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Communication

[*Comunicação*]

Status of paratuberculosis in beef cattle In Rio Grande do Sul, Brazil

[*Status da paratuberculose em bovinos de corte no Rio Grande do Sul, Brasil*]

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Paratuberculosis, or Johne's disease, is an infectious disease caused by *Mycobacterium avium* subsp. *paratuberculosis* (MAP), a resistant acid-fast bacillus that generates granulomatous, incurable enteritis, which progresses to severe cachexia and death (Fernández *et al.*, 2017). The subclinical form of the disease is the most prevalent in herds due to the length of time that elapses before the characteristic clinical manifestations of paratuberculosis begin (Klinkenberg and Koets, 2015).

The disease is more prevalent in dairy herds than in beef herds (Roussel *et al.*, 2005). In the state of Rio Grande do Sul (RS), paratuberculosis has been diagnosed in both dairy (Driemeier *et al.*, 1999) and beef (Fiss *et al.*, 2015) cattle from the metropolitan and southeastern mesoregions, respectively. However, these studies address sporadic disease outbreaks in their clinical form and do not cover more than one herd per outbreak.

To date, there is no information on the epidemiological distribution of paratuberculosis in beef cattle herds with no history of the disease in RS. Thus, our objective was to verify whether paratuberculosis can be detected in herds of beef cattle from different mesoregions of RS using the ELISA test.

The present study was approved by the UPF Ethics Committee on the Use of Animals (CEUA), number 049/2019.

A cross-sectional observational prospective study was carried out from October 2021 to July 2022. Samples were collected from cattle breeding farms geared toward beef production. The selected farms were from the mesoregions of RS with the highest beef cattle production. Samples were collected from 10% of individuals within each herd tested. The age of the cattle varied between 18 and 30 months.

The following formula was used to identify the necessary sample size (Pfeiffer, 2010):

$$n = Z^2 \frac{P(1-P)}{D^2}$$

Where Z is the standard normal distribution value, corresponding to the desired confidence level (Z = 2.56 for 95% confidence intervals); P is the expected prevalence, which has been calculated at 45.5% nationwide (Yamasaki *et al.*, 2013), and D is the maximum acceptable error in the estimate, which we assigned as 0.05.

The target sample size was 357 animals. Based on the availability of farms and to maintain the 10% collection rate in each herd, a total of 393 samples were collected. Blood samples were centrifuged to obtain serum, on

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which the ELISA (Enzyme-Linked Immunosorbent Assay) test was performed using a commercial kit (Paratuberculosis Screening Ab, IDEXX Laboratories, USA) performed as directed by the manufacturer.

Of the 393 animals tested, only two samples were classified as suspicious (0.5%) and the remainder as negative. One of the suspicious samples was collected in Bagé and the other came from Santana da Boa Vista. The remaining samples were negative and belonged to herds from six mesoregions of RS, namely: Southwest (Bagé, 140), Northwest (Soledade, 10; Paim Filho, 29; Cacique Doble, 24; Sananduva, 13), Central-West (Júlio de Castilhos, 20; Santa Maria, 24), Central-East (Rio Pardo, 22; Arroio do Meio, 22), Metropolitan (Arroio dos Ratos, 10; Eldorado do Sul, 10; Capela de Santana, 10), and Southeast (Santana da Boa Vista, 22; Encruzilhada do Sul, 37).

The herds evaluated were composed of beef cattle raised in an extensive system, one in which cattle have a larger area per animal, which contributes to the low or absent MAP infection pressure. The life span of cattle on the farms studied is less than three years, while the average age of clinical manifestation of the disease is over four years, with greater intensity from six years and up. This results in a low probability of clinical cases and little MAP dissemination to the environment. Only beef cattle farms were sampled in this study, none of which had dairy herds present alongside the beef cattle. These factors support most negative cases in the present study.

The low seroreactivity of the tested herds is similar to studies in other countries. However, positivity rates of beef cattle studied in other countries are higher, ranging from 3% (Roussel *et al*., 2005) to 8.75% (Hill *et al.*, 2016), which may be due to the greater number of farms tested in the studies cited, in addition to the sampling rates in each herd. However, the seroreactivity of dairy herds in the state of RS has higher rates (5.6%) (Setim *et al*., 2023), compared with the seroreactivity of beef cattle in the present study. In this sense, the greater detection of the disease in animals reared with a higher concentration of cattle per area is emphasized. These data provide subsidies for further studies on the disease in herds in the state of RS.

The cattle were tested only once due to the management of the properties and the short cycle before slaughter. Thus, it was impossible to collect additional samples from the suspected animals to assess whether there were positive animals at a later date. However, positive animals are likely present in herds where suspected cases were found (Ozsvari *et al.*, 2020).

Paratuberculosis has already been diagnosed in the Southeast region in extensive beef cattle operations (Fiss *et al.*, 2015). Notably, this was where we found the two suspected cases in the present study, highlighting the importance of the Southeast mesoregion for future research, which could result in a fuller picture of the spread of the disease.

In conclusion, the detection of paratuberculosis in the tested herds was extremely low (0.5%). As such, a disease control program indicates considerable feasibility with viable and profitable applicability in the mesoregions studied. More studies are needed and are being developed to understand the epidemiology of the disease within the state's herds, as well as to support the development of a control program.

Keywords: antibodies, ELISA, MAP, Johne's disease

RESUMO

A paratuberculose em bovinos de corte já foi diagnosticada no estado do Rio Grande do Sul, mas relatada em apenas um rebanho. Nesse contexto, o objetivo desta pesquisa é verificar se a paratuberculose pode ser detectada em rebanhos de bovinos de corte de diferentes mesorregiões do RS, por meio do teste ELISA. As amostras foram coletadas de bovinos provenientes de seis

mesorregiões, entre elas as maiores produtoras de bovinos de corte do RS. Apenas duas amostras foram classificadas como suspeitas no teste de ELISA, as outras 391 amostras foram negativas. Os resultados demonstram baixa detecção da doença nas mesorregiões testadas, com apenas 0,5% das amostras com resultado suspeito. Assim, esses dados demonstram que a enfermidade é pouco frequente nos rebanhos testados e que um programa de controle pode ser implementado de forma viável e economicamente lucrativa.

Palavras-chave: anticorpos, ELISA, MAP, doença de Johne

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