

Corynebacterium pseudotuberculosis infection in a 10-year-old male West African Dongola horse in the Zoological Garden, University of Ibadan: a case report

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Abstract

Corynebacterium pseudotuberculosis is a gram-positive bacterium that affects several animals, including horses. The present report presents a case of a 10-year-old male West African Dongola horse that developed an external abscess caused by *C. pseudotuberculosis* in the Zoological Garden, University of Ibadan. The patient was presented to the garden veterinary clinic with a complaint of lameness, pus discharge, and swelling on the right forelimb. The pulse rate and temperature were within normal range. Blood was collected from the jugular vein for a complete blood count (CBC) and serum chemistry. A pus sample was collected and subjected to bacterial culture, gram staining, biochemical tests, and an antimicrobial susceptibility test. *Corynebacterium pseudotuberculosis* was isolated and identified in pure culture from aspirates of the pus discharge based on cultural, gram-staining, and biochemical characteristics. The organism was sensitive to gentamicin, amoxicillin, and ciprofloxacin. A warm compress, wound irrigation, and bandage were applied to the infected area. The antibiotics used were a combination of amoxicillin and gentamycin injections. Other drugs used include tetanus toxoid injection, tetanus antitoxin injection, and diclofenac sodium injection. The therapy was successful, and the patient completely recovered. Good sanitation and prompt wound care are recommended.

Keywords: *Corynebacterium pseudotuberculosis*; Abscess; Bacteria culture; Pus; Biochemical test.

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Introduction

Corynebacterium pseudotuberculosis is a gram-positive, facultative, intracellular, pleomorphic bacterium found across the world (Biberstein and Hirsh, 1999).

Corynebacterium pseudotuberculosis infection has been documented in equids and other animals, including goats, cattle, buffalo, sheep, camelids, and humans (Spier, 2006). There are two genetically separate biovars: *biovar equi*, which affects horses and is nitrate positive, and *biovar ovis*, which affects goats and sheep and is nitrate negative (Spier and Azevedo, 2017).

The route of infection is uncertain; however, it is thought that the bacterium enters the horse through skin abrasions (Spier et al., 2004). Spier et al. (2004) discovered that insects such as the stable fly (*Stomoxys calcitrans*), horn fly (*Haematobia irritans*), and house fly (*Musca domestica*) operate as vectors. There are three clinical manifestations in infected horses: (1) muscular or subcutaneous abscess, primarily in the abdominal and pectoral regions; (2) internal infection resulting in visceral abscess (kidney, liver, spleen, and lungs); and (3) ulcerative lymphangitis, which causes limb inflammation, cellulitis, and severe lameness (Aleman et al., 1996).

Pure cultures can be obtained from the lesions. While abdominocentesis detects abdominal abscesses, normal peritoneal fluid does not rule them out. Ultrasonography can help detect deep abscesses in the inguinal, abdominal, or axillary regions (Jeske et al., 2013).

The treatment is typically curative and comprises antibiotic therapy and abscess drainage. Good hygiene and management, including early wound care and thorough insect control, are the most effective means of prevention (Corbeil et al., 2016).

Here, a case report of a 10-year-old male West African Dongola horse that developed an external abscess in the Zoological Garden, University of Ibadan, caused by *C. pseudotuberculosis* is described.

Case details

History, clinical examination, and sample collection

A 10-year-old male West African Dongola horse, weighing 301kg and stabled at the Zoological Garden, University of Ibadan, was presented to the garden clinic. The main complaints, according to the caregivers, were lameness (Fig. 1), abscess swelling (Fig. 2), and pus discharges (Fig. 3) on the right forelimb. The patient was said to have had a puncture wound on the same right forelimb a few days before presentation. The pulse rate (36 beats/min) and temperature (37.9 °C) were within the normal range. Blood was collected from the jugular vein for complete blood count (CBC) and serum chemistry; the values obtained were compared with the reference laboratory values documented by Radostits et al. (2006).

Aspirates of pus were aseptically collected from the abscess on the right forelimb and subjected to bacterial cultures, gram staining, biochemical tests, and an antimicrobial susceptibility test.



Fig. 1: Horse showing sign of lameness at presentation



Fig. 2: Abscess swelling on the right forelimb (white arrow)



Fig. 3: White arrow showing pus discharge (white arrow)

Laboratory result

The complete blood count showed that the horse had leucocytosis due to neutrophilia (Table 1). The serum

biochemistry results showed that the horse had hyperproteinaemia due to hyperalbuminaemia (Table 2).

Table 1: Haematology result

| Blood parameter | Value | Reference value (source: Radostits et al., 2006) | Inference |
|------------------------------------|---------------------|--|--------------|
| Plasma colour | Clear, straw-yellow | | Normal |
| Fibrinogen (mg/dL) | 300 | 200-400 | Normal |
| Hb (g/dL) | 11.2 | 11-19 | Normal |
| PCV (%) | 35 | 32-53 | Normal |
| RBC ($\times 10^6/\mu\text{L}$) | 7.22 | 6.8-12.9 | Normal |
| MCV (fl) | 57 | 37-59 | Normal |
| MCHC (g/dL) | 32 | 31-38.6 | Normal |
| Platelets ($/\mu\text{L}$) | 118000 | 100000-600000 | Normal |
| Total WBC (μL) | 16700 | 5400-14300 | Leucocytosis |
| Seg. Neutrophils (μL) | 13100 | 2300-8500 | Neutrophilia |
| Band Neutrophils (μL) | 100 | 0-100 | Normal |

| | | | |
|-------------------|------|-----------|--------|
| Lymphocytes (/μL) | 2200 | 1500-7700 | Normal |
| Monocytes (/μL) | 700 | 0-1000 | Normal |
| Eosinophils (/μL) | 600 | 0-1000 | Normal |
| Basophils (/μL) | 0 | 0 | Normal |

Table 2: Serum chemistry result

| Serum parameter | Value | Reference value (source: Radostits et al., 2006) | Inference |
|----------------------|-------|--|-------------------|
| Total protein (g/dL) | 8.1 | 6-7.7 | Hyperproteinaemia |
| Albumin (g/dL) | 4.2 | 2.9-3.8 | Hyperalbuminaemia |
| Globulin (g/dL) | 3.9 | 3.1-3.9 | Normal |
| AST (μL) | 260 | 220-600 | Normal |
| SDH (μL) | 1.9 | 1.9-5.8 | Normal |
| BUN (mg/dL) | 16 | 10-24 | Normal |
| Creatinine (mg/dL) | 1.5 | 0.0-1.9 | Normal |

Nonmotile, gram-positive pleomorphic rods were seen in smears prepared from pure culture. Also, some of the biochemical tests on the organism showed catalase positive, oxidase negative, nitrate reduction positive, urease negative, glucose positive, maltose positive, lactose negative, mannitol negative, and xylose negative (Table 3). Bacterial cultures

followed by gram staining and biochemical tests confirmed *Corynebacterium pseudotuberculosis*.

Based on the antimicrobial susceptibility test, *Corynebacterium pseudotuberculosis* was sensitive to gentamicin, amoxicillin, and ciprofloxacin and insensitive to metronidazole and streptomycin (Table 4; Fig. 4).

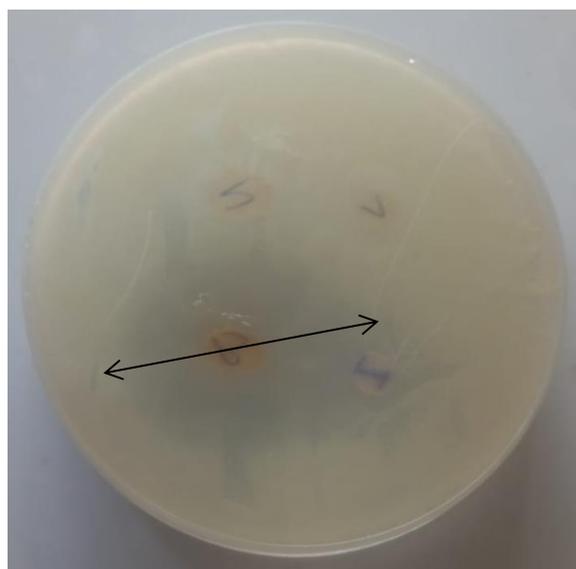
Table 3: Biochemical tests

| Biochemical Characteristics | <i>Corynebacterium spp. Isolate</i> |
|-----------------------------|-------------------------------------|
| Gram Staining | Gram-positive short rods |
| Catalase | Positive |
| Oxidase | Negative |
| Nitrate reduction | Positive |
| Motility | Non-motile |
| Methyl-red | Positive |

| | |
|------------------------------|--|
| Hydrogen sulphide production | Positive |
| Indole production | Negative |
| Citrate | Negative |
| Voges-proskauer | Negative |
| Urease Test | Negative |
| OF (Oxidative-Fermentative) | Fermentative |
| TSI Agar | Alkaline slant, acidic butt, and presence of gas |
| Glucose | Positive |
| Maltose | Positive |
| Lactose | Negative |
| Mannitol | Negative |
| Xylose | Negative |
| Sucrose | Negative |
| Arabinose | Positive |

Table 4: Antimicrobial susceptibility test

| Animal Type | Sensitive | Intermediate | Resistant |
|----------------|--|---|-------------------------------|
| Equine (Horse) | Gentamicin Amoxicillin Ciprofloxacin | Tetracycline, Doxycycline, Chloramphenicol Enrofloxacin Ceftriaxone | Metronidazole Streptomycin |

**Fig. 4: Antimicrobial susceptibility test showing a wide zone of inhibition with gentamycin (black arrow)**

Treatment and outcome

A warm compress was applied to the abscess using a clean washcloth and warm water. It was placed over the abscess for two days to expel the pus, thereby reducing the abscess swelling. The wound was copiously irrigated with normal saline, dried, and covered with a bandage, which was changed every other day.

Tetanus toxoid injection (Tetanus toxoid® 1 ml/horse, Zoetis Inc., Kalamazoo, USA) was administered intramuscularly; tetanus antitoxin injection (Tetanus antitoxin® 1500 IU/horse, Jiangxi Institute of Biological Products

Inc., China) was administered intramuscularly; diclofenac sodium injection (Jawa diclofenac® 1.1 mg/kg, Jawa Group, Lagos, Nigeria) was administered intramuscularly for 3 days; and Amoxicillin and Gentamycin injection (Biogenta® 1 ml/10 kg, Interchemie werken "De Adelaar" BV, The Netherlands) was administered intramuscularly for 7 days.

The patient recovered; the abscess swelling subsided with complete healing (Fig. 5).



Fig. 5: Out come after treatment showing resided abscess swelling (red arrow) and complete healing (white arrow).

Discussion

The present case reveals an external abscess on the horse's right forelimb. External abscesses are most commonly found in the pectoral region and the ventral abdomen (Corbeil et al., 2016). However, Nogradi et al. (2012) established that the

limbs and other regions, such as the prepuce, mammary gland, triceps, and head, can also be affected. It is believed that the puncture wound, which occurred a few days before presentation, predisposed the horse to the infection.

The infected horse had various clinical signs of the disease seen in horses with *Corynebacterium pseudotuberculosis*, which agreed with the findings of Pratt et al. (2005), Zavoshti et al. (2009), and Muñoz-Bucio et al. (2017).

The haematological examination revealed neutrophilia, which was also reported by Gonzalez et al. (2008). The hyperproteinaemia is indicative of dehydration, as established by Boyd (1981). Based on the bacteria culture, gram staining, and biochemical test, *Corynebacterium pseudotuberculosis* was confirmed. The nonmotile, gram-positive pleomorphic rods seen in smears prepared from the pure culture are consistent with the reports of Coyle and Lipsky (1990) and Dorella et al. (2006).

Biochemical tests carried out in this case confirmed the presence of *C. pseudotuberculosis* and are in agreement with studies on the speciation of *C. pseudotuberculosis* (Funke et al., 1997; Dorella et al., 2006; Chahota et al., 2017).

The present case shows that the *Corynebacterium pseudotuberculosis* isolate was nitrate reduction positive, proving the organism to be an *equi-biovar*. This is expected because the organism was isolated from a horse. The nitrate test results for *C. pseudotuberculosis* might vary. This is due to the presence of two biovars: *biovar equi* (from cattle or horses), which reduces nitrate, and *biovar ovis* (from goats or sheep), which does not (Coyle and Lipsky, 1990).

Corynebacterium pseudotuberculosis was only sensitive to gentamicin, amoxicillin, and ciprofloxacin in the present case. Studies have found the bacteria to be resistant to a wide variety of antibiotics (Muckle and Gyles, 1982). This suggests that the susceptibility of *C.*

pseudotuberculosis to antimicrobial drugs differs among isolates derived from different sources.

Warm compress used in the present case was intended to reduce swelling, encourage evacuation of all purulent matter, and speed up healing. Normal saline, a nontoxic, isotonic solution that does not harm healing tissues, was used to irrigate the infected wound to physically remove cellular debris and trapped fluids and lower the bacterial load (Ambe et al., 2020). The bandage was used to collect pus from the wound, provide proper healing conditions, and protect the area until the wound healed. Tetanus antitoxin and tetanus toxoid were both administered to the patient since tetanus antitoxin only protects for 1–3 weeks, whereas tetanus toxoid provides long-term immunity.

The antibiotics employed, combination of amoxycillin and gentamycin injection, were chosen based on antimicrobial susceptibility testing, and they performed admirably. In conclusion, this case reports the diagnosis and successful treatment of a *C. pseudotuberculosis* infection in a West African Dongola horse stabled at the Zoological Garden, University of Ibadan. Hence, good sanitation and prompt wound care are recommended.

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