VLA Miscellaneous Exotic and Farmed Species Expert Group

Annual Report on Camelid Submissions 2009

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INTRODUCTION

The aims of this review are two fold: to provide an overview of the work of the VLA with relation to diseases seen in South American camelids notably alpacas, llamas and guanacos, and to assist vets working with camelids.

In recent years the network of VLA Regional Laboratories has received a growing number of samples for disease investigation which may reflect the growing popularity of these animals.

Submission numbers	2003	2004	2005	2006	2007	2008	2009
Alpaca	243	355	401	489	627	821	997
Number of premises	125	196	254	306	342	440	501
Carcases	42	48	69	100	90	163	187
Llama	26	49	60	80	87	92	73
Number of premises	19	42	53	51	44	62	53
Carcases	2	9	11	28	36	16	13

There appears to be no major seasonal variation in the numbers of submissions which may range from swabs to whole carcases.

Recent statistics from industry organisations estimate that there are 36,000 camelids in the UK with 31,000 alpacas and the remainder mainly being llamas, there are also some farmed guanacos in the UK.

This overview will cover the main types of disease seen in camelids and is by no means meant to be a comprehensive survey of diseases seen in the UK. The report format is based on the quarterly reports published on the VLA website (http://www.vla.gov.uk) for the VLA Miscellaneous and Exotic Farmed Species Expert Group.

1. Notifiable Diseases

Tuberculosis

From January to the end of December 2009, *Mycobacterium bovis* was isolated from 68 submissions from fourteen alpaca herds, including two with infection initially detected in 2008. There were two submissions in which *M. microti* and a single submission in which *M. avium* were isolated from alpacas.

The **spoligotypes** recorded for such *M. bovis* isolates have usually been the same as those seen in cases of bovine TB and from wildlife in the same area, compatible with infection from a local source. However, a case reported in 2009 (Twomey *et al*. 2009a) in which the spoligotype did not match, would suggest that either it is possible for translocation of infected animals from one area to another, or that transmission of infection may occur between alpacas reared in different areas and then co-mingling at a stud or show.

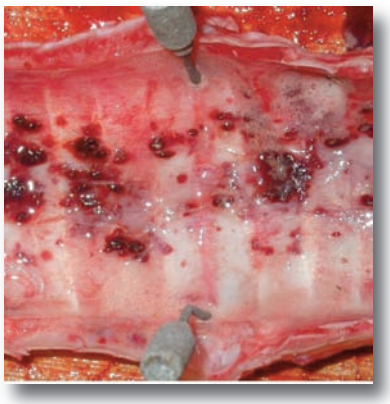
Avian TB was diagnosed in an adult male alpaca, which died after a one-week history of anorexia and terminal watery diarrhoea. At necropsy the mesenteric and submandibular lymph nodes were enlarged with caseous foci and the small intestinal mucosa was thickened. Smears of the lymph nodes contained numerous acid-fast organisms. Samples submitted for culture resulted in the growth of *Mycobacterium avium*. Histopathological lesions of mycobacterial infection were demonstrated in mesenteric and submandibular lymph nodes, and intestine and lung tissues. This is only the second case of avian TB diagnosed by the VLA in South American camelids since 2005.

In the two alpaca cases of ***M. microti* infection**, one died with signs of ill thrift and malaise. A field post-mortem revealed a caseous mesenteric mass and caseous lesions in the liver. In the second case an eight year old female alpaca in a group of 20 had aborted and after shearing a firm nodule was detected in the prescapular region. The private veterinary surgeon had suspected tuberculosis because *M. microti* infection had previously been diagnosed on the premises. Lesions with *M. microti* infection may also be found in the lungs (Zanolari *et al*. 2009).

Clinical signs of mycobacterial infection are non-specific and do not allow discernment of the causative organism. Signs observed may be of variable duration and include any combination of respiratory signs, malaise, anorexia, diarrhoea, weight loss and ill-thrift. Similarly, at post mortem examination it may not be readily possible to distinguish what type of mycobacterial infection is present.

In camelids ***M. bovis* lesions at post mortem** are mainly seen in the lung and thoracic lymph nodes and frequently in the liver though it is also possible to find lesions in many other tissues. Lesions in all tissues show extensive caseous necrosis and little mineralisation. While some camelids with tuberculosis are in poor body condition some with extensive lesions are in good body condition.

In lung sometimes there are multiple foci but in more advanced cases there may be extensive areas of caseous necrosis. Cavities are sometimes present in these large lesions (*see picture*) a feature in humans that indicates that the individual is highly infectious.



The lymph nodes of normal healthy camelids are small and can be difficult to find while tuberculous lymph nodes are often massively enlarged and can show multiple firm foci or the whole lymph node may be a caseous mass.

Lesions in the liver are often multifocal and can coalesce; they are usually not as extensive as lung lesions. Mineralised lesions, resulting from parasitic migration also may be seen in the liver which grossly might be mistaken for tuberculosis lesions.

Lesions have been seen in many other tissues but of note for potential routes of spread are the trachea, skin, gastrointestinal tract and kidney. The tracheal lesions (*see picture*) are raised granulomatous masses and may represent an extension of pulmonary pathology. The cutaneous lesions appear to be the result of enlarged discharging superficial lymph nodes. Scattered lesions in the lung and liver could be mistaken for lymphosarcoma.

Suspect cases of TB infection identified at post-mortem examination of camelids are notifiable to the local Animal Health Office. In such cases, VLA Regional Laboratories will undertake additional laboratory testing for TB (histology and tissue culture in selective media) free of charge.

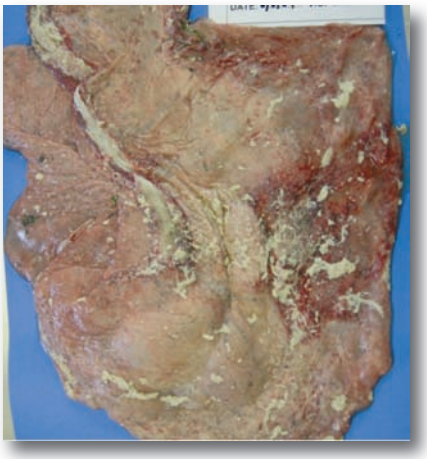
Sporadic cases of **Johne's disease** (which is not notifiable and is associated with *Mycobacterium avium subsp. paratuberculosis (Map)*) are reported in camelids from VLA submissions but there were none in 2009. Ante mortem testing of camelids for Johne's disease may include examination of faecal material including smears with Kinyoun Cold Staining, culture and PCR. In cattle antibody ELISA tests are frequently used for diagnosing Map but they are **not** appropriate for camelids which need species–specific reagents. In sheep and goats the agar gel immunodiffusion test (AGIDT) has been used to screen suspect cases for antibodies. This test does not require species-specific reagents and so could be used in camelids but the specificity and sensitivity of the test is unknown. In general if there are compatible clinical signs of scouring and weight loss then a positive AGIDT result may support a diagnosis of *Map*. A definitive diagnosis would depend upon positive culture and compatible histopathology findings which may include granulomatous enteritis and lymphadenitis of mesenteric lymph nodes.

2. Zoonotic Diseases

Abortions in alpacas due to Campylobacter fetus fetus

Abortions in South American camelids are usually sporadic and samples are not commonly received at VLA RLs. As a result causes of abortion are not well described.

During April and May, one herd experienced four abortions and a live premature cria subsequently died from a group of thirty alpacas due to give birth between March and September 2009. Two fetuses with placentae were submitted for abortion investigation (*see picture*) and *Campylobacter fetus fetus* was isolated from fetal stomach contents of both. Infection was associated with placentitis. A farm visit was undertaken to provide advice on the zoonotic risk and control of this organism. This is the first case of *Campylobacter* associated abortion in alpacas identified in the UK.



E. coli O157

Verocytotoxin-producing *Escherichia coli* (VTEC) O157 has emerged as an important human pathogen during the last 25 years. It causes a broad spectrum of human illness including diarrhoea, haemorrhagic colitis and severe systemic complications, particularly haemolytic uraemic syndrome. Zoonotic transmission can occur via direct contact with asymptomatic carriers or indirectly via contaminated food or water. Cattle are thought to be the main reservoir of infection but it has also been isolated from a wide range of other species including sheep, goats, pigs, horses, dogs and wild rabbits.

An unexpected finding from analysis of VTEC O157 outbreak investigations undertaken by VLA at the request of the public health authorities during the last decade (Pritchard *et al.* 2009) was the apparently high proportion (40-100%) of positive samples amongst alpacas and llamas in the three premises investigated. There are currently no known reports in the literature of VTEC O157 in camelids.

In May 2009 a 12 month survey of verotoxigenic (VTEC) O157 in camelids began using carcases submitted for diagnostic necropsy and faecal samples for routine monitoring or diagnostic purposes to VLA regional laboratories. The information will be used to estimate the extent of VTEC O157 in the sampled population. Provisional data from this survey estimate a prevalence of approximately 3% in the population sampled, which is not dissimilar to what is found in other farmed species.

3. New and Emerging Diseases

Mycoplasma haemolamae infection was detected for the first time by the VLA in 2009 in an anaemic alpaca. It was also detected in an alpaca that was not anaemic. Infected camelids may show acute onset of weakness and recumbency or may show more chronic signs with weight loss, anorexia and pale mucous membranes. Blood mycoplasmas (previously *Eperythrozoon* spp.) are believed to be opportunistic pathogens in other animal species. *Mycoplasma haemolamae* was identified by PCR/DGGE available from the Mycoplasma Group VLA Weybridge and in the first case was confirmed by 16S rDNA analysis. The prevalence and importance of this organism is currently unknown.

Skin Diseases

The diagnosis of skin diseases usually relies on collecting a history, a thorough physical examination and appropriate use of diagnostic tests (for more information see Foster *et al*. 2007a). Often clinicians are presented with chronic skin disease in camelids where the presenting clinical signs can be similar for different causes and include alopecia, crusting and scaling.

The most common skin condition in camelids is considered to be parasitic mange and especially chorioptic mange (Lusat *et al*. 2009; D'Alterio *et al*. 2005). Prior to 2009 less than six submissions per annum to the VLA had a diagnosis of mange; in 2009 there were six herds with scabies and three with chorioptic mange. This increase in the number of diagnoses may merely reflect the general increase in number of submissions. In reviewing the submissions for scabies several factors became apparent:

1. Clinical findings in one herd typically included alopecia, crusting and thickening of the axillae, chest, inguinal areas, perineum and interdigital areas, with extension to the limbs and head in some cases (see picture above). Pruritus can be severe.

2. Sarcoptic mange is usually, in other species, difficult to diagnose because mites are present in small numbers but in some camelids there may be large numbers of mites that are easy to detect with superficial skin scrapes.

3. It is important to establish an accurate diagnosis before embarking on treatment of the whole group. This may be achieved by microscopic examination of superficial skin scrapes from the margins of affected areas (*see picture of adult scabies mite*).

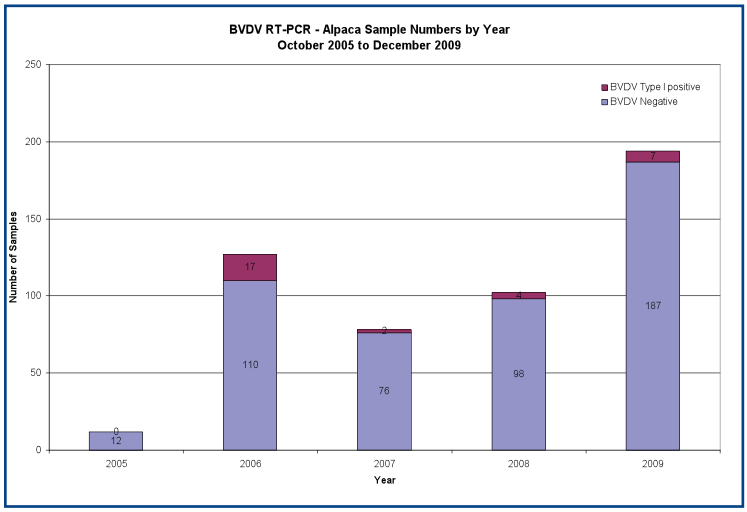
4. Some individuals are so badly affected that they either die or are euthanased. This may suggest that there are underlying predisposing factors that influence the outcome of infestation, such as parasitic burdens in the gastro-intestinal tract, dental disease, BVD, TB infection or mineral deficiency – but there is currently no evidence to suggest that these are predisposing factors.

5. Response to conventional treatments used in other species, such as macrocyclic lactones, can be variable often with disappointing results.

6. The potential for zoonotic spread to people handling affected camelids should always be considered, especially given the large numbers of mites that may be present in some cases.

These factors make biosecurity measures all the more important and when new animals are introduced to a herd they should be isolated, examined for skin disease, undergo appropriate diagnostic testing and thorough treatment before being introduced to the resident herd.

Bovine viral diarrhoea (BVD) virus infection



The histogram shows the number of submissions from alpacas tested for BVD by PCR.

Clinical cases in 2009 included crias with weight loss and diarrhoea. Clinical indicators of infection in South American camelids currently include ill-thrift, diarrhoea, abortion, stillbirths, and congenital neurological disease. Persistent infection has been demonstrated and potentially facilitates transmission of virus to other camelids and possibly other farmed species such as cattle and sheep, although the latter has not been proven. Most camelid isolates are BVDV type 1b (Foster *et al*. 2007b; Kim *et al*. 2009). Nevertheless, BVDV type 2 has also been demonstrated in camelids in Chile (Celedón *et al*. 2006), and it is sensible to continue monitoring British camelids for this virus, particularly as it is significant for cattle. The recommended test for detecting virus in camelids is PCR and for seroconversion from the serum neutralisation test (SNT). The standard serological antigen and antibody tests used in cattle are not validated and may give false results.

4. Endemic Diseases

Liver fluke

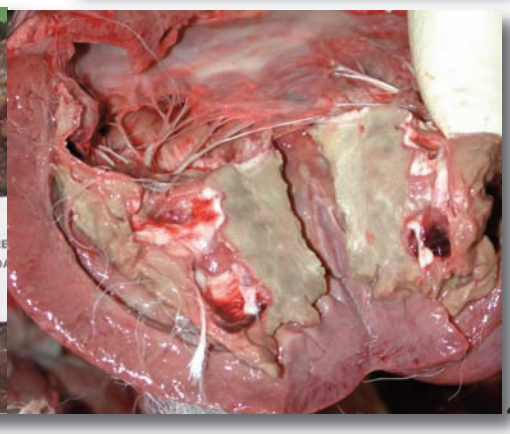
South American camelids appear to be particularly susceptible to *Fasciola hepatica* infection, reasons suggested including a deficient immune response, their relatively small liver size and grazing close to the ground (Leguia 1991).

In 2009, a total of 303 tests for fluke were requested on camelid submissions. Twenty eight cases were diagnosed in alpacas, of which 20 were known to be in adults. Four cases were diagnosed in adult llamas. The most common clinical sign observed was weight loss, other signs reported including apparent pain/discomfort, malaise and recumbency.

Nineteen (out of 32) cases were diagnosed at post mortem examination. Gross pathological findings included ascites, hydrothorax, subcutaneous oedema, hepatic fibrosis, fibrinous perihepatitis, variation in liver size, thickened bile ducts and adult flukes in the liver, occasionally in the absence of gross liver pathology, and significant blood loss into the peritoneal cavity. Endocarditis was seen in two cases and breathing difficulties and exercise intolerance observed clinically in one of these cases was presumed to be associated with this. Mural endocarditis has been described in a series of fasciolosis cases in North America (Firshman *et al*. 2008) and similar cases seen in submissions to VLA laboratories (*see pictures*) were summarised in a poster at the European College of Veterinary Pathology meeting (Schock *et al*. 2009).

Thirteen (out of 32) cases were diagnosed on faecal examination. Fluke eggs can be detected using the VLA sedimentation method, but this does not facilitate diagnosis during the prepatent period of disease.

Pictures: adult fluke in a chronic case (right), nodular hyperplasia (below left) and mural endocarditis (below right)



Parasitic gastroenteritis

Parasitic gastroenteritis (PGE) is considered to be a significant cause of production loss in domesticated South American camelids (SACs) in South America and is also considered a major disease problem in SACs reared in North America, Europe and Australia. Many of the nematode species encountered in domesticated SACs are species also encountered in domestic ruminants, but there are also other species including *Camelostrongylus mentulatus*. This is a common parasite of camelids in North Africa, the Middle East and South America, where it is the most frequently encountered nematode species in the C3 stomach. VLA Winchester provided the first report of this species in camelids in the UK (Welchman *et al*. 2008).

A survey of gut parasites, including nematodes, of alpacas and llamas was carried out by VLA Winchester in 2008-2009 and revealed *Camelostrongylus* spp., *Haemonchus* spp., *Ostertagia/Teladorsagia* sp, *Trichostrongylus axei*, *Trichostrongylus colubriformis, Trichostrongylus* spp., *Nematodirus battus, Nematodirus* spp., *Cooperia* sp, *Capillaria* spp., *Trichuris tenuis* and *Trichuris ovis*. Of 28 camelid carcasses received by VLA Winchester over the survey period, in at least 11 cases PGE was considered to have contributed substantially to the death of the animal, confirming that intestinal parasitism is a significant cause of mortality in VLA camelid submissions included in the study.

In 2009, a total of 675 tests for gastrointestinal nematodes were requested on camelid submissions. Fifty cases of PGE were diagnosed in alpacas, of which 35 were known to be in adults. Seven cases were reported in llamas, of which five were known to be in adults. Parasitic gastroenteritis was also diagnosed in one adult guanaco. Clinical signs associated with gastrointestinal nematodes included soft faeces, diarrhoea, ill thrift, anaemia, malaise and death, although some of these may have been ascribed to concurrent clinical conditions. Anaemia particularly is seen in association with *Haemonchus* spp. A more unusual clinical presentation of *Haemonchus* spp. infestation was seen on one premise, where eight alpacas were infested during the periparturient period, six of which died shortly after giving birth exhibiting marked terminal respiratory distress.

Species and genera detected in 2009 included *Camelostrongylus* spp., *Cooperia curticei, Haemonchus contortus, Nematodirus* spp., including *N. battus, Ostertagia circumcincta, Teladorsagia circumcincta, Trichostrongylus* spp. including *T. axei* and *Trichuris* spp., including *T. vitrinus*. Mixed nematode and coccidial infection was diagnosed in a number of cases and mixed nematode and fluke infestation was diagnosed once.

Thirty of the 58 cases of PGE were diagnosed at post mortem examination. Gross pathological findings included mucosal ulceration, necrosis, thickening, nodules, inflammation/congestion and fibrinopurulent exudates in the C3 compartment and watery contents in the small intestine, coupled with high C3 and/or small intestinal total worm counts and/or high large intestinal/faecal worm egg counts.

Additional findings included subcutaneous oedema and ascites and parasitic granulomata in mesenteric lymph nodes. In animals with haemonchosis, severe anaemia and pulmonary oedema also were seen. Histopathology was utilised occasionally to support gross pathological findings.

Twenty eight of the PGE cases were diagnosed using faecal egg counts. Traditionally, the modified Stoll's test has been favoured by some camelid practitioners for faecal egg counts in camelids (Cebra and Stang 2008). The VLA Aberystwyth's parasitology unit ran a comparison between this method and the Improved Modified McMaster's test, used within VLA, and the results suggested that the latter method, which allows detection of eggs at numbers as low as 10 eggs/gram, was likely to perform more effectively than the modified Stolls method. The existing VLA Modified McMaster's test has been modified further for camelids to incorporate use of both saturated sodium chloride and saturated zinc sulphate solutions, the latter to facilitate improved detection of the heavier *Trichuris* spp. eggs.

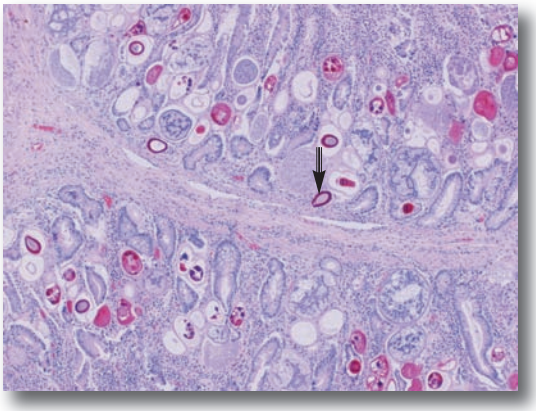
Coccidiosis

Five species of *Eimeria* have been reported in South American camelids: *E. macusaniensis, E. punoensis, E. ivitaensis, E. lamae and E. alpacae*. Prepatent periods vary with the species; *E. punoensis* is as short as 10 days, *E. macusaniensis* may exceed 30 days. *Eimeria macusaniensis* is possibly associated with the greatest severity of disease, causing severe necrotising/ulcerating enteritis.

In 2009, a total of 630 coccidial oocyst counts were requested on camelid submissions. Thirty six cases of coccidiosis were reported in alpacas, of which 12 were known to be in adults and 18 known to be in crias. Three cases were reported in llamas, of which all were known to be in crias. In 6 cases, co-infection with gastrointestinal nematodes was reported. Clinical signs observed with coccidiosis included ill thrift, soft faeces, diarrhoea, anaemia and malaise. Where coccidial oocysts were speciated, *E. macusaniensis, E. punoensis* and *E. lamae* were identified.

Eighteen (out of 39) cases were diagnosed at post mortem examination. Small intestine necrosis was a feature of some cases and this highly pathogenic effect has previously been documented in British alpacas (Schock *et al*. 2007, *see picture)*. In one alpaca cria with dramatic weight loss, histopathology of kidney in addition to intestines detected renal pathology probably secondary to coccidiosis. In this case, faecal parasitology had identified *E. macusaniensis* and other unspeciated coccidial oocysts.

Twenty one (out of 39) cases were diagnosed using faecal oocyst counts. Diagnosis of clinical cases can be complicated by the potential for prepatent infections with *E. macusaniensis* but also by the fact that many asymptomatic alpacas shed coccidia. Therefore, any coccidial oocysts detected in faeces need to be considered in the light of clinical signs. *Eimeria macusaniensis* is a much larger coccidian than the other species, measuring up to 100 microns in length. Due to its size and weight, floatation solutions with specific gravity of 1.20 (e.g. saturated sodium chloride) or less may not detect *E. macusaniensis*. This is a further reason why the VLA Improved Modified McMaster method for camelids uses saturated zinc sulphate solution, which has a higher specific gravity than saturated sodium chloride. For other, smaller *Eimeria* species, saturated sodium chloride is most effective at detecting oocysts and therefore saturated salt solution has been retained for identifying these *Eimeria* species.



Histopathology: enteritis in an alpaca with various developmental stages of *E. macusaniensis* including the thick-walled pear-shaped oocyst (arrow).

Cryptosporidiosis

In the US, it is believed that cryptosporidia are important enteric pathogens of alpaca crias up to six months of age (Waitt *et al*. 2008). In the UK, cryptosporidiosis is a recognized although relatively uncommonly reported cause of diarrhoea in alpacas. Between 2003 and 2008, 14 cases of cryptosporidiosis were diagnosed by the VLA in alpaca crias aged between one week and three months. In 2009, 16 tests for cryptosporidia were requested on camelid submissions, but no cases were reported. Diagnosis in the VLA is primarily by Modified Ziehl Neelson smears of faeces or intestinal contents and, in carcases, histopathology may also be used.

Neoplasia

Round cell tumours (suspected lymphosarcoma) were diagnosed four times in alpacas ranging from six weeks to nine years of age during 2009. Clinical signs included chronic weight loss in two animals, associated with generalised (multicentric) distribution of lesions, and swelling of the neck with dyspnoea in the third (youngest) animal, associated with lesions in the thymus and prescapular lymph nodes. In the fourth case, the spleen was affected in an adult female, but the primary significance of this was unclear as she also had severe sarcoptic mange, hepatic lipidosis, an ovarian teratoma and severe mucometra.

Malignant round cell neoplasia represents a heterogeneous group of tumours, which are amongst the most commonly reported tumours of South American camelids. In the largest review of this type of neoplasia, including 24 camelid cases, the lesions were multicentric in 12 animals, intra-abdominal in nine, intra-thoracic in two and one case involved the neck (Martin *et al*. 2009). The age range was late gestational fetus to 23 years. Using immunohistochemistry, 12 cases were classified as B-cell lymphoma, six as T-cell lymphoma and the others were primitive malignant round cell tumours,

which did not express antigens for either B- or T-cells and possibly originated from neural crest cells. In a similar study of six British cases submitted to the VLA, four cases of T-cell lymphoma, one case of B-cell lymphoma and one mixed T- and B-cell tumour were identified, the latter being a very rare finding in animals (Twomey *et al*. 2008). The results have potential comparative pathological significance, as similar tumours are recognised in humans.

Picture shows a cria with round cell tumour (likely lymphosarcoma) involving the liver

Congenital problems

Congenital problems are believed to be common in South American camelids (Fowler 2010), probably due to the relatively small breeding population, and hence genetic pool, for these species. Anomalies detected during 2009 included choanal atresia (two crias), atresia ani (one of the crias with choanal atresia), ventricular septal defect (one 18 month old alpaca), triple cardiac anomaly, including ventricular septal defect, persistent foramen ovale, tricuspid valve atresia (one cria), renal hypoplasia with hydronephrosis (one cria) and flexor tendon contracture (one cria). The latter anomaly was an incidental finding in a septicaemic cria, and musculoskeletal defects of this type are likely to be more common than indicated by VLA carcase submissions. Choanal atresia is one of the most frequent congenital anomalies in alpacas. The membranous barrier that persists between the pharynx and nasal cavity usually results in respiratory and suckling difficulties at, or soon after birth and usually requires euthanasia on welfare grounds.

Gastric ulceration

Gastric ulceration is commonly seen in South American camelids and is usually associated with stress. Examples of potential stressors identified in 2009 include animal movements, establishing new animal groups and concurrent diseases (usually parasitic). All three stomach compartments can be affected, although lesions are most common in the C3 compartment where they can perforate the stomach wall and lead to peritonitis. Nine such cases were diagnosed in carcases submitted to VLA in 2009, affecting both adults and crias, as well as several cases which had not perforated and were incidental findings. A single case of perforated jejunal ulcer was also associated with peritonitis.

Bacterial diseases

Bacterial infections are mainly of significance in neonates, with septicaemia being an important cause of death. In this age group, risk factors include poor colostrum intake, poor colostrum quality, mismothering, premature birth, poor environmental hygiene and navel infections. Bacterial diseases from all age groups for 2009 are summarised in the following table

Age group	Bacterial isolate	Pathological presentation (no. of cases)
Crias	*Arcanobacterium pyogenes*	Lung abscess (1)
	E. coli	Septicaemia (5); nephritis (1)
	Haemophilus spp.	Septicaemia (1)
	Listeria spp.	Septicaemia (1); heart valve lesion (1)
	Staphylococcus xylosis	Cerebral abscess (1)
	Streptococcus bovis biotype 1 (*Streptococcus gallolyticus gallolyticus*)	Septicaemia (1)
Adults	*Clostridium perfringens*	Enterotoxaemia (2)
	E. coli	Septicaemia (1); pleurisy (1)
	Listeria spp.	Septicaemia (2)
	Streptococcus bovis	Jaw abscess (1)
	Streptococcus equi ruminatorum	Septicaemia (1)
	Unidentified	Multiple abdominal abscesses (1); bronchopneumonia (1)
	Unidentified (presumed secondary to trekking equipment injury)	Multiple fibrinopurulent lesions extending from a ruptured abscess on the body wall (1)
Unspecified	*E. coli*	Nephritis (1)

PRESENTATIONS

1.James Barnett gave a presentation with colleagues from Animal Health about TB and other diseases in camelids to the local Cornish camelid stakeholder group on 7th July. This was mentioned in a report published in the Farmers Guardian: http://www.farmersguardian.com/story.asp?sectioncode=1&storycode=27420

2.Fin Twomey presented an abstract at the annual meeting of the Association of Veterinary Teachers and Research Workers in Scarborough on April 7th entitled "Outbreak of tuberculosis in a Devon llama herd"; Proceedings page 15, abstract number 18.

3.Fin Twomey gave a two hour presentation on South American Camelids: Species of emerging interest to the General Practitioner, on 22nd April 2009 to the Western Counties Veterinary Association in Exeter.

4.Aiden Foster gave a presentation about diseases of camelids including BVD, fasciolosis and TB, on June 18th at VLA Weybridge as part of the New & Emerging Diseases Symposium for industry stakeholders, with a report in the Veterinary Record.

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