Atypical Actinobacillosis in a Dairy Cow

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Abstract

Actinobacillosis is an infectious, chronic, generally non-fatal disease caused by Actinobacillus lignieresii. The etiologic agent is a part of the oral flora and causes pyogranulomatous lesions of the soft tissues. A 5-year old, approximately 500 kg cross-breed Holstein cow was presented for the treatment of a large, ulcerated and hemorrhagic mass at the left side of the neck in vicinity of the jugular furrow. Anamnesis indicated that the condition began three months previously as a small, walnut-size swelling that gradually enlarged over this period. After surgical resection of the mass, histological assessment revealed multiple pyogranulomatous foci-contained radiating eosinophilic clubs surrounded by many neutrophils, lymphocytes, plasma cells, macrophages and connective tissue. Clinical manifestations, bacteriological and pathological examinations of biopsy confirmed atypical actinobacillosis. In this case, the route of entry of organisms probably had been an abrasion or wound in the skin of the neck.

Key words: Actinobacillus lignieresii, Pyogranulomatous, Bacteriological, Pathologic, Biopsy

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Introduction

Actinobacillosis, in the typical presentation, a potentially zoonotic disease caused by Actinobacillus lignieresii, is a sporadic, inflammatory disease of the soft tissue of the gastrointestinal tract in cattle, sheep, goats and other species (Carmalt et al., 1999; Muhammad et al., 2006; Brown et al., 2007; Radostits, 2007), commonly known as “wooden tongue” in cattle. The causative organism is a Gram-negative bacterium, normally present as part of the oral flora (Quinn, 2002) capable to invade mucosal surfaces following trauma caused by abrasive ingesta or the action of the teeth during mastication (Radostits, 2007). Strains of A. lignieresii are reported to vary in their ability to cause disease, both in natural and experimental infections. Cattle, particularly young animals, are very susceptible following experimental subcutaneous inoculation, and lesions (small abscesses) begin to develop within a few days. Lesions begin with an initial leukocytosis followed by formation of a granulomatous reaction with epithelioid cells and some giant cells in the centre. In cattle, the disease typically involves the formation of pyogranulomas in the oral cavity, tongue or fore-stomachs with subsequent spread to regional lymph nodes (Rycroft and Garside, 2000). The involvement of other organs is considered to be atypical (Aslani et al., 1995, Holzhauer and Roumen, 2002). In the atypical form of the disease, the skin of the head, neck and, occasionally, the limbs may be affected (Radostits, 2007). The infection develops following trauma that is capable of altering the integrity of the barrier of the skin (Hebeler et al., 1961). Clinically, lesions appear as nodules, multiple abscesses, ulcers or draining fistulae and characterized by the presence of granulomas with pus-containing small, hard yellow to white granules (Melendez et al., 1999). An epizootic type of actinobacillosis has also been described in dairy heifers appearing as subcutaneous granulomas (Campbell et al., 1975). The diagnosis of wooden tongue is chiefly based upon the isolation and identification of the causative organism.

The present case report describes the clinical manifestations, bacteriologic and pathologic characteristics of atypical actinobacillosis in a mature dairy cow.

Case Description

A 5-year old, approximately 500 kg Cross-breed Holstein (from a dairy with 185 cows) cow was referred to the Department of Clinical Studies, School of Veterinary Medicine, Shiraz University, Iran, for the treatment of a large, ulcerated and hemorrhagic mass at the left side of the neck in vicinity of the jugular furrow (Fig 1). The size of the mass was 11×7×4 cm in diameters. History indicated that the condition began three months previously as a small, walnut-size swelling that gradually enlarged over this period. Physical examination revealed drooling of normal to food-tinged saliva, low quality and quantity of rumen contraction, poor body condition and weight loss of approximately 80 kg. Presence of different degrees of ulceration and hemorrhages on a firm mass was noticed on the left side of the neck in the vicinity of the jugular furrow. The cow was alert, no fever or abnormalities in respiratory and heart rate were observed. The ulcerated and hemorrhagic mass was removed by surgery. After surgical resection of the mass, haemorrhage
was controlled using electrocautery and ligating the large vessels. Antimicrobial treatment was administered for a period of two weeks postoperatively. The cow was treated with parenteral antimicrobial administration of one week each penicillin–streptomycin combination and amoxicillin–clavulinic acid.

The case was followed for six months postoperatively, and no recurrence or new growth was observed. Surgically removed mass and a biopsy of subcutaneous lesions were taken for bacterial culture and histopathological evaluation. Initially, impression smears were prepared from the mass and assessed after Giemsa and Gram staining methods. *Actinobacillus lignieresii* was identified on the basis of colony morphology; microscopic Gram and Giemsa stain characteristics and biochemical tests. For bacteriological examinations, non-discharging lesions were cultured on blood agar and MacConkey agar. The biochemical characteristics and results of biochemical reactions were interpreted following guidelines Lentsch and Wagner (1980). The tissue specimens were fixed in 10% neutral buffered formalin, dehydrated in graded ethanol, cleared in xylene, and embedded in paraffin wax. Sections of 5 µm thicknesses were stained by hematoxylin and eosin (H&E), Ziehl-Neelsen and modified Brown-brenn Gram stains and assessed microscopically.

**Results**

Gram stained smears revealed the presence of small gram negative cocobacilli. Giemsa stained smears showed club colonies, a lot of karyorrhectic, normal neutrophils, a number of cocobacillar bacteria and few reactive macrophages.

Bacteriologically, the growth was observed after incubation under both aerobic and anaerobic conditions, and the isolates were identified using biochemical methods. Small, smooth, non-haemolytic and translucent colonies after 48 hours appeared on the blood agar medium. Growth of bacteria was also present on MacConkey agar. A brief description of the biochemical tests was as follow: catalase and oxidase, urease, sucrose, glucose, galactose, arabinose, mannose and maltose were positive, whilst trehalose, lactose, hydrolysis of aesculin, Methyl red, Voges-Proskauer and indole tests were negative. Colonies were consistent with *A. lignieresii*.

Grossly, the mass contained yellowish white foci measuring 1-2 cm in diameter within a dense fibrous connective tissue. The histopathological examination of the tissue specimen revealed multiple pyogranulomatous foci within the dermis and subcutis centred on densely clustered basophilic bacilli surrounded by eosinophilic rosettes (Fig 2). Bacterial colonies and eosinophilic rosettes in the
Figure 1: Ulcerated and hemorrhagic mass at the left side of the neck in vicinity of jugular furrow. This mass contained yellowish white foci measuring 1-2 cm in diameter within a dense fibrous connective tissue.

Figure 2: Multiple pyogranulomatous foci within the dermis and subcutis centred on densely clustered basophilic bacilli surrounded by eosinophilic rosettes. Pyogranulomatous lesions were surrounded by reactive cells included neutrophils, lymphocytes and macrophages (H & E stain ×80).
pyogranulomatous lesions were surrounded by reactive cells included neutrophils, lymphocytes, macrophages and fibrous connective tissue. Between these pyogranulomas, chronic active inflammatory cells containing mixed populations of lymphocytes, plasma cells, macrophages, and few neutrophils in the connective tissue were observed. Brown-brenn Gram staining of affected tissues showed Gram-negative coccobacilli in the center of pyogranulomas (Fig 3). No acid-fast bacteria were demonstrated by Ziehl-Neelsen staining.

**Discussion**

Bovine actinobacillosis has usually been identified as a sporadic, insidiously developing granulomatous condition (Smith, 2009). Infections and pathological developments are the product of tissue trauma, lesions or prolonged irritation. It is believed that the causal agent penetrates the tissues of the mouth through lesions caused by foods or foreign objects. The organism gains entry to deep tissues via traumatic erosions, ulcers and penetrating lesion induced by hard fibrous hays, straw and plant awns (Radostits et al., 2007, Brown et al., 2007, Smith, 2009). In the cow of this report, actinobacillosis pyogranulomas occurred unusually in subcutaneous tissue of the left side of the neck in vicinity of the jugular furrow. The distribution of lesions over the distal limbs suggests the organism was transmitted through infected saliva during autogrooming, with the pathogen entering the skin through small percutaneous abrasions or wounds. In support to this hypothesis, *A. lignieresii* has been reported in infected wounds of human cases that were bitten by horse and sheep (Dibb et al., 1981; Peel et al., 1991). Young cattle in particular, are susceptible
following subcutaneous inoculation and lesions can develop in a number of days (Rebhun, 1988; Rycroft and Garside, 2000), in the present report, the infected cow was five-year old and the route of entry of organisms probably had been an abrasion or wound of the skin of the neck. Atypical bovine actinobacillosis including granulomas or abscesses formation in the skin and associated soft tissues and lymph nodes, especially in submaxillary region has been reported in cattle (Aslani et al., 1995, Holzhauer and Roumen, 2002). Progress of the granulomatous lesions is slow and chronic. In this report, history indicated that the condition began three months previously as a small, walnut-size swelling that gradually enlarged over this period. Infection can disseminate to the lymphatics and may sometimes spread to deep organs. Although actinobacillosis in cattle is best known as a disease of the tongue, similar to our report, the infection may occur in any of the exposed soft tissues, especially those of the mouth and neck; occasionally it involves the portion of the skin. Taghipour Bazargani (2010) reported six cases of atypical bovine actinobacillosis from a dairy herd occurred unusually in tongue shaft of heifers and soft tissues of oral cavity as well as skin of lower jaw in a short period of time with history of feeding by oat and wheat straw having plant awns as well. In another study, lesions containing odorless pus, were located in the subcutaneous soft tissues of the head and neck, specially the area of the parotid and sometimes the tongue (Rycroft and Garside, 2000). In this report, actinobacillosis granulomas occurred unusually in the dermis and subcutis.

In opinion of authors of this article, skin insults due to skin abrasion with *A. lignieresii* contaminated surface were the most common cause for the atypical actinobacillosis development in this dairy cow.

The case presentation should lead to a recommendation that in future atypical actinobacillosis should be included on the list of differential diagnoses for integumentary or subcutaneous swelling of the neck.

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References


