



CASE REPORT

Severe *Corynebacterium pseudotuberculosis* Infection in a Horse

Areeg Mohamed Al-Mubarak, Yassir Adam Shuaib and Mukhtar Taha Abu-Samra*

College of Veterinary Medicine (CVM), Sudan University of Science and Technology (SUST), Khartoum North, Sudan

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ABSTRACT

A 9-year-old, male draught horse was brought to the Veterinary Teaching Hospital of the Sudan University of Science and Technology for treatment of widespread skin lesions. The horse was anemic and in poor bodily condition. Encapsulated abscesses, nodules and ulcerated lesions were seen on different parts of the horse's body, but were mostly seen on the pectoral region, ventral abdomen, and all four limbs. The horse had sluggish movement and showed swelling and lameness of the right hind limb. The nodules and abscesses discharged viscid purulent exudate. *Corynebacterium pseudotuberculosis* was isolated in pure culture from aspirates of intact encapsulated abscesses and identified in accordance to its cultural, morphological and biochemical characteristics. Fecal examination was negative for internal parasites and complete blood count showed deviation from normal parameters. Histological examination of skin biopsy specimens revealed severe pathological changes. The horse was euthanized and post-mortem examination revealed no internal lesions other than a small fibrous adhesion between the liver and the diaphragm, and the presence of little pathogen-free yellowish fluid in the abdominal cavity. Based on clinical and laboratory findings two forms of the disease caused by *Corynebacterium pseudotuberculosis* were recognized in this horse: "pigeon's breast" and ulcerative lymphangitis.

*Corresponding Author

Mukhtar T. Abu-Samra
mukhtarabusamra@hotmail.com

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INTRODUCTION

Corynebacterium pseudotuberculosis is the classical cause of ulcerative lymphangitis or "pigeon fever" (Spier, 2006; Radostits *et al.*, 2007; Zavoshti *et al.*, 2009). This organism is a gram-positive pleomorphic rod-shaped, intracellular, facultative anaerobe with worldwide distribution (Spier, 2006). Infection with *Corynebacterium pseudotuberculosis* has been reported in sheep, goats, cattle, buffalo, camelids, equids, and humans (Spier, 2006). The portal of entry of this soil-borne organism is thought to be through abrasions or wounds in the skin or mucous membranes. Many insects have been incriminated as vectors for the transmission of the disease to horses, and recent studies have incriminated *Haematobia irritans*, *Musca domestica* and *Stomoxys calcitrans* as vectors of this disease (Pratt *et al.*, 2005; Spier, 2006; White, 2006; Radostits *et al.*, 2007). *Corynebacterium pseudotuberculosis* possesses a cytotoxic surface lipid coat that appears to facilitate intracellular survival and abscess formation. It also produces a phospholipase exotoxin that increases vascular

permeability, has an inhibitory effect on phagocytes and may facilitate spread of infection in the host (Spier, 2006; Radostits *et al.*, 2007).

The most common clinical form of the disease, characterized by external abscesses in the pectoral or ventral abdomen of horses, is often called "pigeon fever" due to the swelling of the horse's pectoral region resembling a pigeon's breast (Spier, 2006). Additional sites for abscess formation are the prepuce, mammary gland, axilla, limbs, and head (Pratt *et al.*, 2005; Spier, 2006). Other less common areas are the thorax, neck, parotid gland, guttural pouches, larynx, flanks, umbilicus, tail, and rectum (Spier, 2006). The disease is characterized by pyogranulomatous abscessation at the site of infection with lymphadenitis and abscessation along drainage lymphatic tracts (Radostits *et al.*, 2007). In the Sudan ulcerative lymphangitis has been diagnosed solely on the clinical appearance of the disease (Annual Reports of the Sudan Veterinary Services (1922-1973). Moreover, ulcerative lymphangitis caused by *Corynebacterium pseudotuberculosis* was reported in the Sudan in 1980 in an 8-year old horse (Abu-Samra *et al.*, 1980).

MATERIALS AND METHODS

Clinical and Laboratory Investigations

A 9-year-old male draught horse was brought to the Veterinary Teaching Hospital (VTH) of the College of Veterinary Medicine (CVM), Sudan University of Science and Technology (SUST) in July/2012. The main complaint according to the owner was severe and extensive skin lesions involving the whole body. Four months before submission for examination; the owner observed solitary nodules involving the lateral and medial aspects of the fore and hind quarters, neck, chest and abdomen.

After carrying out a thorough clinical examination, blood was collected from the jugular vein, complete blood count (CBC) was conducted; the values obtained were expressed in the systeme international d'unités (SI) and compared with the range of values documented by Blood and Studdert (1999). Fecal specimens were examined for internal parasites. Impression smears were prepared from abscesses and stained with Gram's stain. Aspirates of pus were aseptically collected from intact encapsulated abscesses, and abdominal fluid was aseptically collected during post-mortem examination were subjected to bacteriological examination and culture in accordance to Barrow and Feltham (2003).

Biopsy and necropsy specimens were fixed in 10 per cent formal saline, processed, embedded in wax, cut at 5-6 μm and stained with haematoxylin and eosin stain for histological examination.

RESULTS

The horse was in poor bodily condition. Encapsulated abscesses, crust covered and ulcerated lesions were seen on different parts of the horse's body, but are mostly seen on the pectoral region (Fig. 1), ventral abdomen (Fig. 2), and all four limbs extending from elbows down to hooves (Fig. 3) and from thighs down to hooves (Fig. 4). Abscesses and ulcers were also seen on the head concentrating round the mouth commissure (Fig.3) the axilla (Fig. 5) and prepuce (Fig. 6), and solitary lesions were also encountered on the neck (Fig. 7), thorax, flanks and rectum. Enlarged draining lymphatics with many encapsulated abscesses developing along them; were seen on the thorax extending between the elbow joint and sternum (Fig. 8). The nodules and abscesses discharge viscid purulent exudate. The horse had poor appetite, had sluggish movement and was dragging its right hind limb with local rise in temperature. Palpation of the skin was extremely resented by the animal. On physical examination the horse had tachycardia (57/min) and tachypnea (32/min.), the rectal temperature was 37.9 °C and the mucous membranes were faint pink in color.

Fecal examination was negative for internal parasites. The complete blood count (CBC) picture was as follows: Haemoglobin Concentration (Hb) of 83 (g/l), Packed Cell Volume (PCV) was 0.30 (l/l), Total Red Blood Cell Count (TRBC) was 6.81 ($\times 10^{12}/\text{l}$), Total White Blood Cell Count (TWBC) was 2 ($\times 10^9/\text{l}$), While the Differential Cell Count was: 1.22 ($\times 10^9/\text{l}$) Neutrophils, 0.62 ($\times 10^9/\text{l}$) lymphocytes, 0.14 ($\times 10^9/\text{l}$) Eosinophils, 0.02 ($\times 10^9/\text{l}$) Monocytes and 0 Basophils. The results obtained showed



Fig. 1: *Corynebacterium pseudotuberculosis* on the pectoral muscles of a horse showing typical "pigeon breast" form of the disease.



Fig. 2: Abscesses draining on the ventrum of a horse-caused by *Corynebacterium pseudotuberculosis*.



Fig. 3: Lesions of ulcerative lymphangitis in a horse, showing typical pattern of invasion of the lymphatic vessels affecting the entire forelimbs from elbows to hooves and head. Note: concentration of lesions round the mouth commissure



Fig. 4: Lesions of ulcerative lymphangitis in a horse, showing typical pattern of invasion of the lymphatic vessels affecting the entire hindlimbs from thighs to hooves. Note: swelling of the right hindlimb.



Fig. 5: Abscesses and ulcers caused by *Corynebacterium pseudotuberculosis* involving the axilla in a horse.

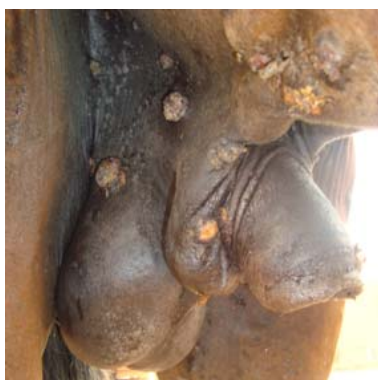


Fig. 6: Abscesses and ulcers caused by *Corynebacterium pseudotuberculosis* involving the prepuce and thigh region of a horse.



Fig. 7: Abscesses and ulcers caused by *Corynebacterium pseudotuberculosis* involving the neck of a horse.



Fig. 8: Encapsulated abscesses developing along an enlarged lymphatic vessel in a horse infected with *Corynebacterium pseudotuberculosis*.

that the horse was anemic and had marked reduction in the total WBC. Impression smears from abscesses revealed Gram positive pleomorphic rods with typical Chinese letters arrangements. *Corynebacterium pseudotuberculosis* was isolated in pure culture from aspirates of intact encapsulated abscesses, and identified in accordance to authentic methods and techniques. Non-motile, Gram positive pleomorphic rods with typical Chinese letters arrangements were seen in smears prepared from pure culture. The organism was also positive for metachromatic (volutin) granules using modified Albert's stain. It was catalase and urease positive, reduced nitrate and produced acid from glucose, maltose and sucrose.

Post-mortem examination revealed no internal lesions other than a small fibrous adhesion between the liver and the diaphragm, and the presence of little pathogen-free yellowish fluid in the abdominal cavity. Histological examination of skin biopsy specimens revealed epidermal degeneration, necrosis and scab formation. The dermis showed severe damage to collagen and adnexal structures and hemorrhage. There was abscess formation involving the subcutaneous tissue and the overlying skin. Central zones of necrosis and pus were surrounded by macrophages, neutrophils and proliferated fibrous tissue.

Based on laboratory findings confirming our tentative diagnosis; two forms of the disease caused by *Corynebacterium pseudotuberculosis* were seen in this horse: The first form was characterized by numerous draining abscesses mainly on the pectoral region "pigeon's breast" and ventral abdomen, and the second form was ulcerative lymphangitis involving the four limbs and was characterized by limb swelling, cellulitis and numerous draining tracts along the lymphatics showing nodular and ulcerative lesions.

DISCUSSION

Our tentative diagnosis based on the clinical picture of the typical lesions was confirmed by the isolation of the causative agent of these lesions in pure culture from pus aspirates of encapsulated abscesses. These findings authenticated the findings of Aleman and Spier (2002), Spier (2006) and Zavoshti *et al.* (2009) who reported that *Corynebacterium pseudotuberculosis* is the classical cause of ulcerative lymphangitis or "pigeon's breast". In a previous investigation in the Sudan only one form of this disease was reported in an 8-year old horse (Abu-Samra *et al.*, 1980). While in the current investigation many external abscesses were seen on the right lateral side of the head extending from mandible curvature to lips, concentrating round the mouth commissure and a few lesions were also seen on the left lateral side of the head. Many external abscesses were noticed on the axilla. Solitary lesions were also encountered on the neck, thorax, flanks and prepuce. These observations supported the findings of Pratt *et al.* (2005) and Spier (2006) who reported that additional sites for abscess formation are the prepuce, mammary gland, axilla, limbs and head, and other less common areas are the thorax, neck, parotid gland, guttural pouches, larynx, flank, umbilicus, tail and rectum. Our observation of enlarged draining lymphatics on the thorax with many nodules developing along them

was in line with what have been stated out by Radostits *et al.* (2007) who reported that lymphatics draining the area become enlarged and hard and secondary ulcers may develop along them.

Infection caused by *Corynebacterium pseudotuberculosis* is characterized by deep intramuscular abscesses in the pectoral and ventral abdomen of horses, infection of limb lymphatics or internal organs, including the liver, kidneys or spleen (Aleman and Spier, 2002; Spier, 2006; Gorman *et al.*, 2010). However, in the current investigation only the first two forms of the disease were observed, and despite the fact that the horse showed tachycardia (57/min) and tachypnea (32/min); no lesions were observed in internal organs except for a small adhesion between the liver and diaphragm. Moreover, culture of the abdominal fluid was negative for *Corynebacterium pseudotuberculosis* or other pathogenic organisms. The deviation of these physical parameters from the normal range was mainly, if not solely attributed to the poor condition of the animal, anaemia and the deleterious effect of the toxins produced by the organism. In support to these findings, Spier (2006) reported that horses with external abscesses do not usually develop signs of systemic illness.

In the current investigation ulcerative lymphagitis lesions were seen involving the four limbs of the horse from elbows and hocks joints to hooves. The horse is a 9-year old draught animal, prone to abrasions and injuries especially on the pastern joints. These abrasions and injuries might have facilitated the entry of the soil borne *Corynebacterium pseudotuberculosis* causing infection of lymphatic vessels and spread on all parts of the body. The virulence of the organism is boosted by the various extracellular exotoxins produced by the organism causing vascular damage and inhibits phagocytes, facilitating spread of infection and exacerbating the disease as was clearly evident from the clinical and CBC picture observed in the current investigation. These findings did confirm the findings of previous workers like Pratt *et al.* (2005), Spier (2006), White (2006) and Radostits *et al.* (2007) who reported that the portal of entry of the soil-borne *Corynebacterium pseudotuberculosis* is thought to be through abrasions or wounds in the skin or mucous membranes, but Valentine and McGavin (2007) reported that the organism can enter the muscle by penetrating open wounds. Our findings also supported the findings of Spier (2006), Radostits *et al.* (2007) and Mamman *et al.* (2011) who reported that the organism possesses a cytotoxic surface lipid coat that appears to facilitate intracellular survival and abscess formation. It also produces a phospholipase exotoxin that increases vascular permeability, has an inhibitory effect on phagocytes and may facilitate spread of infection in the host.

The horse was euthanized after the consent of the owner, for humane reasons because the animal was in poor bodily condition, anemic, with severe lesions involving extensive areas of the body, and manifesting pain and inhibition of phagocytic responses, resulting from the toxins produced by the bacteria. The second reason for sacrificing the horse is for prevention of spread of the disease to other animals in the area by *Musca domestica* and *Stomoxys calcitrans* flies that were seen in large numbers feeding on the lesions. This finding is in

agreement with many other workers Pratt *et al.* (2005), Spier (2006), White (2006) and Radostits *et al.* (2007) who incriminated *Haematobia irritans*, *Musca domestica* and *Stomoxys calcitrans* as vectors for the transmission of the disease to horses. The third reason for sacrificing the animal is our anticipation that treatment might not be successful in producing complete recovery because of the intracellular location of the organism and presence of exudates. In addition to this, the duration of therapy may be long and the cost of the drugs used might be prohibitively expensive for the owner. Moreover, Orsini *et al.* (2010) reported that an antibiotic could be effective *in vitro* that might not be effective for the organism *in vivo*; Spier (2006) reported that several factors should be considered when choosing an antimicrobial: the intracellular location of the organism, the presence of exudates and a thick abscess capsule and the duration of therapy are important as the cost of the drug and the convenience of administration, and Mamman *et al.* (2011) reported that various protocols are being used in the management of ulcerative lymphangitis but the disease seems to always recur in an animal that has once had the clinical disease.

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