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Anaplasmosis in a buffalo: A case report

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Abstract

Anaplasmosis is one of the most common infectious diseases of bovines causing economic losses to farmers due to decreased milk yield, weight loss, abortions and eventually death of the animal. A three year old female buffalo from a farm at Hyderabad was presented with a history of high fever, weakness, decreased milk yield, anorexia, apathy, weight loss and tick infestation. Clinically the animal showed pale ocular, buccal mucus membranes, rapid bounding pulse and lack of rumination. Complete blood picture (CBP) revealed decreased Total Erythrocyte Count (TEC), Hemoglobin concentration (Hb) and Packed Cell Volume (PCV). Blood smear examination revealed the presence of intraerythrocytic *Anaplasma* inclusions at the margins of the corpuscles concluding that the buffalo was suffering from Anaplasmosis. The faecal examination was negative for parasitic ova/oocysts. The animal was treated with oxytetracycline, antipyretics and hematinics for three consecutive days. Prompt improvement in health condition of the animal was noticed post treatment.

Keywords: Anaplasmosis, bovines, complete blood picture

Introduction

Anaplasma marginale is an obligate intraerythrocytic rickettsial organism belonging to the family Anaplasmataceae of the order Rickettsiales [1]. *Anaplasma marginale* causes bovine Anaplasmosis, a hemolytic disease causing severe economic loss to both the dairy and beef industries worldwide. Clinical disease is most notable in cattle, but other ruminants including water buffalo, bison, African antelopes and mule deer can become persistently infected with *A. marginale* [2]. The intracellular pathogen was first described by Sir Arnold Theiler in 1910 in erythrocytes of South African cattle as “marginal points” [3]. *Anaplasma* is one of the most important parasites transmitted by at least 20 ticks species, including *Argas persicus*, *Ornithodoros lahorensis*, *Boophilus annulatus*, *B. decoloratus*, *B. microplus*, *Dermacentor albipictus*, *D. andersoni*, *D. occidentalis*, *D. variabilis*, *Hyalomma excavatum*, *Ixodes ricinus*, *Rhipicephalus bursa*, *R. sanguineus* and *R. simus* [4] but mostly *Boophilus microplus* causes Anaplasmosis [5]. It is transmitted mainly through the bite of ticks or by the mechanical transfer of blood from infected to susceptible cattle from biting flies or by blood-contaminated fomites including needles, ear tagging, dehorning and castration equipment [6]. Recent research documented that biological transmission by ticks was more efficient than mechanical transmission by *Stomoxys calcitrans*, the stable fly [7]. Anaplasmosis is found on six continents and is responsible for high morbidity and mortality in cattle in temperate, subtropical, and tropical regions [8].

After invasion into red blood cells (RBC), they divide into up to eight initial bodies and enlarge within its thin outer membrane forming a large dot. When infected red blood cells rupture, the parasite’s membrane also ruptures, releasing the initial bodies into the blood stream to invade other RBCs. As the infection progresses, more and more RBCs contain parasites and are destroyed [9]. The disease is characterized by fever, severe anemia, jaundice, brownish urine, loss of appetite, dullness or depression, rapid deterioration of physical condition, muscular tremors, constipation, yellowing of mucous membrane and labored breathing [10]. Theiler described the use of *A. centrale*, as a live vaccine to minimize the severity of *A. marginale* infection. However, he concluded that *A. centrale* vaccination did not prevent *A. marginale* infection upon challenge but rather resulted in lower levels of *A. marginale* rickettsemia and a reduction in severity of clinical signs compared to unvaccinated controls [11]. The developmental cycle of *A. marginale* in ticks is complex and coordinated with the tick feeding cycle [12, 13, 14]. Infected erythrocytes ingested by ticks with the blood meal provide the source of *A. marginale* infection for tick gut cells. After development of the pathogen in tick gut cells, many other tick tissues become infected,

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including the salivary glands, the site from which *A. marginale* is transmitted to cattle during feeding [15, 16]. Imidocarb dipropionate has been successfully used for more than 30 years to treat Anaplasmosis in certain territories [17]. Chlortetracycline and oxytetracycline are the only compounds approved for use against acute anaplasmosis in the United States [18]. Previous studies which report successful clearance of persistent *A. marginale* infections in cattle used oxytetracycline, which was administered intravenously at 11-22 mg/kg for 5-12 days [19, 20].

The present case report deals with a three year old female buffalo suffering from Anaplasmosis and its successful recovery post treatment.

Materials and methods

A three year old female buffalo from a farm at Hyderabad was presented.

1. History and Clinical findings: History of the animal was noted. The ocular, buccal mucosa were examined. Temperature, heartrate, respiratory rates and ruminal movements were noted.

2. Complete blood picture: Based on clinical signs, blood sample was collected about 1.5 mL of blood was collected from jugular vein in an anticoagulant coated vacutainers {K3-EDTA tube, 13mm x 75 mm, 4 mL (Rapid Diagnostics Pvt, Ltd., Delhi)} to carry out all hematological parameters. All the blood samples were used for the estimation of Total Erythrocyte Count (TEC), Hemoglobin concentration (Hb), Packed Cell Volume (PCV), Mean Corpuscular Hemoglobin (MCH), Mean Corpuscular Volume (MCV) and Mean Corpuscular Hemoglobin Concentration (MCHC) by using automated whole blood analyzer.

3. Blood smear examination: Blood films were made by placing a drop of blood on one end of a slide, and using a spreader slide to disperse the blood over the slide's length. The aim is to get a region, called a monolayer, where the cells are spaced far enough apart to be differentiated. The slide is left to air dry, after which the blood is fixed to the slide by immersing it briefly in methanol. The fixative is essential for good staining and presentation of cellular detail. After fixation, the slide was stained with Leishmans stain following standard protocols and slides were observed under oil immersion (100X).

4. Faecal examination: Faecal samples were collected and smears were prepared by adding a small quantity of faeces on a clean microscope slide, mixed with a few drops of water thoroughly and covered with coverslip and examined under microscope.

Results and discussion

1. History: A three year old female buffalo from a farm at Hyderabad was presented with a history of high fever, decreased milk yield, weakness, fatigue, anorexia, apathy, labored breathing, frequent urination, weight loss and moderate tick infestation.

2. Clinical examination: The ocular and buccal mucus membranes appeared pale and partially cyanotic (Fig. 1 and 2), increased pulse rate, high temperature about 106 °F, lack of ruminal movements, salivation and rough hair coat. The

disease begins its course by invading and multiplying within red blood cells of the host. The rickettsial bacteria produce endotoxins through its lipopolysaccharide outer membrane. As the disease progresses, infected and even uninfected red blood cells are destroyed predominantly in the liver and spleen, resulting in increasing anemia without hemoglobinemia and hemoglobinuria which is responsible for the pallor mucus membranes and rapid bounding pulse. Similar clinical signs were also reported previously [21]. After erythrocytic infection is detected, the number of parasitized erythrocytes increases geometrically.



Fig 1: Pale buccal mucosa



Fig 2: Pale ocular mucosa

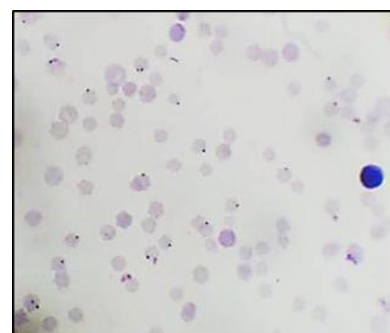


Fig 3: Blood smear showing erythrocytes with Anaplasma inclusions at the margin (100X)

3. Complete blood picture: Blood picture revealed reduction in the levels of TEC, Hb and PCV levels (Table 1). As the rickettsial bacterium mainly affects the red blood cells, the parasitized erythrocytes are removed resulting in decreased TEC, Hb and PCV levels leading to anemia and pale mucus membranes. Due to hemolytic anemia, the animal cannot tolerate stress leading to fatigue and weakness. Similar trends of haematological values in *Anaplasma* infected animals were also observed by previous authors [21, 22].

Table 1: Complete blood picture

| Parameter | Result | Post treatment |
|--|------------------------|-----------------------|
| Total Erythrocytic Count (TEC) | 3 *10 ⁶ /μL | 8*10 ⁶ /μL |
| Packed cell volume (PCV) | 12% | 35% |
| Hemoglobin concentration (Hb) | 6 g/dL | 12 g/dL |
| Mean Corpuscular Volume (MCV) | 50 fL | 45 fL |
| Mean Corpuscular Hemoglobin (MCH) | 17 pg | 16 pg |
| Mean Corpuscular Hemoglobin Concentration (MCHC) | 30 g/dL | 36 g/dL |

4. Blood smear examination: Smears stained with Leishmans stain and examined under oil immersion revealed the presence of *Anaplasma* spp appeared as dense, homogeneously staining blue-purple inclusions 0.3-1 μm in diameter located towards the margin of the infected corpuscle (Fig. 3).

5. Faecal examination: The faecal samples were negative for parasitic ova/oocysts.

6. Treatment: Treatment is most efficient during the bacteremic phase of the infection and is directed at reducing the rate of erythrocyte infection. Oxytetracycline (20 mg/kg BW, once daily for up to 5 days, IV) has been used successfully used to treat the affected animal along with antipyretics (Melonex- 0.5 mg/kg BW, once daily for up to 3 days, IM) and Hematinic preparation (Ferritas- 10mL for 3 consecutive days, IM) to cope up with anemia. Stress should be avoided during handling and treatment.

7. Recovery: After treatment for three consecutive days the animal recovered thoroughly, with normal body temperature (102 °F) and healthy appetite.

Conclusion

Based on the history, clinical signs and clinical pathology it can be concluded that the buffalo was suffering with Anaplasmosis which was successfully recovered after treatment with oxytetracycline. Although clinical signs are rare in buffaloes, their treatment is essential as they may act as carriers for cattle.

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