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Hematobiochemical Disorder in Camels Suffering from different Hemoparasites

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Authors' Contribution
TA and MYT conceived and designed the study. TA, SAA, AA, SA and AI executed the experiment and analyzed the sera and tissue samples. NA, MS, AM and SU analysed the data and drafted the manuscript.

Key words
Haematology, Biochemical analysis, Haemoparasites, Camels.

ABSTRACT

Haemoparasites disturb camel production in tropical countries. Four major haemoparasites in camels are *Trypanosoma*, *Dipetalonema*, *Babesia*, and *Anaplasma*. The current study was executed to investigate haematobiochemical alterations in camels during haemoparasitism. Blood samples were collected from 200 camels of either sex and different ages from district Attock. Samples were processed for haematobiochemical analysis and thin smear stained with Field stain according to standard procedure. Out of 200 camels 46% of them have blood parasites. *Trypanosoma* was the most prevalent (32.5%) followed by diptelonemiasis (8%). *Babesia* and *Anaplasma* was seen in 3% camels. Prevalence of diptelonemiasis was significantly higher in young than older camels. Blood analysis revealed that blood parasites infection in camels results in decrease in PCV, Hb, RBC and PLT values and increase in TLC values. On the other hand, serum analysis revealed increase in ALT, AST, ALP and GGT while decrease in total protein and albumin ($p>0.05$).

INTRODUCTION

Camels are found in arid and semi-arid areas of the world. People of different regions use camels for different purposes such as pack animal, for meat and recreation (Ahmad *et al*., 2004; Durrani *et al*., 2017). The Pakistani camel is well known for higher milk production. The Mareecha breed is considered as the best milk producing camel of the world, with an average milk yield of 4179L per year. According to economic survey of Pakistan, 1 million camels are present in our country (Faraz *et al*., 2013; Pasha *et al*., 2013). Haemoparasitic diseases have devastating effect on productivity, health and working capabilities of camels. Little research has been done on camel disease especially haemoparasitic diseases as compared to other ruminants in Pakistan. Haemoparasitic diseases in the camels have been reported and their prevalence has increased during the past decade. Among vector-borne protozoan diseases, trypanosomiasis or Surra, babesiosis, anaplasmosis, and diptelonemiasis are principally important (Parsani *et al*., 2008; Rabana *et al*., 2011; El-Naga *et al*., 2016) and characterized by elevated body temperature, anemia, emaciation, lymphadenopathy, edema and sudden death. In Pakistan trypanosomiasis and babesiosis are prevalent as a major threat to the camels causing heavy financial losses (Enwezor *et al*., 2005; Sobhy *et al*., 2017; Desquesnes *et al*., 2013; Shah *et al*., 2013). Haemoparasites affect the blood cells and result in change of blood parameters (Chaudhary and Iqbal, 2000; Padmaja, 2012; Pasha *et al*., 2013; Parsani *et al*., 2008; Barghash *et al*., 2106; Ismael *et al*., 2014).

Study of blood components can lead to accurate diagnosis and assist in deciding line of treatment (Farooq *et al*., 2011). Serology indicates the amount of different enzymes present in the serum such as alanine aminotransferase (ALT), aspartate transaminase (AST), alkaline phosphatase (ALP), gamma-glutamyltransferase (GGT). Comparison of serological values of healthy animals with diseased animals can give us clue about the changes inflicted by pathogen and tissues response to that particular stimuli (Tehseen *et al*., 2015).

Keeping in view the economic importance of camel and the losses caused by haemoparasitic diseases this study was designed to find causative pathogens for blood

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protozoan in one humped camel (*Camelus dromedarius*). The aim of this study was to determine the prevalence of haemoparasites in camels, which is the first step necessary before control measures can be implemented against haemoparasites.

MATERIALS AND METHODS

Collection of blood samples

A total of two hundred blood samples were collected under natural conditions from camels present in district Attock. Immediately, smears were prepared from the collected blood for detection of haemoparasites and preserved the smear. For hematology and serology blood was collected in anticoagulant coated vacutainer and plain vacutainer, respectively (Sazmand *et al*., 2016). Transportation of sample was carried out under refrigeration (Vap and Andrea, 2015). Four groups naming A, B, C and D were constructed on the basis of age.

Haemoparasites identification

The diagnostic technique for *Trypanosoma* was wet and thin smear method as described by Khosravi *et al*. (2013). Further confirmation was carried out by formol gel test (Tehseen *et al*., 2015). For dipetalomanaisis, From each sample, Wet and thin blood smears was prepared and stained with Geimsa for light microscopic examination of hemoparasites (Karimi *et al*., 2015; Sazmand *et al*., 2016). Modified Knott method was used for detection of microfilaria of *Dipetalonema evansi* (Karimi *et al*., 2015).

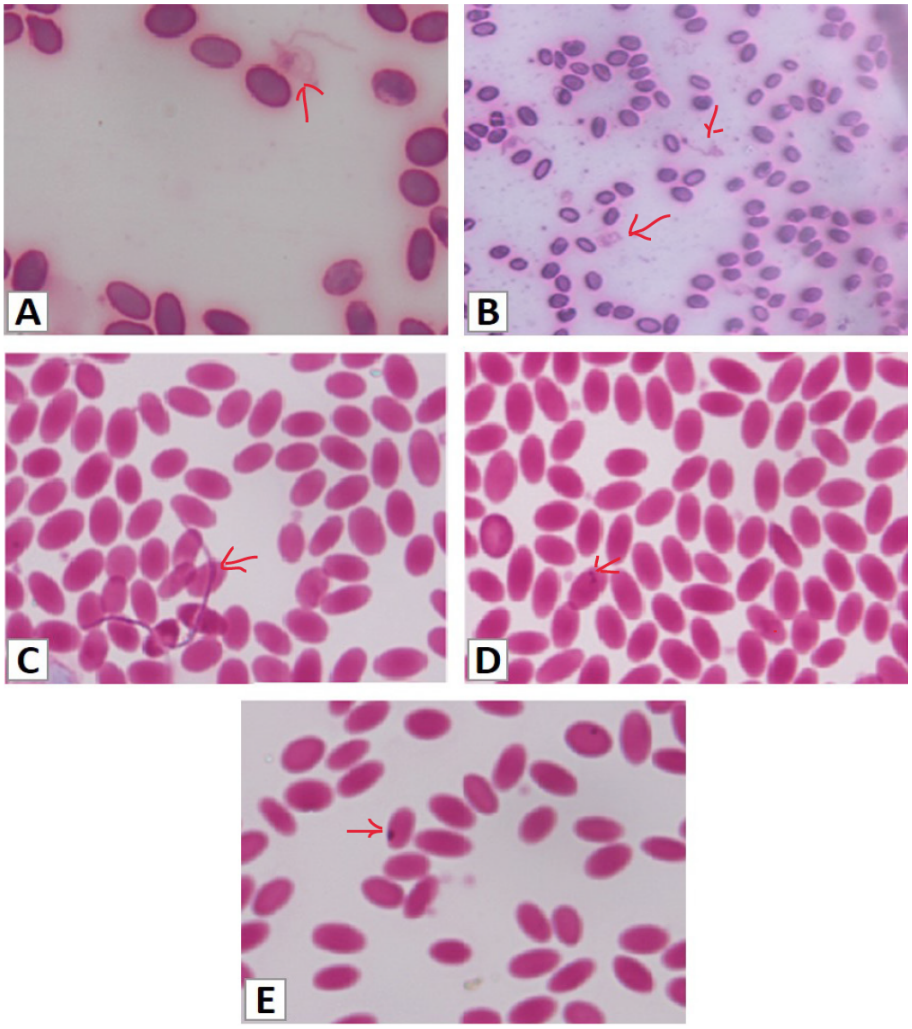

Fig. 1. Blood smear of camels showing haemoparasites: A and B, *Trypanosoma evansi;* C, *Diptelonema evansi;* D, *Bebesia cabuli;* E, *Anaplasma marginale. Magnifying scale for A,C,D and E, 40X; B,10X. Field stain was used for staining of smear.*

Haematology and serology

The blood samples were analyzed for complete blood count including total erythrocytic count (TEC), total leukocytic count (TLC), haemoglobin (Hb) and packed cell volume (PCV) as described by Vap and Andrea (2015). The blood plasma was separated and analyzed for ALT, AST, ALP, GGT, Albumin, total protein and total bilirubin of collected serum samples. Biochemical analyses were carried out using commercial kits (Pars Azmun, Tehran, Iran) according to manufacturer's instructions.

RESULTS

Study revealed presence of five different haemoparasites present in camels shown in Figure 1. In our study prevalence of trypanosomiasis was highest among all five haemoparasites under investigation as shown in Table I. Out of 200 (131 males and 69 females) camels investigated in this study 108 camels were negative for haemoparasites and 92 camels were positive for haemoparasites. Among those 92 camels which were positive for haemoparasites, 64 were males and 28 were females camels. On the basis of haemoparasites 48 males and 17 females were positve for trypnosomiasis, 11 males and 5 females were positive for diptelonemiasis, Bebesiosis was positive in 2 males and 4 females whereas, 3 males and 2 females showed positve results for anaplasmosis.

Haematological analysis of hemoparasitized camel

Haematological results showed that in case of *Trypanosoma evansi* infection the packed cell volume, RBCs and Hb were significantly decreased in haemparasitized camels as compared to healthy camels (Table II). Infection of other three blood parasites also follows the same trend and result in decrease in PCV, Hb, RBC and PLT values whereas TLC was increased significantly (Table II).

Biochemical analysis of haemoparasitized camels

Almost constant trend was observed in case of all haemoparasites that result in considerable increase in ALP, ALT, AST and GGT. On the other hand, total protein and albumin was decreased due to infection of all blood parasites (Table III).

Table I.- Prevalence of trypanosomiasis, diptelonemiasis, bebesiosis and anaplasmosis in different sex and age group of camels.

Risk factor	Trypnosomiasis	P<0.05	Diptelonemiasis	P<0.05	Bebesiosis	P<0.05	Anaplasmosis	P<0.05
Sex								
Male	48	0.085	11	0.776	02	0.092	03	0.793
Female	17		05		04		02	
Age (years)								
0-3	03	0.000	09	0.000	-	0.321	-	0.693
3-6	03		04		-		01	
6-9	26		02		01		01	
Above 9	33		01		05		03	

P<0.05 is considered significant.

Table II.- Mean haematological values ±S.D of healthy adult and haemoparasitized adult camels*.

Parameter	Healthy animals (108)	Trypanosomiasis (65)	Diptelonemiasis (16)	Bebesiosis (6)	Anaplasmosis (5)	P<0.05
Packed cell volume (%)	33.21 ±1.82	24.98 ±0.45	26.03 ±0.74	28.3 ±0.0	27.3 ±0.0	0.00
Haemoglobin (g/dl)	12.37 ±0.42	9.97 ±1.97	9.23 ±1.21	9.61±0.0	8.92 ±0.0	0.004
Red blood cell count (10^6/mm^3)	8.93 ±0.38	7.64 ±0.89	7.50 ±0.57	6.42 ±0.0	5.90 ±0.0	0.00
Total leukocytic count (10^3/mm^3)	12.88 ±0.69	17.01 ±1.87	16.42 ±1.24	14.78±0.0	13.89 ±0.0	0.00
PLT (x10^9/L)	273.7 ±17.54	190.50 ±12.20	185 ±17.23	191 ±0.0	211 ±0.0	0.00

P<0.05 is considered significant. *Blood samples were taken from different age groups randomly. Duration of disease for suffering animals was unknown.

Table III.- Mean serum chemistry values ±S.D of healthy camels and haemoparasitised camels.

Parameter	Healthy animals (108)	Trypanosomiasis (65)	Diptelonemiasis (16)	Bebesiosis (6)	Anaplasmosis (5)	P<0.05
ALP (u/l)	76.36 ±8.13	84.10 ±6.43*	83.47 ±12.2*	83.96 ±0.0*	79.82 ±0.0*	0.00
ALT (u/l)	11.03 ±4.2	14.49 ±1.98*	13.04 ±2.65*	13.78 ±0.0*	13.89 ±0.0*	0.00
AST (u/l)	102.14 ±12.7	123.96 ±6.87*	114.25 ±15.61	118.01 ±0.0*	132.90 ±0.0*	0.729
GGT (u/l)	12.96 ±0.83	14.70 ±0.54	15.81 ±1.51*	15.64 ±0.0*	15.92 ±0.0*	0.00
Total protein (g/dl)	7.32 ±0.79	6.41 ±0.39*	6.37 ±2.50*	5.90 ±0.0*	6.22 ±0.0	0.015
Albumin (g/dl)	4.00 ±0.37	3.15 ±0.19	3.20 ±0.68	3.01 ±0.0	2.94 ±0.0*	0.00
Total bilirubin (g/dl)	0.1 ±0.05	0.2 ±0.05	0.1 ±0.05	0.1 ±0.0	0.1 ±0.0	0.299

*significant change in parameter with respect to healthy animals. P<0.05 is considered significant. *Blood samples were taken from different age groups randomly. Duration of disease for suffering animals was unknown.

DISCUSSION

Infectious diseases and particularly vector-borne protozoan diseases have been serious threat for animal health and productivity in developing countries. The overall prevalence of trypanosomiasis, diptelonemiasis, babesiosis and anaplasmosis during present study was 32.5%, 8%, 3% and 3%, respectively which is not in accordance with the previous studies in Pakistan. Previously, 15% trypanosomes infection was recorded in camels (Durrani *et al*., 2017). This difference in prevalence could be due to different populations' locations, sample size and prior treatment within the different areas of the country. Present study showed higher prevalence of blood protozoan than previously reported studies in Pakistan (Ahmed *et al*., 2004).

Prevalence of diptelonemiasis was significantly higher in young than older camels. However, trypanosomiasis, babesiosis and anaplasmosis prevalence was more in older age camels than young age camels.

The relatively higher prevalence of blood protozoan infection in older camels (above 9 years), could be attributed to the poor management, movement from one place to another and heavy stress owing to the involvement of this age group in transportation (Durrani *et al*., 2017). The results also revealed statistically non-significant difference of infection in male and female animals. Conversely, some studies have reported significantly higher prevalence in female camels (Tehseen *et al*., 2015) as compared to males while few researchers have also reported higher prevalence in the male camels (Hussain *et al*., 2016). In short, all sex wise prevalence findings about blood protozoa showed that both sexes are equally susceptible to the disease. However, higher rates of infection in females could be due to the physiological stress leading to decreased resistance and in turn, higher vulnerability to the infection. In contrast, higher infection rates in males can be attributed to the increased physical work leading to fatigue and stress.

Presence of haemoparasites in camels results in decrease in growth rate and milk production and may changes in haematological parameters (Rabana *et al*., 2011). Blood analysis in present study revealed that blood parasites infection in camels results in decrease in PCV, Hb, RBC and PLT values and increase in TLC values. On the other hand, serum analysis revealed increase in ALT, AST, ALP and GGT while decrease in total protein and albumin (p>0.05).

The results of haematological analysis of trypanosomiasis positive camel samples and healthy camel shows that trypanosomiasis positive samples have decreased PCV, RBCs, Hb and platelets count. These results were similar to, Enwezor and Sackey (2005) and Chaudary and Iqbal (2000). TLC was increased in trypanosomiasis positive camel samples as compared to healthy camel samples. These results were in line with previous studies (Enwezor and Sackey, 2005; Chaudhary and Iqbal, 2000) but did not match to the findings of Ahmad *et al*. (2004) who reported leukopenia in trypnosomiasis positive camel samples. Comparison of Serological analysis of trypanosomiasis positive samples and healthy camels samples illustrated the increase in AST, ALT, ALP, GGT and decrease in Albumin and total protein in case of trypanosomiasis positive camel samples. These results were similar to Chaudhary and Iqbal (2000) and Ahmed *et al*. (2004).

Diptelonemasis positive camel samples shows decrease in PCV, RBCs, Hb and platelets count and increase in TLC as compared to healthy camel samples. These results were similar to the findings of Muhammad *et al*. (2004). A similar study in dogs also gave same results. Comparison of Serological analysis of diptelonemasis positive camel samples and healthy camels samples illustrated the increase in AST, ALT, ALP, GGT and decrease in Albumin and total protein in case of diptelonemasis. Serological

analysis of diptelonemasis in dogs resulted in increase in AST, ALT, total protein and decrease in Albumin (Hashem and Badawy, 2008).

Babesiosis positive camel samples show decline in PCV, RBCs, Hb and platelets count and increase in TLC as compared to healthy camel samples. These finding were similar to studies done previously (Ismael *et al*., 2014; Abd-Elmaleck *et al*., 2015). A study was done by Esmaeil-Nejad *et al*. (2012) on haematological and biochemical parameters in small ruminants naturally infected with *Babesia ovis* also mentioned same changes in PCV, RBCs, Hb and platelets count due to bebesiosis in small ruminants.

Anaplasmosis positive camel samples displayed decline in PCV, RBCs, Hb and platelets count and rise in TLC as compared to healthy camel samples. Similar findings were reported by Al-Saad (2009). Evaluation of serological study of anaplasmosis positive camel samples and healthy camel's samples demonstrated the increase in AST, ALT, ALP, GGT and decrease in albumin and total protein in case of anaplasmosis. Al-Saad (2009) also demonstrated increase in AST, ALT and decrease in total protein due to Anaplasmosis.

CONCLUSION

From the findings of this study it can be concluded that haemoparasitic diseases are present in Attock district of Pakistan and cause disparities in various hematological as well as biochemical parameters of camels. There is dire need of appropriate extension methods and tools to create awareness among camel owners about acaricidal and anti-protozoan drug use for better preventive and control measures against haemoparasitic diseases in camels.

Statement of conflict of interest

Authors have declared no conflict of interest.

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