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Autoimmune haemolytic anaemia in a dog-A case report

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ABSTRACT: A case of autoimmune haemolytic anaemia was observed in 7 year old male Spitz dog brought to TVCC Pantnagar with the history of anorexia, lethargy and weakness. There was no history of previous infection and tick infestation. Vaccination and deworming were proper in schedule. On clinical examination the pale mucus membranes were seen with an increased pulse and respiration rates and normal rectal temperature. There was no hemoprotozoan parasite on examination of blood smear and no parasitic eggs, cyst or oocyst found in faecal samples. Collected blood was slightly agglutinated. Blood picture revealed spherocytosis, agglutination of RBCs and hyper segmented neutrophils. It was diagnosed to be a case of idiopathic autoimmune haemolytic anaemia. Animal was treated with infusion DNS 5% slow intravenously on day first then repeated after 4th day, injection Prednisolone @ 0.5 mg/kg body weight intramuscularly repeated on alternate days for 2 weeks, injection Imferon @ 0.5 ml intramuscularly on alternate days for 2 weeks, tablet Doxycycline @ 100 mg twice a day orally for 2 weeks. Other supportive therapy given were Iron supplement (aRBCe pet) @ 3 ml orally once daily for 2 weeks, multivitamin syrup (Zipvit) @ 3ml orally once daily for 2 weeks and liver tonic (Livotas pet) @ 3ml twice a day orally for 2 weeks was given. The animal died after 2 weeks of treatment.

Key words: Autoimmune haemolytic anaemia, spitz dog

Autoimmune haemolytic anaemia (AIHA) is an immune disorder in which immune system is abnormally over sensitized and auto-antibodies directly or indirectly targets and destroys its own red blood cells (RBC) which leads to haemolytic anaemia (Chauhan, and Tripathi, 2002). AIHA is considered to be the most common autoimmune disease in dogs and cats that results from a type II hypersensitivity reaction against normal glycoprotein molecules present on the surface membrane of the RBCs (Balch and Mackin, 2007). Destruction of RBC occurs either by complement-mediated lysis (intravascular haemolysis) or due to phagocytosis by cells of the mononuclear phagocyte system (MPS) in the spleen and liver (so-called extravascular hemolysis). This destruction of erythrocytes leads to anaemia and in some cases, haemolytic or pre-hepatic icterus due to accumulation of unconjugated bilirubin in tissue (Swann and Skelly, 2016). AIHA may occur as primary (idiopathic) or secondary to a variety of infectious, toxic conditions or neoplastic disorders (Klag *et. al*., 1993). In primary or idiopathic immune mediated haemolytic anaemia (IMHA), immune system mistakenly produces antibodies that attack its own erythrocytes and it is one of the most common immune-mediated diseases of dogs (Giger, 2005). Primary AIHA is a classic example of an autoimmune disorder with no identifiable underlying cause and is the predominant form of IMHA. Secondary IMHA can be caused by bacterial, viral, rickettsial, parasitic, protozoan, toxins, drugs and neo-plastic disorders (Barker and Elson, 1995). Affected RBCs may become infected by pathogens or coated with foreign antigens. Protozoans like *Babesia canis* and *Trypanosoma canis* are the most common cause of secondary IMHA in dogs. Secondary AIHA due to recent vaccination also has been reported (Duval and Giger, 1996). IMHA is more common in dogs than in cats. Primary IMHA can occur in any dog breed, but English Springer Spaniels, Cocker Spaniels, Poodles, Old English Sheep Dogs, Irish Setters and Collies are more suffered breeds (Carr *et al*., 2002). The classical findings of AIHA include weakness, exercise intolerance, lethargy, anorexia, tachypnea, dyspnea, vomiting, diarrhoea and animal not taking any interest. Sometimes polyuria and polydipsia may also be observed. Clinical examination typically reveals pale mucous membranes, tachypnoea, steep pulse and systolic murmur (Mellett *et al*., 2011). Although AIHA can occur at any age (Burgess *et al*., 2000) but the mean age of onset of AIHA is more than 6 years or adult age (McAlees, 2010). The diagnosis is done by examination of blood smear stained by Giemsa stain. The blood picture shows the presence of spherocytes (RBCs without central pallor), nucleated RBCs, RBC agglutination, positive direct Coomb's test and the absence of a detectable underlying cause of haemolytic anaemia (Carr *et al*., 2002). A positive saline agglutination test has been reported in approximately 40% to 89% of dogs with AIHA and it is common outcomes in animals with AIHA (Scott-Moncrieff

et al., 2001). The overall death rate of canine AIHA may be high with a rage of 50-80 % (Reimer *et al*., 1999), and most deaths occur in the first 2 weeks after diagnosis (Piek *et al*., 2008).

Case history

A 7 year old male Spitz dog weighing about 6.2 kg was brought to the Teaching Veterinary Clinical Complex, College of Veterinary and Animal Sciences, Pantnagar from Haldwani city of Uttarakhand with the history of off fed, lethargy and weakness (Fig. 1) for the past 7-8 days. There was no history of previous infection, drug therapy, no sudden diet change and no tick infestation. Vaccination and deworming were proper in schedule. Clinical examination of dog revealed pale mucus membranes (Fig.2), dullness, slightly dark coloured urine with increase pulse and respiration rates. Physiological parameters like rectal temperature, heart rate and respiratory rate were 101.8°F, 180 beats per minute and 42 per minute, respectively. The animal was dehydrated and weak.

Fig. 1: Animal showing weakness

Fig. 2: Animal showing pale anaemic conjunctiva

Clinical examination

A volume of 4 ml of blood was collected from the cephalic vein of the animal in an EDTA coated vial. Faecal sample was also collected. Blood and faecal samples were sent to lab for haematological and parasitological examination. Faecal sample was examined by direct smear method (Soulsby, 1982). Blood sample was examined by the thin blood smear method. Thin blood smear was prepared from fresh blood without anticoagulant on a clean glass slide immediately after the blood being drawn from the vein. Smear was then air dried and fixed with absolute methanol for 1-2 minute. It was then stained with 20% diluted Giemsa stain for 40 minutes. Slide was then washed in running tap water and the smear was air dried and observed under oil emersion. Slide was examined covering about 50 microscopic fields.

Diagnosis

Faecal sample was found negative for parasitic eggs/cysts/ oocysts. Blood smear revealed no haemoprotozoan infection and presence of spherocytes (RBCs without central pallor) (Fig 3), hyper segmented neutrophils (Fig 4) and agglutination of RBCs (Fig 5) were observed in the blood smear. Blood with EDTA started showing slight agglutination in minutes. Haemogram presented in Table 1 indicating a low values than normal such as haemoglobin concentration, packed cell volume (PCV %), total erythrocytes count (TEC). The differential leukocytes count (DLC) indicated 6% lymphocytes, 84% segmented neutrophils, 4% stab neutrophils, 1% eosinophils, 5%

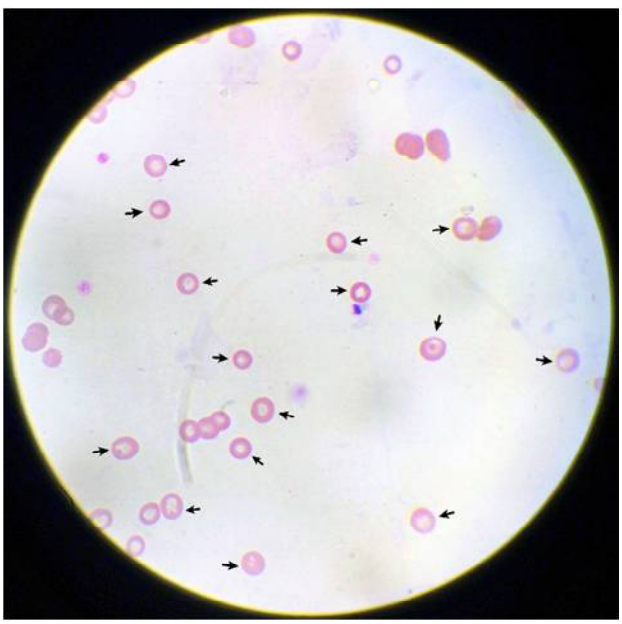

Fig 3: Blood smear showing spherocytes (100x)

monocytes and 0% basophils. Based on the above said facts and laboratory investigations, the case was diagnosed as Idiopathic AIHA.

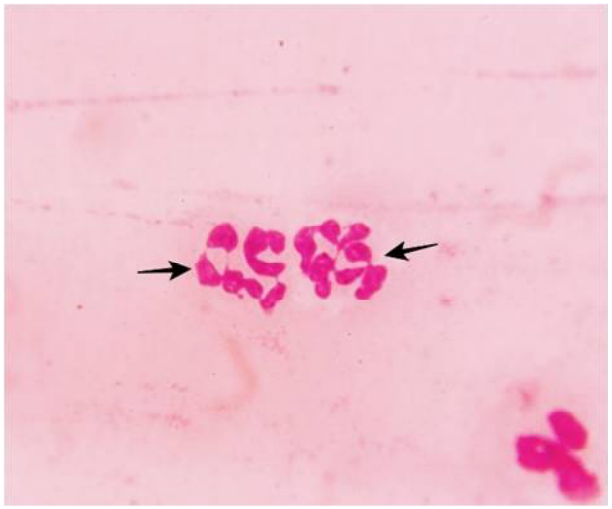

Fig4: Blood smear showing hypersegmented neutrophils (100x)

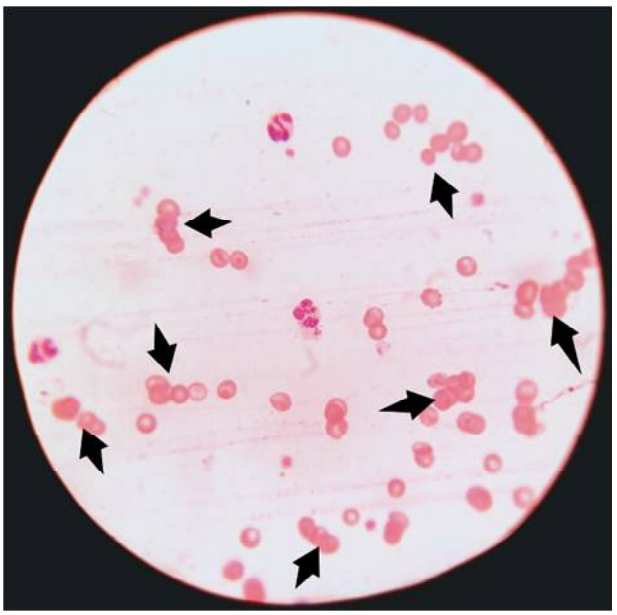

Fig 5: Blood smear showing agglutination of red blood cells (100x)

Treatment

The animal was treated with infusion DNS 5% slow intravenously on day first then repeated after 4th day, injection Prednisolone @ 0.5 mg/kg body weight intramuscularly repeated on alternate days for 2 weeks, injection Imferon @ 0.5 ml intramuscularly on alternate days for 2 weeks, tablet Doxycycline @ 100 mg twice a day orally for 2 weeks. Other supportive therapy given were Iron supplement (aRBCe pet) @ 3 ml orally once daily for 2 weeks, multivitamin syrup (Zipvit) @ 3ml orally once daily for 2 weeks and liver tonic (Livotas pet) @ 3ml twice a day orally for 2 weeks was given. The animal died after 2 weeks of treatment.

Table 1: Various blood parameters of affected dog examined at 0 days post treatment.

Parameters	Values
Hemoglobin (g/dl)	5.8
PCV (%)	17
TEC (Million/Cu mm)	2.9
TLC (cells/Cu mm)	47300
MCV (fl)	58.62
MCH (g/dl)	20.0
MCHC (pg/dl)	34.11

CONCLUSION

Autoimmune haemolytic anaemia (AIHA) is an autoimmune disorder. Animal could not survive even after treatment. Mortality can be delayed and reduced by awareness of disease, speed of diagnosis, new drug treatment therapy approaches and availability of supportive care and haematinics.

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