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POTENTIALLY PATHOGENIC MYCOPLASMAS IN THE EXTERNAL EAR CANAL OF CLINICALLY NORMAL CATTLE IN SOUTHEAST BRAZIL: FIRST REPORT

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ABSTRACT

Mycoplasmas were searched in the ear canal flushing of 60 bovine in Brazil. The prevalence obtained was 80%. The percentages of typified species were 12.5%, for *M. alkalenses*; 2.1%, *M. arginini*; 8.35%, *M. bovirhinis*; 2.1%, *M. bovis*; 25.0%, *M. conjunctivae*; 14.6%, *M. mycoides* subsp. *mycoides* LC and 10.4% *M. capricolum*.

Key words: Mollicutes, bovine, external ear canal, immunoperoxidase.

In the prokaryotes (Class Mollicutes), mycoplasma is the smallest and simplest self-replicating known bacteria that lack a peptidoglycan layer and have a low guanine plus cytosine content in the genome. Mycoplasmas are widespread in nature as parasites of humans, mammals, reptiles, fish, arthropods, and plants (14,15). These organisms have limited metabolic options for replication and survival. Consequently, they are extremely fastidious in their nutritional requirements (14). Many mycoplasmas are pathogenic to animals, humans and plants and they are, therefore, of great concern in human and veterinary medicine as well as in plant pathology. These organisms are often host-specific, and ruminants, especially cattle, harbour a number of different species (5,15). In livestock production these organisms cause economical losses, being *Mycoplasma mycoides* subsp. *Mycoides* SC, the etiological agent of Contagious Bovine Pleuropneumonia, that is from a global point of view, one of the most serious bacterial disease of animals. Others species like *M. bovis*, *M.*

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bovigenitalium, *M. alkalescens*, *M. dispar*, *M. conjunctivae* and *M. capricolum* are capable of infecting cattle and causing mastitis, arthritis, respiratory disease, disorders in the reproductive system and infectious keratoconjunctivitis (2,8,9,10,13). *M. bovirhinis* is a troublesome agent as a secondary invader in respiratory disease, besides being considered the most commonly isolated mycoplasma from the nasal cavity of cattle with respiratory disease (11,15). *M. bovigenitalium* is closely related to *M. bovis* biochemical and culturally. *M. bovis* is a pathogen isolated from otitis media and otitis interna that occur in both dairy and beef cattle (5). *M. alkalescens* was first isolated in cattle in Japan from the lungs of calves with respiratory disease in 1991 (10). In Brazil many mycoplasmas species have been isolated in cattle and goats herds (1,2,6,11,12). In spite of few cattle mycoplasmosis studies in Brazil, *M. bovirhinis* was isolated in vaginal mucus of heifers with symptoms of reproductive disorders, infertility and granular vulvitis. *M. bovis* was isolated in cases of mastitis in cows (11,13). As to goats mycoplasmosis, the species isolated from outbreaks in Brazil were *M. mycoides* subsp. *capri* and *M. mycoides* subsp. *mycoides* LC, being the disease characterized by pneumonia and others respiratory symptoms, keratoconjunctivitis and septic arthritis (9). *M. agalactiae* was isolated in two outbreaks occurred in the Northeastern of Brazil, with the signs of mastitis and agalactia in which the kids yielded fever, lameness, painful joints, anorexia, arthritis and conjunctivitis (2). Reports on infectious keratoconjunctivitis (IKC) also happened from goats flocks in Pernambuco and São Paulo States (1,6). IKC is a disease commonly known as pink-eye, a contagious disease of domestic ruminants, localized in the eyes and characterized by inflammation of the conjunctiva and cornea, caused by *M. conjunctivae*. Although cases of infectious keratoconjunctivitis by *M. conjunctivae*, have not still been described in cattle in this country. The goat's external ear canal can be an unusual source of mycoplasmas, including pathogenic species, where mycoplasmas have been found associated with ear mites (3,4,5,13). In cattle these conditions had not been evaluated yet. The objective of this study was to register the isolation of pathogenic mycoplasmas species in the external ear canal of clinically normal cattle slaughtered for human consumption.

Flushing were carried out in the external ear canal of 60 bovine at slaughter time in an abattoir of Rio de Janeiro State, Southeastern, Brazil. Sterilized syringes (60ml) loaded with buffer solution (PBS, pH 7.2) were used for the cattle ear canal flushing. The obtained samples were diluted in the glycerol (1:2) and stored at -20^{o}C until use. These samples were diluted up to 10^{-5}, inoculated in liquid and solid modified Hayflick´s medium and incubated at 37^{o}C for 2-3 days. Finally, the plates were transferred into a jar to attain a microaerophilia condition. The agar plates were observed every two days under stereomicroscope for the presence of typical colonies "fried-egg". Typical colonies were typified by the indirect immunoperoxidase test (7). Paper discs soaked in hyperimmune rabbit sera against *M. arginini, M. bovis, M. conjunctivae, M. mycoides mycoides* LC, *M. bovirhinis, M. alkalescens, M. capricolum, M. verecundum* and *M. gallisepticum*. The prevalence of *Mycoplasma* spp. in the in the external ear canal of the studied cattle was 80% (48/60). Concomitantly, *Raillietia* spp. was found in the ear of most of these animals. The percentage of *Mycoplasma* species isolated in the flushing ear canals of these bovine were 12.5% (6/48) for *M. alkalenses*; 2.1% (1/48), *M. arginini*; 8.35% (4/48), *M. bovirhinis*; 2.1% (1/48), *M. bovis*; 25.0% (12/48), *M. conjunctivae*; 14.6% (7/48), *M. mycoides* subsp. *mycoides* LC and 10.4% (5/48) for *M. capricolum*. In this study the animals were clinically normal for mycoplasmosis and, although some presented subclinical otitis, which is relatively

common in raillietiosis cases. The results confirm that the cattle's ear canal is also a mycoplasmas source, including pathogenic species.

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RESUMO

Micoplasmas potencialmente patogênicos no canal auditivo de bovinos clinicamente sadios no Sul do Brasil: primeiro relato

Foram pesquisados micoplasmas no conduto auditivo de 60 bovinos no Brasil. A prevalência obtida foi de 80%. A porcentagem das espécies tipificadas foi de *M. alkalenses*, 12,5%; *M. arginini*, 2,1%; *M. bovirhinis*, 8,35%; *M. bovis*, 2,1%; *M. conjunctivae*, 25,0%; *M. mycoides* subsp. *mycoides* LC, 14,6% e *M. capricolum*, 10,4%.

Palavra chave: Mollicutes, bovinos, conduto auditivo externo, immunoperoxidase

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