Ehrlichiosis in leopards (*Leopardus tigrinus*)

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Abstract

Two cases of ehrlichiosis in adult leopards are described and discussed in this report. Rescued leopards from zoological park, Surat were referred at the hospital with fifteen days history of anorexia, prostration and lethargy. Detailed examination revealed pyrexia (103.6 - 105 °F) , anorexia, breathing difficulty, tachycardia, ocular discharge, lethargy, dullness, diarrhoea, stiffness, petechae in buccal cavity and tongue erosions; low haemoglobin, leukocytosis, thrombocytopenia, neutrophilia with shift to left, and intracytoplasmic inclusions in monocytes resembling with those of *Ehrlichia canis* in dogs; and increased levels of alanine aminotransferase (ALT), aspartate aminotransferase (AST) and serum creatinine. Treatment with imidocarb, fluid therapy, steroid, vitamin B-complex and oxytetracycline was promising.

Key words: Ehrlichia, Imidocarb, Leopard, leukocytosis

Ehrlichiosisis caused by *Ehrlichia spp*. (*E. canis* or granulocytic *Ehrlichia spp*.) is transmitted by brown tick *Rhipicephalus sanguineus*. As compared to canines, the disease is not common in domestic and wild felines. Granulocytic ehrlichiosis has earlier been reported in a domestic cat from this hospital (Varshney *et al*., 2009). *E. canis* has been reported responsible for thrombocytopenia and anaemia in felines. Reports on clinical ehrlichiosis in wild cats particularly leopards are scanty in India. Therefore, the present report put on record two cases of ehrlichiosis, caused by *E. canis*, in rescued leopards with recurrent and refractory pyrexia.

Case History Observation and Discussion

Two adult rescued leopards (Shyam and Shama) with recurrent pyrexia refractory to ceftriaxone, amoxicillin, melonex and diminazene aceturate for last 15 days were referred to senior author for the diagnosis and treatment. Detailed clinical examination revealed pyrexia (103.6 - 105 °F), anorexia, breathing difficulty, tachycardia, ocular discharge, lethargy, dullness, diarrhoea, stiffness, petechae in buccal cavity and tongue erosions (Fig.1). Peripheral blood smears and buffy coat blood smear cytology showed the presence of morulae in monocytes (Fig.2) of both Shyam and Shama leopards. Haemato-biochemical investigations revealed low values of haemoglobin, total erythrocytes, packed cell volume and thrombocytes ; and higher values of total leukocytes, neutrophils, ALT, AST and serum creatinine (Table 1.)

Treatment was instituted with intramuscular injection of Imidocarb (@ 5mg/kg at fortnightly interval), intramuscular injection of prednisolone (@ 1.0 mg/kg for initial 5 days), fluid therapy (with Ringers lactate/ normal saline @ 100 ml/kg/day intravenously for 5 to 7 days), oxytetracycline (@ 10 mg/kg intravenously or intramuscularly daily for 15days) and vitamin B complex (3 ml intramuscularly daily for 15 days).

The present report describes a clear cut case of ehrlichiosis, caused by *E. canis,* in two adult rescued leopards diagnosed on the basis of *Ehrlichia*-positive blood smears (Fig.2). We could not trace any report of Ehrlichiosis in leopards in India. It appears to be the first clinical report of ehrlichiosis in leopards, caused by *E. cains*, in India. Pyrexia (103.6 - 105 °F), anorexia, breathing difficulty, tachycardia, ocular discharge, lethargy, dullness, diarrhoea, stiffness, petechae in buccal cavity and tongue erosions in the leopards were nonspecific and resembled symptoms caused by infectious agents, such as respiratory viruses, blood protozoa or rickettsia. Low values of haemoglobin (13.0 and 10.2 g/ dl), total erythrocyte count (7.88 and 5.47 x 10^6/c mm), packed cell volume (38.6 and 31.3 %) and thrombocytes (94000 and 90000/µL) as compared to those (Hb 14.7 ± 2.5 g/dl , TEC , 8.7 ±2.0 x 10^6/c mm · PCV 48.5 ± 6.8 %) reported for healthy leopards (Shanmugam *et al*., 2017) confirmed anaemia and supported clinical suspicion of blood protozoan or rickettsial infection. Detection of intracytoplasmic inclusions in monocytes (Fig. 2) resembling with that of *E. canis* in dogs

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Table 1. Haemato- biochemical indices in leopards suffering from Ehrlichiosis

Haematobiochemical Indices	**Case 1**(Shyam)	**Case 2** (Shama)
Haemogobin (g/dl)	13.0	10.2
Total erythrocytes (10^6)	7.88	5.47
Packed cell Volume (%)	38.6	31.3
MCV (fl)	48.98	57.22
MCH (fmol/cell)	16.75	18.65
MCHC (Hb/L)	34.20	32.59
RDW (fl)	19.50	19.30
Erythrocyte Sedimentation Rate (1 hr.)	100	90
Total leukocyte count (10^3)	30.5	29.8
Neutrophils (%)	92	85
Lymphocytes (%)	3	10
Monocyte (%)	3	02
Eosinophil (%)	2	03
Platelets (µL)	94,000	90,000
ALT (U/L)	82.0	50.0
AST (U/L)	113.0	56.0
SAP (IU/L)	31.0	48.0
Total Bilirubin (mg/dl)	00.5	00.3
Direct Bilirubin (mg/dl)	00.3	00.2
Indirect Bilirubin (mg/dl)	00.2	00.1
Total serum proteins (g/dl)	08.1	06.5
Serum albumin (mg/dl)	04.1	03.1
Serum Globulin (mg/dl)	04.0	03.4
A:G (g/dL)	1.0	00.9
Blood Urea Nitrogen (mg/dl)	37.8	30.8
Serum Creatinine (mg/dl)	03.4	01.3
Na (mmole/l)	169.0	160.0
K (mmole/l)	5.3	5.1
Cl (mmole/l)	134.0	126.0
Blood Smear	*E. canis*	*E. canis*

confirmed that the disease in the leopards was feline ehrlichiosis caused by *E. canis*. Clinical manifestations in the present cases are supported by the observations of Varshney *et al.* (2009) in domestic cat diagnosed with granulocytic ehrlichiosis. Tachycardia and dyspnoea were due to anaemia. Examination of blood smears is the most common method of diagnosing ehrlichiosis but serological and molecular techniques are being increasingly recommended for species identification. Since the morulae in monocytes were detected in the present cases, diagnosis of ehrlichiosis was confirmed. The vector for canine and feline ehrlichia and babesia in India is the tick *Rhipicephalus*. The clinical signs and low values of haemoglobin, erythrocytes and packed cell volume observed in leopards at the time of referral were though nonspecific but have been commonly described in ehrlichiosis (Egenavall *et al*., 1994). Recurrent refractory pyrexia in leopards was possibly due to ehrlichiaemic phase as has been observed in other animal species (Greig *et al.*, 1996). Increased values of ALT, AST and creatinine as compared to those reported for healthy leopards (Shanmugam *et al.,* 2017) indicated liver, muscular (cardiac/other muscle) and kidney insult in leopards suffering with ehrlichiosis.

Fig. 1. Showing lesion on tongue (arrow) of leopard with ehrlichiosis

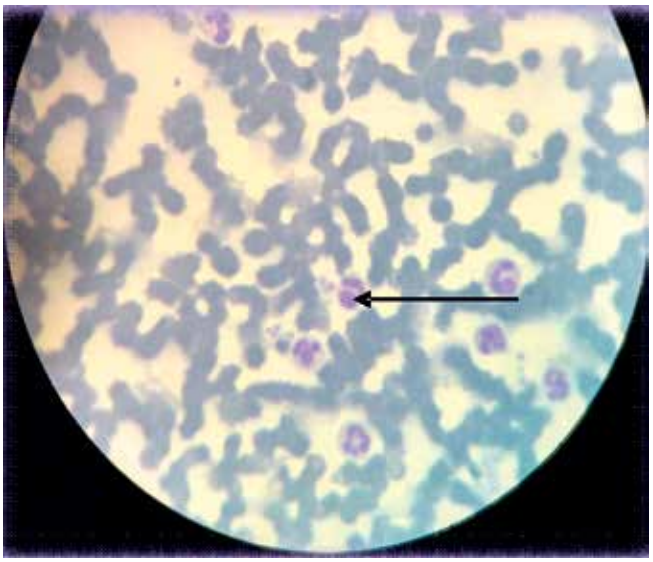

Fig. 2: Blood smear showing morula of *Ehrlichia canis* in monocytes of Leopard with recurrent refractory pyrexia.

The treatment with imidocarb, fluids (Ringer's lactate/DNS), prednisolone ,oxytetracycline, and vitamin-B complex yielded favorable results. Immediate fluid therapy was aimed to restore peripheral circulation and counteract dehydration. Imidocarb and oxytetracycline/doxycycline are regarded as drugs of choice in the management of ehrlichiosis. Temperature started decreasing by 3rd day and became normal by 5thday .

Detection of ehrlichial inclusions combined with case history, low haemoglobin, total erythrocytes and packed cell volume and response to imidocarb provides enough clues for establishing a diagnosis of ehrlichiosis. Increased values of ALT, AST and serum creatinine suggested multi organ dysfunctions in leopards with ehrlichiosis.

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