Case Report

Use of ultrasound in a case of Actinomycosis in a Holstein Friesian cow

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Abstract

Actinomycosis is a sporadic disease of cattle, caused by *Actinomyces bovis*. In presence of oral injuries or openings, *A. bovis* could penetrate tissues and infect maxilla or mandible bones. A Holstein Friesian cow was presented for inappetence, weight loss and a left-sided mandibular swelling. A unilateral swelling was visible on the left jaw as a ventro-lateral and circular in shape. A fine-needle aspiration was collected. Ultrasonography examination was performed. Bacteriological analysis was negative. Due to the history, actinomycosis diagnosis was suggested and the cow was culled. Pathological examination revealed lesions indicative of chronic pyogranulomatous mandibular osteomyelitis with intralesional colonies of gram-positive bacteria, consistent with infection due to *A. bovis*. The importance of an early diagnosis was here highlighted . Since the bacterial culture is not always easy to perform and is usually time-consuming, in case of suspected actinomycosis, the ultrasonographic findings may orient the clinician decision to start adequate and earlier therapy. Further research is needed to evaluate the sensitivity and specificity of ultrasonography for diagnosing actinomycosis.

Keywords: *Actinomyces bovis*, dairy cows, lumpy jaw, ultrasonographic evaluation

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Introduction

Actinomycosis (lumpy jaw) is a sporadic disease of cattle, caused by *A. bovis*, a gram positive, non-encapsulated, filamentous organism which is normally part of the oral and digestive tract microbiota of the ruminant species (Smith, 2015; Peek *et al.*, 2018).

In presence of oral injuries or openings, caused by coarse fibrous feeds, hard plant awns or gingival and dental disease, *A. bovis* could penetrate tissues and infect maxilla or mandible bones (Smith, 2015). The bacterium can also penetrate across contaminated skin wounds and around the alveoli of the teeth (Peek *et al.*, 2018).

A. bovis can cause severe osteomyelitis of the mandible, or less frequently of the maxilla (Smith, 2015), determining an increased culling rate in the bovine herd. Clinically, actinomycosis appears as a warm and painless swelling area on mandible, without other clinical signs. The swelling area slowly increases and becomes harder and usually painful at the digital palpation. The osteomyelitis caused by this bacterium could lead, in untreated cows, to the deformation of infected bones with important problems to the anchored teeth (Peek *et al.*, 2018).

Ultrasonography of the soft tissues is usually indicated when a soft tissue swelling is palpated or identified radiographically. High frequency ultrasonography is considered a useful tool for the evaluation of the skin and other superficial soft tissues affected by focal or diffuse conditions (Rossi, 2011).

In veterinary medicine, computed tomography and radiographic examination are considered the gold standard to fully characterize osseous abnormalities of the head, but they have limited availability in the field. Since many soft tissue structures of the head are superficially located and easily accessible, the ultrasonography is considered a valuable tool in field settings for the diagnosis of these diseases. Although there are many publications describing the use of ultrasound for examination of specific abnormalities or focused regions of the equine head (Rodriguez *et al.*, 2007; Ali, 2000), the same has not yet been described in the bovine.

Case report

A 3-years-old first lactating Holstein Friesian cow was presented for inappetence, weight loss and a left-sided mandibular swelling. At the admission time, the cow was 273 days in milking (DIM) and was not pregnant. The cow belonged to a dairy farm consisting of approximately sixty lactating cows bred in free housing system. Lactating cows were fed with total mixed ration (TMR) two times per day.

At the admission time, the cow appeared alert and responsive. A unilateral swelling was visible on the left jaw as a 15 cm-circular mass at the ventro-lateral area. No cutaneous wounds or fistula were visible, and the swelling area was hard, warm, immovable and painful on digital palpation (Fig. 1). The cow was in poor general condition, with a BCS of 2.25/5 and showed a profound salivation and inappetence. The cow presented a slightly reduced rumen contractions and feces with indigested materials. These findings were probably caused by the reduced capacity of chewing the food due to the swelling area. The hematologic and biochemistry parameters were normal.

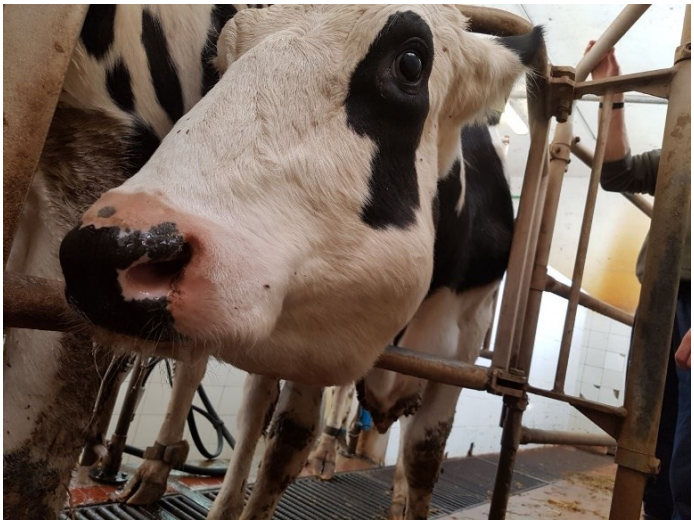

Figure 1 Swelling area on the left jaw visible as a ventro-lateral, circular in shape and about 15 cm of diameter in size mass in a 3-years-old first lactating Holstein Friesian cow.

The farmer reported that the swelling area appeared 2 months before and have been increasing in size, slowly but progressively. The owner also reported a similar case in a lactating cow six months before, which was slaughtered for fertility problems. Blood samples were collected from the jugular vein for running a complete cell blood count (CBC) and biochemistry analysis. Ultrasonography (Z5 Vet, Mindray, China) and X-ray (HF100 M, Gierth, Germany) were performed to examine the swelling area. Radiographs were then reviewed using viewer software program (Horos, Horosproject.org, USA). A convex probe (4.5 MHz) was used. The whole part was scanned in longitudinal and transverse view for evaluating the content, the edges and the layers involved. The radiographic examination was performed using a laterolateral (kVp 66 and mAs 8) and a ventrodorsal (kVp 73 and mAs 8) view. The ultrasonography of the swelling revealed a circular and well-defined structure in the subcutaneous layer,

delineated by a thick, hyperechoic peripheral wall. The content of this structure had a mixed echogenicity, mainly hyperechoic (Fig. 2). The ultrasonographic image was suggestive of an abscess, a granuloma or an organized hematoma. At the radiographic examination, a wide, smooth, continuous, and fusiform periosteal reaction on the ventral edge of the left mandible's body was found. Multiple, circular and radiolucent areas with clear edges were on the body of the mandible (Fig.3). These alterations were suggestive of osteomyelitis.

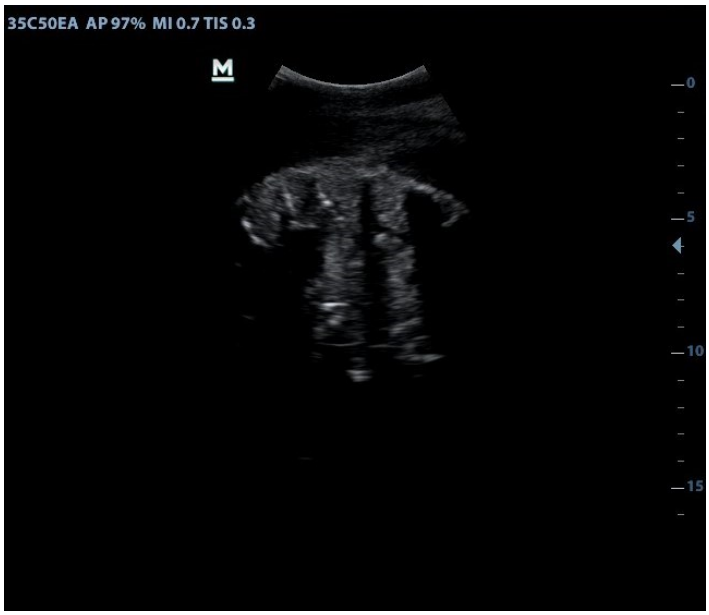

Figure 2 Ultrasound of the swelling area visible on the left jaw of a 3-years-old first lactating Holstein Friesian cow. The content of this structure had a mixed echogenicity, mainly hyperechoic. The ultrasonographic examination was made using a convex probe (4.5 MHz) (Z5 Vet, Mindray, China).

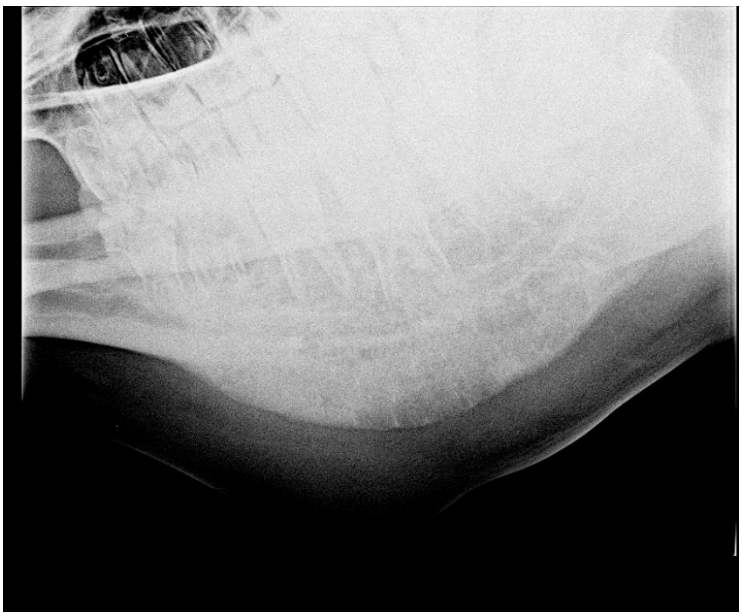

Figure 3 Radiographic examination (HF100 M, Gierth, Germany) performed using a laterolateral view (kVp 66 and mAs 8) of the swelling area visible on the left mandible's body in a 3-years-old first lactating Holstein Friesian cow.

Finally, samples for the bacteriological examination were collected aseptically from the same site by the fine-needle aspiration, using a 20-gauge, 4 cm needle. Bacteriological examination was performed according to protocol suggested by Militerno (2008) and the result was negative. The most important differential diagnosis included actinomycosis, osteomyelitis induced by other pathogens and neoplasia. Due to the poor prognosis, the farmer decided to cull the cow. For completeness of the case, the head of the cow was taken from the slaughterhouse and submitted for pathologic examination. On cut section, there were numerous encapsulated granulomas, up to 5 cm in diameter, in the subcutaneous tissues and mandibular muscles (Fig. 4a). Small abscesses and sequestrated trabeculae of affected bone were also found within the lesions, accompanied by extensive proliferation of granulation tissue. The abscesses contained numerous yellowish particles of less than 1 mm in diameter, interpreted as sulphur granules. After removal of all soft tissues,

necrosis and proliferation of bone were observed giving rise to an enlarged, honeycomb appearance of the body of the mandible (Fig. 4b).

Histologically, most of the mandibular bone was replaced by fibrovascular tissue surrounding many small abscesses with granules composed of large bacterial colonies. Granules were surrounded by radiating eosinophilic clubs, interpreted as Splendore-Hoeppli material.

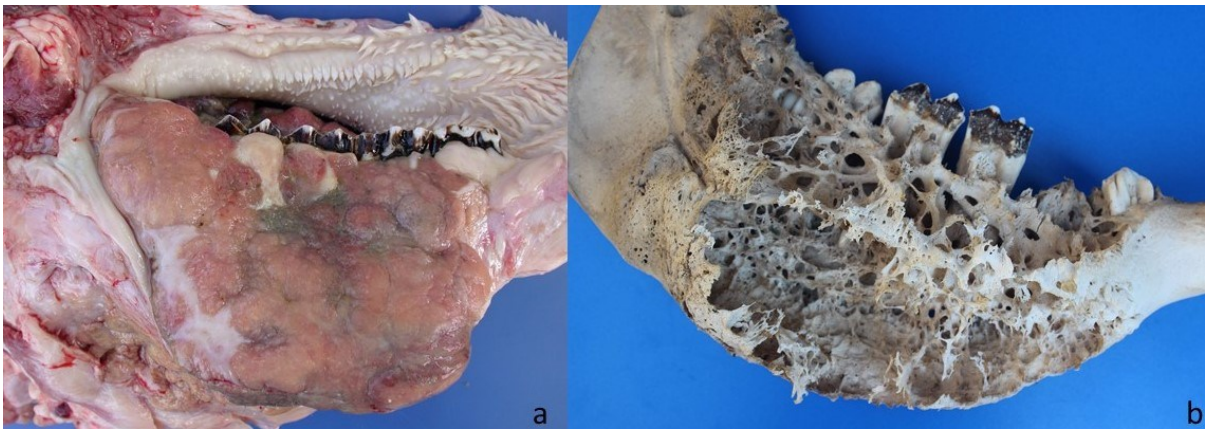

Figure 4a Ventro-lateral aspect of the left mandible in a 3-years-old first lactating Holstein Friesian cow. Numerous granulomas in the subcutaneous tissues and muscles of the mandibular region.

Figure 4b Left mandible of a 3-years-old first lactating Holstein Friesian cow. Honeycomb appearance of the mandibular body caused by extensive necrosis and concurrent proliferation of bone tissue.

The lesions were indicative of chronic pyogranulomatous mandibular osteomyelitis with intralesional colonies of gram-positive bacteria, consistent with infection due to *A. bovis*.

Given the farmer history, a herd visit was also performed. At the time of the inspection, all the animals were in good general conditions and the milk production was in line with the average reference production of a dairy herd. At food inspection of the lactating group, a very fibrous hay, composed mostly by hard plant awns and stickers, was found in the TMR. A complete report was written and sent to the farmer a week later.

Discussion

Actinomycosis is usually a sporadic disease, but it could be an epidemic or endemic herd problem, especially related to the presence of an increasing risk of oral injuries which lead to *A. bovis* penetration into the tissues (Militerno, 2008; Smith *et al.*, 2015). In the farm involved in this study, the occasional use of a fibrous and low-quality hay could have played a key role in the development of actinomycosis.

Given the complexity of the therapy and costs, preventive actions are mandatory. The owner was strongly encouraged to use always excellent quality hay, avoiding the use of forage rich in thorns, stickers and hard plant awns.

The diagnosis of actinomycosis in cows is mainly based on radiographic and bacteriological examination (Peek *et al.*, 2018). The involvement of *A. bovis* was not confirmed by the bacteriologic analysis, however, many authors reported that a conclusive diagnosis made by bacterial culture is rare to achieve (Smith, 2015; Peek *et al.*, 2018).

In the present case, the ultrasonography was easy to perform in field condition, not time consuming and the image was clear and easy to evaluate. The ultrasonographic examination could be also made by a reproductive linear probe which represents an essential tool for the bovine practitioner. The imaging recorded, along with a proper history, should immediately suggest a case of actinomycosis and encourage a conclusive diagnosis through the bacteriological examination.

In conclusion, due to the importance of an early diagnosis of *A. bovis* mandibular osteomyelitis, ultrasound could provide a supportive evidence of *A. bovis* infection. This is important since *A. bovis* is not always easily detected by bacteriological examination. Compared to X-Ray, ultrasound is ~~also~~ easier and safer to perform. Ultrasound can be made by one operator which is a more suitable scenario for those clinicians working in the field. Moreover, an X-Ray machine showed higher cost and its use appear to be more restricted for a field practitioner compared to an ultrasound.

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