



Bovine cervical bursitis co-infection caused by *Brucella abortus* and *Onchocerca* sp.

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Abstract The presence of *Onchocerca gutturosa* in cattle is responsible for lesions similar to those observed in cases suspected of brucellosis, however, *Onchocerca* sp. is not a trade barrier, although it is also responsible for economic losses due to the removal of the affected parts of the carcasses. *Brucella* sp. is a zoonotic agent transmitted to humans through the consumption of contaminated animal products, the contact with infected animals and the handling of carcasses. This agent is also responsible for non-tariff trade barriers. Cervical bursitis is *Brucella* sp. suggestive lesions in bovine carcasses that requires laboratory tests to confirm the diagnosis. The objective of this study was to record the co-infection of *Brucella abortus* and *Onchocerca* sp. as a first report of co-infection of these two

agents in the same lesion. The sample constituted of a nuchal bursitis in the cervical ligament, a suggestive lesion common to these two agents, submitted to histopathology and *Brucella* spp. isolation in the Brucellosis reference laboratory of the Brazilian Ministry of Agriculture, Livestock and Food Supply. Brucellosis serological diagnosis were also performed in the animal's serum sample. *B. abortus* was isolated from the lesion and filarid nematode structures were identified in histopathology. All serological tests were positive for brucellosis. Further studies are needed, however, to understand the co-infection by *Onchocerca* sp. and *B. abortus*.

Keywords Nuchal ligament · Co-infection · Cervical bursitis · Zoonosis · Onchocerciasis · Brucellosis

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Introduction

Many subcutaneous cervical lesions in cattle are caused by adult specimens of filarial nematodes of the genus *Onchocerca* spp., often observed in carcass inspection of animals from tropical regions of Brazil, such as the North, Northeast and Midwest (Almeida et al. 2000). *Onchocerca gutturosa* has a predilection to localize in the nuchal ligament (Cheema and Ivoghli 1978). The nematode is identified and classified through its morphology (Bowman et al. 2010) and PCR techniques (Ferri et al. 2009).

Cervical bursitis, detected only on *post-mortem* inspection, is most commonly associated with suspected cases of brucellosis, since *Brucella* sp. has an affinity for mucosal bags (Freitas and Oliveira 2005; Sola et al. 2014; Figueiredo et al. 2015).

Brucellosis is a contagious chronic infectious disease caused by bacteria of the genus *Brucella* spp. (Figueiredo

et al. 2015). It is a foodborne and occupational disease distributed worldwide and is more prevalent in developing countries (Brasil 2006). It is also considered an international trade barrier to animals and animal products (Sola et al. 2014).

Brucella abortus infection can be detected by isolation and identification of the agent in suggestive lesions found at *post-mortem* inspection, and the collected material can be submitted to bacteriological, molecular biology and/or histopathological diagnostic techniques (Alton et al. 1988; Sola et al. 2014; Macedo et al. 2018). Due to the high biological risks, *Brucella* spp. manipulation is restricted to biosafety level III laboratories (OIE 2018).

At sanitary meat inspection, lesions classified as suspected of occurrence of *Brucella abortus* leads to condemnation of the carcass and raises the possibility that the meat handlers might have been infected (Langenegger et al. 1975; Macedo et al. 2018). On the other hand, onchocerciasis is always neglected because it does not present itself as a barrier to exports (Santos et al. 2014). This agent is therefore a confounding factor in the judgment of carcass lesions in the slaughterhouse, which may be responsible for large economic losses for the meat industry, compromising the status of Brazil as the largest exporter of beef in the world (Santos et al. 2014; Macedo et al. 2018).

The aim of the present study was to report the first case of co-infection of *B. abortus* and *Onchocerca* sp. in a cervical lesion found in a bovine carcass at the slaughterhouse.

Case history and observations

Samples of blood serum and nuchal ligament lesions of a 60-month-old cow of the Nelore breed from the municipality of Cumarú do Norte (Pará State—Brazil) were sent to the Laboratory of Diagnosis of Bacterial Diseases of the National Agriculture and Livestock Laboratory as a suspected *B. abortus* infection by the Federal Meat Inspection Service (FIS). The serum samples were tested by the acidified antigen test (Rose Bengal), the slow serum agglutination test with 2-mercaptoethanol and Complement Fixation test (Alton et al. 1988; Brasil 2006). Lesions were submitted to bacterial isolation (Alton et al. 1988). Also, tissue samples were fixed in 10% formalin, embedded in paraffin and sections stained with haematoxylin and eosin (HE) to describe the lesion microscopically.

The serum sample of the involved animal was positive for brucellosis in the acidified antigen test (Rose Bengal) and showed a title of 200/200 in the slow serum agglutination test with 2-mercaptoethanol (SAT/2-ME). In the Complement Fixation Test (CF), it showed a title of 128. From the lesions identified at the sanitary inspection

procedures, it was possible to isolate and identify *B. abortus* biovar 2, corroborating the findings of serology.

Histologic examination of the lesions revealed the presence of adult nematodes morphologically consistent with genus *Onchocerca* spp. (Gardiner and Poynton 1996) within a chronic mild to moderate tissue inflammatory reaction (Fig. 1).

This co-infection was found just by chance while studying histopathological lesions, initially thought to be caused only by *Brucella* spp. Due to the fact that it was an unexpected finding it was not possible to carry out PCR to confirm the helminth identification because the lesion was not available anymore to perform the test nor was the PCR technique implemented in the laboratory at that time.

Discussion

Nuchal ligament lesions has been reported previously as caused by many agents, especially *O. gutturosa* and *B. abortus*, however, it has been treated as separated entities by bacteriologists and parasitologists (Langenegger et al. 1975; Cheema and Ivoghli 1978; Santos et al. 2014; Macedo et al. 2018). Nevertheless, this is the first time that cervical bursa co-infection by *Onchocerca* sp. and *B. abortus* is identified and recorded.

Based on *Onchocerca* spp. specific location, it is highly probable that the nematode found in the lesion described in this work belongs to *O. gutturosa* species (Cruz et al. 2012). It was suggested that the primary lesions caused by



Fig. 1 Histological section showing female adult filarid nematodes and the adjacent inflammatory reaction in the connective tissue of bovine cervical bursa. Note the cuticular annulations (arrows), paired reproductive tubes (arrowhead) and a very small intestine (short arrow), some of the morphologic features of the genus *Onchocerca*. (HE, obj. \times 10)

Onchocerca gutturosa could predispose to the local secondary infection by some infectious agents, including *Brucella abortus* (Costa et al. 2001). Nevertheless, it is not possible to establish a relationship between these two agents, based only on this report. Further studies and addition of new cases of bursitis co-infection by *Onchocerca* spp. and *Brucella* spp. can bring greater subsidies to confirm the association between these two pathogens.

Due to the possibility of co-infection, it will be necessary to carry out diagnostic tests to detect the presence of *Brucella* sp. or to rule out the infection in the carcasses, which show cervical bursitis, since brucellosis can lead to trade restrictions and pose a high sanitary risk.

According to this finding, a biological risk to parasitologists does exist while manipulating this kind of lesion without proper biosafety precautions, since parasitology laboratories usually require biosafety level II.

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Compliance with ethical standards

Conflict of interest The authors report no conflicts of interest.

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