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Diagnosis of clinical or subclinical mastitis in ewes☆

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ABSTRACT

Objectives of this paper are to review (i) diagnostic methods and procedures available for clinical or subclinical mastitis in ewes and (ii) applications of these procedures in the diagnosis of mastitis. Early and correct diagnosis of the disease is important for identification of affected animals. The following diagnostic procedures can be used: clinical examination, imaging techniques (ultrasonography, endoscopy), bacteriological examination of milk samples, immunological tests, identification of biomarkers (cytological examination of milk and measurement of milk electroconductivity). In most cases, diagnosis of clinical mastitis is straightforward, based on the findings of clinical examination. The differential diagnosis includes primarily (i) bacterial mastitis (usually sporadic occurrence in a flock, usually unilateral, isolation of bacteria from milk samples), (ii) mycoplasmal mastitis [usually epidemic occurrence in a flock, usually bilateral accompanied by other signs (e.g., arthritis), isolation of *Mycoplasma* spp. from milk samples] and (iii) infection by *Small Ruminant Lentivirus* [usually epidemic occurrence in a flock, usually bilateral accompanied by various signs (e.g., respiratory or neurological signs) in the same or other animals of the flock, detection of antibodies to the virus, pro-viral DNA or viral RNA in blood samples]. Subclinical mastitis should be always suspected as one of the primary causes in cases of decreased milk production in dairy flocks; it should also be considered as a possible factor in cases of suboptimal growth rate of lambs in mutton-type production flocks. Diagnosis of subclinical mastitis is based on detection of infection (i.e., isolation of microorganisms from milk samples) and/or inflammatory reaction in the mammary gland. The best method for detection of the inflammatory reaction remains the demonstration of increased cellular content in milk, although various other methods, have been proposed. For individual animals, values $<0.5 \times 10^6$ cells mL^{-1} indicate a healthy mammary gland and values $>1.0 \times 10^6$ cells mL^{-1} indicate a mammary gland with clinical or subclinical mastitis, with no need to perform a simultaneous bacteriological examination of milk samples to confirm the problem; values between 0.5×10^6 and 1.0×10^6 cells mL^{-1} indicate 'suspected disease', hence there is a need for performing bacteriological examination in milk. Two consecutive measurements increase accuracy of results. In bulk milk samples, counts of 0.65×10^6 cells mL^{-1} indicate approximately 15% prevalence of subclinical mastitis in the flock. In the differential diagnosis of cases of reduced milk yield in ewes, other possible causes of the problem should be taken into account (e.g., parasitic infections, chronic wasting diseases, suboptimal level of nutrition); in cases of suboptimal growth rate of lambs, other factors may be responsible (e.g., protozoan or parasitic infections, energy or micronutrient deficiency, viral disease).

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1. Introduction

Mastitis is an important disease of sheep. It reduces production and adversely influences welfare of affected animals. The disease has an obvious financial importance in dairy flocks; in such flocks, it is one of the primary causes of the 'milk-drop syndrome of ewes' (Giadinis et al., 2012). Nevertheless, it is also important in mutton-type production flocks, as reduced milk production can lead to suboptimal growth rate of lambs (Fthenakis and Jones, 1990a).

Effective treatment of the disease requires an early diagnosis (Mavrogianni et al., 2011). Development of disease and subsequent damage to the gland is rapid; histological lesions in the mammary gland are evident within two days after infection (Fthenakis and Jones, 1990b; Mavrogianni et al., 2005). Therefore, early recognition of the disease in order to instigate treatment is important, as this would minimise mammary lesions and restore health of the affected animals. Objectives of this paper are to review: (i) diagnostic methods and procedures available for clinical or subclinical mastitis in ewes and (ii) applications of these procedures in diagnosis of mastitis.

2. Diagnostic procedures

2.1. Clinical examination

For clinical examination of the udder (mammary glands and teats), ewes should be cast and restrained. Possible lesions on the skin of the udder (e.g., colour changes, injuries, disease lesions) should be recorded, as well as changes in the general shape of the udder (e.g., increase in size, atrophy). Then, the mammary glands should be palpated. The shape, size, temperature and consistency of each gland should be assessed and recorded (Fthenakis, 1994). Any pain reaction during palpation should be noted; presence of abnormal structures within the gland (e.g., hardness, nodules) should be recorded. Subsequently, each teat should be held between the thumb and the index finger of the examiner and palpated throughout its length, from the base of the teat to its apex; its shape, size and consistency should be evaluated (Mavrogianni et al., 2005). That way, lesions possibly present inside the teat duct and cistern can be detected. In all cases, the mammary glands and the teats of the same ewe should be compared to each other and any differences should be noted. Finally, the supramammary lymph nodes of the animal should be palpated, in order to detect possible alterations in size or consistency.

Subsequently, the mammary secretion should be evaluated. The first few streams of milk are to be drawn from the teat onto a paddle or on the palm of the gloved hand of the investigator. Expression of milk can be evaluated; for example, expression in drops can be indicative of teat stenosis. Then, presence of abnormal features in mammary secretion (e.g., clots, flakes, tints) should also be recorded.

2.2. Imaging techniques

2.2.1. Ultrasonographic examination

Ultrasonography is useful in detecting and monitoring changes in the teat(s) and mammary gland(s); it provides information about the structure of the udder, whilst being non-invasive, non-ionising, rapid and painless.

Ultrasonographic imaging of the teats can be carried out with ewes in the standing position, with an ultrasound scanner fitted preferably with a 12.0 MHz linear transducer (Franz et al., 2001; Lazaridis et al., 2012), although use of a 6.0 MHz sector transducer has also been reported (Mavrogianni et al., 2004). For improved quality images, teats should be immersed into a plastic cup filled with water (Lazaridis et al., 2012). Nevertheless, successful imaging after direct application of the transducer directly on a teat has been reported (Franz et al., 2001; Mavrogianni et al., 2004); in that case, it is advisable to fill the teat with milk during scanning. Sonograms can be obtained in the vertical and the horizontal plane. The probe should be moved along the axis of the teat starting from its base towards the orifice, in order to image the teat cistern and the teat duct. The entire teat can be imaged in one field per level. The method is particularly useful to identify lesions within the teat, e.g., fibrous tissue that develops in case of stenosis (Mavrogianni et al., 2004), and complements well the clinical examination.

Ultrasonographic imaging of the mammary parenchyma is performed by direct contact of the transducer on the udder skin. For improvement of the image, it is advisable to clip hair on the udder skin. Images are taken on the horizontal or the vertical plane. The probe is placed on the caudal surface of each mammary gland, along its longitudinal axis, and is moved from left to right to evaluate the parenchyma; 80 and 120 mm scanning depths can be used. Then, the probe is moved upwards and downwards to image the *sinus lactiferous* of the gland at the base of the teat. The procedure may be repeated after placing the probe on the lateral surface of the udder. Then, the entire procedure is restarted for the other mammary gland and, finally, paired transverse sections of the udder can be taken. The mammary parenchyma is imaged as a homogeneous, echogenic structure, with some anechoic areas therein, which correspond to large milk ducts or to vessels. The *sinus lactiferous* is imaged as an anechoic to hypoechoic structure at the lower part of the mammary gland (Ruberte et al., 1994). The technique is useful for identification of structures, which, due to their size and location, cannot be identified during clinical examination, as well as for differentiation of structures identified in the parenchyma (e.g., haematoma, abscess, granuloma) (Lazaridis et al., 2012).

Benefits of using Doppler ultrasonography in the udder of ewes as a diagnostic method have not yet been fully evaluated.

2.2.2. Endoscopy

Teat endoscopy is useful for diagnosis of teat lesions (Kiossis et al., 2009, 2012). The technique is performed by insertion of the endoscope into the teat through the teat orifice and teat duct or through the teat skin (Kiossis et al.,