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Diagnostic pathology in microbial diseases of sheep or goats

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ABSTRACT

Post-mortem examination is a key step in the diagnostic process of infectious diseases in sheep and goats. Diagnostic pathology deals with identification and study of lesions, at the same time providing also significant clues regarding pathogenesis of the diseases. This article reviews the salient pathological findings associated with the most significant infectious diseases of sheep and goats present in countries where small ruminants are a relevant agricultural industry. Lesions are reviewed according to the different organ systems where they occur. Emphasis has been given in the description of the salient lesional patterns than can be identified in each organ and which can be of help in the differential diagnosis of the lesions caused by bacteria, viruses, fungi or prions. Finally, a review of the usefulness of ancillary tests that may be used on various tissue samples for performing an aetiological diagnosis, is included; the application of various techniques, from immunohistochemistry to molecular biology-based tests, is described.

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1. Introduction

Diagnostic pathology deals with study and diagnosis of disease through examination of organs, tissues or cells from affected animals. Pathology is a key tool in the diagnosis, understanding and control of infectious diseases and essential when studying their pathogenesis (Caswell and Callanan, 2014).

A thorough post-mortem examination is probably the most useful tool for achieving a correct pathological diagnosis and the only way to identify the majority of lesions (King et al., 1999). Tissue or other (e.g., gastrointestinal content, fluid) samples are collected for further analysis and to establish a diagnosis of the disease. In small ruminants, due to their size and economic value, post-mortem examination is an affordable and frequently used method of diagnosis of infectious diseases or other conditions affecting the animal. In order to avoid overlooking gross changes, it is recommended that the necropsy should be performed methodically always following the same routine (King et al., 1999).

On the basis of the authors' experience in the field of diagnostic pathology in small ruminants, this article reviews the relevant literature on pathological findings associated with the most relevant infectious diseases of sheep and goats. The review does

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http://dx.doi.org/10.1016/j.vetmic.2015.07.012 0378-1135/© 2015 Elsevier B.V. All rights reserved. not contain an exhaustive list of lesions, but aims to provide some guidelines that permit the identification of pathological changes that appear in the different organ systems, paying special attention to the differential diagnosis. Finally, some information is provided regarding ancillary techniques, based on pathological methods, which can be used to arrive to a diagnosis from examining the affected tissues.

2. Alimentary system

2.1. Oral cavity

Superficial stomatitis (i.e., with lesions limited to the mucosa) is mainly caused by viruses leading to the formation of vesicles, erosions-ulcers or papules. Foot-and-mouth disease is caused by a virus of the Picornaviridae family that results in a systemic disease primarily of cloven-hoofed animals. Small ruminants are, in general, less susceptible than pigs or cattle and lesions in the mouth appear initially as blanched areas that subsequently develop into fluid-filled vesicles, which rapidly progress to shallow erosions (Alexandersen et al., 2003). The dental pad and the tongue are the most common locations for lesions and erosions usually heal after a few days with little exudate or scarring. Pestedes-petits-ruminants virus of the Paramyxoviridae family causes a disease currently recognised in several parts of Africa and Asia. The virus has an affinity for the alimentary epithelium where it causes necrosis. Subsequent abrasions in the affected area are responsible for the sloughing of necrotic tissue resulting in erosions and ulcers, primarily in the caudal oral cavity (Truong et al., 2014). Goats and sheep are also susceptible to bluetongue, a viral disease caused by a virus of the Reoviridae family, which induces vascular damage leading to oedema, haemorrhage and thrombosis in several organs (MacLachlan et al., 2008), including the oral mucosa. A relevant detailed review of the lesions has been published recently (MacLachlan et al., 2009). During the acute phase, lesions are characterised by catarrhal stomatitis, punctuate haemorrhages in the lips and gums and oedema and congestion of the tongue resulting in cyanosis, giving the disease its name. Subsequently, small erosions and ulcers are present especially in the hard palate and dental pad. Contagious ecthyma, or for contagious pustular dermatitis, is probably the most common viral disease affecting the oral cavity in small ruminants. It is caused by a parapoxvirus that affects primarily young lambs and kids (Reid and Rodger, 2007). Outbreaks can also occur in adult sheep. Initially, lesions appear as raised reddened or greyish areas, surrounded by hyperaemia, that rapidly evolve into papules (Supplementary material S1), which become necrotic and slough, originating therefore erosions and ulcers (McElroy and Basset, 2007). These lesions are most frequent in the gums, associated with erupting teeth, as well as in the hard and soft palate and in the lateral and dorsal zones of the tongue (McElroy and Basset, 2007). In long-standing clinical cases, we have observed that lesions become chronic and progress to proliferative stomatitis, with pseudo-papillomatous growths in the affected areas (Supplementary material S2), comprised of epithelial hyperplasia and granuloma formation that can persist for several months (Reid and Rodger, 2007). This may result in deformation of the tongue. Microscopically, there is epithelial hyperplasia with ballooning degeneration and eosinophilic cytoplasmic inclusions (McKeever et al., 1988).

Supplementary material related to this article found, in the online version, at http://dx.doi.org/10.1016/j.vetmic.2015.07.012.

A common consequence of mucosal lesions causing disruption of the oral epithelium, especially in orf-affected lambs, is secondary bacterial infection. Fusobacterium necrophorum, the aetiologic agent of oral necrobacillosis, is probably the most common cause of deep, often necrotising, stomatitis (i.e., lesions extend to deeper layers of the oral wall) in small ruminants (Nagaraja et al., 2005). The early lesions are characterised by welldemarcated white-grey-yellowish foci of necrosis surrounded by an hyperaemic ring and are commonly seen on the lateral or dorsal aspects of the tongue (Brown et al., 2007). Lesions can extend to form a locally extensive or even diffuse fibrino-necrotic stomatitis affecting the gums, palate, lingual aspects of the cheeks and the pharynx (Supplementary material S3). Microscopically, the necrotic tissue shows loss of histological architecture and peripheral vascular hyperaemia, leucocytes and, very chronic cases, granulation tissue (MacLeod and Watt, 1967). Bacteria within the lesions can be identified as Gram-negative filamentous rods around the periphery of the necrotic areas. Actinobacillosis, caused by Actinobacillus ligneriesii, can affect sheep causing chronic multifocal pyogranulomatous glossitis and stomatitis (Rycroft and Garside, 2000).

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2.2. Forestomachs and abomasum

The only lesion caused by an infectious agent of some significance in the forestomachs is necrotising inflammation caused by *F. necrophorum* or fungi, usually zygomycetes (Chiyaya et al., 1988; Nagaraja et al., 2005). In both cases, acidosis is a significant predisposing factor. Visible lesions include necrotic foci

surrounded by a hyperaemic halos (Supplementary material S4) that frequently sloughs resulting in ulcers.

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Haemorrhagic abomasitis is the most common lesion observed in the abomasum caused by infectious agents and are usually associated with clostridial infections. Exotoxins produced by *Clostridium septicum* infection are the cause of 'bradshot' or 'braxy', in sudden deaths in lambs in cold climates. At post-mortem examination (Ellis et al., 1983), besides other signs of toxaemia, the abomasum is severely and diffusely congested together with oedema of the folds, in which haemorrhages, ulcers or emphysema may be present. Infection by *Cl. sordelli* in lambs can also result in haemorrhagic abomasitis (Lewis, 2007). In our experience, multifocal punctuate mucosal haemorrhages together with abomasal milk overload can be seen in young sucking lambs dving from colibacillosis (Supplementary material S5).

Supplementary material related to this article found, in the online version, at http://dx.doi.org/10.1016/j.vetmic.2015.07.012.

2.3. Intestine

2.3.1. Enteritis in young lambs or kids

Enteritis in young animals is usually of multifactorial aetiology, with influences from management procedures, climatic factors, infectious agents, etc. Among these, Escherichia coli is an important cause of enteritis in lambs or kids during the first weeks of life (Muñoz et al., 1996). Newborn lambs are affected in their first hours of life by 'watery mouth disease', an endotoxic colibacillosis the predisposing factor of which is the inadequate intake of colostrum (Gilmour et al., 1985). At post-mortem examination, there is absence of colostrum in the abomasum, which is filled with the mucus seen typically in the foetuses, the presence of meconium in the large intestine and, occasionally, dilation of areas of the jejunum. During the first weeks of life, E. coli is also an important cause of diarrhoea that can also cause septicaemia. The most typical lesion is sero-mucous (catarrhal) enteritis, characterised by diffuse or segmental dilation of the jejunal loops and ileum due to abundant yellowish sero-mucous content (Supplementary material S6) (Tzipori et al., 1981). In our experience, the abomasum frequently contains coagulated milk with dark red spots due to small haemorrhages (Supplementary material S5). When septicaemia is present, many petechiae and larger haemorrhages can be observed on serosal surfaces. A sero-mucous enteritis is also a characteristic of cryptosporidiosis or rotavirus infections. The latter, although reported in lambs and kids, are not considered to be a significant cause of enteritis (Muñoz et al., 1996; Chatzopoulos et al., 2012). The main pathological distinction is made microscopically where, in contrast to colibacillosis, a marked villous atrophy is seen in the small intestine.

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The role of the several types of *Cl. perfringens* causing enteritis in small ruminants has been reviewed in detail by Uzal and Songer (2008). *Cl. perfringens* type B is the aetiologic agent of 'lamb dysentery', characterised by haemorrhagic and necrotising enteritis (Supplementary material S7), predominantly in the ileum, with ulcer formation and presence of blood and fibrin strands in the intestinal content, in the small intestines of lambs younger than 14 days. *Cl. perfringens* type C can also cause haemorrhagic enteritis in neonatal lambs. Occasionally, *Salmonella* species, mainly *S. arizonae*, or *Yersinia enterocolitica* have been incriminated in cases of diarrhoea in young lambs or goat kids and are characterised by a fibrinous enteritis (Richards et al., 1993). A condition termed 'terminal ileitis ' or 'regional ileitis', for which the aetiology has not been established, has been described in lambs up to 6-month-old

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