidos. Administramos el fármaco o bien mediante sonda rígida directamente en el estómago o mediante la comida (tortugas de gran tamaño, *Centrochelys spp*) que ingieren las cápsulas de Humatin® enteras mezcladas con la comida.
   En Lagartos es fácil que acepten la desparasitación oral mediante jeringuilla.
   Las serpientes las desparasitamos mediante sonda flexible (sondas uretrales de perro por ejemplo).

5. Protozoos ciliados: *Nyctotherus spp y Balantidium colli*: este tipo de parásitos unicelulares ciliados los vemos con mucha frecuencia en los análisis de heces directos de tortugas terrestres. No solemos dar ninguna importancia a la presencia de tales microbios, ya que está descrito que son parte integrante de la flora microbiana normal de los quelonios. Ayudan a digerir la celulosa de los vegetales que ingiere la tortuga, y únicamente recuerdo un caso en el que tratamos un animal con heces muy pastosas y mal digeridas. En caso de tratarlos, utilizamos el metronidazol.

6. Amebas: la presencia de estos parásitos, en nuestra experiencia, siempre es indicativo de patología intestinal. A menudo, animales con retraso en el crecimiento, mala digestión, presencia de alimento no digerido en las heces, es indicativo de amebiasis intestinal. Las amebas las tratamos con paromomicina (Humatin®), a dosis de 50 mg/kg vía oral 10 días consecutivos. Posteriormente, analizamos las heces del reptil en 2 o más ocasiones para cerciorarnos de la eliminación total de estos parásitos.

7. Microsporidios: coccidios y criptosporidios. Los coccidios, a día de hoy, son un tipo de parásito que no hemos visto prácticamente nunca en heces de tortuga: en caso de detectarlos las desparasitamos con toltrazurilo (Baycox®). Los criptosporidios se han descrito en numerosas especies de tortugas (criptosporidiosis gástrica), sin embargo, en análisis coprológicos nunca hemos visualizado criptosporidios en tortugas (sí en cambio en serpientes y lagartos). La criptosporidiosis es relativamente frecuente en tortugas jóvenes, con retraso en el crecimiento y en muchas ocasiones con problemas de calcificación. El único fármaco útil para controlar a los criptosporidios es la paramomicina (Humatin®), aunque recientemente se ha probado tratar animales diagnosticados de criptosporidios con suero fetal bovino obteniéndose buenos resultados.
   Los coccidios se ven con mucha frecuencia en camaleones y en pogonas. Los tratamos con Toltrazurilo (Baycox® 5%), la pauta que seguimos es: 5-15 mg/kg V.O. 3 días seguidos.

8. Los reptiles con parásitos externos los tratamos con ivermectina inyectada (Ivomec®) a dosis de 200-400 microgramos por kg (saurios y serpientes). Se repite la dosis a los 15 y 30 días. Los ácaros aislados sobre la piel del animal, son arácnidos que provienen de la madera o de los troncos recogidos en el campo que usan como enriquecimiento ambiental para el terrario. Es de suma importancia eliminar del terrario los troncos y desinsectizarlo. Para ello utilizamos una mezcla de agua con ivermectina (Oramec®: ivermectina 0.08%), mezclando enérgicamente en una botella pulverizadora el agua con la ivermectina en solución oral (500 ml de agua/6ml de Oramec®) y luego rociando con esta mezcla todo el terrario, los recodos, las piedras, etc.
   En las tortugas debemos evitar las "vermectinas" debido a su alta toxicidad. Podemos aplicar la ivermectina sobre las garrapatas y extraer pasados dos días a los parásitos una vez han fallecido.

Xavier Valls

Estasis reproductiva: ¿Pre-ovulatoria o post-ovulataoria? ¿Cómo tratarlas?

Clínica Veterinaria Exotics, Barcelona

Resumen:

Los lagartos, tortugas y serpientes mantenidos en cautiverio suelen tener problemas reproductivos por el mero hecho de no disponer de lugares para realizar la ovoposición. En esta conferencia se explicará mediante casos reales la manera de detectar cuando un reptil está gestante o en fases iniciales de la gestación (activación de los ovarios), cómo detectamos si hay algún problema reproductivo (distocia) y como resolvemos estos problemas de forma médica o quirúrgica.

ESTASIS: ¿Pre-ovulatoria o post-ovulataoria?

Difícil respuesta si no se dispone de los medios adecuados para llegar a tal respuesta con certeza. Una vez conocemos los ciclos normales de puestas para une especie concreta y que proviene de un lugar concreto (hemisferio norte o sur), nos debemos plantear las siguientes preguntas:

a. ¿Qué edad tiene?
b. ¿Qué época del año es?
c. ¿Dónde vive el reptil, indoor, outdoor? ¿Aspecto del terrario?
d. ¿Cuáles son las condiciones de mantenimiento: Tª, humedad, ciclo de luz y tipo de luz, etc.?
e. ¿Tiene algún sitio para la puesta de huevos? ¿Hay sustrato en el terrario o en el jardín, de qué tipo y cuanto grosor de sustrato?
f. ¿Este ejemplar ha hibernado?
g. ¿Cuál es su dieta?
h. ¿Vive con otros animales de la misma especie, hay machos, cuantos, peleas?
i. ¿Es la primera vez que pone huevos o es la primera vez que se sospecha que va a poner huevos?
j. Describe los síntomas más relevantes que tiene el animal: comportamiento excavador, nerviosismo, pérdida de apetito, etc.

Una vez explorado al animal y preguntado la extensa anamnesis, debemos plantear las siguientes pruebas para decidir si hay o no un problema distócico:

1. Radiografía: nos va a permitir ver si hay huevos calcificados y qué grado de calcificación tienen éstos. En lagartos y serpientes la radiografía permite ver si los huevos están calcificados o están en estasis preovulatoria. En tortugas solamente veremos si hay huevos si éstos están en el oviducto y calcificados.

2. Ecografía: muy útil la ecografía en tortugas, ya que nos permite evaluar a través de las diferentes ventanas acústicas si hay presencia de folículos ováricos preovulatorios y la fase de desarrollo de éstos. También la usamos para valorar el aspecto ecográfico de estos folículos y determinar si se trata de una estasis o de una ovulación.

3. Determinar los valores de Calcio. Valores superiores a 20 mg/dl indican que el ovario está lleno de folículos y que todavía no ha ovulado la tortuga/lagarto o serpiente.

   Valores elevados de Calcio en animales jóvenes puede indicar otras patologías.

   Valores normales (entorno a 10 mg/dl), indican que el animal ha ovulado y debemos plantear una Rx para ver si se detectan huevos calcificados en el oviducto.

4. En ocasiones se puede plantear realizar un TAC para valorar el estado interno de una tortuga, en busca de huevos poco calcificados/masas intracelómicas, presencia de líquido libre y estado de los ovarios, etc., que nos ayuden a orientar un diagnóstico (folículos libres en cavidad abdominal, líquido libre, masas que retienen contraste, etc.).

Tenemos diferentes situaciones en las cuales sospechamos de una distocia y generalmente debemos tomar una decisión que implicará un tratamiento poco confortable para el reptil y que en numerosas ocasiones implicará una cirugía. Mostramos a continuación diferentes situaciones en las que nos encontramos en la clínica diaria y explico la manera como las resuelvo.

¿Cómo tratar las distocias en tortugas? Basándonos en las imágenes radiográficas/ecográficas nos encontramos con estas situaciones:

- La presencia de huevos calcificados **rotos** en tortugas, casi siempre nos indica una distocia post-ovulatoria. En este caso el tratamiento de elección sin duda es la Celiotomía y extracción de estos huevos "rotos". Sin embargo hay numerosos propietarios que por motivos económicos (son la mayoría) no quieren realizar la cirugía. Nuestra experiencia en estos casos es que en un 50 % funciona el tratamiento médico y el quelonio es capaz de expulsar los huevos todo y estar rotos dentro del oviducto. El otro 50%, suelen empeorar tras la estimulación farmacológica (oxitocina, prostaglandinas, atenolol, etc.) y requieren una cirugía de urgencia o a veces una eutanasia.

- La presencia de huevos poco calcificados en tortugas aparentemente sanas, sugiere un problema metabólico. Se debe revisar su calcemia y valorar el tratamiento con calcio inyectable para restablecerla si es baja (niveles inferiores a 8 mg/dl). Posteriormente se puede plantear un tratamiento médico o quirúrgico para resolver el problema de retención de huevos.

- Observamos huevos con cáscara muy gruesa y otros menos calcificados. Esta situación nos indica que hay huevos retenidos de años anteriores y huevos del presente año. En este caso recomendaríamos como primera opción la cirugía, ya que probablemente hay algún problema en la ovoposición y probablemente la distocia sea obstructiva.

- Vemos un solo huevo en la mitad de la cavidad celómica, muy calcificado. Suelen ser tortugas que han puesto ya huevos esta temporada pero siguen intranquilas y con comportamiento de puesta. Puede ser un huevo retenido en un oviducto y ha eliminado correctamente los huevos del otro oviducto o un huevo en la vejiga de la orina. Yo recomiendo realizar endoscopia cloacal. En caso de ser un huevo en vejiga, se puede extraer si la necesidad de intervención quirúrgica!!!

- Tortugas con radiografías normales, sin huevos calcificados, pero con comportamiento de gestación (intranquilidad, pérdida de apetito, etc.). Recomendamos realizar analítica sérica. Si los niveles de Ca son muy elevados, esperar un tiempo prudencial (1, 1.5 meses) y repetir Rx y/o análisis. También evaluamos el frotis sanguíneo en busca de leucocitosis para descartar posibles problemas infecciosos/inflamatorios (celomitis por yema de huev por ejemplo). Si en animal sigue sin comer, Calcio elevado y no hay huevos calcificados, recomiendo realizar ecografía y probablemente celiotomía para extirpar los ovarios que contienen los folículos preovulatorios retenidos.

- Tortuga sana y bien alimentada que en época de la puesta, está intranquila, cava agujeros en el suelo, se mueve de un lugar a otro y deja de comer. Observamos en la radiografía un número de huevos acorde con la especie, de tamaño y aspecto normal. En este caso iniciamos tratamiento médico con Oxitocina (2 UI/kg IM inicialmente). Se puede administrar 1 hora antes una dosis de Calcio IM (100 mg/kg). Una vez inoculada la oxitocina, se envía al animal a casa y generalmente hace la puesta durante la noche. Si no hay puesta de los huevos, se repite el proceso y vamos aumentando la oxitocina, 5 UI/kg hasta 10 UI/kg en sucesivas inyecciones.

- Cada presentación clínica es diferente, ya que las tortugas suelen ser distintas, de edades comprendidas entre los 7 y los 90 años, con un manejo y dieta particular y unas condiciones de mantenimiento distintas. La mayor parte de reptiles de la familia de los quelonios cuando tienen un problema reproductivo, se suele manifestar como mostramos en los apartados mencionadas anteriormente, pero cada animal es un mundo y cada vez que sospechamos de una estasis reproductiva, tendremos que evaluar cada animal y entorno de éste.

¿Cómo tratar las distocias en lagartos?

El problema de los saurios es la amplísima variedad de especies y de subespecies (y "Fases") que nos llegan a la consulta. También es difícil convencer a los propietarios de estas mascotas (sobre todo a los criadores) de llegar a un diagnóstico del problema que éstas padecen y de poner soluciones individuales a grupos de hembras que se mantienen para la cría.

En animales que se mantienen solitarios o en grupos reducidos como "mascota", es más fácil convencer al propietario para que esterilice su Iguana, Geco o Pogona, sin embargo los criadores evitan estos procedimientos ya que el objetivo de un saurio hembra es que crie.

Dicho esto, vamos a exponer diferentes situaciones y casos en los cuales se sospecha de estasis folicular/distocia en lagartos, basándonos en las imágenes de radiografías o las ecografías realizadas al saurio.

- Presencia de huevos calcificados, ovales, sin alteraciones significativas en la forma, tamaño, número y calcificación de los huevos. En estos casos, solemos recomendar la estimulación de la puesta aportando un ambiente adecuado y un grosor suficiente de sustrato para que puedan ovopositar. Si el manejo de las condiciones

ambientales no es posible, podemos estimular la puesta con inyecciones de calcio intramuscular o intravenoso y posteriormente oxitocina y/o otros agentes estimulantes de la contractibilidad del aparato genital (dosis del Formulario de Carpenter). Este procedimiento funciona en alguna ocasión si se trata a Iguanidos y Agámidos, sin embargo los camaleones no suelen dar buen resultado y es preferible plantear la cirugía.

- Huevos deformados, mal calcificados o doblados/rotos. Generalmente planteo un análisis de sangre e inmediatamente una cirugía: ovariosalpingotomía. En esta ocasión, si el lagarto no está ya muy debilitado, la cirugía suele ser curativa ya que retiramos los oviductos y los dos ovarios.

- Se observan numerosos folículos preovulatorios, formando una especie de racimo de uvas en la zona dorso caudal del animal. En ocasiones estos folículos se aprecian externamente (camaleones, iguánidos y agámidos). Mi recomendación, cuando apreciamos en la Rx o en la ecografía una retención preovulatoria, es extirpar los dos ovarios mediante una Ovariectomía bilateral.

- En algunos gecos, apreciamos en una rx la presencia de 3 huevos en lugar de 2, que es lo común en esta familia de reptiles. Se puede pensar que es una gestación normal, sin embargo en la mayor parte de ocasiones, un huevo suele ser ectópico y vagar por la cavidad celómica. La solución es la cirugía abdominal.

- Hay numerosas especies de camaleones de montaña que son vivíparos, en éstos es difícil tomar una decisión de si actuar de forma médica o quirúrgica. Generalmente son animales de muy reducido tamaño y esto dificulta la decisión a tomar cuando sospechamos de una distocia.

- Los animales que se suelen mantener como mascota, individualizadas y sin el objetivo de hacer criar, solemos plantear una Ovariosalpingotomía preventiva. En reptiles de una cierta edad y tamaño, la esterilización es una buena solución a todos los problemas reproductivos y de comportamiento ligados a las hormonas sexuales.

¿Cómo tratar las distocias en serpientes?

No solemos ver problemas reproductivos en serpientes. Probablemente es por un tema estadístico (es el grupo de reptiles que vemos con menos frecuencia) pero además, son animales que pueden activar el ovario y reabsorber los folículos preovulatorios con más acierto que los lagartos y las tortugas. A veces se detectan problemas reproductivos ligados al crecimiento corporal excesivo de estos animales en cautividad (sobrealimentación). Hemos visto pitones birmanas alcanzar la edad púber en tan solo 3 años, por el exceso de aporte de alimento. Estos crecimientos acelerados, facilitan que el animal llegue a la pubertad de forma prematura y esto desencadena con mucha probabilidad una distocia obstructiva a la hora de realizar la puesta.

También tenemos numerosas especies de serpientes vivíparas (los Bóidos), las cuales no suelen manifestar problemas a la hora de la puesta.

La cirugía para eliminar los huevos retenidos en casos de distocia, requiere de una serie de incisiones laterales para acceder a los huevos retenidos, los cuales se localizan a lo largo del último tercio del cuerpo del animal.

En comparación a los quelonios y saurios, tenemos una incidencia muy baja de distocias en serpientes. También hemos constatado, que las serpientes si no ovulan, suelen reabsorber los folículos ováricos con cierta facilidad, sin empeorar su estado corporal (siguen en su mayoría comiendo cuando están reabsorbiendo los folículos preovulatorios)

Xavier Valls

Huston, tenemos un problema: ¿Qué hacer con una urgencia de un reptil cuando lo que se de mamíferos no lo puedo extrapolar?

Clínica Veterinaria Exotics, Barcelona

Resumen:

En numerosas ocasiones, los centros veterinarios para perros y gatos deben atender emergencias de animales poco convencionales. Los reptiles suelen ser un reto a la hora de ser atendidos por profesionales acostumbrados a los mamíferos domésticos. En esta conferencia trataré de explicar las emergencias más frecuentes y la manera de actuar para estabilizar inicialmente a la tortuga caída o mordida, al camaleón deshidratado o a la iguana con temblores (por poner unos ejemplos).

IIntroducción:

Una urgencia en un reptil no tiene nada que ver con una urgencia de un mamífero!!!

Son animales ectotérmicos, no generan calor y su estado de actividad/anímico depende de la temperatura ambiental, fotoperiodo, presión atmosférica y de las fuentes de iluminación que se usan.

Hay más de 7000 potenciales especies de reptiles que pueden llegar a nuestro centro, desde animales minúsculos (crías de tortugas, camaleones de tamaño pequeño, gecos, etc., hasta reptiles que superan los 50 kg, por ejempla Tortugas de tierra centroafricanas, caimanes, cocodrilos o pitones birmanas entre otras).

En la mayor parte de ocasiones, nos traen animales que llevan enfermos semanas e incluso meses, aunque el propietario asegure que el animal ayer estaba bien.

El gran problema no es estabilizar o curar al animal de una urgencia puntual, el gran caballo de batalla es convencer al propietario de la mascota de mantenerla en unas condiciones adecuadas de espacio, temperatura, humedad, irradiación y alimentación. Las recidivas suelen ser muy comunes y desesperantes para el veterinario que atiende a estas mascotas repetidamente por el mismo motivo.

La mejor defensa en estas visitas es un buen ataque, debemos explicar al propietario que el animal no está enfermo porque sí, lo hemos "puesto enfermo" nosotros por no aportar las condiciones adecuadas de mantenimiento. Es decir, la culpa recae casi siempre sobre la persona que te trae el animal enfermo, y se debe remarcar esto en la consulta, pero sin pasarse, ya que si ofendemos al cliente no volverá con su mascota al veterinario.

Haré a continuación un listado de urgencias habituales de reptiles, intentando explicar en cada caso el protocolo de actuación que usamos en cada caso.

1. Tortuga paracaidista: muchos propietarios mantienen a sus mascotas en balcones. La mayor parte de la población vive en ciudades y las mascotas, al igual que sus propietarios, conviven en pisos. Es muy común tener a la tortuga en un balcón para que le dé el sol, pero las tortugas terrestres y acuáticas tienen la manía de escalar o colarse por los sumideros o desagües de estos balcones o terrazas. Cualquier planta cerca de la pared o las paredes con relieves (ladrillos por ejemplo), facilitan la escalada de los animales y su posterior caída. Atendemos a lo largo del año un gran número de animales caídos de las casas de los propietarios!!!

2. Tortuga mordida por un cariñoso perrito: los perros son el amigo más fiel del hombre, pero a las tortugas las muerden. "Evitar perros y Tortugas juntos".

3. Accidentes en jardines: la tortuga está enterrada y viene el jardinero.... El resultado es generalmente fatal para el animal, ya que suelen tener heridas en caparazón y/o miembros, tienen cortes profundos y de difícil solución.

4. Tortugas que están en plena hibernación y aparecen por el jardín con lesiones en las extremidades: lesiones por roedores. Suelen requerir amputaciones de los miembros/cola. En primavera, suelen venir con la complicación de larvas en las heridas, miasis.

5. Tortugas de agua que tienen los ojos hinchados: es una de las visitas más comunes, no la podemos catalogar como una urgencia, sin embargo el propietario ve que el animal en muy poco tiempo deja de ver y tiene un gran hinchazón en los párpados, lo que le impide abrir los ojos.

6. Iguanas que salen a tomar el sol a la terraza, balcón o que las suben al árbol del jardín. Al igual que las tortugas, muchos de estos animales caen y suelen fracturarse los huesos largos de las extremidades o la cola.

7. Lagartos que pierden la cola: es frecuente que nos llegue a la clínica gecos, lacértidos o iguánidos que han perdido una parte de la cola cuando era manipulado por su amo. El propietario suele estar muy asustado y nos traen la parte de la cola cortada para ver si la podemos "PEGAR".

8. Saurios que tienen temblores: hace una década estas urgencias eran muy comunes. Los reptiles más afectados de este problema eran las iguanas (Iguana iguana) y en general los saurios de las familias de los camaleones y los agámidos. Los animales llegaban a la consulta con temblores y en ocasiones con las extremidades y la mandíbula hinchada. En camaleones se ve actualmente con cierta frecuencia animales que tienen las extremidades deformadas, éstos suelen ser hembras grávidas!.

9. El lagarto o la tortuga tienen "algo" que les sale por el ano: una urgencia que atendemos con cierta frecuencia es la del prolapso cloacal/ del pene o del hemipene. En raras ocasiones se ven prolapsos de intestino o de la vejiga de la orina. Los prolapsos de oviducto los contemplamos en la ponencia **"Estasis reproductiva: ¿Pre-ovulatoria o post-ovulataoria? ¿Cómo tratarlas?"**

10. La serpiente tiene toda la zona ventral descamada y/o con vesículas: las quemaduras. El manejo de las quemaduras es complejo y en numerosas ocasiones muy desalentador. Explicaré cómo manejo la urgencia de una quemadura y la evolución que suele tener.

11. Las serpientes de un criador empiezan a respirar mal tras una desparasitación colectiva, se desorientan y pierden el equilibrio: suele suceder cuando se utilizan antiparasitarios a dosis elevadas (por ejemplo el amitraz y los órganofosforados).

1,2,3 y 6: Urgencias traumáticas

Los traumatismos de los reptiles, ya sea por caídas, mordiscos, golpes o por otras causas, solemos tratarlos siempre siguiendo el siguiente protocolo:

- Exploración Física General/Anamnesis
- Valorar estado de hidratación (piel seca y adherida, ojos hundidos, mucosas secas,...
- Valoración de la posible pérdida de sangre: coloración de las mucosas, TRC, Hematocrito!!!! (muy importante el hematocrito).
- En casos de caparazones rotos, limpiar heridas y estabilizar las fracturas con tiras de esparadrapo, cintas cohesivas (3M), etc.
- Si hay presencia de larvas de mosca, limpiar bien la zona afectada y retirar las larvas manualmente, desinfectar las lesiones y cubrir con pomadas tipo Blastoestimulina®, Silvederma® o similar.
- En el caso de fracturas cerradas de extremidades (lagartos), estabilizar el miembro afectado atándolo al cuerpo o a la cola.
- En casos de lesiones muy graves, con amplia pérdida de caparazón o evisceración (por ejemplo), valorar eutanasia del animal
- Manejo del dolor con Morfina (de 1 a 10 mg/kg/24 h)
- Si la lesión es por mordisco de otro animal, tratar de inmediato con antibióticos; Ceftazidima o Ceftazidima+enroflozacina son los fármacos de elección.
- Hospitalizar al animal, mantenerlo hidratado y mantener en un ambiente adecuado con su rango de temperatura óptima (generalmente 24-28 ºC).
- Si no se dispone de material o conocimientos para reparar los caparazones o huesos largos rotos, derivar una vez estabilizado el reptil, a un centro de referencia especializado en medicina y cirugía de animales exóticos.

4: Urgencias por mordisco de rata

Estas lesiones tienen difícil solución si no se tratan a tiempo. Por norma general, se deben amputar las extremidades afectadas de mordiscos por roedor para poder cicatrizar correctamente y que se forme el muñón. Es de vital importancia la antibioterapia y despertar al animal de su letargo, mediante baños con agua tibia, sueros intra-abdominales/epicraneales, inyectar vitaminas del complejo B, etc. Estos animales requieren ser hospitalizados (de 1 a 4 semanas).

5: Tortugas de agua que tienen los ojos hinchados

Suelen ser quelonios de agua dulce mal alimentados de forma crónica (sobre todo tortugas alimentadas a base de gambitas liofilizadas (Gammarus) y/o pienso. Las tratamos eliminando la secreción blanca y espesa que se forma

en el ojo entre párpados y córnea, pinchando 2-3 dosis de vit A (2000 UI/kg) cada 15 días y cambiando la dieta. Las tortugas llamadas de florida, tienen una dieta muy variada y podemos aportarle proteína y vitamina A, a partir de la carne de vacuno, ovino, conejo, pollo etc. Recomiendo que coma una vez a l mes hígado de ternera, ovino, conejo o pollo. Normalmente, tras la primera inyección, el ojo que suele tener la córnea blanquecina/opaca, se aclara un poco. Se produce en los párpados una metaplasia escamosa y un edema.

7: Lagartos si su cola

Las manipulaciones bruscas, el estés y/o una mala sujeción, suelen provocar la pérdida de una parte de la cola o de la cola entera. **NO** se deben sujetar nunca los reptiles agarrándolos de la cola. Iguánidos, Lacértidos y Gecónidos tienen en las vértebras coccígeas, zonas preformadas de ruptura. En casos en los que la cola está parcialmente rota, no es recomendable suturar, ya que éstos saurios tienen la cola diseñada para soltarla voluntariamente. En estos casos es mejor acabar de "romper" la cola por la zona fracturada y coser el muñón con puntos de sutura en U.

8: Saurios con temblores

Cuando acuden a la clínica lagartos con temblores, lo primero que hacemos, si se puede, es tomar una muestra de sangre y medir los niveles de calcio y fósforo sérico. Los temblores son generalmente por bajadas de calcio sérico. Es importante recuperar estos niveles hasta su normalidad, por el contrario estos animales pueden morir de hipocalcemia. Si comprobamos que el calcio está bajo, recuperamos la calcemia mediante inyecciones intravenosas de calcio gluconato. Las inyecciones intramusculares son muy dolorosas y debemos evitarlas. Una vez conseguimos recuperar la calcemia, tenemos que realizar una extensa anamnesis para saber por qué motivo el animal manifiesta estas tetanias (falta de luz UVB, falta de exposición solar, fuente de luz gastada, dieta rica en fósforo y baja en calcio, etc.). Una vez detectamos la causa que ha llevado al reptil a la hipocalcemia, debemos actuar para evitar que estos síntomas vuelvan a aparecer.

9. Prolapsos cloacales

Los prolapsos son frecuentes en los reptiles. Las causas que los provocan son muchas y los tejidos prolapsados pueden ser la cloaca, el intestino grueso, el oviducto o la vejiga de la orina, el pene de las tortugas y cocodrilos o los hemipenes de los escamosos. El primer paso a seguir siempre es el de reducir el tejido prolapsado, mediante torundas lubrificadas, disminuir el hinchazón ayudándonos de frío, soluciones sobresaturadas de sal o de azúcar o con gasas impregnadas con manitol. Una vez reducido, solemos colocar un par de puntos en los extremos de la cloaca o una sutura en forma de bolsa de tabaco. Los penes y hemipenes prolapsados se recomienda amputarlos. Una vez estabilizados los animales, se debe buscar la causa del prolapso para evitar la recidiva.

10. Quemaduras

Las quemaduras son cada vez menos frecuentes en nuestra clínica, probablemente por la adecuada información que dan las tiendes que venden reptiles y por las fuentes de calor que se usan actualmente. Aún así, vemos quemaduras en ocasiones y debemos saber cómo actuar ante una quemadura de segundo o tercer grado. Los reptiles más afectados por las quemaduras son las serpientes, suelen quemarse con las esterillas o los cables calefactores. También se queman con cierta frecuencia los camaleones, cuando se acercan demasiado a las fuentes de iluminación o de calor.

Las serpientes se queman las escamas ventrales, la gravedad depende del grado de la quemadura, que al igual que en los mamíferos se tipifican como quemaduras de primer, segundo y tercer grado. Las quemaduras de primer

grado, resuelven bien aplicando a los enrojecimientos de las escamas pomada a base de sulfadiazina argéntica. Las quemaduras de segundo gado, tenemos la formación de vesículas que revientan y exudan, de tal manera que es muy fácil ver contaminación secundaria. Estas quemaduras tienen un pronóstico más reservado ya que si no se manejan correctamente las heridas ventrales, el animal puede desarrollar una sepsis y/o una deshidratación grave. Estos animales requieren hospitalización durante varios días, rehidratación parenteral y tratamientos prolongados con antibióticos, pomadas cicatrizantes, etc.

11. Intoxicaciones

Las intoxicaciones son frecuentes en criadores con muchos animales, sobre todo criadores de serpientes y lagartos. Al usar antiparasitarios altamente tóxicos, pueden exceder con facilidad las dosis tóxicas. En numerosas ocasiones se usa el Amitraz o los pesticidas organofosforados. Cuando atiendes animales sospechosos de estar intoxicados, lo importante es saber qué tóxico provoca los síntomas (suelen ser síntomas neurológicos como la incoordinación de los movimientos). Cuando sabemos el tóxico, podemos ver si tiene o no antídoto. Los casos que hemos tenido nosotros, respondieron bien a la atropina o al tratamiento de soporte.

Cristina Almeida

Como estabilizar uma ave doente?

Resumo

As aves como animais de companhia sempre existiram ao longo da história. Os seus cuidados e sobretudo o estudo das suas doenças tem sido alvo de grande evolução na medicina veterinária. Enquanto clínicos, devemos estar familiarizados com a identificação das principais espécies de cativeiro, presentes como animais de companhia, criação e coleção. A distinção dos comportamentos normais e dos sinais clínicos iniciais é para os nossos clientes um enorme desafio e requer aprendizagem. Esse conhecimento pode e dever ser partilhado nas primeiras consultas de rotina. É sempre mais complexo gerir a urgência, reabilitar a ave debilitada e também corresponder às expectativas do proprietário se não dominarmos as técnicas básicas de manuseamento, contenção e recolha de amostras biológicas destas espécies. É com expectativa que esta apresentação possa servir de partilha e de revisão dos aspetos essências para estabilizar uma ave doente. A nossa atenção deve prender-se com múltiplos aspetos que não podem ficar em espera como : a inanição, a correta oxigenação, a temperatura, a hidratação e o estado mental. Deve-se recordar que lidamos com 25% de mortalidade numa situação de urgência como tal "mãos à obra!"

Introdução

Muitas vezes o primeiro contato entre o cliente de uma ave e a clínica é feito através do atendimento telefónico e email. Quando lidamos com aves é importante ter uma equipa treinada com linhas orientadoras de triagem para que de forma rápida e inequívoca possamos distinguir as verdadeiras urgências. Isto significa que devem ser vistas, o mais rápido possível, as aves que evidenciem as seguintes alterações:

- mudança súbita nos hábitos alimentares
- vómito ou regurgitação
- perda de sangue ou feridas visíveis
- mudança da frequência e aspeto das fezes
- mudança de comportamento, atitude e temperamento
- penas eriçadas e corpo em forma de bola
- corrimento anormal olhos, boca, narinas
- alteração dos sons respiratórios
- alteração da voz

Estas merecem uma atenção imediata do médico veterinário sendo que as indicações e as recomendações devem ser feitas apenas mediante consulta do paciente.

Antes de se dirigirem à clínica deve ser solicitado sempre que possível o seguinte

- trazer a ave na sua própria gaiola ou pela menos imagens vídeo ou foto das instalações.
- Amostras de fezes recentes
- Amostra da comida habitual
- Registo de medicamentos previamente administrados
- Vitaminas, minerais ou outros suplementos que tenham sido fornecidos.
- caso venha referenciado de outro colega, as informações devem ser transmitidas aquando da consulta.

Anamnese

A observação da ave inicia-se antes mesmo da sua captura. O levantamento de uma história detalhada pode ser o início da detecção de problemas graves. O comportamento, o movimento e postura do corpo, a observação dos movimentos respiratórios bem como o estado de alerta da ave devem ser alvo de cuidadosa apreciação.

Existem dados que devem recolhidos numa fase inicial que enumeramos em seguida sobre a identificação.

- idade, se conhecida
- sexo, se foi determinado e por qual método
- história reprodutiva, sendo mais importante em fêmeas
- espécie e enquadramento(ave silvestre, doméstica, capturada, criada à mão)
- origem (criador, loja de animais, outro)
- duração de pertença (outros donos, mudanças)

A propósito do habitat em cativeiro tentar obter uma descrição detalhada das instalações, maneio e higiene, tais como:

- gaiola(tamanho, localização, material, posição relativa a outras aves)
- poleiros (dimensão, posição, composição)
- higiene (frequência, materiais e métodos)
- interação homem-ave-outros animais
- exposição a toxicidade (plantas, alimentos, fatores ambientais)

A dieta e a forma como é providenciada deve ser revista:

- tipo de alimentação
- comida avulso, sacos fechados
- uso de germinado
- armazenamento
- preparação de alimentos frescos
- rotina de higiene

Deve-se registar o motivo da consulta e da vinda ao veterinário. Nem sempre a maior preocupação do proprietário corresponde ao problema de maior gravidade. Fazer um levantamento sobre o aparecimento dos sinais em termos de tempo, frequência, duração e persistência. É importante também rever historial médico, tratamentos prévios, exames efetuados anteriormente de relevância.

Emergência em aves

Os pássaros são mestres em esconder as suas doenças, como uma resposta da preservação e defesa dos predadores. Contudo a crise surge quando o pássaro não pode mais compensar a doença e os sinais do problema tornam-se evidentes.

As aves criticamente doentes são frequentemente apresentadas num estado avançado de descompensação. Quando um proprietário afirma que "o pássaro estava bem ontem" e hoje ele está sentado no fundo da gaiola, o proprietário geralmente pode ser verdade.

Outro assunto que deve ser discutido é se o pássaro deve ser levado para trás para tratamento, ou se o proprietário deve estar presente durante a avaliação inicial. Muitos proprietários estão muito nervosos e não querem estar separados da ave, especialmente durante uma crise. No entanto, sob nenhuma circunstância deve-se evitar um proprietário restringir ou conter o seu pet durante o tratamento e exame.

As técnicas de contenção devem ser praticadas, se necessário, para que as aves doentes não sejam feridas ou forçadas indevidamente, por restrição estranha ou perigosa. Além disso, aves com pele facial nua são propensas ao aparecimento de hematomas, contusões ou abrasões na mesma.

Deve ser fornecido calor de imediato, por exemplo uma incubadora. Se dispneico, evitar manipulação excessiva e preparar oxigénio complementar. O paciente deve ser mantido quente, ambiente calmo e bem ventilado com mínimo ou nenhum distúrbio.
Pesar o pássaro em gramas é muito importante para o cálculo das doses de emergência e fluídos.
Realizar um exame físico completo, mas breve, incluindo a auscultação dos pulmões, sacos cardíacos e aéreos, usar ampliação e fonte de luz para examinar a oro faringe, fenda cutânea, olhos, orelhas, pele, penas, cloaca, palpação, abdômen, membros.
Administrar medicamentos, fluidos, desde que o proprietário esteja informado do nosso plano e de um presuntivo prognóstico.
Claro, se houver sangramento excessivo, dispneia ou outros problemas que requerem atenção imediata, tratá-los imediatamente.
Organização e preparação antecipada são necessários para fornecer cuidados necessários com a menor quantidade de manipulação e máxima rapidez de execução com uma equipa devidamente treinada.

O objetivo do veterinário de emergência deve ser para estabilizar o paciente. Embora seja sempre uma boa ideia para adquirir amostras para cultura e sangue para testes na apresentação inicial, muitas vezes tem riscos e dever ser comunicado. Dito isto, a menos que haja circunstâncias especiais, provavelmente é melhor não tirar sangue durante o período de exame inicial para despistes moleculares por exemplo.

A maioria dos casos de aves descompensadas são : o descompensado adulto ou neonato, lesões traumáticas , toxicoses, mordida feridas de outros animais de estimação da família (cães, Gatos, furões, etc.), queimaduras elétricas, distócia, dispneia grave.

Fluidoterapia em Aves

Frequentemente e qualquer paciente ave sofre de algum grau de desidratação. Diarreia, poliúria, regurgitação / vômito e consumo de água diminuído resultarão em desidratação.

Os fluidos subcutâneos são administrados nas áreas da axila e flanco lateral. A área intraescapular também pode ser empregada, no entanto, é importante evitar a área em torno da base do pescoço, devido às extensas comunicações do sistema de saco de ar cervicais. Use uma agulha de calibre pequeno, 25-28 ga. Para evitar fugas de fluidos nos locais de injeção. O volume total de líquido (5-10 ml / kg / local) deve ser administrado em vários locais, para evitar a interrupção do fluxo sanguíneo e subsequente má absorção.

A importância de manter o equilíbrio de fluidos e eletrólitos deve ser uma prioridade. A maioria dos pacientes hospitalizados deve receber fluidos de reposição, com base nas perdas estimadas de evaporação, hemorragia e vasodilatação presumida. O volume de fluido de substituição equivale bem a requisitos de manutenção, desde que não haja perda significativa de sangue e a ave não regurgitar ou poliúrica. Os volumes de substituição de fluidos calculados para doentes gravemente doentes ou traumatizados devem incluir fluidos perdidos do compartimento vascular resultantes da resposta fisiológica à doença e fluidos sequestrados da circulação como resultado de um trauma. Perdas absolutas (sangue perdido por hemorragia) e mudanças de fluidos em áreas de lesão, infecção ou tecidos com fluxo sanguíneo comprometido também devem ser consideradas. O deficit de fluido pode ser calculado com base no peso corporal e no grau de desidratação percebido com a seguinte fórmula:

Deficit de líquido (ml) = grau de desidratação (%) x peso corporal (g)

As necessidades diárias de fluidos de manutenção para aves de rapina e psitacídeos foram estimadas em 50 ml / kg / dia (5% do peso corporal). Estimar o estado de hidratação é baseado em sinais clínicos e história. As aves severamente desidratadas (10% +) podem ter olhos enfraquecidos e membranas mucosa pegajosas.

Metade do deficit de fluido total pode ser dado ao longo das primeiras 12-24 horas, juntamente com os requisitos de fluido de manutenção diária. Os restantes 50% são divididos nas 48 horas seguintes, juntamente com os fluidos de manutenção diária.

A solução de Ringer de lactato ou uma solução isotópica equilibrada similar deve ser administrada, aquecida. Os líquidos aquecidos são mais importantes para neonatos e para qualquer administração intravenosa ou intraóssea de fluidos, especialmente em casos de hipotermia ou choque.

Situações clínicas frequentes

Rutura de papo
Se apresenta rutura deve ser reparado cirurgicamente. Existem várias técnicas mas o mais importante é prevenir infeções secundárias e alterações a nível do esvaziamento do papo.

Convulsões
As convulsões podem ser causadas por traumatismos, hipoglicémia, hipocalcémia, virus, parasitas, toxinas, aspergilose, granulomas, otite evoluindo para encefalite, clamidiose, septicémia bacteriana ou podem ser idiopáticas. Convulsões idiopáticas são mais comuns em agapornis e Amazonas. Otite é mais comum em agapornis.

As convulsões em papagaios cinzentos africanos é causada mais frequentemente pela hipocalcémia. Existe uma relação complexa entre a absorção de cálcio, a secreção das glândulas do uropígeo, a ingestão de cálcio, a ingestão de vitamina A e a exposição a luzes de espectro total (especialmente UV-B) ou luz solar natural (não filtrada através de vidro ou plástico). Se possível, extrair sangue para verificar o nível de cálcio no sangue. Um nível normal de cálcio no sangue num cinzento não descarta hipocalcémia.

Hipertermia
Pássaros hipertérmicos (deixados num carro quente, deixados numa gaiola em pleno sol sem sombra, por exemplo) terão pés muito quentes, muitas vezes a pele vermelha, geralmente ofegante. Refrigere o pássaro com compressas frias nos pés, ou mergulhe a metade inferior do pássaro em água fria.

Hipotermia
Hipotermia geralmente ocorre secundariamente à doença, ou em recém-nascidos, pode ocorrer numa chocadeira se não é fornecido com calor adequado. Aquecer lentamente na incubadora.

Retenção de ovo ou dystocia
A maioria das aves estão desidratadas até certo ponto e devem receber fluidos aquecidos, quer pela via SQ, IV ou IO. Os fluidos SQ devem ser administrados com hialuronidase. O lactato de cálcio / glicerofosfato deve ser administrado a 5-10 mg / kg IM ou gluconato de cálcio 50-100 mg / kg IV lentamente ou IM, diluído a 50:50 com soro fisiológico estéril ou LRS. Vitaminas A, D3 e E, 10.000 UI de vitamina A e 1.000 UI de vitamina D3 / 300 gm IM, são dadas para facilitar a absorção de cálcio.

As aves podem apresentar o abdómen inchado, sujidade na cauda, e estará com sinais de prostação e desconforto. Um ovo pode ser palpável no abómen ventral. As radiografias podem confirmar a presença de um ovo retido. Ovos que não são bem calcificados podem ser difíceis de visualizar radiograficamente.

Às vezes, após a rehidratação do pássaro e administração de cálcio parenteral e vitaminas, e colocá-lo com calor e humidade, a ave pode passar com êxito o ovo. Em situações em que o ovo é muito grande, deformado ou a devido à inércia uterina tais medidas não vão ser suficientes.

Dependendo do estado da ave, pode ser aconselhável apenas mantê-la, fornecendo fluidos, calor e humidade, até que ela possa ser transferida para o seu veterinário habitual posteriormente. No entanto, em muitos casos, a ave pode estar bastante debilitada, e no seu melhor interesse deve-se lidar com a distócia imediatamente.

Embora a oxitocina tenha sido classicamente utilizada para distócias, não é o método mais seguro ou mais eficaz de tratamento. Se o ovo parece ser capaz de passar através da pélvis, a entrega do ovo intacto pode ser tentada.

Por vezes recorre-se à ovocentese ou ainda outras abordagens mais invasivas se existe risco de rutura e peritonite.

Queimaduras
Queimaduras de papo ocorrem a partir de aves criadas à mão, fórmula que é usada muito quente, geralmente de pontos quentes em que recorreu ao uso microoondas. Se ele acabou de ocorrer, pode-se lavar com água fria, e começar a terapia com antibióticos e antifúngicos. A cirurgia deve ser adiada até que se tenha desenvolvido fístula. Se executada precocemente existe probabilidade de deiscência.

Conclusão

Embora nem sempre podemos apurar numa primeira abordagem todas as razões pelas quais um pássaro se apresenta numa clínica em situação de urgência podemos também abranger o básico. Em caso de dúvida, mantenha o paciente aquecido, hidratado e contate um veterinário dedicado a aves.

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2. Ritchie, Harrison and Harrison

John Chitty

A Practical Guide to Gut Stasis in Rabbits

This presentation will cover management of gut-related signs and their underlying causes in pet rabbits

The following will be discussed

- Anatomy/ physiology of rabbit gut
- Causes and management of acute gut stasis
- Causes and management of chronic / relapsing gut stasis/ hypomotility

Some reference will be made to other small mammal species where of interest

Acute gut stasis is life-threatening and can be considered an emergency situation in this species. The following notes form this author's step-by-step approach to such presentations

Acute Gut Stasis

1. Clinical Examination. See as soon as possible and assess demeanour before taking history. If necessary place in oxygen chamber, give subcutaneous fluids and warm first.
    a. When handling do not turn over as may lead to gut torsion
    b. Palpate abdomen very carefully to avoid gastric rupture
    c. Assess mucous membranes for cyanosis and observe respiratory pattern in case of excess pressure on thorax.
    d. Percutaneous needle decompression may be used for gastric dilation but should be avoided in caecal dilation. Radiograph first!
    e. If becomes distressed, stop handling and place in oxygen chamber
    f. History: concentrate on
        I. Feeding history- especially house rabbits: check if possibility of foreign body ingestion
        II. Faecal output
        III. Recent management/ temperature changes
        IV. Moulting?

2. Temperature- I only measure if feels cold. If <92˚F then very poor prognosis

3. Bloods-
    a. Glucose
        I <4 hypoglycaemic- chronic malnutrition?
        II 8-15 – normal/ stress
        III 15-20 – needs regular monitoring: if rising may be surgical
        IV 20-25 – probable surgical
        V >25 likely needs immediate surgery

    b. Electrolytes and PCV- assess fluid needs

4. Anxiolytics – midazolam 0.25-0.5mg/kg sc/im.

5. Fluids-
    a subcutaneous – "mild" cases: bright, stable. 30ml/kg
    b Intravenous- more severe cases: rate based on hydration need. If v collapsed assess blood pressure and give colloid as well as crystalloid until blood pressure normal
    c Intra-osseous – if too collapsed for intravenous

6. Analgesia – NSAID best (carprofen 5mg/kg sc or meloxciam 1mg/kg sc). May use buprenorphine (0.01-0.5mg/kg im) / butorphanol (0.2-0.4mg/kg im) if wish to sedate for radiography too

7. Radiograph- usually possible under anxiolytic +/- opiate. If necessary give small amount sevoflurane. If v collapsed perform asap with no sedation. Generally recommend abdominal lateral + DV. However, standing lateral may show fluid lines
    a Foreign body- stabilise quickly and surgery unless glucose <20 (reassess regularly!)
    b Ruptured gut- poor prognosis- operate asap
    c Ileus/ gastric or caecal dilation try medical management

8. Feed- Oxbow Critical Care or Supreme Recovery if capable of tolerating feed. BEWARE hepatic lipidosis

9. Anti-toxin- cholestyramine gel

10. Assess progress!

11. Prokinetics- once sure that no foreign body and properly hydrated
    a Metoclopramide – 0.5-1mg/kg sc bid-tid (or daily dose calculated and given via iv drip) for gastric/ si stasis
    b Cisapride – 1mg/kg po bid caecal stasis
    c Ranitidine – 2mg/kg iv or po; gastric dilation/ suspected ulceration

12. Antibiotics- often not needed. If wish to give then hydrate and give anti-toxins first. Consider trimethoprim-sulphonamide (15-30mg/kg sc bid) or metronidazole 20mg/kg over 12h CRI

13. Collapsed bloated animals/ lack of response.
    a. Anaesthetise – midazolam/ opiate/ sevo or iso
    b. Pass red rubber tube into stomach (gastric bloat) or via rectum (caecal) or both
    c. Gently wash with warmed saline
    d. Insert relevant prokinetic
    e. May add probiotic or antibiotic + fluid (caecum)- decision can be based on cytology of evacuated fluid
    f. May place critical care feed in stomach after emptying

14. Role of probiotics?

15. Once stabilised, investigate underlying factors

John Chitty

Problems of the Geriatric Rabbit

Based on the author's chapter in the BSAVA Manual of Rabbit Medicine

If we do our job right, we will hopefully see a lot of older rabbits! Just as with people, older pets have problems of their own and may be beginning to suffer the consequences of the wear and tear of a long life

So, what is an older rabbit? For the "average" rabbit we would hope to see them live to 10-12 years old. We may therefore start to see geriatric disease from approximately 7 years old. Dwarf and Giant breeds do not have the same life expectancy so these older age diseases may often seen from 4-5 years old

While this talk will discuss the problems generally seen in the older rabbit, we must remember that there are many conditions that affect rabbits of all ages – ie. We must never assume that older animals only get geriatric disease... we must simply add more diseases to the differential list as they get older.

Many of these old-age problems link to pain and so it is important that owners are aware of signs of pain

- Reduced activity
- Weight loss
- Altered mood- either reluctant to move or come to owner, or may become aggressive
- Reduced appetite
- Faecal and urinary changes
-

In summary, any deviation from what is known as normal behaviour is worthy of attention as may be linked to pain

Arthritis

This is one of the most common age-related complaints and can result in periodic bouts of gut stasis or urinary stasis problems as well as generalised pain signs (see above)

Rabbits rarely if ever vocalise pain- just because they do not complain does not mean they are not in a lot of pain and radiography is indicated for any animal showing potential pain signs or disease that may be linked to an underlying focus of pain

Joints typically affected by osteoarthritis in the older rabbit are

- Spine (all parts!)
- Stifle joints
- Hips
-

Giant breeds have particularly bad problems

Arthritis is managed, not cured, and many approaches are taken

- Pain relief- non-steroidal drugs and oral opiates are most commonly used
- Diet- a good quality high fibre diet to avoid obesity
- Nutraceuticals- glucosamine and essential fatty acids are unproven, but may be of some benefit in reducing NSAID dose
- Acupuncture has benefitted some spinal cases in this author's opinion
- Daily management. Address flooring to avoid slipping and injury of smooth floors. Avoid climbing steps or having to jump up stairs or on furniture. Careful handling is always required to avoid further injury and gentle ramps can be used to avoid the need to jump up/ down steep inclines

Dental disease

Older rabbits may actually be at less risk of dental disease than younger- having got this far without problems, they are presumably unlikely to be faced with the predominant risk factors for dental disease!

However, many rabbits may enter older age with pre-existing dental disease and this may require more sensitive management as other concurrent diseases occur. In particular, altered anaesthetic risk with certain disease may mean more careful attention, or altered regimes when anaesthetising older rabbits for regular dental care

Heart

Cardiac disease is increasingly diagnosed. This likely reflects improved diagnostic capabilities and understanding as well as an increasing number of animals living long enough to develop such problems

Typical signs are of a quieter animal often losing weight (or failing to gain weight). There are often few other signs! On examination they are often slightly thin and heart murmurs may or may not be present- these are usually soft. If present and not associated with anaemia they are normally significant

Ultimately diagnosis depends on imaging- radiography and ultrasonography. The latter is especially useful in monitoring disease progress. ECG can be very useful where dysrhythmias are present, and also provide a non-invasive means of monitoring.

Bloods are of use in determining renal function- heart and renal disease are often seen together and will affect prognosis

Most cases appear to be typical of a hypertrophic cardiomyopathy. Long term prognosis is therefore poor. However, in many cases quality of life can be significantly improved in the short-to medium-term. This author typically uses ACE-inhibitors with dieresis if congestion and pimobendan used when signs worsen

Liver

Liver disease is relatively unusual in older age and is certainly difficult to diagnose on bloods or imaging
Most importantly, many overweight rabbits may be suffering from subclinical hepatic lipidosis. Anorexia for any reason may therefore trigger clinical disease

Kidney

Renal disease is common. As well as chronic renal failure associated with renal fibrosis, disease may also be seen as a result of chronic (or previous) encephalitozoonosis and secondary to urinary flow problems (see above) with development of uroliths throughout the urinary tract and chronic inflammatory changes that may result in an ascending infection

Renal disease may be diagnosed by blood biochemistry or radiography. It is certainly to be suspected in all thin quiet animals especially if there is polyuria/polydipsia and/or apparent urinary incontinence/ scalding

The cause of any urinary flow problem must be addressed in order to alleviate problems. Uroliths should be managed medically or surgically as necessary. Encouragement to move around and to drink (water should be provided in both bowls and drinkers to enable the rabbit to choose its own preferred route) are important aspects of management as is analgesia. Diuresis can be a help in many cases- dandelions, dandelion extracts and thiazide drugs may all have a place

Benazepril appears an effective aid in chronic renal disease

Fluid therapy is required in dehydrated animals

As with heart disease, the condition is often managed rather than cured and owners should be advised accordingly

Eyes

Ocular disease, especially cataract formation, is common

Some cataracts in larger animals may be amenable to surgery. Diabetic cataracts are rare. Cataract secondary to encephalitozoonosis are seen but are more likely in younger rather than older animals. They are certainly unlikely to respond to fenbendazole which should be reserved for cases where encephalitozoonosis has been definitely identified and there is active infection

Otherwise, many rabbits do very well with reduced or no vision. Care must be taken not to make major changes in environment as this may cause distress. Food and water bowls must be left in the same familiar places

Neoplasia

Tumours are a natural consequence of living longer. Reproductive tumours (eg testicular or uterine adenocarcinoma) are avoided by early neutering. However, many other neoplasms are seen – benign and malignant
Any unusual swelling or mass should be investigated in much the same way as in other species

John Chitty and Aidan Raftery

Diseases of the Ear

(based on the authors' chapter in the BSAVA Manual of Rabbit Imaging, Surgery and Dentistry)

Introduction

Ear disease is commonly seen in practice. Traditionally this is described as otitis, and this may be the case in some instances, however there is a growing body of evidence to show that the majority of cases are linked to non-drainage and build up of cerumen in lop-eared rabbits, or those with short noses and occluded Eustachian tubes.

External ear disease (EED) may manifest as irritation of the ear or head shaking; or may be detected on auriscopic examination in asymptomatic rabbits. It should be stressed that subclinical EED should not be treated but monitored on a regular basis- therapy can be given either on onset of clinical signs or if cytology of the exudates confirms it is inflammatory rather than cerumen

Subclinical EED can be defined as

- Presence of larger than normal amounts of white cerumen in the canal
- Variable degrees of canal wall erythema
- No clinical signs associated with the ears

Middle ear disease (MED) may be seen as a cause of chronic poor-doing (weight loss, persistent/ recurrent gastrointestinal hypomotility, reduced appetite (possibly due to the proximity of the tempero-mandibular joint to the middle ear, or due to facial neuritis), parasitic dermatoses, etc. It may also be seen as a cause of head tilt or torticollis. Atrophy of facial muscles caused by facial neuritis may be evident in some rabbits as a sequel to MED , though initially this may present as drooping of muscles on the affected side before atrophy occurs resulting in "twisting" of the face toward the affected side

Otitis interna signs are very similar to MED with head tilt and torticollis and, potentially, evidence of a vestibular syndrome

Underlying causes of ear disease include

- Mite infestation with Psoroptes cuniculi. This will cause a distinctive crusting otitis externa. Secondary bacterial infection may cause further external ear inflammation or a secondary otitis media
- Irritation (or even potential, though as yet unproven, allergy) of the external ear canal. While analogous to the situation in other species, there appears little evidence for this at present
- Amputation or damage of the ipsi-lateral hindlimb. This makes it hard for the rabbit to scratch the ear as part of normal grooming, and contributes to the non-clearance of cerumen.
- Anatomy. EED and MED are highly prevalent in lop-eared rabbits compared to those with "normal" ears. The anatomy and predisposing factors will be discussed.

Investigation

Otoscopy

- An otoscope with channels for instruments and for flushing will make otoscopy more effective. The light source used needs to be very bright for good visualisation
- In most cases the tympanic membrane is obscured due to purulent accumulations and stenosis due to inflammation
- General anaesthesia is required in most rabbits to fully evaluate the external ear canal and the tympanic membrane especially of cleaning is required to allow visualisation
- Normal ears are a pink colour with smooth lining and with minimal exudate present. Lop eared rabbits usually have much more ceruminous exudate present and the lining is often a red colour
- Otoscopy may reveal abnormal discharge/exudate, masses, ulceration, stenosis, foreign body
- Flushing may be necessary to view and evaluate the tympanum but the exudate is usually very thick and requires gentle removal with cotton tips. The lining of the external ear canal may be inflamed and rough technique will cause further swelling and bleeding
- If there is otitis media with the middle ear filled with material then the tympanum will look darker than normal and sometimes may be seen to bulge outwards
- Otoscopy facilitates the accurate collection of samples for cytology, culture, and the biopsy any abnormal mass.

Culture

- A swab is inserted through a sterile otoscope cone or
- A sterile catheter (attached to syringe) is passed through working channel of endoscope and a sample aspirated
- The first sample should be used for culture and the second sample for cytology.
- If otitis media is present with no otitis externa then take a deep nasal swab in addition to a sample direct from the middle ear via a myringotomy incision
- The use of an otoendoscope facilitates safe myringotomy. A 3 French Tom cat catheter is passed through the instrument channel and used to make an incision through the caudoventral quadrant of the tympanic membrane. Samples are collected by aspiration.
- NB. Studies in the authors' clinics have revealed a very wide range of bacteria cultured from "normal" rabbit ears. Many of these (especially Pseudomonas) would be regarded as pathogenic in other species- it is therefore vital that cytology is performed alongside culture to assist in interpretation.

Imaging

Radiographs are essential in the investigation of otitis. With otitis media there may be an increased fluid density in the bullae. Lysis and remodelling of the bones may be visible in some cases. The following views are usually required

- DV
- Lateral
- Obliques X 2
- I

Radiography often gives false negatives as the changes are not always sufficiently advanced or mineralised to be visible radiographically

CT evaluation if available will provide further diagnostic information. In particular, CT can provide detailed information regarding the bullae and middle ear- it is generally regarded as the most sensitive diagnostic tool in such cases especially as it shows the presence of soft exudates (pus/ exudates) more clearly than radiography.

Contrast Radiography

- Canalography
- Of use when investigating otitis usually to evaluate if the tympanic membrane is intact and it may provide information about the the tympanic cavity and its current margins, though cerumen may also block passage of contrast, and this technique is inferior to CT

Haematology, biochemistry and serology

- To establish a possible underlying cause e.g. serology for encephalitozoonosis as a negative result will effectively rule this out as a cause of neurological signs
- To identify other ongoing and/or underlying disease processes
- These tests will assist with formulating a treatment plan that is in the best interests of the animal

Neurological exam

It is important to identify any neurological deficits before surgery. They will alter the prognosis and may be misidentified after the surgery as surgical complications.

- Facial nerve
- Vestibular deficits
- Paralysis of the facial muscles and an exposure keratitis due to inability to blink. See the complications section of the bulla osteotomy section
- Head tilt, nystagmus and torticollis.
- Paralysis of the tongue.
- Horners syndrome.

Techniques

All cases of true otitis (other than Psoroptes- associated) will require systemic antibiosis and non-steroidal anti-inflammatory drugs. In some cases these will be sufficient to control clinical signs. These are difficult cases to manage –decision-making for antibiotic choice is similar to that for abscesses and it is hard to determine how long these should be used for as these rabbits will always have subclinical aural disease.

Topical squalene based ear cleaners have proved of great use in helping reduce build up of cerumen- these are most effective when used after saline canal flushes.

Syringing

General anaesthesia is normally required to allow full examination of external ear canal and, especially, visualisation of the tympanic membrane

Sampling of cerumen/ aural discharges is essential at this stage (see above)

In severe cases it is also necessary in order to properly examine the ear canal

Removal of pus and aural discharges will also assist in management of the case by reducing infective load, enabling better penetration of therapies (both topical and systemic) and reducing localised irritation caused by the build up of discharge. In cases of otitis media where there is rupture of the tympanic membrane, syringing of the ear will also remove debris from the bulla thereby reducing pressure on the facial nerves

It is not, however, sufficient as primary therapy as it does not address primary causes of otitis externa/ media

Syringing of the ear is carried out in similar fashion to other species.

Saline is used as ototoxicity of ear cleansing solutions used in dogs and cats does not appear to have been evaluated in the rabbit (NB. Chlorhexidine must NEVER be used- this author (JC) has seen one case of this causing ototoxicity). This is especially important where the tympanic membrane may be ruptured

Wick placement
Polyvinyl acetate sponges (Ear Wick, Dermapet) may be used in bothEED and MED.

These are used following thorough cleaning of the ears under anaesthesia (as above)

In essence they are dry sponges that are inserted in the ear canal (or deeper into the bulla if the tympanic membrane is ruptured) and then "filled" using antibiotic solution. This is then held in situ over a period of time enabling continuous localised antibiotic therapy deeper in the ear

In rabbits, the standard dog sponge is cut in half

Choice of antibacterial is based, as ever, on culture and sensitivity combined with cytological findings. However, choices must be tempered with avoiding ototoxicity

Therefore trimethoprim-sulphonamide injectable solutions (eg. Norodine 24, Norbrook, UK) appear safe and suitable as a first choice in most cases. If required, TRIS-EDTA solution may be added to the wick as well, in order to potentiate the antibiotic's effect, but avoid preparations containing chlorhexidine.

The wick should be removed after a week- longer periods in place may result in excessive granulation around the wick making it hard to locate and remove. In one case this author (JRC) has seen, failure to remove the wick resulted in abscessation and "expulsion" of the wick via the skin from the vertical canal. However, removal at 7 days is normally straightforward

Wick removal should be carried out under anaesthesia allowing for further cleaning of the ear at the same time
If necessary another wick may be placed at this stage.

The main complications seen are

- Irritation caused by presence of the wick. However, this is unusual and normally stops after the first 24-48 hours. If it continues then the rabbit should be re-anaesthetised and the wick removed
- Premature loss of the wick. Some wicks may be removed by head shaking/ grooming

While this technique is relatively new and evaluation largely subjective it appears useful and certainly appears to prolong intervals between ear cleaning episodes in chronic cases

Ear diverticula
Lop eared rabbits often present with fluid-filled swellings at the base of the ear. These are often described as abscesses and treated by lancing

In reality they represent a build up of cerumen at the junction of cartilaginous auditory meatus and tragus. As this region is soft walled at this point it is liable to become distended forming a diverticulum or even hernia of the ear canal wall

Some cases will become secondarily infected

If there are no associated clinical signs, the lump may be left alone other than teaching the owner to massage it gently to move the cerumen into the more distal parts of the ear canal. This may prevent "stagnation" of the cerumen and also reduce the rate of further stretch of the ear canal- squalene based cleaners may assist with this.

If there are clinical signs, rather than lancing, which will further damage the ear canal wall, the material should, initially be treated as in any case of otitis (see above). The presence of bacterial infection should be determined by culture and cytology. Otherwise material may be removed by syringing and wicks may be placed if necessary

In cases that are refractory to this therapy or persistently recur, possibly due to chronic over-distension of the ear canal wall, lateral wall resection may be indicated.

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Lateral wall resection

In severe or chronic cases of EED where regular cleaning and wick placement in combination with systemic therapy are not controlling the condition, or where cleaning is required at such frequency that it is felt the rabbit's welfare may be compromised, lateral wall resection may be considered. This may also be indicated for tumours of the lateral wall of the ear canal, though these are extremely rare, and diverticula.

As in other species, this technique is not curative (other than for tumour removal)- instead it is designed to improve exposure of the vertical canal and improve access to the horizontal canal

This enables easier cleaning and application of topical therapies greatly facilitating conservative management of these cases. In rabbits, where the primary problem is one of anatomy, then this surgery may be more effective.

Total ear canal ablation

Indications

- MED non responsive to non-surgical treatments
- Benign neoplasms

Contraindications

- Otitis interna
- Lytic bone changes of the medial aspect of the tympanic bulla or deeper
- Extensive proliferative bone changes
- Neoplasia (unless there is a rational treatment plan)
- Vestibular symptoms
- Other severe intractable disease processes

Preparation

A comprehensive workup is essential before embarking on surgery of the middle ear.

- Radiology of the skull to review the extent of any bone changes
- Computed tomography (CT) where available can provide important information to aid in decision making.
- A neurological examination is important to identify any neuropathies already present before the surgery
- Ideally the pathogenic bacteria present should be cultured and its antimicrobial sensitivity identified before the surgery

Lytic changes to the bone of the lateral wall of the bulla due to chronic osteomyelitis will require thorough debriding. A thorough review of the complex anatomy of this area is strongly advised before the surgery.

A ventral bulla osteotomy is indicated when there is an intact tymphanum and no involvement of the external ear canal. The ventral approach provides better access to the bulla facilitating a more reliable removal of tissue debris, necrotic material and the epithelial lining of the bulla. There is also better drainage after the surgery and the risk of damage to the facial nerve is reduced. However there is risk of damage to the hypoglossal nerve. Occasionally on curettage of the bulla the tympanic membrane ruptures. This will increase the risk of ongoing chronic otitis media requiring a repeat procedure. If there is rupture of the tympanic membrane during a ventral bulla osteotomy then the surgeon should also perform a total ear canal ablation.

Total ear canal ablation and a lateral bulla osteotomy is indicated when the tymphanum is ruptured and the disease process involves the external ear canal.

Complications

Neurological deficits are common and will resolve within 14 days unless a nerve has been transected. Facial nerve deficits and peripheral vestibular signs are most commonly seen.

The facial nerve can easily be damaged during the surgery as the external ear canal is being dissected. In some cases there is lysis of the lateral wall of the tympanic bulla which may just crumble during the surgery. This poses a greater risk to the facial nerve as it exits through the stylomastoid foramen on the caudal border of the lateral wall of the bulla. The clinical signs seen include facial paralysis and an absent or reduced palpebral reflex. Ocular lubricants will reduce the risk of exposure keratitis. If there is drooling due to a facial nerve damage regular cleaning of the chin will help prevent dermatitis in the area.

Peripheral vestibular deficits are caused by damage to the dorsomedial area of the bulla during curettage. Signs seen are horizontal nystagmus, ataxia and head tilt.

Also incisional infection, pinna necrosis, haematoma, hearing loss and salivary gland damage can be seen. Pinna necrosis is due to vascular damage and in the rabbit may require removal of the pinna

Fistula formation is a chronic complication that can be seen months after an apparently successful surgery. A repeat bulla osteotomy is usually required to resolve it

Futuro e a Medicina Veterinária

Mário Almeida

Projecto para a Presença de Animais de Companhia em Ambiente Hospitalar

Resumo

Abordagem à presença de animais de companhia em ambiente hospitalar. Conceitos de Atividades Assistidas por Animais (AAA) e Terapia Assistida por Animais (TAA).

Vantagens para os doentes, seleção de animais adequados aos fins a que se destinam e controlo de riscos envolvendo o Médico Veterinário como pedra basilar dessa segurança.

Experiências noutros países, nomeadamente o projecto–piloto "Pet Therapy" no Hospital Albert Einstein, em S. Paulo, Brasil .

Proposta de documento único que acompanha tutor/animal nas visitas. Contextualização, regras e restrições, atestado sanitário médico-veterinário, termo de compromisso do tutor, decisão final e limitações.

Project for the implementation of a Pet Presence program within Health Care Facilities
Summary
Overview of the benefits of the use of pets in Health Care Facilities to improve patient recovery and\or welfare.
Explanation of the concepts of Animal-Assisted Activities and Animal-Assisted Therapy.
Benefits for the patients, adequate animal selection, risk assessment and control by the Veterinary Surgeon to ensure correct implementation of the project.
The use of Animal-Assisted Activities and Animal-Assisted Therapy in other Countries, namely the trial project "Pet Therapy" at São Paulo´s, Brazil, Albert Einstein Hospital.
Proposal for a unique document to accompany handler and animal during visits that will contain all necessary information, i.e. best practices, Veterinary Health Certification, handler´s responsibilities and ultimate decision-making requirements by the Medical Staff.

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Joaquim Henriques

O Futuro no Tratamento das Doenças Oncológicas

ORIGINAL RESEARCH ARTICLE

Front. Vet. Sci., 30 March 2015 | http://dx.doi.org/10.3389/fvets.2015.00004

Regenerative approach to bilateral rostral mandibular reconstruction in a case series of dogs

 **Boaz Arzi**[1]*, **Derek D. Cissell**[1,2], **Rachel E. Pollard**[1] and  **Frank J. M. Verstraete**[1]

[1]Department of Surgical and Radiological Sciences, School of Veterinary Medicine, University of California Davis, Davis, CA, USA

[2]Department of Biomedical Engineering, University of California Davis, Davis, CA, USA

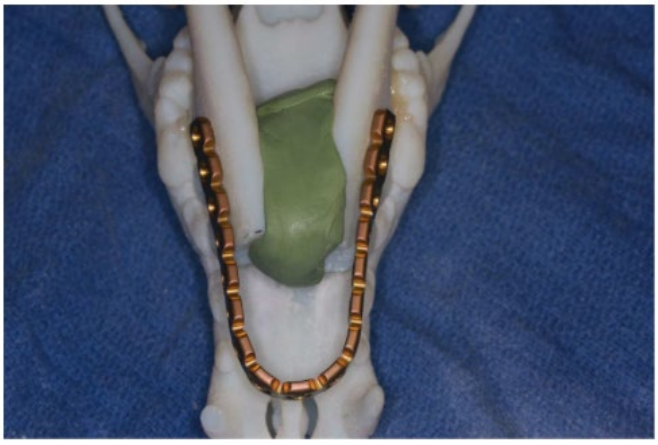
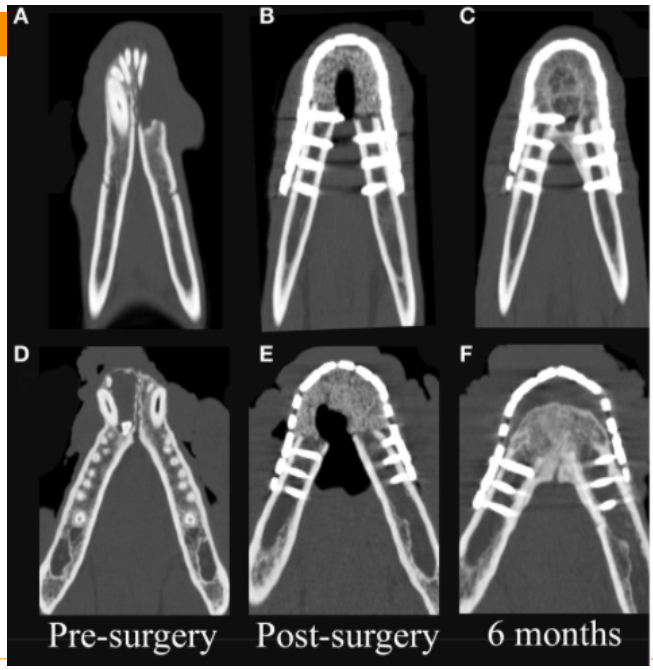


FIGURE 1 | Surgical planning on a 3D printed skull demonstrating the adjustment and adaptation of the titanium locking plate to the model of the rostral mandibles.



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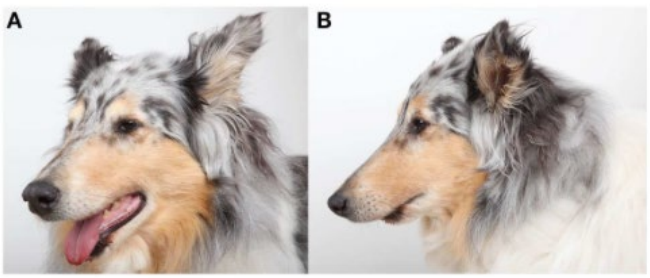
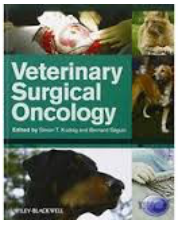
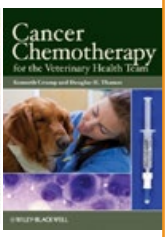


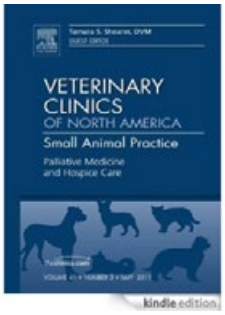
FIGURE 3 | Photographs of dog 2 at 6 months recheck illustrating the appearance of the mandibles and skull following successful reconstruction of the rostral mandibles.

6 6

E o futuro...?!

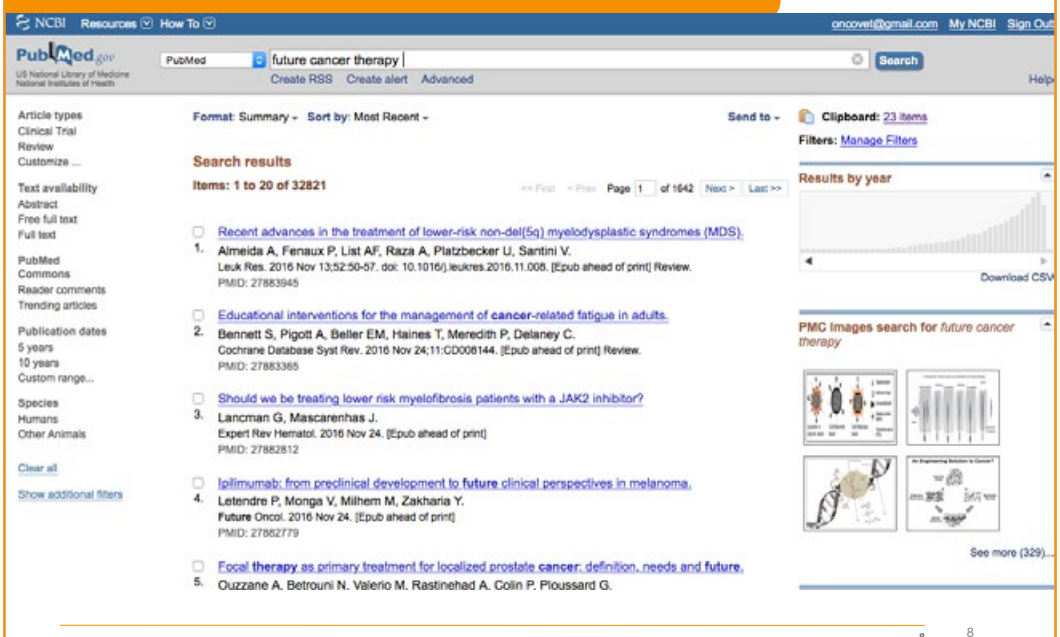






http://www.theprosperitycommunity.com/blog/tag/teamwork/

7 7



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Immunotherapy with a HER2-Targeting Listeria Induces HER2-Specific Immunity and Demonstrates Potential Therapeutic Effects in a Phase I Trial in Canine Osteosarcoma.
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future veterinary cancer therapy (154)
PubMed
future cancer therapy (32821)
PubMed
future cancer treatment (63410)
PubMed
See more...

9 9

Conhecer o Passado...

O Cancro

Particularidades Biológicas
Particularidades Clínicas

Compreender como a doença ocorre e como se alastra no organismo são a chave para o desenvolvimento de novas terapêuticas e para o adequado acompanhamento clínico ao paciente.

Mas o que é o cancro e como se desenvolve?

11

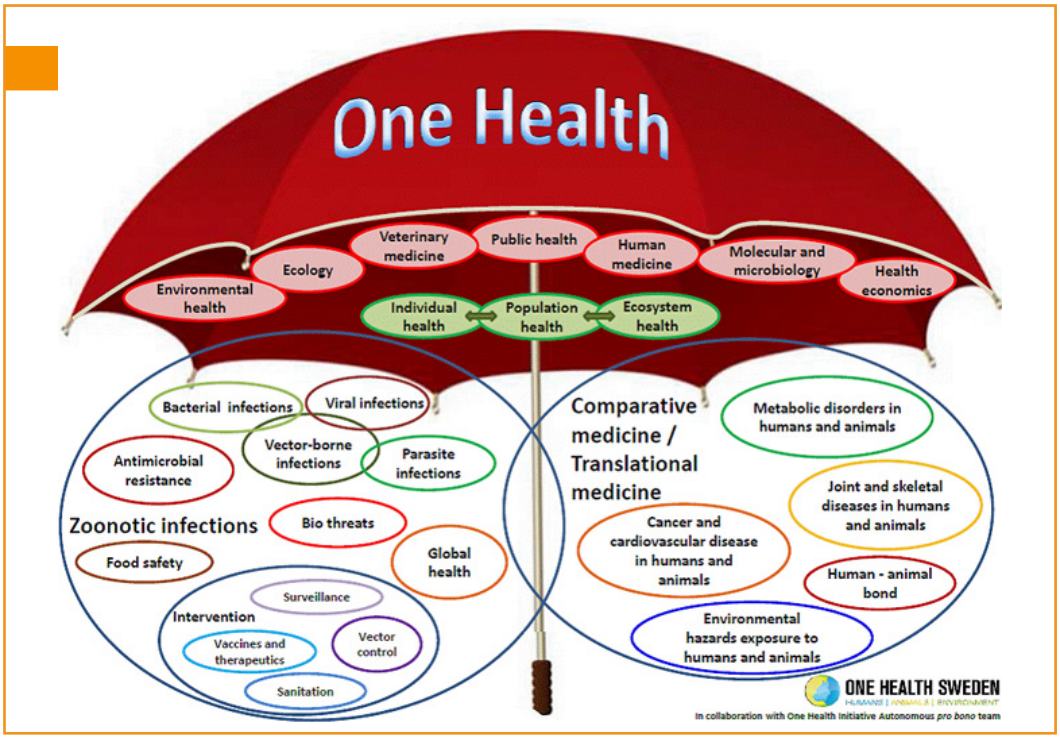
Background

Durante décadas o cancro foi aceite como sendo o produto final da acumulação de anomalias genéticas

- Desenvolvimento tecnológico desponivel após a década de 60 do século passado permitiu a identificação e descoberta de anomalias genéticas associadas a determinados tipos de tumores
- Mais recentemente, e de forma mais ampla, a sequenciação do genoma tumoral permitiu identificar aberrações genómicas associadas aos tumores

Novos conceitos e áreas de investigação estão em desenvolvimento permitindo um olhar mais profundo na biologia tumoral (oncobiologia) (-omics)

12



Background

A Oncologia Veterinária está de mãos dadas com a Oncologia Humana

14

Background

Como médicos veterinários a nossa missão é contribuir para a saúde animal sem compremeter o seu bem-estar

Na Oncologia esta filosofia é muito importante:

Os animais não percebem que têm uma doença mortal:
- não ficam com insónias ou stress associado

Os animais não vivem no futuro ou passado:
- Vivem no presente!

Os animais não podem decidir o rumo da sua vida:
- Os cuidadores têm a primeira e última palavra
- A escolha terapêutica é "cuidador-condicionada)

Os animais dependem dos cuidadores para a medicina preventiva
- Exames frequentes em doentes de risco
- Consultas geriátricas

15

Background

Portanto, tendo em conta o bem estar do doente:

Os veterinários devem ter consciência da "dose terapêutica":

- Dose de terapia cirúrgica
- Dose de terapia médica
- Dose cumulativa de "terapia"

Objectivo: Máxima qualidade de vida e não quantidade de vida sem qualidade

- Quando e como?
- Despedir-se...

17

Background

A medicina é a ciência onde os avanços acontecem todos os dias!

A Investigação Clinica e Básica revelam descobertas constantemente muitas vezes implicando mudanças de diagnóstico ou terapia , com vista , sempre, ao melhor desfecho clínico.

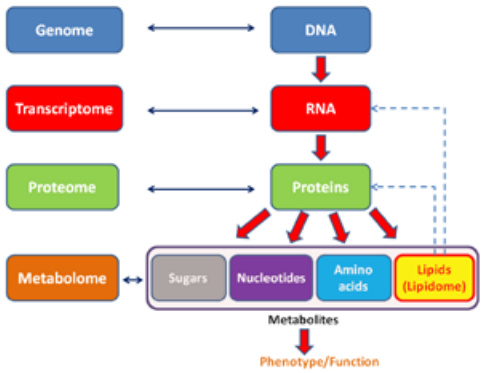
A Oncologia é uma das áreas em que mais se investe em Ciência:

- **Impacto Social**
- **Impacto Económico**

18

Background

Tecnologias complexas permitem-nos descobertas maravilhosas na biologia celular (genomics, metabolomics, proteomics, etc)



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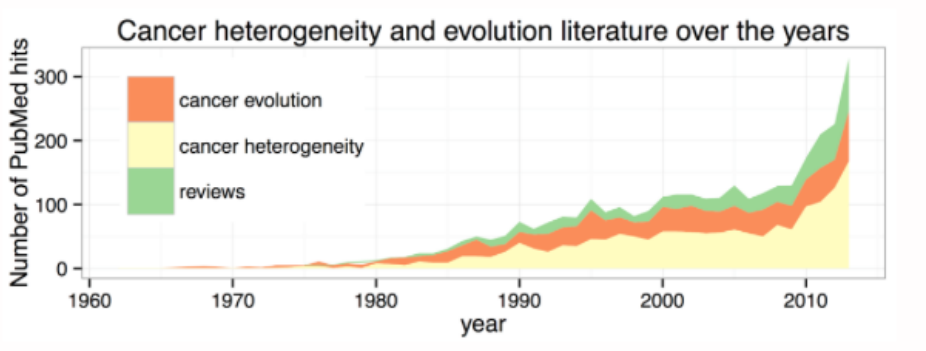
Wikipedia.org

Background



https://scientificbsides.wordpress.com/author/florianmarkowetz/

20

Background

Mas...apesar de tanto conhecimento, ainda nos deparamos com o insucesso...

- A incidência do cancro continua a aumentar
  - Porque os animais vivem mais?
- A taxa de mortalidade em determinados tipos de tumores não diminuiu
  - Linfoma; OSA, HSA
- A descoberta e utilização com novos fármacos ou protocolos nem sempre está associada a sucesso
  - OSA, Linfoma
- Alguns (se não, todos) tumores têm comportamento clínico atípico
  - MCT
  - SCC
  - Melanoma

21

Background

22

Background

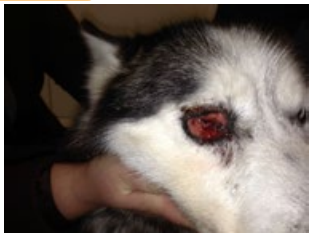
Diagnóstico:

Argumentos clínicos:

Sintomas , História Clínica, Imagiologia

Argumentos moleculares:

Informação laboratorial



Veterinary and Comparative Oncology

Original Article

European consensus document on mast cell tumours in dogs and cats

L. Blackwood[1,2], S. Murphy[2,3], P. Buracco[4], J. P. De Vos[5], P. De Fornel-Thibaud[6], J. Hirschberger[7], M. Kessler[7], J. Pastor[8], F. Ponce[9], K. Savary-Bataille[10] and D. J. Argyle[11]

23

Table 2. Cell proliferation markers with a prognostic value in cutaneous mast cell tumours in dogs

Parameter	Significance	Comment	References
Mitotic index (MI)	>5 prognostic for reduced survival independent of grade. >7 predictive for recurrence.	Useful test that can be carried out on routine histological sections. Some recommend 7 as cut off, rather than 5.	24, 36–38
Ki-67	High Ki-67 expression is associated with increased mortality, recurrence and metastasis. Prognostic factor independent of histological grade.	Useful if available as proven independent of grade.	39–41
Agyrophilic nucleolar organiser regions (AgNORs)	Higher AgNOR counts associated with increased likelihood of death, recurrence and metastasis.	Not a prognostic indicator independent of histological grade, but may support decision making for grade II tumours.	40, 42
Proliferating cell nuclear antigen (PCNA)	Increased PCNA expression associated with increased mortality. Not consistently with increased risk of recurrence or metastasis.	Not a prognostic indicator without histological grade. Not predictive of survival.	39, 40, 42

24

Background

Diagnostico:

Os argumentos moleculares estão a sobrepor-se aos argumentos clínicos!!

Published OnlineFirst June 19, 2013; DOI: 10.1158/0008-5472.CAN-12-3546

Molecular and Cellular Pathobiology

Cancer Research

Gene Profiling of Canine B-Cell Lymphoma Reveals Germinal Center and Postgerminal Center Subtypes with Different Survival Times, Modeling Human DLBCL

Kristy L. Richards[1,3,4], Alison A. Motsinger-Reif[4,5,9], Hsiao-Wei Chen[3,6], Yuri Fedoriw[2,3], Cheng Fan[3], Dahlia M. Nielsen[5,10], George W. Small[1,3], Rachael Thomas[4,7], Chris Smith[5], Sandeep S. Dave[12], Charles M. Perou[2,3], Matthew Breen[3,4,7], Luke B. Borst[4,11], and Steven E. Suter[3,4,8]

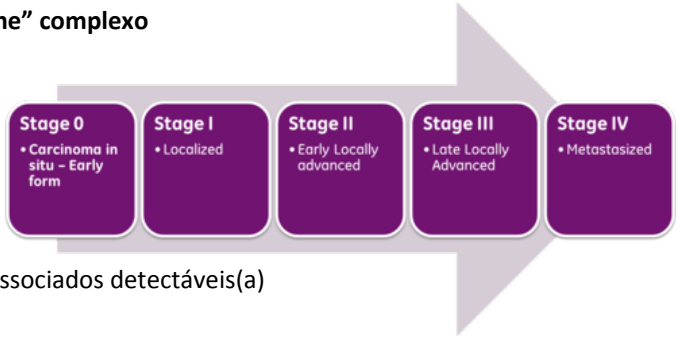
25

Background

O Cancro é um "síndrome" complexo

Tumor
Paraneoplasia
Caquexia

Estadiamento:



Pré- clinico: sem sinais associados detectáveis(a)
Clinico (b)
compensado
descompensado

26

Background

24 February 1956, Volume 123, Number 3191

SCIENCE

On the Origin of Cancer Cells

Otto Warburg

Injuring of Respiration

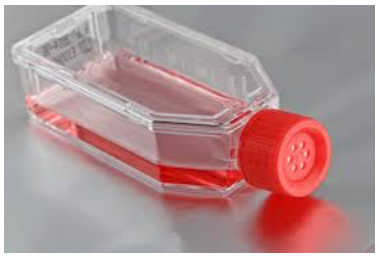
Since the respiration of all cancer cells is damaged, our first question is, How can the respiration of body cells be injured? Of this damage to respiration, it can be said at the outset that it must be *irreversible*, since the respiration of cancer cells never returns to normal. Second, the injury to respiration must not be so great that the cells are killed, for then no cancer cells could result. If respiration is damaged when it forms too little adenosine triphosphate, it may be either that the oxygen con-

27

Background

"Cancro Clínico) – Bedside

" Cancro Molecular"– Benchside



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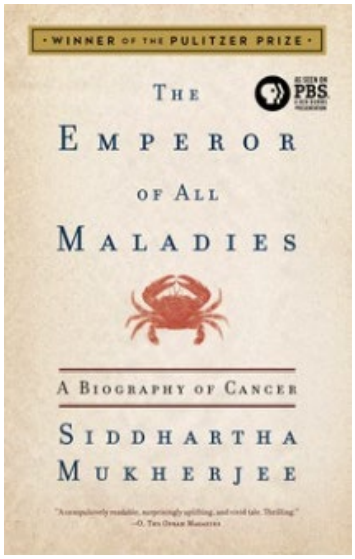
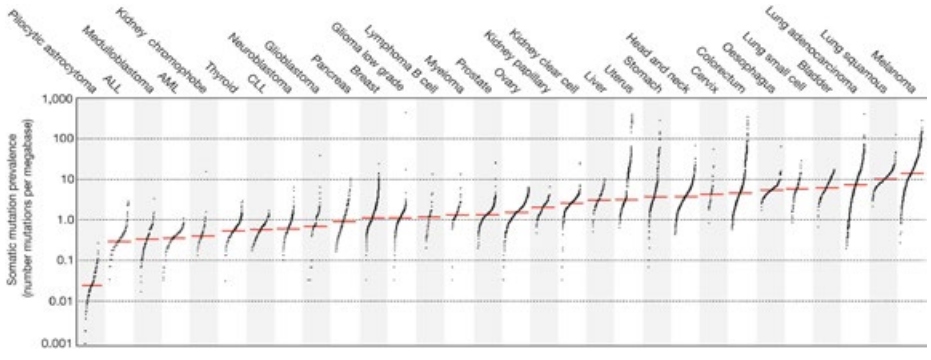
Background



29 29

Prevalência de mutações somáticas em determinados tipos de cancro.



LB Alexandrov *et al. Nature* **000**, 1-7 (2013) doi:10.1038/nature12477

nature

Background

Brücher and Jamall *BMC Cancer* 2014, **14**:331
http://www.biomedcentral.com/1471-2407/14/331

BMC Cancer

HYPOTHESIS **Open Access**

Epistemology of the origin of cancer: a new paradigm

Björn LDM Brücher[1,2,3,4,5,6,7*] and Ijaz S Jamall[1,2,3,4,5,6,8*]

Abstract

Background: Carcinogenesis is widely thought to originate from somatic mutations and an inhibition of growth suppressors, followed by cell proliferation, tissue invasion, and risk of metastasis. Fewer than 10% of all cancers are hereditary; the ratio in gastric (1%), colorectal (3-5%) and breast (8%) cancers is even less. Cancers caused by infection are thought to constitute some 15% of the non-hereditary cancers. Those remaining, 70 to 80%, are called "sporadic," because they are essentially of unknown etiology. We propose a new paradigm for the origin of the majority of cancers.

Presentation of hypothesis: Our paradigm postulates that cancer originates following a sequence of events that include (1) a pathogenic stimulus (biological or chemical) followed by (2) chronic inflammation, from which develops (3) fibrosis with associated changes in the cellular microenvironment. From these changes a (4) pre-cancerous niche develops, which triggers the deployment of (5) a chronic stress escape strategy, and when this fails to resolve, (6) a transition of a normal cell to a cancer cell occurs. If we are correct, this paradigm would suggest that the majority of the findings in cancer genetics so far reported are either late events or are epiphenomena that occur after the appearance of the pre-cancerous niche.

Testing the hypothesis: If, based on experimental and clinical findings presented here, this hypothesis is plausible, then the majority of findings in the genetics of cancer so far reported in the literature are late events or epiphenomena that could have occurred after the development of a PCN. Our model would make clear the need to establish preventive measures long before a cancer becomes clinically apparent. Future research should focus on the intermediate steps of our proposed sequence of events, which will enhance our understanding of the nature of carcinogenesis. Findings on inflammation and fibrosis would be given their warranted importance, with research in anticancer therapies focusing on suppressing the PCN state with very early intervention to detect and quantify any subclinical inflammatory change and to treat all levels of chronic inflammation and prevent fibrotic changes, and so avoid the transition from a normal cell to a cancer cell.

Implication of the hypothesis: The paradigm proposed here, if proven, spells out a sequence of steps, one or more of which could be interdicted or modulated early in carcinogenesis to prevent or, at a minimum, slow down the progression of many cancers.

Keywords: Cancer, Paradigm, Inflammation, Fibrosis, Carcinogenesis, Tumor, Neoplasm

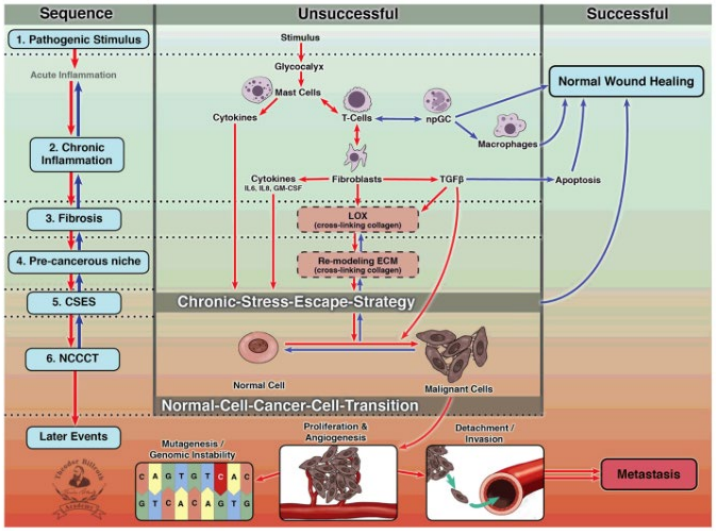


Figure 1 Schematic drawing of "Epistemology of the Origin of Cancer". Abbreviations: CSES, chronic stress escape strategy; NCCCT, normal cell cancer cell transition; npGC, neutrophil Granulocyte; TGFβ, tumor growth factor beta; LOX, Lysyl oxidase; ECM, extracellular matrix.

Brücher and Jamall *BMC Cancer* 2014, **14**:331

Background

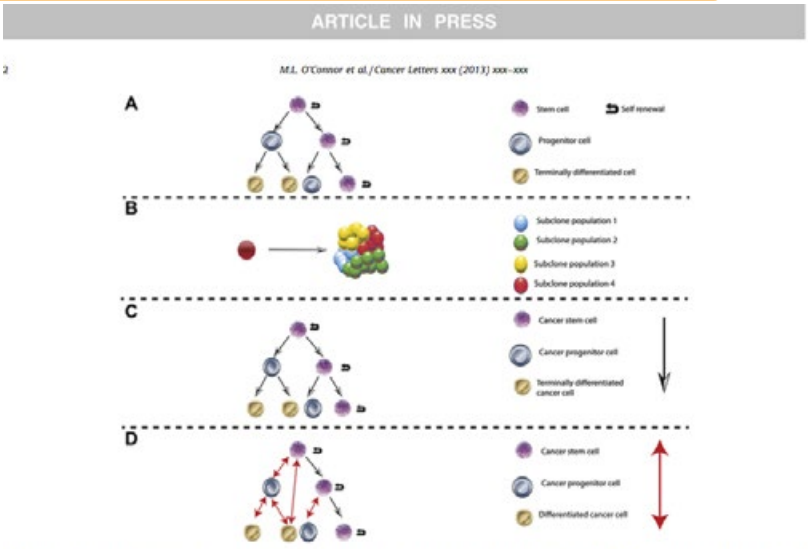


Fig. 1. Normal tissue hierarchy and evolving cancer models. (A) Normal stem cell hierarchy. Normal tissue composition is defined by a cellular hierarchy stemming from a single cell that can undergo unlimited self-renewal and create entire progenies of differentiated cells. (B) The clonal evolution model, where all cells are derived from a single cell and are divided into subpopulations resulting from specific mutations and selection fitness. (C) The original CSC model, a mirage to the cellular hierarchy in normal tissue, with the exception of unregulated control of CSC driving tumourigenesis. (D) The fluid CSC model, in which both progenitor cells and differentiated cells are able to re-acquire self-renewal potential.

33 33

Background

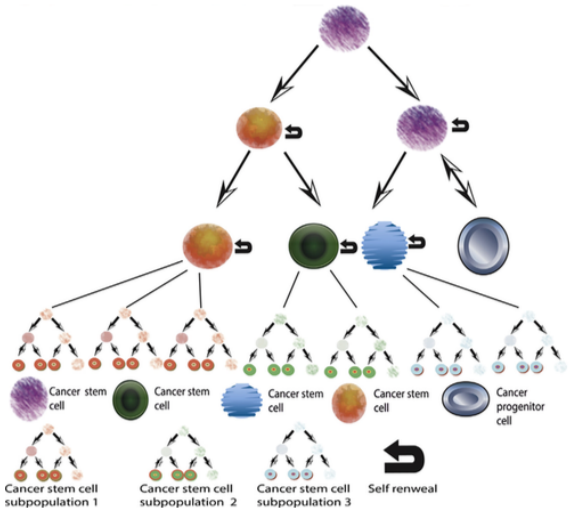


Fig. 3. The convergent CSC model. This model suggests that clonal evolution and the CSC model are both involved in tumour development forming a cellular hierarchy within a tumour and are likely to involve different subclones depending on the mutations/epigenetic changes occurred in specific cells with self-renewal capacity.

34 34

Background

Um tumor....

Diferentes acontecimentos.....

35 35

Background

As células normais têm um número variado de defesas contra a transformação neoplásica:

- Várias alterações têm que ocorrer antes que comecem a crescer descontroladamente para formar um tumor.

Em Janeiro de 2000 foi elaborado um documento cientifico que congrega as mais recentes descobertas sobre a evolução fenotipica que fazem do cancro “um cancro”.

Este Documento ficou conhecido como os 6 pilares do cancro (Hallmarks)

Acredita-se que estes principios governam a biologia de todos os tumores , simplificando e unificando uma miríade de doenças que caem debaixo da designação de cancro.

36

Background

The Hallmarks of Cancer:

Documento científico fundamental em oncobiologia!

Acessado 20.000 vezes(2004-2007)
15.000 citações

T

37

Background

Princípio do "Hallmarks of Cancer":

A complexidade molecular e clínica do cancro pode tornar-se compreensível através de um numero pequeno de princípios comuns.

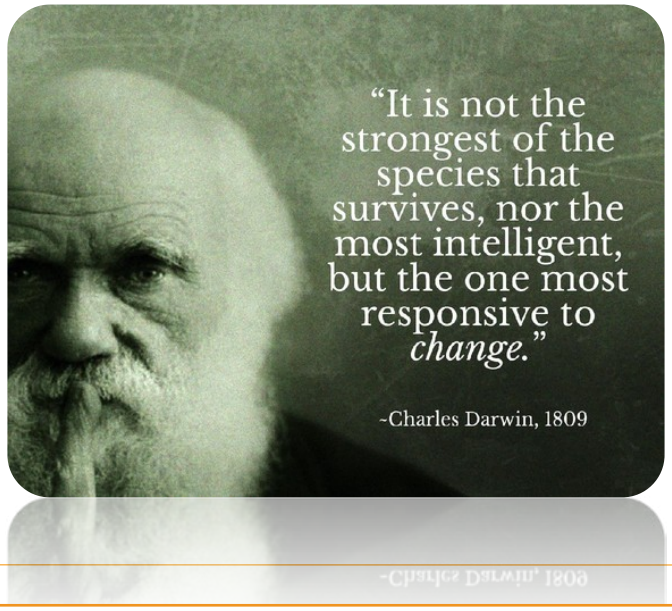
- Capacidades adquiridas transversais a todos os tumores;
- Simplificação da oncobiologia(celular, molecular e bioquímica)
- Tumorigénesis como processo multipasso

A observação da oncobiologia demosntra que o cancro evolui numa biologia semelhante à Darwiniana.

38

38

Background



39 39

Background

Cell, Vol. 100, 57–70, January 7, 2000, Copyright ©2000 by Cell Press

The Hallmarks of Cancer

2000

Review

Douglas Hanahan* and Robert A. Weinberg†
*Department of Biochemistry and Biophysics and Hormone Research Institute
University of California at San Francisco
San Francisco, California 94143
†Whitehead Institute for Biomedical Research and Department of Biology
Massachusetts Institute of Technology
Cambridge, Massachusetts 02142

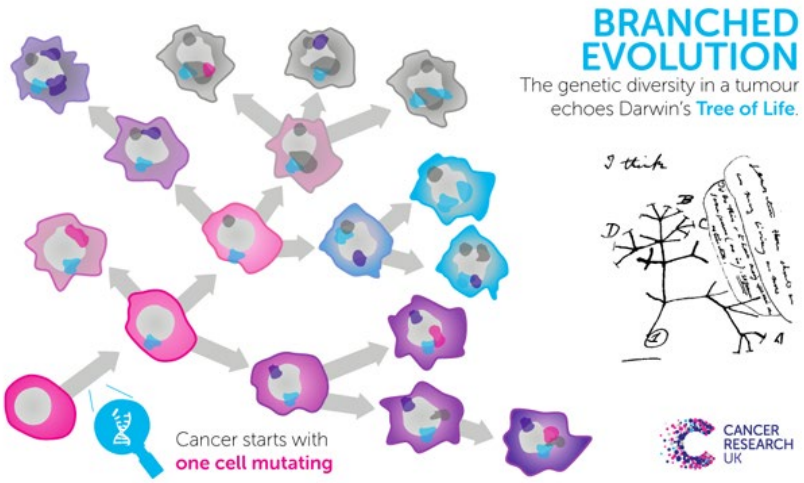
After a quarter century of rapid advances, cancer research has generated a rich and complex body of knowl-

evolve progressively from normalcy via a series of preneoplastic states into invasive cancers (Foulds, 1954).

These observations have been rendered more concrete by a large body of work indicating that the genomes of tumor cells are invariably altered at multiple sites, having suffered disruption through lesions as subtle as point mutations and as obvious as changes in chromosome complement (e.g., Kinzler and Vogelstein, 1996). Transformation of cultured cells is itself a multistep process: rodent cells require at least two introduced genetic changes before they acquire tumorigenic competence, while their human counterparts are more difficult to transform (Hahn et al., 1999). Transgenic

40 40

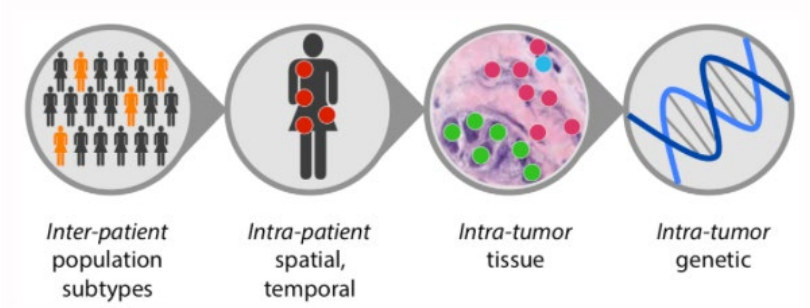
Background



41 41

Background

Heterogeneidade tumoral

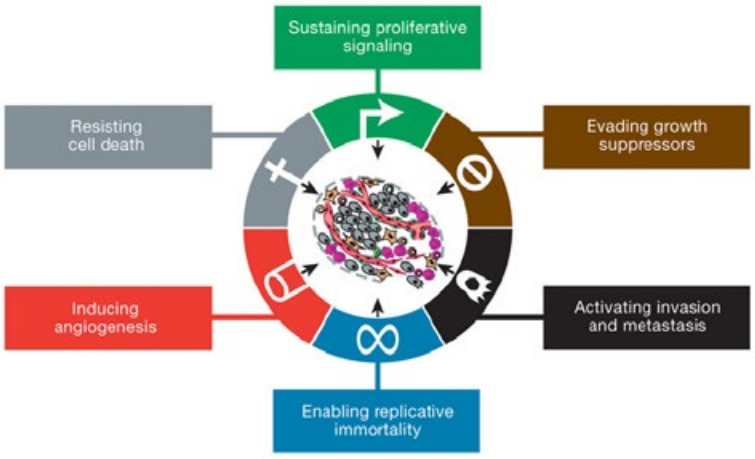


https://scientificbsides.wordpress.com/author/florianmarkowetz/

42 42

Background

Os génotipos das células tumorais são resultado de 6 alterações essenciais na sua fisiologia:



43 43

1-Auto-Suficiência em Factores de Crescimento

https://crowdfavorite.com/blog/2014/11/17/purposeful-growth/

44 44

1-Auto-Suficiência em Factores de Crescimento

- Quakquer célula nbormal necessita de sinais de crescimento mitogénicos para sair do seu estado de quiescência para um estado proliferativo.

Os sinais são recebidos e transmitidos através de receptores membranares

- Podem ser difusíveis;
- Componentes da matriz extracelular
- Moleculas de interação /adesão célula-a célula

As células tumorais produzem a maioria dos seus sinais de crescimento

Estimulo contínuo e independente de crescimento

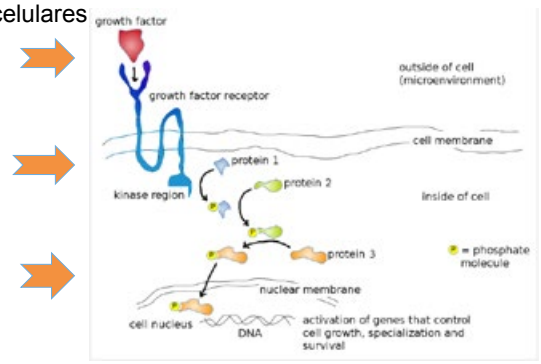
45 45

1-Auto-Suficiência em Factores de Crescimento

- How they do it?

Através de 3 estratégias moleculares:

1. Alteração dos fact cresc. extracelulares;
2. Alteração dos transdutores trasmembranares;
3. Alteração dos circuitos intracelulares



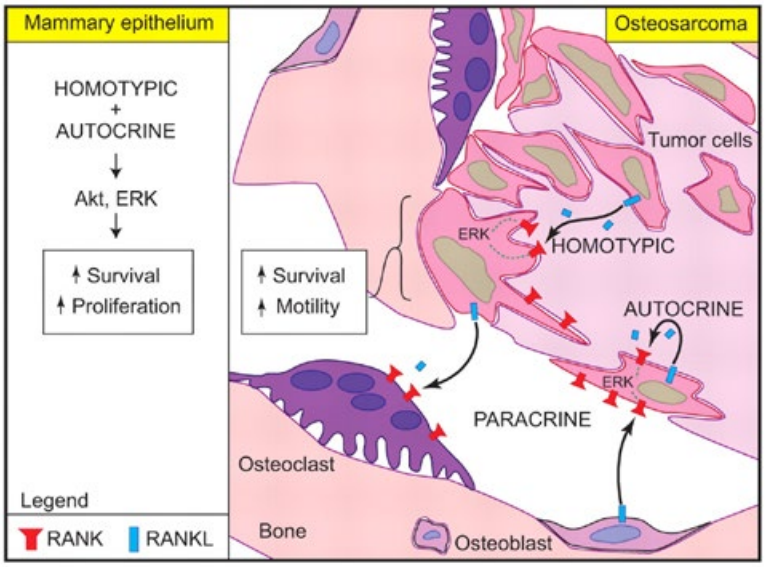
http://www.jargonwall.com/cancer/hallmarks-cancer-1-self-sufficiency-growth-signals/

46 46

1-Auto-Suficiência em Factores de Crescimento

PDGR

TGFα

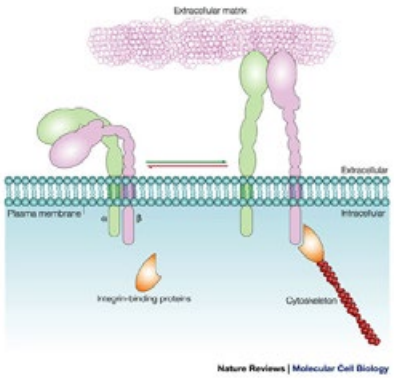


http://jcs.biologists.org/content/125/4/943/F7.large.jpg

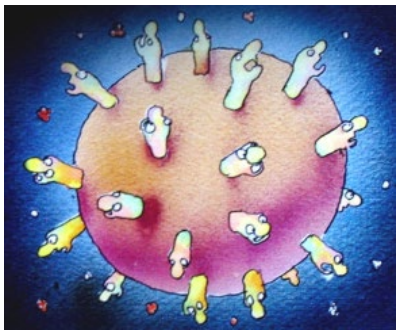
47 47

1-Auto-Suficiência em Factores de Crescimento

Modulação das Integrinas



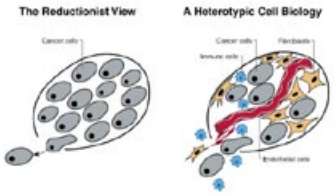
Aumento do número receptores



48 48

1-Auto-Suficiência em Factores de Crescimento

As células tumorais recrutam e subvertem células normais para actuar como colaboradores



Focus nas células e seus genes

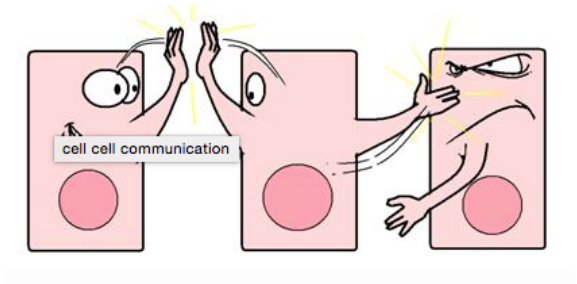
Focus no cancro como tecido complexo

- Os factores de crescimento dos carcinomas vêm do estroma adjacente
- Em alguns tumores estas células deixam o seu estado de dormência ou mesmo o seu nicho para cooperar com o tecido maligno (ex: Neuts/ Macrofagos)

Cell, Vol100,57-70,2000

49 49

2- Insensibilidade aos factores anti-proliferação



http://betterbodychemistry.com/31-days-body-chemistry/

- Os sinais anti-proliferativos são realizados através do uso de inibidores solúveis ou insolúveis presentes na matriz extracelular

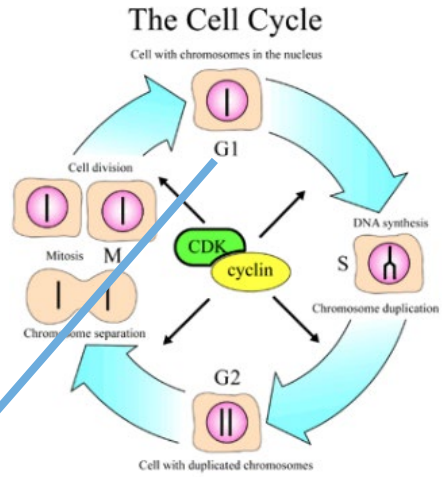
50 50

2- Insensibilidade aos factores anti-proliferação

As células monitorizam constantemente o seu ambiente

STOP
or
GO

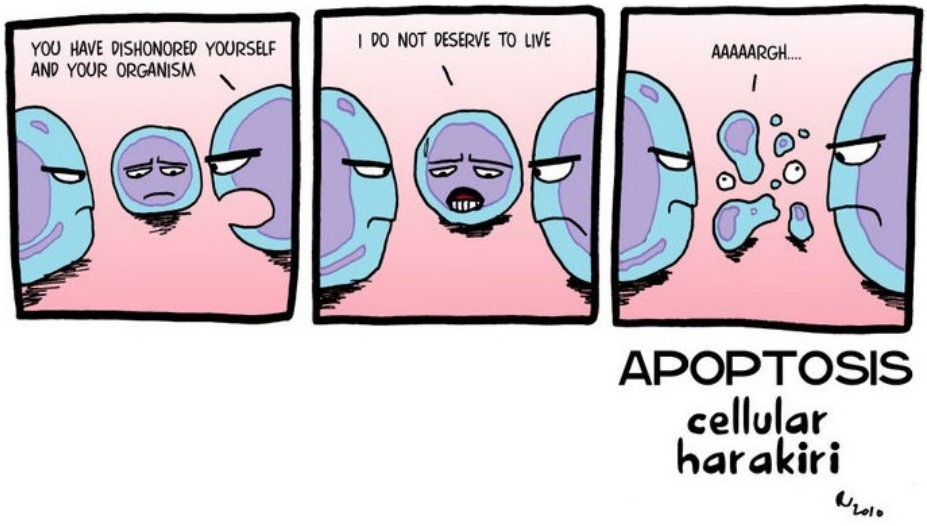
pRb (retinoblastoma protein)
(Especialmente nos carcinomas induzidos por vírus)
TGFβ (Tumor Growth Factor)



www.nobelprize.org

51 51

3-Evasão à apoptose



52 52

3-Evasão à apoptose

Bypass à morte celular programada

Resultado de:

Perda de genes supressores pró-aptópticos

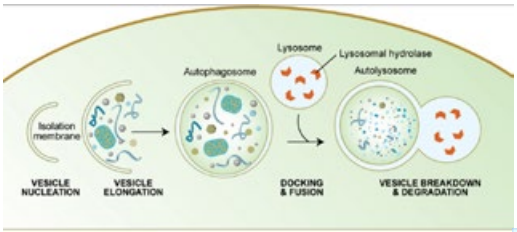
- PTEN
- p53

Ganho de função de genes anti-apoptóticos

BCL

53 53

3-Evasão à apoptose



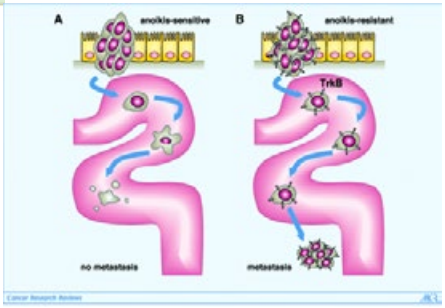
http://www.wormbook.org/chapters/www_autophagy/autophagyfig1leg.jpg

Autofagia

Quando stressadas ou em ambientes com baixo teor nutritivo

Anoikis

Capacidade de sobrevivência após perda de adesão celula-a-célula ou contacto com Matriz Extra Celular

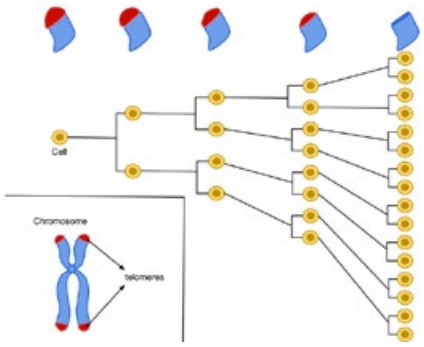


54 54

4- Potencial Replicativo Infinito

O Crescimento tumoral expansivo não se faz unicamente à custa da perda de adesão celular

Todas as células vêm programadas com capacidade limitada intrinseca de replicação



Hayflick (1961)

$2^{40\text{-}60}$

Senescence

Crisis

Immortalization (Wright et al,1989)

Chromossomal fusion
DNA deletion
Karyotipic variation

1 in 10^7

55 55

4- Potencial Replicativo Infinito

Hayflick work (1961)

$2^{40\text{-}60}$

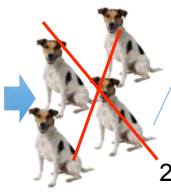
Senescência

Crise

Imortalização (Wright et al,1989)

Fusão cromossómica
Delecção ADN
Variação Cariotípica

1 in 10^7



$2^{40\text{-}60}$

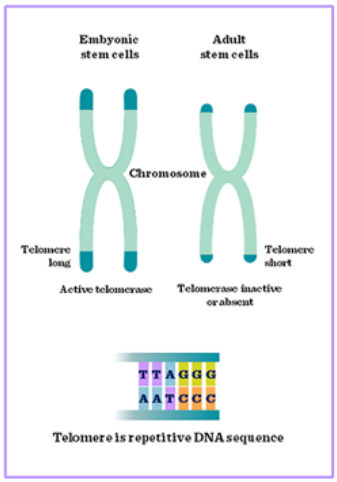
$2^{40\text{-}60}$

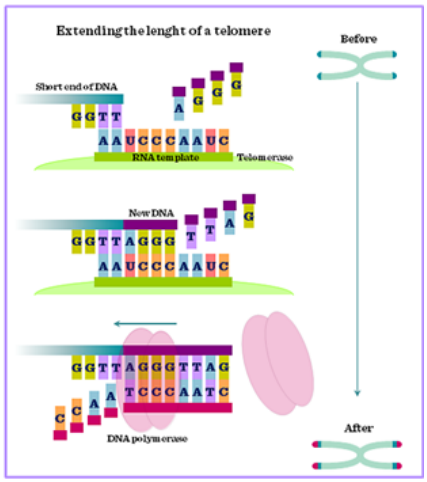
Apoptose

56 56

4- Potencial Replicativo Infinito

A manutenção do comprimento telomérico é essencial para a multiplicação infinita





Lookfordiagnosis.com

57 57

5-Promoção da Angiogénese

A sobrevivência celular é dependente do fornecimento de Oxigénio e Nutrientes

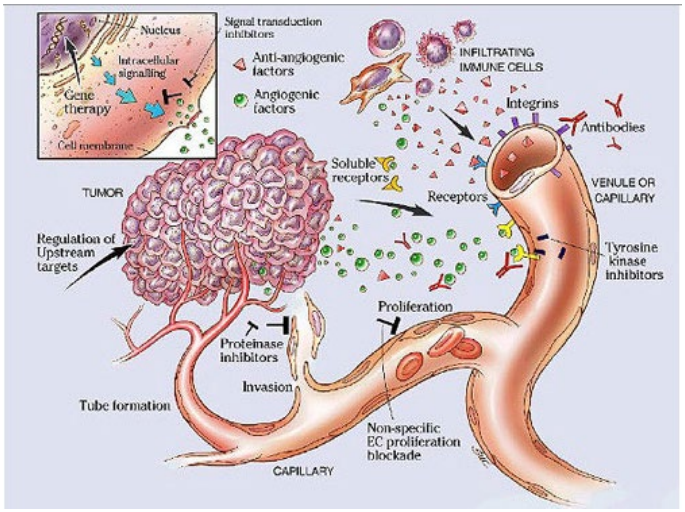
O Crescimento normal dos órgãos é feito com crescimento coordenado dos vasos sanguíneos e parêquima

É de um balanço de sinais positivos e negativos que se promove ou bloqueia a angiogénese

VEGF – promotor
Thrombospondin-1 – inibidor

58 58

5-Promoção da Angiogénese



http://www.mdanderson.org/education-and-research/research-at-md-anderson/basic-science/research-programs/liver-tumor-study-group-research/research2lg-2.jpg

59 59

6- Invasão Tecidular e Metastização

Noventa por cento das mortes são associadas à metastização(Sporn, 1996).

É desconhecido o momento em que um tumor começa começa a espalhar células pioneiras para invadir e formar novas colónias em tecidos adjacentes ou distantes.

O processo de invasão e mestastização depende dos “hallmarks” anteriores

Várias classes de proteínas que estão envolvidas em manter a homeostasia célula-tecido estão alteradas

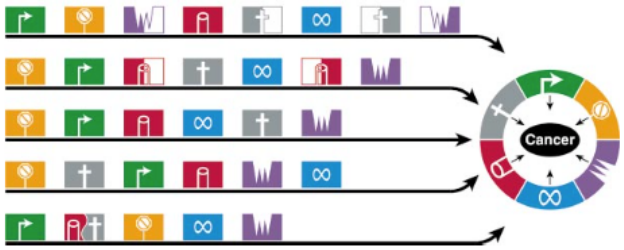
CAM’s
- Imunoglobulinas
- Caderinas (E- Cadherin)

- Integrinas

60 60

Component	Acquired Capability	Example of Mechanism
	Self-sufficiency in growth signals	Activate H-Ras oncogene
	Insensitivity to anti-growth signals	Lose retinoblastoma suppressor
	Evading apoptosis	Produce IGF survival factors
	Limitless replicative potential	Turn on telomerase
	Sustained angiogenesis	Produce VEGF inducer
	Tissue invasion & metastasis	Inactivate E-cadherin

B



61 61

Cell

2011

Leading Edge
Review

Hallmarks of Cancer: The Next Generation

Douglas Hanahan[1,2,*] and Robert A. Weinberg[3,*]
[1]The Swiss Institute for Experimental Cancer Research (ISREC), School of Life Sciences, EPFL, Lausanne CH-1015, Switzerland
[2]The Department of Biochemistry & Biophysics, UCSF, San Francisco, CA 94158, USA
[3]Whitehead Institute for Biomedical Research, Ludwig/MIT Center for Molecular Oncology, and MIT Department of Biology, Cambridge, MA 02142, USA
*Correspondence: dh@epfl.ch (D.H.), weinberg@wi.mit.edu (R.A.W.)
DOI 10.1016/j.cell.2011.02.013

63 63

The Next Generation

Os "Hallmarks" e a metáfora do circuito

Postulam-se subcircuitos
A lista de sinais e coneções ganhou novas designações
Conceito de interconectividade

64 64

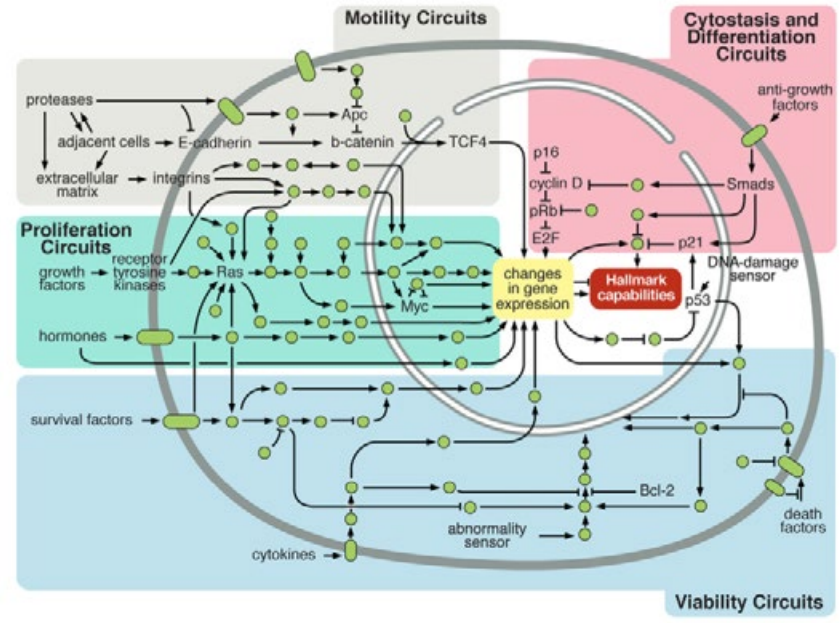


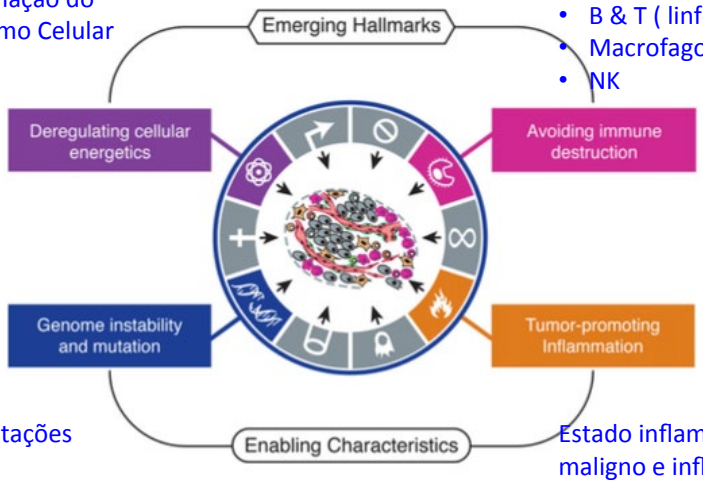
Figure 2. Intracellular Signaling Networks Regulate the Operations of the Cancer Cell
An elaborate integrated circuit operates within normal cells and is reprogrammed to regulate hallmark capabilities within cancer cells. Separate subcircuits, depicted here in differently colored fields, are specialized to orchestrate the various capabilities. At one level, this depiction is simplistic, as there is considerable crosstalk between such subcircuits. In addition, because each cancer cell is exposed to a complex mixture of signals from its microenvironment, each of these subcircuits is connected with signals originating from other cells in the tumor microenvironment, as outlined in Figure 5.

65

The Next Generation

Reprogramação do Metabolismo Celular

Escape :
- B & T (linfócitos)
- Macrofagos
- NK



Geração de mutações aleatórias
Rearranjo cromossómico

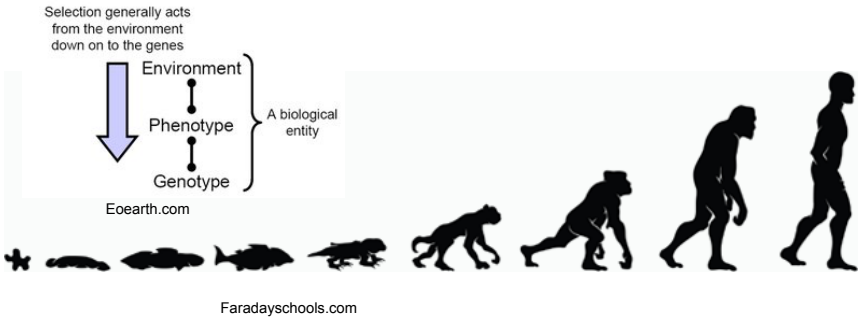
Estado inflamatório pre´-maligno e inflamatório mantido por células recrutadas

66

Caracteristicas Permissoras: Instabilidade genómica

As células cancerosas aumentam o seu número de mutações (Salk et al, 2010)

Estes defeitos na maquinaria que mantém a integridade genómica são vantagens selectivas das células pré-malignas de forma a acumularem genótipos favoráveis



Faradayschools.com

67 67

Caracteristicas Permissoras: Inflamação

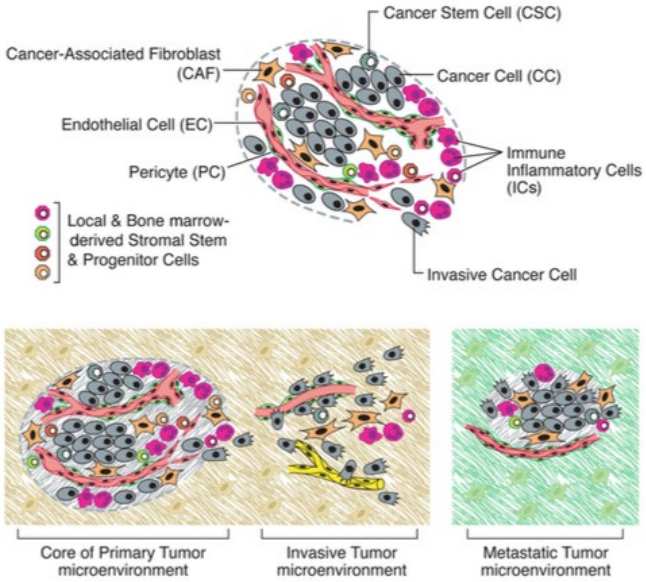
Está mais que reconhecido que os tumores são densamente populados por células do sistema imune inato e adaptativo.

Este fenómeno era interpretado no passado como uma resposta imunitária do hospedeiro para erradicar o cancro.

A Inflamação contibui para a manutenção de vários “hallmarks”:
Fornece factores tróficos, pró-angiogénicos, proteinas extracelulares, etc.

A inflamação leva também à libertação de “Reactive Oxigen Species” (ROS), altamente mutagénicos, que aceleram a evolução genética.

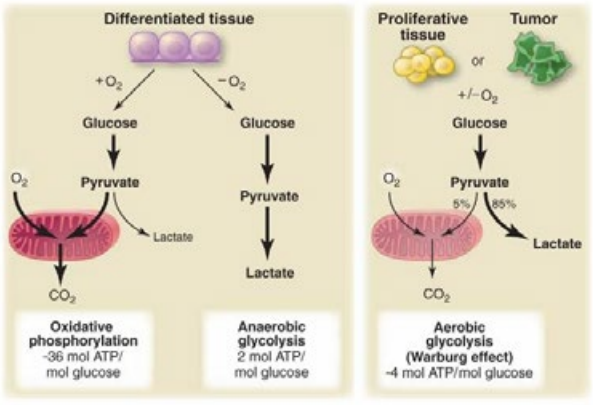
68 68



69 69

Características Permissoras: Reprogramação Metabólica

O crescimento crónico e descontrolado de uma massa tumoral necessita de ajustamentos para suprir as suas necessidades metabólicas.



http://www.esscientificconsulting.com/research.htm

70 70

Características Permissoras: Reprogramação Metabólica

Porque é que isto lhes traz vantagens?

O metabolismo glicolitico está associado à activação de oncogenes (ras, myc) e supressores de mutações(TP53)

Muitos tumores são ambientes hipóxicos

Como compensam a desvantagem em cerca de~18X menos eficácia na produção de ATP?

- Aumentam expressão de receptores de glucose(GLUT1)
- Têm populações heterogéneas:
  - Glicose dependentes que excretam Lactato
  - Lactate excretado é usado pelas outras células como fonte de metabolismo

Semelhante ao que acontece no tecido muscular!

71 71

Características Permissoras: Evasão Imunitária

Porque falha o sistema imunitário no comabte aos tumores?

Existe o conceito de imuno-vigilância validado pelo desenvolvimento de tumores em doentes imunocomprometidos

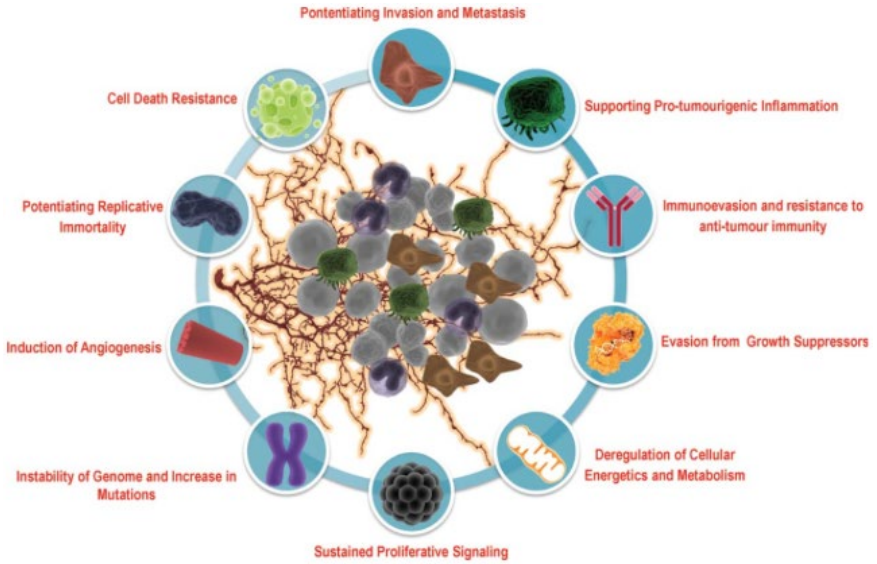
Estes tumores são normalmente de origem viral e podem ser controlados, se controlada a carga viral

Células tumorais imunogénicas são frequentemente erradicadas em hospedeiros imuno competentes, mas podem desabilitar a resposta imune:

Paralizam NK and CTL's segregando TGFβ
Recrutam Tregs and MDSC's

72 72

E o futuro…?!



Abhishek D Garg, Hannelore Maes, Alexander R van Vliet & Patrizia Agostinis (2015) Targeting the hallmarks of cancer with therapy-induced endoplasmic reticulum (ER) stress, Molecular & Cellular Oncology, 2:1, e975089, DOI: 10.4161/23723556.2014.975089

73 73

The NEW ENGLAND JOURNAL of MEDICINE

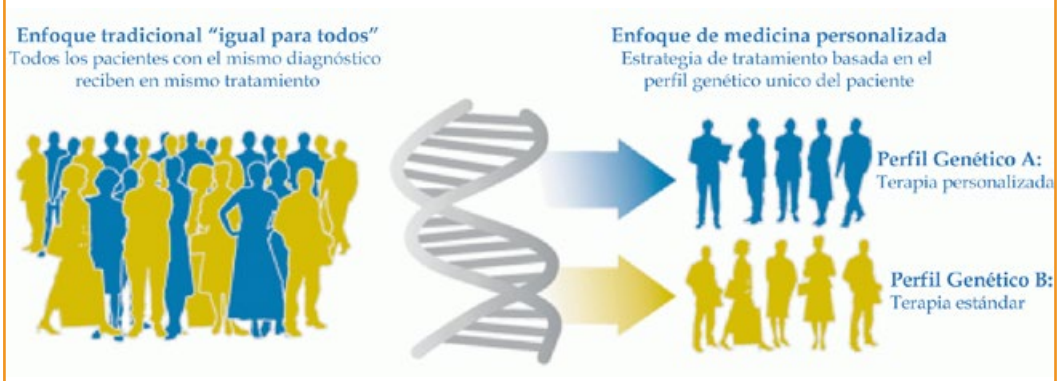
Perspective

JULY 22, 2010

The Path to Personalized Medicine

Margaret A. Hamburg, M.D., and Francis S. Collins, M.D., Ph.D.

74 74

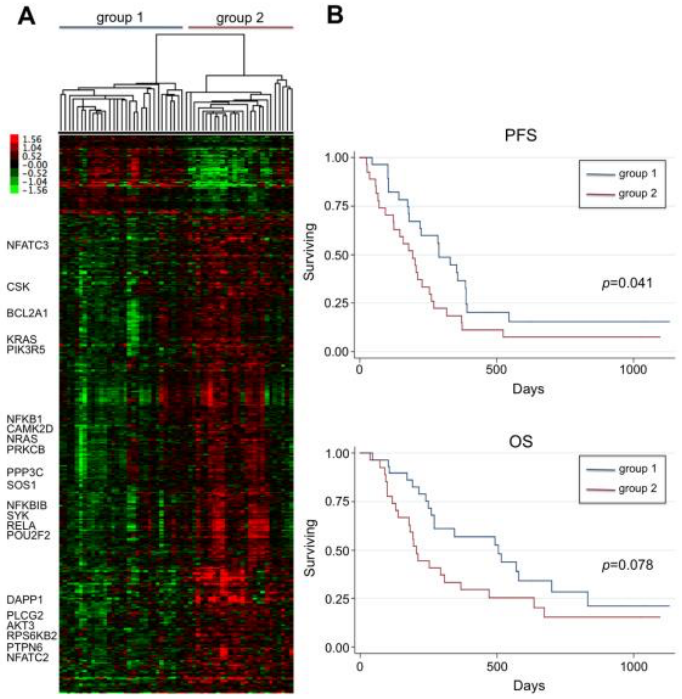


75 75

Gene profiling of canine B-cell lymphoma reveals germinal center and post-germinal center subtypes with different survival times, modeling human DLBCL

Kristy L. Richards,[1,2,3] Alison A. Motsinger-Reif,[2,4,5] Hsiao-wei Chen,[3,6] Yuri Fedoriw,[3,7] Cheng Fan,[3] Dahlia M. Nielsen,[5,8] George W. Small,[1,3] Rachael Thomas,[2,9] Chris Smith,[5] Sandeep S. Dave,[10] Charles M. Perou,[3,7] Matthew Breen,[2,3,9] Luke B. Borst,[2,11] and Steven E. Suter[2,3,12]

76 76



77 77

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Top Companion Anim Med. Author manuscript; available in PMC 2010 August 1.

Published in final edited form as:
Top Companion Anim Med. 2009 August ; 24(3): 113–121. doi:10.1053/j.tcam.2009.03.002.

Update on Genomics in Veterinary Oncology

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Dept. of Molecular Biomedical Sciences, College of veterinary Medicine, North Carolina Sate University, 4700 Hillsborough Street, Raleigh, NC 27606, Center for Comparative Medicine and Translational Research, NCSU, Raleigh, NC 27606 UNC Lineberger Comprehensive Cancer center, Chapel Hill, NC

78 78

OPEN ACCESS Freely available online

PLOS ONE

Prospective Molecular Profiling of Canine Cancers Provides a Clinically Relevant Comparative Model for Evaluating Personalized Medicine (PMed) Trials

Melissa Paoloni[1☯], Craig Webb[2☯], Christina Mazcko[1], David Cherba[2], William Hendricks[3], Susan Lana[4], E. J. Ehrhart[4], Brad Charles[4], Heather Fehling[5], Leena Kumar[5], David Vail[6], Michael Henson[7], Michael Childress[8], Barbara Kitchell[9], Christopher Kingsley[3], Seungchan Kim[3], Mark Neff[2], Barbara Davis[3], Chand Khanna[1*☯], Jeffrey Trent[2,3*☯]

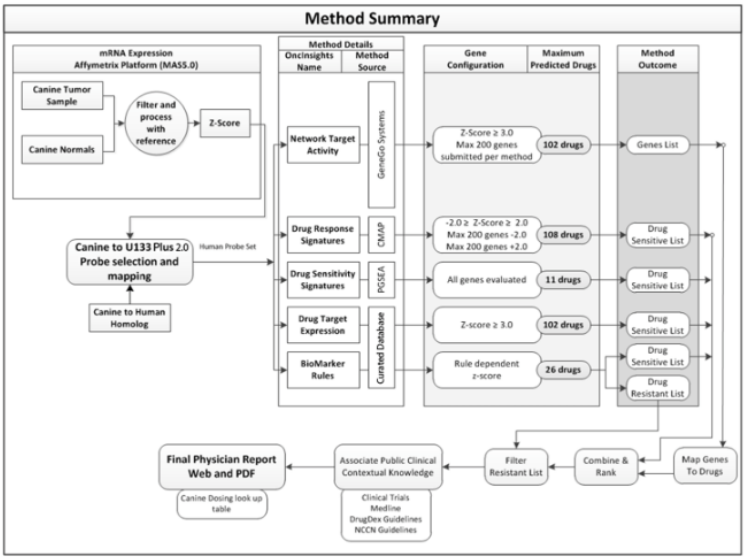

Figure 2. Bioinformatic analysis defines the platform for PMed report generation. Gene expression data from each tumor was compared to a reference sample set (canine normal tissue compendium, GSE20113 from Gene Expression Omnibus) to obtain a relative gene expression profile. Each gene probeset was represented by a z-score depicting its expression in the tumor in terms of the number of standard-deviations from the mean expression in the reference set. In the iteration of the PMed tools used in this study, data were analyzed by six distinct predictive methodologies (Drug Target Expression, Drug Response Signatures, Drug Sensitivity Signatures, Network Target Activity, Biomarker-Based-Rules-Sensitive, Biomarker-Based-Rules-Insensitive) to identify (or exclude in the case of biomarker resistant rules) potential agents for consideration. All predictions were based on the conversion of canine genomic data into human homologs (for both patient tumor samples and the reference set of normal tissues) prior to the application of the specific algorithms that rely exclusively on human knowledge and/or empirical drug screens using human cell lines (see Methods). While individual patient tumor PMed report generation and distribution was the final step in this process, this specific study did not have therapeutic intent and drug prescription was not performed.
doi:10.1371/journal.pone.0090028.g002

Comparative Optimization of Precision Medicine

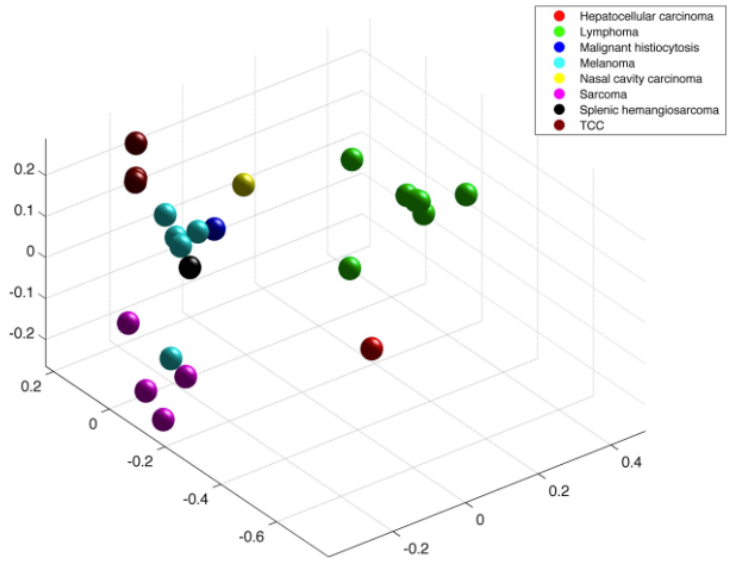

Figure 4. Cancer type defines canine tumor gene expression signatures. Multidimensional scaling (MDS) coordinates were generated using individual tumor gene (mRNA) expression z-scores to define relationships within the dataset. Tumor gene expression clustered by tumor type. Additionally, histologic categories share genomic signatures, with carcinomas (bladder TCC, nasal carcinoma, hepatocellular carcinoma (HCC)), mesenchymal (soft tissue sarcomas, hemangiosarcoma, histiocytic sarcoma, melanoma), and round cell (lymphoma) tumors clustering together in subgroups.
doi:10.1371/journal.pone.0090028.g004

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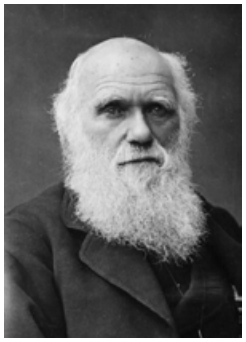
Futuro…?!

O paradigma vai mudar?



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Science & Society

Cell

Cancer research, a field on the verge of a paradigm shift?

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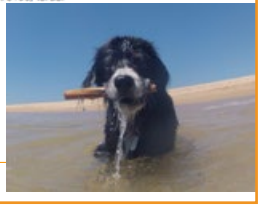
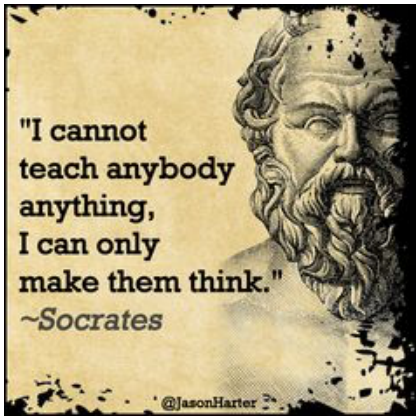


THANK YOU !



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Yolanda Vaz

Nova Lei de Saúde Animal

Direção Geral de Alimentação e Veterinária – Direção de Serviços de Proteção Animal

Resumo

A nova Lei da Saúde Animal

Direção Geral de Alimentação e Veterinária – Direção de Serviços de Proteção Animal

Foi publicado este ano o Regulamento (EU) n.º 429/2016 do Parlamento Europeu e do Conselho de 9 de Março relativo às doenças animais transmissíveis e que altera e revoga determinados atos no domínio da saúde animal, conhecido como **Lei da Saúde Animal**.

Este Regulamento irá substituir os diplomas comunitários relacionados com a saúde animal simplificando-os num só ato legislativo e está integrado na estratégia da saúde animal 2007-2013, "**É melhor prevenir do que remediar**" e implementa ainda os objetivos da Estratégia Europa 2020 nomeadamente a obtenção de "... elevados padrões de saúde animal no intuito de convergência com as normas internacionais e tendo presente as interligações da saúde animal com a saúde pública, bem-estar animal, ambiente, segurança dos alimentos e aspetos económicos, sociais e culturais".

Faz parte de um pacote de regulamentos proposto pela Comissão em Maio de 2013, para reforçar a aplicação das normas de saúde e segurança para toda a cadeia agroalimentar. Destacam-se dentro desse pacote os regulamentos Financeiro e o dos Controlos Oficiais que se interligam com a Lei da Saúde Animal.

Realçam-se os aspetos mais relevantes da Lei da Saúde Animal:

- Introduz regras mais simples, claras e harmonizadas que habilitam as autoridades veterinárias e os destinatários a focarem-se nas principais prioridades: a prevenção e erradicação das doenças dos animais;
- Clarifica as responsabilidades dos operadores (produtores, comerciantes, entre outros), detentores de animais de companhia, médicos veterinários, pessoal dos laboratórios e autoridades veterinárias nacionais em matéria de saúde animal, de biossegurança e da utilização prudente e responsável dos medicamentos veterinários;
- Permite maior flexibilidade de ajustamento das regras às circunstâncias locais e às questões emergentes, nomeadamente por meio das derrogações previstas;
- Melhora a deteção e controlo de doenças animais, incluindo doenças emergentes ligadas às alterações climáticas e de evolução de resistências a antimicrobianos, com o objetivo de reduzir a ocorrência e os efeitos das epidemias de doenças dos animais;
- Permite e estimula uma maior utilização de tecnologias de informação nas atividades de saúde animal como por exemplo o uso do certificado sanitário eletrónico nos movimentos intracomunitários de animais.

As primeiras discussões sobre a Lei da Saúde Animal iniciaram-se em 2007, na presidência Portuguesa da UE, e com a implementação em 2008 do "Plano de Ação para a implementação da Estratégia de Saúde Animal da União Europeia (UE) (COM (2008) 545 final). Entre 2008 e 2013, vários grupos de trabalho estiveram dedicados à preparação da proposta legislativa da Comissão Europeia. Em maio de 2013 a Comissão Europeia enviou a proposta para o Parlamento Europeu e Conselho Europeu para dar início ao processo legislativo intitulado "Co-decisão" com participação das três instituições. A proposta foi discutida entre 2013 e 2016 pelo trílogo constituído pelo Parlamento Europeu, o Conselho Europeu e a Comissão Europeia, contando com o contributo de vários grupos como o de peritos veterinários, o dos Chefes de Serviços Veterinários (CVO) e as Representações Permanentes de Conselheiros e Adidos Agrícolas. Em setembro de 2015, o Conselho alcança acordo político sobre o texto de compromisso da 1ª leitura, tendo a concordância da Comissão em janeiro de 2016. O Parlamento Europeu adotou o regulamento a 8 de Março de 2016. A Lei da Saúde Animal (Regulamento (UE) n.º 429/2016) é publicada no dia 31 de março.

Na sua introdução, a Lei da Saúde Animal apresenta **179 considerandos** que detalham o extenso percurso legislativo e justifica as medidas adotadas e aplicadas às doenças infeciosas de animais terrestres e aquáticos. Realça-se:

- a fundamentação da necessidade de regras relativas a vários aspetos como mercado interno, saúde pública, segurança dos alimentos, bem-estar animal, ambiente, biodiversidade, aspetos económicos, sociais e culturais, para que se alcancem os objetivos estratégicos;
- a manutenção das estratégia "Mais vale prevenir que remediar", "Uma Só Saúde" e "Plano de ação contra a resistência antimicrobiana";
- o respeito pelos acordos internacionais da OIE - Organização Internacional da Saúde Animal e da OMC - Organização Mundial do Comércio;
- a definição de responsabilidades de operadores, detentores e entidades oficiais assim como de médicos veterinários, onde se reforça o seu papel chave na investigação de doenças e na ligação com as autoridades competentes;
- a noção de que a gestão da saúde animal só é conseguida através de estreita cooperação de todos interessados;
- a necessidade de adoção de medidas imediatas para a erradicação de doenças de emergência e de planos a médio e longo prazo para o controlo e erradicação de outras doenças, tendo em consideração determinados critérios de seleção de prioridades.

A Lei da Saúde Animal está dividida em 9 partes, que refletem as matérias versadas, e que se enumeram:

> PARTE I – Regras gerais
> PARTE II – Notificação, vigilância, erradicação e indemnidade
> PARTE III – Sensibilização, preparação e controlo
> PARTE IV – Registo, aprovação, rastreabilidade e circulação
> PARTE V – Entrada na União e exportação
> PARTE VI – Circulação sem caráter comercial de animais de companhia com destino a um Estado-Membro a partir de outro Estado-Membro ou de um País Terceiro ou território
> PARTE VII – Medidas de emergência
> PARTE VIII – Disposições Comuns
> PARTE IX – Disposições transitórias e finais

Em linhas gerais explicam-se os conteúdos de cada uma das partes.

PARTE I – Regras gerais

São descritos, no Capítulo 1 o objeto e as finalidades do Regulamento, seus âmbitos de aplicação e definições. No Capítulo 2 descrevem-se as regras e parâmetros de avaliação das doenças para a sua categorização em classes de doenças sujeitas a regras de:

- emergência (relativamente às quais estão já definidas a febre aftosa, as pestes suínas clássica e Africana, a gripe aviária de alta patogenicidade e a peste equina)
- erradicação obrigatórias
- controlo e erradicação voluntárias
- vigilância e notificação obrigatória
- comércio intracomunitário

Os critérios de categorização das doenças, com vista à sua seleção para a erradicação obrigatória e co-financiamento, incluem o fato de serem transmissíveis, existirem na União animais das espécies suscetíveis ou vetores, terem efeito negativo na saúde animal ou na saúde pública, existirem instrumentos de diagnóstico e medidas de mitigação eficazes e proporcionais. Para além destes critérios é ainda necessário que se adicione um dos seguintes: os efeitos negativos na saúde animal e pública devem ser significativos, ou o agente desenvolveu resistências ou representa perigo significativo, ou tem impacto económico negativo na produção agrícola ou aquícola, ou é susceptível de gerar situação de crise ou é agente usado em bioterrorismo ou ainda tem impacto negativo significativo no ambiente ou biodiversidade. Na legislação de suporte à LSA serão ainda elaboradas listas de espécies animais relevantes para cada doença.

A Parte I finaliza com um Capítulo 3 dedicado às responsabilidades de operadores, profissionais, detentores de animais de companhia, médicos veterinários e outros profissionais trabalhando na saúde dos animais aquáticos e entidades oficiais.

Resumidamente os operadores são responsáveis pela aplicação de boas-práticas de criação dos animais, e pela manutenção da sua saúde, pelo uso prudente e responsável do medicamento veterinário e pela minimização de risco de propagação de doenças. Devem dar particular atenção à biossegurança das explorações, através de manutenção de barreiras físicas e medidas de gestão e à atualização de conhecimento sobre os aspetos acima referidos (boas-práticas, biossegurança) e sobre as doenças, a interação entre a saúde animal, a saúde pública e o ambiente e as resistências a antimicrobianos.

Os médicos veterinários e profissionais que trabalham com animais aquáticos devem adotar todas as medidas necessárias à prevenção de introdução, desenvolvimento e propagação de doenças, através de uma deteção precoce (diagnóstico etiológico e diagnóstico diferencial) e da sensibilização dos operadores para os seus domínios de conhecimento. Têm o dever de cooperação com a Autoridade Veterinária Competente (DGAV) e com os profissionais, operadores e detentores de animais de companhia na aplicação das medidas previstas no Regulamento.
A Autoridade Veterinária Competente deve incentivar, profissionais e operadores a adquirirem e manterem os conhecimentos adequados de saúde animal, através de desenvolvimento de programas de ensino formal. A Lei da Saúde Animal possibilita ainda a delegação de atividades oficiais como as medidas dos programas de erradicação, ações de vigilância, registo, identificação animal, rastreabilidade e informação do público a médicos veterinários não oficiais.

Refere ainda as responsabilidades de laboratórios e instalações que manuseiem agentes de doença.

PARTE II – Notificação, vigilância, erradicação e indemnidade

Esta é a parte mais importante no que se refere às medidas de saúde animal.

O Capítulo 1 diz respeito às obrigações de notificação de suspeitas e investigação de suspeitas de doença e as formas de reporte à Comissão. O Capítulo 2 refere-se às obrigações de vigilância por parte dos operadores, que devem observar a saúde e o comportamento dos animais, identificando alterações dos parâmetros de produção e mortalidade anormal, bem como devem ainda assegurar o acompanhamento dos animais por um médico veterinário. Refere ainda as obrigações das autoridades competentes, métodos, frequência e intensidade da vigilância. O Capítulo 3 tem como tema a submissão de programas de erradicação obrigatórios e opcionais e as respetivas medidas assim como as propostas e relatórios a submeter à Comissão para co-financiamento. Finalmente o Capítulo 4 versa sobre a obtenção, manutenção, suspensão e retirada de estatuto de indemnidade de países, zonas e compartimentos.

PARTE III – Sensibilização, preparação e controlo

A parte III, referente à sensibilização e preparação relativamente a doenças, às medidas de controlo de doenças de emergência e de erradicação obrigatória ou facultativa encontra-se subdividida:

No Título 1 "Sensibilização e preparação" são referidos os planos de contingência e os exercícios de simulação, o uso de medicamentos veterinários para o controlo de doenças infecciosas e os bancos de antigénios, vacinas e reagentes.

No Título 2 "Medidas de controlo", Capítulo 1, descrevem-se as medidas e obrigações para as doenças listadas (de emergência), nomeadamente a declaração e investigação de suspeita, as medidas preliminares, os inquéritos epidemiológicos, a confirmação de surto ou levantamento das medidas preliminares. Seguem-se as medidas de eliminação dos focos e atuação nos estabelecimentos relacionados, e o estabelecimento das zonas de restrição e respetivas medidas. Refere as delegações de poderes, a vacinação de emergência a e atuação em populações de animais selvagens. Uma das seções deste título é ainda dedicada ao papel de coordenação da Comissão nestas ocorrências sanitárias.

No Capítulo 2 do Título 2, determinam-se as ações em caso de erradicação obrigatória, sendo referidas as obrigações, a investigação que a autoridade competente deve realizar, as medidas de controlo de doenças em caso de suspeita e em caso de confirmação de doença de erradicação, quer em animais com detentor, quer em animais selvagens. Refere ainda o papel de coordenação da Comissão e a possibilidade de regras especiais temporárias.

PARTE IV – Registo, aprovação, rastreabilidade e circulação

Esta parte da Lei da Saúde Animal versa tema do registo de explorações e dos locais de comércio, dos transportadores e dos operadores que efetuam operações de agrupamento de forma independente dos estabelecimentos, bem como a identificação animal, e está separada em 3 Títulos, o primeiro relativo a animais terrestres, produtos germinais e produtos de origem animal, o segundo relativo aos animais aquáticos e aos produtos deles originados, e um terceiro relativo a animais de espécies que não sejam abrangidas pela definição de animais terrestres ou aquáticos, e produtos germinais, e produtos de origem animal provenientes desses outros animais.

Cada parte divide-se em capítulos relativos aos seguintes temas: registo, aprovação e conservação de arquivos, requisitos de rastreabilidade, circulação de animais detidos terrestres e aquáticos na UE, circulação de animais terrestres selvagens, circulação de produtos germinais e produção transformação e distribuição de produtos. Finaliza com determinações sobre o âmbito das medidas comunitárias e nacionais. São explicitadas as obrigações de quem mantém animais, os transporta e os recebe no destino, assim como as operações de agrupamento.

PARTE V – Entrada na União e exportação

O Capítulo 1 desta parte apresenta os requisitos para a entrada na União, de animais, produtos germinais e produtos de origem animal provenientes de países terceiros e territórios, a listagem de países terceiros e territórios e aprovação de estabelecimentos, da entrada na União de produtos germinais e de produtos de origem animal, assim como para a certificação veterinária, as declarações e documentos que devem acompanhar os movimentos de animais, produtos germinais e produtos de origem animal.

O Capítulo 2 refere-se à entrada na União de determinadas mercadorias que não sejam animais, produtos germinais e produtos de origem animal a partir de países terceiros e territórios e o Capítulo 3 à exportação.

PARTE VI – Circulação sem caráter comercial de animais de companhia com destino a um Estado-Membro a partir de outro Estado-Membro ou de um País Terceiro ou território

Após um primeiro capítulo com as disposições gerais, os Capítulos 2 e 3 referem-se às condições para movimentos entre Estados-Membros e para movimentos a partir de um país terceiro ou território para um Estado-Membro, respetivamente. Os Capítulos 4 a 6 referem-se à identificação de animais de companhia e às medidas de prevenção e de mitigação dos riscos e às obrigações de informação.

PARTE VII – Medidas de emergência

Na primeira seção são descritas as medidas de emergência relativas à circulação de animais e produtos na União e meios de transporte e outros materiais que possam ter estado em contato com esses animais e produtos são vertidas nesta Parte. A segunda seção aborda os mesmos aspetos mas relativos a animais, produtos e transporte originários de países terceiros e territórios.

Finalmente as duas últimas **Partes VIII e IX** são relativas às disposições comuns e disposições transitórias e finais, onde se listam os atos legislativos revogados as medidas entretanto aplicadas.

Os Anexos da Lei da Saúde Animal referem-se às espécies de animais de companhia à qual se aplica o Regulamento, à lista de doenças consideradas de importância na UE, às espécies de ungulados a que se referem determinados artigos, aos critérios de aplicação de certas regras e à correspondência entre os artigos do Regulamento em relação aos atos existentes.

A Lei da Saúde animal irá ser aplicável a partir de **21 de Abril de 2021** e os atos delegados e de execução (os regulamentos e as decisões) de suporte à Lei da Saúde Animal irão ser adotados pela Comissão Europeia até **Abril de 2019**. Este processo vai implicar uma profunda revisão da legislação nacional onde se contam com mais de 55 peças legislativas nacionais com matérias relacionadas. Será necessário especificar as entidades competentes para a fiscalização, o regime sancionatório e as normas revogatórias e ainda preparar a legislação nacional correspondente a todos os atos delegados e de execução.

José Angélico e Graça Amaral

O Médico Veterinário e Aquicultura

DGRM – Direção Geral de Recursos Naturais, Segurança e Serviços Marítimos

Resumo

A aquicultura é a criação ou cultura de organismos aquáticos aplicando técnicas concebidas para aumentar a produção dos organismos, para além das capacidades naturais do meio. Nas últimas três décadas, foi o sector de produção de alimentos que mais rapidamente cresceu, a nível mundial. Embora a maioria da produção mundial seja originária da Ásia, sobretudo da China, a produção europeia tem algum significado. Entre nós a produção inclui sobretudo peixes e bivalves marinhos, mas Portugal tem ainda uma cota produtiva relativamente pequena no seio UE, muito abaixo do seu potencial. Na verdade, apesar de termos o maior consumo de pescado per capita da UE a aquicultura representa apenas 2% do consumo. A oportunidade de crescimento é imensa. Contudo, as doenças e consequente mortalidade em aquicultura, são um dos constrangimentos mais significativos ao sucesso da atividade. A perda de produção relacionada com doenças, em animais de aquicultura, é de cerca de 25% representando um enorme fardo económico. Para o sector poder crescer de forma sustentada, necessita da abordagem global destes problemas, por médicos veterinários com formação específica e a trabalhar diretamente com a produção. Para tal é indispensável encarar frontalmente a necessidade formação altamente especializada de médicos veterinários, focado nos objectivos da produção em aquicultura.

Palavras-chave: Aquicultura, saúde nos animais de aquicultura, formação complementar de médicos veterinários.

Someia Umarji

Medicina Veterinária Integrativa

Resumo

O conceito de Medicina Veterinária Integrativa resume uma prática da Medicina Baseada em Evidência (MBE) com particular ênfase ao recurso a técnicas terapêuticas complementares.

Como se sabe a MBE é a utilização conscienciosa, explícita e criteriosa da evidência científica actualizada, na tomada de decisões clínicas referente ao doente individual[1]. Só no caso de ausência de estudos publicados sobre um problema (ausência de evidência) é que a experiência clínica isolada poderá servir de base exclusiva de atuação[1].

A prática deste tipo de Medicina tem sido divulgada desde a década de 80 tornando-se nas décadas seguintes mais popular, sendo um conceito suportado pela realização de ensaios clínicos aleatórios e controlados.
O exercício diário de clínica médica baseada na MBE é extremamente compatível com o recurso ao diagnóstico convencional e às medicinas denominadas tradicionais como é o caso da Medicina Tradicional Chinesa (MTC).

As técnicas terapêuticas complementares mais comumente utilizadas vão desde Acupunctura em todas as suas vertentes, Implantes de Ouro, Células Estaminais e Plasma Rico em Plaquetas, Ozono, Hirudoterapia, Fitoterapia, Laser, Mesoterapia, Quiropraxia entre outras.

Introdução

A Medicina Veterinária Integrativa consiste numa prática médica convencional baseada em MBE em que desde o processo de diagnóstico ao tratamento do paciente são utilizadas todas as ferramentas científicas disponíveis para o melhor conhecimento do paciente e sintomas bem como a escolha da(s) terapêutica(s) mais adequada(s).

O uso dos conhecimentos adquiridos pela prática da MTC são compatíveis na totalidade com a Medicina Convencional permitindo um diagnóstico mais amplo e em simultâneo mais preciso de acordo com cada paciente e mesmo o estadio da patologia.

Atualmente são conhecidos variados estudos científicos que suportam as teorias denominadas energéticas baseadas em meridianos e pontos de Acupunctura.

É com base nestes conhecimentos que é formulado um Diagnóstico Integrativo que por sua vez permite na fase terapêutica obter um leque variado de técnicas terapêuticas que operam em conjunto promovendo o tratamento da patologia em simultâneo com a manutenção do bem-estar e qualidade de vida do paciente.

Alguns exemplos da simbiose terapêutica de sucesso em prol dos pacientes é a utilização da Acupunctura no decorrer de tratamentos quimioterápicos em pacientes oncológicos.

O surgimento atual das terapias baseadas no uso de Células Estaminais é um dos exemplos de como o conhecimento científico continua a evoluir e a comunidade científica procura baseado na evidência estabelecer os protocolos terapêuticos em patologias ainda desafiantes da nossa realidade clínica diária.

A prática de uma Medicina Veterinária Integrativa é vantajosa em prol de uma prática clínica ética e em prol da qualidade de vida do paciente.

As Terapêuticas Integrativas

No âmbito da Medicina Integrativa são de uso frequente as técnicas como Acupunctura, a aplicação de Implantes de Ouro, Ozono, Células Estaminais, Hirudoterapia, Laser, Mesoterapia, Quiropraxia e a selecção e recomendação de Fitoterapia

Acupunctura

A acupuntura (do latim acus - agulha e punctura - colocação[2]) é um ramo da medicina tradicional chinesa (MTC) e, de acordo com a nova terminologia da OMS - Organização Mundial da Saúde, um método de tratamento complementar. Foi também declarado Patrimônio Cultural Intangível da Humanidade pela Unesco em 19 de novembro de 2010.

Os efeitos da Acupunctura são descritos em documentos que estudam a neurofisiologia por trás da colocação das agulhas de Acupunctura, sendo de referir que a OMS publicou os resultados num documento "Acupuncture: review and analysis of reports on controlled clinical trials.". Neste documento consta uma lista de 41 doenças e os excelentes resultados com o recurso do tratamento com a Acupunctura.
As outras vertentes do tratamento com a Acupunctura: moxabustão, electroacupunctura, laser-puntura, sangria, massagem Tuina e magnetopunctura são também descritos neste documento em comparação com o tratamento convencional para 147 patologias, sintomas e condições.

Por este motivo é talvez a técnica mais bem aceite pela comunidade médica com maior relutância ao recurso de tratamentos complementares.

No âmbito da Medicina Veterinária Integrativa a Acupunctura, a electroestimulação, a moxa, a fármaco-acupunctura e a laser-puntura são práticas diárias.

A Acupunctura simples pelos seus conhecidos efeitos analgésicos e anti-inflamatório[3] possui indicação para tratamento e maneio de dor aguda e crónica. Esta é a forma ocidental mais usada da técnica.
De acordo com a Teoria dos Meridianos de Acupunctura, baseada na MTC a seleção de pontos pode ser utilizada para obtenção do seu efeito sistémico e energético, tratando em simultâneo a origem de uma patologia e o sintoma manifestado.

A Electroestimulação confere à técnica de Acupunctura Simples a aplicação de impulso elétrico (semelhante à usada na técnica de TENSE - Transcutaneous Electrical Nerve Stimulation). O efeito da aplicação de corrente de baixa intensidade ao nível da fibras Abeta-aferentes induz analgesia[4]. Ao aumentar a intensidade da corrente as fibras Agama e C são estimuladas e o efeito analgésico é mais potente[5].

A Farmaco-acupunctura é uma modalidade que consiste na administração de um fármaco num ponto de acupunctura com o objectivo de potenciar o efeito do fármaco. Um exemplo estudado e de grande utilidade na prática clínica diária é o "Efeito sedativo da dexmedetomidina administrada no acuponto YinTang"[6] em que a mesma dose de fármaco administrada em acuponto possui maior efeito sedativo que a administração via sub-cutânea.
No caso da acepromazina 1/10 da dose administrada no ponto YinTang produz o mesmo efeito sedativo que a dose convencional administrada via intramuscular[7].

A Moxa consiste na aplicação de calor por via de combustão de Artemisia sinensis e Artemisia vulgaris. O princípio é baseado na Acupunctura e portando a aplicação ocorre em pontos ou meridianos sendo que beneficia o tratamento de patologias de dor em que é benéfico o aumento do fluxo sanguíneo e o relaxamento muscular. A sua aplicação é também útil em patologias dermatológicas como úlceras (também o tratamento de úlcera de córnea) e escaras, lambedura psicogénica. A temperatura atingida à superfície da pele e em camadas mais profundas foi estudada, porém o efeito da Moxabustão não está apenas associado à irradiação de calor, mas também à radiação infravermelha[8] e à emissão de fumo que contém substâncias que possuem propriedades antivirais e antifúngicas. São também conhecidos os efeitos da exposição dos pacientes à moxa no sentido de segurança dos mesmos[8].

A utilização do Laser nos pontos de Acupunctura é uma outra modalidade terapêutica em que o efeito obtido pela inserção de agulha de acupunctura é conseguido pela administração de Laser de intensidades estudadas[9]. As aplicações desta modalidade são semelhantes às indicações terapêuticas da Acupunctura Simples, desde maneio da dor, analgesia e obtenção de efeitos sistémicos através da seleção de pontos baseados na Teorias dos Meridianos.

Implantes de Ouro

Os Implantes de Ouro (IO) consistem em fragmentos de Ouro de 24 quilates que são introduzidos em locais específicos selecionados com o objetivo de auxiliar em processos inflamatórios e/ou degenerativos.
Esta técnica começou a ser usada nos animais de estimação nos anos 70 pelo Dr. Terry Durkes para o tratamento de displasia de anca, displasia de cotovelo e espondilose. O sucesso da terapêutica em humanos é também notória sendo denominada Auroterapia ou Terapia Local com Ouro.

Existem variados estudos que comprovam a eficácia do tratamento e comparam o efeito analgésico com os de fármacos convencionalmente utilizados, como é o caso do estudo da Displasia de Cotovelo[10].
Neste estudo em que foram avaliados 1320 pacientes cerca de 80% dos pacientes mostrou melhorias significativas (score elevado).

Outros acompanham o percurso dos pacientes implantados para tratamento de artrose coxofemoral, por um período de 2 anos[11].

O processo de atuação dos IO não era na altura em que começou a ser utilizado conhecido na totalidade. Atualmente graças às tecnologias de microscopia e a possibilidade de acompanhamento de pacientes implantados por longos períodos bem como a realização de exames pós-morte, foi possível conhecer todo o processo químico na origem do efeito anti-inflamatório dos IO.

Na pratica clínica diária esta técnica é uma mais valia seja como prevenção de quadros de artrose em pacientes com grau baixo de Displasia (anca e cotovelo), bem como em pacientes com sinais moderados a severos candidatos ou não a procedimento cirúrgico. O fato de se tratar de um procedimento não invasivo e sem um pós-operatório restritivo aumenta a qualidade de vida do paciente.

Atualmente a ZENVET está a desenvolver um estudo sobre o tratamento do Complexo Estomatite Gengivite Felina e o tratamento com os IO, tendo em conta os casos de sucesso que já tem registado.

Ozono

O Ozono é uma forma de Oxigenoterapia em que um átomo extra de oxigénio é adicionado. Historicamente os relatos datam de 1870 em que há registo de eliminação de microrganismos por contacto com o ozono. Em 1900 Nikola Tesla cria uma fábrica de geradores de Ozono.
Os relatos médicos surgem em 1902 no Dicionário de Práticas Médicas de Londres em que é registado o sucesso de tratamentos com recurso a água ozonizada.

Apesar do sucesso da terapêutica politicamente forma tomadas decisões que conduziram à restrição do uso da técnica durante décadas. Atualmente o processo encontra-se em inversão em muitos países sendo reconhecida pelos Sistemas de Saúde na Alemanha, Suíça, Itália, Espanha, Rússia, Grécia, Austrália entre outros. Nos Estados Unidos é praticada em 15 Estados.

Estudos em Medicina Veterinária que comparam a eficácia da administração intra-rectal do Ozono e a administração em Pontos de Acupuntura versus o uso de meloxican em fêmeas esterilizadas revelam o sucesso desta terapêutica em termos analgésicos[12].

O mecanismo de acção do Ozono é conhecido sendo que os efeitos biológicos produzidos são dependentes de dose, conseguindo-se a ativação de enzimas antioxidantes, efeito antiviral, estimulação de libertação de citocinas específicas[13], interferão e fator de necrose tumoral[14].

É com conhecimento do modo de atuação desta substância que nos tratamentos de maneio de dor crónica, inflamação crónica, patologias autoimunes e oncológicas, o Ozono é usado pela administração via Insuflação Rectal, subcutânea, ou através de técnicas de Auto-hemoterapia menor e/ou maior.

Células Estaminais e Plasma Rico em Plaquetas

As células estaminais são atualmente uma realidade ao alcance do Médico Veterinário pela facilidade de colheita de células e pela existência de laboratórios já especializados na sua produção.

O uso desta terapia recente no âmbito da Medicina Veterinária tem, pelo seu mecanismo de atuação, um futuro promissor. Apesar de ainda não existirem dados suficientes sobre o tratamento de todas as patologias em que se apresentam como candidatas ao tratamento é sabido que os estudos já se encontram a ser realizados.

Atualmente os protocolos existentes são relativos ao tratamento de patologia ortopédica (artrose) e renal.

Hirudoterapia

A terapia é baseada no uso de sanguessugas do tipo Hirudo Medicinalis, tendo sido aprovada para uso médico em Junho de 2004 nos Estados Unidos.
As sanguessugas possuem na saliva que segregam variados compostos entre os quais a Hirudina, um anticoagulante potente e um vasodilatador semelhante a um anti-histamínico. Em conjunto estas substâncias aumentam

o fluxo sanguíneo e aliviam a congestão venosa. Outras substâncias são encontradas na saliva do parasita são Destabilase e Hialuronidase que destroem a fibrina e tecido conectivo, um análogo da morfina, um componente com características bacteriostáticas e uma molécula inibidora da Tryptase o que lhe confere propriedades anti-inflamatórias.

Para além da presença de todos estes componentes as sanguessugas possuem ainda a capacidade de deteção das zonas do organismo hospedeiro em que ocorrem alterações que beneficiam da sua atuação. Por este motivo podem também ser utilizadas como auxílio no diagnóstico de algumas condições.

As indicações terapêuticas são variadas desde a revascularização de tecidos, o tratamento de hematomas como é o caso do sucesso de tratamento de otohematomas, resolução de quadros inflamatórios severos como a laminite nos equinos, hematoma peri-orbital, tromboembolismo cortiço felino, etc.

Recentemente foi reconhecido o seu potencial no tratamento de quadros de osteoartrite.

Quiropraxia

Consiste na manipulação das diversas zonas anatómicas com o objetivo de correção anatómica garantindo o alívio de sintomas como a dor permitindo maximizar a mobilidade e aperfeiçoar a função do sistema nervoso no sentido da troca de informação entre o cérebro e todo o organismo.

A manipulação é feita seguindo as indicações precisas de manipulação garantindo a segurança do paciente.
Esta técnica é frequentemente utilizada em conjunto com a acupunctura permitindo potenciar o seu efeito em particular em quadros de luxação, subluxação, neuralgia do nervo isquiático, espondilose, Síndrome de Cauda Equina e trauma.

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Helena Vala

Interação Médico Veterinário – Enfermeiro Veterinário no exercício da profissão

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Introdução. Perspectiva Histórica

A procura do Curso de Medicina Veterinária pelas classes jovens prende-se essencialmente com o gosto pela área da saúde e o interesse pelos animais. A inexistência de alternativa à Medicina Veterinária, único Curso no nosso país em 2002, para quem gostasse de tratar e cuidar de animais, num âmbito diferente do da produção, fez com que muitos jovens passassem pela frustração de se dedicarem a outras áreas. Com o objetivo de colmatar essa lacuna do panorama Veterinário nacional, proporcionando aos amantes dos animais uma opção alternativa a quem aspirava poder praticar cuidados de saúde veterinários e não tinha possibilidade de aceder à carreira de Medicina Veterinária, fazia sentido preencher este vazio com a criação de uma nova carreira intermédia, de índole técnico, profissionalizante e superior e, enquanto por todo o país se optava pela criação de mais cursos de Medicina Veterinária, os Politécnicos despertaram para este novo nicho de mercado promissor, inspirados pela Europa, que permitiria a formação de quadros bacharéis, integráveis em equipas Médico-veterinárias e às centenas de profissionais que já exerciam essas funções, por todo o país, sem formação adequada, a oportunidade de dignificação da sua profissão.

O Curso de Enfermagem Veterinária foi aprovado em Portugal em 2002/03 para funcionar na Escola Superior Agrária de Elvas e reprovado para funcionar na Escola Superior Agrária de Viseu, no mesmo ano, sendo na altura Ministro da Ciência e Ensino Superior o Doutor Pedro Lynce Faria. Em 2003/04 funcionava o seu primeiro ano na Escola Superior Agrária de Elvas, quando finalmente a Escola Superior Agrária de Viseu também obteve a aprovação para a proposta enviada no ano anterior, colocando o curso em funcionamento um ano mais tarde (2004/05). Posteriormente, viria a entrar em funcionamento nas Escolas Superiores Agrárias de Bragança (Tecnologia Veterinária 2005/06; Enfermagem Veterinária 2010/11), Castelo Branco (2006/07) e Ponte de Lima (2006/07).

Até então cabia exclusivamente ao Médico Veterinário uma grande diversidade de funções, que incluíam o atendimento ao público, a receção dos animais e seus proprietários, a procura e preenchimento de fichas clínicas, a contenção e pesagem, o cálculo e administração de dosagens terapêuticas, a monitorização anestésica, a preparação e disposição de material cirúrgico, a preparação dos animais para cirurgia e a colheita de amostras. Quando não podia realizar todas estas funções, recorria a pessoal não especializado que o próprio tinha que formar e preparar, despendendo tempo e recursos, em detrimento do atendimento e prestação de serviços Médico-Veterinários mais especializados.

A nível Europeu o Curso de Enfermagem Veterinária foi contemplado no tratado de Bolonha e a nível internacional, vários países consideravam, desde há muito, que a Enfermagem Veterinária, a par com os colégios de especialidade, seriam a chave para o desenvolvimento da boa prática dos cuidados veterinários e para a evolução da Medicina Veterinária, permitindo ao Médico Veterinário uma dedicação maior à clínica da especialidade, concentrando-se exclusivamente no diagnóstico, tratamento e na cirurgia em si.

A 11 de Outubro de 2012, a Enfermagem Veterinária no Reino Unido fazia 50 anos, celebrados com pompa e circun-

stância na Casa dos Comuns, enquanto em Portugal, esta jovem profissão, a dar os seus primeiros passos, era vista como uma ameaça por muitos, incluindo Médicos Veterinários, Ordem dos Enfermeiros, entre outros, e nem sequer era ainda reconhecida nos Centros de Emprego.

Contudo, foi o advento desta nova profissão que levou ao surgimento da equipe Médico-veterinária que pressupõe a existência de vários profissionais, com competências distintas mas que devem interagir em prol do objetivo comum da prestação de cuidados especializados de saúde e de bem-estar ao paciente animal.

Em 2014, o curso de Enfermagem Veterinária da Escola Superior Agrária de Viseu obteve a acreditação europeia pela ACOVENE e Portugal junta-se assim à Bélgica, Holanda, Itália, Noruega e Reino Unido, os países onde são atualmente acreditados cursos de Enfermagem Veterinária pela ACOVENE.

A equipe Médico-veterinária

Uma equipe não é um grupo de pessoas que partilham o mesmo ambiente laboral mas, para que interajam em prol de um objetivo comum, a formação e os conhecimentos de cada profissional constituirão um contributo importante, assim como o reconhecimento dos papéis de cada membro da equipe, a existência de uma autoridade bem definida, da capacidade de adaptação, de interacção e de comunicação.

As competências do Enfermeiro Veterinário encontram-se bem definidas pela ACOVENE (2013) e pela AVMA (2012) e estão assentes no papel de apoio ao Médico Veterinário nos procedimentos de rotina cirúrgicos, preparação de material cirúrgico, monitorização anestésica, desinfecção de feridas, aplicação de pensos e bandagens e realização de exames complementares, centrando-se também no paciente animal e no seu bem-estar geral: mantê-lo aquecido, confortável, levá-lo a movimentar-se, mantê-lo bem nutrido e higienizado, gastando mais tempo com um paciente que está desconfortável ou com medo. O seu papel de comunicador privilegiado é também reconhecido, na relação com o cuidador do paciente animal e na prestação de cuidados de enfermagem, graças à formação que o dota de conhecimentos de base suficientes e de competências capazes de o fazer superar, com dignidade e excelência, os desafios dos novos paradigmas da qualidade em prestação de cuidados veterinários.

Conclusão

Equipas funcionais, com boa interação têm potencial para melhorar os resultados da empresa, tornar mais eficaz a prestação de cuidados de saúde de qualidade e ainda poder usufruir de um bom ambiente no local de trabalho, reduzindo os níveis de ansiedade e de stresse de todos os profissionais.

Agradecimentos

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Medicina Veterinária na Avicultura

Jan Willems

Broiler Signals: how to approach a poultry farm visit & how to optimize broiler management

vetworks

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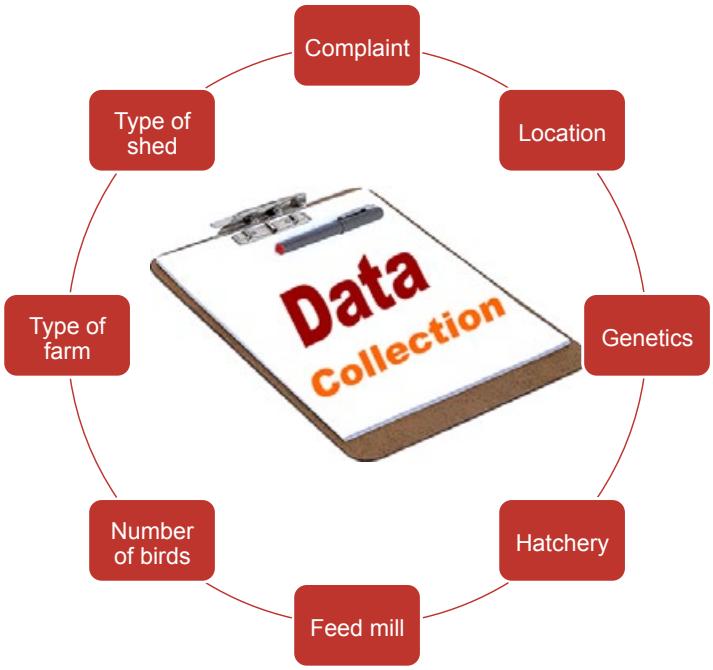
2

How to approach a poultry farm visit

Before going to the farm: ACT



4

Before going to the farm: THINK

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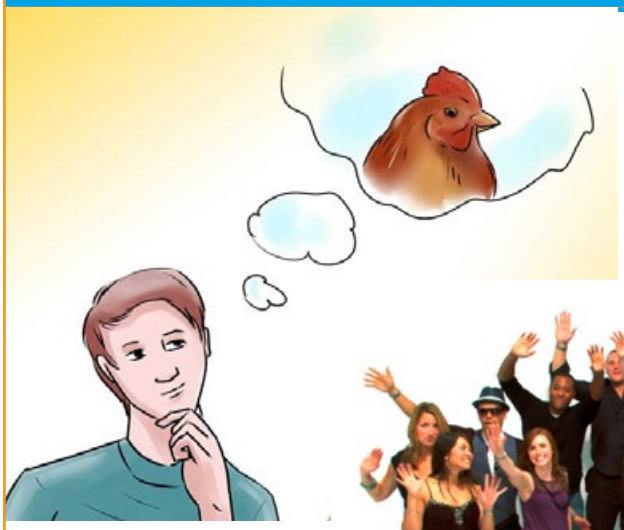

5

Arrival on the farm: LOOK

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ANIMALS – PEOPLE – INFRASTRUCTURE

6

Before you enter the houses: LOOK

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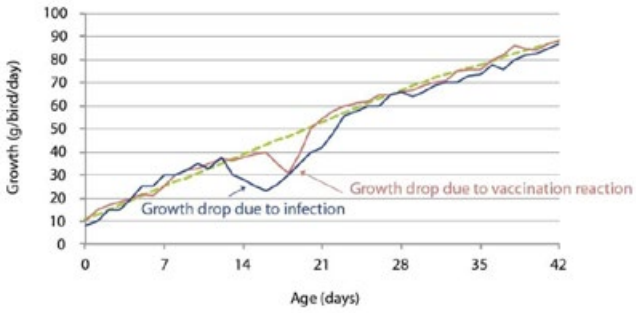
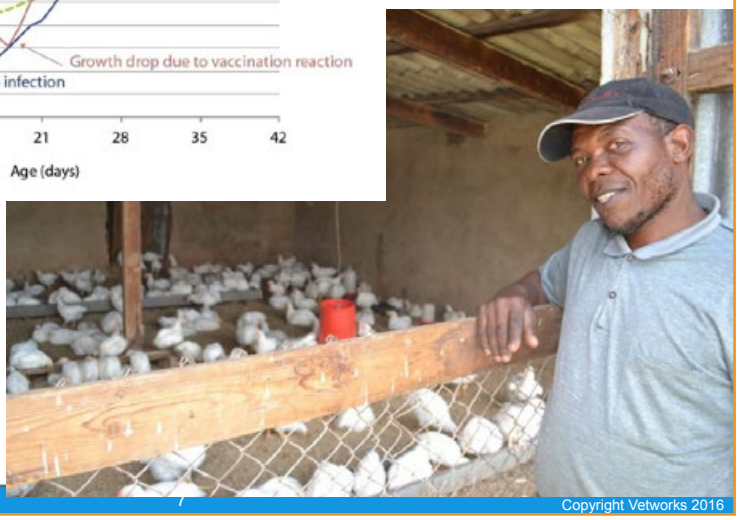
Recognise cause of growth retardation





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Entering the house: LOOK

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8

Entering the house: LOOK

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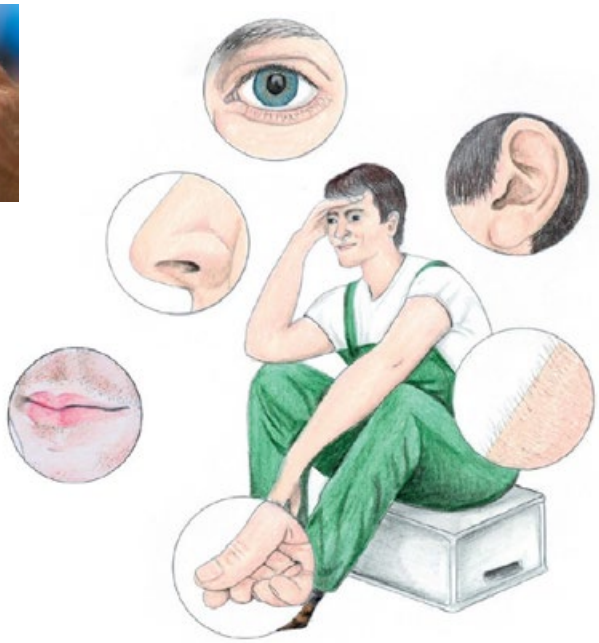

9

Necropsy: LOOK

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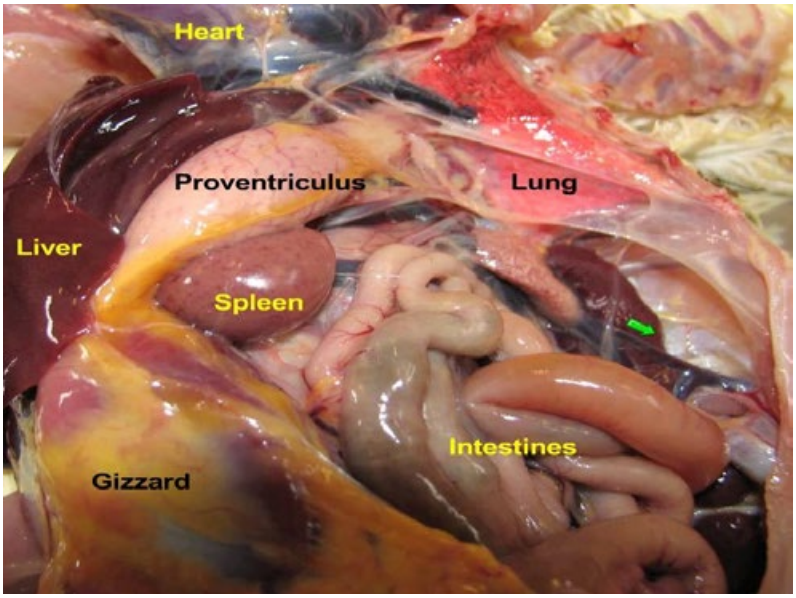

10

Necropsy: ACT

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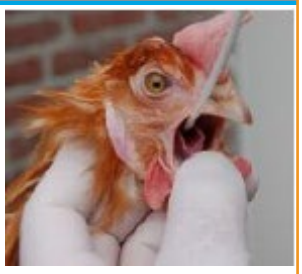
- Samples: don't think, act!
- 1. SWAB
  - Bacteriology (sensitive for contamination!)
  - PCR

- 2. Collect & Cool
  - PCR



- 3. Collect & preserve in formaldehyde
  - histology

- (4. Blood samples)
  - antibodies

11

Report: THINK

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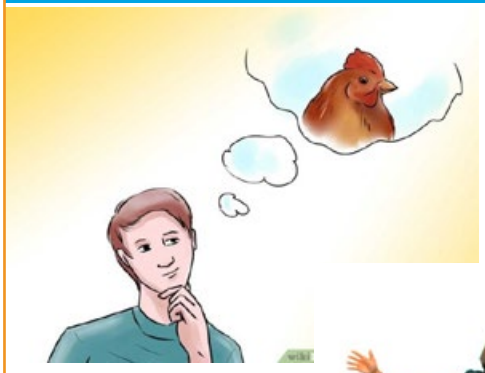

12

Conclusion

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- **LOOK:** knowledge at hand but take your time
- **THINK:** take your time but don't make mistakes
- **ACT:** immediate action (veterinarian) + long term solution (team)

Take your time, then you have the time!

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Questions?

14

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How to optimize broiler management

Overview

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- Getting of to a good start
- Bird signals
- Litter quality
- Infectious diseases
- Climate
- Biosecurity

Introduction

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- Modern poultry system:

Challenges for intestinal health

High density – high feed intake – high performance

17

Preparations

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- d -7: Cleaned and disinfected house + start drying

- d -2: Start heating the house
- d -1: Put down the litter, ideal floor temperature: 28 – 30°C
  - Floor completely covered, at least 0.5 to 1.3 kg / m², at least 3 cm thick, leveled

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Preparations

- d 0: install and check systems
  - Check Feed lines & fill feeding pans
  - Check Drinking lines:
    - pressure, nipples functioning, disinfectants flushes, 80 nipples / 1000 animals
  - Check Ventilation system
    - Fans, thermostats, thermometers, …
  - Put down chick paper close to but not under nipples
  - Put food on the chick paper
    - 40 grams / chick
  - Check temperature
    - 33 – 35°C at CHICK LEVEL!

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Preparations

20

Preparations

vetworks

Feed accessibility and nipple height

21

Day 1

vetworks

- Quality of day-old chicks: Check 10 crates
  - Check number of chicks per crate
  - Weigh chicks: average weight: light chicks (< 40 g) ➔ **+ 1°C**

Check	Right	Wrong
Reflex	Lay chick on its back. It should stand up within 3 seconds	Chick takes more than 3 seconds to stand up: chick is listless
Eyes	Clean, open and shiny	Closed, dull
Navel	Navel should be closed and clean	Bumpy: remnants of yolk; open navel; feathers smeared with albumen
Feet	Feet should be a normal colour and not swollen. Feel warmer than your cheek.	Red hocks, swollen hocks, malformations, deformed toes, cold
Beak	Beak clean with closed nostrils	Red spots on beak; dirty nostrils; malformations
Yolk sac	Stomach soft and malleable	Stomach hard and skin taut
Down	Should be dry and shiny	Down wet and tacky
Uniformity	All chicks the same size	More than 20% of chicks are 20% heavier or lighter than average
Vent temperature	Should be 40°C 2-3 hours after arrival	Above 42°C: too high, below 38°C: too low

  - Chicks should start to eat and drink very fast

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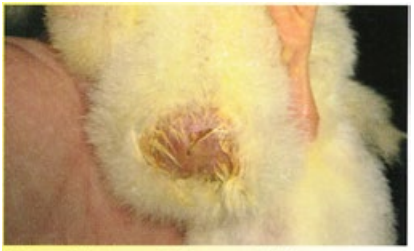
Day 1

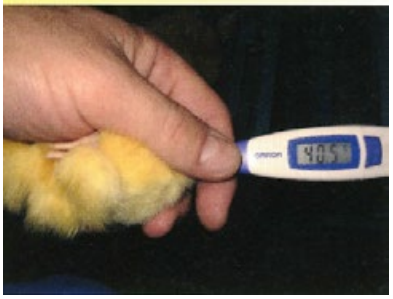
vetworks

- Quality of day-old chicks



Not good but acceptable: navel will close.

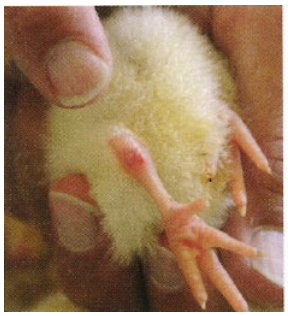
Unacceptable: cannot close because there is some yolk is in the way.



Day 1

vetworks

- Quality of day-old chicks



- Collect dead chicks and search for cause of death

Day 1

vetworks

No gut flora from mother hen

The young chick has not yet developed its own gut flora and picks up all kinds of bacteria, both bad and good. In nature the chick gets the flora from its mother and her environment and is therefore more resistant. The lack of a mother hen on the broiler farm is a risk that is often underestimated.

25

Day 1

vetworks

- Check crop filling
  - 8 hours on the farm: 80% with filled crop
  - 24 hours on the farm: 95% with filled crop
- Light: 23 hours
  - Slowing reduce to 16 – 6 by d7

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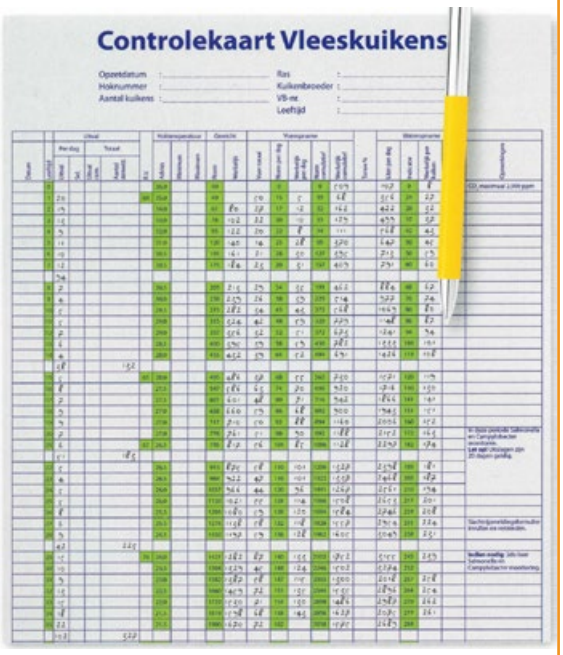
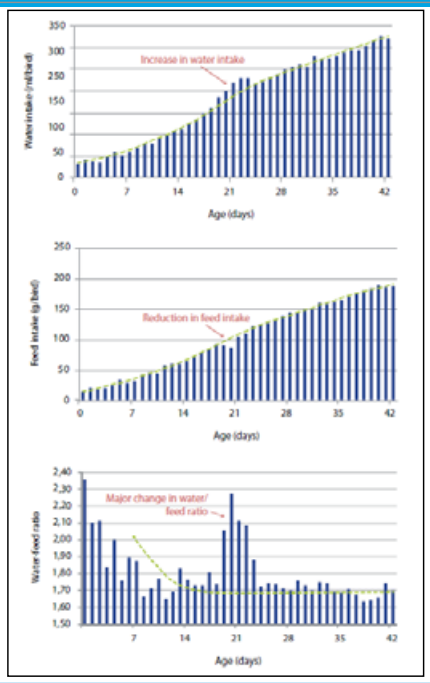
Monitoring

- Registration
  - Daily weight gain
  - Feed intake
  - Water consumption
  - Measurements



Monitoring: Water / feed ratio



Vaccination reaction?

Bird Signals

vetworks

- Week 3 is a critical period
  - Feed intake increases
  - Change of feed
  - Risk coccidiosis
- Development of intestinal problems: signs
  - Droppings
  - Litter
  - Feed/water intake
  - Decreased uniformity

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Droppings

vetworks

Scores for intestinal droppings

Right *Reasonable* *Not right* Source: A. Slaats

Scores for caecal droppings

Right *Reasonable* *Not right*

30

Orange mucus

Orange mucus in the droppings is a sign that the lining of the intestine is damaged. Find out the cause and make sure you provide easily digestible feed.

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Litter quality

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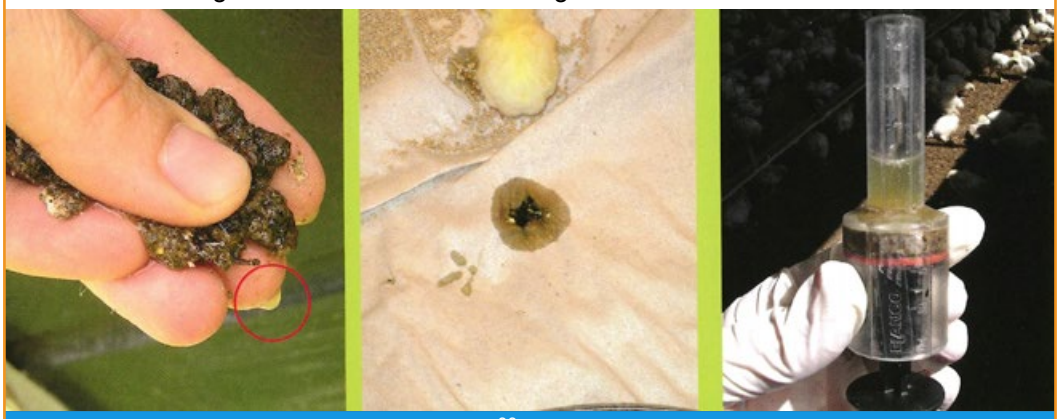
Climate

vetworks

- Litter quality

Humid Faeces

- Gut health problems
- Kidney problems (Gumboro & IB)
- Too high mineral / salt in the drinking water or feed



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Climate

vetworks

- Air Composition: Gasses

	Desirable	Toxic	
Oxygen	> 20 %	< 16 %	used by bird (+ heating)
CO_2	< 2500 ppm	> 12000 ppm	produced by bird (+ heating)
CO	0	>35 ppm	improper combustion
NH_3	< 25 ppm	> 10 ppm	bact. breakdown manure
H_2S	< 40 ppm	> 500 ppm	bact. breakdown manure
Methane	< 1%	> 5%	bact. breakdown manure

- A decrease in one gas implies an increase in at least one other gas and vice versa.
- Exposure time → minimal ventilation

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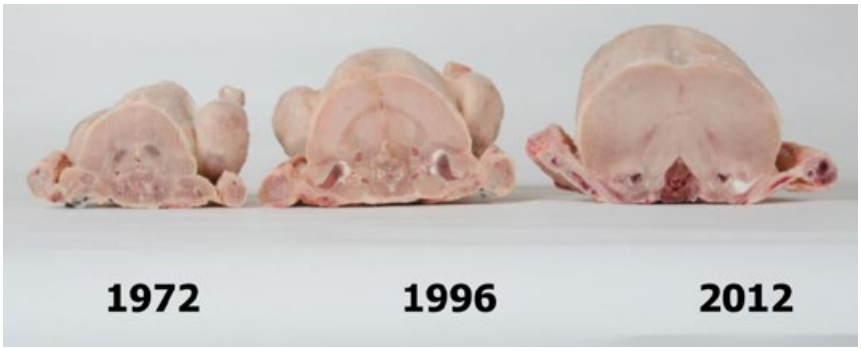
Climate

vetworks

- Air Composition: Gasses
  - **O_2 – Oxygen**
    - Birds have high O_2 demand because of high metabolism
      → reason for adapted respiratory system: O_2 rich air in lungs insp. & exp.
    - Broilers extra high metabolism due to fast growth



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Climate

vetworks

- Ascites
  - Mainly due to management
  - Problems occur at 4 – 5 weeks of age but originates in week 1!
  - Shortage of oxygen
    → heart tries to compensate by pumping more blood around
    → heart overload → heart failure → venous congestion → ascites

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Climate

vetworks

- Ascites
  - Main causes for (relative) oxygen shortage and consequently ascites:
    - **Climate**
      - Lack of oxygen in the air (f.e. high altitudes)
      - Ammonia, CO^2 and dust: reduces respiratory efficiency
      - High temperature x humidity
      - Cold (higher metabolism for heat production)
    - **Genetics**
      - Very fast growth
      - Males
    - **Feed**
      - High protein & high energy
      - High levels of Sodium
      - Pelleted feed x mash feed
    - Lung inflammation / consolidation (aspergillosis, IB…live vac.)
    - Low phosphorus rickets (inadequate lung function)
    - Poor levels of oxygen during incubation

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Climate

vetworks

- Air Composition: Gasses
  - **CO^2 – Carbon dioxide**
    - < 2500 ppm desirable
    - Sources: birds, heaters
    - Symptoms: sleepy, heavy breathing, incoördination
    - Headache
    - Typically young birds in cold climates (minimal ventilation + heaters)
  - **CO – Carbon monoxide**
    - 0 !
    - Heating inside the houses
    - Improper combustion
    - Autopsy: beak & face cyanotic, bright pink lungs

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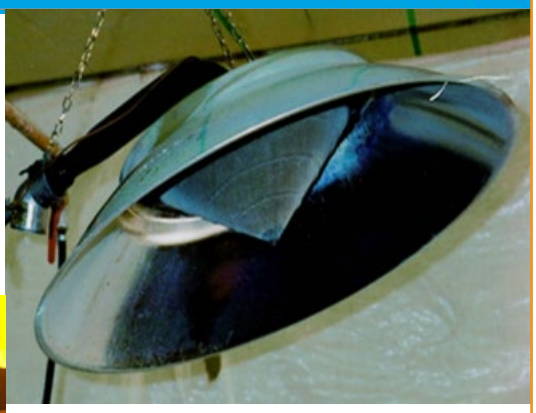
Climate

vetworks



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Climate

vetworks

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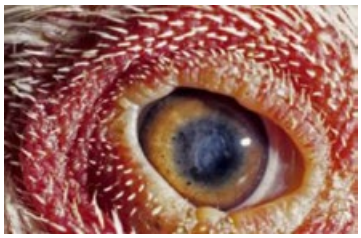
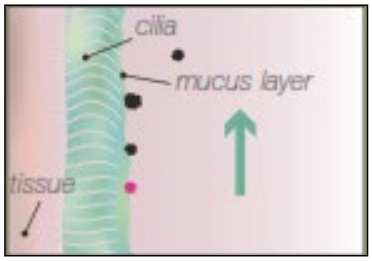
Climate

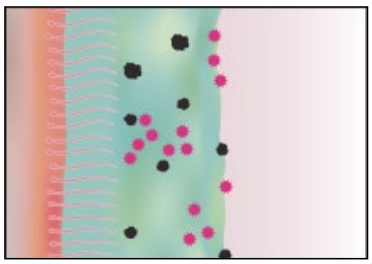
vetworks

- Air Composition: Gasses
  - **NH_3 – Ammonia**
    - Source: Wet (& warm) litter
    - < 10 ppm desirable
    - > 10 : damage to mucosa & cilia
      ➔ more sensitive to infections
    - 50 - 75 ppm : reduced feed intake, growth & egg production
    - >100 ppm : damage to the eye (cornea)





Normal



Thick mucus, cilia not working

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Climate

vetworks

- Air Composition: Dust

- < 3,4 mg / m^3
- Mucosal damage
- RH ↑ = NH_3 absorption ↑
- Max concentration from day 21
  - 1000 E. coli CFU/m3
- Feed particles
- Litter & faeces
- Feathers & Skin
- Bacteria, Viruses, Fungi
  - endotoxins

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Climate

vetworks

- Air Composition: Dust

Concentration depends on:

- Animal density
- Animal activity
- Litter moisture
- Humidity
- Feed composition

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Climate

vetworks

- Litter quality

Humid Faeces

- Gut health problems
- Kidney problems (Gumboro & IB)
- Too high mineral / salt in the drinking water or feed

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Litter quality

vetworks

This is what you see	Caused by	Actual cause
		
An indirect signal is encrusted or compacted manure. This indicates...	*...a poor distribution of birds in the house, which may be caused by...*	*... a cold downdraught, which makes birds avoid cold places in the house.*

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Climate

vetworks

- Litter quality
  - Choice: Absorption ⟷ Drying capacity

fan or water pump?

Caking

- Water leaks
- Condensation
- Relative humidity too high
  - Too little air exchange
  - Unsufficient heating
- Humid faeces
- Differences in light intensity
- Overstocking

Relative humidity dries the litter, not the temperature!

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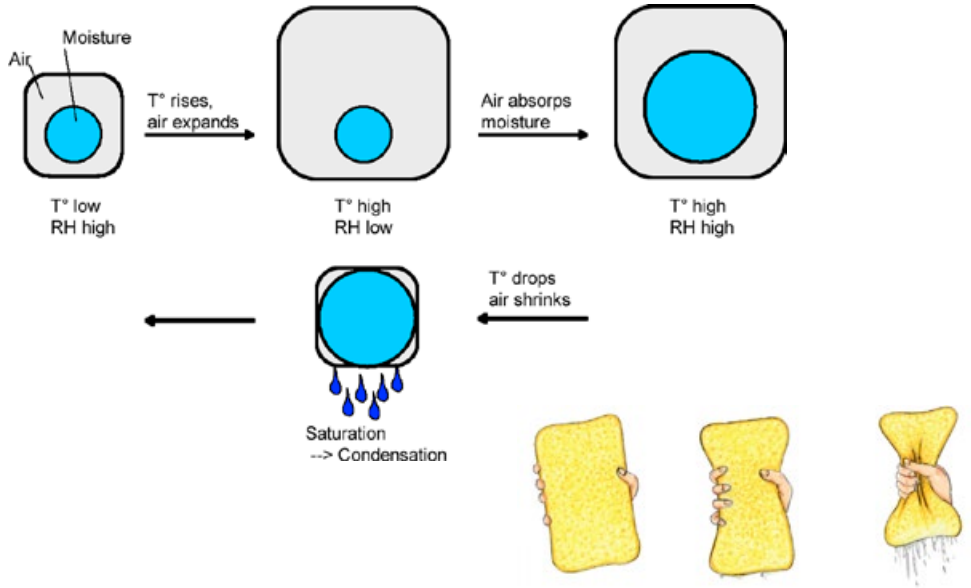
Climate

vetworks

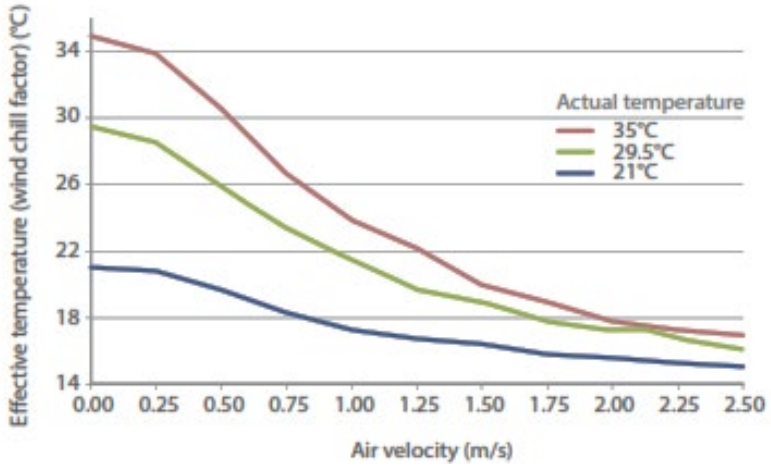
- Relative humidity



Climate

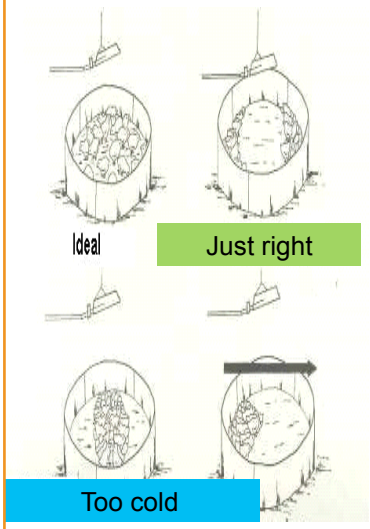
vetworks

- Effective temperature & wind chill factor
  - Air speed (wind chill factor)
  - Relative humidity
  - Stocking density
  - Feather cover
  - Radiant heat



Climate

vetworks



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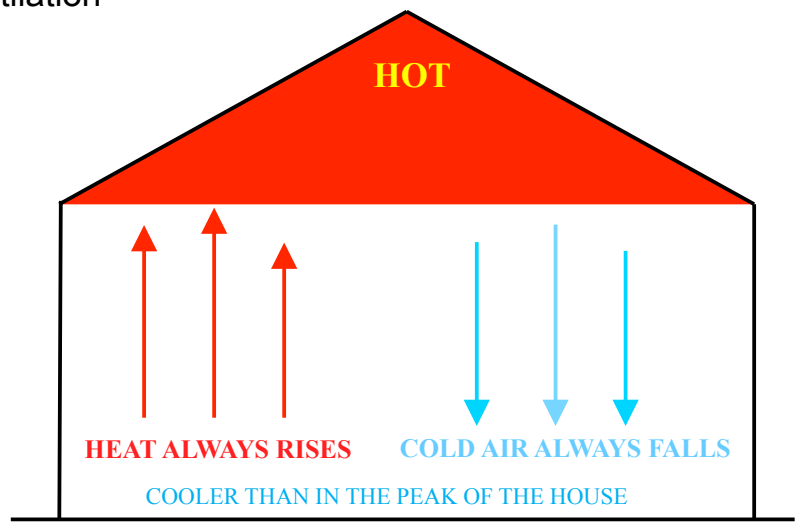
Climate

vetworks

- Ventilation



Source: Broiler manager guide Cobb, 2010

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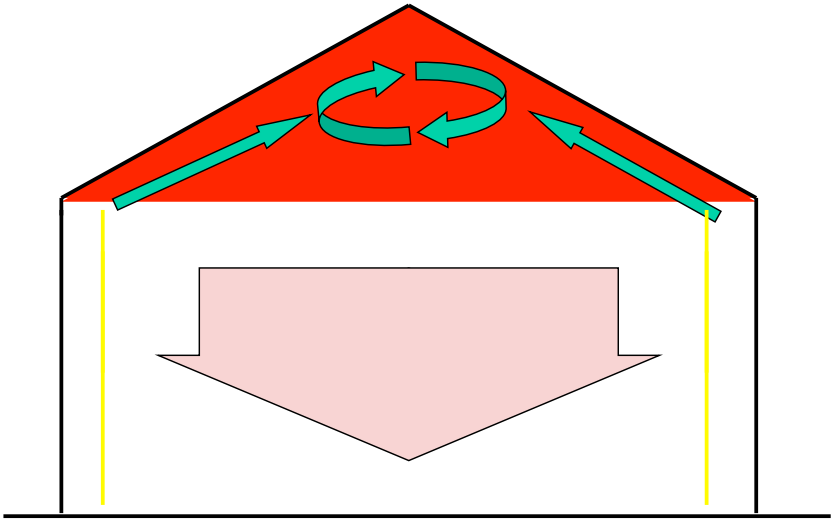
Climate



Source: Broiler manager guide Cobb, 2010

Climate

- Litter quality
  - Choice: Absorption ⟷ Drying capacity

fan or water pump?

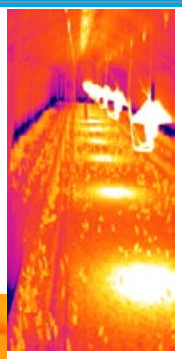
Caking

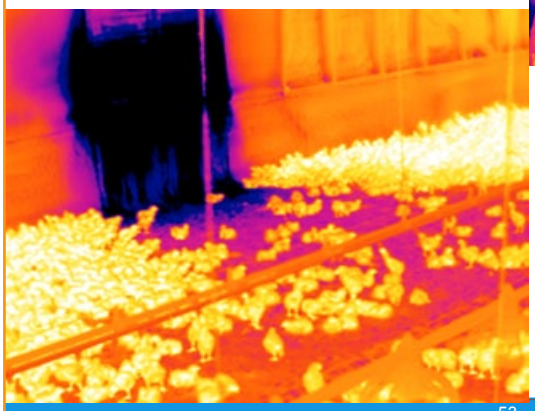
- Water leaks
- Condensation
- Relative humidity too high
  - Too little air exchange
  - Unsufficient heating
- Humid faeces
- Differences in light intensity
- Overstocking

Relative humidity dries the litter, not the heat!

Climate

vetworks





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Climate

vetworks

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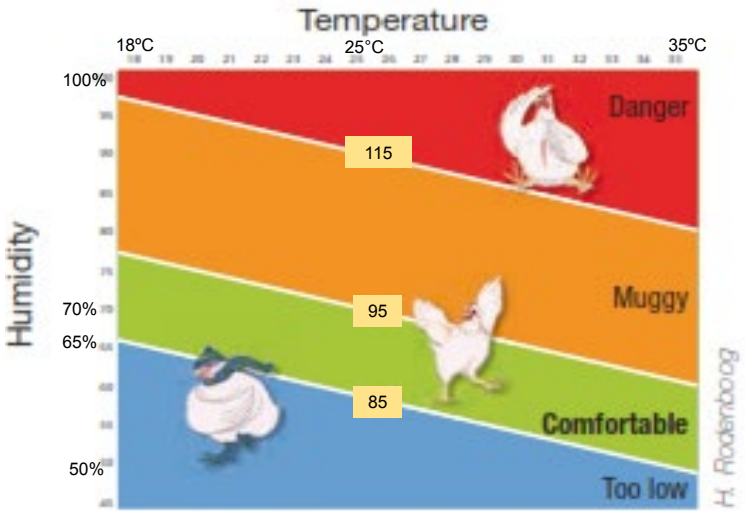
Climate

vetworks

- Temperature / Humidity index



Temperature + Relative humidity = 90 + age of chickens in weeks

Climate

vetworks

- Temperature / Humidity index
  - Example 1:
    - 3 weeks old chicks → optimal = 90 + 3 = 93
    - RH = 73%
    - T°= 24°C

    → 24 + 73 = 97
    - Conclusion: The air is too moist → more ventilation is needed
  - Example 2: 1st week
    - Optimum: 90 + 1 = 91
    - Temperature: 35°C
    - → RH: 91 – 35 = 56 %
    - → Range: 51 – 61 %
  - Example 3: 28 days
    - Optimum: 90 + 4 = 94
    - Temperature: 24°C
    - → RH: 94 – 24 = 70 %
    - → Range: 65 – 70% (= max.)

Temperature + Relative humidity = 90 + age of chickens in weeks

Climate

vetworks

- T/H-index & Heat Stress

	Conductive Cooling	Evaporative Cooling
Temperature ↑	-	+
Relative Humidity ↑	+	-

T/H-index too high: failure to compensate

- > 5 points too high → Heat stress
- > 115 points → Death

Open beak & wings wide open

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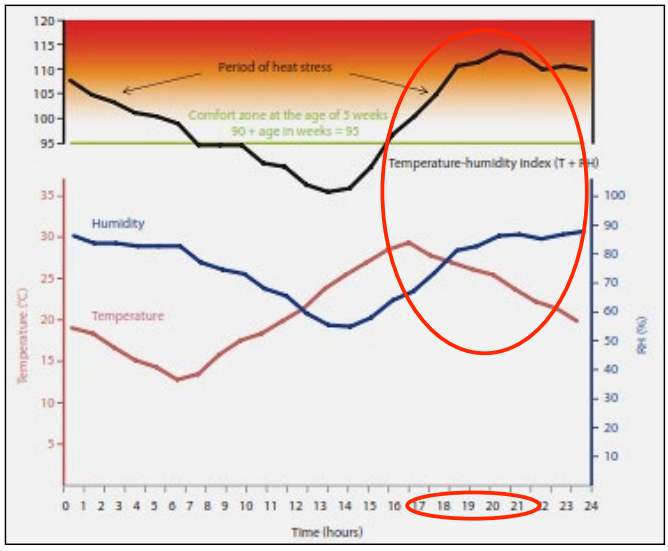
Climate

vetworks

- T/H-index & Heat Stress

- T° ↓ then RH ↑↑

➔T/H-index rises!



- The danger zone is not when the temperature is at its highest but later when the temperature starts dropping!

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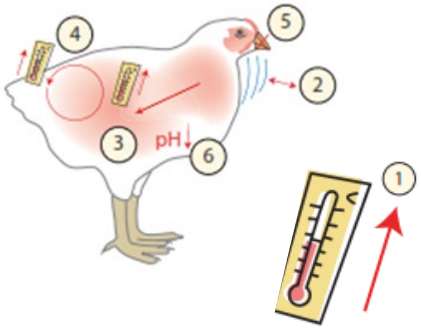
Climate

vetworks

- T/H-index & Heat Stress

Heat Stress:

- Ambient temperature too high 1
  - → Conductive cooling becomes less efficient
  - → **Hyperventilation** 2 (up to 240 breaths / minute)
  - → Muscle contractions heat up the body even more 3
  - → Metabolism rises (+25 % for 1°)
  - → Body temperature rises even further
  - → loss of appetite 5 → **weight loss**
  - → CO^2 in blood ↓
  - → body acidifies 6
  - → retention of bases
  - → CO^2 ↑ Ca↓in blood
  - → **decreased egg quality**
  - → eventually **death**



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Climate

vetworks

- T/H-index & Heat Stress

How to prevent heat stress?

- Better air circulation
- Easily accessible drinking water
- Cooling
- Adjust feed schedule
- Adjust feed composition
- Extra vitamin C
- Extra electrolytes (Kalium)
- Lower stocking density

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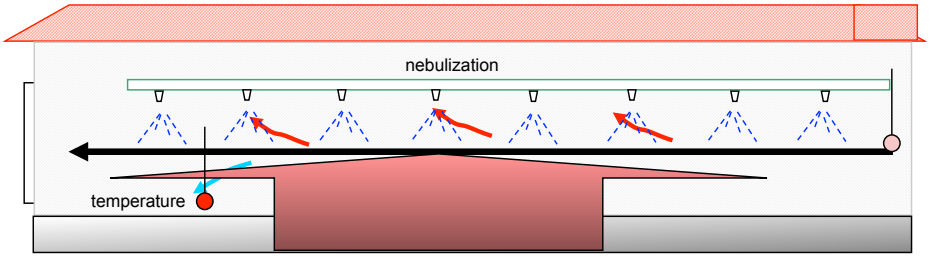
Climate

vetworks

- T/H-index & Heat Stress
  - **Nebulization / fogging system**
    - Reduction 6ºC in 5 min.
    - Humidity rises : turn off the system when 60 to 70% !!!



Source: Broiler manager guide Cobb, 2010

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Climate

vetworks

- Cooling pad

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Climate

vetworks

- Ventilation

Minimum ventilation ALWAYS needed

- **Air exchange** : air quality
  - Heaters: waste gases, CO_2 , H_2O
  - Chickens: H_2O, CO_2
- **Air velocity**
  - Mixing hot and cold air
  - T/H-index controllable
  - Cold air directly to the floor = condensation = wet litter

The birds are always right!

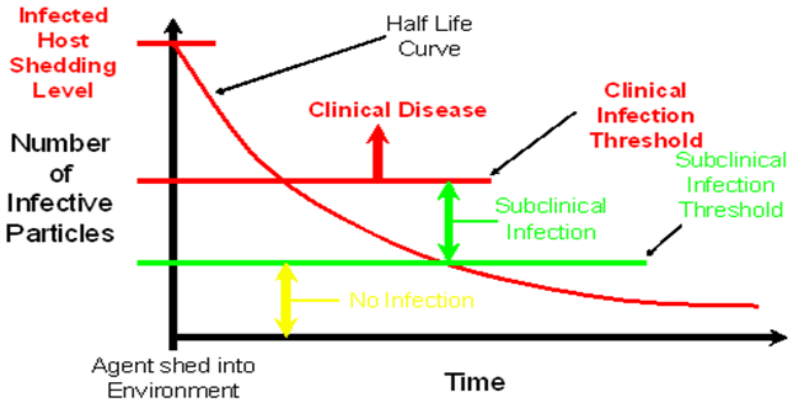
Climate

vetworks

- Ventilation

Minimum Ventilation Fan With House Closed

Infection pressure



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Vaccination

- Protection against disease NOT infection
- Immune reactions are very energy consuming
  - Balance protection – energy loss
- Importance well thought-out vaccination scheme
  - Health benefits >> energy loss
  - Improved production results: good return on investment
  - Avoiding double loss: vaccinated but still sick

Vaccination can not compensate for management flaws!

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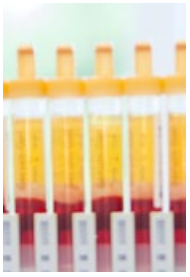
Monitoring

vetworks

- Serology & Microbiology
  - Subclinical infections
  - Choose vaccination programs
  - Vaccine efficacy
  - Maternal immunity levels

- Regional disease information
  - Colleagues
  - Results from regional monitoring programs
    → Rapid detection rapid eradication



- Other farmers and their problems / solutions
  - Same house build
  - Same climate system

Biosecurity

vetworks

- Goal: Keep diseases **OUT & IN**
- External & Internal biosecurity
- Structural & Operational biosecurity

ANIMALS – PEOPLE – INFRASTRUCTURE

Biosecurity

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Biosecurity

- People
  - Bird – free
    - Personnel does not visit other farms
    - Personnel does not have birds at home
  - Visitors and their vehicles → Parking
  - Disinfection: boots / shoes need to be clean first !
  - Shower + new clothes
  - Change clothes per house

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Biosecurity

vetworks

- Animals
  - New birds / eggs
  - All in / All out
  - Pest control: rodents & insects
  - No pets
  - Act fast when sick: infection pressure!
    - Veterinarian
    - Strict selection
    - Try to keep it in one house: extra ‘operational’ biosecurity measurements
  - Respect management guidelines
  - Vaccination program

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Biosecurity

vetworks

- Infrastructure
  - Strict separation between on & off site
  - Strict separation between ‘Clean’ and ‘Dirty’ parts of the farm
  - Dead bird disposal
  - Litter storage & disposal
  - Water source & distribution
  - Vermin-free feed storage
  - Choice of materials
  - Every house its own equipment

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Biosecurity

vetworks

- Choice of materials & infection pressure

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Biosecurity

vetworks

- Choice of materials & infection pressure

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Biosecurity

vetworks

- Cleaning & Disinfection

Ideal scheme

- Dry cleaning: all visible dirt
- Fill up water lines with disinfectant
- Soaking with detergent preferably foam (incl. all materials and boots)
- Wet cleaning with hot water under high pressure (12 – 30 L / min)
- Flush water lines
- Dry out
- Visual control
- General disinfection
- Dry out
- Targeted disinfection (if needed)

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Biosecurity

vetworks

- Cleaning & Disinfection
  - Always bottom – down
    - Ventilation → ceiling → water- & feeding lines → walls → floor
  - Double check

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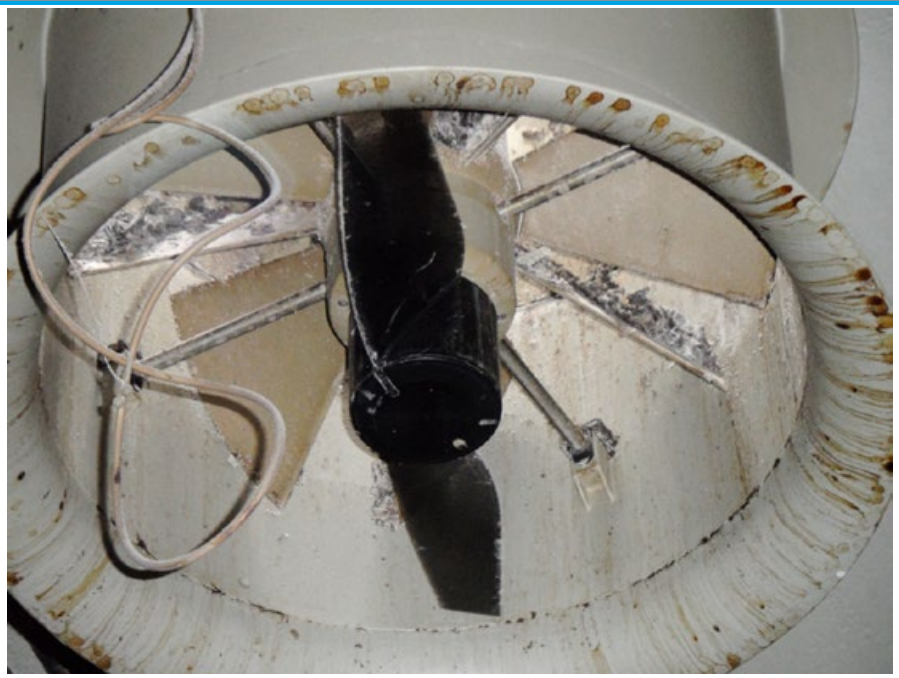
Biosecurity

vetworks



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Biosecurity

vetworks

- Disinfection

  Temperature – Contact Time – Concentration

  - Useless without proper cleaning

  - Choice of disinfectant: product information!
    - Spray, Fog, Foam, Cooling, Burning, Gas
    - Surfaces, water temperature & quality, activity, safety

  - Success =
    - Proper cleaning
    - Quality and suitability of disinfectant
    - Correct application

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Biosecurity

- Organization
  - **Assign one responsible person + authority**
  - Warning signs as reminders
  - Colors
  - Overview map of site
  - Checklists
  - Calendar
    - Pest control
    - Foot baths
    - Maintenance (f.e. bearings fans)
    - Training

Assume there is always a risk!

Thank you for your attention

Jan Willems

The art of vaccination: keys to success

Overview

vetworks

- Aim of vaccination
- Immune respons in poultry
- Different types of vaccines: pro's and con's
- How to perform successful vaccination
- Monitoring success of vaccination and identify mistakes

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Vaccination in poultry

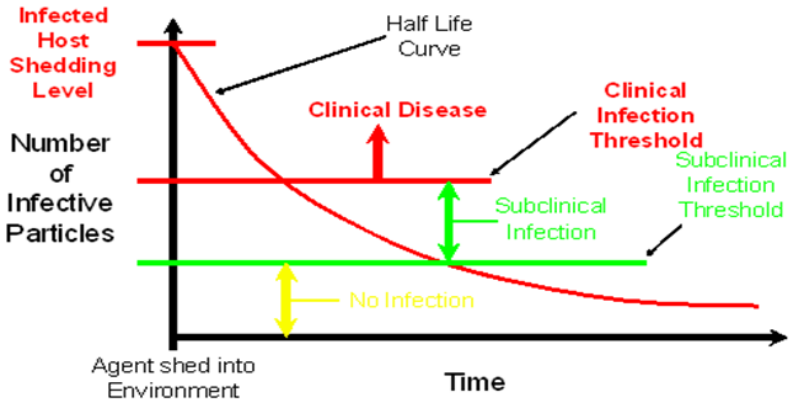
Special Features:

- Poultry Industry: mass production
  - Health care: based on prevention
- Vaccination application
  - Practical: mass application
  - Cost effective

Aim of vaccination

- Production of specific antibodies and immune cells
- Generate **sufficient** immunity in **all** animals in the flock
  - To protect the animal itself
  - To protect the progeny
    - Maternal immunity lasts about 3 weeks
  - To prevent clinical infection, but they can still be carrier
    - AI, Salmonella
  - To prevent zoonoses (*Salmonella*)
- **Never 100% protection especially not in field conditions!**

Infection pressure



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Vaccination

- Protection against disease NOT infection
- Immune reactions are very energy consuming
  - Balance protection – energy loss
- Importance well thought-out vaccination scheme
  - Health benefits >> energy loss
  - Improved production results: good return on investment
  - Avoiding double loss: vaccinated but still sick

Vaccination can not compensate for management flaws!

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What can go wrong?

Why do we get outbreaks in vaccinated flocks?

- Challenge by variant?
- Immunosuppression?
- Very short or long interval between vaccination and challenge?
- Inadequate application/take
- Interference between vaccines? Field infections?

Why monitoring? How to choose the right test?

- Clinical disease : diagnosis
- Detection of subclinical infections
- Showing freedom of infection
- Detection of the take of a vaccine
- Epidemiological research

Immune respons in poultry

- General defense mechanisms
- Specific Immune respons

General defense

- Health = good defense mechanism
- Good feed should offer the ingredients and structure
  - Balanced intestinal flora: so pathogens can not overgrow
  - Build-up of antibodies
  - Local immunity in the intestine

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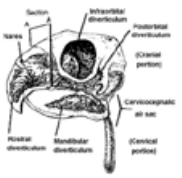
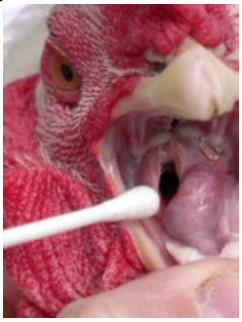
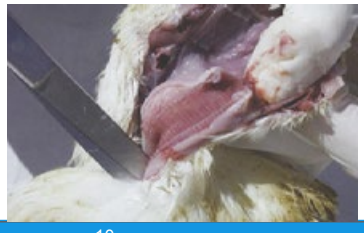
Anatomy: Look

- Nares, nasal cavity with conchae
  - Smell
  - Mucus production



- Oropharynx
  - Mouth +pharynx
  - From choanal opening at the bottom of the nasal cavity to the base of tongue
  - Glottis – opening into the larynx
    - Can open wide in respiratory distress
    - No vocal cords or epiglottis like in mammals





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Respiratory issues – easy to recognise...




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Bird Signals: Listen

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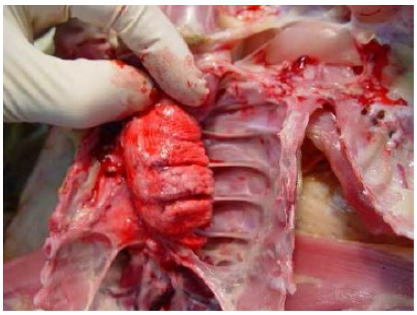
Look

vetworks

trachea



lung

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Anatomy of respiratory system

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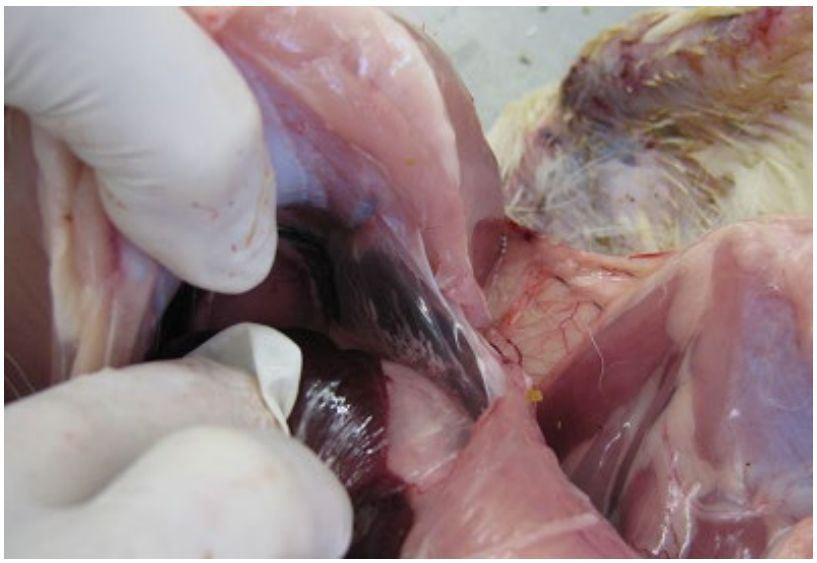
Air sacs, broiler



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Introduction to avian immune system

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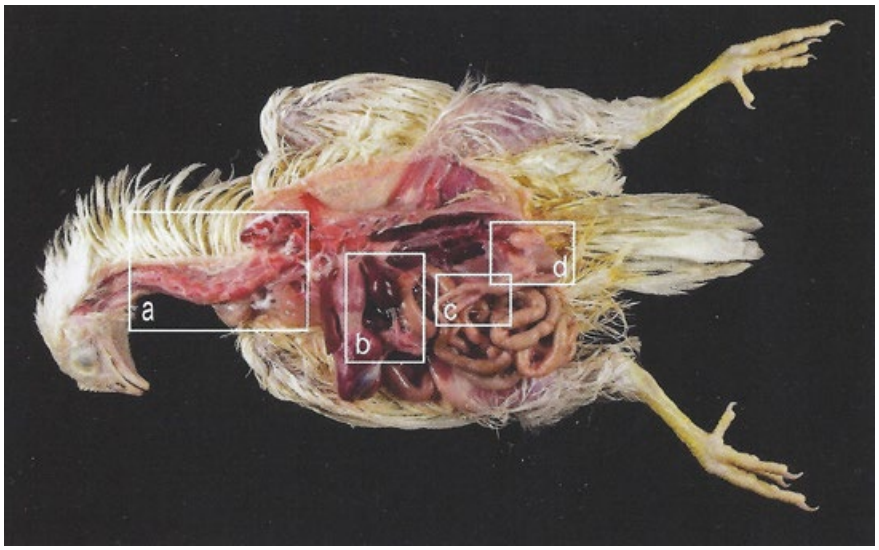
- Main immune organs of the bird



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Main immune organs of the bird

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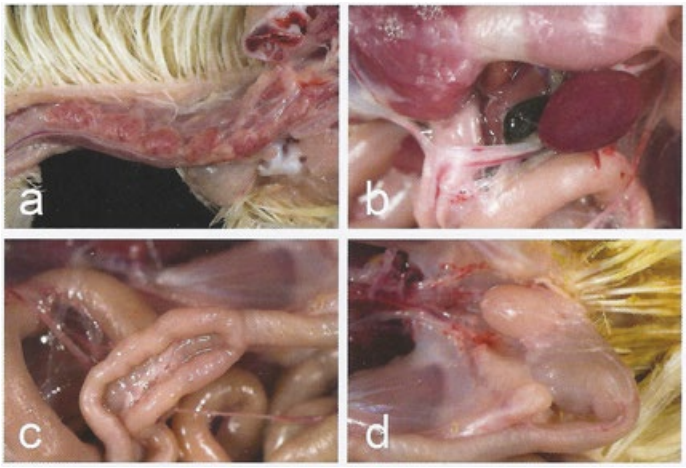
Thymus

Spleen



Cecal tonsils

Bursa Fabricii

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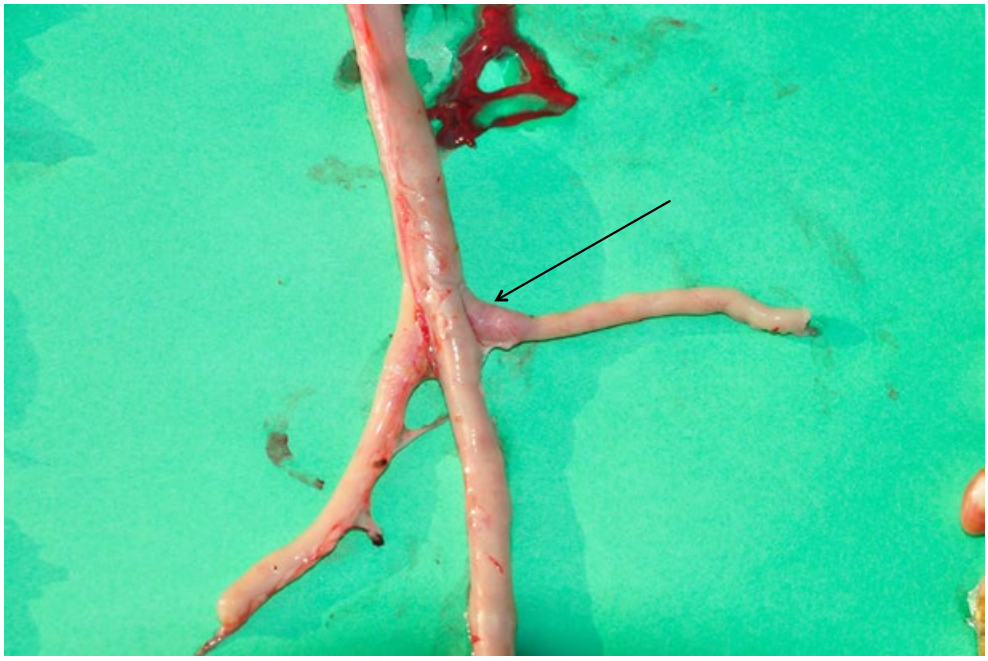
Cecal tonsils



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If this is not enough?

First line of defense: Innate immunity

- Quick and nonspecific

Second line of defense: Adaptive immunity

- Slow but very specific

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Second line of defense: antibodies

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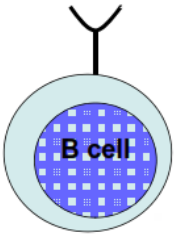
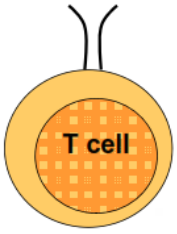
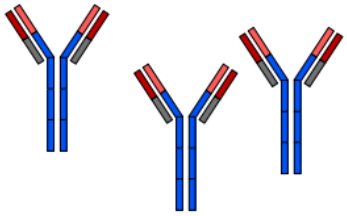
The adaptive immune system





Make antibodies



- Help B-cells
- Control infections

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How do we get antibodies?

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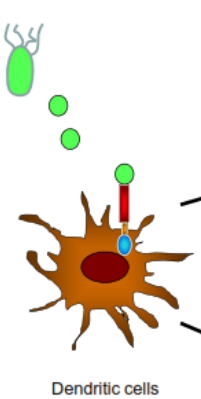
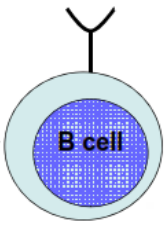
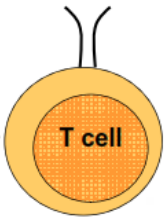


Dendritic cells



Make antibodies



- Help B-cells
- Control infections

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Humoral immunity

- Antibodies – Immunoglobulins (Ig) produced by B cells
  - IgM
    - First antibody produced after immunisation
    - 'class switch' to IgG or IgA in progress of immune response
  - IgG (IgY)
    - Predominant in chicken blood
    - Also produced after second immunisation
  - IgA
    - Most important Ig in mucosal immunity
- Maternal immunity
  - IgG transferred from oviduct to yolk sac
  - IgA and IgM transferred via amniotic fluid

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Memory response of immune system

- Specific B and T memory cell produced after contact with antigen
  - **Vaccination** is inducing memory response to protect from field challenge!
- Each following contacts with the same antigen
  - Quicker immune response
  - Higher affinity immune response

=> Used in re-vaccination (booster vaccination)

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Defence system of avian resp. tract

vetworks

High pressure on resp. system in intensive poultry production
-> dust, pathogens

Intact mucociliar cells

Damaged mucociliar cells

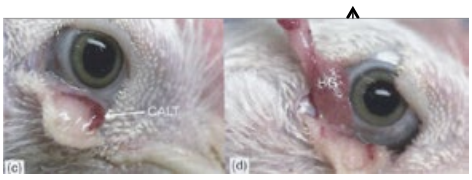
- Non-specific innate defence system
  - Cilia
    - Tiny hair-like structures in the trachea
    - Remove in direction of mouth entrapped particles
  - Continuous mucus secretion in trachea
    - If the mucus too thick, cilia cannot function properly
- Respiratory immune system in birds

Advantage of hatching in house? No damage to the mucosa by formalin/ dust?

Respiratory immune system

vetworks

- Paraocular lymphoid tissue: Harderian gland and CALT
  - Important for both T-dependent and humoral immune response (**IgA** !)
  - Eye drop vaccination



- Trachea - no special lymphoid tissue
  - But: extensive lymphocyte infiltration in *M. gallisepticum* and IBV models
- Bronchus-associated lymphoid tissue
  - Highly organised lymphoid nodules and diffuse lymphoid cells
  - Junctions of primary bronchus, caudal sec. bronchi and ostia of air sacs
  - Fully develops only after 6 weeks of age!
    - After life span of a broiler

Respiratory immune system

- Immune response in the lungs
  - Diffusely distributed leukocytes
    - Majority – macrophages and dendritic cells
      - Uptake and presentation of small size (<0.1µm) inhaled antigen
      - Mainly induction of systemic antigen-specific response
- Particles in respiratory tract
  - High load of aerosolized particle in poultry houses
  - Site of deposition depends on particle size
    - Nasal cavities and trachea – larger particles (3.7-7 µm)
    - Lung and cranial air sacs – smaller particles (ca. 1.1 µm)
    - Caudal air sacs – less than 0.1 µm
  - Upper resp. tract, trachea, till prox. part of sec. bronchi – mucociliary transport to mouth
  - Lungs, air sacs – phagocytosis by macrophages and epithelial cells

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Response of respiratory system to vaccination

- Many vaccinations are done via spray
  - First contact of the bird with the live vaccine via respiratory mucosa
  - Induction of local and systemic immune response
  - Immune response depends on the pathogen and application route
- Important role of local antibody response in protection from respiratory disease
  - Aerosol challenge via mucosal route (IBV, NDV)
    - Induction of local IgA in the Harderian gland, lacrimal IgA and IgG and local IgG in respiratory tract
  - Live mycoplasma vaccines
    - Local mucosal antibodies play a role in protection

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Specific Immune System

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- **Local Immunity**
  - After contact with live agent
  - Superficial in respiratory or intestinal mucosa
  - Protection of short duration
  - Usually group specific
- **Systemic Immunity**
  - After contact with live or dead agent
  - Not superficial, antibodies in blood, white blood cells
  - Protection of longer duration
  - Strain-specific

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Live and inactivated vaccines:

vetworks

Live vaccines

- easier (eye-drop, spray drinking water)
- local and systemic I
- sometimes pathogenic, vaccination reaction
- mostly spreading
- priming
- cheaper (per dose)

Killed vaccines

- more difficult (individual injection)
- only systemic I
- safe
- no spreading
- long protection
- booster
- more expensive
- more stress

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Newest vaccine technology

Recombinant vaccines

- **Immune complex vaccine**
- **Vector vaccine**

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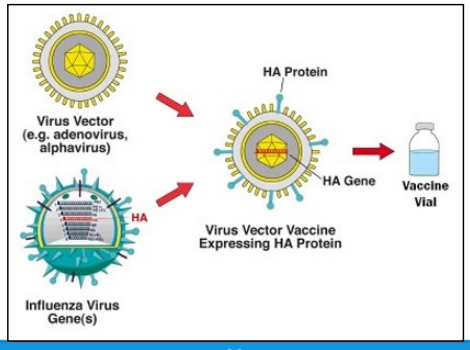
Recombinant vaccines – vector vaccines

- Live vaccines
- Genes coding for antigens are introduced into the vector genome - virus or bacteria -
- When the vector multiplies, the inserted segment is replicated.
- Vaccination with a recombinant vaccine: Immune response against the vector but also against the antigen included without using the actual disease agent.



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Vector vaccine

- Level of protection similar to conventional vaccines
- Stimulate cellular and humoral immunity

- The main viral vectors:
  - Herpes virus of turkey (HVT) and the Poxvirus
  - Because their genomes are large enough to accept large inserts

- Examples:
  - HVT expressing NDV
  - HVT expressing ILT
  - HVT expressing IBDV
  - FP expressing AI
  - FP expressing NDV
  - FP expressing ILT

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Immune complex vaccines Icx

- The antigen is bound to specific antibodies that protect it from the host recognition system and action of MDA
- The antigen is released exactly at the right time on an individual bird per bird basis

- Very interesting for IBDV!
- Live attenuated IBDV strain + homologous specific antibodies
- This vaccine is insensitive to high levels of MDA
- The potential adverse effect of the IBDV strain having considerable residual virulence in chickens with low MDA levels is prevented by the homologous antibody added to the vaccine.
- The vaccine virus can start colonising the bursa, when MDA are low

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Recombinant vaccines

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- **Advantages**
  - No post vaccination reactions
  - No local reactions caused by the use of inactivated vaccines
  - No recombination of wild and vaccine viruses in the environment
  - No interference with maternal immunity
  - No latent carriers after vaccination
  - The use of a recombinant vaccine based on a continuous replicating virus (such as Herpes virus for example) could eliminate the need for booster vaccinations with either live or inactivated vaccines in the field.
  - The use of recombinant vaccines also allows the use of the so-called “DIVA” system (English: Differentiating Infected from Vaccinated Animals) to differentiate vaccinated birds from infected birds.
- **Disadvantages**
  - High cost to develop: the genes for the desired antigens must be located and expressed efficiently in the new vector

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Inactivated vaccines

vetworks

- Advantages
  - No systemic vaccination reactions
  - Long lasting protection
  - No transmission of the agent
  - Less danger of interference
  - More combinations possible
- Disadvantages
  - More expensive per dose
  - Retarded response
  - Individual application
  - In general “primer” vaccination needed
- Longer lasting immunity and more homogenous antibody levels achieved after the use of inactivated vaccines

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Composition of inactivated vaccine

- Antigen particles are in the aqueous phase
- Adjuvant :
  - Enhances the immunogenicity of the antigen
  - Long lasting immune respons
  - Stability, safety, injectability
- Adjuvants
  - Saponins
  - Aluminum salts
  - Mineral oil
    - Water in oil emulsions
    - Oil in water emulsions
    - Water in oil in water emulsions
- **Different types of adjuvants produce different immune responses and different local reactions**

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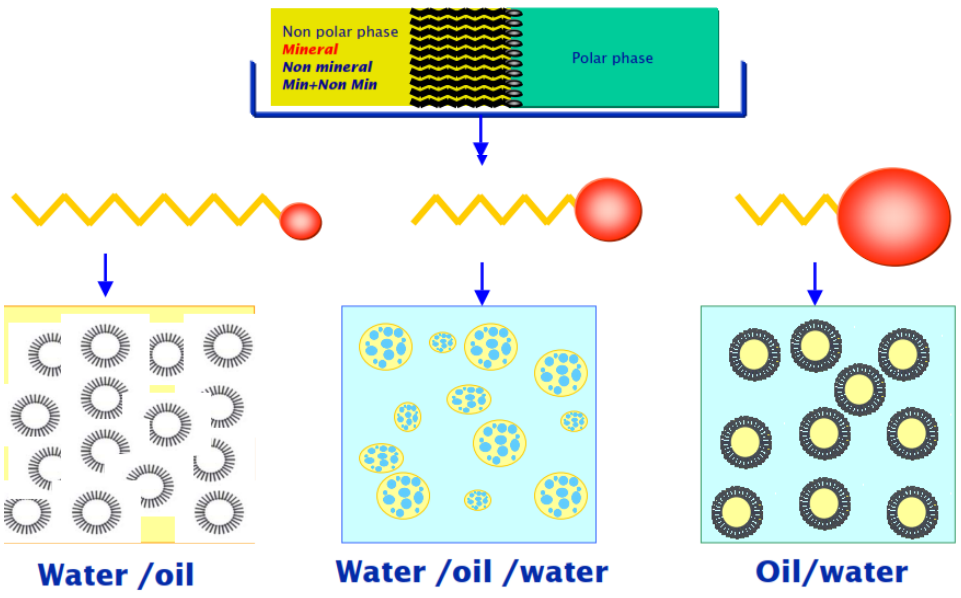
Adjuvants



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How to perform successful vaccination

- Only vaccinate healthy animals
- Take into account maternal immunity
  - Can inactivate the vaccine
- No immunosuppression
  - Immunosuppresive viruses
    - eg Gumboro, CAV
  - Stress, deficiencies, mycotoxins…
- Good quality vaccine, good storage (cold chain)
- Proper vaccination technique
  - For respiratory diseases, cox: administration via natural infection route

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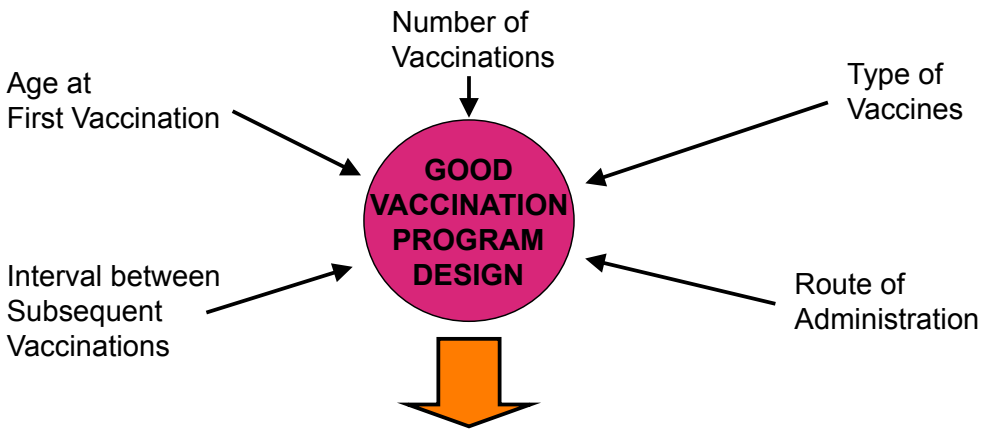
Successful Vaccination Program



1. Stimulation & Maintenance of Protective Immunity
2. Development of Immunologic Memory

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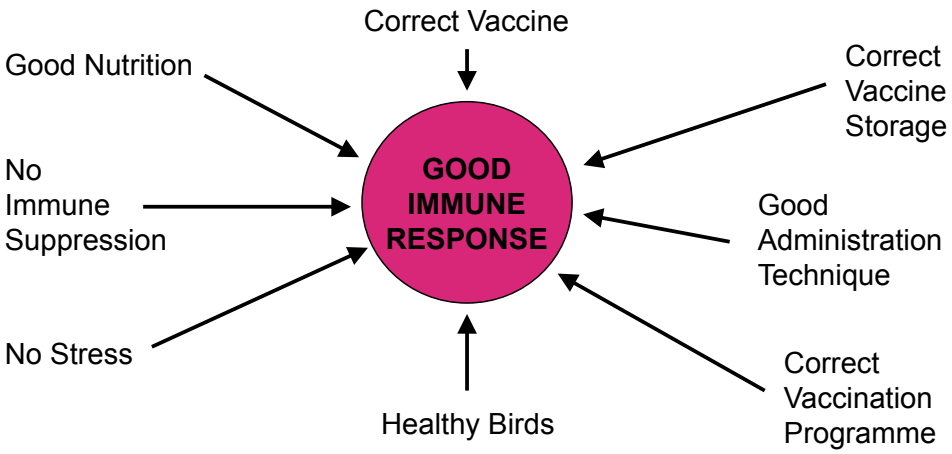
Requirements for Good Immune Response



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Successful vaccination

- Proper administration
  - In ovo
  - Drinking water
  - Spray on birds
  - Spray on feed
  - Aerosol
  - Eyedrop
  - Wing web
  - Injection

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Vaccination technique

- Inactivated vaccines:
  - No multiplication in the animal
  - Individual injection
- Live vaccines:
  - Multiplication in the animal, spread in the flock
  - Different techniques are possible
    - eg NCD drinking water, spray, aerosol, in ovo
  - Sometimes 1 technique
    - Pox: wing web , CAV injection

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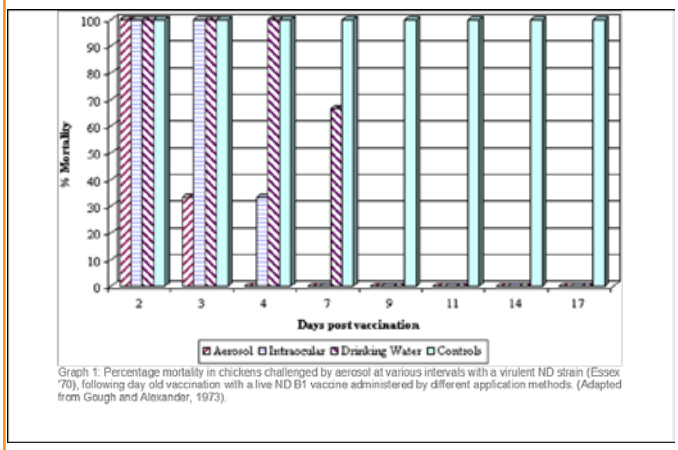
Vaccination technique

- Succes of vaccination depends on the vaccine strain being presented to the correct target cells.
- Efficacy of a live Newcastle Disease (ND) vaccine administered by different routes: aerosol, drinkwater, eyedrop 1973 Gough and Alexander
- ND virus challenge within 3 to 4 days of a ND vaccination



Graph 1: Percentage mortality in chickens challenged by aerosol at various intervals with a virulent ND strain (Essex '70), following day old vaccination with a live ND B1 vaccine administered by different application methods. (Adapted from Gough and Alexander, 1973).

Time needed for protection
Aerosol 4 days
Intraocular 7 days
DW 9 days

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Which technique?

- Depending on:
  - Type of vaccine: live or killed
  - Equipment
  - Experience of vaccinator
  - Efficacy: respiratory pathogen most effective if administered via respiratory system
  - Risk of vaccination reaction

Vaccination technique

- Dose: calculate exactly
  - Number of animals = number of doses
  - Drinking water: test or calculate how much water is needed for all animals to drink in 2 h
  - **Yesterday's water intake: registration!**
  - **Age in days x 1.5L = number of litres/1000 birds**
- Material, equipment:
  - Must be clean and well functioning
  - No products that can inactivate the virus 48 h before vaccination
  - Best to use for vaccination only for eg bucket, backpack sprayer .....

In ovo

- In ovo: individual injection very precise
- Hatchery at 18D transfer between setter and hatcher
  - Injection at the right site amniotic fluid or right breast area ideally
    - D18 amniotic fluid, D 19 in the embryo (P.Wakenell et al. 2002)

- Advantage
  - Vaccination in the embryonic stage: early immunity of DOC
  - Individual application
  - No risk of contamination, no individual handling DOC (labour intensive)

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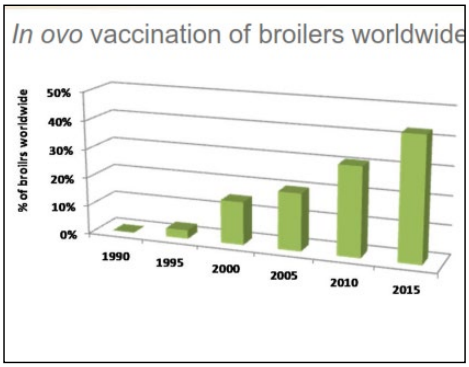
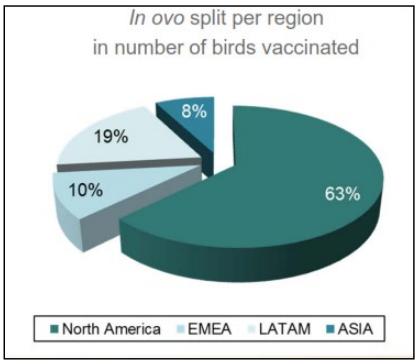
In ovo vaccination





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In ovo vaccination

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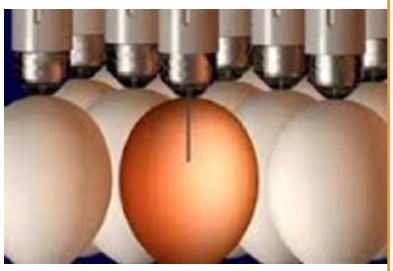
- Restrictions
  - Not all vaccines can be given in ovo (no killed oil vaccines)
- Available vaccines:
  - Live vectored Md, IBD,FP, ILT
  - Coccidiosis
- High standard sanitation in hatchery!
  - Equipment, maintenance!
  - Environment , Aspergillosis



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Hatchery: spray cabinet

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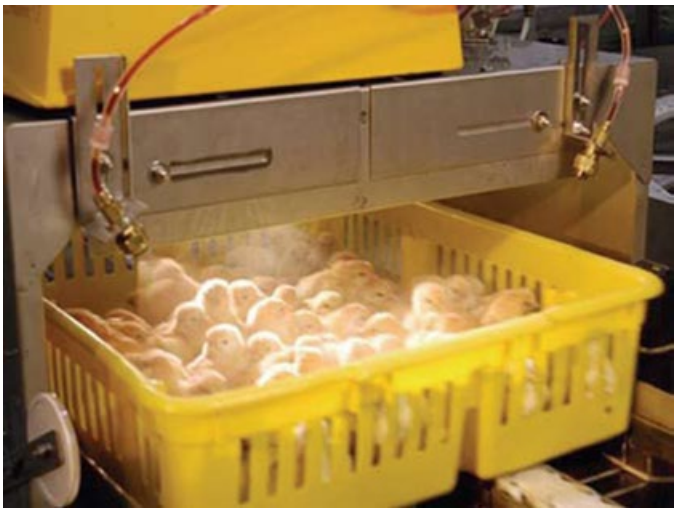
DOC spray vaccinations in Hatchery



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Drinking water

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- Live vaccine
- Relatively easy to perform
- Not labour intensive
- Less suitable for respiratory infections

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Drinking water: attention points

vetworks

- No disinfectant or acid 48h before
- Check if all animals can drink
  - Enough space and no blocked or leaking nipples
- Empty the drinking line completely (check at end point)
- Check water consumption day before or calculate
  - **Age in days x 1.5L = number of litres/1000 birds**
- Stabiliser
  - Skimmed milk: 2L/100L or 20gr powder/100L
  - Aviblue type (or other) protector 1 dosingcap/200L
- Vaccination while feeding, morning after dark period
- To stimulate drinking birds should be deprived of water 2h prior

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Drinking water: attention points

- Stabiliser
  - Skimmed milk: 2L/100L or 20gr powder/100L
  - Commercial protector
  - Add to solution BEFORE vaccine
- Vaccination while feeding, morning after dark period
-
- To stimulate drinking, birds should be deprived of water for two hours prior to the vaccination

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Drinking water

- Control water intake: blue dye
  - Blue dye stains tongue and crop blue: if 90% colored, well vaccinated

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Spray

- Live vaccine is sprayed over the animals
  - NCD, IB, TRT
- Spray reaches the upper respiratory tract
  - Drop: coarse >100µ or fine 50µ-100µ 0.5 – 2l/1000birds
  - Smaller drops go deeper in the airways
- Better and faster immune response than drinking water but also more reaction

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Aerosol: atomist

- Live vaccine is sprayed over the animals (NCD) by atomist
- Reaches the lower respiratory tract: drop < 50µ
- 0.3-0.6 L/1000 birds
- More reaction
- Better and faster immune response
- Reasonably fast implementation
- Some loss in the environment

54

Day old chicks

- Vaccination on the farm: spray
  - In the crates
  - Put chick paper with feed in a limited space so they stay close together

55

Hygiene!

- YES

- NO

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Spray: attention points

vetworks

- Clean equipment
  - But no disinfectant!
- Quality of water: add skimmed milk or vaccine protector
- Make sure the vaccine reaches the birds
  - Put out the ventilation until 20 minutes after spraying
  - Best when chickens are perching
  - There should be enough light

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Spray: attention points

vetworks

- Availability vaccine
  - Good storage
  - Dissolve the vaccine in clean environment, first add blue dye
  - Open the vials under water
  - Wear gloves
  - Check right dosage, adapt speed if necessary
  - Ambient conditions: temperature & humidity
    - Evaporation rate

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Spray vaccination: how much water needed?

- Number and age of chicken.
- Equipment to be used.
- Ambient conditions: temperature & humidity (rate of evaporation).
- All chicks have to be hit by the spray (shake the head for a moment).
- All chicks heads have to get slightly moist.
- Vaccination in two applications might be necessary.
- Perform a sham vaccination in order to assess time and volume of water needed!

Control blue dye

good

bad

Eye drop

vetworks

- Live vaccine (AE, ILT, MG, MS)
- Very good immune response (Harderian gland)
- Little reaction
  - sometimes unilateral conjunctivitis
- Individual administration
  - each animal gets his dose
  - Very labour-intensive

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Eye drop

vetworks

- Check dosage
  - Compare with 1000doses injection or pox
  - Some devices have a automatic counter
- Do not touch the eye
  - The drop will not be completely formed
  - The dosage is not correct
  - Damage to the eye
- Use the correct diluent
- Calibrated dropper

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Wing web

- Live vaccine pricked into wing membrane (Fowl pox)
- Individual application: each animal gets his dose
- Very labour intensive: each animal individually

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Injection

- Live or dead vaccine (IB, NCD, CAV end rearing)
- In ovo at the hatchery
- Subcutaneous or intramuscular in breast or leg
  - Sometimes necrosis lesions at site of injection
  - Careful with birds for meat production (meat turkeys, broilers)
- Individual administration: each animal gets his dose
- Very labour intensive

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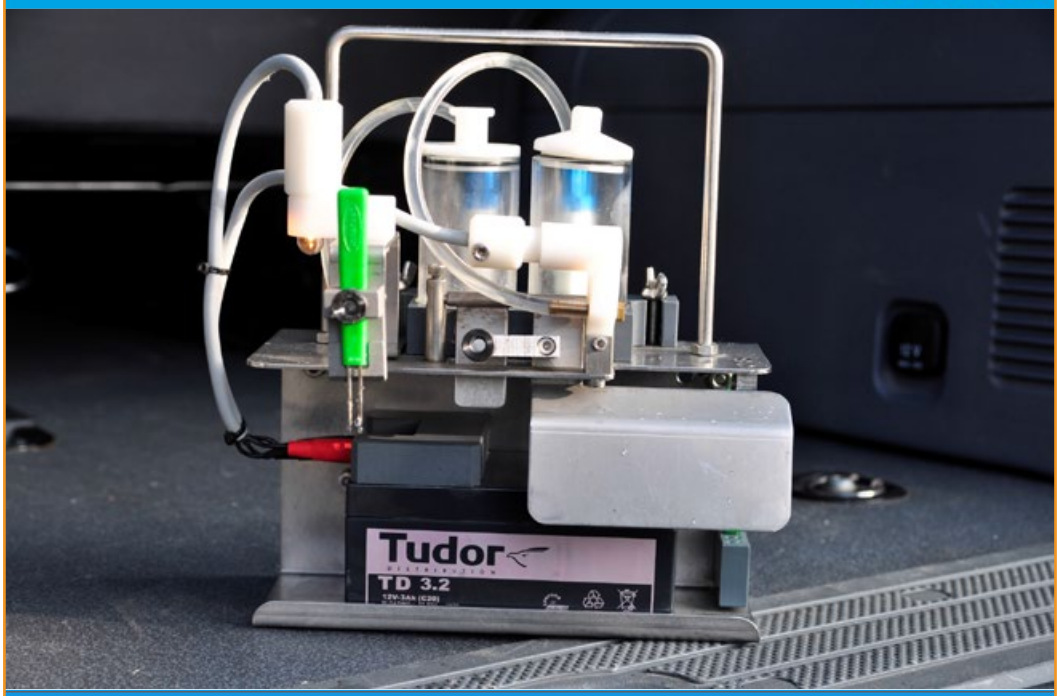
Injection

- Use sterile equipment only.
- Change needle every **800 birds.**
- Dilute live vaccines in their **appropriate diluent.**
- Oil vaccines should have room-temperature before application.
- Needle diameter:
  - 1,2 mm = 18 G; Length of 0,7 cm for s.c.
  - 1,0 - 1,3 cm for i.m.
- Subcutaneous route: Inject into the lower part of the neck.
- Intramuscular route: Inject tangential into the breast muscle.

65

Eye drop and wingweb



66

Eye drop

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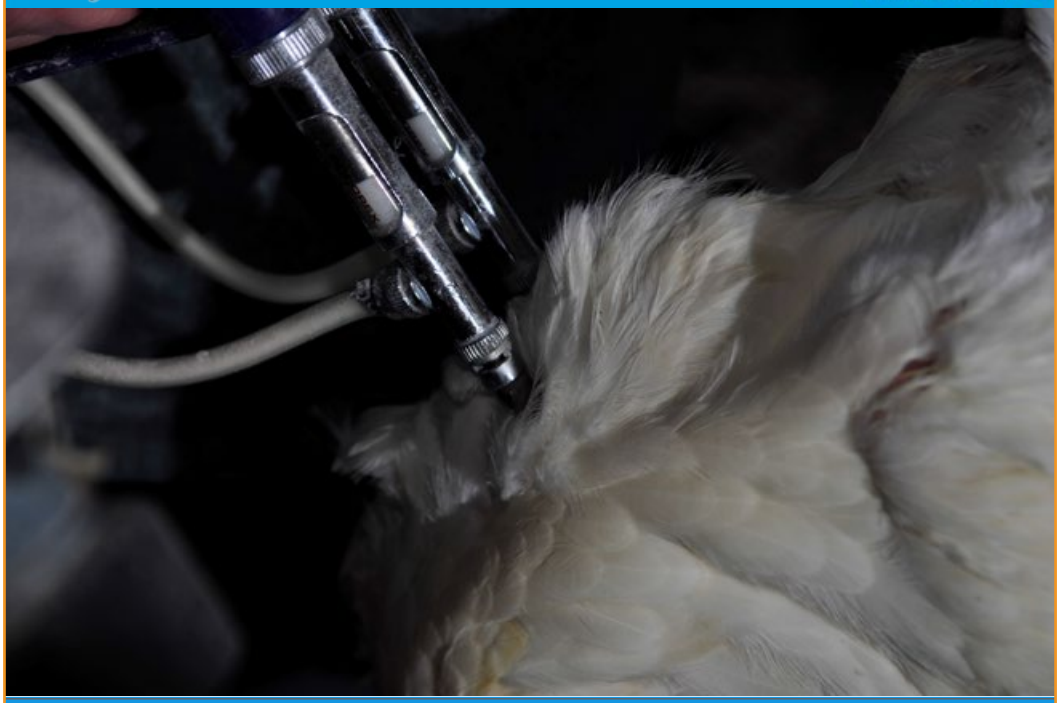
2 injections in the breast



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1000 birds per hour
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69
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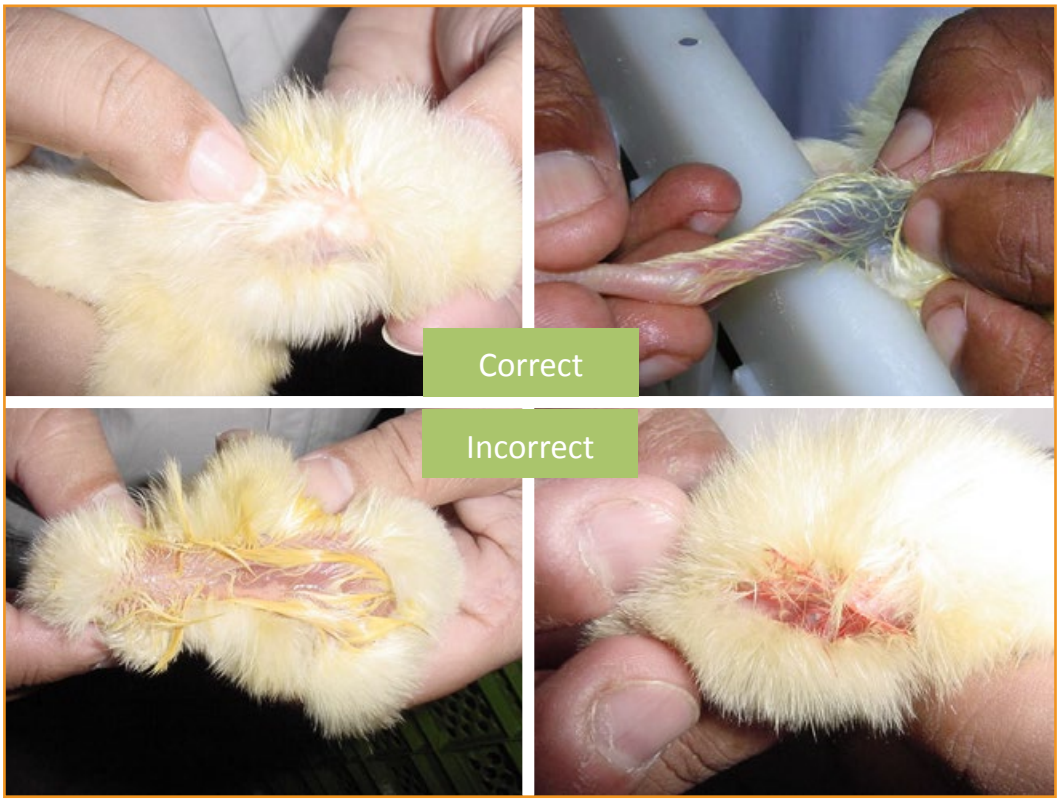
Correct
Incorrect

Storage

- Live vaccines!
- Freeze dried
  - Stored at 4 – 8 °C
  - Standard refrigerator
- Frozen
  - Liquid Nitrogen
  - - 80°C freezer

Storage

Temperature logger during transport

Check temperature of the fridge
- On arrival of vaccine
- During storage

Do not store vaccine in the door
- Diluent OK

Do not store food or beverage in the refrigator with vaccines. The door will be frequently opened

Storage: Pitfalls

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- Summer in Belgium: true story
  - Vaccine delivery by the vet day before arrival of the DOC
  - Refrigerator is not plugged in
  - Breach in cold chain!
  - Refrigerator is plugged in.
  - Some one needs to do some repairs and unplugges the refrigerator again !
  - Next day: vaccines are not considered to be stored well
  - To be sure, all vaccine is replaced (at the expense of the farmer)
  - **Use temperature loggers!**

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Overview

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- Aim of vaccination
- Immune respons in poultry
- Different types of vaccines: pro's and con's
- How to perform successful vaccination
- **Monitoring success of vaccination and identify mistakes**

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Respiratory issues – easy to recognise…

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Monitoring

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- Serology & Microbiology
  - Subclinical infections
  - Choose vaccination programs
  - Vaccine efficacy
  - Maternal immunity levels

- Regional disease information
  - Colleagues
  - Results from regional monitoring programs
    → Rapid detection rapid eradication
- Other farmers and their problems / solutions
  - Same house build
  - Same climate system

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Monitoring

- Efficacy of vaccination
  - On farm: correct administration of vaccine
  - Retrospective evaluation based on serology
- Differentiate suspected infections
  - New variant strains eg IB –QX
  - Complicated interpretation: live vaccines can give higher response if concurrent infection
  - Check for all respiratory diseases: mixed infections TRT, ORT, Ms..
- Serology
  - Live vaccines: 3-5 weeks after vaccination
  - Inactivated: 5-8 weeks after vaccination
- Pox: check after 7- 10 days pox lesion

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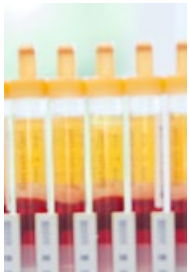
Monitoring

Interpretation of vaccination results by ELISA:

Key components of immune response after vaccination



1. **Intensity of Response**: **Mean Titer**.

2. **Uniformity of Response: % CV** (coefficient of variation).

3. **Persistency of Response: Mean Titer response over Time**

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Interpretation of vaccination results by ELISA

1. **Intensity of Response**: **Mean Titer**

- Do the birds develop sufficient titers levels that are in the expected range for the vaccine used?
- **" Baseline Titers"**
- Baseline titer values may vary according to
  - type of bird
  - age
  - vaccine type, method of application
  - vaccination program
  - other factors
  - testkit used
- Therefore, one should make their own baselines for there own vaccination programs and local conditions.

Interpretation of vaccination results by ELISA

2- Uniformity of Response: % CV

- Coefficient of variation: is the vaccine actually getting to the all birds or not?
- General guidelines for % CV following vaccination

% CV	Uniformity
Less than 30 %	Excellent
From 30-50 %	Good
Greater than 50 %	Need to Improve

- Vaccination Index = Mean titer/ %CV
  - Higher is better

Interpretation of vaccination results by ELISA

3. Persistency of Response: Mean Titer response over Time

- Do titers persist long enough over time, or is another vaccination needed to boost titers above minimum protective levels.

Interpretation of serology for diagnosis

- High titres compared to the baseline indicate a challenge
  - It does not say there was a problem (if the vaccination works well)
- To confirm relation challenge and clinical problem
  - Paired serology (start problem and 2-3 weeks later)
  - Detection of the pathogen and differentiate from vaccine
  - PCR and sequencing

Vaccination Baselines Titers in Broiler

Test	Vaccine Type	Mean titer range at 35 - 40 days	Suspect Titer Infection
NDV	-Live, 2x D.W	2000 – 5000	More than 7,000
	-Live, 2x Spray	4000 – 8000	More than 10,000
IBV	-Live, 1x (H120)	800 – 1500	More than 3,000
	-Live, 2x (H120)	2000 – 4000	More than 6,000
IBD	-Live, 1x (intmed.)	2500 – 4500	More than 7,000
	-Live, 2x (intmed.)	3000 – 6500	More than 9,000

Vaccination Baselines Titers in layers or Breeders:–

Test	Vaccine Type	Mean titer range	Wks after Vac. To test
NDV	-Live (Lasota)	2,000 – 8,000	2 – 3 wks
	-Inact.	10,000 – 15,000	4 – 7 wks
IBV	-Live (H120)	2,000 – 4,000	3 – 5 wks
	-Inact.	6,000 – 17,000	5 – 7 wks
IBD	-Live (intmed.)	2,500 – 7,000	3 –5 wks
	-Inact.	7,000 – 12,000	4 – 7 wks

Example for Organized Monitoring Program Breeders / Layers

Age	Sample	Test
Day 1	-Transfer box paper - Serum	- Salmonella. - MG – IBD – SE-SP/G - AI
Week 9	- Cloaca swabs - Serum	-Salmonella - ND – IBV - etc
Week 16	- Droppings - Serum	-Salmonella - Se/St- MG –ND – AI -etc
Week 22	- Droppings - Serum	-Salmonella - SP/G-ND – AI – MG -etc
Week 45	- Droppings - Serum	-Salmonella - Se/St- MG –ND – AI -etc
Week 62	- Droppings - Serum	-Salmonella - Se/St- MG –ND- AI -etc

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Example for Organized Monitoring Program Broilers

Age	Sample	Test
Day 1	-Transfer box paper - Serum	- Salmonella. - MG – IBD - AI
- 10 days before exit	- Droppings	- Salmonella
- Marketing Age	- Serum	- ND – IBV – AI - IBD

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Monitoring: audit vaccination team

- **Audit point one: Equipment**
  - Multi-dose syringes compatible with mineral oils used as adjuvants.
  - Syringes must be calibrated prior to use and at regular intervals during vaccination procedure.
  - Syringes and all ancillary equipment must be sterilized prior to use.
  - Correct size needle must be selected according to age of birds being vaccinated, site of injection and type of vaccine being administered.
  - Needles should be regularly replaced, at least once every thousand birds.

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Monitoring: audit vaccination team

- **Audit point two: Vaccination technique**
  - Check vaccinated birds for deposition of vaccine at the correct site
  - Wet feathers and vaccine on the floor: part of the dose is lost
  - Blue dye: eye drop, drinkwater, spray

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Monitoring: audit vaccination team

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- **Audit point three: correct dosage**
  - Check the doses of vaccine used to the number of birds.
  - Overdosing is costly, under-dosing results in poor immunity.
  - Serological response: indication of the accuracy of vaccination.
  - A poor %CV in combination with a low mean titre: a large percentage of birds are being missed or are not receiving the full vaccine dose
  - The key to vaccine administration by injection is: "Quality is more important than speed!" (be careful with payment per dose applied!)

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Vaccination program: considerations

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- What vaccinations are required by law? Example Belgium
  - NCD, *Salmonella* in layers
  - Trade restrictions
- Diseases at risk
  - Virulent field strains in the area or previous flock infection
  - Protection needed for offspring – AE
- Availability of vaccines
- Best age to vaccinate?
  - Consider interference of maternal antibodies
- Booster
  - Best immune response: priming live vaccine and booster inactivated
  - Booster with field strains IB
- Right interval between vaccinations targetting the same organs
  - IB and NCD

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Vaccination program: considerations

- Vaccine residual pathogenicity?
  - In the vaccinated bird: IB, NCD
  - Vaccine can cause drop in egg production, effect on progeny AE
- Plan monitoring vaccination
  - On the date of vaccination plan when and how to monitor efficacy

Vaccination program broilers – example Belgium

D1	**Newcastle Disease**	Live vaccine - full dose - coarse spray
D1	**Infectious Bronchitis**	Live vaccine - full dose - coarse spray
D10-18 (legally required period for ND vaccination) D14-18	**Newcastle Disease** **Infectious Bronchitis**	Full dose - fine spray or drinking water Full dose - coarse spray
D21-28 Dependent on serology on D1	**Gumboro**	Full dose – drinking water

Vaccination program layers

vetworks

FARM location House NR :			date of birth breed
age	**vaccine**	***DOSE***	***METHOD***
STANDARD vaccinations			
age	vaccine	dose	method
D1	Vaxxitek	1	SC
	Rismavac	1	SC
	IB Ma5	1	spray
	IB 4/91	1	spray
D7	Salmonella E+T	1	drinking water
D14	ND	1	spray
32D	IB QX	1	drinking water
6 weeks	Salmonella E+T	1	spray
7 weeks	ND	1	spray
	IB primer	1	spray
12 weeks	PD	1	wingweb
	AE	1	eye drop
	ILT	1	eye drop
	ND	1	intramuscular
15 weeks	H52	1	spray
16 weeks	Salmonella E+T	1	drinking water

Optional vaccinations			
8 weeks	TRT	1	spray
12 weeks	ND+EDS	1	intramuscular
	IB+ND	1	intramuscular
	RT+IBmulti+ND +EDS	1	intramuscular
16 weeks	TRT	1	spray

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Vaccination program Broiler Breeders

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STANDARD vaccinations			
D1	Vaxxitek	1	SC
	IB primer	1	spray
D7-10	Paracox8	1	drinking water
D12	IB 4/91	1	drinking water
	ND Clone 30	1	drinking water
D14	Salmonella E+T	1	drinking water
3 weeks	Gumboro D78	1	drinking water
6 weeks	IB primer	1	spray
	ND Clone 30	1	spray
8 weeks	PD	1	wingweb
	AE	1	eye drop
	ILT	1	eye drop
	REO	1	intramuscular
	autovaccin E. coli	1	intramuscular
	CAV	1	intramuscular
9 weeks	Salmonella E+T	1	drinking water
10 weeks	IB-QX	1	spray
12 weeks	RTV 8544	1	spray
16 weeks	Salmonella E+T	1	drinking water
17 weeks	IB multi-ND-RT-EDS	1	intramuscular
	autovaccin E. coli	1	intramuscular
	REO inac	1	intramuscular
19 weeks	IB primer	1	spray

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Monitoring

- Monitoring stimulates people to do a good job
- 'To measure is to learn'
- Detect mistakes and improve
  - part will result in better results next flock
  - part will result in a more appropriate vaccination schedule
- **Excellent opportunity for the professional to show his/her added value**

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Thanks for your attention

Jan Willems

Coccidiosis life cycle and importance in poultry production

Overview

vetworks

- Coccidiosis : the parasite and life cycle
- Coccidiosis: the disease and link with intestinal health
- Economical importance of coccidiosis

1

What is coccidiosis?

vetworks

- **Parasitic** disease, **protozoa** (unicellular) of genus ***Eimeria***
- In **chickens** - 7 species, 6 important (*E. praecox*???)
- **Broilers** (till +/- 6 weeks):
  - *E. acervulina*
  - *E. maxima*
  - *E. tenella*

  } **Lesion scoring**
  - *E. praecox*
  - *E. mitis*
- For **older chickens** also important 2 species
  - *E. necatrix*
  - *E. brunetti*

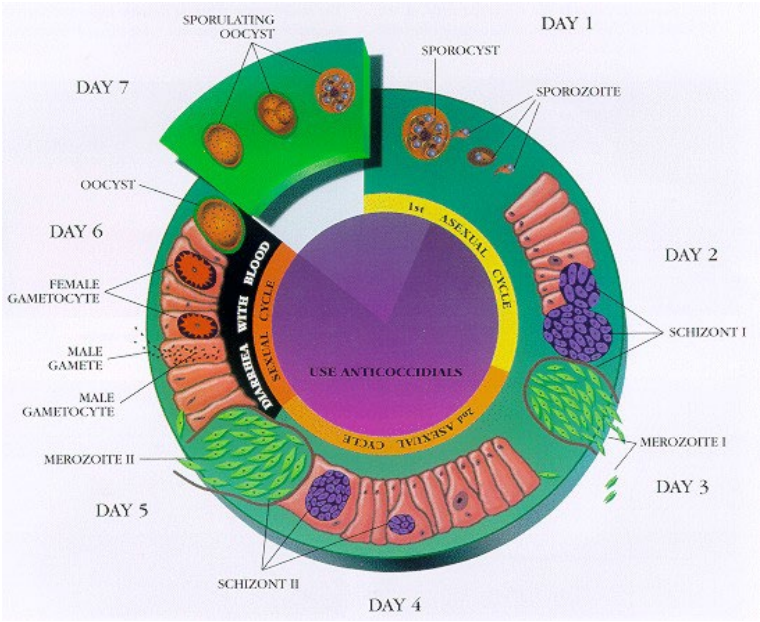
  } **Lesion scoring**

Eimeria spp.

vetworks

- **Direct life cycle** (no intermediate host)
- **Species specific**
  - Morphology
  - Host specific: chicken – turkey – rabbits - ...
  - Immunity (no cross-immunity)
  - Tissue tropism / tissue lesions

Life cycle of *Eimeria* species



New cycle every **4-7 days** depending on species/strain

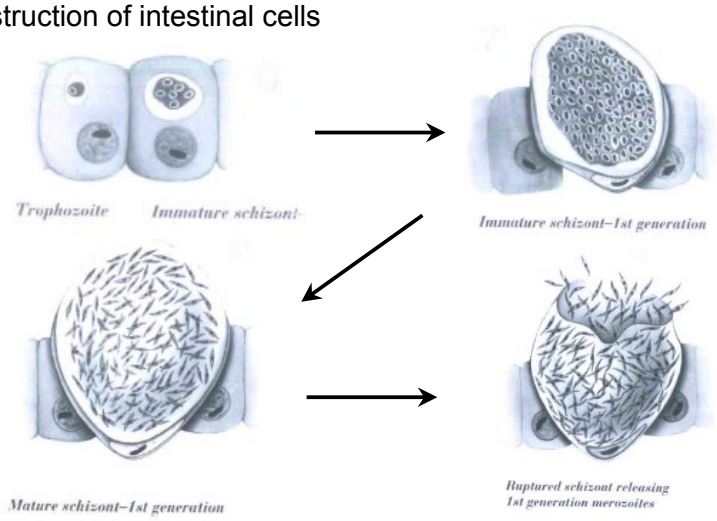
Eimeria species: lifecycle

- Oral uptake oocyst
- Crushed in gizzard: sporozoites free
- Sporozoites enter intestinal epithelial cells

Pictures from « Poultry Coccidiosis 3rd edition, Conway and McKenzie »

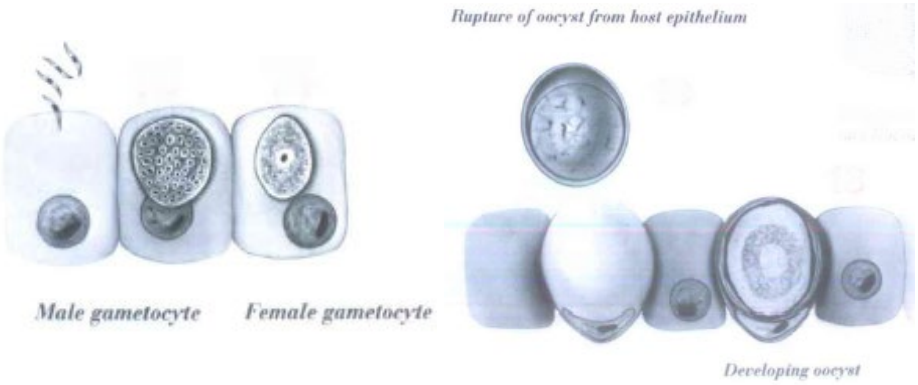
Eimeria species: lifecycle

- 2-3 asexual stages; trophozoite-schizont-merozoit
- Destruction of intestinal cells



Eimeria species: lifecycle

- 1 sexual stage : micro-macro-gametocyt
- Leads to unsporulated oocysts
- Excreted with faeces



Eimeria species: lifecycle

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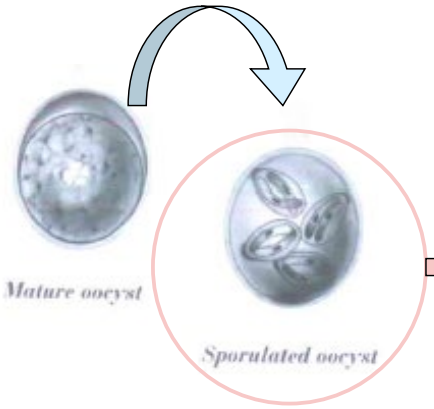
- Released with faeces : oocyst sporulates in litter



Very resistant !!!

- A new cycle can begin
- In theory each oocyst can lead to 2.52 million parasite cells

Sporogony

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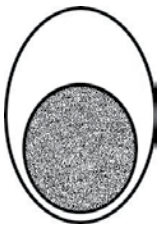

Non infectious unsporulated form

Temperature

Gas exchange (Oxygen)

Humidity

Infectious sporulated form

Very resistant !!!

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Sporulation

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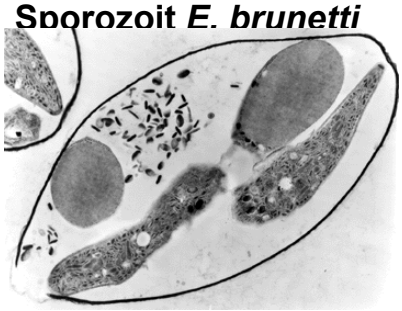
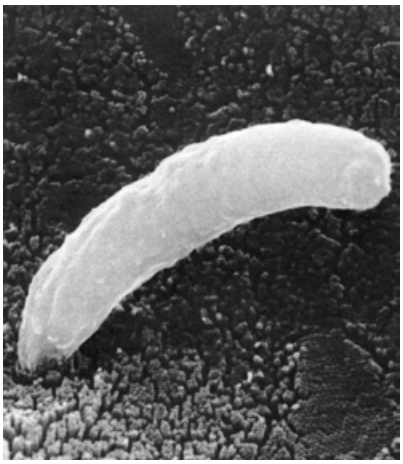
Sporozoit *E. brunetti*



Sporozoit entering intestinal cell



Life cycle of *Eimeria* species

vetworks

	Minimum prepatent period in hours and in days		Optimal sporulation time (in hours)
Eimeria acervulina	89	3,71	17
Eimeria maxima	121	5,04	30
Eimeria tenella	132	5,50	18
Eimeria brunetti	120	5,00	18
Eimeria necatrix	138	5,75	18
Eimeria mitis	93	3,88	15
Eimeria praecox	83	3,46	12

Reproductive potential and immunogenicity

Species	Max output/d (x1000)	To induce clinical coccidiiosis (x1000)	Pathogenicity	Immunogenicity
E. acervulina	432 000	100-2 000	+++	+++
E. maxima	36 000	20-200	++++	++++
E. tenella	65 000	50-200	++++	+
E. necatrix	12 000	20-100	+++++	+
E. brunetti	53 000	20-200	++++	+++

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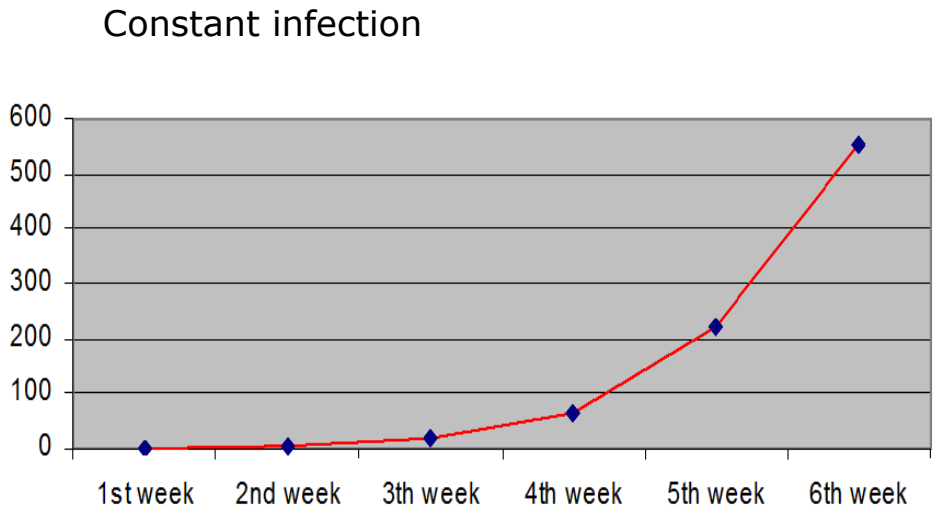
Theoretical shedding of oocysts



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Real situation

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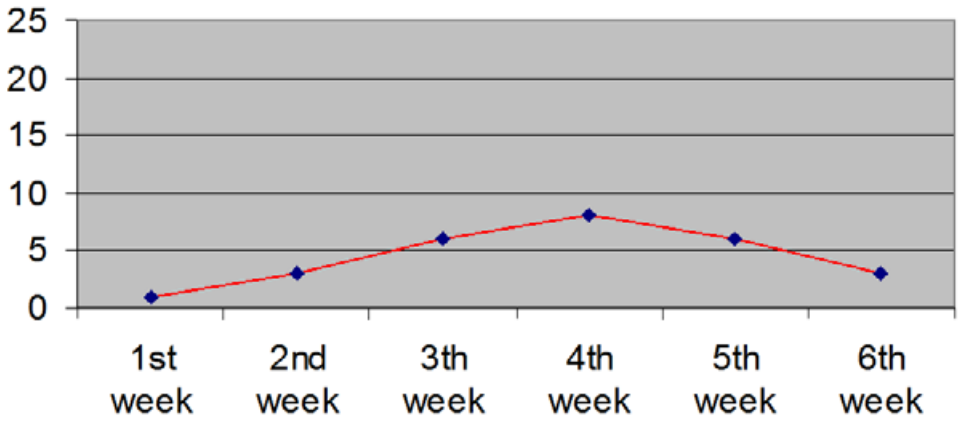
Oocysts shedding + Immunity + Competition



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Coccidiosis – self-confined infection

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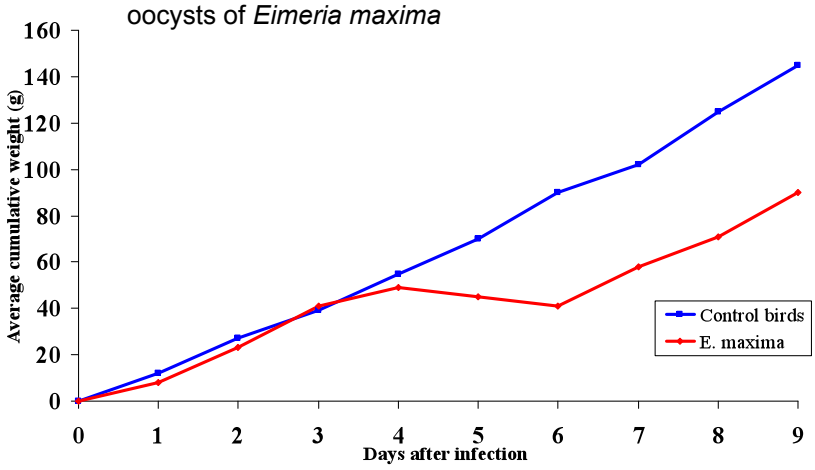
Average weight gain of chicks artificially infected with sporulated oocysts of *Eimeria maxima*



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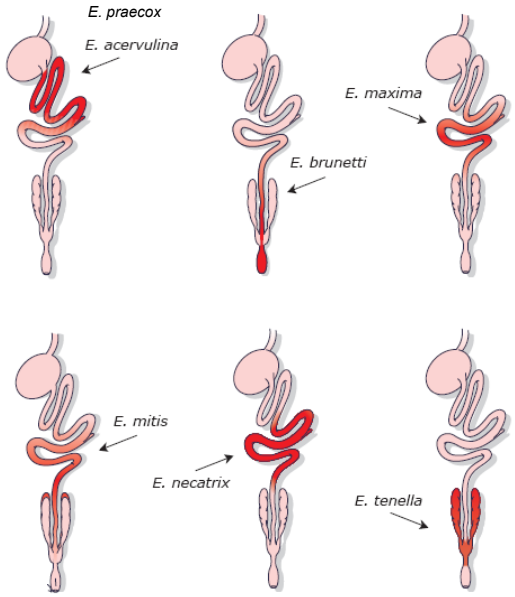
Eimeria species: location of lesions



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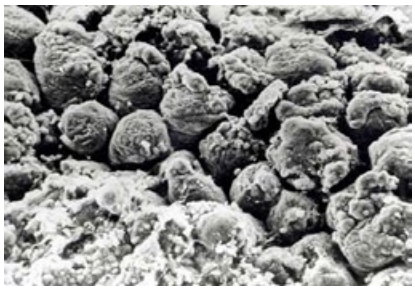
Intestinal damage: subclinical coccidiosis

Ileum, **uninfected** turkey poult, 3 weeks old (650X)



Ileum, turkey poult with **subclinical** coccidiosis, 3 weeks old (650X)

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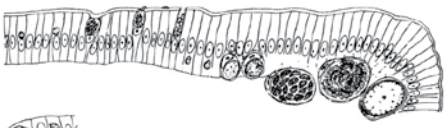
Locations of *Eimeria spp*. in the intestinal mucosa

Eimeria acervulina

Eimeria mitis

Eimeria maxima

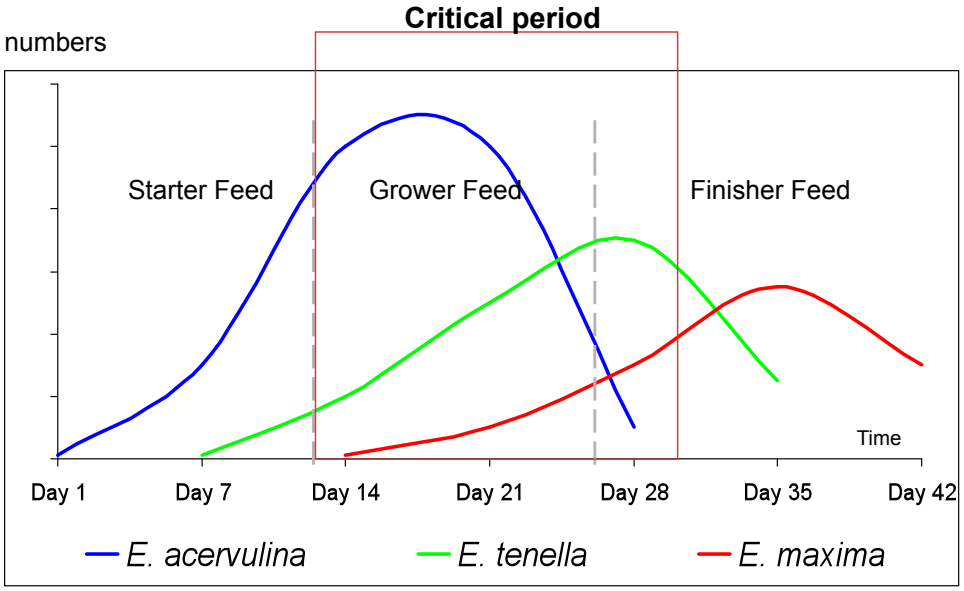


Eimeria necatrix
Eimeria tenella

Intestinal damage

- *E. acervulina*
  - Lesions by schizonts, gametocystes and developing oocysts
- *E. maxima:*
  - Minimal damage first 2 asexual stages
  - Lesions by sexual stages (5-8 days PI)
- *E. tenella*
  - 2e generation schizont
  - Schizont develop deep in lamina propria
- *E. brunetti*
  - Asexual stages in upper small intestine
  - Sexual stages in lower small intestine and ceca
- *E. necatrix*
  - 1e and 2e generation schizonts large and deep in (sub)mucosa small intestine -> severe damage

Development of *Eimeria spp*. throughout broiler growth cycle



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Real life example gut health scoring

Farm: Jade Oplok House: 1, parcox Age: 5 weken	Coccidiosis			Bacterial Enteritis											vetworks Dr. Jan Willems
		Eimeria		Overall	Cranial				Caudal				Overall		Date:
Chick	*acervulina*	*maxima*	*tenella*	Gut ballooning	Inflammation/ blood vessels dilated	Fluid	Abnormal Contents	Thickness/ Translucency/ Fragility	Inflammation/ blood vessels dilated	Fluid	Abnormal Contents	Thickness/ Translucency/ Fragility	Undigested feed		BES
1	0	1	0		0	1	1	1	0	1	1	1	1		3,2
2	0	2	0		1	0	0	0	1	0	0	0	1		1,2
3	0	1	1		0	1	0	0	0	1	0	0	1		1,2
4	1	0	1		0	1	0	1	0	1	0	0	1		2
5	0	1	0		0	0	0	0	0	0	0	0	0		0
6															0
MLS	0,2	1	0,4	Previous anticoccidial and growth promotor program (unit:	Vaccinated										
TMLS	1,6	TMBES	1,266667	Current anticoccidial and growth promotor program :	Vaccinated										

Farm: Jade oplok House: 1, Evalon Age: 5 Weken	Coccidiosis			Bacterial Enteritis											vetworks Dr. Jan Willems
		Eimeria		Overall	Cranial				Caudal				Overall		Date:
Chick	*acervulina*	*maxima*	*tenella*	Gut ballooning	Inflammation/ blood vessels dilated	Fluid	Abnormal Contents	Thickness/ Translucency/ Fragility	Inflammation/ blood vessels dilated	Fluid	Abnormal Contents	Thickness/ Translucency/ Fragility	Undigested feed		BES
1	1	4	0		1	0	1	0	1	0	1	0	0		1,6
2	0	1	0		1	0	0	0	1	0	0	0	0		0,8
3	0	0	0		1	1	1	1	0	1	1	1	1		1,6
4	0	0	2		0	1	1	1	1	1	1	1	1		1,6
5	0	1	0		1	1	1	1	0	1	1	1	1		1,6
6															0
MLS	0,2	1,2	0,4	Previous anticoccidial and growth promotor program (unit:	Vaccinated										
TMLS	1,8	TMBES	2,2	Current anticoccidial and growth promotor program :	Vaccinated										

BES: "De Gussem, M. (2010). Macroscopic scoring system for bacterial enteritis in broiler chickens and turkeys. In WVPA Meeting 01/04/. Merelbeke, Belgium."

coccidiosis J&R

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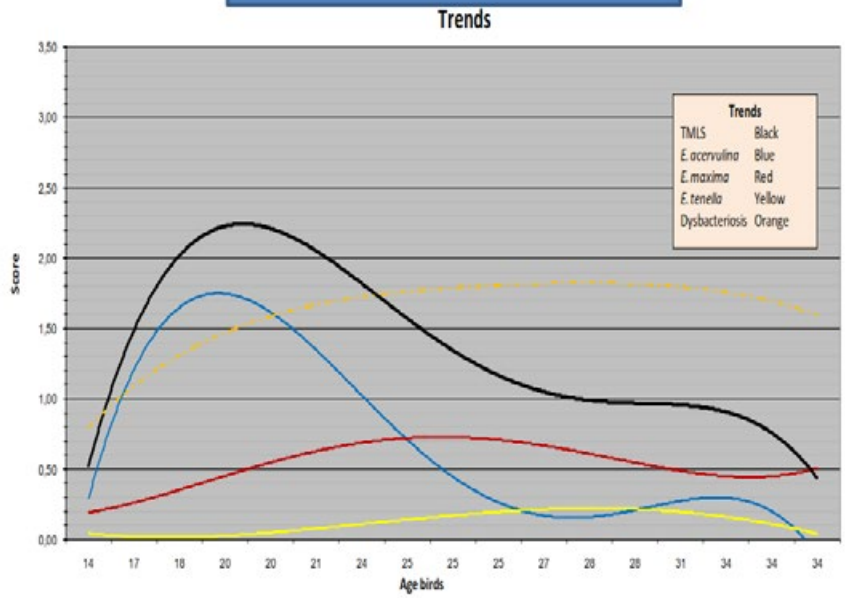
Real life example gut health session



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Consequences of coccidiosis

Clinical disease

- Mortality
- Blood in faeces
- Ruffled feathers
- Loss of appetite
- Poor performance
- Diarrhea

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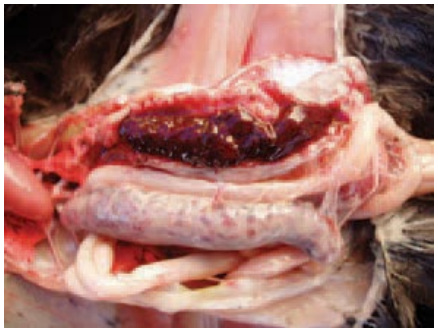
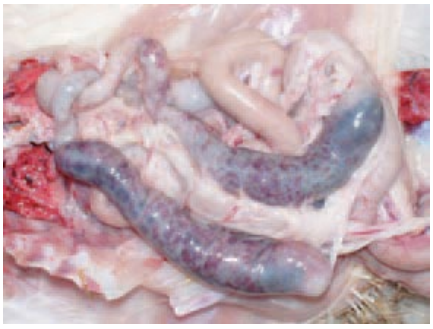
Intestinal damage: *E. tenella*

vetworks





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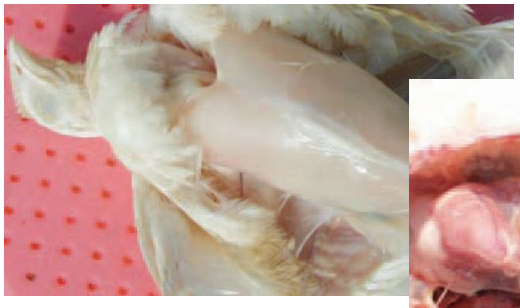
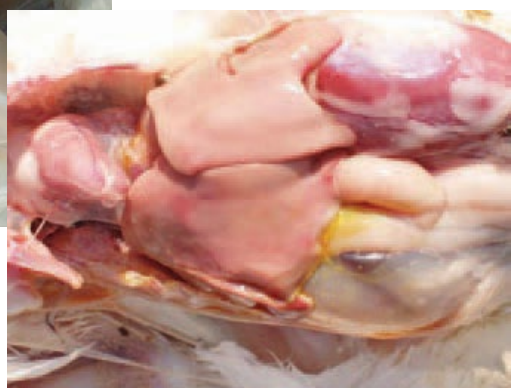
General anemia: *E. tenella*

vetworks





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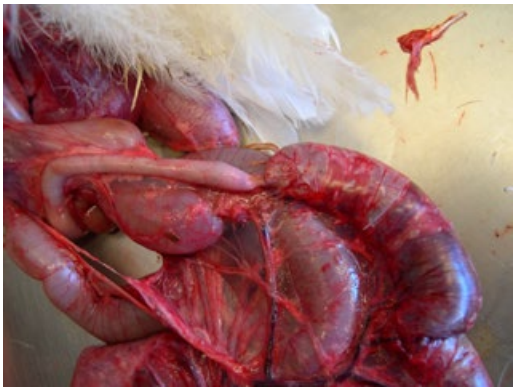
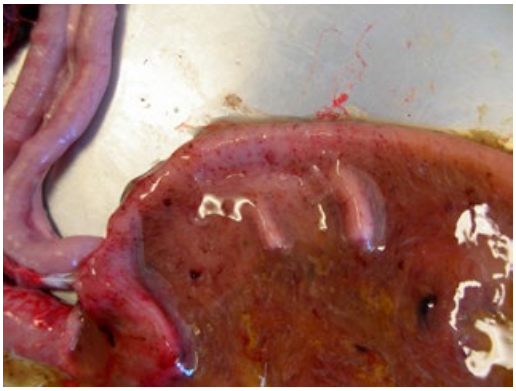
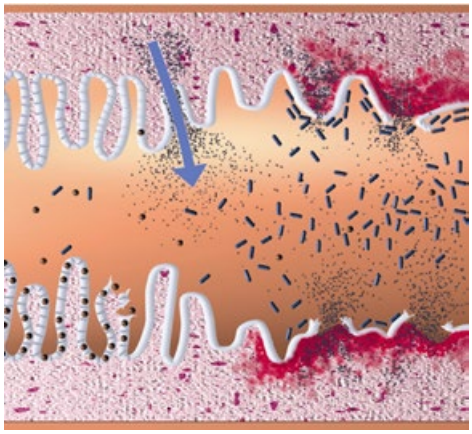
Intestinal damage: *E. necatrix*





Consequences of coccidiosis



Subclinical disease

Damage epithelial cells:

- Reduction nutrient absorption
- Poor performance: weight , FCR
- Poor uniformity
- Predisposing sec. diseases e.g. ***C. perfringens*** (NE/BE)

Effects of coccidiosis on performance:

- Increase mortallity: up to 40 %
- Decrease weight gain: up to 10 %
- Increase feed conversion: up to 10 %

Economical importance

vetworks

Waldenstedt *et al*, 2005: costs associated with coccidiosis in Swedish broiler production

=> **+/- 70 %** due to subclinical symptoms

Category		Cost (€)	%
Prevention		790 053	30.0
Therapy		0	0
Clinical symptoms	Mortality	4 255	0.2
	Condemnations	54 067	2.0
Subclinical symptoms	Reduced weight gain	1 118 834	42.5
	Increased FCR	666 814	25.3
Total		2 634 023	100

+/- 70 %

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Economical importance

vetworks

- Improving economical results by controlling and reducing subclinical coccidiosis
- Improvements that seem small may have a huge impact on integration level
- Eg:
  - 1 mio broilers/month grown up to 2.3 kg
  - +/- 4000 tonnes feed needed per month
  - Suppose FCR reduces from 1,72 to 1,70 by improved coccidiosis control = 48 tonnes feed saved
  - At 300 euro/tonne: savings13 000 € / month

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Economical importance

vetworks

Clinical signs = visible part of a (much bigger) problem!

Clinical coccidiosis: mortality, blood in feces,…

Subclinical coccidiosis: reduction of weight gain, decrease of performance, increase of FCR, …

Special features of coccidiosis in broilers

vetworks

- Big stocking density
- Broilers are slaughtered at the age, when the infection just starts to be controlled by immunity
  - Accumulation of infection throughout the cycles in the house
- Why different Eimeria spp. prevail at different ages?
  - Different length of the lifecycle
  - Different ability to multiply
  - Interaction between species: some species can manifest themselves after the other species is pushed to background by immunity
    - E.g. *E. acervulina* versus *E. maxima*
    - Immunity is species dependant and (partialy) strain dependant!

Coccidiosis in pullets, layers and breeders

- First 6 weeks similar to coccidiosis in broilers
  - *E. acervulina*, *E. maxima*, *E. tenella*, *E. praecox* and/or *E. mitis*
- Differences from broilers
  - In general, much better management
  - Lower stocking density
    - Lower spread of infection
  - Impact of coccidiosis is more difficult to evaluate
  - Clinical outbreaks are very devastating, loss of expensive birds
- Coccidiosis in breeders – the main reason of bad flock uniformity (weight)

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Conclusions

- Intestine: very big surface, very important for feed absorption and Immunity
- Clinical coccidiosis: mortality, blood in faeces, …
- Subclinical coccidiosis: link with bacterial enteritis
- Negative consequences
  - Weight gain
  - FCR
  - Predisposing factor for BE/ NE
  - Mortality
- Very resistant parasite in the environment
  - Destruction – is not a solution !!!
  - Prophylaxis is needed

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7 EFOMV
Encontro de Formação da Ordem dos Médicos Veterinários
26 e 27 de Novembro de 2016
Ponto de Encontro da Medicina Veterinária

vetworks

Prevention of poultry coccidiosis, rotation concept

Agrihealth, september 2016

Overview

vetworks

- Coccidiosis control tools
- Rotation concept
- Conclusions

Coccidiosis control tools

- Alternative products (phytoproducts)
- Therapeutics: e.g. sulfonamides, toltrazuril, amprolium
- Vaccines
- Anticoccidials
  - Chemical products (synthetically produced)
  - Ionophores (produced by fermentation)

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Coccidiosis control tools

- Alternative products (phytoproducts)
  - Probiotics, herb extracts (immunomodulation, antioxidant activities)
  - Direct action parasites? Might act on general gut health.
  - Numerous *in vitro and in vivo* (floor pen) studies but no stable results
  - Not yet proven under high infection pressure in field conditions
  - Need for more data to support claims (trials)
- Therapeutics (sulfonamides, toltrazuril, amprolium)
  - Usually used to control outbreaks
  - Limited spectrum of actives, dangerous as a prevention tool (if resistance, then no control tool is left)

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Vaccines

- Alternative option for coccidiosis control
- Mode of action – replacement of field strains by vaccine strains sensitive to anticoccidials
- EU, sold mainly in slow growing broilers, organic farming, breeders, layers and standard broilers with high infection
  - Broilers – expensive, problems with bacterial and necrotic enteritis
  - Breeders – very reliable, market leader for this segments
- Problem solver for many farms – but need blanc feed
- Have 'green' image compared to anticoccidials
- No cross-immunity, limited species immunity!
- Price difference with anticoccidials

(production in SPF birds)

Coccidiosis vaccines

- LIFE VACCINES
  - Non attenuated (vaccines with selected field strains)
    - Less expensive
    - Very good immunity
    - Easily provoke Necrotic Enteritis
  - Attenuated (usually «precocious» strains, with shorter reproduction cycle)
    - More expensive
    - Less intestinal damage (less virulent)
    - Less risk of Bacterial and Necrotic Enteritis
    - Immunity as in normal infections, but controlled epidemiology
    - Low incidence of coccidiosis outbreaks
    - Improve flock uniformity
- Administration
  - Day 1-7, in water, spray on feed, hatchery spray, eye drop (best results)
  - In ovo

Coccidiosis vaccines

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Commercial Name	Company	Country	Target Species	Attenuation	Composition
			LIVE VACCINES		
Coccivac B	SPAH	USA	Broilers	Non-attenuated	Eac, Emax, Emit, Eten
Coccivac D	SPAH	USA	Breeders	Non-attenuated	Eac, Ebru, Eh, Emax, Eprae, Eten
Coccivac T	SPAH	USA	Poults	Non-attenuated	Eaden, Emeleag, Egallop, Edispersa
Paracox 5	SPAH	UK	Broilers	Attenuated	Eac, Emax (x2), Eten, Emit
Paracox	SPAH	UK	Breeders	Attenuated	Eac, Emax (x2), Eten, Emit, Enec, Eprae, Ebru
Immucox I	Vetech	Canada	Broilers	Non-attenuated	Eac, Emax, Enec, Eten
Immucox II	Vetech	Canada	Breeders	Non-attenuated	Eac, Emax, Enec, Eten, Ebru
Livacox T	Biopharm	Czech republic	Broilers	Attenuated	Eac, Emax, Eten
Livacox Q	Biopharm	Czech republic	Breeders	Attenuated	Eac, Emax, Eten, Ebru
Advent	Viridus Animal Health	USA	Broilers	Non-attenuated	Eac, Emax, Eten
VAC M	Elanco	USA	Broilers	Non-attenuated	Emax
Nobilis Cox ATM	Intervet		Broilers	Non-attenuated	Eac, Emax (x2), Eten
Eimeriavax 4m	Bioproperties	Australia	Breeders & layers	Attenuated	Eac (RA), Emax (MCK_{+10}), Eten (Rt_{3+15}), Enec ($mednec_{3+8}$)
Huveguard start **Huveguard plus**	Huvepharma		broilers Breeders & layers	Attenuated Attenuated	Eac, Emax,Eten, Emit Enec, Ebru
Bio-Coccivet R	Biovet	Brazil	Breeders	Attenuated	Eac, Ebru, Emax, Enec, Eprae, Eten, Emit
Inmuner Gel-Coc	Inmuner	Argentina	Broilers		Eac, Ebru, Emax, Eten
Hipracox	Hipra	Spain	Broilers	Attenuated	Eac, Eten, Emax, Emit, Eprae
Evalon+ **Hipramune T**	Hipra	Spain	Breeders and Layers	Attenuated	Eac, Emax, Eten, Ebru, Enec




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Vaccine against coccidiosis

vetworks

- WHAT ABOUT EXTENDED USE (more than 3 production cycles) AND FIELD STRAINS?
  - On the long term possible escape of immune system of field strain (antigenic drift)
  - Breeders: complaints of outbreaks when always vaccinating with same vaccine
    - Check the application, rotation between vaccines, use of anticoccidials
- LIVE VACCINES: PROBLEMS DYSBACTERIOSIS/N.E.
  - Stimulate bacterial/necrotic enteritis, because of direct damage to enterocytes
  - Anticoccidials: ionophores reduce dysbacteriosis, chemicals are neutral against dysbacteriosis (except robenidine)
  - Using additives that improve the intestinal microbial ecosystem and development of the intestinal epithelium can help

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Why vaccine “does not work”

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- Wrong application
  - Change time of vaccination (day 1 in hatchery to day 7-8 in the house)
  - Check vaccination procedure
- Poor cross-protection to field strains
- Impared bird immunity
  - Mycotoxins, viruses, stress (onset of lay, transfer to production site)
  - Improve by using vitamins, ß-glucans others…..to improve general gut health
- High infection pressure
- Insufficient humidity of litter (sporulation delay, some slow multiplying species (*E. brunetti , E. necatrix*) may be pushed away by fast multiplying coccidia (*E. acervulina, E. maxima*)
  - RH should not be lower than 40% for first three weeks p.v.
- Anticoccidial in feed by accident

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Vaccination in broilers

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- Economically less efficient for most integrations where anticoccidials have good efficacy
- Interesting tool
  - Often as an additional to anticoccidials tool in problem farms (presence of multi resistant strains)
  - Rotation tool (we will give examples how to make sound programs later)
- Green image!

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Coccidiosis control tools

vetworks

- Alternative products (phytoproducts)
- Therapeutics: e.g. sulfonamides, toltrazuril, amprolium
- Vaccines
- Anticoccidials
  - According to Chapman, 2005
    - **Anticoccidials in feed : 95% broilers**
    - **Vaccination and others : 5%**

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Anticoccidials

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- Used for different animal species
  - Vaccines – only for certain species(no vaccines for turkeys)
- Work against all *Eimeria* species
  - Vaccine – not always contains all the species
- The favorable side effect of ionophores
- BUT : Avoid reduction of the sensitivity!
- No new actives development!!

→ Rotation

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Anticoccidials : classification

- Synthetic = 'Chemicals':
  - robenidine (Cycostat®) / diclazuril (Clinacox®) / nicarbazin (+narasin) (Maxiban®) / decoquinate (Deccox®) / (halofuginon)
- Polyether ionophore= 'Ionophores':
  - Monovalent:
    - Salinomycin (Salinomax®/Biocox®, , Sacox®)
    - Monensin (Elancoban®, Coxidin®)
    - Narasin (Monteban®, Maxiban®)
  - Monovalent glycoside:
    - Maduramicin (Cygro®)
    - Semduramicin (Aviax®)
  - Divalent:
    - Lasalocid (Avatec®)

Anticoccidials authorised in EU: Broilers

Brand Name	Compound	Company	Dose (ppm)	WT (d)
Avatec	Lasalocid sodium	Zoetis	75-125	3
Robenz	Robenidine HCl	Zoetis	30-36	5
Cygro	Maduramicin ammonium	Zoetis	5-6	3
Deccox	Decoquinate	Zoetis	30-40	0
Salinomax	Salinomycin sodium	Zoetis	50-70	1
Elancoban	Monensin sodium	Elanco	100-125	1
Maxiban	Narasin/nicarbazin	Elanco	80-100	0
Koffogran	Nicarbazin	Phibro	125	1
Kokcisan	Salinomycin sodium	KRKA	60-70	3
Monteban	Narasin	Elanco	60-70	0
Sacox	Salinomycin sodium	Huvepharma	60-70	1
Clinacox	Diclazuril	Huvepharma	1	0
Aviax	Semduramicin	Phibro	20-25	5
Stenorol	Halofuginone	Huvepharma	2-3	5
Coxidin	Monensin sodium	Huvepharma	100-125	1

Anticoccidials authorised in EU: Turkey

Brand Name	Compound	Company	Dose (ppm)	WT (d)	Max age weeks
Avatec	Lasalocid sodium	Zoetis	75-125	5	16
Robenz	Robenidine HCl	Zoetis	30-36	5	
Cygro	Maduramicin ammonium	Zoetis	5	3	16
Elancoban	Monensin sodium	Elanco	100-125	1	16
Clinacox	Diclazuril	Huvepharma	1	0	12
Stenorol	Halofuginone	Huvepharma	2-3	5	12
Coxidin	Monensin sodium	Huvepharma	100-125	1	16

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Anticoccidials authorised in EU: Layers

Brand Name	Compound	Company	Dose (ppm)	WT (d)	Max age weeks
Avatec	Lasalocid sodium	Zoetis	75-125	3	16
Elancoban	Monensin sodium	Elanco	100-125	1	16
Sacox	Salinomycin sodium	Huvepharma	60-70	1	12
Clinacox	Diclazuril	Huvepharma	1	0	16
Coxidin	Monensin sodium	Huvepharma	100-125	1	16

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Anticoccidials

- Anticoccidial tools
- Dangers: resistance & cross-resistance
- What to do: rotation

Anticoccidials: What are the issues ?

- **Reduced sensitivity/resistance**

After some time of use

↓

the efficacy of anticoccidials decreases

- **Cross-resistance**

If resistance to one product arises

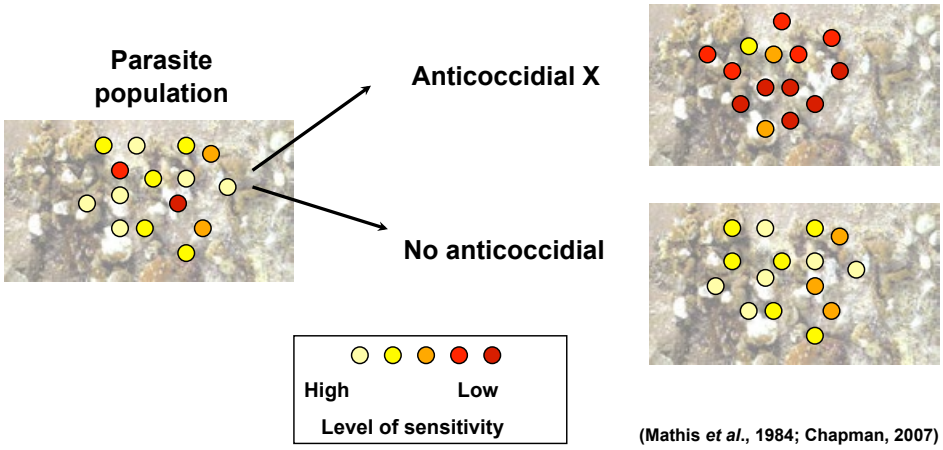
↓

other similar products will also work less efficient

(Mathis *et al.*, 1984; Chapman, 2007)

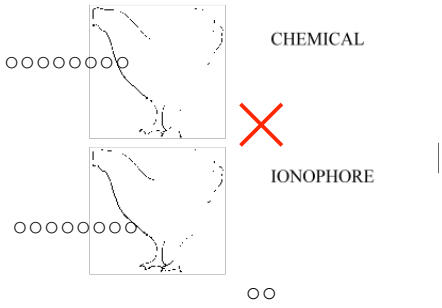
Anticoccidials: What are the issues ?

- **Reduced sensitivity/resistance**
- **After some time of use the efficacy of anticoccidials decreases**



(Mathis *et al.*, 1984; Chapman, 2007)

Reduced sensitivity/resistance

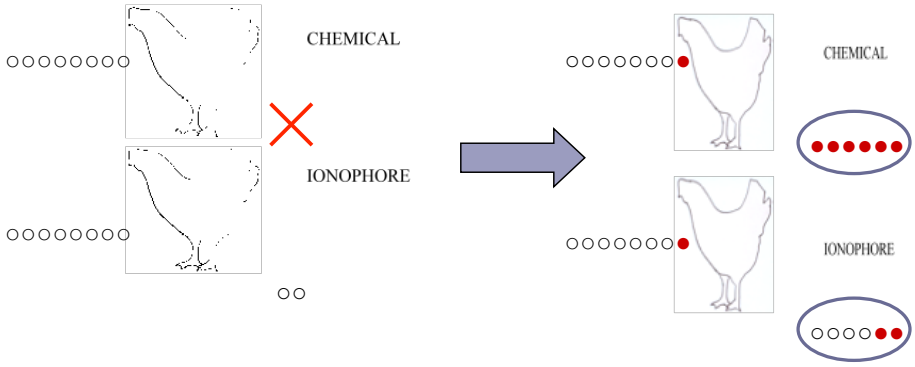


coccidial leakage

Danforth *et al.*, 1977, Poultry Science 56:926-932

Chapman and Johnson, 1992, Poultry Science 71:1342-1347

Reduced sensitivity/resistance: coccidial leakage

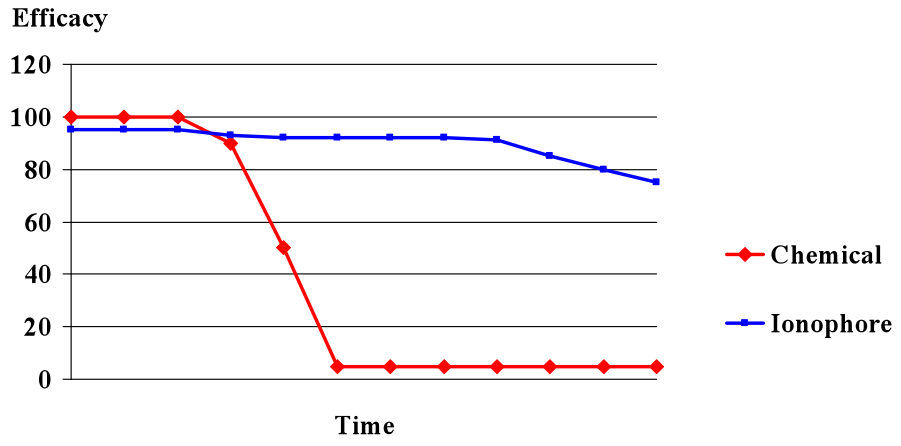


coccidial leakage

Danforth *et al.*, 1977, Poultry Science 56:926-932

Chapman and Johnson, 1992, Poultry Science 71:1342-1347

Timing of resistance development (theoretic timeline)



Anticoccidials: What are the issues ?

vetworks

Reduced sensitivity/resistance

After some time of use

↓

the efficacy of anticoccidials decreases

Cross-resistance

If resistance to one product arises

↓

other similar products will also work less efficient

(Mathis *et al.*, 1984; Chapman, 2007)

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Cross-resistance

vetworks

- **Chemicals**:
  - All products have different mode of action for parasite destruction
    - Diclazuril: disruption oocyst wall formation
    - Nicarbazine: inhibition oxidative phosphorylation
    - Robenidine: inhibition nucleus division schizont

→ cross-resistance does not appear

- **Ionophores**:
  - Have similar mode of action
  - Cross-resistance may become a problem
  - But: big difference between ionophores of **different classes**

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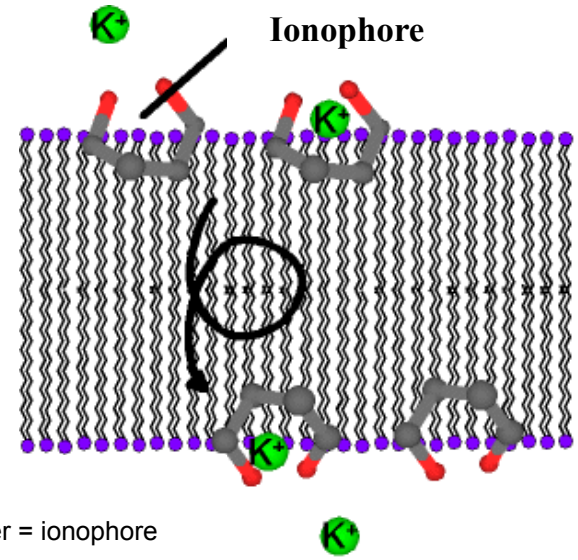
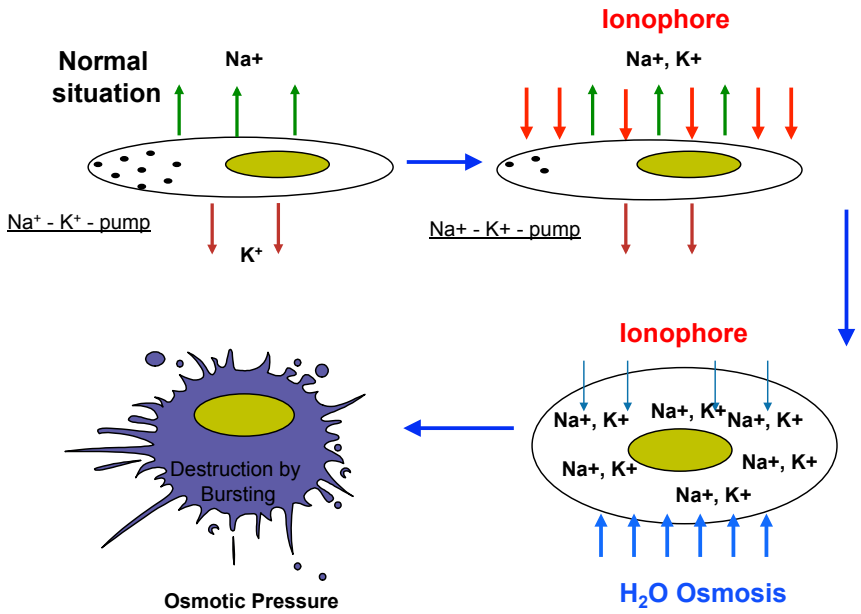
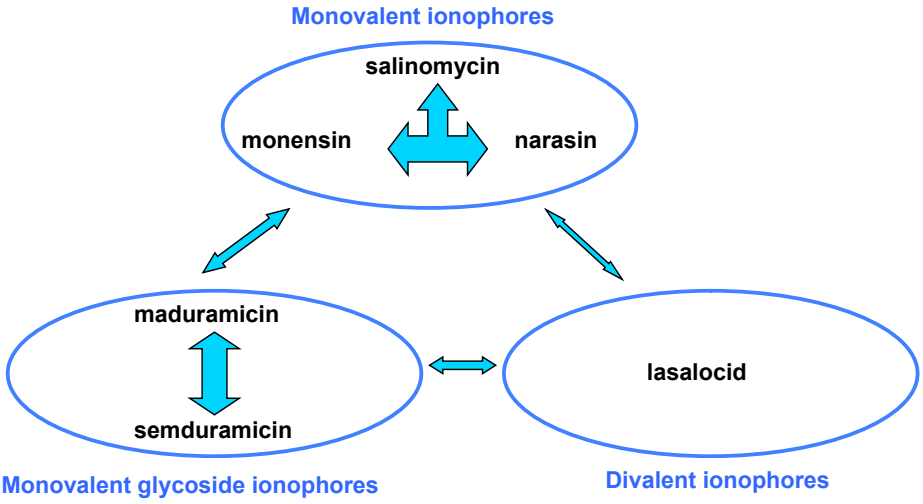
Ionophore mode of action



Ionophore mode of action



Potential for cross-resistance amongst ionophores



(Weppelman et al., 1977; Bedrnik 1983, 1989; Hamet 1986; Mc Dougald 1981; Chapman 1994)

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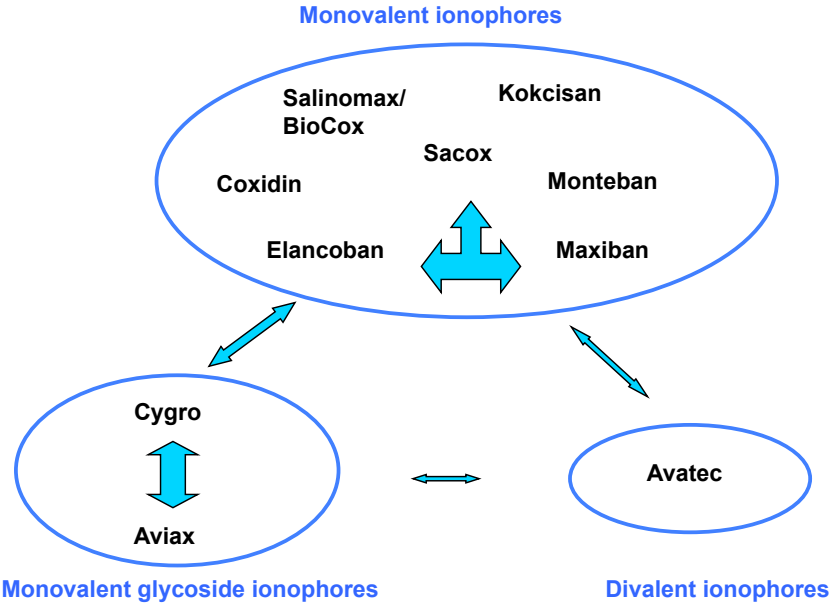
Potential for cross-resistance amongst ionophores



61

Cross-resistance between ionophores

vetworks

- **Cross-resistance** between monovalent ionophores: narasin, monensin and salinomycin **very often seen** in field conditions
  - Example: resistance against **narasin** observed prior to its use due to cross-resistance with **monensin** and **salinomycine**! (Weppelman et al., 1977)
- Coccidia strains resistant to monovalent ionophores usually sensitive to lasalocid (= divalent) (Weppelman et al., 1977; Bedrnik 1983, 1989; Hamet 1986; Mc Dougald 1981; Chapman 1994)

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Anticoccidials

vetworks

- Anticoccidial tools
- Dangers: resistance & cross-resistance
- What to do: rotation

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How to solve these problems?

vetworks

ROTATION PROGRAM

What ?

Rotation from one class of anticoccidials to another class after few cycles

(can be in **full** program or **shuttle**)

Why ?

- Makes it possible to restore the efficacy of anticoccidial
- Limits the development of resistance

64

Why to rotate? Reason 1

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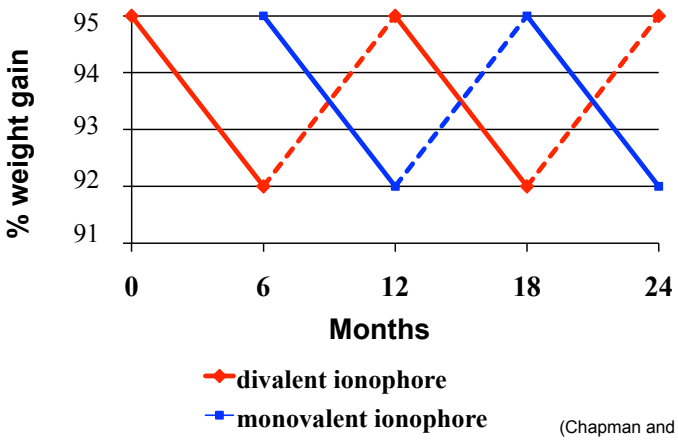

(Chapman and McFarland, 2003)

Rotation (= break) helps anticoccidials to restore their efficacy!!!

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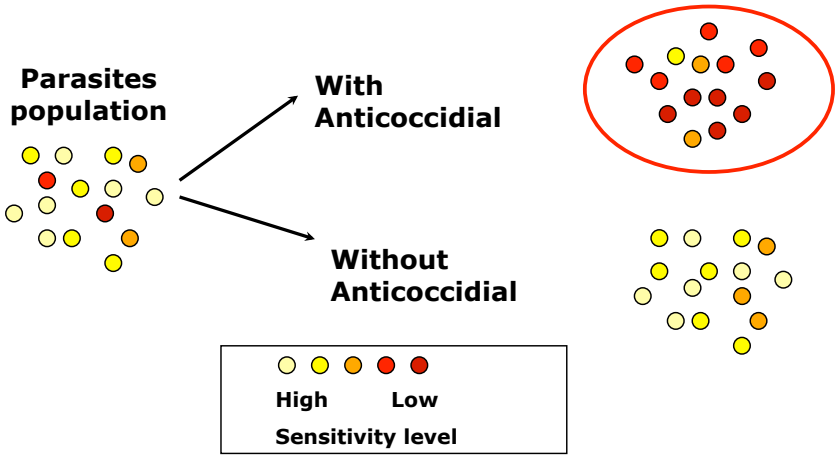
Reduced sensitivity / resistance

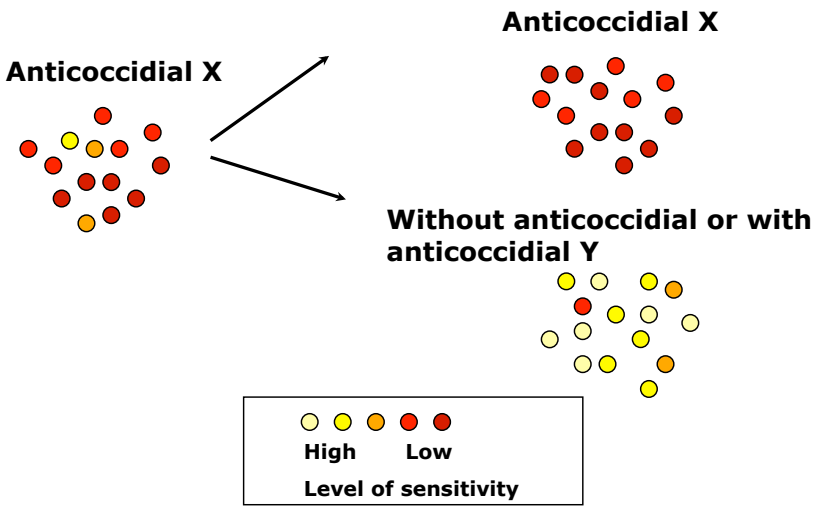
- After some use the efficacy of anticoccidial decreases



66

Break for coccidiostatics

- Break in some anticoccidial = reverse selection



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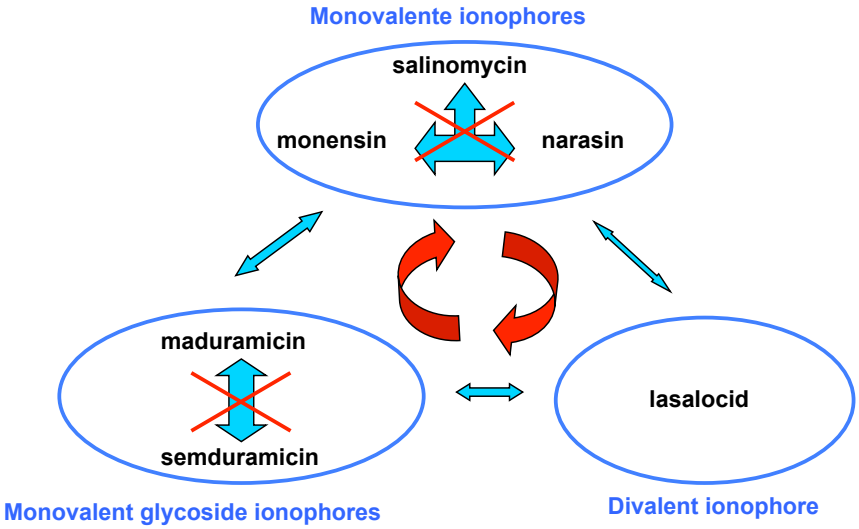
Correct rotation



Don't forget cross-resistance

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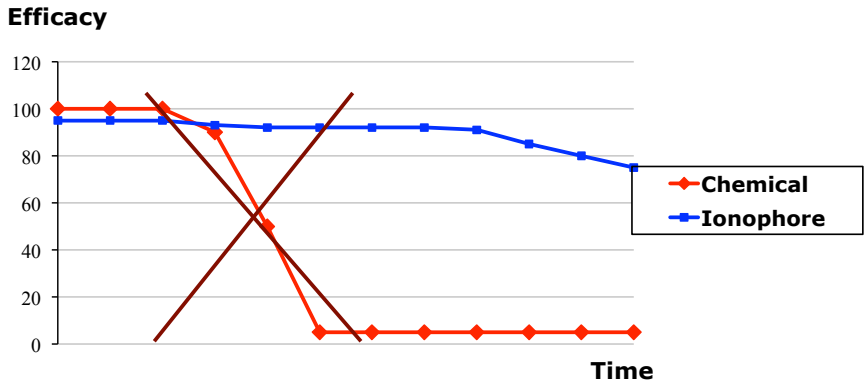
Why to rotate? Reason 2



Rotation reduces the risk of resistance!!!

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Rotation - conclusions

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Giving rest anticoccidials helps to restore their EFFICACY!

- This is the main concept of rotation
- For salinomycin, Chapman has shown that 25% restoration of efficacy occures after 5 cycles (10 month) of rest
- Restoration is slow, better not to bring to the point of loss of efficacy!
- Rotation helps to reduce the risk of resistance

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Coccidiosis control: Four basic rules!!!

- Do not use same anticoccidial for **long** period
- After using a product, allow **long enough** rest
- When rotating, rotate between **classes** and not just products
- Use a **chemical clean-up** once a year

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Optimal duration of program in broilers

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- Ionophore: up to 6 months
- Chemical full: 3 months (1 cycle)
- Chemical shuttle: 4.5 months (2 cycles)

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ANTICOCCIDIAL STRATEGIES

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- Chemicals: will inhibit excretion of oocysts
  - Very effective, significant reduction infection pressure level: clean up program
  - Mainly resistant parasites will survive
  - Need strict limit of control in time
- Ionophores: allow leakage of oocysts
  - Significant % of (sensitive) oocysts can escape action of drug
  - Less risk for resistance!
  - But after continuous use, levels of subclinical coccidiosis are increased
- Use both types in combination (shuttle, rotation) in order to maximize subclinical coccidiosis control in a safe way!

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Advised resting period

vetworks

- After the use of ionophore, do not use it (or some other ionophore of the same class) at least for 6 months
- Reduce the use of certain chemical till once a year
  - If it was used for 3 months in full program, make a break for 9 months

Conclusion

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Any of the product types (ionophores, chemicals, vaccines) have advantages and disadvantages, and no single one of them can be implemented on its own without losing performance on the long-term

BUT…

When implementing these strategies/products by rotating them, disadvantages are minimized and advantages are maximized.

Overview

vetworks

- **Anticoccidial strategies**
- Trial with coccidiosis vaccine
- Importance of chemical Clean–up
- Conclusions

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Coccidiosis control tools

- Vaccines
- Anticoccidials
  - Chemical products
  - Ionophores

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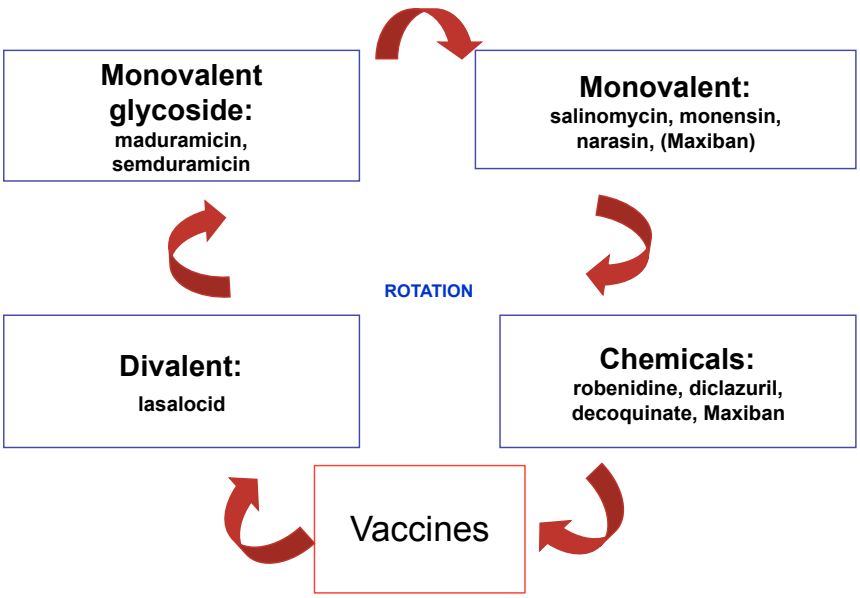
Rotation



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Overview

- Anticoccidial strategies
- **Trial with coccidiosis vaccine**
- Importance of chemical Clean–up
- Conclusions

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Overview

- Anticoccidial strategies
- Trial with coccidiosis vaccine
- **Importance of chemical Clean–up**
- Conclusions

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Clean-up, as part of strategic coccidiosis control program in broilers

Why to clean-up?

1. **For prevention of (sub)clinical coccidiosis**

2. **For reduction of the infection pressure**
Minimized shedding of oocysts in broilers

Important part of rotation program

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Clean-up

vetworks

reduce coccidiosis pressure by reduction of the oocysts shedding

○○○○○

○○

Ionophore:
Coccidia leakage

○○○○○

Chemical

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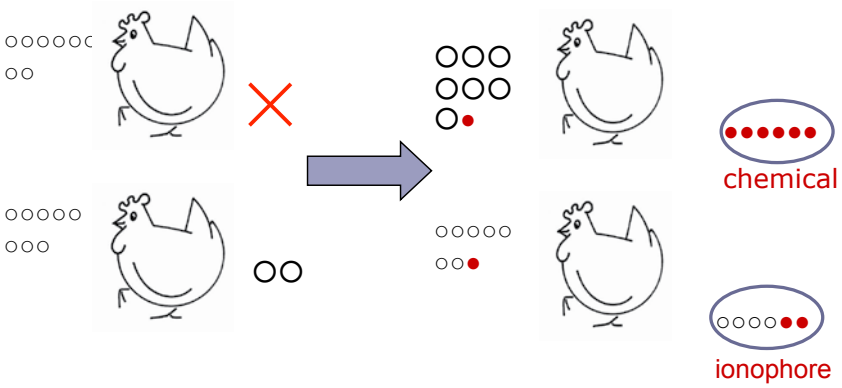
Clean-up

vetworks

leakage/resistance development



Danforth *et al*., 1977, Poultry Science 56:926-932
Chapman and Johnson, 1992, Poultry Science 71:1342-1347

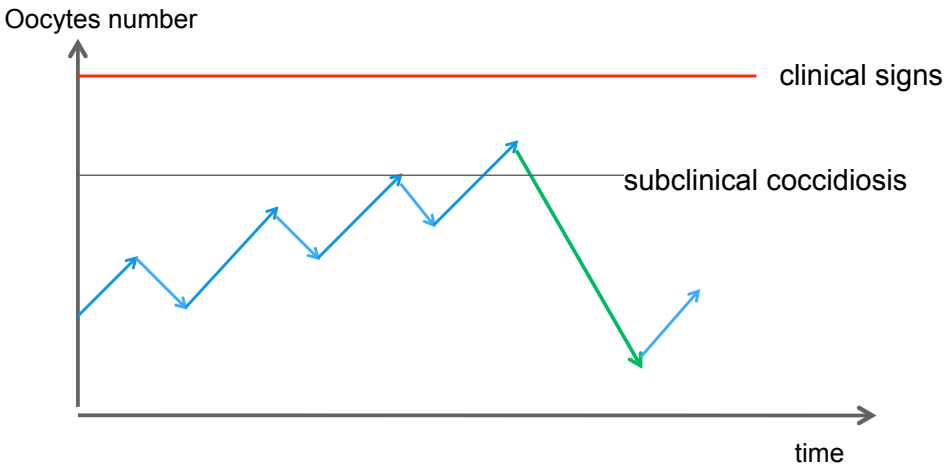
84

Clean-up

vetworks

Ionophore: slow development of resistant population, allow the development of subclinical coccidiosis

Chemical: reduces oocysts shedding, reduces coccidiosis pressure



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2 ways of clean-up

In FULL program	In Shuttle program
• At **house** level **1 full cycle** • At **feed mill** level for **3 months**	• At **house** level **2 full cycles** • At **feed mill** level for **17/18 weeks**

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Anticoccidial programs :

- Dual or Shuttle

- Straight or Full

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Clean-up

vetworks

1 **full** cycle in each house of integration

12 weeks to reach every single house in the integration

	WEEK	1	2	3	4	5	6	7	8	9	10	11	12	13
FARMS														
Group 1		S	S	G	G	G	W	E	E	S	S	G	G	G
Group 2		E	S	S	G	G	G	W	E	E	S	S	G	G
Group 3		E	E	S	S	G	G	G	W	E	E	S	S	G
Group 4		W	E	E	S	S	G	G	G	W	E	E	S	S
Group 5		G	W	E	E	S	S	G	G	G	W	E	E	S
Group 6		G	G	W	E	E	S	S	G	G	G	W	E	E
Group 7		G	G	G	W	E	E	S	S	G	G	G	W	E
Group 8		S	G	G	G	W	E	E	S	S	G	G	G	W

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Clean-up

vetworks

2 cycles **shuttle (starter feed)** in each house of integration

17 weeks to reach every single house in the integration

WEEK	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
FARMS																	
Group 1	S	S	G	G	G	W	E	E	S	S	G	G	G	W	E	E	S
Group 2	E	S	S	G	G	G	W	E	E	S	S	G	G	G	W	E	E
Group 3	E	E	S	S	G	G	G	W	E	E	S	S	G	G	G	W	E
Group 4	W	E	E	S	S	G	G	G	W	E	E	S	S	G	G	G	W
Group 5	G	W	E	E	S	S	G	G	G	W	E	E	S	S	G	G	G
Group 6	G	G	W	E	E	S	S	G	G	G	W	E	E	S	S	G	G
Group 7	G	G	G	W	E	E	S	S	G	G	G	W	E	E	S	S	G
Group 8	S	G	G	G	W	E	E	S	S	G	G	G	W	E	E	S	S
This is 2 shuttle cycles /year on a farm level																	

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Clean-up

vetworks

2 cycles **shuttle (grower feed)** in each house of integration

18 weeks to reach every single house in the integration

WEEK	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
FARMS																		
Group 1	G	G	G	W	E	E	S	S	G	G	G	W	E	E	S	S	G	G
Group 2	S	G	G	G	W	E	E	S	S	G	G	G	W	E	E	S	S	G
Group 3	S	S	G	G	G	W	E	E	S	S	G	G	G	W	E	E	S	S
Group 4	E	S	S	G	G	G	W	E	E	S	S	G	G	G	W	E	E	S
Group 5	E	E	S	S	G	G	G	W	E	E	S	S	G	G	G	W	E	E
Group 6	W	E	E	S	S	G	G	G	W	E	E	S	S	G	G	G	W	E
Group 7	G	W	E	E	S	S	G	G	G	W	E	E	S	S	G	G	G	W
Group 8	G	G	W	E	E	S	S	G	G	G	W	E	E	S	S	G	G	G
This is 2 shuttle cycles/year on a farm level																		

3 way chemical shuttle

vetworks

- Eg Deccox – Robenz – Clinacox
- Nicarb – Robenz – Clinacox
- (0 day withdrawal in the end, most expensive in the starter or the one with age toxicity (nicarb) in the starter....
- Max. 2 cycles
- - risk of “burning up” three chemicals
  - logistic effort for the feed mill
  + ideally before vaccinating

Comparison of cost of anticoccidial programs

Program	Dose, kg/ton	Period, days	Price 1000 birds, €
Deccox	0,5	0-35	25.9
Vaccine	-	-	40
Salinomycin	0,5	0-35	5.7
Maxiban/Salinomycin	0,5/0,5	0-28/29-35	13.4
Deccox/Robenz/Clinacox	0,5/0,5/0,2	0-18/19-32/33-42	21.4

Important: the average costs of the program should be calculated **for 2 years period**!

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Example of anticoccidial program

Year	2013					
Month	Jan	Feb	Mar	Apr	May	Jun
Active	Salinomax	Salinomax	Salinomax	Salinomax	Salinomax	Avatec/Clinacox
Class	mon. Ion.	mon. Ion.	mon. Ion.	mon. Ion.	mon. Ion.	Div. Ion/chem
Comments	*70 ppm*				*from 15 May*	*Avatec 0-24 days*
					Avatec/Clinacox	*Clinacox 25-37 days*
Year	2013					
Month	Jul	Aug	Sept	Oct	Nov	Dec
Active	Avatec/Clinacox	Avatec/Clinacox	Avatec/Clinacox	Avatec	Avatec	Deccox
Class	Div. Ion/chem	Div. Ion/chem	Div. Ion/chem	div. Ion	div. Ion	Chem
Comments						
Year	2014					
Month	Jan	Feb	Mar	Apr	May	Jun
Active	Deccox	Deccox	Robenz/Cygro	Robenz/Cygro	Robenz/Cygro	Robenz/Cygro
Class	Chem	Chem	chem/mon glyc	chem/mon glyc	chem/mon glyc	chem/mon glyc
Comments			*Robenz 0-21 days*			
			Cygro 22-35 days			
Year	2014		*Robenz 36 ppm*			
Month	Jul	Aug	Sept	Oct	Nov	Dec
Active	Robenz/Cygro	Cygro	Maxiban/salinomycin	Maxiban/salinomycin	Maxiban/salinomycin	Maxiban/salinomycin
Class	chem/mon glyc	chem/mon glyc	chem/mon ion	chem/mon ion	chem/mon ion	chem/mon ion
Comments	*from 15.06 Cygro alone*					*use till March*

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Example of anticoccidial program

vetworks

Year	2012					
Month	Jan	Feb	Mar	Apr	May	Jun
Active	Deccox/ Salinopharm	Deccox/ Salinopharm	Deccox/ Salinopharm	Deccox/ Salinopharm	Deccox/ Salinopharm	robenidin/lasalocid
Class	chemical /mon.ion.	chemical /mon.ion.	chemical /mon.ion.	chemical /mon.ion.	chemical /mon.ion.	chem./div. Ionophor
Comments						*Avatec 125 ppm*
Year	2012					
Month	Jul	Aug	Sept	Oct	Nov	Dec
Active	robenidin/lasalocid	robenidin/lasalocid	robenidin/lasalocid	robenidin/lasalocid	lasalocid	Clinacox/Cygro
Class	chem./div. Ionophor	chem./div. Ionophor	chem./div. Ionophor	chem./div. Ionophor	div. Ionophor	Chem./Mon. glyc.
Comments	*Avatec 125 ppm*	*Avatec 125 ppm*	*Avatec 125 ppm*	*Avatec 125 ppm*	*Avatec 125 ppm*	
Year	2013					
Month	Jan	Feb	Mar	Apr	May	Jun
Active	Clinacox/Cygro	Clinacox/Cygro	Clinacox/Cygro	Clinacox/Cygro	Cygro	Deccox/ Avatec
Class	Chem./Mon. glyc.	Chem./Mon. glyc.	Chem./Mon. glyc.	Chem./Mon. glyc.	Mon. glyc.	chemical /div.ion.
Comments				Stop Clinacox at mid April		
Year	2013					
Month	Jul	Aug	Sept	Oct	Nov	Dec
Active	Deccox/ Avatec	Deccox/ Avatec	Deccox/ Avatec	Deccox/ Avatec	Robenz/ Salinomax	Robenz/ Salinomax
Class	chemical /div.ion.	chemical /div.ion.	chemical /div.ion.	chemical /div.ion.	chem /mon. Ionophor	chem /mon. Ionophor
Comments						*use till February*

General Conclusion

vetworks

- **Subclinical coccidiosis** is very important!!! → big economic impact + risk of bacterial enteritis/ necrotic enteritis
- **Maintaining the effectiveness of anticoccidials** is very important to prevent other factors …
  - Use the products in optimal doses

    higher dose → less coccidiosis + less enteritis → better performance !!!
  - Perform rotation to keep the efficacy of the products
  - Use of chemical clean-up may improve performance parameters

vetworks

Thank you for your attention!

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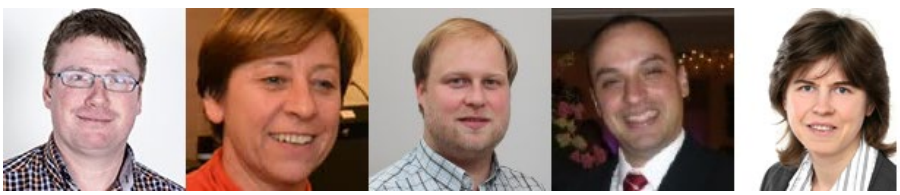
Jan Willems

Bacterial Enteritis in broilers is a key component compromising gut health

vetworks

- Dr. Jan Willems, DVM
  - Poultry veterinarian
  - Graduated as DVM in 2011 at University of Ghent, Belgium
  - 2011 – 2015: Poultry Veterinarian in The Netherlands, Germany, Belgium
  - 2015 – now : Vetworks
  - www.poultrytechnicaltraining.com www.vetworks.eu



Poultry production 2016

vetworks

- Global poultry industry is very competitive compared to other meat sources – expected to grow most aggressively
  - Healthy protein source
  - Very efficient Feed Conversion Ratio
  - Main grow market : Africa!
  - 2050 > 40% of births will be in Africa!
  - Who is going to provide meat?
    - Poultry industry
    - Aquaculture
    - red meats

3

Poultry production 2016

vetworks

- Cost of feed: ±70% of total cost
- Intestine = most important organ to convert feed to meat
- Gut health is key in financial return of investment (ROI)
  - Last ten years: increased understanding gut health
  - →pressure on classic AGP
  - - banned in EU but
  - - still best performance: FCR and weight gain!
  - Genetics are similar worldwide
  - Climate: climate control
  - Raw material: quality and prices: world market: no influence
  - Feed composition: additives for improving gut health (Additives: anticoccidials, AGP's, mycotoxin binders, enzymes, anti-oxidants, immunostimulants, gut development products)
  - Winner : right choice of additives (efficacy ánd ROI)

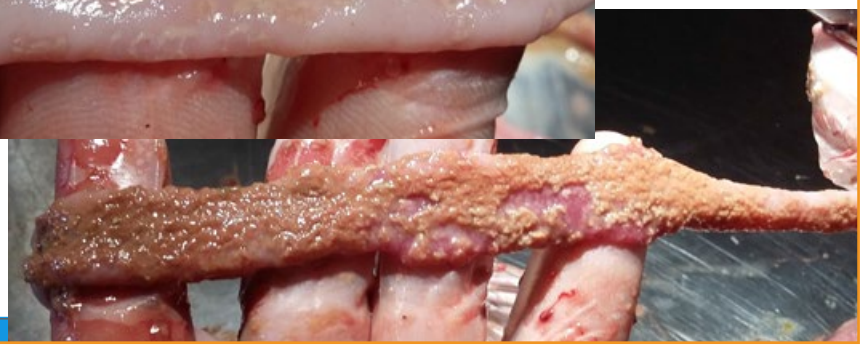
4

BE / NE

- Bacterial Enteritis / Necrotic Enteritis
  - Main indication for antibiotic treatments in Europe for the last 15 years - estimated 50% - 70%
  - Meat and bone meal & AGP free
- last 5 years drastic reduction of AB use!
  - Pressure from society
  - But mainly because we are starting to understand how to solve the problems that occurred when we switched to vegetarian & AGP free
- No BE models available until recently – increase of knowledge expanding rapidly last 5 years

BE / NE

- BE or NE?
  - Due to common predisposing factors
  - Due to similar AB with good response



Macroscopic signs of BE

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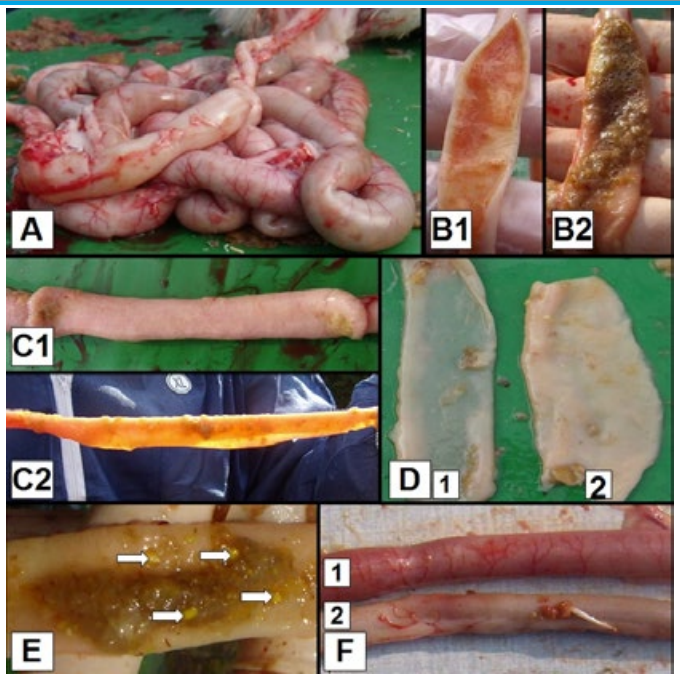

Intestinal morphology

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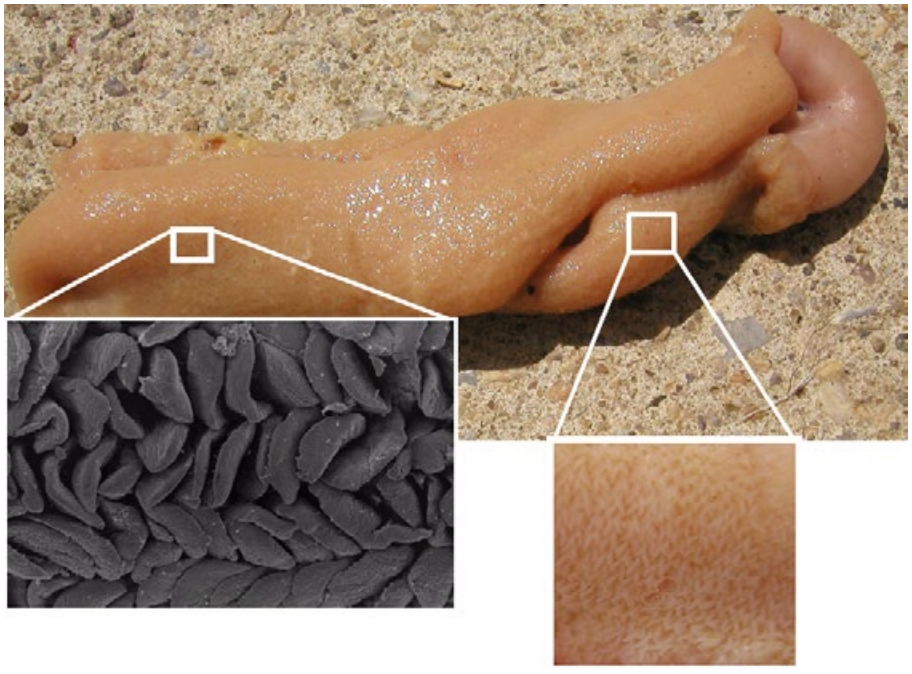

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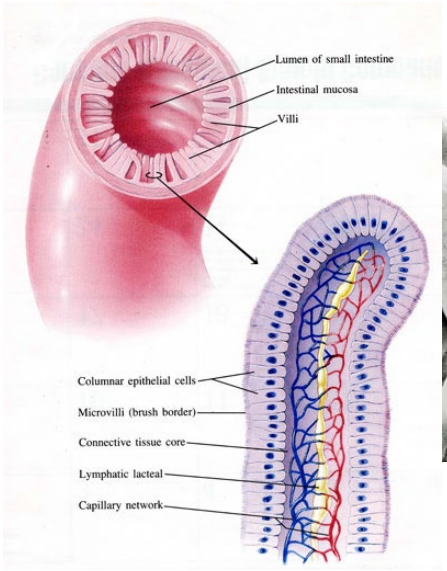
Intestinal morphology



9

Overview

- What is Bacterial Enteritis/Necrotic Enteritis?
  - Introduction to the disease
  - Contributing factors – link with coccidiosis
  - Consequences
- Conclusions

10

Paradox to solve in broilers

- Improved genetics over 50-60 years
- Selection for maximum growth rate = selection for maximum feed intake => increase of feed passage
- Even minor violations of the intestinal digestion and absorption capabilities increases number of nutrients in intestines, available to potentially harmful bacteria
- All factors that cause an initial damage/disbalance to the intestine under this high pressure may lead to

Bacterial Enteritis and/or Necrotic Enteritis

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Bacterial enteritis

- Main indication for antibiotic treatments in Europe for the last 10 years - estimated 50% - 70% of antibiotic (AB) use for BE
- Initially a lot of confusion with NE
  - Due to common predisposing factors
  - Due to similar AB with good response
- No drugs registered for BE poorly described condition, models not available until recently

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Bacterial enteritis

- Importance increased after ban on:
  - meat and bone meal (MBM): vegetarian diets
  - antimicrobial growth promotors (AGP)
- Much less acute problems in countries producing in presence of AGP but also in these countries BE is important, often goes unnoticed due to AGP use – losses maybe even higher?

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Bacterial Enteritis

- Very confusing terminology:
  - Dysbacteriosis
  - Bacterial enteritis
  - small intestinal bacterial overgrowth(SIBO)
  - subclinical NE
  - Dysbiosis
- Confusion with MAS, coccidiosis and NE
- No consensus on diagnostics:
  - no pathognomonic lesions
  - which of the typical lesion are relevant, what combination of lesions is typical

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Causes

- Multi-factorial:
  - Bacterial factor
  - Feed factors
  - Management: ventilation, litter quality
  - Gut Stressors:
    - Coccidiosis
    - Mycotoxins
    - Viral agents?

2016: main routes for impaired gut health?

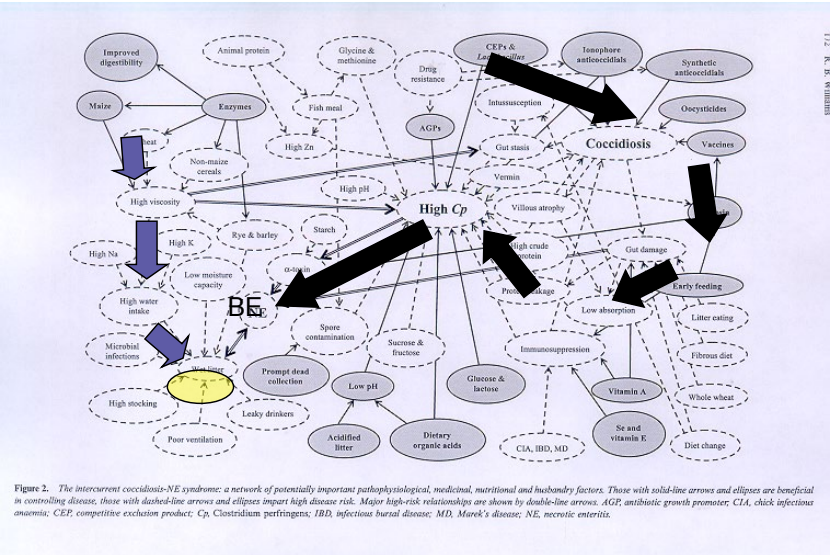


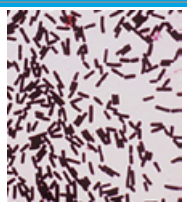
Figure 2. *The intercurrent coccidiosis-NE syndrome: a network of potentially important pathophysiological, medicinal, nutritional and husbandry factors. Those with solid-line arrows and ellipses are beneficial in controlling disease, those with dashed-line arrows and ellipses impart high disease risk. Major high-risk relationships are shown by double-line arrows. AGP, antibiotic growth promoter; CIA, chick infectious anaemia; CEP, competitive exclusion product; Cp, Clostridium perfringens; IBD, infectious bursal disease; MD, Marek's disease; NE, necrotic enteritis.*

Williams, 2001

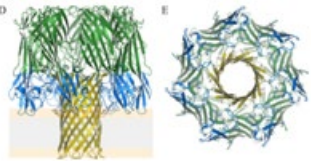
NE bacterial causes

vetworks

- Clostridium perfringens
- Gram positive, spore forming rod



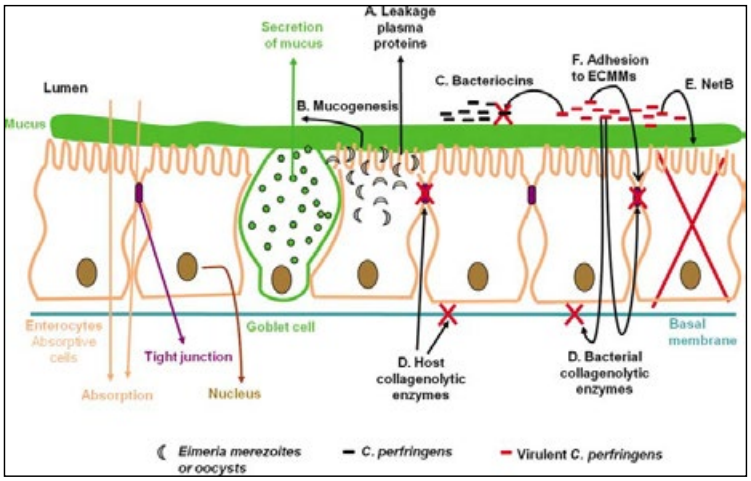
- NetB poreforming toxine
  - Only Cp strains with netB virulence factor can produce NE?



- A single clone of virulent Cp
  - Different strains of Cp are found in healthy birds
  - Produce bacteriocin to eliminate other Cp strains
  - Produce collagenolytic enzymes to destroy tight junctions
  - Better ability for adhesion to ECMM(extracellular matrix molecules)
    - ECMM are exposed if tissue damage (coccidiosis)

NE and coccidiosis

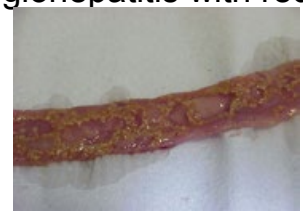
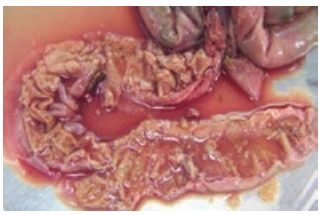
vetworks



L.Timbermont et al 2011

NE clinical symptoms – lesions

- Acute or chronic enterotoxaemia
  - Mortality +/- 10 % (5-50%)
  - Layers and breeders often when production almost at peak production
- Rather rare
- Acute clinical disease: prompt response to AM treatment
  - Pathognomonic lesions: small intestine necrosis, inflammation, haemorrhage
- Same contributory factors as BE
- Liver lesions: cholangiohepatitis with red or white foci

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NE predisposing factors

= Intestinal environment favorable to the growth of C. perfringens

- Nutrition
  - High NSP: rye, wheat, oats, barley versus maize diet
  - High protein: fish meal, poorly digestible protein
  - Animal fat versus vegetable oil
- Stress
  - Changes in feed regime (strater to grower)
  - Immuno suppression: viral diseases CAV, IBDV,Marek
  - Stocking density
- Coccidiosis
  - NE models use coinfection of cox and Cp

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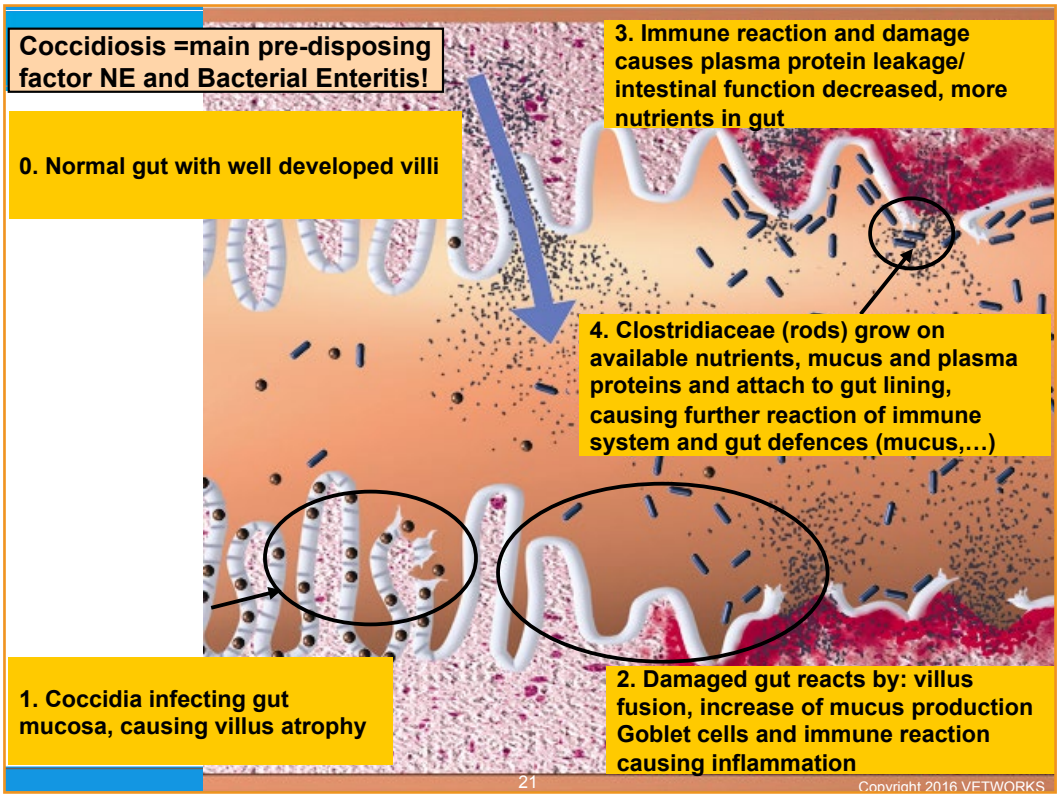

BE vs. NE

vetworks

- Some triggers or contributing factors with BE are similar as with NE (coccidiosis and feed quality)
- Antibiotics used for therapy are similar : amoxicillin, tylosin, bacitracin, lincomycin, penicillins….hence the confusion…. ?
- NE is not common, BE is omnipresent!
- Models used for evaluating solutions for NE are useful in BE!
  - Feed composition + trigger with cox

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BE versus NE

- But BE is not the same as NE:
  - Not one single bacterial species causing problems but different groups (Clostridiaceae) and probably more groups will be discovered in the future
  - No clonal multiplication of one species
  - More complicated interaction of microbiota and mucosa, not causing necrosis, but inflammation and morphological changes leading to favourable environment for further multiplication of involved bacterial groups
  - Never sure what AB/preventive tools will work best as different group are involved?

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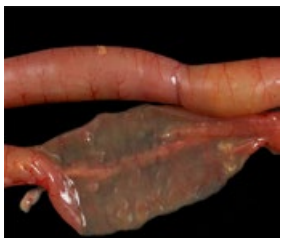
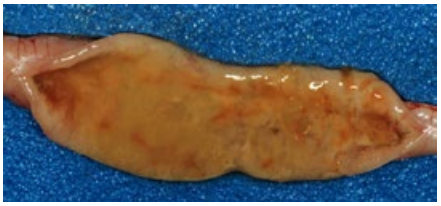
How to diagnose BE

- Typical Signs
  - Wet litter in house
  - 'Diarrhea' loose faecal droppings
  - Feed intake stable for a couple of days
  - Increased water/feed ratio



- Necropsy (not always all of these apply!!)
  - Gut strength: fragile, thin, transparent
  - Poor tonus of gut wall
  - Inflammation
  - Blood vessels dilated
  - Excessive fluids in small intestine
  - Ballooning of intestinal wall
  - Undigested feed in hindgut



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How to diagnose BE

- Histological signs
  - Normal lining and/or tight junctions are damaged
  - More villus fusion as natural reaction on breach of gut barrier
  - More inflammatory cells in the gut mucosa
    - Higher number of T-Lymphocytes and heterophils infiltrated
    - Try to stop invading bacteria
  - More mucus-producing Goblet cells
    - Try to replace the physical gut barrier
    - But good feeding source for Clostridiaceae
  - Decreased villus length
  - Decreased villus/crypt ratio
    - Larger crypt as turnaround of intestinal cells is increased and villi are shorter
- All together signs of poorer gut health.

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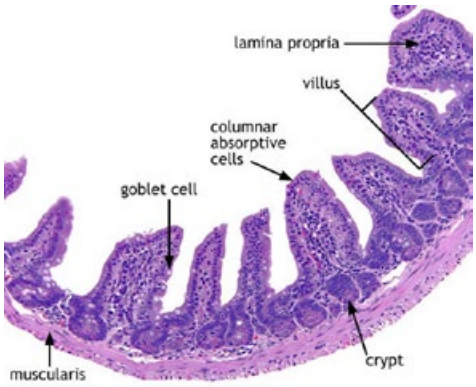
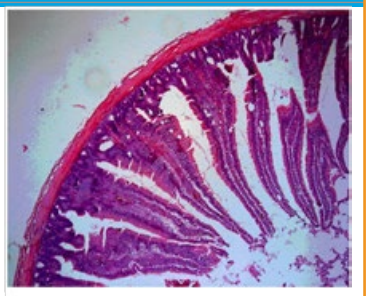
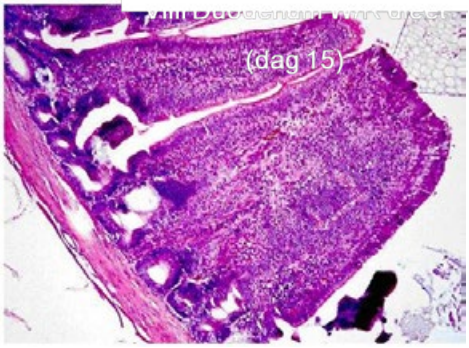
Histology







Inflammation:
- Gobletcells, heterophile infiltration

Villi: fusion,length
Crypt hyperplasia

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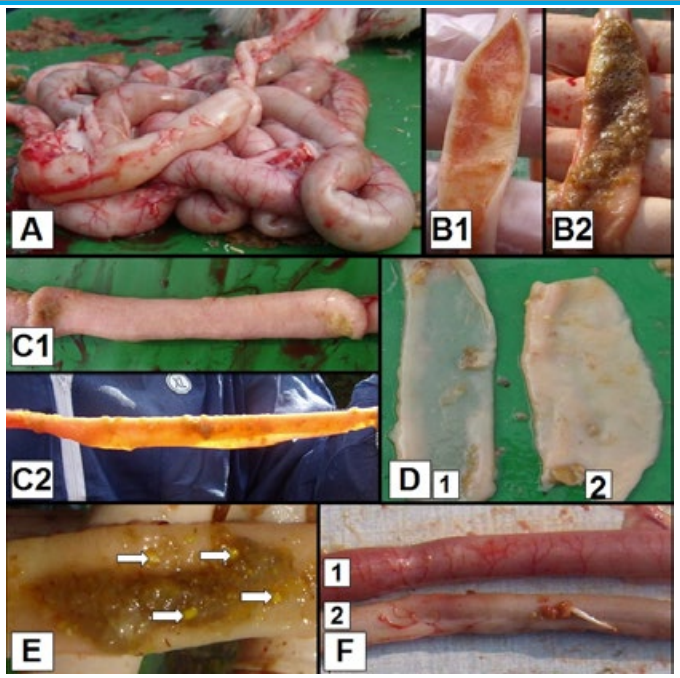
Macroscopic signs of BE

vetworks



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Vicious cycle of pathogenesis of BE

vetworks

High feed intake, high NSP levels, coccidiosis

1

oversupply of nutrients in lumen

Presence of nutritional factors that favour some bacterial groups and disfavour others causing disbalance

Increase in lumen of *Clostridium perfringens* and Clostridiaceae

2

Vicious circle Hypothesis

4

poor digestion of feed and absorption of nutrients

Morphological and functional alterations in the gut

3

Less functional gut

Inflammation and oxidative stress caused by interference of microbiota with mucosa

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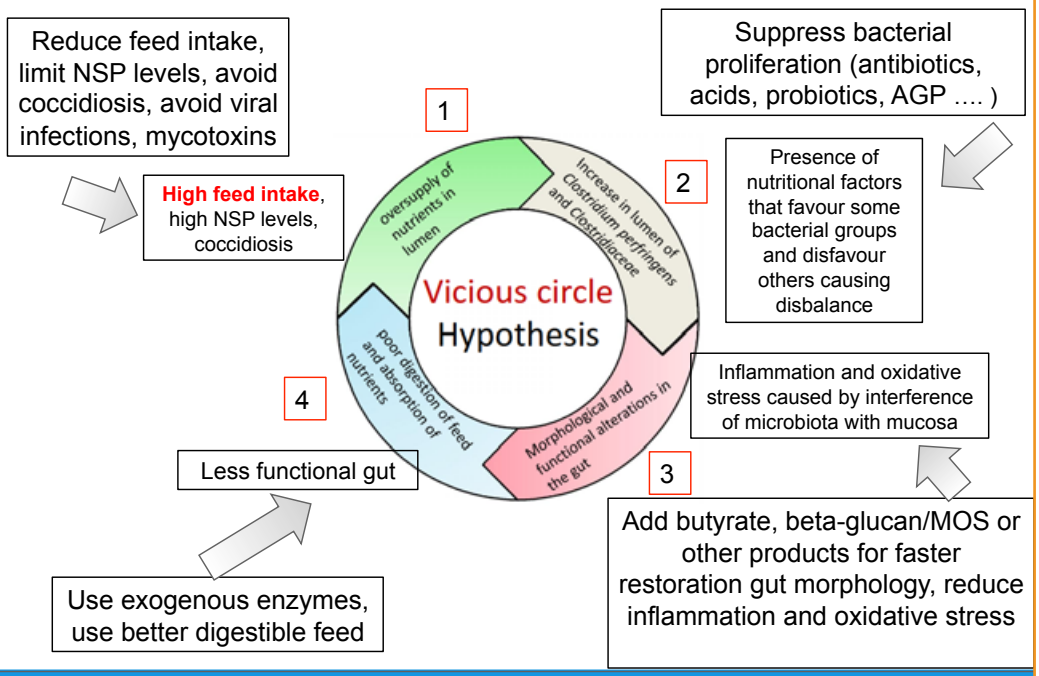
Solutions to Vicious circle of BE



BE trials

- Using a combination of trigger (coccidiosis) and difficult-to -digest feed (high NSP)
- Floor pen facilities only (or field trials)
- Control group with antimicrobial growth promotor
- Additionally parameters can be assessed such as macroscopic or histologic scoring systems
- Main impact on BWG and FCR, up to 100 gr BW reduction and 8 points higher FCR

Susceptible ages for BE

vetworks

- Broilers: from 15 days of age onwards
- Turkeys: between 2-10 weeks
- Breeder pullets : restricted feeding no BE
- Layers : genetically not susceptible for BE, but NE yes
- Every integration affected, prevalence per integration varies
- (feed quality, feed management, coccidiosis management, …)

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Consequences BE

vetworks

= most important reason for wet litter

- Pododermatitis: stress for birds (higher FCR)
- Breast blisters → rejections at slaughter
- Faster oocysts sporulation : increase of coccidiosis
- Increase of air-ammonia concentration (respiratory diseases)
- Welfare issue!
  - Not treating = against principles of Good Veterinary Practice (GVP) and ethical code veterinarians but AB use reduction !?

➔Big economic consequences

(↑ FCR, ↑ slaughter house rejections, etc.)

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Consequences BE

- BE is also seen in good performing flocks
- Flocks with too low feed intake usually no problems (layer pullets…)
- Financial consequence due to higher FCR
  - 70% of cost of broiler meat is feed
  - Digestion of feed is impaired (more undigested feed in faeces)
  - Absorption of nutrients is decreased (more nutrients in faeces)
  - Slower growth rate leads to longer fattening period for same age, and thus more feed needed for maintenance (higher FCR)
  - Higher inflammatory status of most important immunity organ of the body requires higher FCR

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Consequences BE

- Financial consequence due to lower body weight
  - Proteins used for immune reaction and tissue regeneration not available for muscle growth
  - Based on BE model, on average:
    - 150 gr lower BW at 42 when birds are not treated
    - 5 points higher FCR
- This is, based on 400€/ton feed price and 1€ per kg LW price 100 € impact on income per 1000 birds placed
- Per house of 20000 birds, 2000€ higher income per cycle
- Not counting costs of rejections and respiratory health problems, more coccidiosis development for next cycle

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Economics of Gut Health problems

vetworks

- Vetworks performing own trials to investigate how performance can be boosted
- Based on own floor pen trials and field trials we can estimate average cost of coccidiosis and BE:
  - 2.5 kg chicken
  - 0.1 Euro / chicken
  - from sub-clinical coccidiosis
  - +
  - 0.1 Euro / chicken from BE
- Total 20 cent per bird on average!
- 51 B birds = 10.2 B € losses / year! WW

35

AB used for NE/BE

vetworks

- Amoxycillin/penicillin
  - 80%
  - Broad spectrum!? Interferes with efficacy to amoxy for other pathogens!
  - Resistence (ESBL,...) pressure to stop use of beta-lactam AB
  - Cheapest AB on the market for this indication in many countries….
  - Works fast and good…
- Macrolides (tylosin and tylvalosin)
  - 15%
  - Small spectrum (G+)
  - More expensive than amoxycillin….
  - Resistance
- Lincosamides & other (5%)
  - Small spectrum and broad spectrum
  - Resistance

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Alternatives to enhance gut health

- Anticoccidials: indirect (less coccidiosis) and direct (if ionophores or robenidin)
- Alternative approaches supporting
  - Host defence (VC #3): gut barrier management: integrity and recovery of intestine
  - Control the microbial ecosystem directly (VC#2)

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Bacterial Enteritis scoring system

- There is a need for a uniform scoring system:
  - Although BE is on this moment the main indication for therapeutic use of antimicrobials in poultry in EU(-like) poultry production, and diagnosis is done very frequently, no uniform scoring system is accepted
  - Uniform system could be important for evaluating (in a scientific way) feeding strategies, providing guidelines and thresholds for treatments for BE, evaluating impact management measures (e.g. ventilation) on occurrence and consequences of BE and wet litter, research-linked evaluation of the efficacy of antimicobials and 'alternatives' (probiotics, acids, etheric oils,….)

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Diagnose better!

vetworks

- Very often, when wet litter occurs, BE treatment is started without proper diagnosis
- No isolation possible, no antibiogram, only macroscopic possible in field conditions. Need for veterinarian = cost.
- Improve/standardise diagnostics through scoring system (0 low -10 high)
  - Set thresholds for AB treatment - example
    - 0 – 2 no treatment needed
    - 3 - 5 only non-AB alternative treatments allowed
    - 6 - 10 AB allowed, only narrow spectrum G+
  - Document scoring for each treatment !!
  - Audit procedures : check threshold levels

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Macroscopy – Lesion scoring in the field

vetworks

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Strategy to deal with BE?

- Diagnose more to understand problem linked to certain operation
- Cost of diagnostics is increased, but estimated 30-40% of AB treatments on house level can be avoided, so pay back is expected
- Why is it not done today?
  - Scoring system is new, training needed in the industry
  - No uniform guidelines from authorities or stakeholders: more easy to 'ban' Antibiotics…. Some producers don't want to take the effort….believe it is cheaper to use AB
  - Audits don't focus enough on this particular aspect of AB use for gut health (lack of in-depth knowledge ?)
  - But welfare is compromised today when AB are simply banned….
  - Urgency needed!

Conclusions

- NE and BE important diseases in poultry
- Multifactorial with several predisposing factors
- Often antibiotic therapy
- Prevention is key:
  - Ruling out predisposing factors (coccidiosis, feed quality, ...)
  - Antimicrobials help but we must use them prudently to preserve efficacy
    - Negative image using antibiotics, resistance, legislations
  - Use of functional feed additives to improve gut health
  - Feed formulation and management

AB and 'alternatives' in use against BE

vetworks

- Classify them according to Vicious Circle (VC) so you can judge the additive or synergistic potential of alternatives
- Break VC by having solutions on all 4 steps of VC!
- Historically focus on step 2: suppress bacterial overgrowth
  - AGP – flavo, bacitracin, virginiamycin, macrolides VC#2 (&VC#3?)
  - Therapeutics Amoxycillin/penicillin/linco/macrolides VC#2 (&VC#3?)
- Today: Nutriad Solutions that are brought to Egyptian market are discussed in this role

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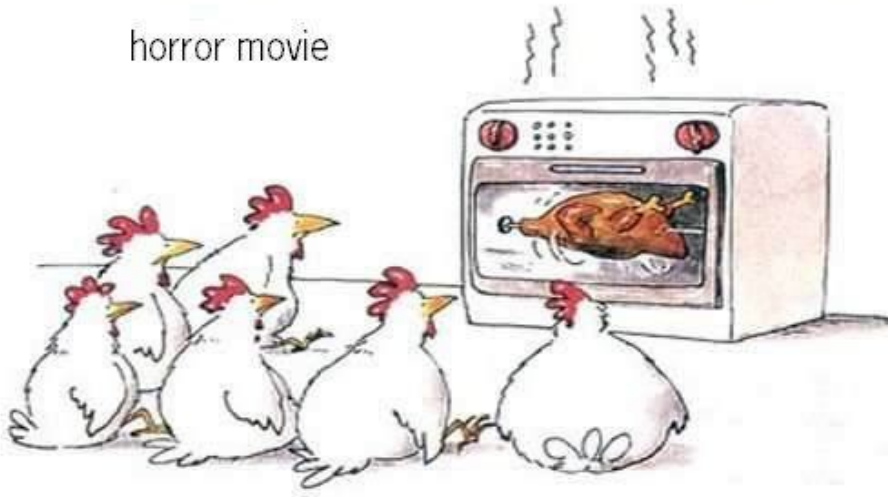
Thank you for your attention!

vetworks



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Bem Estar Animal

Malcom Mitchell

Myopathy in modern meat poultry mechanisms and solutions

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Summary

Rapidly growing and efficient modern meat poultry may develop growth associated and stress inducible myopathies. These pathologies may have welfare consequences and underlie a variety of meat quality problems such as wooden breast, white striping and PSE like conditions. Understanding the fundamental mechanisms responsible for this "metabolic disease" is central to the future development of genetic, nutritional and environmental strategies and approaches to reduce the incidence and severity of the condition and to optimise bird health, welfare and productivity.

Review of myopathy and mechanisms in meat poultry:

Current global production of broiler chickens is 59 billion birds per annum (FAO-Stat 2014). It is predicted that global demand for poultry meat will increase by 1.6% in 2014 and by an average 2.3% per annum until at least 2023 (OECD and FAO 2013) In the UK approximately 900 million broiler chickens are slaughtered per annum at an average body weight of around 2.2 kg. and the production of 1.6 million tonnes of meat and a value of £1.8 billion to the UK economy.

This staggering scale of output in the poultry sector has been supported by tremendous improvements in bird production traits achieved by genetic selection for high levels of performance and feed conversion efficiency, the development of optimum nutrition strategies and defined and accurate environmental control. It has been proposed that in meat animals selection for continued improvement in growth and efficiency should take account of potential physiological limits to growth and consider that approaching such limits may account for problems with animal health and product quality (Webb and Casey 2010). Such problems are described as "production and growth related disorders or metabolic diseases". Metabolic disorders or disease appear to result frequently from an excessive demand being imposed upon the animals' physiological or metabolic adaptive capacity by maximisation of the production performance and efficiency. It might be proposed that a simplistic operational definition of metabolic disease is a "mismatch in supply and demand" and that a failure of the some supply organs or systems e.g. cardiovascular, respiratory or digestive systems to meet the requirements of the demand systems e.g. skeletal muscle, reproductive functions or the supply organs themselves may result in compromise of function and pathology. An important example of such metabolic disease in meat poultry is a range of muscle pathologies often described in terms of their most apparent effects upon meat quality e.g. pale soft exudative meat, haemorrhagic muscle, "white striping" and "wooden breast". Muscle pathology in poultry has been reviewed previously Mitchell (1999), Kuttappen (2013) Petracci et al (2013)

Modern, rapidly growing strains of meat poultry exhibit an elevated incidence of spontaneous or idiopathic myopathy and an increased susceptibility to stress induced myopathy (Mitchell, 1999; Sandercock et al., 2006). Pectoral

and focal myopathy, dietary deficiency myopathies and toxic myopathies have long been recognized and described in poultry. These pathologies are attributable ultimately to alterations in intracellular cation regulation and calcium homeostasis (Sandercock and Mitchell 2003; Sandercock et al., 2006; Sandercock et al 2009) and consequent changes in sarcolemmal integrity and may result from excessive myofibre hypertrophy and inadequate development of support tissues and vascular supply (MacRae et al., 2006, 2007). Rapidly growing lines of birds may exhibit a reduced thermoregulatory capacity compared to their genetic predecessors and may thus be more susceptible to heat stress during production and in transport and to consequent problems, including muscle damage, acid-base disturbances and reduced meat quality (Sandercock et al., 2006). Genetic selection for improved growth rate and feed conversion efficiency may be associated with altered mitochondrial function and changes in the production of reactive oxygen species (ROS). In this context acute heat stress has been demonstrated to induce oxidative stress in broiler chickens, depress the mitochondrial respiratory chain complex and increases superoxide free radical production in skeletal muscle. This process is mediated by altered mitochondrial function and down-regulation of "uncoupling protein content". It has been proposed also that enhanced mitochondrial substrate oxidation plays a crucial role in increased production of reactive oxygen species in heat stressed broiler muscle probably via altered mitochondrial membrane potential. Transportation stress in broiler chickens has been demonstrated also to increase skeletal muscle metabolism, stimulate mitochondrial superoxide production and accelerate lipid metabolism resulting in tissue damage. A common feature of all myopathic and dystrophic conditions is the leakage of the intracellular muscle enzyme creatine kinase (CK) into the circulation. Thus increased plasma activity of CK is a useful diagnostic indicator of muscle pathology and altered sarcolemmal integrity (Mitchell 1999). It thus appears that the elevated enzyme efflux and muscle damage in the selected line broilers are associated with increased growth rate and not body size (Mitchell, 1999). Similar findings have been reported in comparisons of slow growing traditional line turkeys and a more rapidly growing commercial male line (Mills et al., 1999). Other studies have also demonstrated that rapidly growing broiler lines are more susceptible to stress-induced myopathy than genetically slower growing ones (Sandercock et al., 2001). Myopathy as evidenced by elevated plasma CK, is associated with demonstrable histo-pathological changes in muscle tissue. The condition is characterized by histological changes indicative of muscle degeneration including hyaline (hypercontracted) fibres, fatty infiltration, and fragmentation of the sarcoplasm, mononucleocyte infiltration and focal necrosis. Indicators of tissue regeneration such as basophilic fibres and internalized nuclei have also been observed (Mahon, 1999; Mills et al 1998 a and b; 2000; MacRae et al 2006, 2007). The onset of pathological changes appears to correlate with the attainment of a specific fibre diameter regardless of age or body weight suggesting a limit for fibre hypertrophy beyond which muscle function may be compromised (Mills et al., 2000). Successful genetic selection for muscle fibre hypertrophy accounts for the majority of increased muscle size. Many studies have shown a negative correlation between muscle fibre diameter and meat quality. Giant fibres (GF) are a post mortem observation resulting from hyper-contraction and fibre structural disintegration reflecting existing pathology. The incidence of GF is higher in rapidly growing lines of birds and in the pectoralis superficialis muscle of broiler birds. It has been reported that there is a positive relationship between plasma CK activity and muscle fibre diameter (cross sectional area or CSA) in broilers. Higher fibre CSA in females than in males has been observed and CK activity appeared to be higher in females. Those studies reported no obvious differences in meat quality traits associated with differences in CSA. Muscle fibre hypertrophy and increased radial fibre size is associated higher degrees of muscle damage in broiler birds compared to layers (MacRae et al 2006; Petracci 2012, Kuttappan et al 2012; 2013 a and c). In turkeys higher muscle fibre diameters in lines selected for improved growth rate are associated with more muscle damage and reduced growth rate and lower fibre diameter during food restriction decrease the extent of the myopathy (Mills et al 1998 a and b; 2000). Some studies have suggested that within two lines of rapidly growing broilers the genetic correlations between the incidence of breasts muscle myopathies (BMM) and breast meat weight or yield were low and propose that environmental factors contribute greater than 65% of the variance of the incidence of BMM.

More recently it has been proposed that muscle pathologies in rapidly growing meat birds and the attendant meat quality problems such as "white striping" and "wooden breast" may have a common aetiology associated with an

underlying idiopathic myopathy. Thus, the myodegeneration observed in pectoralis major muscle in broiler chickens may be associated with the subsequent development of fibrosis and regeneration. In muscle affected by white striping or hardness (wooden breast?) there may be moderate to severe polyphasic myodegeneration with regeneration as well as a variable amount of interstitial connective tissue accumulation (fibrosis). There was multi-focal degeneration and necrosis with hyper-eosinophilic amorphous fibres, a loss of striations and infiltration of inflammatory cells mainly macrophages and heterophils around the degenerative fibres. The factors released from these inflammatory cells will activate satellite cells initiating the regeneration of damaged myofibres. It has been proposed that when the extent or degree of idiopathic myopathy is excessive and sustained that the regenerative process will be ineffective (Mahon, 1999) and this will result in fatty degeneration. In that case, the pluripotent stem cells in the muscle tissue differentiate to fibroblasts or adipocytes due to the influence of the degenerating muscle fibre which ultimately results in fibrosis and lipidosis in the tissue as described in "white striping".

In young meat birds (broilers) the peak incidence of myopathies and meat quality issues occur around the normal age of slaughter (5-7 weeks of age. If birds of these lines or related breeding bird lines are grown to sexual maturity then myopathy becomes increasingly severe with increasing age and body weight but decreases at sexual maturity (Hocking et al 1998, 1999 &2001; MacRae et al 2006).This may be a consequence of ovarian steroid production and reduced reproductive performance after sexual maturity is associated with increased indices of myopathy in female birds (Hocking et al 1998, 1999 & 2002). Both treatment with oestrogen and increased oestrogen secretion at he onset of ovarian activity and sexual maturation in the female bird appear to reduce the extent of myopathy as evidenced by a decreased plasma CK activity and reduced incidence of histologically demonstrable abnormal fibres (Mitchell et al 1997, 1998 Carlisle et al 1999; MacRae et al 2006).

The present review has addressed the patho-physiological mechanisms underlying the aetiology and of these metabolic muscle diseases in meat poultry and the main risk factors for their development have been identified, characterised and described. Key issues are the regulation of intracellular cations specifically calcium and the alterations in cellular redox state and the production of reactive oxygen species and the consequent oxidation of cellular and membrane components and the resultant cellular and tissue dysfunction and subsequent regeneration and repair. Failure to regulate intracellular calcium results in a cascade of increased production of ROS, altered lipid peroxidation, impaired mitochondrial function, inflammatory responses, tissue remodelling, repair and regeneration but also increased apoptosis and tissue necrosis. The role of all of these processes in the both pathological changes and the associated effects upon bird health, welfare and product quality have been considered in detail. It is suggested that an improved understanding of the underlying mechanisms of metabolic diseases of poultry will constitute the basis of improved selection and interventions to minimise the incidence and impacts of these conditions.

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Malcom Mitchell

Animal transport: improving welfare and systems through physiological modelling

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Summary

The transportation of livestock is a major health and welfare issue as the procedure and conditions in transit may be the source of significant stress resulting in both health and welfare consequences. In addition transport stress can have profound effects upon production efficiency and product quality. It is proposed that identifying the specific stressors responsible for transport stress and the creation of appropriate quantitative, integrative, predictive models facilitates the development of practical solutions to the problems and sound scientifically based policy and legislation as well as forming the basis of education and training programmes for all those involved in livestock transport.

Animal Transport – Physiological modelling

The EU Strategy for Protection and Welfare of animals (2011 – 2015) states that "The Commission has prepared the second EU strategy for the protection and welfare of animals adopted in December 2011..... and Science is the foundation of the new Animal Health (and Welfare) policy.....it will be on the basis of solid scientific advice and information that future animal health and welfare rules are developed". Thus, it is essential that animal welfare directed research provides the sound scientific basis i.e. Evidence for all current and future policy and legislation in addition to providing the foundation of improved practices, procedures and animal transport vehicle and container design and operation. Indeed, Andrea Gavinelli (EC Commission for Animal Welfare) has indicated that:-"changes and amendments to animal welfare Regulations and any new or proposed Regulations must be based on sound and proven scientific principles and research outputs". The scientific evidence provided by research, therefore, constitutes the basis the review and assessment functions of the EFSA Working Groups and their reports to the AHAW Panel. The latter has performed scientific assessments on the welfare of a number of animal species. AHAW has proposed that "Further work will be needed to update those assessments in light of new scientific evidence and to develop new assessments in response to risk management demands". The Scientific Opinions of the AHAW Panel are used as a scientific basis for many of the EU legislative measures on animal welfare. For example, Council Regulation (EC) No 1/2005 on the protection of animals during transport is essentially based on conclusions and recommendations of the 2004 EFSA opinion on the welfare of animals during transport. This opinion has been updated (AHAW, 2011) and has contributed to a comprehensive report to the European Parliament proposing additional management options. The scientific evidence that underpins Scientific Opinion and ultimately legislation, regulation and codes of practice as well as improved or best practice and equipment must fulfil certain criteria. It is no longer useful for research involving characterization and analysis of animal transportation to merely identify or report apparent welfare problems and issues. It is now considered essential that research should quantify both the stressors imposed upon livestock in transit AND the consequences for the animals in terms of measurable outcomes i.e. physiological and behavioural responses and/or pathological consequences. This approach may be regarded as a quantification of imposed stress and the quantified consequences of that stress and as such may be described as

"physiological (and behavioural) stress response modelling". In turn, this may offer a way of accurate assessment of the effects of stress upon the welfare of animals in transit and constitute the basis for the identification and quantitative description acceptable ranges and limits for exposure to quantified stressors. Such models may constitute a central component of risk assessment in relation to the welfare of animals in transit. Biologists develop and use mathematical models to understand observed phenomena more fully, to characterize mechanisms and to predict the behaviour of systems under a range of conditions or in the face of changing inputs. Models may be derived from theory or empirical data or both, but ultimately should be applicable to "real world" situations and systems. Models may be generally classified according to their theoretical origins, the nature of the input-output variables and probability components and their descriptive or predictive functions. Thus, models describing a process or response may be regarded as analytical, numerical or observational. During transportation animals may be exposed to a range of concurrent stressors, which individually or in combination may compromise their health and welfare and have profound effects upon their subsequent performance or productivity and in the case of slaughter animals their product quality and thus overall production efficiency. Clearly losses due to mortality in transit represent a highly undesirable consequence of transport stress. The primary stressors that animals experience in transit include, thermal micro-environments (heat or cold stress), inadequate ventilation, vibration, acceleration, impacts, postural instability, fatigue, withdrawal of feed and water, exposure to novel stimuli, noise, contaminants, social disruption and the challenges associated with handling, loading and unloading.

The physiological modelling approach employed in research on animal transport may be described as having three specific objectives, and involving three fundamental approaches or phases. Thus,

Objectives:-

- To employ measurements of animals' physiological and behavioural responses to quantified stressors to characterise the effects of environmental, production and social challenges upon welfare status.
- To provide a sound scientific basis for the definition of acceptable ranges and limits for stressors to which animals are exposed in routine commercial agricultural practice.
- To provide the sound scientific basis for European and national legislation and codes of practice relating to animal production, methods, procedures and environments.
- Generally based upon three approaches or phases
- Full characterisation of the relevant commercial environments or procedures and identification and quantification of the primary stressors imposed in routine practice.
- Development of "physiological stress response" models under controlled laboratory conditions in which selected physiological, patho-physiological and behavioural responses to quantified individual stressors or their combinations are determined. From these models the magnitude of changes in physiological controlled variables or homeostatic effort expended can be determined for each "level" of stress imposed. The acceptable and unacceptable levels of stress can then be defined in terms of the magnitude of the physiological adaptive or compensatory responses or degree of dysfunction induced.
- Evaluation and validation of the models under field conditions and/or in commercial practice.

Such research programmes involve the development of a wide range of physiological and behavioural monitoring techniques. The outputs from these approaches and research programmes can then be employed as the basis for improvements in or modification of practices, procedures, environments, housing, and equipment in addition to informing codes of practice and regulations.

These strategies and approaches have been very successfully applied to improvements of animal welfare during transportation including the development of new vehicle designs, ventilation systems, and definition of acceptable thermal envelopes in transit for several species, journey duration optimisation and animal monitoring. Completed work has contributed to both current EU regulations and national legislation. The present review illustrates the nature of research and modelling with specific examples and case histories and demonstrates the impacts of the approaches on practice, guidelines and legislation and welfare monitoring and enforcement.

George Stilwell

A adaptação e uso de protocolos de avaliação de bem-estar animal por médicos veterinários

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Temas como a segurança alimentar, o impacto ambiental e o bem-estar animal, têm estado no topo das preocupações dos consumidores, que neste momento exigem saber onde, como e por quem são produzidos os alimentos de origem animal, desde o "prado ao prato". Mesmo que o criador e aqueles que o apoiam (e.g. médicos veterinários) não concordem com o conceito ou com as suas regras, a produção animal estará condenada a um futuro pouco risonho se estas exigências não forem levadas em conta.

Como resposta a estas reivindicações a legislação e uma série de programas certificadores têm-se multiplicado nos países desenvolvidos de forma a proporcionar algumas certezas à sociedade, nomeadamente quanto à qualidade e segurança dos alimentos, mas também quanto ao bem-estar dos animais de pecuária. Assim, o bem-estar animal tem-se estabelecido solidamente como um dos pilares da pecuária sustentável e eficiente.

Todas as definições mais consensuais de "bem-estar animal" consideram este conceito como sendo multidimensional, considerando-o como um estado de saúde física e mental completo e em que o animal vive em harmonia com o seu ambiente. Tradicionalmente são usadas as "Cinco Liberdades" para traduzir estas dimensões:

1. Livre de fome e sede;
2. Livre de doenças, lesões e dor;
3. Livre de stress e medo;
4. Livre de desconforto;
5. Livre para expressar comportamentos naturais.

Para conseguir transmitir de uma forma fidedigna o grau de bem-estar de um animal ou de uma população, deverão ser utilizados e integrados diferentes indicadores e sinais, de forma a cobrir todas estas diferentes dimensões. Sendo o bem-estar um conceito tão vasto e complexo, a avaliação não poderá estar limitada a um ou dois componentes mais óbvios (e.g. ausência de doença), mas terá de compreender uma abordagem multivalente. O resultado final será um protocolo, ou um sistema composto por uma sequência de protocolos, mais ou menos complexo dependendo dos objectivos.

Há cinco áreas onde a aplicação de protocolos de avaliação de bem-estar animal tem maior potencial – na investigação; como forma de monitorizar o cumprimento da legislação; em programas de certificação, normalmente estabelecidos para prestar informação ao consumidor; como ferramenta de gestão da exploração; e para perceber a prevalência e/ou evolução de doenças subclínicas numa população.

Nesta palestra serão apresentados os passos necessários à criação de um protocolo de avaliação de bem-estar, tomando como exemplo a nossa experiência pessoal no projecto AWIN-Animal Welfare Indicators que foi um projecto financiado pela Comissão Europeia (20011-2015). De uma forma resumida: um protocolo é constituído por uma

série de indicadores que procuram abarcar todas as dimensões já referidas do bem-estar animal. Por exemplo, ao se avaliar a condição corporal poderemos ter uma ideia da nutrição (Livre de fome) e também do impacto de doenças crónicas ou parasitismo (Livre de doenças). Ou quando avaliamos a distância de fuga de vacas leiteiras poderemos ficar com a noção da qualidade das relações animal-humanos (Livre de stress e medo).

Actualmente os protocolos usam, quase em exclusividade, indicadores baseados nos animais. Ou seja, a avaliação do bem-estar deixou de ser feita através de listas infindáveis de medições objectivas (e.g. dimensão de gaiolas; níveis de amoníaco; estado do piso), mas de significado controverso, e passaram a ser autênticos questionários aos animais – qual o impacto que as instalações, o maneio, os humanos, os coabitantes... estão a ter naquele momento sobre aqueles animais em concreto.

Estes indicadores, colhidos a partir do animal, podem ser vistas como diferentes peças acumuladas numa caixa-de-ferramentas. Para garantir a credibilidade dos resultados a caixa tem de estar cheia de ferramentas de qualidade, ou seja, que foram alvo de estudos que garantem a sua validade (medem o que pretendemos que meçam), a sua repetibilidade (os resultados serão iguais independentemente do utilizador ou do momento da observação), e a sua exequibilidade (podem ser recolhidos na exploração em tempo útil). Este conceito da caixa-de-ferramentas traduz perfeitamente a necessidade de flexibilidade e adaptabilidade dos protocolos às diferentes circunstâncias. Assim, conforme as características dos animais ou da exploração, serão seleccionadas ferramentas diferentes. Por exemplo, não faz sentido avaliar o impacto das estruturas dos edifícios em animais em pastagem mas fará se estivermos a trabalhar numa vacaria com cubículos.

Como é facilmente demonstrável o médico-veterinário é aquele melhor preparado para criar, desenvolver, adaptar e aplicar protocolos com indicadores baseados nos animais. Não só o médico-veterinário é o que melhor consegue identificar os sinais que os animais exibem (tantas vezes de forma subtil), como é o melhor (ou único?) que percebe o que significam. O médico-veterinário experiente é de certeza o leitor e o tradutor mais perfeito.

A criação de protocolos

Serão apresentados os protocolos criados no seio de dois grandes projectos europeus – o já mencionado AWIN e o Welfare Quality. Nestes projectos que juntaram investigadores de dezenas de instituições foram criados protocolos de avaliação de bem-estar animal para quase todas as espécies de produção. Em Portugal estes protocolos já foram aplicados, por equipas com as quais colaborámos, em diversos tipos de exploração como ruminantes leiteiros, suínos, frangos e galinhas poedeiras. Serão ainda apresentadas adaptações entretanto desenvolvidas para sistemas muito particulares – pequenas explorações de cabras em regime extensivo; pequenas explorações leiteiras não convencionais em regime "low-input"; e, explorações de vacas leiteiras mantidas permanentemente em pastagem nos Açores.

Todos estes estudos permitiram destacar as importantes diferenças destes sistemas em comparação com os sistemas mais intensivos, mas também permitiu detectar os seus maiores defeitos tornando a procura de soluções mais fácil e próxima.

As vantagens dos protocolos

O potencial da aplicação dos protocolos é enorme e por isso sugerimos que a formação e treino passem a ser procurados pelos Colegas envolvidos na área da produção animal.

Uma das principais vantagens dos protocolos é a capacidade de transmitir um imagem imediata e bastante clara do bem-estar, da incidência e impacto de certas doenças e do efeito de intervenções ou mudanças no maneio. Serve também para fornecer dados objectivos que permitem comparações entre explorações (benchmarking) ou entre diferentes momentos no mesmo efectivo.

Por outro lado permitem aceder a projectos de certificação voluntária, abrindo novas perspectivas de negócio ou promovendo mais-valias de um tipo ou sistema particular de produção. Por exemplo, a certificação através da aplicação de um protocolo de avaliação de bem-estar de vacas leiteiras em pastagem nos Açores permitirá "vender" esta imagem, chegando a um nicho de consumidores que provavelmente estaria a abandonar o consumo de leite cru. De nada serviria a sensibilização do público para os benefícios ou qualidade de produtos de origem animal se esta informação não pudesse ser certificada por entidades imparciais usando ferramentas de medição validadas.
De referir que os protocolos também são úteis para a investigação por reduzir a variabilidade na avaliação permitindo comparações, entre dois momentos de um estudo ou entre estudos de diferentes equipas, essenciais a uma análise robusta e significativa.

Aumentar a exequibilidade

Um dos maiores entraves à aplicação dos protocolos é a duração quando se quer tirar uma fotografia completa abrangendo todas as dimensões do bem-estar. Esta duração pode tornar inviável uma boa utilização dos protocolos ou restringir em demasiado o número de explorações avaliadas. Para circundar este inconveniente tem sido explorada a possibilidade da abordagem em dois tempos.

Ao desenhar e validar dois protocolos – um rápido e mais superficial e outro mais demorado e completo – podemos mais facilmente detectar as explorações com problemas graves. Para a primeira fase de avaliação serão retiradas da caixa-de-ferramentas os indicadores mais fáceis de usar mas também aqueles que podem traduzir o estado de mais do que uma dimensão de bem-estar. É, por exemplo, o caso da classificação da condição corporal que nos dá indicações quanto à nutrição ou prevalência de doenças debilitantes. A estas medidas deu-se o nome de "indicadores icebergue".

Aquelas explorações que demonstrem problemas significativos na primeira fase serão subsequentemente avaliadas com o protocolo mais complexo.

Uma outra possibilidade será a de usar dados recolhidos no matadouro e que servem de alerta para a detecção das explorações às quais se deve aplicar posteriormente o protocolo. Esta é uma forma bastante útil de abordar o bem-estar animal em explorações de suínos e aves. Devido ao grande número de animais abatidos, é relativamente fácil criar uma caixa de ferramentas a usar na linha de abate (e.g. número de fracturas ou hematomas, estado das penas ou lesões pulmonares). Este filtro permite identificar explorações problemáticas favorecendo um sistema de vigilância mais justo, constante e eficaz.

Também o uso de registos centrais podem ajudar a identificar as explorações a necessitar de uma auditoria mais

urgente. Indicadores como taxa de refugo/mortalidade, intervalo entre partos ou nascimentos, não são obrigatoriamente sinonimo de problemas de bem-estar mas, depois de validados, podem ser sinais de alerta.

Em conclusão, os protocolos são extremamente úteis para a identificação, monitorização e certificação do bem-estar de animais de produção. Cada vez mais estes protocolos usam indicadores baseados nos animais, sendo por isso essencial que se conheça e se reconheça alterações de comportamento e fisiologia e sinais de doença. O médico-veterinário é, sem dúvida, o profissional mais habilitado para recolher e interpretar todos estes sinais e portanto deverá procurar obter formação e experiência de forma a aplicar e liderar todo o processo de avaliação de bem-estar animal.

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Saúde Pública

Inês Almeida

Autobioresistência e o papel do médico veterinário

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Os antimicrobianos são essenciais para a medicina veterinária e humana para o tratamento de doenças infecciosas e zoonóticas, mas o uso inapropriado pode resultar na ineficácia destas moléculas. A manutenção da disponibilidade de todas as classes de antimicrobianos existentes e o desenvolvimento de novas moléculas para uso veterinário são essenciais para a manutenção da saúde e bem-estar quer dos animais de companhia quer dos de produção.

As comunidades médica e veterinária partilham responsabilidades no desenvolvimento da resistência microbiana. Cada utilização de antimicrobianos aumenta o risco de seleção de bactérias resistentes e, por isso, é necessário o desenvolvimento e aplicação de práticas sobre o uso responsável de antimicrobianos.

Não é possível o crescimento de animais em ambiente estéril; as infeções nos animais são uma realidade e os antimicrobianos continuam a ser vitais para o tratamento de um único animal ou de um grupo animais.

A nível europeu foi publicada a Lei da Saúde Animal que atribui responsabilidades ao Médico Veterinário no desempenho do papel ativo na sensibilização para a resistência aos tratamentos, incluindo a resistência antimicrobiana e as suas implicações e considera que a resistência aos antimicrobianos deve ser tratada como uma doença transmissível. A legislação europeia relativa ao medicamento veterinário e ao alimento medicamentoso está em revisão, sendo que estão a ser discutidas várias medidas no âmbito da resistência aos antimicrobianos.

Consciente desta problemática, a DGAV implementou os seguintes Planos: desde 2008 o Plano Nacional de Controlo de Utilização de Medicamentos, desde 2010 o Plano Nacional de Monitorização do Consumo de Antimicrobianos e desde 2014 que se encontra em execução o Plano de Ação Nacional para a Redução do Uso de Antimicrobianos em Animais.

Em Portugal, as vendas de antimicrobianos variaram entre 2010 e 2014. No total houve um aumento de 5% durante este período e entre 2011 e 2014, o aumento foi de 15%. O aumento de vendas entre 2011 e 2014 foi devido essencialmente às tetraciclinas, polimixinas, penicilinas e lincosamidas, tendo-se verificado uma redução substancial na venda de pleuromutilinas.

O padrão de venda de CIA* de categoria alto risco em Portugal, especialmente cefalosporinas de 3ª e 4ª geração, fluoroquinolonas e colistina é diferente da média europeia, sendo para as fluoroquinolonas quase 3 vezes superior (em mg/PCU, population correction unit).

A metafilaxia continuará a ser necessária face a surtos de doenças em grupos de animais de forma a minimizar a disseminação da doença.

Desta forma, a importância do Médico Veterinário no reconhecimento do seu papel na promoção e adoção de boas

práticas clínicas, na notificação de falhas de eficácia de antimicrobianos, na prescrição de antimicrobianos de espectro reduzido e na utilização responsável dos Critically Important Antimicrobials (CIA) é essencial.

* Critically Important Antimicrobials - Únicos ou em que a disponibilidade de medicamentos para tratamento de doenças graves no Homem é limitada ou ainda quando são medicamentos para tratamento de doenças no Homem causadas por agentes que se transmitam ao Homem através de fontes não humanas ou causadas por agentes que podem adquirir genes resistentes de fontes não humanas.

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Leptospirose, uma zoonose desafiante para a medicina humana e veterinária

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Resumo

A leptospirose é uma zoonose emergente que face à diversidade de reservatórios animais que disseminam leptospiras patogénicas no ambiente, requer uma visão holística e uma estratégia de controlo alicerçada no conceito "One Health". A casuística humana e animal será apresentada, bem como as principais dificuldades do diagnóstico e interpretação dos resultados laboratoriais.

Introdução

A leptospirose é uma zoonose emergente com distribuição mundial, registando-se hoje cerca de 1.000.000 de casos por ano (Costa et al., 2015). Atualmente considerada no grupo das doenças negligenciadas, a leptospirose representa um problema de Saúde Pública quer nos países desenvolvidos quer nos em desenvolvimento (ou de baixa renda).

A leptospirose ou "leptospiroses" são infeções causadas por bactérias patogénicas do género Leptospira, que afetam humanos e animais (WHO, 2012). A designação global de "leptospiroses" é um conceito que decorre do reconhecimento de que o quadro clínico observado e o subjacente quadro lesional são função da relação serovar-hospedeiro, o que representa um desafio ao diagnóstico clínico e na interpretação dos resultados laboratoriais, na medicina humana e animal.

As leptospiras patogénicas têm como habitat o rim e o trato genital de espécies animais que atuam como reservatórios (hospedeiros de manutenção), eliminando o agente para o ambiente via urina (Ellis, 2015). Apesar de se admitir que a maioria das espécies de mamíferos possa ser potencialmente infetada por vários serovares, sabe-se que efetivamente que apenas uma ou poucas espécies podem ser consideradas hospedeiras de manutenção para cada serovar (Bolin, 2000), assim como, apenas um pequeno número de serovares são endémicos numa determinada região (Ellis, 2015). Os pequenos mamíferos (insectívoros e roedores) são considerados os principais reservatórios de leptospiras, e constituem a fonte primária de infeção para humanos, que como hospedeiros acidentais são pouco eficientes para perpetuar a doença (WHO, 2012).

As infeções acidentais cursam com maior frequência com quadros agudos da doença e a excreção renal de leptospiras via urina é de duração limitada (Ellis, 2015).

Da interação serovar-hospedeiro resultam quados clínicos diversificados e compatíveis com outras doenças infe-

ciosas, pelo que o subdiagnóstico é muito frequente. Por um lado, ocorre uma sobrevalorização de alguns sinais clínicos, tais como a anemia, hemorragias e icterícia, muito característicos de infeções provocadas por determinados serovares (i.e., os dos serogrupos Icterohaemorrhagiae e Pomona) e, por outro, a desvalorização de quadros clínicos sem estes sinais.

Nos humanos a infeção ocorre pelo contacto direto ou indireto com urina, secreções, fluidos e anexos placentários, leite cru, etc. de animais domésticos e selvagens infetados por leptospiras. Pode também ocorrer pela ingestão de água ou alimentos contaminados com o agente infecioso (WHO, 2012).

Casuística humana e animal

A leptospirose animal vem sendo estudada em Portugal de forma mais sistemática desde a década de 80, depois dos primeiros inquéritos serológicos em animais terem tido lugar entre as décadas de 40 e 60, por iniciativa de Fraga de Azevedo e colaboradores (Fraga de Azevedo & Silva, 1942; Fraga de Azevedo & Palmeiro, 1943, 1961, 1964, 1969). Porém, já em 1926, Ricardo Jorge, havia diagnosticado retrospetivamente um surto epidémico ocorrido no país em 1914, como "Leptospirose icterohemorrágica" ou "Doença de Weil" (citado por Pacheco et al., 2000) e em 1931, foi reportado, o 1º caso humano com confirmação bacteriológica (Figueira, 1931). Contudo, o maior incremento da investigação da leptospirose humana e (em parte também animal), teve lugar a partir da década de 80, devendo-se a Collares-Pereira inúmeros trabalhos realizados quer no continente quer nas ilhas dos Açores (Collares-Pereira, 1982), estas últimas, consideradas áreas endémicas de leptospirose. Já em meados da década de 90 e nos primeiros anos deste século a referida investigadora e a sua equipa, prosseguiram os estudos epidemiológicos e laboratoriais (Collares-Pereira, 1991, 1992; Collares-Pereira et al. 1996, 2000a), tendo no período compreendido entre 2004 e 2008, sido desenvolvido um projeto sob o título "Epidemiologia e Controlo da Leptospirose na Região Autónoma dos Açores," ao abrigo de um acordo bilateral entre os EUA e o Governo Regional (Collares-Pereira et al., 2007), que procurou responder ao grande desafio com que a referida Região se confrontava há mais de uma década, face à elevada incidência de casos de leptospirose humana e animal, com o consequente aumento das taxas de morbilidade e, sobretudo de mortalidade no Arquipélago, em particular nas ilhas de São Miguel e Terceira (Pacheco et al., 2000; Vieira, 2006). Na altura o número de óbitos por leptospirose entre a população humana açoriana constituía, se não o principal, um dos principais problemas de Saúde Pública da referida Região insular (Vieira et al., 2006).

Acresce, que o referido projeto, pioneiro até então, nas valências que integrou: i) a epidemiologia, incluindo estudos retrospetivos e prospetivos da leptospirose humana e inquéritos KAP na população das duas ilhas; ii) o diagnóstico clínico; iii) o diagnóstico laboratorial, com obtenção de isolados humanos, e ainda uma componente de transferência de tecnologia de Lisboa (IHMT) para os dois hospitais (em S. Miguel e Terceira); iv) o estudo dos reservatórios silváticos, com a determinação do quadro referencial da leptospirose, incluindo a georreferenciação das principais espécies de roedores existentes, e o conhecimento das espécies de Leptospira que os mesmos veiculavam no ambiente; e v) populações animais e, em particular de produção pecuária, com obtenção de isolados de Leptospira spp em bovinos com o apoio do Laboratório Regional de Veterinária (Collares-Pereira et al., 2007; 2009).

Na Europa a leptospirose humana, nos últimos anos, tem vindo a manter uma taxa média de incidência de 0,13 /100 000 habitantes não sendo por isso uma doença com grande expressão (Dupouey et al., 2014). Porém, em Portugal, no Arquipélago dos Açores, entre 1993 e 2003, foi registada uma incidência de 11,1/100 000 habitantes, taxa muito superior à registada no continente, no mesmo período (Vieira et al., 2006). Recentemente o laboratório de Leptospirose e Borreliose de Lyme, do IHMT/UNL, um dos dois lab's de referência no país, para o diagnóstico laboratorial de leptospirose humana, levou a efeito uma atualização dos dados de 2003, através de uma série casuística compreendida entre 2004-2015. Esta incluiu 2539 indivíduos (1801 homens e 738 mulheres) com suspeita clínica de

leptospirose. Avaliou-se pelo teste de referência (TAM), um total de 3.304 amostras de soro, sendo 2.161 (65,4%) e 1143 (34,6%), provenientes do continente e ilhas açorianas, respetivamente. Observaram-se anticorpos anti-Leptospira interrogans sensu lato (s.l.) em 19,4% da amostra (n=493+/2539) que, de acordo com os critérios clínicos e epidemiológicos foram considerados como 'casos' (=doentes) de leptospirose, com uma média global de 41 casos/ano, sendo 188 (38,2%) no continente e 305 (61,8%) nos Açores. Os homens foram o grupo mais afetado em particular, aqueles com idades compreendidas entre os 37 e 61 anos e com profissões associadas à agricultura, criação de gado, saneamento básico e construção civil. Os serogrupos mais prevalentes foram Icterohaemorrhagiae (21%) e Ballum (47%). No entanto, foram ainda determinados outros serogrupos (Sejroe, Australis, Cynopteri e Pomona, este último, apenas registado no continente).

Quando comparado com a série anterior, este estudo mostrou um decréscimo do número de casos no Arquipélago açoriano, mas ainda com uma incidência elevada (10,2/100 000 habitantes), destacando-se positivamente a ausência de casos fatais desde 2008, data da conclusão do projeto anteriormente referido. Também no continente se observou um decréscimo da incidência da leptospirose humana (0,3/100 000 comparativamente ao valor encontrado em 2003 (0,7/100 000 habitantes).

Relativamente à leptospirose animal, a casuística existente referente às últimas décadas, reflete a contribuição de inúmeros investigadores para o conhecimento da epidemiologia da doença em Portugal. Apesar de alguns estudos sobre a leptospirose animal versarem casos clínicos em espécies domésticas (Lança, 2011; Duarte, 2015) e selvagens (Sargo et al., 2015), a maioria dos estudos realizados em Portugal têm assumido a forma de rastreios ou estudos retrospetivos de amostras provenientes de bancos de soros analisados pela técnica de aglutinação microscópica (TAM). Estes estudos vêm incidindo em espécies domésticas, tais como canídeos (Paiva-Cardoso et al., 2000a; Nóbrega, 2001; Paiva-Cardoso, 2011), gatos (Ato, 2015), bovinos (Collares-Pereira e Rocha, 1991; Flôr et al., 1998; Paiva-Cardoso et al. 2000b; Paiva-Cardoso, 2000 e 2009; Borrego et al., 2012), equídeos (Rocha et al., 2004), suínos (Rocha et al., 1998; Faria et al., 2016), e também sobre espécies selvagens, tais como roedores (Collares-Pereira et al., 1996, 2000a, 2007; Paiva-Cardoso et al., 2005b; Paiva-Cardoso, 2009), javalis (Vale-Gonçalves et al., 2015), ouriços-cacheiros (Paiva-Cardoso et al., 2016), corço, raposas, caprinos (Paiva-Cardoso, dados não publicados).

O isolamento de leptospiras patogénicas em Portugal a partir de animais e humanos tem sido de importância primordial no desvendar da epidemiologia da doença, permitindo identificar as espécies animais que funcionam como hospedeiros de manutenção no ecossistema (nos diferentes biótopos) e, adicionalmente, aumentar a sensibilidade e especificidade da prova de aglutinação microscópica pela sua inclusão na bateria de antigénios a testar.

Dado que o isolamento de leptospiras patogénicas é uma técnica muito exigente do ponto de vista técnico, morosa e dispendiosa, considerou-se fundamental sublinhar os desenvolvimentos alcançados neste âmbito em Portugal. Destacam-se os isolamentos de serovares do serogrupo Pomona, serovar Mozdok em suínos (Rocha et al., 1990) e em pequenos mamíferos (Collares-Pereira et al., 2000b), serovar Tsaratsovo em equinos (Rocha et al., 2004) e o "novo" serovar Altodouro a partir de roedores (Paiva-Cardoso et al., 2005b; 2005c, 2013). O serovar Hardjo foi também isolado a partir de bovinos (Collares-Pereira, 1991) e o serovar Icterohaemorrhagiae de roedores e humanos dos Açores (Collares-Pereira et al., 2000b; 2007, 2009; Gonçalves et al., 2009 e 2010). O serovar Copenhageni foi isolado de ouriços-cacheiros dos Açores (Collares-Pereira et al. 2000b). O serovar Ballum isolado de roedores na região de Trás-os-Montes (Paiva-Cardoso, 2009) e o serovar Arborea isolado de roedores dos Açores e do continente (Collares-Pereira et al. 2000b). O serovar Bratislava foi isolado de cavalos (Rocha, 2004) e o serovar Saxkoebing de Mus musculus (Paiva-Cardoso, 2009).

Em Portugal continental, a região de Trás-os-Montes tem sido alvo particular de estudos versando não só a leptospirose em animais de produção (bovinos, suínos e caprinos) e canídeos, mas também do seu estudo em espécies selvagens. A partir de 1999, a Universidade de Trás-os-Montes e Alto Douro (Vila Real) passou a dispor da

capacidade de realizar a técnica de referência (TAM), bem como da aptidão de realizar o isolamento, entre outras técnicas do diagnóstico da leptospirose. A região apresenta elevadas taxas de seropositividade em bovinos (61%) (Paiva-Cardoso, 2009) e suínos criados em regime extensivo (23%, TAM) (Faria et al., 2016), enquanto a taxa de positivos em canídeos doentes suspeitos de leptospirose ronda os 54,5% (Paiva-Cardoso e Nunes-Pereira, dados não publicados). Também, como parte integrante do ecossistema, as espécies selvagens têm sido alvo de estudo, revelando elevadas taxas de positividade em roedores (55% pela TAM e 52,3% pela PCR) (Paiva-Cardoso, 2009), javalis (65,4%; TAM) (Vale-Gonçalves et al., 2015) e ouriço-cacheiro (53,3%; TAM) (Paiva-Cardoso et al., 2016). Outras espécies como o corço, raposas e lobo têm mostrado títulos de anticorpos relevantes, que serão brevemente alvo de publicação. De forma geral, a maior atividade aglutinante ocorre com o serovar Altodouro, estirpe 139, isolada de Mus musculus capturado na região de Trás-os-Montes (Paiva-Cardoso et al., 2013) e que viria a ser reconhecido pelo "Subcommittee on the Taxonomy of Leptospiraceae" como um novo serovar, pertencente à espécie de Leptospira kirschnerii (Levett e Smythe, 2014).

A divulgação de resultados e as ações de sensibilização das populações escolares, rurais, de produtores e junto das associações de caça tem permitido obter a colaboração dos vários "atores" nesta região, possibilitando reunir esforços no combate aos prejuízos económicos devidos à leptospirose nos animais de produção e aos riscos de saúde pública.

Diagnóstico Laboratorial

As técnicas laboratoriais com maior utilidade e relevância no contexto do diagnóstico são: a técnica de aglutinação microscópica (TAM); a observação direta de leptospiras na urina por microscopia de fundo escuro e a reação em cadeia da polimerase (PCR) realizada no sangue e na urina.

A TAM permanece como o teste serológico de referência devido à sua elevada sensibilidade e especificidade, quando a bateria de serovares/estirpes, usadas como antigénios vivos, é alargada e adaptada à área de estudo. A não inclusão dos isolados nacionais na prova serológica de referência ou o envio de soros de animais suspeitos de leptospirose, para laboratórios estrangeiros em regime de "Outsourcing" que, por regra, testam um número reduzido de serovares e incluem nas baterias de antigénios apenas os isolados locais do seu país, têm gerado muitos resultados falsos negativos, conduzindo em larga medida ao subdiagnóstico de casos clínicos da leptospirose animal em Portugal.

A interpretação dos resultados serológicos é uma tarefa complexa, em particular na leptospirose animal, cujas dificuldades se encontram associadas a: i) existência de reações cruzadas entre os diversos serovares; ii) presença de títulos de anticorpos induzidos pela vacinação; iii) falta de consenso sobre os títulos de anticorpos aglutinantes considerados indicativos de uma infeção em curso e; iv) falta de correlação entre as prevalências serológica e bacteriológica (Paiva-Cardoso, 2009).

A observação de leptospiras na urina em microscopia de fundo escuro pode revelar-se muito profícua na definição do estado de portador crónico de leptospiras e determinar a aplicação de estreptomicina para eliminar as leptospiras dos túbulos contornados proximais do rim e, assim, prevenir a infeção de animais e humanos coabitantes. Esta metodologia apresenta como principais limitações: i) dever ser realizada nas primeiras horas após a colheita; ii) observada em microscópios de fundo escuro com uma elevada intensidade de luz; e iii) efetuada por técnicos muito experientes. Em caso positivo, a urina suspeita deve ser inoculada em meios apropriados, por forma a tentar o isolamento do agente.

O diagnóstico molecular pela PCR convencional ou "nested" pode ser muito proficiente se efetuada no sangue,

na fase inicial da doença, e na urina nos estádios mais avançados da enfermidade. Contudo, os resultados positivos ou negativos não podem ser interpretados como confirmatórios da presença ou ausência de infeção ativa, se não forem coadjuvados pela observação do aumento do título de anticorpos em amostras pareadas ou um título anormalmente elevado, bem como da presença de sinais clínicos compatíveis com a doença. A PCR efetuada, post mortem, em tecido renal, é muito útil dado que permite a identificação do estado de portador renal (Paiva-Cardoso et al., 2005a). A técnica de PCR apresenta muitas limitações em termos de informação epidemiológica, pois não permite, só por si, identificar a estirpe de Leptospira infetante o que limita o prognóstico da possível evolução da doença (quadro lesional e desfecho). Acresce que, o seu valor diagnóstico é mais adequado nos primeiros dias de infeção, quando ainda não existe produção de anticorpos específicos contra o referido agente. No entanto, quando a qualidade do produto amplificado permite a sequenciação molecular é possível identificar a genoespécie. Porém, também nem sempre há uma correlação direta entre a caracterização molecular e serológica, já que coexistem dois sistemas de classificação, um de base molecular, atualmente com 21 espécies genómicas conhecidas, e outro de base antigénica (serológica) com as duas espécies clássicas (L. interrogans- patogénica, e L. biflexa - saprófita), ambas divididas em serogrupos e serovares.

A técnica de real-time PCR tem mostrado resultados promissores relativamente à identificação das genospécies de Leptospira que infetam animais (Picardeau, 2013; Ferreira et al., 2015) e humanos (González et al., 2013), mas a sua aplicação a amostras clínicas ainda apresenta resultados com algumas limitações, desde logo, as já referidas para a PCR convencional, a que acresce também a necessidade de recursos especializados, quer laboratoriais (equipamento) quer humanos.

Profilaxia vacinal em animais

Apesar da vacinação dos animais não impedir a infeção por leptospiras patogénicas é uma ferramenta indispensável para reduzir a mortalidade e morbilidade das leptospiroses em animais de produção, de desporto e de companhia. Dado que a vacinação só confere imunidade para os serovares presentes na vacina ministrada, é de extrema relevância que a escolha das valências siga o princípio de imunizar para os serovares mais prevalentes na área geográfica e/ou os que se traduzem em quadros lesionais mais graves para a espécie animal em causa.

Considerações Finais

Em termos globais parece poder dizer-se que em Portugal se verifica atualmente uma redução da leptospirose humana, devendo porém, ser mantida a vigilância e controlo epidemiológico por parte das autoridades de saúde locais (Collares-Pereira et al., 2009), de modo a poderem manter-se os "ganhos para a saúde" que começaram a verificar-se a partir de 2009, decorrentes precisamente da ativação de linhas de orientação deixadas pelo projeto, na Região Autónoma dos Açores (RAA) (Collares-Pereira et al., 2009).

Importa, no entanto, relevar, por aparentemente poder ser paradoxal, que o valor elevado da incidência da leptospirose atualmente observado na RAA, se deverá mais à maior sensibilização dos profissionais de saúde no que respeita ao diagnóstico, ao mais efetivo apoio dos laboratórios locais e à atitude da população que recorre a assistência médica mais precocemente aquando dos primeiros sinais e sintomas, do que a um aumento do caráter endémico da leptospirose na Região, pelo que importa enfatizar esta melhoria registada desde 2009, até ao presente.

É assim possível, falar-se hoje de um exemplo bem-sucedido de um sistema ativo e permanente de controlo e vigilância epidemiológica no Arquipélago dos Açores, onde os protagonistas são primeiramente as populações locais e as Autoridades de Saúde (humana e veterinária), numa demonstração inequívoca da importância de reunir Institui-

ções e saberes em projetos integrados de investigação, o que na prática, significa concretizar o conceito, hoje tão atual, de "One Health".

Na sequência dos trabalhos realizados desde 1999 na região de Trás-os-Montes, a epidemio vigilância da leptospirose animal estende-se atualmente no contexto "One Health" a toda a região norte do país. Esta linha de investigação liderada pelo grupo de Ecologia Médica, do Laboratório de Ecologia Aplicada (CITAB/UTAD), financiada pelo Projeto INTERACT/BEST, linha "Predicting the regional occurrence of zoonotic diseases and the potential consequences for agri-food chains" n.º da operação NORTE-01-0145-FEDER-000017 (INTERACT/BEST, 2016), continuará a ser assegurada nos anos vindouros por uma equipa multidisciplinar que conta com diversas parcerias intra e extrauniversitárias, associações, serviços de saúde, entre as quais se salienta a estreita colaboração com o IHMT.
A manutenção de agentes zoonóticos na fauna selvagem, apenas recentemente tem mobilizado atenção dos médicos e médicos veterinários, habitualmente mais focados na resolução de casos clínicos. O alerta tem resultado da emergência e reemergência de zoonoses, nalguns casos devido há falha de alguns programas de controlo e erradicação aplicados a animais domésticos, por não terem sido tidos em conta os reservatórios selvagens como focos de disseminação dos agentes patogénicos e não considerarem as alterações ambientais em curso.
Como a História vem demonstrado, só as ações colaborativas entre profissionais com vários saberes permitem uma efetiva mitigação dos custos associados às zoonoses em termos de Saúde Global, exigindo uma abordagem devidamente enquadrada no interface humanos-animais-ecossistema.

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Jardins, Espaços Públicos e Áreas Caninas: Fontes de Infestação e Infeção Parasitária para Animais de Companhia*

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Resumo

O desenvolvimento de zonas urbanas e suburbanas, com grande densidade humana e animal, tem proporcionado o aparecimento de grande número de jardins/espaços públicos de lazer para humanos e/ou animais de companhia. A criação destas zonas públicas de recreio, à qual se juntaram recentemente as áreas caninas, não são isentas de riscos, pois também potenciam o contacto dos animais com os parasitas, que são veiculados diretamente por outros animais ou indiretamente pelo ambiente contaminado, como é o exemplo dos dejetos dos canídeos não recolhidos pelos respetivos proprietários. O risco de contaminação parasitária dos espaços públicos compreende artrópodes (como ixodídeos e pulgas, bem como as respectivas doenças transmissíveis), protozoários (géneros Giardia, Cystoisospora e Cryptosporidium) e helmintes (Echinococcus spp., Toxocara spp. e Ancylostomatidae). Neste trabalho serão destacados estes e outros parasitas que são relevantes não só pela sua importância na Saúde dos Animais de Companhia, mas também pela sua vertente de Saúde Pública e de Saúde dos Ecossistemas. Será dado relevo à realidade nacional, no que respeita à prevalência dos parasitas e ao seu controlo, assim como à regulamentação destes espaços e propostas de solução desta problemática.

Introdução

Os animais domésticos de companhia têm aumentado vertiginosamente de densidade e importância nas últimas décadas, sendo considerados como mais um elemento do agregado familiar. Em particular, o número de carnívoros domésticos aumentou substancialmente nas grandes cidades, situação mais notória nos últimos 30 anos. A sua confraternização e proximidade com os proprietários são de tal forma intensas, que alguns autores são mesmo da opinião que "os animais de companhia mudaram-se do jardim para o quarto", acabando por se tornar membros da família (Becker, 2002). Este aumento acabou por se repercutir ao nível da saúde animal e pública, principalmente nos grandes aglomerados urbanos, pelo maior número de pessoas e respetivos animais. Esta consequência de carácter sanitário pode ter várias causas, entre as quais a pouca informação dos proprietários relativamente aos agentes transmissíveis de doença dos seus animais, a não observância de regras básicas de higiene e sanidade animal, o elevado número de animais errantes como consequência do seu abandono e a deficiência nos programas de saúde animal, nomeadamente quando não recomendados pelo Médico Veterinário ou quando o sejam, não são cumpridos com eficácia (Madeira de Carvalho et al., 2005; Matos et al, 2015).

Os autores farão de seguida uma descrição de alguns dos resultados obtidos a nível nacional em vários espaços públicos e espaços dedicados especificamente a canídeos, dando especial ênfase a alguns parasitas reconhecidamente com importância em Saúde Animal e Saúde Pública.

Metodologias de isolamento de Parasitas com Importância em Espaços Públicos

Os ixodídeos são provavelmente os ectoparasitas com maior importância em zonas públicas, não só pela sua frequência de detecção, mas pelas consequências patogénicas diretas (espoliação sanguínea, inoculação de toxico-alergenos) e indiretas (transmissão de vírus, bactérias e parasitas) da sua infestação (Cruz-Cabezas et al., 2016). A colheita de ixodídeos em cães presentes a consultas em CAMV em Portugal, que frequentaram espaços diversos, incluindo públicos, promoveu um melhor conhecimento da sua frequência (por identificação morfológica) e dos agentes potencialmente transmitidos (através de métodos moleculares) (Pereira da Fonseca et al., 2016).

No âmbito dos endoparasitas e em particular dos gastrointestinais (GI), trabalhos recentes têm procurado avaliar a prevalência de ovos de helmintes GI dos canídeos no solo dos parques públicos da Grande Lisboa, em particular os de Toxocara spp., averiguando a viabilidade de infeção dos respetivos ovos. Foram efetuadas colheitas de amostras fecais e de solo em 12 parques públicos e 7 caixas de areia de áreas semi-públicas na Grande Lisboa. No total, foram colhidas 151 amostras de solo e 135 amostras fecais de Outubro de 2013 a Setembro de 2014. As amostras de solo foram analisadas segundo a técnica de Centrifugação, Sedimentação-Flutuação (CSF) (Santarém, Magoti, & Sichieri, 2009), enquanto as amostras fecais foram analisadas pelo método adaptado de Cornell-Wisconsin (Egwang & Slocombe, 1982; Levecke et al., 2012). A incubação dos ovos recolhidos foi realizada em H2SO4 0,05M durante um período de 60 dias (Otero et al., 2014; Otero, 2015).

Em paralelo e com o intuito de avaliar o risco parasitário gastrointestinal dos canídeos em zonas de exercício e lazer próprios, foram avaliados alguns parques caninos na zona da Grande Lisboa, como novas áreas por excelência de convívio animal e dos respectivos proprietários. Para o efeito foram colhidas amostras fecais e de solo em três parques caninos, dois em Lisboa (Benfica e Campo Grande) e outro em Oeiras (Algés). As colheitas decorreram nos meses de Outubro a Dezembro de 2014, totalizando 369 amostras fecais (125 de Algés, 124 de Benfica e 120 do Campo Grande) e 18 amostras de solo (6 de cada parque) (Ferreira, 2015; Ferreira et al., 2015, 2015a). Estas foram sujeitas a pesquisa de ovos/(oo)cistos de parasitas pelas técnicas de CSF (Dryden, Payne, Ridley, & Smith, 2005), e esfregaço fecal corado pela técnica modificada de Ziehl-Neelsen (Casemore, Armstrong, & Sands, 1985).

Com o propósito de avaliar a prevalência de Angiostrongilose canina, doença cardiopulmonar grave transmitida através da ingestão de moluscos gastrópodes (caracóis e lesmas) infetados pelo parasita Angiostrongylus vasorum (Helm et al., 2010), foi realizado um rastreio serológico nacional, utilizando a técnica de ELISA em 906 cães, mantido em canis de Norte a Sul de Portugal (Alho et al., 2016). Este parasita deve ser considerado como mais uma ameaça à saúde dos canídeos, dado que os seus veículos de infecção (moluscos gastrópodes) estão facilmente disponíveis nos espaços públicos e a sua componente patogénica (cardiovascular, neurológica e hemática), pode revelar-se fatal (Alho e Madeira de Carvalho, 2016).

Principais parasitas em locais públicos

No trabalho efectuado com cães presentes a consultas em CAMV, no total de cerca de 1140 ixodídeos colhidos, 95,5% eram da espécie Rhipicephalus sanguineus, 1,8% da espécie Ixodes ricinus e 1,1% de I. hexagonus. Outras espécies como R. pusillus, Dermacentor reticulatus e Hyalomma marginatum, também foram identificadas com frequências entre 0,1 -0,6%. Os agentes patogénicos identificados incluíram Babesia/Theileria spp. (estimativa de

10-65% dos ixodídeos examinados), Anaplasma/Ehrlichia spp. (11-64%), Rickettsia spp. (5-38%) e Coxiella burnetti (0,54%). Sendo R. sanguineus a espécie com maior representatividade, não será de estranhar que todos estes agentes tenham sido isolados neste ixodídeo (Pereira da Fonseca et al., 2016).

No que respeita a prevalência de Toxocara no solo dos parques públicos da Grande Lisboa, 50.0% dos parques (6/12) e 85.7% das caixas de areia de parques infantis (6/7) estavam contaminados com ovos de Toxocara spp. A prevalência foi especialmente elevada nas caixas de areia dos parques infantis. Observou-se também uma taxa de viabilidade de 56% dos ovos recolhidos, com uma densidade média de 4,2 ovos por cem gramas de solo. Relativamente às amostras fecais, 15,8% de todos os locais estudados e 5,9% das amostras foram positivas a Toxocara spp. Dos ovos de Toxocara spp., em 50,8% não foi possível distinguir se eram T. cati ou T. canis, enquanto 41,7% foram identificados morfologicamente como T. cati e 7,5% como T. canis (Otero et al., 2014; Otero, 2015).

Relativamente aos parques caninos testados, a prevalência global de amostras fecais positivas foi de 33,1%. Os agentes parasitários encontrados foram Ancylostomatidae (16,5%), Cryptosporidium spp. (11,9%), Giardia sp. (11%), Toxascaris leonina (1,1%), Cystoisospora spp. (1,1%), Toxocara spp. (0,5%) e Sarcocystis spp. (0,3%). A prevalência de protozoários e nemátodes foi de 23,6% (87/369) e de 16,8% (62/369), respectivamente. Das amostras de solo, 27,8% (5/18) estavam contaminadas com ovos de Ancylostomatidae, sendo os três parques positivos. Adicionalmente, foram observadas amostras fecais em todos os parques caninos analisados, indicando que muitos proprietários não apanham os dejetos dos seus animais, um comportamento fulcral para reduzir a contaminação ambiental e salvaguardar a saúde pública e animal (Ferreira, 2015; Ferreira et al., 2015, 2015a).

Quanto à situação de prevalência de Angiostrongylus vasorum em Portugal, 0.66% dos canídeos testados foram positivos simultaneamente no teste de antigénio e de anticorpo, indicando uma infeção ativa por A. vasorum; 1.32% foram positivos apenas no teste de anticorpo, indicando um contacto prévio com este agente. Desse modo, casos de infeção ativa e de exposição prévia foram observados na zona Norte, Centro e Sul de Portugal, confirmando a endemicidade desta doença no nosso País (Alho et al., 2016).

Discussão e Conclusão

Em Portugal, devido às alterações climáticas, o estudo dos ixodídeos é extremamente importante pois algumas das suas doenças transmissíveis podem ser particularmente favorecidas, tanto mais que são conhecidas 12 espécies de carraças parasitas dos canídeos domésticos em Portugal (Casimiro et al., 2006; Santos-Silva et al., 2011). As espécies de ixodídeos mais assinaladas no estudo recente efetuado em canídeos são consideradas aquelas com maior variedade de hospedeiros, incluindo o Homem, sendo R. sanguineus e Ixodes ricinus muito associados à Doença de Lyme e à Febre Escaro-Nodular em Humanos (Santos-Silva et al., 2011). Mas há que considerar também a uma disseminação latente e patente de vários agentes responsáveis por Doenças Transmitidas por Vectores, com especial referência para Babesia/Theileria spp., Anaplasma/Ehrlichia spp. e Rickettsia spp, em particular de Norte para Sul do país, tanto como agentes de elevada patogenia para os cães, como de zoonoses graves (Cardoso et al., 2012; Pereira da Fonseca et al., 2016).

A elevada contaminação e prevalência de formas parasitárias do género Toxocara encontrada nos parques públicos da Grande Lisboa é preocupante, em particular nas caixas de areia dos parques infantis, um meio crítico de contágio e perpetuação da infeção em crianças. Estes dados sugerem que as medidas em curso (cercados, proteção física e desinfeção da areia) são pouco eficientes. Recorde-se que a Toxocarose é a zoonose parasitária mais comum dos países desenvolvidos, senda a ingestão de ovos presentes no solo a via de infeção mais frequente para o ser humano (Madeira de Carvalho et al., 2005). Em Portugal, estudos ambientais referentes a este parasita são praticamente inexistentes. Em 2006, no conselho do Seixal, foi realizado um estudo que reportou um total de 40,4% de parques

estudados; 10,8% das amostras de solo recolhidas; e 1,3% das amostras fecais positivas para ovos deste parasita (Fernandes, 2006).

Nos três parques caninos analisados na zona de Lisboa, foi observada um terço das amostras fecais positivas a pelo menos um agente parasitário. Ancylostomatidae foi o agente parasitário detetado com maior prevalência, o que vai ao encontro do observado em áreas rurais, quintas e cães de caça do noroeste de Portugal (Mateus, Castro, Ribeiro, & Vieira-Pinto, 2014). A prevalência de protozoários foi superior à de nemátodes, uma tendência comum registada nas últimas décadas e possivelmente explicada pela maior consciencialização dos donos e consequente aplicação de antiparasitários profiláctico que normalmente não cobrem protozoários intestinais (Robertson, Irwin, Lymbery, & Thompson, 2000). Ainda assim, a elevada prevalência detetada neste estudo de hemintes geralmente abrangidos por produtos de desparasitação regular, sugere que apesar do contacto frequente com outros animais, poucos cães são desparasitados internamente com a frequência recomendada (mínimo trimestralmente) (ESCCAP, 2010).

Até o ano passado, dados publicados sobre a ocorrência de A. vasorum em Portugal eram praticamente inexistentes, algo justificado pelo facto de se tratar de uma parasitose subestimada na prática clínica de pequenos animais. Para além disso, estudos indicam que apesar da maioria dos donos desparasitar os seus animais de companhia, fá-lo em intervalos irregulares e consequentemente ineficazes (Matos et al, 2015). Todos estes fatores mostram a importância de educar os proprietários de animais sobre métodos de prevenção e mecanismos de transmissão para controlar a situação atual.

Estes resultados alertam para o potencial dos parques caninos e dos jardins e espaços públicos como fonte de transmissão de diversas parasitoses, em particular, quando práticas efetivas de desparasitação (tanto externa, como interna) e medidas de limpeza apropriadas estão em falta. Em suma, torna-se fundamental envolver os cidadãos promovendo uma melhor informação por parte do Médico-Veterinário, uma desparasitação (realmente) regular dos animais e a promoção da limpeza dos espaços urbanos, que pertence a todos nós, mas cujos serviços oficiais (centrais e municipais) devem acompanhar de perto, consciencializando-os para a necessidade de um esforço conjunto no sentido de melhorar a qualidade higio-sanitária dos espaços públicos e promover a saúde pública e animal.

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Tecnologia Alimentar

Natasha Planas Dias

Refeições Halal no Inflight Catering

Qualidade e Segurança Alimentar

As refeições Halal são as refeições consumidas pelos Islâmicos e que estão de acordo com as Leis Islâmicas.

Será demonstrado todo o processo envolvido na produção deste tipo de refeições desde o abate ritual, aos ingredientes que as compõem, a todo o processo e layout da sua produção até ao fornecimento a bordo da aeronave.

Halal Meals are the meals consumed by Islamic people and that are in compliance with Islamic laws.

We will show you everything that is involved in the production of these kind of meals since the ritual slaughter, the ingredients that can be used, the layout, until loading the aircraft.

Natasha Planas Dias

Perda e Desperdício Alimentar

Qualidade e Segurança Alimentar

A aplicação de legislação (Reg1774/2002) obriga todos os caterings internacionalmente a destruírem todos os "restos de cozinha e de mesa provenientes de meios de transporte que efectuem transportes internacionais."

Será demonstrado o longo processo que levou a Cateringpor juntamente com a TAP e Associação Dariacordar a aplicar o chamado "Zero Desperdício" no catering da aviação.

In order to accomplish the EU Reg 1774/2002, all the inflight caterings have to destroy "all catering waste from means of transport operating internationally".

It will be shown the long process taken by Cateringpor together with TAP and "Dariacordar" to apply the so called " Zero Waste" in an Inflight Catering.

Daniela Silva

As novas tecnologias moleculares e o seu impacto na avalição da autenticidade alimentar

Nos últimos anos tem-se assistido a um aumento significativo de casos de fraude nos diferentes sectores da área alimentar, sendo a verificação da autenticidade de produtos alimentícios um dos requisitos mais solicitados pelos distribuidores e consumidores. Considerada uma das áreas da investigação de ponta, a Biologia Molecular apresenta várias técnicas de elevada fiabilidade que permitem dar resposta a esta questão, tais como PCR (reação de polimerase em cadeia) e sequenciação de ADN. Estas tecnologias têm acompanhado o processo evolutivo com inovações que permitem dar respostas essenciais e de forma expedita. Como exemplos de aplicações destacam-se o controlo de autenticidade de produtos, o controlo da rotulagem, a detecção de OGM, a identificação de espécies, entre outros.

Reconhecida como sendo um dos mais importantes avanços científicos do século 20, a técnica de PCR é uma maneira rápida e fácil de criar um número ilimitado de cópias de um ADN alvo permitindo a detecção / identificação de uma dado organismo/gene. Milhões de cópias de uma seção de ADN são obtidas em apenas algumas horas. O ADN copiado pode então ser usado com fiabilidade em uma ampla variedade de testes. Nas últimas duas décadas, a técnica de PCR foi modificada de modo a tornar-se mais versátil e de aplicação mais ampla. A base da técnica mantém no entanto os mesmos princípios, havendo introdução de avanços tecnológicos à medida que o ADN amplificado é detectado. Assiste-se assim à emergência das técnicas de PCR em tempo real onde a amplificação de ADN é visualizada por monitorização de fluorescência e mais recentemente à evolução para métodos de PCR digital que permitem uma quantificação absoluta do número de moléculas de ADN alvo presentes numa determinada amostra.

No entanto, as metodologias baseadas na tecnologia de PCR têm a desvantagem de necessitar que seja conhecido à priori qual a espécie/organismo/gene a identificar, não permitindo detetar espécies não esperadas e não sendo por isso aplicável na detecção de fraudes ou de contaminações cruzadas de ocorrência desconhecida. Por outro lado, a sequenciação de ADN permite detetar as espécies de uma forma totalmente inequívoca e imparcial, e sem informação prévia. A aplicação desta técnica iniciou-se com a tecnologia de Sanger que durante mais de duas décadas foi o método de eleição para sequenciar genomas. A capacidade de gerar dados é no entanto limitada, bem como a sua aplicação no caso de ocorrência de misturas de espécies. Nos últimos anos, assistiu-se a um enorme progresso em tecnologias de sequenciação de ADN, tendo sido várias as abordagens inovadoras que evoluíram para substituir o método de Sanger como o principal fornecedor dos dados de sequenciação. Destaca-se assim a mais recente tecnologia designada por sequenciação de nova Geração ou "Next Generation Sequencing" (NGS), desenvolvida para superar as limitações da tecnologia de Sanger, e permitindo uma abordagem holística ao problema da detecção/identificação de organismos. Com a tecnologia NGS, todo o ADN presente na amostra é sequenciado e, com recurso a ferramentas informáticas adequadas e ainda sujeitas a desenvolvimento, irá ser comparado com bases de dados de genomas de seres vivos. Desta forma, torna-se possível apurar quais as espécies presentes na amostra e, assim confirmar, ou não, a lista de ingredientes que compõe o alimento.

Inspecção e Tecnologia Alimentar

Paulo Costa

Desafios (antigos e novos) na inspeção sanitária o paradigma das aves de capoeira

ICBAS - Instituto de Ciências Biomédicas Abel Salazar
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Resumo

A evolução da inspeção sanitária dependerá muito da nossa capacidade técnica em dar corpo aos objetivos e métodos plasmados na legislação e, igualmente, da nossa capacidade em empreender uma revisão metódica de todas as contingências que ladeiam esta importante missão veterinária.

Abstract

A avaliação da segurança e da higiene dos géneros alimentícios (higiene pública veterinária) é uma das incumbências matriciais dos médicos veterinários. O crescimento das cidades no final do século XVIII, conjuntamente com o descontentamento popular face à capacidade dos facultativos (médicos) e dos cirurgiões para inspecionarem os géneros alimentícios de origem animal, reclamaram a criação de um corpo profissional mais vocacionado e competente para fazer "o exame sanitário de todo o gado, (...) inspeccionar o serviço de matança, (...) examinar toda a carne vísceras e miudezas, (...) dar parte diária do serviço a seu cargo", conforme expresso no primeiro regulamento normativo das funções dos médicos veterinários citado por Bárbara Pinto de Araújo em "Breves notas sobre a evolução da higiene pública alimentar".

Desde então, o bom desempenho dos médicos veterinários no fomento da produção pecuária e na inspeção dos produtos de origem animal tem sido fulcral na satisfação da mais vital das necessidades humanas: o acesso alimentos em quantidade suficiente e mais seguros para a sua saúde. Com as prateleiras dos supermercados repletas de géneros alimentícios e com o afastamento das comunidades urbanas em relação ao meio rural, a gratidão pelo desempenho deste corpo profissional tem-se vindo a desvanecer. Sem um reconhecimento social direto, é justo e necessário que exista um reconhecimento institucional para quem defende simultaneamente a saúde do homem e dos animais, a economia, o bem-estar animal e o ambiente. Tudo isto feito num contexto de desconforto vocacional (assistir ao abate de animais) e enfrentando um conjunto de dificuldades físicas inerentes a uma atividade realizada num ritmo frenético, em horário muitas vezes noturno, num ambiente ruidoso, frio, húmido e muito conspurcado.

Sem a objetividade da fome, a satisfação das necessidades alimentares da humanidade não representa um avanço civilizacional óbvio. Entretanto, ganham protagonismo todas as situações de risco que resultam da utilização excessiva de sal, hidratos de carbono e gorduras ou escassa de fibras, vitaminas e sais minerais; as quais são responsáveis por transtornos metabólicos e pela redução da esperança e qualidade de vida. Neste contexto, o Homem ocidental apela à ciência que baixe o teor calórico aos alimentos, suprima todos os riscos inerentes ao seu consumo, cesse de experimentar novos "venenos" e dissipe todos os efeitos nefastos associados à sua produção. Se é verdade que esta pretensão criou um sistema racional e eficiente para a avaliação, gestão e comunicação dos

riscos associados ao consumo de alimentos, também não deixa de ser verdade que, levada a excessos delirantes ou a omissões seletivas, estigmatizou ou submergiu, respetivamente, o papel dos intervenientes neste campo.

Hoje, sob a designação de Médico Veterinário Oficial, o veterinário faz o exame clínico dos animais conduzidos para o abate, avalia a sua condição higiénica e promove as melhores práticas para minimizar a sua aflição e sofrimento. Recebe dos veterinários assistentes das explorações um conjunto precioso de informações sobre a sua história clínica e sobre as condições de criação desses animais, nomeadamente os alimentos compostos fornecidos, os programas profiláticos e metafiláticos aplicados e os resultados zootécnicos alcançados. Todos estes elementos irão contextualizar as observações realizadas no âmbito do exame post-mortem; em minutos, ou numa fração de segundo, o veterinário integra toda a informação, recorda a lei e decide. Cabe ainda a este auditar as condições de processamento, transformação, distribuição dos produtos de origem animal bem como o tratamento de subprodutos e de águas residuais produzidos. Este vigilante noturno precisa de convicções firmes, parceiros responsáveis, independência e uma imensa competência para escrutinar os potenciais pontos de introdução, amplificação e controlo dos mais de cinco mil perigos sanitários veiculáveis através dos alimentos, repartidos entre moléculas químicas, agentes biológicos e perigos físicos. Tem, em contraste com outros domínios da veterinária, objetivos de trabalho muito precisos, erigidos em torno do interesse comum, selados numa pilha de diplomas legais que identifica claramente os caminhos permitidos e interditos. Sem espaço para o relativismo, importa melhorar a realidade: a que existe e a que deveria existir.

Os veterinários são o único grupo profissional com funções específicas na segurança alimentar. Existe porém uma enorme pressão para que as muitas competências necessárias à prossecução deste objetivo deixem de estar disponíveis e dispersas por diversos grupos profissionais (veterinários, nutricionistas, microbiologistas, engenheiros zootécnicos, engenheiros alimentares, biotecnologias, técnicos veterinários, enfermeiros veterinários, etc.) e se concentrem numa profissão específica, reconhecida institucionalmente para o desempenho transversal de todas as tarefas a empreender no campo da segurança alimentar.

Não valorizar devidamente estas dinâmicas nas nossas práticas e competências aumenta a nossa vulnerabilidade no confronto com outras formações, e agrava o risco de acantonamento definitivo dos veterinários nos matadouros. Seria uma perda para os veterinários, e para a sociedade, não acompanhar de uma forma convicta a estratégia plasmada na profunda reforma do quadro legal europeu para os alimentos, consagrando em substância a ligação entre o abastecimento alimentar (food security) e a segurança dos alimentos (food safety) do "prado" ao "prato". Nesse novo paradigma esbatem-se as fronteiras entre o clínico de espécies pecuárias e o inspetor sanitário. Afigura-se evidente que o domínio e a integração de todas estas competências não se farão por intermédio de um qualquer prodígio veterinário, mas sim através trabalho pluridisciplinar realizado por um grupo de profissionais bem preparados e irmanados na linguagem e nos valores éticos.

Inspeção sanitária de carnes de aves de capoeira

Uma cadência de abate extremamente elevada – que pode alcançar 12 000 aves/hora – determina que a equipa de inspeção sanitária disponha apenas de 0,3 segundos para observar a superfície das carcaças e das miudezas, realizar quaisquer avaliações suplementares através da palpação ou incisão e, no imediato, tomar uma decisão sanitária informada.

Mesmo com muito apreço pela capacidade dos médicos veterinários e seus auxiliares, não é possível aceitar que tal feito seja possível, pelo menos à luz dos métodos tradicionais empregues na inspeção sanitária de ungulados domésticos.

Acontece que esta inconciliabilidade entre a mecanização do processo de abate (e concomitante aceleração do mesmo) e o exame individual das carcaças de aves de capoeira obrigou, ad initio, a importantes alterações metodológicas: (i) a avaliação sanitária deve incidir sobre o grupo de animais (bando), (ii) o controlo e verificação devem ser realizados ao longo de toda a cadeia produtiva, devendo (iii) ser responsabilizados todos os intervenientes que, através da sua ação, possam evitar quaisquer riscos.

É ao abrigo desta abordagem que o exame em vida na exploração de origem assume notável preponderância. Neste espaço, o médico veterinário pode consultar os preciosos registos da exploração (e.g. mortalidade, crescimento, consumo alimentar e de água, exames realizados, vacinas e medicamentos administrados), observar as condições de criação (infraestruturas e maneio, contemplando o aquecimento, ventilação, iluminação, distribuição de alimento e água, estado da cama e densidade vital), apreciar as medidas de biossegurança praticadas na exploração e examinar in loco o comportamento e a distribuição das aves, a uniformidade do bando, o desenvolvimento corporal, o nível de emplume e de pigmentação das patas, a higiene exterior e o estado clínico das aves.

Todo este manancial de informação estará presente no momento da realização do exame post mortem, contextualizando tudo aquilo que possa estar a ser observado e, como tal, dando maior travamento lógico àquilo que o médico veterinário venha a decidir.

Acresce que a realização do exame em vida na exploração de origem permite ao médico veterinário definir a ordem de abate segundo critérios sanitários e higiénicos, mitigando os incontornáveis riscos de contaminação cruzada durante o abate, particularmente ao nível das operações de escaldão, depena e evisceração. No presente, a ordem de abate é sobretudo condicionada pelos resultados obtidos no programa nacional de controlo de salmonela.

Finalmente, para além de ajudar a prevenir a contaminação da linha de abate, a realização do exame em vida na exploração de origem possibilita uma racionalização dos recursos humanos envolvidos no exame post mortem das carcaças. Muito naturalmente, bandos com desvios na performance produtiva ou com registo de episódios clínicos deverão ser alvo de um reforço na avaliação. Em termos formais, a legislação prevê que a velocidade da linha de abate possa ser reduzida de forma a cumprir este objetivo. Porém, na prática, este princípio conflitua com os compromissos comerciais do operador, com a eficiência mecânica dos equipamentos de abate (e.g. atordoamento e depena) e, mais problemático, pode comprometer o cumprimento dos requisitos higiénicos e de bem-estar animal plasmados nos regulamentos. Neste contexto, possuir informações que ajudem a definir atempadamente os recursos humanos a alocar às tarefas inspetivas permitirá a realização de um exame proporcional ao risco, dando cumprimento ao preceito claramente expresso no Regulamento (CE) nº. 854/2004: "O pessoal oficial envolvido deve ser em número suficiente para que possam ser cumpridos todos os requisitos."

Toda esta argumentação sustenta aquele que poderá ser considerando o primeiro princípio na inspeção sanitária de aves de capoeira: privilegiar o exame em vida, na exploração de origem, como forma de conhecer o verdadeiro estado de saúde de um bando.

O segundo princípio – não infringir qualquer sofrimento desnecessário aos animais – é um valor matricial dos veterinários, independentemente da espécie, contexto produtivo ou valor económico do animal. A defesa do bem-estar animal tem sido reforçada com legislação bastante pormenorizada sobre as condições de criação, manipulação, transporte, contenção e atordoamento de animais destinados a abate. Nos países economicamente mais prósperos, há muito que despontou uma consciência social de repúdio pelos métodos de exploração animal excessivamente intensivos e artificiais. Uma justa preocupação em virtude da importância destes animais para o abastecimento alimentar e no fomento das exportações, sendo os mesmos um sustentáculo do bem-estar económico destas sociedades. Ironicamente, num contexto de economia aberta, o efeito poderá ser o inverso, mediante a redução da competitividade da produção pecuária nacional (vinculada ao cumprimento de normas de bem-estar

"mais restritivas") e pelo aumento na necessidade de importar produtos a partir de países que não incorporam algum destes princípios na sua produção animal.

Estes paradoxos são igualmente visíveis no momento da aplicação prática de algumas das regras de bem-estar animal. Por exemplo, num cais de receção de aves vivas de um centro de abate é recomendável que se reduza a intensidade da luz para um valor inferior a 5 lux e que se usem apenas focos de luz azul (menos percetível pelas aves). Este requisito técnico visa reduzir a agitação e o stress das aves enquanto aguardam a sua entrada na linha de abate e, como tal, defender o seu bem-estar. Tem igualmente um impacto positivo no repouso das mesmas e na reposição do glicogénio a nível muscular (essencial para uma maturação normal das carnes). Porém, uma fraca intensidade luminosa conflitua com obrigações de natureza sanitária (e.g. realização do exame em vida e avaliação da higiene exterior das aves que, por sua vez, condiciona a própria ordem de abate que, também, poderá estar em conflito com o bem-estar, se a ordem definida por critérios de natureza sanitária estiver em oposição à recomendável em função da ordem de chegada ou da distância percorrida por cada lote de aves). Noutra perspetiva, a baixa intensidade luminosa poderá estar em contradição com a própria defesa do bem-estar das aves, em virtude de não haver luz suficiente para uma observação adequada das aves e do seu comportamento enquanto aguardam o abate, obstaculizando a aplicação de quaisquer medidas corretivas. Este requisito técnico tem, igualmente, reflexos negativos na saúde do pessoal encarregue de manipular as jaulas e de suspender as aves na linha de abate: as regras de higiene e segurança no trabalho preconizam que tarefas com riscos para a integridade física do trabalhador devam ser realizadas em condições de iluminação adequadas. Também não se poderá considerar benéfico para a saúde de qualquer pessoa a exposição à obscuridade durante horas consecutivas. Adicionalmente, nestas condições de trabalho, será mais difícil uma manipulação precisa e adequada das aves, bem como a sua monitorização pela equipa de inspeção. Igualmente comprometido fica o requisito sanitário de não introduzir na linha de abate aves dentes, mortas por causa estranha e, ironicamente, aves feridas.

O terceiro princípio basilar da inspeção sanitária de aves de capoeira contempla os objetivos do exame após o abate, podendo ser formulado nos seguintes termos: no exame post mortem, qualquer órgão, ou outra parte da carcaça, afetado por um processo inflamatório deve ser rejeitado, e se existirem repercussões sistémicas, ou se existirem evidencias de ser provocado por agentes infeciosos transmissíveis ao homem, toda a carcaça deve ser rejeitada.
Uma vez mais, a aplicação desta regra poderá estar em conflito com a elevada cadência de abate e, consequentemente, com o escasso tempo para a apreciação. Como tal, a legislação autorizou que o pessoal envolvido nas operações de abate pudesse assistir os controlos oficiais, desempenhando determinadas funções específicas, sempre sob a supervisão do veterinário oficial. Esta medida deve ser entendida como um reforço da capacidade operacional da equipa de inspeção oficial, multiplicando os olhos e braços atentos a qualquer inconformidade ou suspeita e, por essa via, reequilibrando as limitações subjacentes à redução do tempo para observar e identificar inconformidades. Adicionalmente, este pessoal encontra-se distribuído por todo o espaço físico do matadouro, aumentando assim, o campo de vigilância da equipa de inspeção.

Emerge, porém, uma questão tão sensível quanto óbvia: poderemos reconhecer a estas pessoas capacidade e independência para um desempenho satisfatório no auxílio das tarefas inspetivas? A resposta seria um rotundo não se a sua intervenção visasse substituir algum dos membros da equipa oficial. Porém, a legislação é clara ao determinar o seu carácter auxiliar, prescrever a necessidade de serem formados de forma adequada e de serem sujeitos a testes de desempenho regulares e, finalmente, desta estratégia somente ser possível num matadouro que funcione segundo padrões de higiene satisfatórios e que aplique um código de boas práticas de higiene e HACCP há pelo menos 12 meses. Ou seja, é necessário que o operador também esteja sintonizado no mesmo objetivo que norteia a ação do serviço de inspeção oficial: proteger a saúde animal e humana, independentemente do fato de o poder estar a fazer por motivações meramente comerciais.

O quarto, e último, grande princípio decreta que o inspetor deve julgar em conformidade, devendo permitir todos os procedimentos que visem recuperar a maior quantidade de carne possível, desde que tal não represente perigo ou a possibilidade de introdução de carne contaminada na cadeia alimentar humana. Para tal é necessário que o médico veterinário tenha conhecimentos técnicos, experiência, bom senso e acesso a meios complementares de diagnóstico e de monitorização da higiene e segurança. No seu conjunto, estes predicados garantem um desempenho satisfatório da sua missão, uniformidade de critérios e um acrescento na interação com todos os intervenientes na cadeia alimentar: operadores, colegas, técnicos, manipuladores.

Pedro Vieira

Identificação e movimentação animal

A identificação e o registo (movimentação, nascimentos, morte, desaparecimentos) animal são suportados pelas comunicações efetuadas pelos detentores de animais sobre o Sistema Nacional de Informação e Registo Animal (SNIRA).

O SNIRA estabelece as regras para a identificação, registo e circulação dos animais das espécies bovina, suína ovina, caprina e equídeos.

A Direção Geral de Alimentação e Veterinária (DGAV) é a entidade responsável pela definição da informação necessária ao funcionamento do SNIRA, sendo o Instituto de financiamento de Agricultura e Pescas (IFAP) a entidade responsável pela gestão informática das bases de dados que integram aquele sistema.

A identificação e registo animal no SNIRA têm por base:

1. O registo de explorações;
2. A identificação dos animais (marcas auriculares, tatuagem, identificadores eletrónicos)
3. O registo das ocorrências na exploração no "Registo de Existências e Deslocações" (RED);
4. Movimentação através de guias de circulação (em vida, para abate, morte na exploração);
5. As declarações de existências (suínos, ovinos e caprinos).

As comunicações destas ocorrências são declarativas e efetuadas pelos detentores dos animais obrigatoriamente sobre a aplicação informática que suporta o SNIRA, diretamente via "web" ou através das organizações de agricultores protocoladas com o Estado Português no âmbito do SNIRA. Os detentores de animais devem comunicar e fazer registar na Base de Dados SNIRA as ocorrências acima identificadas até 7 dias corridos após a verificação dos factos.

No caos dos animais mortos na exploração deverá o produtor contactar o Sistema de Recolha de Cadáveres (SIRCA) nas doze horas seguintes ao conhecimento da morte. O contacto é efetuado via telefone.

Os detentores dos animais deverão manter em arquivo, pelo menos durante três anos, toda a documentação de suporte ao registo efetuados e disponibilizá-la, se necessário, às autoridades competentes quando por estas solicitado.

Identificação e Movimentação de Bovinos.

Os animais são identificados individualmente por duas marcas auriculares, uma em cada orelha, com um número único iniciado pelo código de país, PT para Portugal, até 20 dias após o nascimento.

Sempre que se verifica a queda de uma Marca Auricular deverão ser apostas novas marcas auriculares com o mesmo número único que as originais.

Além das marcas auriculares é emitido um passaporte individual para cada bovino com informação referente às características do animal (identificação, sexo, data de nascimento, raça, identificação a mãe e outras informações não obrigatórias), a exploração de nascimento e resultado de rastreio sanitário executado na exploração. O passaporte deve ser atualizado sempre que o animal é transferido de detentor/exploração, devendo também ser aposta informação referente aos rastreios sanitários efetuados.

As movimentações em vida e para abate são efetuadas diretamente sobre a BD SNIRA, através do sistema iDigital (www.ifap.pt), pelos detentores previamente ao transporte dos animais.

A aplicação informática valida o movimento referente aos animais em termos da posse dos mesmos, em termos sanitários e do transportador.

Os detentores de origem e destino/matadouro validam o registo dos animais na BD SNIRA até ao prazo de 7 dias.

A informação referente a todo o efetivo bovino, entradas e saídas da exploração deverão estar refletidos no livro de registo de existências e deslocações de bovinos (RED bov).

Identificação e Movimentação de Suínos

Os suínos deverão ser marcados através de tatuagem ou marca auricular na orelha direita, o mais cedo possível, pelo menos até ao desmame, e sempre antes de saírem da exploração. A esta marcação pode ser acrescentada uma marca no dorso ou na anca, ou uma identificação eletrónica.

Os detentores de Suínos devem ter atualizado o registo de existências e deslocações de suínos (RED Suínos), que refletirá o número de suínos existentes diariamente na exploração. O RED Suínos deve ser atualizado mensalmente.

Os documentos de circulação para abate são emitidos, "on line", pelo produtor diretamente sobre BD SNIRA, ou através das Organizações de Agricultores. A movimentação efetuada em lote com especificação de identificação individual dos suínos positivos à doença de Aujesky.

A movimentação em vida é efetuada com recurso a guias sanitárias de trânsito, disponíveis nos serviços oficiais e emitida pelos detentores de suínos protocolados para o efeito com a DGAV, tendo em consideração as condicionantes impostas pelo rastreio da exploração à doença de Aujesky.

Os detentores de suínos declaram, periodicamente, de 4 em 4 meses, e sempre antes de iniciarem atividade, as - Declaração de Existências de Suínos, "on –line" diretamente ou através das Organizações de Agricultores.

Identificação e Movimentação de Ovinos e Caprinos

Os pequenos ruminantes são identificados individualmente até aos 6 meses de idade, ou 9 meses em explorações extensivas, através de uma marca auricular convencional e de um identificador eletrónico (bolos reticular, ou brinco).

Os animais destinados a abate com uma idade inferior a 12 meses podem ser identificados com uma marca auricular contendo o código de exploração de nascimento.

Sempre que se verificar a queda de uma marca auricular os animais serão reidentificados com uma nova identificação, devendo ser efetuada a rastreabilidade da mesma na BD SNIRA.

As movimentações em vida e para abate são efetuadas diretamente sobre a BD SNIRA, através do sistema iDigital (www.ifap.pt), pelos detentores previamente ao transporte dos animais.

A aplicação informática valida o movimento referente aos animais em termos da posse dos mesmos, em termos sanitários e do transportador. Os detentores de origem e destino/matadouro validam o registo dos animais na BD SNIRA até ao prazo de 7 dias.

Identificação e Movimentação de Equídeos

Todos os equídeos nascidos em Portugal ou introduzido livremente em Portugal devem ser identificados.

A identificação de equídeos pressupõe:

- Um passaporte individual onde conste a identificação do animal através de um número único e Universal – Universal Equine Life Number (UELN).
- Um método que identifique o passaporte e o animal, através do resenho gráfico e descritivo e de identificador eletrónico.
- Registo de informação do animal e do seu detentor numa base de dados informatizada.

Em Portugal a base de dados informatizada onde se efetua a gestão de identificação de equídeos é o "Registo Nacional de Equídeos" (RNE)que efetua validações sobre o SNIRA relativamente às explorações e aos detentores dos animais.

O UELN é constituído por quinze dígitos em que os primeiros três identificam o código de país, 620 Portugal, os três dígitos seguintes identificam a BD RNE (001), 8 dígitos que identificam individualmente o animal e por ultimo um dígito de controlo.

Os equídeos são identificados como animais registados, em livro genealógico ou em "studbook" e em animais de produção e rendimento, todos aqueles que não se encontram registados. Em Portugal estes passaportes distinguem-se pela cor azul e pela cor verde, respetivamente.

A movimentação de equídeos em vida e para abate efetua-se apenas com recurso ao passaporte do animal.

Trocas Intracomunitárias

Para todas as espécies objeto de trocas intracomunitárias é necessário para além dos documentos de identificação e circulação, um certificado sanitário emitido pelo TRACES, sistema informático desenvolvido pela Comissão Europeia.

Exportações

As exportações de animais são objeto de certificação sanitária especifica de acordo com os requisitos estabelecidos por cada país.

Conclusão

O SNIRA é ferramenta essencial para monotorização e controlo de saúde e do bem-estar animal, bem como das políticas de apoio à agricultura.

A rastreabilidade dos animais poderá ser assim assegurada promovendo a confiança dos consumidores sobre os alimentos de origem animal colocados no mercado.

O SNIRA é considerado e utilizado pelos produtores como ferramenta de apoio à gestão das suas explorações pecuárias.

Fontes:

1. Decreto-Lei n.º 142/2006 de 27 de julho; Decreto-Lei n.º81/2013 de 14 de junho; Decreto-Lei 123/2013 de 28 de agosto.

Marta Borges

Regulamentação dos materiais e objetos em contacto com os alimentos e dos aditivos alimentares

Materiais e objetos em contacto com os alimentos

Quando um género alimentício entra em contacto com um material de qualquer natureza, verifica-se uma interação entre eles. Ocorre uma absorção de constituintes do género alimentício pelo material e, fundamentalmente, uma migração dos constituintes do material para o género alimentício, pois que a inércia química total não existe. Todavia, em muitos casos, ela é mínima e desprezável.

Considera-se materiais e objetos destinados a entrar em contacto com os alimentos são toda e qualquer superfície que esteja em contacto com os géneros alimentícios ou que a isso se destinem, todos os tipos de embalagem, louça de mesa e de cozinha, tubagens, depósitos, mesas de trabalho e a maquinaria e equipamento para processar alimentos.

Regulamento (CE) n.º 1935/2004, relativo aos materiais e objetos destinados a entrar em contacto com os alimentos, tem como objetivo garantir o funcionamento eficaz do mercado interno no que respeita à colocação no mercado comunitário de materiais e objetos destinados a entrar direta ou indiretamente em contacto com os alimentos, constituindo simultaneamente a base para garantir um elevado nível de proteção da saúde humana e dos interesses dos consumidores.

A este regulamento está subjacente o princípio segundo o qual qualquer material ou objeto destinado a entrar em contacto direto ou indireto com os alimentos deve ser suficientemente inerte para excluir a transferência de substâncias para os alimentos em quantidades susceptíveis de representar um risco para a saúde humana ou de provocar uma alteração inaceitável na composição dos alimentos ou uma deterioração das suas propriedades organoléticas. Assim os materiais e objetos devem ser fabricados cumprindo as boas práticas de fabrico definidas no Reg. nº 2023/2006.

O Reg. 1935 /2004 inclui disposições genéricas, previsão da publicação de disposições específicas (Regulamentos ou Directivas), disposições de rotulagem (indicações obrigatórias), disposições relativas à Declaração de conformidade (comercialização), regras de rastreabilidade e de Inspecção e medidas de controlo por parte dos Estados-Membros.

Os materiais abrangidos pelo Regulamento nº 1935/2004 são diversos havendo alguns que já dispõem de requisitos específicos designadamente Plásticos (Reg. nº 10/2011) , Cerâmicas (Decreto-Lei nº 190/2007 que transpõe a Diretiva 2005/31/CE) , Materiais e objetos ativos e inteligentes (Reg. nº 450/2009).

Os Materiais e objetos ativos destinam-se a alargar a validade dos alimentos e os Materiais e objetos inteligentes destinam-se a monitorizar o estado dos alimentos.

Aditivos Alimentares

Aditivo alimentar é qualquer substância não consumida habitualmente como alimento em si mesmo e normalmente não utilizada como ingrediente característico na alimentação, cuja adição intencional ao alimento tem objetivo tecnológico determinado na fase de fabrico, transformação, transporte (etc.), e que ela própria ou os seus derivados se tornam direta ou indiretamente um componente desses alimentos;

Os aditivos alimentares estão regulamentados pelo Reg 1333/2008, que incorpora num REGULAMENTO ÚNICO os corantes, edulcorantes e miscelânea de aditivos. A maioria das provisões estão em aplicação desde 20/01/2010. Cria um procedimento legal de autorização de aditivos mais simplificado e Estabelecimento de programa de re-avaliação para os aditivos existentes.

O regulamento prevê Listas comunitárias de aditivos alimentares autorizados que estão definidos na lista comunitária constante do anexo II relativa aos aditivos podem ser colocados no mercado enquanto tais e utilizados nos géneros alimentícios nas condições de utilização nele especificadas e na lista comunitária constante do anexo III relativa aos aditivos podem ser utilizados nos aditivos alimentares, nas enzimas alimentares e nos aromas alimentares nas condições de utilização nele especificadas.

Nas listas está definido o teor de utilização para os aditivos alimentares que é fixado no nível mais baixo necessário à obtenção do efeito desejado, de acordo as disposições relativas à utilização dos aditivos no alimentos do Regulamento .

Os requisitos relativos aos Materiais em contacto e aos Aditivos são alvo de controlo pelas autoridades competentes, os quais a nível da agro-industria são realizados pela DGAV e pelas Direções Regionais da Agricultura. São requisitos complexos, muito exigentes quer relativamente à sua aplicação pelos operadores, quer para os técnicos que realizam as ações de controlo. Inerentes às ações de controlo está uma importante tarefa de comunicação com os operadores económicos e a fundamentação da necessidade dos requisitos legais , o que promove a melhor aplicação das regras, papel que os técnicos no terreno tem vindo a desenvolver com sucesso.

Ana Martins

Métodos moleculares ao serviço da deteção de agentes patogénicos de origem alimentar

As doenças transmitidas por alimentos são um importante problema de saúde pública em todo o mundo. Embora seja difícil estimar a incidência global de doenças transmitidas pelos alimentos, especialmente nos países em desenvolvimento, verifica-se um aumento significativo da incidência de doenças transmitidas por alimentos foram relatados a nível mundial. Os agentes patogénicos de origem alimentar incluem bactérias, vírus, fungos e parasitas e o desenvolvimento de doença vem associado ao consumo de alimentos ou água contaminada por estes agentes ou pelas suas toxinas.

Por quase um século, a detecção de agentes patogénicos bacterianos baseou-se exclusivamente em métodos convencionais microbiologia para o seu isolamento e detecção. No entanto o diagnóstico alimentar foi e continua a ser uma tarefa desafiadora devido à complexidade e heterogeneidade das várias matrizes alimentares. Os avanços científicos conduziram ao desenvolvimento de uma série de técnicas moleculares com um enorme impacto na detecção de agentes patogénicos transmitidos pelos alimentos. Comparativamente com os métodos microbiológicos, as técnicas baseadas na análise de moléculas de DNA, RNA e/ou proteínas têm maior sensibilidade e especificidade e não dependem unicamente dos meios de cultura para enriquecer, seleccionar e detectar patogénicos bacterianos nos alimentos.

O uso de técnicas moleculares possibilita também a detecção de toxinas e outros factores de virulência, via detecção dos genes codificantes (pela técnica de PCR) ou pela detecção directa da toxina por métodos imunológicos. É possível assim inferir sobre o potencial patogénico dos agentes bacterianos identificados. Como exemplos específicos podem ser referidas as verotoxinas de E. coli, a enterotoxina estafilocócica, toxina emética do Bacilus cereus, micotoxinas, toxinas botulinicas, toxinas ***α β ε í*** do clostridium perfringens entre outras.

Por outro lado a técnicas moleculares permitiram a abertura ao universo dos agentes patogénicos virais como os Norovírus agentes etiológicos das gastroenterites vírias e do vírus da Hepatite A, até então virtualmente indetectáveis.

Adélia Pereira

Papel do Médico Veterinário Municipal

O Médico Veterinário Municipal tem a sua atividade regulada pelo Decreto-Lei nº 116/98 de 5 de maio. Este diploma determina que estes profissionais são a autoridade sanitária veterinária concelhia a nível da respetiva área geográfica de atuação.

O exercício do poder de autoridade sanitária veterinária concelhia traduz-se na competência de, sem dependência hierárquica, tomar qualquer decisão, por necessidade técnica ou científica, que entenda indispensável ou relevante para a prevenção e correção de fatores ou situações suscetíveis de causarem prejuízos graves à saúde pública, bem como nas competências relativas à garantia de salubridade dos produtos de origem animal.

Os Médicos Veterinários Municipais (MVM) têm o dever de colaborar com o Ministério da Agricultura, Florestas e Desenvolvimento Rural (MAFDR), na área do respetivo município, em todas as ações executadas nos domínios da saúde e bem-estar animal, da saúde pública veterinária, da segurança da cadeia alimentar de origem animal, da inspeção hígiossanitária, do controlo de higiene da produção, da transformação e da alimentação animal e dos controlos veterinários de animais e produtos provenientes das trocas intracomunitárias e importados de países terceiros.

As relações funcionais dos MVM com o MAFDR são asseguradas através das Direções Regionais de Alimentação e Veterinária e da articulação destas com a Direção Geral de Alimentação e Veterinária.

Os médicos veterinários municipais dependem, hierárquica e disciplinarmente, do presidente da câmara da respetiva área da sua intervenção. Em caso de concorrência de obrigações entre o serviço do ministério da agricultura e o serviço municipal, prevalece o serviço municipal.

A par das competências técnicas e científicas inerentes à profissão médico-veterinária a legislação é uma ferramenta de trabalho para estes profissionais.

Descriminam-se seguidamente alguns diplomas legais relevantes na atividade dos MVM:

- Decreto-lei n.º 28/84, de 20 de Janeiro, com as respetivas alterações e republicações, refere-se a infrações antieconómicas e contra a saúde pública.
- Regulamento (CE) n.º 852/2004, do Parlamento Europeu e do Conselho, de 29 de Abril, estabelece as regras relativas à higiene dos géneros alimentícios.
- Regulamento (CE) n.º 853/2004, do Parlamento Europeu e do Conselho, de 29 de Abril, estabelece as regras específicas de higiene aplicáveis aos géneros alimentícios de origem animal.
- Regulamento (CE) nº 1069/2009 do Parlamento Europeu e do Conselho de 21 de Outubro de 2009 define regras sanitárias relativas a subprodutos animais e produtos derivados não destinados ao consumo humano.

- Decreto-lei nº 147/2006, de 31 de julho, aprova o Regulamento das Condições Higiénicas e Técnicas a Observar na Distribuição e Venda de Carnes e Seus Produtos.
- Decreto-lei 113/2006, de 12 de Junho, estabelece as regras de execução, na ordem jurídica nacional, dos Regulamentos (CE) nº 852/2004 e 853/2004, do Parlamento Europeu e do Conselho, de 29 de Abril, relativos à higiene dos géneros alimentícios e à higiene dos géneros alimentícios de origem animal, respetivamente.
- Lei n.º 92/95, de 12 de Setembro, com as respetivas alterações e republicações, refere-se à proteção aos animais.
- Decreto-lei n.º 276/2001, de 17 de Outubro, com as respetivas alterações e republicações, estabelece as normas legais tendentes a pôr em aplicação em Portugal a Convenção Europeia para a Proteção dos Animais de Companhia.
- Decreto-lei n.º 313/2003, de 17 de Dezembro, aprova o Sistema de Identificação e Registo de Caninos e Felinos (SICAFE).
- Decreto-lei n.º 314/2003, de 17 de Dezembro, aprova o Programa Nacional de Luta e Vigilância Epidemiológica da Raiva Animal e Outras Zoonoses (PNLVERAZ) e estabelece as regras relativas à posse e detenção, comércio, exposições e entrada em território nacional de animais suscetíveis à raiva.
- Portaria n.º 421/2004, de 24 de Abril, aprova o Regulamento de Registo, Classificação e Licenciamento de Cães e Gatos.
- Portaria n.º 422/2004, de 24 de Abril, determina as raças de cães e os cruzamentos de raças potencialmente perigosos
- Decreto-Lei n.º 315/2009, de 29 de Outubro, com as respetivas alterações e republicações, aprova o regime jurídico da detenção de animais perigosos e potencialmente perigosos enquanto animais de companhia
- Portaria n.º 968/2009 de 26 de Agosto, estabelece as regras a que obedecem as deslocações de cães, gatos, pequenos roedores, aves de pequeno porte, pequenos répteis e peixes de aquário, que sejam animais de companhia, em transportes públicos.
- Lei n.º 69/2014, de 29 de Agosto, procede à trigésima terceira alteração ao Código Penal, aprovado pelo Decreto-Lei n.º 400/82, de 23 de setembro, criminalizando os maus tratos a animais de companhia, e à segunda alteração à Lei n.º 92/95, de 12 de setembro, sobre proteção aos animais, alargando os direitos das associações zoófilas.
- Lei n.º 27/2016, de 23 de agosto, aprova medidas para a criação de uma rede de centros de recolha oficial de animais e estabelece a proibição do abate de animais errantes como forma de controlo da população.
- Decreto-Lei n.º 59/2003, de 01 de abril, alterado pelo Decreto-Lei n.º 104/2012, de 16 de maio. Transpõe para a ordem jurídica nacional a Diretiva n.º 1999/22/CE, do Conselho, de 29 de Março, relativa à detenção de animais da fauna selvagem em parques zoológicos, estabelecendo as normas para a manutenção e bem-estar dos animais, o licenciamento e inspeções dos parques, a gestão das coleções, a promoção de estudos científicos, a salvaguarda da biodiversidade e a educação pedagógica dos visitantes.
- Decreto-Lei n.º 255/2009, de 24 de Setembro alterado pelo DL n.º 260/2012, de 12 de Dezembro, relativo ao estabelecimento das condições de polícia sanitária aplicáveis à circulação de animais de circo e outros números com animais entre Estados membros, e aprova as normas de identificação, registo, circulação e proteção dos animais utilizados em circos, exposições itinerantes, números com animais e manifestações similares em território nacional.
- Decreto-Lei n.º 81/2013 de 14 de junho, aprova o novo regime do exercício da atividade pecuária (NREAP).
- Decreto-Lei 184/2009 de 11 de Agosto, estabelece o regime jurídico aplicável ao exercício da atividade dos centros de atendimento médico-veterinários e os respetivos requisitos quanto a instalações, organização e funcionamento.
- Decreto-Lei n.º 38 382 de 7 de Agosto de 1951, aprova o Regulamento geral das edificações urbanas.
- Decreto-Lei n.º 4/2015, de 07 de Janeiro, aprova o novo Código do Procedimento Administrativo

Apesar da pesada carga legal que serve de base à atuação destes profissionais, as características sociais, demográficas, físicas, económicas, políticas e culturais de cada município levam a que os desafios diários enfrentados pelos MVM possam ser muito distintos em diferentes municípios.

Tal como em outras áreas da Medicina veterinária também os MVM são confrontados com a necessidade de atualização constante para suportar as suas decisões. Recentemente a consciencialização social das necessidades dos animais de companhia coloca como áreas emergentes de atuação a medicina veterinária forense, o comportamento animal e a medicina de abrigos.

Ricardo N. Pereira, José A. Teixeira and António A. Vicente

Ohmic heating – an emergent technology in milk pasteurization

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Abstract

Ohmic heating is nowadays considered one of most promising and emergent technologies in thermal processing, favoring the technological and nutritional quality of several food products such as the case of pasteurized milk for human consumption.

Introduction

Physical and chemical changes occur in milk after thermal processing affecting its nutritional, organoleptic and functional properties Different types of thermal processing protocols are applied to milk with different purposes such as: sterilization, for preservation longer than 5 months (Raynal-Ljutovac, Park et al. 2007); thermization, which is generally used to kill temperature-sensitive microorganisms, such as psychrotrophs, and by this means reduce the micro-flora during low-temperature storage; ultra-high temperature (UHT), in which temperatures above 100 ºC are applied being successful in extending shelf life of milk but also leading to organoleptic changes; and pasteurization, typically applied in the range of 60 - 80 ºC for a few minutes or seconds, depending on the treatment temperature. Pasteurization aims a reduction of the number of a high variety harmful microorganisms or undesirable enzymes to a level that they do will not constitute a significant health hazard. Currently, two methods of pasteurization are still in use in the dairy industry, the low temperature long time (LTLT) and the high temperature short time (HTST) processing (Dumalisile, Witthuhn et al. 2005). LTLT is an almost a disused batch process adopted only by small rural dairy farms and laboratories. This "old-fashioned" method is being replaced by continuous HTST pasteurization. However, for an effective m pasteurization, it is still considered that milk should be heated in properly operated equipment to 62.8 ºC for 30 min (LTLT pasteurization) or 71.7 ºC for 15 seconds (HTST pasteurization). Traditional pasteurization of milk is carried out using HTST process in plate-and-frame heat exchangers (PHE). These are widespread in dairy industry for heating and cooling applications offering high degrees of compactness and effectiveness (Bansal and Chen 2006, Ghosh, Sarangi et al. 2006). In these units the milk and the heating medium (i.e. superheated steam or hot water) are separated by a pack of metal plates pressed together being the heat transferred to the product by conduction and convection mechanisms. These indirect heating treatment methods often result in the deposition of milk solids – commonly known as fouling - on the inner surfaces of heat exchangers and pipe work (Kastanas, Lewis et al. 1995). This phenomenon is particularly enhanced through the use of indirect heat transfer technologies due to the greater contact between the product and the heating hot surfaces. Fouling of heat exchanger surfaces by milk is still a major problem experienced by the dairy industry (Simmons,

Jayaraman et al. 2007), because addresses not only technological issues but also economical and quality losses, namely: increases pressure drop which can affect the economy and efficiency of the processing plant; it reduces heat transfer efficiency compromising the safety of the food product, once the process fluid is not heated properly

(Bansal and Chen 2006); the deposits composed mainly of proteins and fat that are often dislodged by the flowing fluid can cause microbiological contamination; it requires cleaning operations procedures which can be time consuming and lead to significant increases in maintenance costs. Food product composition such as pH, mineral content, presence of fat and protein composition also influence occurrence of fouling. In case of the milk one of main role in fouling is played by heat denaturation of whey proteins, in particular of *β*-lactoglobulin, which is one of the most thermo-labile protein found in milk. In reason of these problems the development of new and emergent technologies for continuous thermal dairy food treatment is being encouraged. Currently, Ohmic Heating (OH) technology, also known by electro-heating, direct resistance heating or Joule effect, is raising a great industrial and scientific interest

Ohmic heating, called Joule heating or electro-heating, is one of the earliest applications of electricity in food thermal processing and is defined as a process where electric currents are passed through foods with a purpose of heating them. During OH heat is internally generated due to electrical resistance inherent to food products (de Alwis and Fryer 1990). Ohmic heating technology has gained interest recently because the products are of a superior quality to those processed by conventional technologies (Castro, Teixeira et al. 2003, Pereira, Pereira et al. 2007). Given its volumetric and direct way of heating together with absence of hot surfaces, it is possible to overcome the process limitations and reduce the problems associated with overheating and fouling for example. Moreover, the OH assembly can be easily incorporated into a complete product sterilization or cooking process line. Among the advantages claimed for this technology the most outstanding are the uniformity of heating and improvements in quality with minimal structural, nutritional or organoleptic changes. The major benefits of this technology are as follows:

I. Temperature for HTST processes can be achieved very quickly;
II. Suitable for continuous processing without hot transfer surfaces;
III. Uniform heating of liquids with fast heating rates;
IV. Reduced problems of surface fouling and overheating;
V. Fresher-tasting products;
VI. No residual heat transfer after the current is shut off;
VII. Useful in pre-heating products before canning;
VIII. Low maintenance costs (no moving parts) and high energy conversion efficiencies (> 90%);
IX. Environmentally friendly system.

Considering the specific characteristics and advantages of this technology it is expected to high-quality pasteurized milk with minimal structural, nutritional, or organoleptic changes in a short operating time. The objective of this work is to review some of the most striking aspects of OH on quality of pasteurized milk. Microbiological aspects, as well as physical and chemical properties of milk related with protein and lipid composition will be addressed.

Results and Discussion

Microbiological Aspects

In OH the mechanisms of microbial inactivation are mainly thermal but the presence of alternating moderate electric field with and an associated electrical frequency should not be neglected. Recent research is claiming that a mild electroporation (i.e. disruption of cell membranes that can be reversible or irreversible, depending on intensity) may take part during OH treatment and thus contribute to cell damage or lysis, bringing a non-thermal and synergistic effect to inactivation. Among several microorganisms Escherichia coli is considered a reliable indicator of faecal contamination generally found in milk and other dairy products in unsanitary conditions (Diliello 1984). Regarding death kinetics of Escherichia coli artificially inoculated in milk, results show that with OH the time required for a given

thermal treatment can be reduced thus confirming a non-thermal killing effect over vegetative cells (Pereira, Martins et al. 2007). Table 1 shows the death kinetics curve parameters for Escherichia coli during conventional and OH performed under identical thermal profiles. D-values correspond to the time needed to cause 90% of inactivation in a microbial population at a given temperature. While z-value is the temperature increase required to reduce D-value by 90%. OH offers a great potential for shorter and less aggressive thermal treatments without compromising food safety.

Table 1.
Death kinetic parameters of Escherichia coli in milk under conventional and Ohmic thermal treatments

Kinetic Parameters	Temperature (°C)		
	55	63	65
Conventional Heating			
D-value (min)	10.9 ± 1.8	3.9 ±0.5	3.5 ± 0.2
z-value (°C)		23.1 (R2=0.98)	
Ohmic Heating			
D-value (min)	14.2 ± 0.2	1.9 ± nd	0.86 ± nd
z-value (°C)		8.4 (R2=0.99)	

2.2. Physical and Chemical Characterization

2.2.1. Free Fatty Acids

In general, main physical and chemical features milk did not change with application of OH. When ohmic and conventional pasteurization operations were compared under identical thermal profiles, properties such as pH, total solids, ash content, titratable acidity and total fatty acids did not change significantly ($p<0.05$) (Pereira, Pereira et al. 2007). In particular, it was expected that OH and its electric fields may cause physical disorders in milk fat globules during pasteurization resulting in an increase of free fatty acids (FFA) and development of rancid off-flavors due to bacterial lipases, which are thermo-resistant at the temperatures of pasteurization and ultra-high temperature treatments, but this was not confirmed. Table 2 shows the FFA composition of untreated raw milk, and milk pasteurized by PHE heat exchanger and an ohmic heater. OH did not impose modifications to general composition of FFA of short and medium chain thus indicating that this technology can be introduced in milk pasteurization without affecting negatively its quality (Pereira, Martins et al. 2008).

Table 2.
Mean relative percentage weight of FFA in different goat milk samples

	FFA composition (%)			
Treatment	Butyric ($C_{4:0}$)	Caproic ($C_{6:0}$)	Caprylic ($C_{8:0}$)	Capric ($C_{10:0}$)
Raw milk	24.49 ± 1.08^{a}	19.67 ± 0.75^{a}	19.80 ± 0.19^{a}	36.04 ± 1.43^{a}
Heat exchanger	22.82 ± 0.73^{b}	18.67 ± 0.38^{a}	19.98 ± 0.17^{a}	38.53 ± 1.08^{b}
Ohmic heater	23.36 ± 0.71^{b}	18.99 ± 0.44^{a}	20.15 ± 0.23^{a}	37.51 ± 1.22^{a,b}

$^{a-b}$ means in columns followed by the same letter are not significantly different ($p > 0.05$)

2.2.2. Milk Proteins

Excessive denaturation of whey proteins during thermal treatment of milk, together with a consequent loss its nutritional value, may give rise to the formation of deposits on heat exchangers (i.e. fouling) or affect functional for technological properties of milk for manufacture of cheese (i.e. increased rennet clotting time and low gel strength). β-Lactoglobulin (β-LG) constitutes more than 50% of total whey protein in bovine milk, presenting a high nutritional and biological value (i.e., digestibility, amino acid pattern, and sensory characteristics) and is mainly responsible for the gelation and emulsification properties of whey derivatives (such as whey ingredients Whey Protein Concentrate and Whey Protein Isolate). The effects of OH on denaturation of whey proteins have been extensively studied over the last decade (Pereira and A. 2009, Pereira, Teixeira et al. 2011, Rodrigues, Martins et al. 2015, Pereira, Rodrigues et al. 2016). It was possible to conclude that OH induces less whey protein denaturation thermal treatments at temperatures ranging from 75 to 90 °C. When indirect and ohmic heating were compared under identical thermal profiles, significant differences ($p < 0.05$) were found between the native total contents of whey at for short time thermal processing at 85 °C. Figure 1 shows contents of native whey proteins measured through High-Liquid Performance Chromatography (HPLC); in particular after 30 s at 85 °C, WPI solutions treated by OH presented more 8 and 10% of soluble native and β-Lg and α-Lactalbumin (α-La), respectively. The effects of indirect heating (conventional) and OH on casein protein fraction of milk, heated at temperatures between 75 and 85 °C was also evaluated. In this case, it was noticed that the size increase of casein micelles during OH treatment was less dependent from heating temperature, when compared with a conventional thermal process ($p < 0.05$). Increase in particle size of casein may result from denaturated whey protein deposition on micelle surface. Consequently, the less increase of micelle size during OH may be related with the lower levels of whey protein denaturation. Figure 2 shows the size increase of casein micelle at different temperatures.

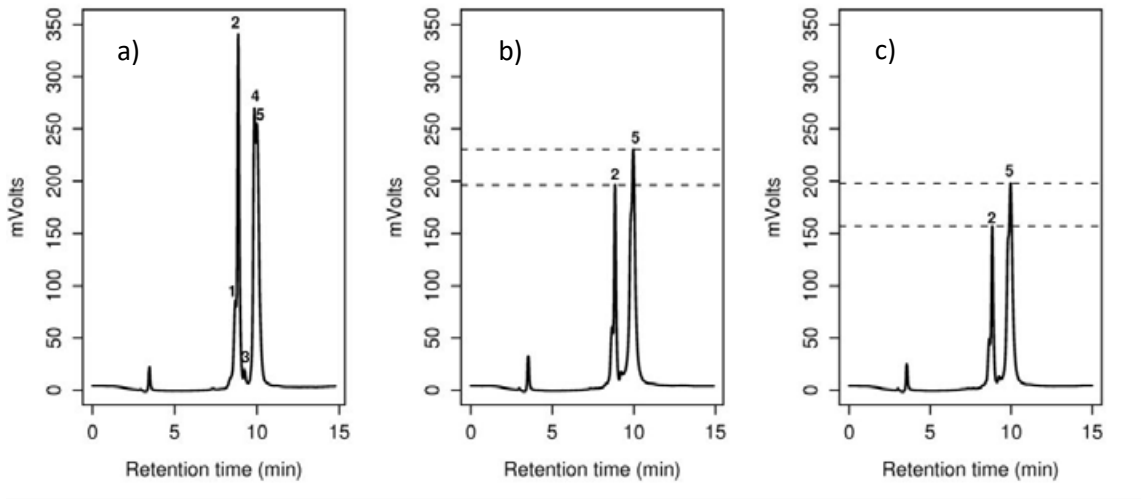

Figure 1. Examples of HPLC chromatograms obtained for unheated (a), and heated WPI samples at 85 °C for 30 s through ohmic (b) and conventional heating (c): [1] BSA; [2] α-La; [3] Immunoglobulins (IG); [4] β-LgB; and [5] β-LgA.

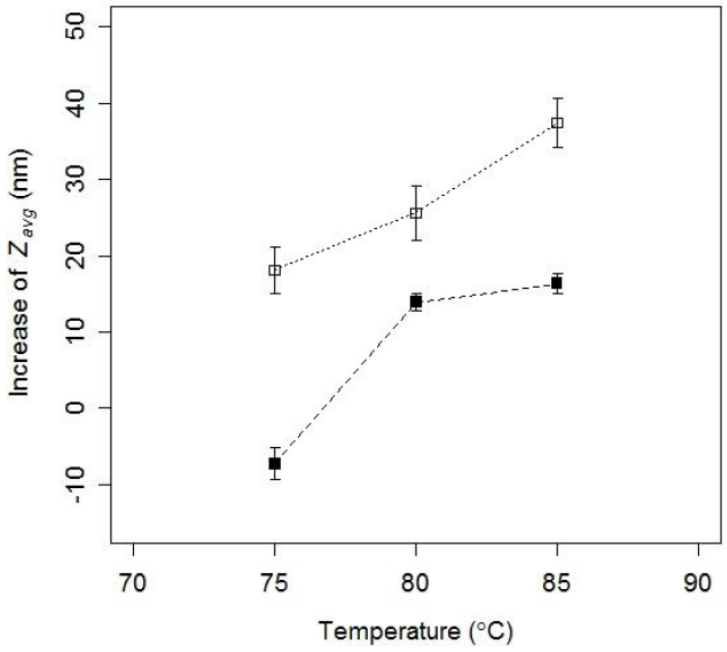

Figure 2. Effect of heating temperature on casein micelle average size (Zavg), measured by dynamic light scattering technique, after 30 minutes of holding time at different temperatures through conventional heating (white squares) and OH (black squares).

Conclusions

OH technology opens a new paradigm for the high temperature short time thermal processing of foods, such as milk and its derived products. It has been demonstrated that even in a worst case scenario (when increase of temperature is made slow) OH is still less aggressive than indirect heating methods. When compared with conventional thermal processing OH heating offers the potential to reduce whey protein denaturation levels and maintain higher stability of casein micelle size. Further, microbiological safety is also enhanced due to the combination of an effective binomial time-temperature treatment with and non-thermal effect due to the presence of moderate electric fields contacting the food.

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Painéis Científicos

Avaliação da qualidade do ar em estabelecimentos de restauração e bebidas universitários em Portugal

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Os objetivos deste estudo consistiram em avaliar a população de bactérias e fungos no ar interior de estabelecimentos de restauração e bebidas (ERB) de campus universitários do Norte de Portugal.

Foi utilizada a metodologia passiva de sedimentação das partículas em placa para avaliação da população de bactérias e fungos, fazendo a comparação da população entre as diferentes zonas: preparação, laboração, armazenamento e empratamento de cada ERB e entre os ERB localizados nos campus universitários diferentes (L1 e L2). Foi identificado o género de fungos, a presença de cocos ou bacilos e afinidade Gram. No total dos ERB em estudo, verificou-se a presença de cocos Gram-positivos (45,8%), cocos Gram-negativos (4,5%), bacilos Gram-positivos (21,50%) e bacilos Gram-negativos (22,60%). A avaliação da qualidade do ar através das contagens fúngicas em placa, revelou que os ERB analisados apresentaram no geral um nível satisfatório ou aceitável. No entanto, foram observados níveis insatisfatórios de acordo com o Decreto-Lei nº 79/2006 (> 500 UFC/m^3) para o total de Eumyces em cantinas no L2. Ainda, os resultados indicaram maiores contagens fúngicas em L2 (p<0,05) e em cantinas (p<0,05). Os géneros mais comuns de fungos isolados foram *Acremonium*, *Cladosporium*, *Aspergillus* e *Penicillium* em L2 e os géneros *Rhizomucor*, *Rhizopus* e *Epicoccum* em L1. O género *Penicillium*, um dos envolvidos na deterioração dos alimentos e frequentemente associado a sintomas respiratórios alérgicos, foi isolado em L2 e L1 representando 92% e 4% dos casos, respetivamente. O género *Fusarium* sp., que possui características patogénicas, foi isolado no campus L2, em valores superiores nas cantinas (51 UFC/m3) e inferiores nos bares (39 UFC/m3), tendo sido predominante nas zonas de lavagem, de preparação de produtos crus e de confeção. A vertente legislativa não menciona quaisquer critérios de avaliação no que concerne às espécies fúngicas identificadas, sendo reforçada pelo diploma legal (NT-SCE-02, 2009) que determina o parâmetro de não conformidade quando se verifique a existência de crescimento visível de fungos em qualquer superfície ou a concentração fúngica no interior superior à detetada no exterior. Nos casos em que se verifique a condição anterior, deve-se ter atenção: 1) a presença de espécies pouco comuns, desde que as misturas de espécies pouco comuns seja ≥ 150 UFC/m3 e uma só espécie pouco comum seja ≥ 50 UFC/m3; 2) a presença confirmada de *Aspergillus fumigatus*, *Stachybotrys* spp. ou outros fungos toxigénicos ou patogénicos. Porém no diploma apenas se refere como comuns: *Cladosporium* spp., *Alternaria* spp. e *Penicillium* spp..

Com este estudo, salienta-se a importância de conhecer de forma qualitativa e quantitativa os géneros de fungos no ar nomeadamente em ERB universitários que servem grandes quantidades de refeições diárias. A informação obtida permitiu implementar um sistema útil de avaliação e controlo do ar (tais como, manter uma humidade relativa inferior a 70% e uma temperatura não superior a 20 ºC e a instalação nos ERB mais afetadas de filtros de ar de alta qualidade.

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Avaliação de hipertensão pulmonar em cães com doença mixomatosa da válvula mitral

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Objectivos

Determinar a prevalência de hipertensão pulmonar em canídeos com doença mixomatosa da válvula mitral (DMVM) e avaliar se o rácio átrio esquerdo:aorta (AE:Ao) e se a classificação de acordo com American College of Veterinary Internal Medicine (ACVIM) estão associados à presença de hipertensão pulmonar em canídeos com DMVM.

Métodos

Foram incluídos 31 canídeos. A realização do exame ecocardiográfico foi precedido de uma abordagem à história pregressa do animal e um exame sumário do estado geral. Os canídeos foram divididos em dois grupos com base na ausência (grupo 1.1) e presença (grupo 1.2) de hipertensão pulmonar. Todos foram classificados de acordo com a classificação de ACVIM.

Através de ecocardiografia transtorácica foi avaliado o rácio AE:Ao e o diagnóstico de hipertensão pulmonar foi baseado na presença de regurgitação da válvula tricúspide ≥2,8 m/s. A pressão arterial pulmonar sistólica foi calculada através da equação de Bernoulli modificada tendo sido os canídeos posteriormente classificados de acordo com a gravidade da hipertensão pulmonar.

Resultados

O grupo 1.1 foi constituído por 28 canídeos (22 pertenciam à classe B (78,6%) e 6 à classe C (29,0%)) e o grupo 1.2 por 3 canídeos (classe C).

A prevalência obtida de hipertensão pulmonar foi de 9,7% numa amostra de 31 canídeos.

Foi encontrada uma associação estatisticamente significativa entre as classes B e C entre os grupos, sugerindo que a hipertensão pulmonar possui tendência para se desenvolver com a progressão da doença. Todavia, não foi encontrada uma diferença estatisticamente significativa do rácio AE:Ao entre o grupo 1.1 e 1.2.

Os canídeos do grupo 1.2 apresentaram hipertensão pulmonar classificada como ligeira.

Conclusões

A prevalência obtida de hipertensão pulmonar em cães com DMVM sugere que a evidência ecocardiográfica de hipertensão pulmonar não é um achado raro em canídeos com DMVM, podendo representar um papel importante na progressão desta doença.

A gravidade da hipertensão pulmonar associada à DMVM foi ligeira. No entanto, esta manifestou-se em canídeos com insuficiência cardíaca congestiva direita sugerindo que deve ser suspeita principalmente nestas situações.

A avaliação da presença de hipertensão pulmonar deve fazer parte do exame ecocardiográfico num canídeo com DMVM uma vez que já existem terapias dirigidas à hipertensão pulmonar arterial.

Avaliação dos níveis de proteína c reativa em cães com doença periodontal

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A doença periodontal é um processo inflamatório do periodonto bastante prevalente em cães e associado a um estadio crónico de inflamação sistémica, podendo este ser alterado após tratamento adequado. A proteína C reativa (CRP) é uma proteína de fase aguda utilizada por rotina como diagnóstico de doença sistémica em humanos e em cães, os seus níveis séricos já foram avaliados na doença periodontal.

Este estudo teve como objetivo determinar os efeitos sistémicos da doença periodontal através do doseamento da CRP sérica e avaliar o impacto do tratamento periodontal em cães com a doença, comparando os níveis de CRP com cães saudáveis e sem doença periodontal (grupo controlo).

A população estudada incluiu cães que foram submetidos a um exame físico e estomatológico-dentário detalhados, análises hematológicas e radiografias intraorais. Trinta e nove cães (22 machos e 17 fêmeas) foram selecionados e divididos em dois grupos: (i) grupo controlo constituído por 28 animais com idade inferior a 2 anos e sem qualquer doença inflamatória, incluindo doença periodontal e (ii) grupo com doença constituído por 11 animais com mais de 2 anos de idade e com doença periodontal. O doseamento da CRP sérica foi realizado com recurso ao kit Canine-CRP (Randox, República da Irlanda). No grupo controlo, obteve-se uma amostra de sangue e no grupo com doença, duas amostras (uma antes e outra 30 dias após o tratamento periodontal).

Diferenças significativas (p=0,001) foram encontradas entre o grupo controlo (2,947 ± 1,428 mg/L, 95% intervalo de confiança (CI)) e pré-tratamento do grupo com doença (13,824 ± 17,273 mg/L, 95% CI), mas entre o grupo controlo e pós-tratamento do grupo com doença (3,376 ± 1,235 mg/L, 95% CI) não se verificou. Os valores de CRP entre o grupo controlo e pós-tratamento foram significativamente mais baixos do que os obtidos no grupo pré-tratamento. Não foi observada associação estatística entre o peso vivo e o estadio da doença (p=0,130; CI 95%), bem como entre a raça e o estadio da doença (p=0,114; CI 95%). Não foi também identificada qualquer relação da idade com o estadio da doença (p=0,28; CI 95%).

O doseamento de CRP em cães com doença periodontal foi avaliado em estudos anteriores, incluindo pré e pós-tratamento periodontal. Contudo, os valores de CRP reportados na presença de doença periodontal nunca foram comparados com um grupo de controlo. Este estudo permitiu caracterizar a associação entre a doença periodontal e o estadio inflamatório sistémico associado. A análise utilizada neste estudo foi de elevada especificidade para o cão, permitindo uma determinação mais precisa dos valores séricos, sendo que, de acordo com o conhecimento dos autores, não foi utilizada anteriormente em estudos clínicos semelhantes.

Em conclusão, os níveis de CRP estão aumentados em cães com doença periodontal e existe um efeito mensurável no impacto inflamatório sistémico do tratamento periodontal, quando comparado com um grupo control.

Este estudo reforça a importância da tratamento periodontal no controlo da doença no cão, no sentido de reverter o seu impacto sistémico.

Avulsão do plexo braquial em cães: estudo descritivo de nove casos clínicos

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Objectivos

A avulsão traumática das raízes nervosas do plexo braquial é a causa mais comum de monoparesia ou monoplegia em animais de companhia, sendo o atropelamento e a queda em altura as causas mais comuns. Com base nos sinais clínicos, no exame neurológico e nos estudos electrodiagnósticos é possível, não só elaborar o diagnóstico, como localizar a que nível se encontra a lesão e emitir o prognóstico.

Métodos

Este estudo consistiu na descrição de nove casos clínicos em cães observados na Clínica Ani+ entre 2010 e 2011 e na Clínica Referência Veterinária entre 2011 e 2015 e com diagnóstico de avulsão do plexo braquial. O objectivo deste trabalho foi o de caracterizar os dados epidemiológicos da população estudada, a etiologia da doença, a apresentação clínica e alterações no exame neurológico, bem como os dados obtidos nos exames complementares de diagnóstico e a evolução da doença.

Resultados

Após a análise dos dados obtidos, verificou-se que a maior parte dos animais em estudo eram cães machos, sem raça definida, com menos três anos e com peso vivo inferior a 15 Kg. Quase todos os animais tinham história de traumatismo, sendo o atropelamento a etiologia mais frequente. Os sinais clínicos observados e os resultados do exame neurológico foram semelhantes entre os cães em estudo. Constatou-se assim que, de uma forma geral, estes apresentavam monoparesia ou monoplegia, com aparecimento súbito e não progressivo, reacções posturais e reflexos espinhais diminuídos ou ausentes e ausência de sensibilidade profunda no membro torácico afectado. A anisocoria e reflexo do músculo cutâneo do tronco ausente ipsilateral à lesão foram registados na maior parte dos animais. Quanto ao tratamento, este consistiu, essencialmente, numa abordagem conservativa que incluiu a reabilitação física e a prevenção de feridas do membro afectado.

Conclusões

Este estudo permitiu constatar que a doença tem mau prognóstico em relação à recuperação funcional do membro, tendo apenas um animal recuperado totalmente a sensibilidade profunda e a capacidade funcional. Dos nove animais em estudo, três foram amputados, embora só se conheça o motivo desta decisão em um deles

Biodiversidade (também) parasitária em suínos de produção biológica no minho e no alentejo

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As infecções por parasitas gastrointestinais são frequentes em suínos em todos os regimes de produção, assim como a sua influência negativa na saúde e no peso vivo dos animais. O modo de produção biológico (MPB) tem crescido em Portugal e as restrições ao uso de fármacos, aliado ao facto de estes animais serem produzidos em regime extensivo, incentivou este estudo, cujo objectivo geral foi fazer um levantamento da diversidade e carga parasitária em suínos criados em MPB em Portugal.

Foram recolhidas amostras de fezes em cinco explorações, que foram devidamente rotuladas e armazenadas até serem analisadas no laboratório (técnicas de análise qualitativa - flutuação e sedimentação - e quantitativa).

Das 45 amostras analisadas, em 29 (64,4%) foram identificadas formas parasitárias, variando a percentagem de amostras positivas por exploração entre 37,5% e 100%. Uma grande diversidade de formas parasitárias foram identificadas: *Balantidium coli* (22/45, 48,9%), *Strongylida* (21/45; 46,6%), *Ascaris suum* (21/45; 46,6%), *Coccidia* (8/45, 17,8%), *Trichuris suis* (4/45; 8,9%) e *Ascarops strongylina* (3/45; 6,7%). A Tabela 1 apresenta a média e o desvio padrão do número de ovos/oocistos por grama (OPG) de fezes de cada forma parasitária encontrada por exploração.

Tabela 1 – Média e desvio padrão do número de OPG de fezes por forma parasitária por exploração

	Balantidium coli		*Ascaris suum*		Strongylida		Coccidia		*Trichuris suis*		*Ascarops strongylina*	
	Média	Desvio Padrão	Média	Desvio Padrão	Média	Desvio Padrão	Média	Desvio Padrão	Média	Desvio Padrão	Média	Desvio Padrão
Alentejo (n=13)	150	133	3900	3356	800	5795	3575	10627	300	0	1750	1202
Minho 1 (n=8)	400	468	2000	1178	350	141	2200	2546	0	0	0	0
Minho 2 (n=8)	0	0	375	304	50	0	200	0	50	0	0	0
Minho 3 (n=8)	550	614	375	304	50	0	200	0	0	0	0	0
Minho 4 (n=8)	700	212	100	484	50	0	0	0	0	0	0	0

Os resultados evidenciam uma provável elevada contaminação ambiental com formas parasitárias que além do impacto negativo que têm na saúde animal e ambiental, tem também impacto na saúde pública pelo potencial zoonótico de algumas (*Balantidium coli*, *Ascaris suum* e *Ascarops strongylina*).

Biodiversidade parasitária gastrointestinal em ovinos da raça Churra Galega Mirandesa – dados preliminares

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O parasitismo gastrointestinal em ovinos produzidos em sistema extensivo pode ter um forte impacto na produtividade das explorações, nomeadamente em pequenas explorações familiares onde muitas vezes se pratica uma pecuária de sobrevivência. Esta, associada a alguns erros de maneio, leva à redução no peso vivo e predispõe os animais a outras doenças, o que é grave em áreas geográficas onde o acesso a cuidados veterinários é escasso. No Planalto Mirandês a criação de ovinos da raça Churra Galega Mirandesa (CGM) é realizada principalmente em pequenas parcelas com partilha de terrenos comunitários, favorecendo a contaminação constante das pastagens com formas parasitárias. Perante a evidência de que há poucos estudos sobre estes parasitas em ovinos de raças autóctones em geral e da raça CGM em particular, o objectivo deste estudo foi avaliar a diversidade de carga parasitária gastrointestinal dos animais desta raça.

Para o efeito, acompanhamos a Associação de Criadores da raça CGM e recolhemos até à data amostras de fezes de 9 explorações, dos concelhos de Miranda do Douro e Mogadouro, num total de 61 amostras. A amostragem foi de conveniência, contudo recolhemos um número mínimo de três amostras por exploração. Foram usados métodos coprológicos qualitativos (flutuação e sedimentação) e quantitativos (McMaster).

Foram identificadas formas parasitárias em 60 (98,4%) amostras, nomeadamente estrongilídeos (58/61; 95,1%), *Nematodirus* spp. (21/61; 34,4%), *Eimeria* spp. (48/61; 78,7%), *Moniezia benedeni* (1/61; 1,6%), *Eimeria intricata* (1/61; 1,6%), *Skrjabinema* spp. (1/61; 1,6%) e *Dicrocoelium* spp. (1/61; 1,6%). %). As cargas parasitárias (número de ovos por grama de fezes - OPG) dos estrongilídeos têm um valor mediano de 50, com um máximo de 1300, 55,7% dos valores estão entre 50 e 800 OPG, 1,6% entre 801 e 1200 OPG e 4,9% com mais de 1200 OPG correspondendo aos graus de infecção leve, moderado e grave, respectivamente.

Os resultados mostram uma grande biodiversidade nas formas parasitárias encontradas e uma carga parasitária aparentemente baixa o que sugere alguma capacidade de adaptação do parasita e uma resiliência que tão bem caracteriza estes animais também noutros contextos. Este estudo está a decorrer estando também a ser avaliadas também as práticas profilácticas dos produtores.

Bypass ureteral subcutâneo: estudo retrospetivo

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O principal objetivo deste estudo foi a avaliação retrospectiva dos resultados obtidos com a implantação do *Bypass* Ureteral Subcutâneo (SUB) para correção cirúrgica de obstruções ureterais no Hospital Veterinário do Restelo.

Foram incluídos dados de 35 intervenções em pacientes da espécie *Felis catus*, desde outubro de 2013 a março de 2016, inclusive. Foi recolhida informação a partir dos registos dos historiais médicos e os dados foram analisados utilizando o programa Excel.

De forma a incluir o máximo de dados possíveis o estudo foi dividido em vários períodos, cada um com os seus próprios critérios de inclusão e exclusão.

Obteve-se um total de 35 intervenções em pacientes com idade média de 8,2±3,4 anos. Não se encontrou predominância clara de nenhum dos sexos, contudo, a raça predominante foi a Europeu Comum (79%). Cerca de 1/3 dos pacientes necessitou de colocação bilateral do SUB. Comparativamente ao seu valor de entrada, no momento da alta clínica, cerca de 97% da totalidade dos casos diminuiu o valor sérico de creatinina. Foram necessários, em média, 5,2 dias de hospitalização pós-cirúrgica.

Tabela 1 - Taxas de complicações observadas

Período	Nº complicações direta/indiretamente relacionadas com a técnica	Nº total de complicações	% total de complicações
Cirúrgicas	0	3	8,6
Pós-cirúrgico imediato	10	14	29,4
Pós-cirúrgico retardado	7	14	43,3
Curto-prazo	2	3	12,0
Longo-prazo	15	18	47,8

As complicações observadas que poderão estar diretamente relacionadas com o SUB ncluíram, por ordem decrescente de importância: obstrução do sistema, disúria, líquido livre subcapsular, lavagens difíceis, líquido livre peri-renal, vinco do cateter de nefrostomia, portal de acesso virado para o lado interno, desconexão do cateter de nefrostomia do portal, incontinência urinária e hematúria.

Obteve-se uma taxa de mortalidade perioperatória de 8,6% (3 em 3 causas de morte foram sugestivas de complicações anestésicas) e até aos 6 meses de 16,7% (3 em 4 foram submetidos a eutanásia por questões monetárias).

O SUB é uma alternativa cirúrgica útil para resolução de obstruções ureterais, principalmente quando outros procedimentos falham, não estão disponíveis ou estão contraindicados, evitando lesão relevante do parênquima renal e permitindo descompressão ureteral eficaz com imediato abaixamento da azotémia.

Caracterização genética de giardia duodenalis em cães da região de Lisboa

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Giardia duodenalis é um protozoário unicelular que parasita o intestino de diversos mamíferos, incluindo o Homem. O número de infeções subclínicas é muito superior ao número de casos de doença. Apesar de aparentemente saudáveis, os cães portadores são capazes de transmitir o parasita ao Homem e outros animais.

O presente estudo visou a deteção de Giardia duodenalis em amostras fecais de cães oriundos da região de Lisboa, tendo como objetivo específico, a caracterização genética de sequências amplificadas através do uso de primers distintos, dirigidos para secções alvo dos genes que codificam as proteínas glutamato desidrogenase e β-giardina (bg), e do gene SSU-rDNA (pequena sub-unidade ribossomal).

Para tal, entre 2014 e 2015 foram analisadas 114 amostras com base na técnica coprológica de Willis modificada, tendo-se detetado por microscopia ótica, a presença de quistos de *Giardia* spp. em 13,2% (15/114) das mesmas. Obtiveram-se sequências parciais dos genes SSU-rDNA e bg em 86,7% (13/15) e 20,0% (3/15), respetivamente, das amostras positivas por microscopia. Doze das 13 sequências obtidas da amplificação parcial do gene SSU-rDNA e duas das três sequências obtidas da amplificação parcial do gene bg apresentaram elevada homologia com o genótipo D. O genótipo C foi detetado nas duas sequências remanescentes.

Este estudo vem demonstrar a presença de G. duodenalis em cães da região de Lisboa, e a existência de genótipos específicos da espécie canina. Estudos adicionais devem ser realizados de forma a avaliar quais os fatores de risco para a ocorrência de infeção canina, bem como levar a cabo uma caracterização genética do parasita multilocus.

Case report: Intoxicação por Vitis vinífera complicada por intussusceção intestinal

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Objectivos

O objetivo é descrever o primeiro caso de intussusceção jejuno-jejunal associado a uma intoxicação por Vitis vinífera,
mais concretamente uvas, e discutir possíveis fatores de risco para a sua ocorrência.

Métodos

Cão macho, 7 meses, 14,5 kg, labrador retriever, com história de ingestão de uvas há mais de 48h, seguida de anorexia, vómitos incoercíveis e diarreia líquida com uvas não digeridas. A quantidade de uvas ingeridas estimou-se em cerca de 1 quilograma. As analíticas sanguíneas revelaram uma insuficiência renal aguda com aumento da creatinina (8,7 mg/dL) e ureia (96 mg/dL) e presença de proteinúria.

Resultados

Foi instituída terapêutica de suporte gastrointestinal (ranitidina, sucralfato e metoclopramida), benazepril (5 mg) e fluidoterapia endovenosa com NaCl 0,9% (duas taxas de manutenção). Ao fim de 48h de internamento, repetiram-se as análises renais e os valores apresentados foram imensuráveis. Manteve-se o mesmo tratamento e ao 4.º dia de internamento a creatinina baixou para 2,1 mg/dL e a ureia para 65 mg/dL e os vómitos e diarreia foram controlados, suspendendo-se a metoclopramida. No entanto, a anorexia manteve-se e, ao exame físico, à palpação abdominal detetouse uma massa intestinal. Foram realizadas radiografias (Figura 1, 2 e 3) com e sem contraste baritado e ecografia abdominal (Figura 3 e 4), compatíveis com intussusceção intestinal. Realizou-se uma laparotomia exploratória com enterectomia da porção invaginada (jejuno com sinais de necrose) e uma anastomose entero-entérica, seguida de plicagem intestinal. No pós-operatório, privilegiou-se a analgesia e houve uma introdução gradual da água e alimento. 4 dias pós-cirurgia, os valores de creatinina (2,8 mg/dL) e ureia (50 mg/dL) mantinham-se alterados. O animal permaneceu internado mais 10 dias com monitorização constante dos parâmetros renais, normalizando a creatinina (1 mg/dL) e ureia (38 mg/dL). O animal teve alta com dieta renal e benazepril, mantendo-se sem sinais de novas intussusceções até à data.

Conclusões

Na literatura não existe descrição de um caso semelhante, por isso torna-se importante identificar possíveis fatores de risco que possam ter originado a intussusceção, que não podemos concluir com o relato de um único caso. Assim, é necessário realizar mais estudos epidemiológicos da toxicidade das uvas/taninos e da sua associação no desenvolvimento de intussusceção intestinal.

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Caso de uveíte na espécie acanthosaura capra

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Objectivos

Apesar de doenças oftálmicas serem comuns em répteis (sobretudo em tartarugas), a informação que existe publicada acerca de Uveítes e a sua progressão em répteis é pouco detalhada. De igual modo, apesar de existirem bastantes relatos médicos envolvendo espécies de lagartos, são bastante escassos os que referem a espécie *Acanthosaura capra*.

Métodos

Foi apresentado à consulta um lagarto da espécie *Acanthossaura capra*, macho, 5 anos de idade com queixas de anorexia e apatia que duravam há 1 semana. Apresentava várias dermatites na face ventral do abdomen e nas faces palmares dos membros anteriores, com formação de crostas de coloração amarelada. O globo ocular direito apresentava um depósito esbranquiçado na câmara anterior, sugestivo de hipópion, que por sua vez seria indicativo de um processo de uveíte. Foi também observada uma opacidade da córnea, sugestivo de edema. A amostra recolhida por zaragatoa ocular não permitiu qualquer conclusão citológica ou cultura bacteriana. Iniciou-se antibioterapia sistémica (Marbofloxacina 1% 10mg/kg q24h) e terapia oftálmica tópica (Prednisolona 1 gota q12h; Cloranfenicol 1 gota q12h). Devido ao estado geral do paciente (sobretudo a desidratação extrema) optou-se por não fazer anti-inflamatório sistémico. Foi instituída terapia de suporte (NaCl 0,9% SC 30ml/Kg/dia; Oxbow Carnivore Care PO 0,5g/dia).

Resultados

O animal foi reavaliado semanalmente, havendo melhorias ligeiras em cada reavaliação. Passados 28 dias, era visível uma diminuição clara do hipopion e o edema de córnea reduziu bastante. Passado um total de 42 dias desde o início do tratamento a única alteração visível era edema de córnea com um diâmetro bastante reduzido, pelo que se manteve apenas a administração do ntibiótico tópico.

Conclusões

A aplicação de antibiótico sistémico em conjunto com antibiótico e corticosteróide tópicos demonstrou ser eficaz na resolução de uveíte e edema de córnea nesta espécie. O edema de córnea poderá ser secundário a uma lesão endotelial resultante da uveíte, o que é corroborado pela sua boa resposta à medicação aplicada. Ao conhecimento dos autores, esta é a primeira descrição de uma uveíte na espécie *Acantosaura capra*.

Deteção de anticorpos anti-leptospira em soros de ouriços-cacheiros (erinaceus europaeus)

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Objectivos

A leptospirose é uma zoonose reemergente com distribuição mundial, que afeta o homem e animais (domésticos e selvagens), e é provocada pela infeção por espécies patogénicas do género *Leptospira*.

O habitat natural das leptospiras patogénicas é o rim, sendo a urina a principal via de excreção. Estas são mantidas na natureza pela adaptação de determinados serovares a espécies específicas de mamíferos, considerados hospedeiros de manutenção, nos quais o agente provoca uma resposta imunológica reduzida.

O ouriço-cacheiro (*Erinaceus europaeus*) assume um papel relevante como hospedeiro de manutenção de estirpes do serogrupo Australis na Europa, podendo ainda ser hospedeiro acidental para outros serovares, tais como Icterohaemorrhagiae, Canicola, Pomona, Bataviae e Grippothyphosa.

O trabalho realizado teve como objetivo conhecer os possíveis serovares circulantes na população de ouriços no Norte de Portugal.

Métodos

Efetuou-se a pesquisa de anticorpos anti-Leptospira em 15 amostras séricas de ouriços-cacheiros capturados na região (licença 452/2015/CAPT, ICNF), ou provenientes de centros de recuperação/parques de vida selvagem, no período de maio a outubro de 2015. Os soros foram analisados pela técnica de referência, o Teste de Aglutinação Microscópica, utilizando uma bateria de 14 serovares patogénicos de *Leptospira*.

Resultados

Foram detetados anticorpos em 53,3% (8+/15) dos soros analisados, os quais mostraram reatividade específica para 7 serogrupos diferentes: Pomona, Canicola, Australis, Autumnalis, Icterohaemorrhagiae, Grippotyphosa e Sejroe. As maiores taxas de reatividade foram observadas para os serovares Altodouro e Canicola. O título de anticorpos mais elevado foi de 1:800 e observado para o serovar Bratislava (serogrupo Australis).

Conclusões

Os resultados obtidos evidenciam uma taxa de seropositividade elevada e destacam-se as reatividades com os serovares Altodouro e Canicola. O novo serovar Altodouro foi isolado na região de Trás-os-Montes a partir de *Mus musculus*, não se conhecendo ainda se terá outros hospedeiros de manutenção no ecossistema. Devido à sua enorme plasticidade e adaptabilidade, o ouriço-cacheiro circula com facilidade entre zonas rurais e peri-urbanas, tornando-se uma fonte de fácil de infeção para outros animais selvagens e para animais de companhia mais curiosos. Pode constituir fonte de infeção por leptospiras para pessoas que os recolhem durante o período pós-hibernatório e para médicos veterinários que os manipulam nos centros de recuperação.

Determinação da frequência cardíaca e respiratória do cão através da técnica de fotopletismografia e descrição espetral do sinal

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Objectivos

Este estudo teve como objetivos determinar quais os elementos espetrais que constituem o sinal de FPG obtido no cão e avaliar a atividade cardíaca e respiratória durante a anesthesia pré-operatória.

Métodos

A população estudada incluiu quatro cães atendidos no Hospital Veterinário do Porto, entre novembro de 2015 e março de 2016, e que foram submetidos a uma pré-medicação anestésica através da administração intramuscular de dexmedetomidina (Dextomitor, Zoetis, EUA) na dose de 5 µg/Kg e metadona (Semfortan, Esteve, Espanha) na dose de 0,35 mg/Kg. O sensor de FPG, colocado na base da cauda após tricotomia da zona, foi ligado a uma placa hardware Bitalino (Plux, Portugal) conectada a um computador por via bluetooth de forma a visualizar *in loco* o sinal de pulso periférico.

O sinal foi analisado antes e depois da administração da pré-medicação e os dados foram recolhidos durante 3 a 5 minutos em ambos os momentos. Posteriormente, os dados obtidos foram analisados com recurso ao programa informático MatLab (MathWorks, EUA) e o sinal de FPG decomposto nas suas várias frequências com recurso à transformada de wavelet. Finalmente, foram analisados e comparados os valores de atividade cardíaca e respiratória, mínima e máxima, encontrados antes e depois da administração da pré-medicação anestésica.

Resultados

Neste estudo, foram encontradas diferenças significativas quando comparados os grupos referentes à atividade cardíaca (p = 0,043), sendo que nos restantes parâmetros, embora não tenha sido detetada nenhuma diferença significativa, detetou-se um decréscimo dos valores após a administração da pré-medicação. Na avaliação espetral no sinal de FPG, foram detetadas 7 ondas de frequência e estas caraterizadas segundo estudos já efetuados em humanos (Tabela 1).

Conclusões

A técnica de FPG de reflexão, embora já bastante descrita em Medicina Humana, ainda não foi analisada em animais de companhia. Assim sendo, este trabalho piloto permitiu contribuir para o conhecimento do espetro do sinal de FPG em Medicina Veterinária e permitiu avaliar as potencialidades que a técnica de FPG oferece na monitorização de sinais vitais no cão, como por exemplo, na monitorização ambulatória das frequências cardíaca e respiratória.

Diagnóstico e remoção de corpo estranho associado a trajetos fistulosos em dois cavalos

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Descrevem-se dois casos de cavalos apresentados ao HV-UTAD com história de feridas crónicas, com trajeto fistuloso, não responsivas ao tratamento com limpeza da ferida, aplicação de penso e antibioterapia. Em ambos os casos foi detetada a presença de um corpo estranho como causa da não cicatrização das feridas.

O primeiro animal apresentava um trajeto fistuloso proximalmente à articulação femuro-tibio-patelar direita, que se manteve após resolução de uma ferida profunda na mesma região. O segundo animal apresentava um ponto de supuração intermitente na face medial da articulação tarsometatársica esquerda.

Em ambos os animais foi necessário recorrer a estudos radiográficos e ecográficos da região da ferida para fins de diagnóstico. No primeiro animal, à radiografia, detetou-se um corpo estranho de radiopacidade semelhante à do osso nas projeções latero-mediais, tendo-se posteriormente realizado uma fistulografia, com solução de bário, para determinar a extensão do trajeto fistuloso e da loca onde o corpo estranho estava alojado. À ecografia era visível uma linha hiperecogénica, que provocava efeito de reverberação. A cirurgia foi realizada com o animal em estação, sob sedação profunda, tendo-se verificado que o corpo estranho era um fragmento de vidro. No segundo animal, não se detetou qualquer alteração à radiografia. À ecografia, no entanto, era visível uma loca com cápsula definida, contendo uma estrutura hiperecogénica com sombra acústica, compatível com um corpo estranho, rodeada por material anecoico. A cirurgia foi realizada com o animal sob anestesia geral, tendo-se apurado que o corpo estranho era um fragmento de madeira. Em ambas as cirurgias foi de grande utilidade a colocação de uma agulha adjacente ao corpo estranho, de forma ecoguiada, para servir de guia durante o desbridamento cirúrgico. Sem este procedimento, a pesquisa cirúrgica do corpo estranho seria mais difícil e traumática. Ambos os animais recuperaram sem complicações após a cirurgia.

A existência de feridas crónicas não responsivas ao tratamento médico convencional é comum, sendo a presença de corpos estranhos uma das possíveis causas subjacentes. Nestes casos deve ser realizada uma abordagem diagnóstica com recurso a técnicas imagiológicas para confirmar esta hipótese. A remoção do corpo estranho é, normalmente, suficiente para a recuperação completa do animal.

Doença do nariz branco: resultados preliminares da pesquisa do agente em morcegos dos distritos de Vila Real e Bragança

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Objectivos

A doença do "nariz branco" (WND) é uma doença fúngica emergente, que afeta morcegos durante a sua hibernação. A doença ocorre via colonização da membrana das asas, focinho e orelhas dos morcegos por um fungo psicrófilo de cor branca, denominado de *Pseudogymnoascus destructans*. A WND tem originado mortalidade em massa nas populações de morcegos que ocorrem na América do Norte e o seu carácter epidémico poderá mesmo provocar extinções regionais. Não obstante a ausência de registos de mortalidade significativa na Europa, este microrganismo é reconhecido a nível global como constituindo uma potencial ameaça para populações de morcegos hibernantes. Com o objetivo de conhecer a distribuição deste agente na região de Trás-os-Montes, realizou-se um rastreio da presença deste fungo em populações de morcegos dos Distritos de Vila Real e Bragança.

Métodos

Foram recolhidas amostras da membrana das asas, focinho e orelhas de 26 espécimes de morcegos, pertencentes às espécies *Myotis blythii*, *M. myotis*, *M. emarginatus*, *Rhinolophus ferrumequinum*, *R. hipposideros, R. euryale, Barbastella barbastellus, Plecotus austriacus*, presentes em hibernáculos da região, utilizando zaragatoas esterilizadas (Licença nº 05/2014/CAPT,ICNF). As amostras foram inoculadas em meios apropriados e os isolados identificados por métodos diretos. Os procedimentos de manipulação dos animais foram realizados no pleno respeito pelas "Guidelines of European Community Directive 92/43/EEC" (http://ec.europa.eu/environment/nature/legislation/habitatsdirective/index_en.htm) e "the Agreement on the Conservation of Populations of European Bats "(www.eurobats.org)".

Resultados

O isolamento de P. destructans ocorreu em 8,3% (5+/60) das amostras recolhidas de 26 morcegos. O agente foi isolado de dois exemplares de *M. blythii* em abrigos com duas localizações geograficamente diferentes (Mondim de Basto e de Aldeia de Montes, ambos no Distrito de Vila Real) e de um Rhinolophus sp também no abrigo da localidade de Mondim de Basto.

Conclusões

Dado o papel que as populações de morcegos desempenham no equilíbrio e dinâmica dos ecossistemas terrestres e o seu estatuto de conservação desfavorável (espécies em perigo ou criticamente em perigo em Portugal), é fundamental efetuar a epidemiovigilância de doenças que possam causar mortalidade em massa nestas populações. Os resultados obtidos confirmam a presença do *P. destructans* num abrigo com uma localização diferente daquela onde foi isolado pela primeira vez em Portugal, em 2013.

Doenças estomatológicas do gato diagnosticadas por histopatologia: estudo retrospetivo de 6 anos

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As lesões estomatológicas são frequentes no gato. Estas podem ser divididas, de acordo com a sua natureza, em doenças inflamatórias e doenças neoplásicas. A caracterização exata destas afeções deve basear-se no seu exame histopatológico.

O presente trabalho teve como principal objetivo determinar a frequência das doenças estomatológicas em gatos. Nesse sentido, foi realizado um estudo retrospetivo a partir dos relatórios de exames histopatológicos realizados nos laboratórios de Investigação Científica e Análises Moleculares (DNAtech, Lisboa) nos anos de 2010 a 2015. As variáveis recolhidas incluíram a raça, o sexo e a idade dos gatos, a localização das lesões na cavidade oral, a técnica de recolha utilizada, a natureza das lesões (inflamatória ou neoplásica) e o diagnóstico definitivo obtido.
De um total de 297 exames de lesões da cavidade oral, encontraram-se 186 lesões inflamatórias (62,6%) e 111 (37,4%) lesões neoplásicas, das quais 81,1% eram malignas.

As lesões estudadas foram mais frequentes em gatos do sexo masculino (173, 58,4%) e com idades entre os 7 e os 10 anos (88, 33,0%). Com o avançar da idade, observou-se um aumento das doenças neoplásicas e, pelo contrário, o diagnóstico de doenças inflamatórias diminuiu. As neoplasias benignas ocorreram tendencialmente em gatos mais jovens comparativamente com as neoplasias malignas. Relativamente à raça, predominaram os indivíduos de raça Europeu Comum (206, 73,6%), seguindo-se os de raça Persa (32, 11,4%) e de raça Siamesa (21, 7,5%), de entre um total de 9 raças puras observadas.

A gengiva foi a localização anatómica mais afetada pelas lesões estomatológicas com 127 casos (43,1%), seguindo-se a mucosa oral e os lábios. A biópsia incisional foi a técnica de recolha selecionada para a obtenção de 256 (86,2%) amostras, tendo 36 (12,1%) sido recolhidas por biópsia excisional e 5 (1,7%) com recurso a punch. De entre o total de diagnósticos histológicos registados, destacou-se o complexo gengivite-estomatite-faringite felino (115, 39,0%), o carcinoma espinocelular (49, 16,5%) e o complexo eosinofílico (34, 11,4%).

Este estudo permitiu contribuir para o conhecimento mais aprofundado da epidemiologia das doenças da cavidade oral do gato em Portugal.

Efeito da adição de bacteriocinas no controlo de listeria monocytogenes inoculada em queijo de ovelha

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Este trabalho tem como objetivo avaliar o efeito da adição de bacteriocinas na redução de *Listeria monocytogenes* inoculada em queijos de ovelha.

Procedeu-se ao fabrico de queijos de ovelha a partir de leite cru (3 lotes) obtido em explorações de Portugal, o qual foi transportado para o Laboratório TeQSA da UTAD, sob as adequadas condições de refrigeração. Cada lote de leite foi subdividido em 3 grupos: 1 grupo controlo (C) e 2 grupos para adição de bacteriocinas a 2 concentrações diferentes (B1; B2 em concentração 4 vezes superior a B1). Obtiveram-se assim queijos controlo e queijos fabricados com leites com bacteriocinas B1 ou B2, os quais foram por sua vez subdivididos em 6 grupos de queijos: 1) sem adição de bateriocinas mas inoculados com *L. monocytogenes* (controlo positivo); 2) sem adição de bateriocinas e não inoculados com *L. monocytogenes* (controlo negativo); 3) e 4) queijos apenas com bacteriocinas B1 ou B2; 5) e 6) queijos inoculados com *L. monocytogenes* e adição de bacteriocinas B1 ou B2 respetivamente, totalizando 108 amostras.

Logo após a produção dos queijos e antes do início da sua maturação, inoculou-se 80 µL de uma suspensão bacteriana de *Listeria monocytogenes* (5×10^5 UFC/g) em cada queijo inoculado, sendo depois armazenados a 8ºC. Foram feitas análises microbiológicas imediatamente após a inoculação e em vários tempos de armazenamento (2, 24, 72, 168, 336 e 672 horas) para contagem de *L. monocytogenes* em todas as amostras e de Enterobacteriaceae, bactérias do ácido lático (BAL) e mesófilos nas amostras controlo negativo. Nos queijos que continham bacteriocinas B1 não se verificou um efeito inibidor sobre estes microrganismos, uma vez que ocorreu o aumento da microbiota ao longo do tempo enquanto nos queijos com bacteriocinas B2 verificou-se um efeito inibidor ($p<0,001$), principalmente sobre *L. monocytogenes*, uma vez que ocorreu uma redução desta bactéria comparativamente aos queijos com a adição de bacteriocinas na concentração B1 e nas amostras apenas inoculadas com *L. monocytogenes*. Concluiu-se que a adição das bacteriocinas teve efeito antimicrobiano ($p<0,001$), sugerindo a sua potencial utilização como substâncias conservantes em alimentos suscetíveis à contaminação e multiplicação por *L. monocytogenes*.

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Endoscopia e ingluviotomia na remoção de corpo estranho numa arara azul e amarela (ara ararauna) evitando ventriculotomia

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Objectivos
Existem diferenças consideráveis na cirurgia do trato gastrointestinal realizada em aves ou em mamíferos. O esófago das aves é dividido apenas em duas regiões e ao contrário dos mamíferos é composto por quatro camadas, mucosa, submucosa, muscular e serosa. Não apresenta esfíncter superior e inferior. O estômago é constituído por duas câmaras, o proventrículo ou estômago glandular e o ventrículo ou estômago muscular.

A cirurgia do proventrículo e ventrículo apresenta grandes riscos e deve ser cuidadosamente ponderada. A ventriculotomia é geralmente evitada. O ventrículo possui paredes altamente musculadas e a atividade muscular fisiológica leva a uma frequente rejeição das suturas, tal como apresenta uma grande probabilidade de dificuldades de encerramento da ferida cirúrgica com padrões de inversão. As principais complicações incluem a perda da estanquicidade da sutura, deiscência e peritonite.

A remoção de CE por ingluviotomia é uma abordagem cirúrgica mais segura e simples. Como alternativa, a remoção do CE pode ser realizada por métodos não invasivos, utilizando-se endoscópios rígidos ou flexíveis.

Métodos
Vinte e quatro horas após a ingestão acidental de uma sonda de alimentação uma arara azul e amarela (ara ararauna), de 2 meses de idade, deu entrada no serviço de urgências hospitalares. A ave apresentava-se alerta, com apetite mas regurgitava. À palpação o inglúvio estava vazio. Pela apresentação clínica, optou-se pela abordagem não invasiva endoscópica, para a remoção da sonda. Ao entrar no inglúvio não se observou nenhum CE. Pelo flexibilidade e pequeno diâmetro do endoscópio (5.2 mm) e com a ajuda de uma pinça de crocodilo foi possível atingir o proventrículo e arrastar o CE até ao esófago torácico, no entanto, não foi possível a sua extração. O CE foi extraído por uma ingluviotomia simples.

Resultados
Neste caso a recuperação do CE por endoscopia foi apenas parcialmente bem-sucedida, no entanto, permitiu evitar a ventriculotomia.

Uma ingluviotomia também pode ser realizada para a introdução intraoperatória do endoscópio de forma a atingir a entrada do proventrículo e ventrículo.

Conclusões
A endoscopia, mesmo que não permita a extração completa do CE, pode ser utilizada como método complementar para evitar as técnicas cirúrgicas mais invasivas e com maior risco de complicações.

Download: CE após extração para esófago torácico.jpg
http://efomv.guc.pt/PaginaF.aspx?Tipo=ResumoSingle&Chave=000242EF4C&Janela=Nao

Estudo preliminar da ocorrência de anticorpos anti-*borrelia burgdorferi sensu lato* em javalis abatidos em montaria nos distritos de Vila Real e Bragança

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Objectivos
A borreliose de Lyme (BL) é uma zoonose emergente e com distribuição mundial, causada por bactérias do complexo *Borrelia burgdorferi* sensu lato, transmitidas através da mordedura de carraças do género *Ixodes*. Na Europa, o principal vetor é a espécie Ixodes ricinus, que faz a sua refeição sanguínea em inúmeros vertebrados domésticos e silvestres entre os quais o javali (*Sus scrofa*), cuja importância no ciclo epidemiológico da *B. burgdorferi* s.l. tem vindo a ser estudado nos últimos anos.

O principal objetivo foi aprofundar o conhecimento sobre a circulação destas espiroquetas no javali pela deteção de anticorpos anti-*Borrelia burgdorferi* s.l., e determinar os fatores de risco para a sua presença.

Métodos
Soros de 60 javalis abatidos em montarias nos distritos de Vila Real e Bragança (amostragem de conveniência entre os meses de outubro e fevereiro das épocas venatórias de 2012/13, 2014/15 e 2015/16) foram analisados por imunofluorescência indireta (IFA) para deteção de anticorpos anti-*Borrelia burgdorferi* s.l. (antigénio produzido a partir de culturas de estirpes de referência de *B. afzelii*, *B. garinii*, *B. lusitaniae* e *B. burgdorferi* sensu stricto). Os resultados obtidos foram analisados estatisticamente em função das variáveis distrito, sexo e idade, pelo método de regressão logística nominal e a força de associação entre as variáveis foi estimada pelo cálculo do *odds ratio* (OR), para um intervalo de confiança de 95% (JMP 10.0 Software SAS Institute).

Resultados
A taxa de seropositividade foi de 51,7% (31+/60), com títulos de anticorpos entre 1:64 e 1:256. Das variáveis estudadas apenas a idade mostrou diferenças significativas entre os grupos (P<0,05), podendo o aumento da idade constituir fator de risco para a ocorrência de aglutininas anti-Borrelia burgdorferi s.l. na população estudada (OR=2,975; 95%IC=1,036-8,545).

Conclusões
A elevada taxa de seroreatividade anti-*Borrelia burgdorferi* s.l. observada na IFA (elevada sensibilidade) é um forte indicador da importância do javali como potencial reservatório destas espiroquetas no ecossistema, para os humanos e outros animais (silvestres e domésticos), devido à partilha de habitat. Particularmente porque na mesma área geográfica, a presença do agente em carraças e javalis, já foi confirmada por métodos moleculares. Os caçadores e cães de caça estão particularmente expostos, pelo que estes resultados recomendam profilaxia contra o vetor, nomeadamente o uso de produtos repelentes (coleiras, pipetas, etc.), na área de estudo. A emergência da BL no Hemisfério norte requer o desenvolvimento de ferramentas epidemiológicas integrativas, alicerçadas no conceito "uma só saúde" no contexto espácio-temporal.

Factores de risco para colonização nasal por staphylococci meticilina- resistente em portadores humanos saudáveis em contacto diário com animais em Portugal

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Objectivos
Os objetivos deste estudo consistiram em investigar a frequência, persistência e fatores de risco associados a colonização nasal por *staphylococci meticilina*-resistente (MRS) em pessoas em contacto diário com animais.

Foram recolhidas zaragatoas nasais de 71 médicos veterinários, 34 estudantes (de Medicina Veterinária e Enfermagem Veterinária) e 24 enfermeiros/auxiliares veterinários. A pesquisa de MRS foi realizada em agar manitol com sal e em agar Brilliance™ MRSA2. A identificação das espécies foi obtida por PCR espécie-específico. A resistência à meticilina foi confirmada por PCR com amplificação dos genes *mecA* e *mecC*. As estirpes identificadas foram caracterizadas a nível molecular para identificação dos clones. Os fatores de risco foram determinados por regressão logística. As pessoas colonizadas por MRS foram contactadas para participarem no estudo de seguimento.

Dos 129 participantes, 79 (61%) estavam colonizados por, pelo menos, uma espécie de MRS [*S. epidermidis* (MRSE, n=68), *S. aureus* (MRSA, n=19), *S. haemolyticus* (MRSH, n=7), *S. pseudintermedius* (MRSP, n=2) e outros *staphylococci* coagulase-negativo (n=4)]. As linhagens clonais de MRSE, MRSA e MRSP detetadas correspondem aos clones mais frequentes nos animais de companhia em Portugal. Os profissionais veterinários (médicos veterinários e enfermeiros/auxiliares veterinários) tiveram maior probabilidade (P<0.0001, OR=6.369, [2.683-15.122]) de estarem colonizados por MRS que os estudantes. Contudo, a comparação entre os médicos veterinários e os enfermeiros/auxiliares veterinários não evidenciou diferenças significativas (P=0.7635). Foi também identificado como fator de risco o ter contactado com um animal positivo para MRSA ou MRSP (P=0.0361, OR=2.742, [1.067-7.045]). O estudo de seguimento, realizado em 54 pessoas, permitiu verificar que a maioria (85%) continuava colonizada, ao fim de 1 a 4 meses, por uma das espécies de MRS iniciais, devendo-se esta persistência à presença da mesma estirpe ou aquisição sucessiva de estirpes.

A elevada prevalência de colonização por MRS, os clones detetados e os fatores de risco identificados sugerem exposição ocupacional, realçando a importância e necessidade de implementar medidas de prevenção e controlo de infeção para minimizar a propagação destes agentes patogénicos na prática clínica veterinária.

Feocromocitoma em canídeos – estudo de 24 casos clínicos

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O feocromocitoma é um tumor endócrino raro, do sistema nervoso simpático, com origem nas células cromafins da medula adrenal e produtor de catecolaminas. O diagnóstico *ante mortem* é complexo e raro, exigindo um alto índice de suspeita por parte dos médicos veterinários, devido à natureza paroxismal e inespecífica de sinais clínicos assim como à presença de doenças concomitantes e ainda carência de meios de diagnóstico específicos e sensíveis em cães. Como resultado de potenciais consequências fatais, o diagnóstico precoce é essencial de forma a garantir qualquer possibilidade de sobrevivência do animal.

O objetivo deste trabalho consistiu na caracterização da apresentação clínica, laboratorial, imagiológica e anátomo-patológica de 24 cães com o diagnóstico histopatológico de feocromocitoma realizado no laboratório de Anatomia Patológica da Faculdade de Medicina Veterinária da Universidade de Lisboa, entre junho de 2003 a junho de 2016. Constatou-se a existência de uma vasta variedade de sinais clínicos, sendo que os mais comuns consistiram na fraqueza generalizada, na letargia e na taquipneia/arfar. Os animais apresentaram uma média de 11 anos de idade e a presença de doenças concomitantes (FR=54%) e de tumores de outros tecidos (FR=42%) foi bastante expressiva, o que dificultou a interpretação dos sinais clínicos e dos parâmetros laboratoriais. A medição da pressão arterial foi efetuada apenas em três animais e em nenhum animal foram realizados testes funcionais. A causa da morte foi associada à prática de eutanásia em 65% dos cães, devido ao deterioramento do estado clínico dos mesmos, e à morte natural em 35%, sendo que, apesar da existência de doenças concomitantes, a presença de um feocromocitoma pareceu justificar o quadro clínico apresentado na maioria dos animais.

A ecografia abdominal apresentou um papel valioso, na medida em que consistiu no único meio de diagnóstico que permitiu suspeitar deste tumor ante mortem, através da observação de uma massa adrenal. Esta suspeita apenas ocorreu em 8% dos cães, o que reforça o desconhecimento atual da presença deste tumor e das suas consequências.

Fibroplasia esclerosante eosinofílica gastro-intestinal felina (FEEGF): a propósito de 3 casos clínicos

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Recentemente descrita, a FEEGF caracteriza-se pela presença de massas/lesões focais de natureza inflamatória mista mas predominantemente eosinofílica no tracto digestivo. O prognóstico é reservado e a resposta ao tratamento médico é variável. Os casos descritos são escassos devido à fraca prevalência e ao desconhecimento da doença.

Esta série de casos visa sensibilizar o clínico de animais de companhia para esta doença emergente.

Métodos
Foi efectuado um estudo retrospectivo que incluiu os casos de FEEGF diagnosticados entre Março e Outubro de 2015 no CHV Fregis, Arcueil, França.

Resultados
Três gatos foram incluídos no estudo: uma fêmea maine-coon de 13 meses e dois machos de raça europeu comum de 7 e 9 anos. Os três gatos foram referenciados por vómito crónico. Foi efectuada ecografia abdominal em todos os animais, a qual revelou a presença de um espessamento parietal focal ao nível do duodeno proximal e linfadenomegália loco-regional. A gata maine-coon apresentava concomitantemente um corpo estranho jejunal (tricobezoar). Esta gata foi submetida a laparotomia tendo sido feita a exérese do tricobezoar por enterotomia e realizadas biópsias do duodeno e linfonodos loco-regionais. Os outros dois gatos foram submetidos a gastro-duodenoscopia, a qual permitiu a inspecção da região piloro-duodenal e realização de biópsias digestivas. Macroscopicamente, ambos apresentavam um espessamento importante da junção piloro-duodenal e num dos casos foi ainda observada uma ulceração focal da mucosa com reação proliferativa concomitante. A análise histológica foi similar nos três casos e revelou uma FEEGF (enterite mista predominantemente eosinofílica associada à presença de fribroblastos reactivos e colagénio).

Foi iniciado tratamento medico nos três casos (alimentação hipoalergénica, prednisolona e omeprazole). Um gato apresentou melhoria clinica e regressao ecográfica da lesão duodenal (ultimo controlo clinico e ecográfico um ano após diagnostico). Um gato recidivou 3 semanas pós-diagnóstico com progressão da lesão e subsequente oclusão/sub-oclusão digestiva. Apesar de ter sido feita gastro-duodenostomia, o gato morreu 48h após a intervenção. A gata Main-coon recidivou 2 meses após o diagnostico e os donos optaram pela eutanasia.

Conclusão
Mais do que aumentar o número de casos descritos de FEEGF, este trabalho sobre-eleva a importância de incluir esta doença no diagnóstico diferencial de lesões digestivas focais em gatos.

Frequência dos tipos de sangue felinos na Península Ibérica

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Objectivos
Documentar a prevalência dos tipos de sangue felinos na Península Ibérica e determinar o risco potencial de reações transfusionais resultantes de incompatibilidade do sistema AB em transfusões entre animais não tipificados e o risco potencial de isoeritrólise neonatal em gatinhos descendentes de pais de tipo de sangue desconhecido.

Métodos
As amostras de sangue foram recolhidas de animais dadores do Banco de Sangue Animal (BSA - Banco de Sangue Animal Lda). A tipificação sanguínea foi realizada através de um sistema de cartão (RapidVet-H Feline Blood Typing, MDS).

Resultados
A população estudada compreende 1070 gatos de Portugal e Espanha, de ambos os sexos e com idades compreendidas entre um e oito anos. As frequências dos tipos de sangue A e B foram 96.5% e 3.5%, respetivamente. Não foram encontrados gatos AB. Com base nestes dados, o risco potencial de reação transfusional em transfusões entre animais não tipificados e o risco potencial de isoeritrólise neonatal foram calculados, sendo 6.8% e 2.8%, respetivamente.

Conclusões
Contrariamente a estudos anteriores, não foram encontrados gatos do tipo AB neste estudo. Embora o risco potencial calculado de reação transfusional em transfusões entre animais não tipificados e de isoeritrólise neonatal em gatinhos descendentes de animais não tipificados sejam baixos, a tipificação dos felinos antes da transfusão e a tipificação de gatos para criação é recomendada.

Download : Tabela Frequência dos tipos de sangue felinos na Península Ibérica.docx
http://efomv.guc.pt/PaginaF.aspx?Tipo=ResumoSingle&Chave=00027B28E4&Janela=Nao

Infeção canina por *Cyniclomyces guttulatus*

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A levedura comensal do trato gastrointestinal dos lagomorfos, *Cyniclomyces guttulatus* (família Saccharomycetaceae; ordem Saccharomycetales), tem vindo a ser apontada como um putativo patogénico oportunista em cães.

O presente trabalho visa a descrição de um caso clínico de infeção canina por *C. guttulatus*. Em Setembro de 2015, uma cadela da raça Golden Retriever, com 12 anos de idade, apresentou-se à consulta na Clínica Veterinária Aristocão (Torres Vedras, Lisboa), tendo o estimulo iatrotrópico consistido num quadro de diarreia intermitente. O animal encontrava-se devidamente desparasitado e vacinado, habitava numa casa com terraço e era alimentado bi-diariamente com alimento composto completo seco, juntamente com frutas e vegetais. Ao exame clínico não se observaram sinais inequívocos de doença. Constatou-se, junto do tutor, que o quadro de diarreia intermitente permanecia há duas semanas, tendo-se iniciado após um período de férias numa casa de praia onde o canídeo teve acesso permanente a um jardim exterior. O tratamento preconizado consistiu na administração de metronidazol (20 mg/kg PO q12h durante 10 dias consecutivos), contudo sem sucesso terapêutico. Seguidamente efetuou-se uma análise coprológica através da técnica de Willis modificada, tendo-se observado a presença de inúmeras formas vegetativas de *C. guttulatus*. Prescreveu-se, como tratamento, a administração de nistatina (150000 UI PO q8h durante 5 dias consecutivos) e de pre/probióticos (PO q24h 10 dias consecutivos). Realizou-se nova análise coprológica, cinco dias após o término do tratamento, não se observando a presença de *C. guttulatus*. A consistência das fezes normalizou.

A expressividade clínica da infeção canina por *C. guttulatus* continua por elucidar. Não obstante, tem vindo a ser descrita a presença desta levedura em amostras fecais de cães com alterações gastrointestinais. Apesar de não estar documentado nenhum tratamento efetivo contra *C. guttulatus*, os resultados terapêuticos por nós alcançados sugerem que o uso concomitante de nistatina e pre/probióticos possa ser uma hipótese viável.

Infecção por *Mycobacterium avium* num gato em Portugal

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Objetivos

As micobactérias que infectam gatos dividem-se em 3 grupos: micobacterioses do complexo tuberculose (*Mycobacterium bovis, M. tuberculosis, M. microti*); lepra felina (*M. lepraemurium*) e outras micobactérias não causadoras de tuberculose (*M. fortuitum, M. avium*). Neste trabalho descreve-se um caso de infecção cutânea granulomatosa por *M. avium*. Um gato, Europeu, não esterilizado, de 2 anos, não vacinado e com acesso ao exterior, apresentou-se para consulta de segunda opinião com tumefacção subcutânea dura, não ulcerada e não alopécica que se estendia pelas regiões cervical, dorsal e escapular. O restante exame físico, hemograma, bioquímica geral, radiografia torácica e ecografia abdominal não apresentavam alterações. O gato tinha sido previamente sujeito a terapêutica com diferentes antibióticos e anti-inflamatórios durante dois meses. Na avaliação citológica da tumefacção observou-se inflamação piogranulomatosa, com estruturas não coradas fagocitadas em macrófagos. A presença de microorganismos álcoolácido resistentes foi confirmada pela coloração Ziehl-Neelsen. O diagnóstico definitivo de micobacteriose foi efectuado com recurso às seguintes análises: cultura em meio sólido Lowenstein- Jensen e líquido Middlebrook 7H9 (em curso), extração de DNA a partir de aspirado da massa cutânea, amplificação parcial dos genes rpoB e hsp65 específicos para o género *Mycobacterium* spp. (resultado positivo), amplificação do IS6110 do complexo *M. tuberculosis* (resultado negativo). A sequenciação dos genes rpoB e hsp65 permitiu a identificação do complexo *M. avium* e, através das sequências de inserção específicas para a diferenciação dentro deste complexo, foi identificada a espécie M. avium. Iniciou-se tratamento com rifampicina (13 mg/Kg SID PO), mas como não houve diminuição do tamanho da tumefacção, associou-se pradofloxacina (7,5 mg/Kg SID PO). São raras as descrições de infecções por micobactérias em animais, particularmente em animais de companhia em Portugal. O diagnóstico etiológico é habitualmente difícil e requer confirmação e identificação da espécie por técnicas de biologia molecular devido à necessidade de avaliar o potencial zoonótico da micobactéria. As infecções por *M. avium* acarretam outra dificuldade que se prende com a baixa eficácia do tratamento médico. O presente caso demonstra a importância da análise citológica e alerta para a inclusão de infecção por *Mycobacterium* na lista de diagnósticos diferenciais de lesões cutâneas granulomatosas/piogranulomatosas em gatos.

O leite e o consumidor: preferências de compra e consumo

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A reestruturação do setor leiteiro Português ocorrida nos últimos anos, principalmente desde o anúncio do término das quotas leiteiras, levou a alguma desorganização do tecido produtivo nacional. Aliado a isto, denota-se uma diminuição acentuada do consumo de leite, acompanhada pela crescente mediatização de informação de cariz negativo. A diminuição do consumo *per capita* relaciona-se assim com os excedentes de leite em natureza e, com a entrada de leite importado no mercado Português, colocando em risco a vitalidade do setor produtivo leiteiro.

Pretendeu-se com este trabalho avaliar o conhecimento e opinião de uma amostra de consumidores sobre o setor leiteiro, bem como entender o seu comportamento de compra e consumo. Os objetivos incluíam também perceber potenciais razões para a diminuição, ou extinção, do consumo de leite por parte de muitos consumidores, bem como avaliar quais os fatores que poderiam levar ao aumento do consumo. Finalmente, visou-se perceber as motivações de quem não consome leite no dia-a-dia e se há algum fator que possa alterar esse hábito.

Para tal, recorreu-se a um questionário validado, disponibilizado numa plataforma online, disseminado através de redes sociais, tendo sido obtidas 535 respostas completas e consideradas válidas para este estudo.

Como resultado, detetou-se que este alimento se está a distanciar principalmente dos habitantes de meio urbano, enquanto crescem as ofertas de mercado e o conhecimento relativamente a bebidas alternativas, nomeadamente vegetais. Adicionalmente, os não consumidores de leite apresentam como principal motivação a perceção de que o leite é pouco saudável e faz mal à saúde.

Apontam-se sugestões para a necessidade de dinamização do setor, abordando todo o conhecimento ou falta dele por parte dos consumidores, que paira em torno da atividade leiteira, focada fundamentalmente nos habitantes de meio urbano, classes etárias jovens, incidindo estudantes, e mulheres.

É necessário encontrar novos mercados e desenvolver produtos que acrescentem valor ao leite, indo ao encontro das necessidades e preferências do consumidor, de modo a manter o setor em atividade de forma sustentável.

Polirradiculoneurite aguda em cães: estudo descritivo de 20 casos clínicos

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Objetivos
A polirradiculoneurite aguda é a forma mais comum de polineuropatia aguda em cães e é considerada o equivalente à síndrome de Guillain-Barré em Humanos. É uma doença imunomediada que afeta as raízes nervosas ventrais dos nervos espinhais, apresentando-se geralmente com um quadro agudo de lesão generalizada de neurónio motor inferior, possivelmente acompanhada de disfunção de nervos cranianos. Apesar do seu estudo em Medicina Humana ser mais aprofundado, em Medicina Veterinária muitos factos sobre a doença permanecem por esclarecer. Este trabalho teve como objetivo caracterizar uma população de 20 cães com diagnóstico de polirradiculoneurite aguda na clínica Referência Veterinária (Portugal) durante o período entre outubro de 2011 e novembro de 2015.

Métodos
Em relação aos parâmetros avaliados, foram estudados os dados demográficos, a história pregressa, a apresentação clinica, os resultados dos meios complementares de diagnóstico, o tempo de recuperação e a subclassificação da doença consoante a causa. Quatro cães realizaram a terapêutica com imunoglobulina humana intravenosa. Os resultados observados revelaram que toda a população estudada apresentou um quadro agudo não ambulatório, tendo 17 cães (85% do total) apresentado um quadro ascendente progressivo.

Resultados
Quanto ao exame neurológico, 19 cães (95%) apresentaram tetraparesia não ambulatória, um cão (5%) tetraplegia e toda a população apresentou fraqueza cervical. Alguns animais revelaram défices nos nervos cranianos (13, 65%) e alterações na vocalização (17, 85%), com hiporreflexia generalizada (18, 90%). Oito cães obtiveram resultados positivos para Neospora caninum e/ou Toxoplasma gondii. O tempo médio de recuperação foi de 41,07 dias. Quatro animais que realizaram tratamento com imunoglobulina intravenosa apresentaram uma média de 42,75 dias e os 11 cães que apenas fizeram tratamento de suporte apresentaram 40,45 dias de recuperação.

Conclusões
Neste estudo foi possível concluir que os resultados acerca da progressão da doença, apresentação clínica e tempo de recuperação são semelhantes aos referidos na bibliografia consultada. Parece observar-se também uma associação entre a doença e infeções por Neospora caninum e Toxoplasma gondii. Em relação ao tratamento com imunoglobulina intravenosa, os resultados apresentaramse inconclusivos, carecendo de estudos futuros para um maior esclarecimento do seu papel no tratamento desta

Primeiro restreio de parasitas cardiopulmunoares e gastrointestinais em cães e gatos da região do Algarve utilizando a técnica de Flotac

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Objetivos

Felinos e caninos são hospedeiros de vários parasitas alguns dos quais com potencial zoonótico. O contacto íntimo que muitos donos têm com os seus animais, associado a uma desparasitação por vez deficiente aumenta o risco de transmissão destes agentes aos humanos. Atualmente há poucos dados sobre a incidência parasitária em cães e gatos no Algarve. Por esta razão foi realizado um estudo epidemiológico da fauna parasitológica gastrointestinal e cardiopulmonar presente em cães e gatos da região.

Métodos

Entre Fevereiro e Abril de 2016, 140 amostras fecais (Caninos n=64, Felinos n=76), foram colhidas aleatoriamente de 7 canis e 6 gatis. As amostras foram suspensas em água, filtradas e centrifugadas. Uma solução de sulfato de zinco foi adicionado a cada pellet, 5ml foi colocado em cada câmara do flotac. Procedeu-se à centrifugação do flotac. O disco de leitura foi removido e colocado num microscópio para visualização.

Resultados

22% (14/64) dos caninos estavam parasitados, 3% (2/64) com infeções mistas. *Toxocara* spp. foi o parasita mais frequente 14% (9/64), seguido de *Taeniidae*. 5% (3/64), *Ancylostomatidae* 3% (2/64) e *Trichuris* spp. 2% (1/64). Nos felinos 41% (31/76) estavam parasitados, 9% (7/76) eram infeções mistas. *Toxocara* spp. foi o parasita mais frequente 32% (24/76), seguido de *Isospora* 7% (5/76), *Ancylostomatidae* 5% (4/76), *Aelurostrongylus abstrusus* 4% (3/76) e *Taeniidae* 1% (1/76).

Conclusões

Os resultados obtidos demonstram um maior grau de parasitismo nos gatos estudados, quando comparado com os cães. No entanto ambos os grupos apresentam uma variedade de parasitas com potencial zoonótico incluindo *Toxocara* spp e *Ancylostoma*. A elevada incidência de *Toxocara* spp nas duas espécies é particularmente preocupante e apresenta uma verdadeira ameaça não só para o bem-estar animal mas também a saúde pública e alerta para necessidade de desparasitação regular. A maior expressão nos gatos pode ser devido a questões de maneio como a maior proximidade dos animais, que vivem grandes grupos nos gatis, quando comparado com os caes que são geralmente mantidos em unidades com poucos elementos. Mas em ambos os casos a dependência de doações financeiras para a implementação de medidas profiláticas regulares torna difícil a tarefa de erradicação destes agentes.

Privaprol na interrupção da gestação: quando os fins questionam os meios

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Objetivos

O Privapro®l (Lotrifen 50mg/mL; FATRO) é um produto comercializado para interrupção precoce da gestação em cadelas, que atua induzindo a necrose do embrião e sua posterior reabsorção. Descrito como um produto não-hormonal, o Lotrifen (2-(4-Clorofenil)-(1,2,4)triazolo(5,1-a) isoquinolina] atua como um inibidor dos estrogénios. Apesar de terem sido introduzidos novos produtos no mercado, com maior eficiência e segurança acrescida para o animal, a sua utilização na prática clínica mantém-se, possivelmente por não ser demasiado dispendioso, mesmo em animais de grande porte, e por ser administrado próximo do momento do salto. Contudo, apesar de cumprir as recomendações do fabricante, em muitos casos o clínico depara-se com sequelas graves da sua administração, que colocam a vida do animal em risco.

Será então o Privaprol® uma forma adequada de induzir o aborto em cadelas?

Métodos

Baseado em 30 casos clínicos de aplicação do Privaprol®, este trabalho faz revisão dos principais efeitos secundários e sequelas associados à utilização do produto. O Privaprol® foi administrado a cadelas com idades entre os 18 meses e os 8 anos de idade. Em 25 dos casos, a administração foi feita sob a supervisão de um veterinário, na dose e esquema ecomendado pelo fabricante, mas 5 dos casos deram entrada como emergência clínica sendo descrita a administração deste abortivo pelo proprietário.

Resultados

O aborto completo foi conseguido em 93,3% (28:30) dos casos, mas em 6,7% (2:30) das situações o aborto foi parcial. Foram observados efeitos secundários idiossincráticos (rigidez do pescoço, apatia, anorexia, problemas digestivos e alteração na marcha) em 23,3% (7:30) das cadelas, que em 5 (16,7%) dos animais perduraram por 15-30 dias. Foram observados sinais compatíveis com a existência de processo inflamatório no útero em 63,3% (19:30) dos animais, dos quais 40% (12:30) desenvolveram piómetra até um mês pós-tratamento. Os restantes apresentaram um corrimento esverdeado semelhante a lóquias no final do período a que corresponderia o termo da gestação.

Conclusões

Uma reflexão estratégica (matriz SWOT adaptada) mostra que os principais fatores de risco no uso de Privaprol® são: o momento da administração, a sensibilidade individual, a idade da cadela e a inexistência de antibioterapia adjuvante.

Rastreio de anticorpos anti-*Leptospira* e anti-*Brucella* em suínos de raça Bísara da região de Trás-os-Montes

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Objetivos
A leptospirose e a brucelose são zoonoses reemergentes com grande distribuição mundial, a primeira causada por espiroquetas patogénicas do género *Leptospira* e a segunda por cocobacilos do género *Brucella*. Têm como denominador comum o facto de provocarem graves problemas reprodutivos com notável repercussão económica na produção animal e de afetarem um diversificado número de espécies animais, entre os quais os suínos. Os suínos da raça Bísara (porco bísaro), raça autóctone criada em regime extensivo, são bem conhecidos pelas qualidades organoléticas da sua carne e fumeiro, atingindo elevados preços no mercado.

O objetivo principal deste trabalho foi avaliar a ocorrência de anticorpos anti-*Leptospira* e anti-*Brucella* em suínos de raça Bísara da região de Trás-os-Montes, um estudo inédito.

Métodos
Foram analisados 57 soros de suínos bísaros provenientes de três concelhos do Distrito de Bragança - Vinhais (39 amostras), Miranda do Douro (4 amostras) e Mirandela (14 amostras) - 84,2% (48/57) dos quais fêmeas. Os animais apresentavam idades compreendidas entre os 6 meses e os 18 meses, com uma média correspondente de 12 meses. O diagnóstico da leptospirose foi realizado usando a técnica de aglutinação microscópica, e os testes de Rosa de Bengala, aglutinação em tubo e c-ELISA para a deteção de anticorpos anti-Brucella.

Resultados
O teste de aglutinação microscópica permitiu a deteção de anticorpos anti-*Leptospira* em 23% (13+/57) dos soros testados, tendo as principais reatividades ocorrido para os serogrupos *Ballum* e *Icterohaemorrhagiae*. Observou-se uma relação estatisticamente significativa entre a idade dos suínos e a presença destes anticorpos.

O teste de Rosa de Bengala detetou anticorpos anti-Brucella em 7% (4+/57) dos soros testados, enquanto o método de aglutinação em tubo não revelou títulos significativos. Apenas um soro (1,75%) foi positivo para a presença de anticorpos anti-Brucella pelo teste de c-ELISA e os três outros soros apresentaram resultados muito próximos do limite de positividade.

Conclusões
A deteção de anticorpos contra estes dois agentes zoonóticos sugere que os suínos da raça Bísara da região de Trás-os-Montes podem constituir uma potencial fonte de infeção para o Homem e outros animais (domésticos ou selvagens). Os resultados obtidos poderão estar relacionados com um contacto estreito com javalis (*Sus scrofa*) que habitam na mesma área geográfica, já que esta espécie de ungulado selvagem apresentou valores elevados de seropositividade com a utilização dos mesmos métodos de diagnóstico para as duas zoonoses em estudo.

A estreita vigilância epidemiológica destas doenças nestes animais poderá evitar perdas ecológicas e financeiras, bem como reduzir o risco de saúde pública.

Serological and molecular profile of Spotted Fever Group Rickettsiae in cats from Luanda, Angola.

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Background:
Cats are usually parasitized by ticks or fleas. Besides a panoply of pathogens which these arthropods can transmit, they are the main vectors of spotted fever group of *rickettsiae* (SFGR). Cats can be used as sentinels for identification of circulating *Rickettsiae* although their role in the transmission cycle of *Rickettsia* spp. is not well elucidated. A considerable percentage of the feline population are pets and, therefore, they are in close contact with their owners. Understanding which *Rickettsia* spp. are circulating in cats and in their ectoparasites might disclose the potential infections that can occur in humans.

Our aim was to assess the prevalence of IgG antibodies against SFGR and molecular detection of *Rickettsia* spp in cats from Luanda.

Methods
All the cats that were admitted at veterinary clinic in the city of Luanda between 1st of May of 2014 and 30th of February of 2016 were included in the study. Information from each cat was collected in questionnaire and an informed consent was authorized by the cat's owners. Blood specimens were collected from cats and plasma and buffy-coat were separated. Plasma was tested by serological assay using an in-house immunofluorescence assay using *R. africae* as antigen and buffy-coat was tested by a nested –PCR using specific primers for *Rickettsia* spp.

Results
Of a total of 100 cats admitted at the clinic twenty-two (22%) lives exclusively indoor and the other seventy-eight (78%) lives indoor with access to outdoor or exclusively outdoor. Almost 50% of these cats have contact with rodents and 86% had no protection against ectoparasites. Fifty-seven (57%) were females. Four (4%) were kittens (<6 months), 58 (58%) juveniles (6-12 months), 23 (23%) young adults (1-3 year), and 15 (15%) adults (>3 years).

In addition, blood samples from the same cats were also assessed to detect *Rickettsia* spp. by nested-polymerase chain reaction (PCR).

None of the cats had antibodies against *R. africae* and only one (1%) cat was PCR-positive for *Rickettsia* spp. The infected animal was a domestic short-haired female cat, that co-habited with other animals, with access to rodents and no protection against ectoparasites.

Conclusions
These results indicate that prevalence of Rickettsia spp. in cats from this study is very low. The prevalence of feline Rickettsia infection in the present study is much lower when compared with the numbers reported in other studies. However this result is in agreement with the low prevalence of Rickettsia spp. (5.8%) infection reported in a group of dogs from the same area.

Information on canine and feline ectoparasites in dogs and cats from Angola is lacking, however, we hypothesise that the vectors that most often parasitize canines and felines do not commonly harbour *Rickettsia* spp.

Seroprevalência da Fasciolose nos bovinos leiteiros da ilha de São Miguel – Açores

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Objetivos
A fasciolose é uma parasitose provocada pelo trematode, Fasciola hepatica, que afeta vários ruminantes e também o Homem. A ilha de São Miguel (ISM) apresenta condições climatéricas, solos e sistema de pastoreio favoráveis ao desenvolvimento desta parasitose. No presente estudo pretendeu-se determinar a seroprevalência e a distribuição geográfica da fasciolose bovina na ISM.

Métodos
Foram rastreadas 149 (10,5%) explorações de bovinos leiteiros da ISM de um total de 1424 explorações e de 51 590 vacas, durante o período de janeiro a julho de 2016. As explorações incluídas neste estudo foram distribuídas, aleatoriamente, por três áreas consideradas de risco baixo, médio e elevado para fasciolose, com base em estudos anteriormente realizados sobre esta parasitose na ISM. Foram selecionadas três vacas em produção por exploração, para colheita de amostras de sangue e fezes para rastreio de fasciolose. Foram colhidas 443 amostras de sangue para pesquisa de anticorpos contra a *F. hepatica*, pela técnica de ELISA indireto. Foram ainda colhidas fezes de 465 bovinos e constituído um pool de três vacas por exploração, resultando um total de 149 amostras, para posterior análise coprológica pelas técnicas de McMaster, Willis e sedimentação simples. A compilação e organização dos dados foram realizadas numa folha de cálculo do *Microsoft Office Excel* e a distribuição geográfica dos casos foi realizada comrecurso a suporte de informação geográfica (*ArcGis Desktop-ArcMAP softwares*).

Resultados
Os resultados obtidos revelaram que 46,7% (207 em 443) das vacas analisadas apresentaram anticorpos contra a *F. hepatica* e que 74,5% (111 em 149) das explorações apresentaram pelo menos uma vaca com serologia positiva para esta parasitose. No exame coprológico constatou-se que 13,4% (20 em 149) das explorações amostradas apresentaram ovos de rematódos.

Conclusões
A distribuição geográfica dos casos positivos permitiu concluir que a fasciolose está disseminada por toda a ISM e não apenas na metade oriental da ilha como referido em estudos anteriores. Os dados obtidos permitem concluir que a fasciolose é uma parasitose emergente, à semelhança do constatado em outras áreas do Globo.

Três casos fatais de pneumonia verminosa em gatos domésticos por *Aelurostrongylus Abstrusus*

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Aelurostrongylus abstrusus é um nemátode pulmonar de felídeos, agente de pneumonias verminosas de gravidade variável. É frequentemente subdiagnosticado por não ser incluído no diagnóstico diferencial na presença de sinais respiratórios e pela raridade de utilização da técnica Baermann, a recomendada para identificação do parasita. Os estudos *post-mortem* em gatos infetados por *A.abstrusus* em Portugal são escassos, pelo que se procedeu à caraterização das lesões pulmonares encontradas na necrópsia de três felídeos naturalmente infetados.

No âmbito de um projeto de mestrado, detetou-se *A. abstrusus* pela técnica de Baermann em amostras fecais de três felídeos de raça indefinida, com idades compreendidas entre 4-8 meses. Apresentavam anorexia, letargia, pirexia, corrimento nasal purulento, dispneia, taquipneia e diarreia. Tinham sido resgatados da rua por voluntários de um gatil que procederam à sua desparasitação utilizando a associação praziquantel e milbemicina oxima. Apesar da terapêutica instituída, o quadro clínico foi-se deteriorando, tendo os animais sido submetidos a eutanásia e doados para necrópsia, que se realizou na Faculdade de Medicina Veterinária da Universidade de Lisboa. Efetuaram-se cortes histológicos de pulmão para exame histopatológico, esfregaço por aposição e técnica de Baermann a partir de fragmentos de pulmão.

Foi observado através do esfregaço pulmonar de aposição, ovos embrionados e larvas de primeiro estádio (L1) com comprimento entre 300-390 µm e extremidade posterior em forma de "S", com entalhe dorsal. Observou-se também pneumonia granulomatosa associada a congestão e lesões nodulares múltiplas nos três animais, bem como pleurisia serofibrinosa, hemorrágica e sinfisária num dos casos. A nível histológico observaram-se ovos embrionados, larvas L1 e formas adultas no lúmen alveolar. Identificaram-se lesões difusas de pneumonia exsudativa, intersticial e granulomatosa. Outras alterações encontradas foram alveolite e hiperplasia do músculo liso assim como presença de infiltrados linfóides, eosinofílicos e de células inflamatórias macrofágicas. Pela técnica de Baermann observaram-se ovos e larvas L1 com movimentação ativa.

As lesões observadas post-mortem permitem complementar o conhecimento sobre o potencial patogénico do A. *abstrusus* em felídeos domésticos. Deste modo, a partilha do conhecimento e a sensibilização da comunidade médico-veterinária torna-se indispensável para um controlo dirigido e adequado a esta parasitose em Portugal.

Urolitíase em coelhos - A importância do maneio e alimentação

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Objetivos
A urolitíase é uma doença que acontece frequentemente em coelhos domésticos. Os rins são uma parte vital da regulação do cálcio nos coelhos. Durante períodos de privação de cálcio, a reabsorção tubular renal do cálcio aumenta, o contrário acontece em casos de excesso de cálcio no organismo, havendo um aumento considerável de cálcio excreto na urina. O elevado teor em cálcio, normal na urina do coelho, pode, em casos extremos, causar doença geniturinária como a "sludgy urine" e, eventualmente urolitíase.O objectivo
deste trabalho é fazer uma avaliação preliminar, clínica, da relação entre os vários factores predisponentes e a formação de urolitiase em coelhos

Métodos
Esta doença, em geral aparece relacionada com coelhos sedentários, com excesso de peso, alimentados com ração seca ad libitum e com história prévia de suplementação de minerais, podendo no entanto surgir em animais sem nenhum destes fatores predisponentes. Serão aqui descritos sete casos de urolitíase, vesical, uretral e renal, e discutidos o diagnóstico, tratamento e prevenção da sua recorrência, em comparação com o que é efetuado nos pequenos animais.

Resultados
Dos animais tratados e após mudança de maneio e alimentação, em nenhum deles houve recorrência do problema ao fim de 2, 4 e 5 anos respectivamente.

Conclusões
Embora não seja suficiente para se aferir a real importância do maneio e da alimentação na prevenção deste problema, é sugestivo que terão alguma influência. Serão no entanto, necessários estudos mais abrangentes e comparativos para conseguir perceber se existe alguma relação estatisticamente significativa.

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