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Concurrent infection of theileriosis and anaplasmosis in crossbreed cattle

Manu Jaiswal, Vivek Kumar Singh and Kuldeep Saini

Abstract
A 5-year-old crossbred cattle with history of anoraxia, pyrexia, salaivation was presented to Veterinary Clinical Complex, PGIVER, Jaipur, India. On clinical examination, it was revealed that the animal was dull and depressed with rectal temperature of 105.2°F. The cow also exhibited nasal discharge, coughing, moist rales on auscultation of lungs, ruminal atony, dehydration, rough body coat, paler mucous membrane of conjunctiva and vulva and enlargement of prescapular lymph nodes. Heavy tick infestation was also observed. History and clinical signs suggested haemoprotozoan infection. Laboratory examination revealed mixed infection of *Theileria* spp. and *Anaplasma* spp. with low haemoglobin level (4.2g/dl). The cattle was treated with buparvaquinon, fluid therapy, liver extracts, antipyretic and antihistaminics for three days, but eventually succumbed at 8th day. Post-mortem examination revealed severe anaemic carcass with congestion and petechiae in the visceral organs. Lungs showed severe congestion suggesting that the animal might have died due to secondary bacterial pneumonia together with toxaemia.

Keywords: Cattle, moist rales, post-mortem examination, toxemia

Introduction
The arthropod borne hemoparasitic diseases are of great economic impact on livestock affecting 80% of the world cattle population and causes economic loss due to morbidity and mortality (Kasozi *et al*., 2014) [8] Theleriosis is one such commonest haemoprotozoan diseases of Indian subcontinent causing severe economic losses. Theileriosis is caused by Theileria annulata and transmitted through the bites of tick *Hyalomma anatolicum.* It has been considered as the most important blood protozoan parasite with higher incidence in exotic breeds and the crossbred cattle of all age groups. *Theileria annulata*, an intracellular obligate hemoprotozoan, causes tropical theileriosis in both wild and domestic animals. The most frequently observable clinical symptom of theileriosis are high fever, enlargement of regional superficial lymph nodes, inappetence, cessation of rumination, cachexia, anemia, leucopenia, laboured breathing, lacrimation and conjunctivitis. After clinical recovery from acute infections, animal may become carriers with long-term persistent infections (Brown, 1990) [2].
Bovine anaplasmosis caused by the obligate intra-erythrocytic rickettsia, Anaplasma marginale (Ristic, 1980) [15] is responsible for great economic losses in developing countries. Anaplasmosis is endemic in tropical and subtropical areas but is frequently reported in temperate regions of the world (Kuttler, 1984) [10]. Anaplasmosis is currently classified in list B of the Office International des Epizooties (OIE) Terrestrial Animal Health Code due to its socioeconomic importance and significance in terms of restriction and animal products (OIE, 2003) [14]. The number of infected erythrocytes increases logarithmically and removal of these infected cells by phagocytosis results in development of haemolytic anaemia and icterus. Acute anaplasmosis causes anaemia, abortion, decreased milk production and death (Ajayi *et al*., 1978) [1]. Cattle recovered from acute infection remain persistently infected and protected from clinical disease, serving as reservoirs for A. marginale, which are not detectable by microscopic methods in the carrier stage of infection (Ewing, 1981; Kieser *et al*., 1990) [7, 9].

Case history, clinical observation and diagnostic work -up
A 5-year-old crossbred pregnant cattle with history of anoraxia, pyrexia, salaivation was presented to Veterinary Clinical Complex, PGIVER, Jaipur, India. On clinical examination, it was revealed that the animal was dull and depressed with rectal temperature of 105.2°F. The cow also exhibited nasal discharge, coughing, moist rales on auscultation of lungs, ruminal atony, dehydration, rough body coat,

paler mucous membrane of conjuctival and vulvar and enlargement of prescapular lymph nodes. Closed physical examianation of medial aspect of pinna and udder were completely yellowish which indicate towards jaundice. Heavy tick infestation was also observed. On the basis of clinical examination, blood sample was collected aseptically from juglar vein in a sterilized EDTA vial for haematological examination. The Haemoglobin (Hb) was 4.2 g/dl indicative of marked anaemia. A peripheral blood smear was made from ear tip and stained with Giemsa and microscopic examination done under oil immersion for the presence of any haemoprotozoan. Peripheral blood smear examination revealed mixed infection of *Theileria* spp. And *Anaplasma* spp. (Fig 1)

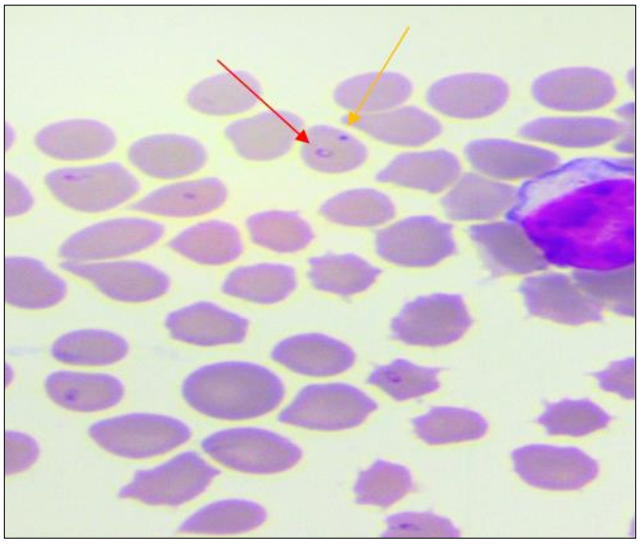

Fig 1: Signet ring piroplasms of Theileria spp. in RBCs (red arrow) and Anaplasma spp. (Yellow arrow)

Treatment and post-treatment follow up

The cow was treated with the single dose of injection Zubion 20 ml deep intramuscular (Buparvaquone, Intas Animal Health) @ 2.5 mg/kg b.wt single dose, injection Melonex Plus, 10 ml intamuscular (Meloxicam and Paracetamol, Intas Animal Health) @ 0.2 mg/kg b.wt. and injection Avilin Vet 07 ml (Pheniramine Maleate, MSD, Intervet India Pvt. Ltd) @ 0.5 mg/kg body weight, along with three doses of oxytetracycline long acting (Oxytetracycline-LA, Zydus AH) deep I/M @ 20 mg/kg BW at every 48 hours interval. A pour on solution of Flumethrin 40ml (Bayer Animal Health) was applied topically for control of ticks population and supplemented with Liquid Vitakind Liv @50ml orally in a day. A total 5 ml of a commercial formulation containing vitamin A 2.5 lac IU, vitamin D_3 25,000 IU, vitamin E 100 IU, and biotin 12.4 mcg/ml (Intavita-H) was also given intramuscularly. After 4-5 days of treatment cattle show drooping of left ear pinna and continuing salivation from mouth, these signs indicate partial paralysis of half side of face. The rectal temperature was gone lower side (98.6 ºF) and cattle and eventually succumbed after 8th day of treatment.

Post-mortem examination

Upon post mortem examination, the gross lesions encountered included pale mucous membrane with emaciated carcass; serosal surfaces of the abdominal organs had yellowish discoloration indicating icterus. Splenomegaly with congestion and hepatomegaly were also seen. The lungs were congested. (Fig 02, 03, 04)

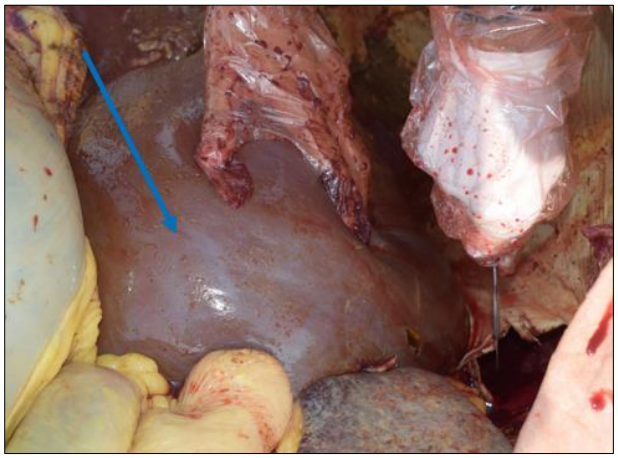

Fig 2: Gross lesion of the liver showing marked enlargement (hepatomegaly) blue arrow

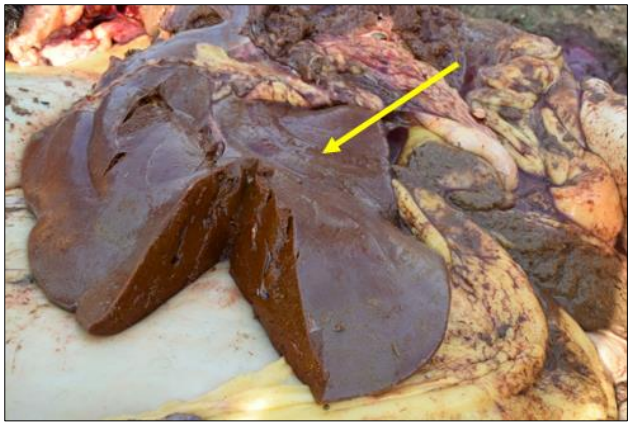

Fig 3: Necrosed liver

Fig 4: Gross lesion of the spleen showing enlargement (splenomegaly) red arrow, with yellowish discoloration of the omentum (Yellow arrow).

Discussion

A significant feature of the disease is anemia owing to overproduction of cytokines and reactive oxygen species (Nazifi *et al*., 2009; Saleh *et al*., 2011) [12, 16]. Presence of oxidative stress for development of anaemia in animals affected with Theileriosis has been reported by the previous scientific workers (El-Deeb and Younis, 2009; Nazifi *et al*., 2011) [6, 13]. Previously antioxidant potential of vitamins A, D_3, E and H has been demonstrated (Singh *et al*., 2012) [18]. Buparvaquone is a promising compound for the therapy and prophylaxis of all forms of theileriosis. The adjunction of

antioxidants and anti-theilerial agents can pervent the animals from fatal theileriosis. Moreover, large scale clinical studies are on adjunctive antioxidant is required to provide a novel therapeutic regimen of theileriosis. However, to prevent the infection of *Theileria* infection in animals *in vitro* attenuated schizontal cell culture vaccine is available with trade name of 'Rakshavac T' manufactured by Indian immunologicals limited, Hyderabad (Singh *et al*., 2014) [17]. It is further recommended that the high valued animals or high yielding animals may be vaccinated with this vaccine to prevent them with *Theileria* infection. Tetracyclines are effective against the schizonts of *T. annulata* (Mallick *et al*., 1987) [11] and of *T. parva* (Dolan, 1981) [5], but only when used in large doses during the prepatent/incubation period of infection.

Oxytetracycline and Chlortetracycline are effective drugs against bovine anaplasmosis, provided they are given early in the course of the infection, before the onset of severe anemia (Campbell and Rew, 2013) [3]. The other treatment options such as diminazene aceturate (dosed at 3 to 5 mg/kg, IM), amicarbalide (dosed at 5 to 10 mg/kg, IM), and imidocarb (dosed at 1 to 3 mg/kg, IM) are most often used (Campbell and Rew, 2013) [3].

The most useful postmortem lesions are marked splenic enlargement and congestion (Kieser *et al*., 1990) [9] and this is consistent with the lesions we observed in this report. The jaundice and anaemia reported in this case is due to the destruction of red blood cells and their contents being released into the blood stream. Similarly, splenomegaly with congestion and discoloration of the spleen reported in our case is in agreement with the results of (Devos *et al*., 2006) [4], who reported that the spleen of infected cows was enlarged following anaplasmosis. This is associated with excessive destruction of defective erythrocytes by macrophages within the spleen (Devos *et al*., 2006) [4]. Anaplasmosis is an infectious disease of cattle that causes anaemia, abortions, and death. Adult cattle have the most severe symptoms of the disease.

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