

DIAGNOSTIC SURVEY ON CONTAGIOUS EPIDIDYMITIS OF RAMS IN PIEDMONT (ITALY) *

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SUMMARY: *Brucella ovis* is the causative agent of contagious epididymitis of rams. It produces a clinical or subclinical disease in sheep that is characterised by genital lesions in rams and the main consequence is reduced fertility.

We have tested 634 animals by serological test (CFT). Two positive rams were slaughtered and their samples were further processed with other laboratories researches: bacteriological examination and PCR.

Twenty eight animals among 634 were resulted seropositive, both male and females.

Bacteriological testing has elicited the presence of *Corynebacterium* spp in both rams and the presence of *Brucella* spp in one of two rams.

Keywords: Ram, *Brucella ovis*, serology, bacteriology, PCR.

RESUME : *Brucella ovis* est l'agent de l'épididymite contagieuse du mouton. Elle peut produire une maladie clinique ou subclinique chez les ovins, est caractérisée par des lésions génitales chez le mouton et, en conséquence, entraîne une fertilité réduite. Nous avons testé 634 animaux par fixation du complément.

Sur la totalité des animaux, 28 mâles ou femelles ont fourni un résultat positif.

Les deux béliers positifs ont été envoyés à l'abattoir et soumis à des examens bactériologiques et à la PCR.

La bactériologie a permis d'isoler *Corynebacterium* spp. sur les deux béliers et *Brucella* spp sur l'un d'eux.

Mots-clés : Bélier, *Brucella ovis*, sérologie, bactériologie, PCR.



* Communication affichée lors des Journées AEEMA, 19-20 mai 2005

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I - INTRODUCTION

Brucella ovis is the causative agent of contagious epididymitis of rams. It produces a clinical or subclinical disease in sheep that is characterised by genital lesions in rams and the main consequence is reduced fertility.

The disease is world-wide distributed and in Europe has been reported in France, Germany, Hungary, Romania, Russia, the Slovak Republic, Spain, but probably occurs in most sheep-raising countries [AA. VV., 2000] and in Northern Italy.

In 1998, after having done researches in order to establish the prevalence of epididymitis in rams on national territory, we pointed out in 2003 many positive serologic reactions to *B. ovis*.

Flocks in which we have diagnosed the infection, were officially brucellosis free since 1996 and usually go to mountain pasture during summer in regional territories.

II - MATERIAL AND METHODS

We have tested 634 animals by serological test (CFT). Two positive rams were slaughtered and their samples were further processed with other laboratories researches: anatomopathological testing to elicit the presence, on these two animals, of the purulent orchids-epididymitis. Further more we used bacteriology and molecular biology techniques on tissue samples (spleen, testicles, epididymus) considering the difficulties in isolating *B. ovis*.

Serology – We used technique according to bibliography [Blasco, 1990] and optimized by National Health Superior Institute.

Bacteriology – Tissue were macerated in sterile saline in a Stomacher. The whole material was inoculated onto selective medium developed by Farrell [Farrell, 1974], with the addition of 10% horse serum and incubated at 37°C in an atmosphere containing 5% CO₂ for 15 days. All colonies resembling *Brucella* were subcultured onto blood agar medium with 5% sterile ovine blood and incubated for a further 2 days before re-examination. If *Brucella* was suspected using Stamp's staining [Alton *et al.*, 1992], then the colonies were identified to species by classical techniques [Alton and Jones, 1967].

III - RESULTS

Twenty eight animals among 634 were resulted seropositive, both male and females. Bilateral orchids-epididymitis (ascessual form) was detected on slaughtered rams at necropsy.

Bacteriological testing has elicited the presence of *Corynebacterium* spp in both rams

and the presence of *Brucella* spp in one of two rams. Species identification by PCR [Casanas *et al.*, 2001] done in Brucellosis Reference Centre Laboratories, confirmed our diagnosis and the presence of *B. ovis*. Molecular biology has detected *Brucella* spp on two examined rams.

IV - DISCUSSION

Indirect diagnosis based on serological tests is preferred for routine diagnosis.

The demonstration of the existence of genital lesions (bilateral orchid-epididymitis) by palpating the testicles of rams was indicative of the presence of *B. ovis* infection in this flock.

However, this clinical diagnosis is not sensitive enough because only about 50% of rams infected with *B. ovis* present epididymitis [Blasco, 1990]. Moreover, the clinical diagnosis is extremely unspecific due to the existence of many other bacteria causing clinical epididymitis, e.g. *Corynebacterium* spp.

V - CONCLUSIONS

Bacteriological and molecular biology methods let us to confirm the presence of *B. ovis* on two seropositive rams. This case-report is an

important contribute for evaluation of *B. ovis* presence in Italy.

BIBLIOGRAPHY

AA.VV. - Ovine Epididymitis (*Brucella ovis*) in Manual of Standards Diagnostic Tests and Vaccines, OIE, 2000.

Alton G.G., Jones L.M. - Laboratory Techniques in *Brucella*, WHO, 1967.

Alton G.G., Jones L.M., Angus R.D., Verger J.M. - Techniques for the Brucellosis Laboratory. INRA, Paris, France, 1988. Baily G.G. *et al.* - Detection of *Brucella melitensis* and *Brucella abortus* by DNA amplification. *J. of Trop. Med. And Hyg.*, 1992, **95**, 271-75.

Blasco J.M. - *Brucella ovis*. In Animal Brucellosis, Nielsen K. & Duncan J.R., eds. CRC Press, Boca Raton, Florida, USA, 1990, 371-378.

Casanas M.C. *et al.* - Specificity of a Polymerase Chain Reaction Assay of a Target sequence of the 31 kD *Brucella* Antigen DNA used to diagnose Human Brucellosis. *Eur. J. of Clin. Microbiol. & Inf. Dis.*, 2001, 20 (2), 127-31

Farrell I.D. - The development of a new selective medium for the isolation of *Brucella abortus* from contaminated sources. *Res. Vet. Sci.*, 1974, **16**, 280-86

