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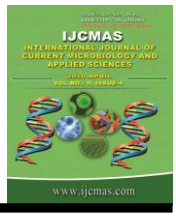

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Haemato-Biochemical Profile of Black Bengal Goat on Different Gastro-Intestinal Parasitic Load

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A B S T R A C T

A study was conducted of a total of one thousand four hundred sixteen blood samples collected from one hundred eighteen Black Bengal does in every months during July, 2004 to June, 2005 to assesses haemato biochemical profile of black Bengal goat in accordance to the level of gastrointestinal nematodes as a good indicator of parasitic load to take decision for selection of goat to conserve the genetic resources with their up gradation for better production and supply of healthy, disease free products for human use. In this aspect, the present study was conducted with the objective to assess the haemato-biochemical profile of Black Bengal goat. The does of base stock having EPG count more than 1200 (Group 3) showed anaemia and erythrocytopenia with significant decrease in PCV and TEC and hypoproteinaemia with reduction in TSP and SA. The Group 3 animals also showed significant increase in alkaline phosphatase and AST activity. Though the affected goats do not show clinical illness but EPG count more than 1200 can lead to anaemia, erythrocytopenia, hypoproteinaemia and increased activity of serum AST and alkaline phosphatase enzymes give birth to inferior progenies generation after generation and the progenies are also susceptible to infection while benefit can be obtained by rearing goats of low EPG count along with corresponding high PCV, Hb and TSP value perform better generation after generation.

Introduction

Among the livestock of India, goats have fulfilled agricultural, economic, cultural and even religious roles from very early times in human civilization. They are the most adaptable and geographically widespread livestock species ranging from the high altitude to desert and humid coastal areas of India and contribute to the rural economy where all other means of agriculture is a failure.

Goats make an important contribution to the sustenance of small and marginal landholders and landless rural people by their contribution towards marketable commodities such as meat, milk, fibre and skin. It also plays a leading role in eradication of poverty in small farmers and landless labourers by self-employment and thus improvement of goat production is necessary to benefit the rural community. (Dhara *et al.,* 2011)Though most of the Indian breeds of goats are well adapted to the harsh environment, low nutrition, tropical disease and poor water quality, gastrointestinal parasites are considered to one of the top ten ranked disease in goats (Rout and Chauhan, 2004).

In the world when goat producers are facing increasing problems due to the rapid spread of antihelmintic resistance, the battle against gastrointestinal nematodes is a difficult one. Widespread infection with internal parasites in grazing animals, associated production losses, antihelmintic costs and death of infected animals are some of the major concerns. India is paradise of many parasites due to its hot and humid climatic condition. The climatic factor may favour the development of gastro-intestinal parasites during the period of nutritional stress and wet season in the tropical area (Hawlader *et al.*, 2002) like India. The most important helminthes infections of livestock are infections of grazing ruminants by nematodes residing within the gastrointestinal (GI) tract of the vertebrate host.

Strongyle infestation significantly affects the body weight gain of sheep and goat and this reflects in haemato biochemical profile of the animal which ultimately a good indicator of parasitic load to take decision for selection goat to conserve the genetic resources with their up gradation for better production and supply of healthy, disease free products for human use. In this aspect, the present study was conducted with the objective to assess the haemato- biochemical profile of Black Bengal goat in accordance to the level of gastrointestinal nematodes

Materials and Methods

The study was conducted at Mohanpur village under Haringhata block in the Nadia district of West Bengal state which is under New Alluvial zone of Lower Gangetic Plain region of India with a total of one hundred eighteen Black Bengal does with age of three months and above. All the one hundred eighteen does were divided into three groups depending upon the mean annual egg per gram of faeces (EPG) count of gastrointestinal nematode, *Strongyle* sp. from July 2004 to June 2005 and they were considered as base stock. The groups were - Group 1 (Low EPG < 600), Group 2 (Moderate EPG >600 < 1200) and Group 3 (High EPG > 1200).

All the animals utilized for the present study were maintained in the farmers' house with almost uniform management practice. All the animals were dewormed four times in a year. Deworming was performed on the basis of faecal examination. Vaccination against PPR, Goat pox and FMD was done in every year. A part of individual faecal samples were subjected to direct microscopic examination as well as floatation and sedimentation methods (Soulsby, 1982) for prevalence study of gastrointestinal parasites. All the positive samples were subjected to eggs per gram of faeces (EPG) count for strongyle group of gastrointestinal nematodes, by employing modified McMaster technique (Coles *et al.*, 1992).

Collection of blood

Blood were collected from a total of one hundred eighteen does in each month during the year July, 2004 to June, 2005. Eight

millimeter of blood was drawn out by jugular vein puncture of the goats using standard aseptic procedure after proper marking with animal number.

Out of 8 ml of blood drawn, 3 ml was taken in a sterilized screw capped vial containing the required amount of EDTA @ 1 mg/ml of the blood as anticoagulant. Out of 8 ml blood drawn, 5 ml of blood (without anticoagulant) was kept undisturbed for two to three hours in same syringe to clot for separation of serum.

The serum was taken in a sterilized screw-capped vial and stored at -20°C in a deep freezer after proper marking with animal number till further use. The enzyme activities were estimated within forty eight hours of collection, whereas the other biochemical parameters were estimated within a week after collection.

Haematological estimation

Haemoglobin (Hb)

Hemoglobin was estimated by Sahli's method as described by Schalm *et al.,* (1986). The result was expressed in gram per deciliter (g/dl) of blood.

Packed cell volume (PCV)

PCV of the blood samples were estimated in Wintrobe's haematocrit tube and reading were taken after centrifuging blood at the rate of three thousand r.p.m. for thirty minutes as per standard method of Schalm *et al.,* (1986) and were expressed in percentage (%) of total volume.

Total erythrocyte count (TEC)

TEC was estimated by using haemocytometre as described by Schalm *et al.,* (1986). RBC was differentiated by the characteristic shape and intensity of stain taken. The result of TEC was expressed as millions per cubic millimeter (106/cu.mm).

Total leukocyte count (TLC)

TLC was estimated by using haemocytometre as described by Schalm *et al.,* (1986). WBC was differentiated by the characteristic shape and intensity of stain taken. The result of TLC was expressed as thousand per cubic millimeter (103/cu.mm).

Differential leukocyte count (DLC)

DLC was performed following the method described by Schalm *et al.,* (1986).

Serum biochemical estimation

Total serum protein (TSP)

TSP was estimated spectrophotometrically by Biuret method (Reinhold, 1953) using the diagnostic reagent kit (SPAN, Code No. B0211) and the values were expressed in gram per deciliter (g/dl).

Serum albumin (SA), serum globulin (SG) and albumin and globulin ratio (SA: SG)

Serum albumin was determined spectrophotometrically by the method described by Dumas *et al.,* (1971). The value of serum globulin was estimated by subtracting the value of SA from that of TSP. The value of SA and SG were expressed in gram per deciliter (g/dl).

Aspertate aminotransferase (AST)

Serum AST was estimated spectrophotometrically by 2, 4-DNPH method (Reitman and Frankel, 1957) using the diagnostic reagent kit (SPAN, Code No. 25913) and result was expressed in IU/L of serum.

Alanine aminotransferase (ALT)

Serum ALT was estimated spectrophotometrically by 2, 4-DNPH method (Reitman and Frankel, 1957) using the diagnostic reagent kit (SPAN, Code No. 25912) and result was expressed in IU/L of serum.

Serum alkaline phosphatase

Serum alkaline phosphatase was estimated spectrophotometrically by Kind and Kings method (Kind and King, 1954) using the diagnostic reagent kit (SPAN, Code No. 25904) and result was expressed in IU/L of serum.

Statistical methodology

The effect of the intensity of GI nematode infection (EPG) of the base stock on the haemato biochemical profile was estimated by analysis of variance (Snedecor and Cochran, 1994). The formulae used for statistical analysis were:

$Yijk = \mu + Ai + eik$

Where:
Yik = kth animal of the ith EPG group
μ = overall mean
Ai = Effect of EPG (j = 1 to 3)
eik = Random error on observation distributed NID (0, $\sigma 2e$)

From the above model, it was calculated
CF = Grand Total (GT) ^{2}/N (Number)
Sum Squire (SSA) (Between EPG group) = $\sum$ Ai2 - CF
Sum Squire (SST) (Total) = $\sum$ Yik2 – CF
Sum Squire (SSe) (error) = SST – SSA

The critical difference test (CD test) was carried out for the traits, which showed significant differences. This is done to compare between the means of sub-classes by applying the following formula.

$| B1- B2 | > t1/2\alpha (N-5) X \sqrt{MSe}$ (Mean sum square error) (1/N1 + 1/N2)

Results and Discussion

Haemato-biochemical study

The haemato-biochemical values of a total of one thousand four hundred sixteen blood samples collected from one hundred eighteen Black Bengal does in every months during July, 2004 to June, 2005 were estimated and the results were analyzed in accordance with the level of EPG.

Haematological study

The result of haematocrit values has been presented in Table 11 and analysis of variance has been presented in Table 12. The result of haemoglobin concentration revealed a gradual decrease in Hb percentage from Group 1 to Group 3 (10.15%, 9.94% and 9.03% respectively). The Hb concentration of Group 3 was statistically significant (P< 0.01) from both Group 1 and Group 2. The difference between Group 1 and Group 2 was non-significant. The statistical analysis of PCV (%) and TEC (106cumm) values showed that the decrease in PCV and TEC of Group 3 (24.96% and 10.25 X 106cumm) were significant (P< 0.01) in comparison to Group 1 (28.85% and 12.62 X106cumm) and Group 2 (28.04% and 12.45 X 106cumm).

It was further evident that changes in the values of TLC did not follow a consistent trend and the variation among Group 1 (9.78 x 103cumm), Group 2 (9.86 x 103cumm) and Group 3 (9.56 x 103cumm) were non-significant. The statistical analysis of DLC values revealed non- significant changes in the individual cells between the three groups.

Table.1 Haematological values of Black Bengal goat in different level of EPG count (Mean ± SEM)

Groups	**Hb (g/dl)**	**PCV (%)**	***TEC* (10^6/cu.mm)**	**TLC (10^3/cu.mm)**	**DLC (%)**				
					Lymph (%)	**Ne (%)**	**Eo (%)**	**Baso (%)**	**Mono (%)**
Group-1 (N = 49)	10.15 ± 0.10 [a]	28.85 ± 0.44 [a]	12.62 ± 0.13[a]	9.78 ± 0.15	58.73 ± 0.61	38.55 ± 0.58	2.06 ± 0.11	0.16 ± 0.05	0.49 ± 0.09
Group-2 (N = 58)	9.94 ±0.09 [a]	28.04 ± 0.38 [a]	12.45 ± 0.11[a]	9.86 ± 0.13	57.79 ± 0.48	39.28 ± 0.45	2.17 ± 0.13	0.26 ± 0.06	0.50 ± 0.07
Group-3 (N =11)	9.03 ±0.14 [b]	24.96 ±0.34 [b]	10.25 ± 0.34 [b]	9.56 ± 0.20	57.91 ± 1.37	39.09 ± 1.39	2.36 ± 0.15	0.18 ± 0.12	0.45 ± 0.16
F Value	11.74**	8.39**	32.06**	0.42	0.77	0.49	0.64	0.75	0.03

Values bearing same or no superscript within a column do not differ significantly (P< 0.01).
* P < 0.05 ** P < 0.01

Serum biochemical study

The result of total serum protein (TSP), albumin (SA), globulin (SG) and SA : SG ratio and the statistical analysis of variances have been presented in Table 13 and Table 14. The result revealed a significant (P< 0.01) decrease in TSP of Group 3 (5.99 g/dl) than that of Group 1 (6.68 g/dl) and Group 2 (6.66 g/dl). There was gradual decrease in the value of albumin from Group 1 to Group 3.

The statistical analysis of the values showed a significant (P< 0.01) decrease in albumin content of Group 3 than Group 1 and Group 2. The changes between Group 1 and Group 2 were non-significant. The changes in globulin values of the three groups did not show a definite trend and the changes were non-significant. The SA: SG ratio showed a decreasing trend from Group 1 to Group 3. The analysis of variance indicated that the SA : SG ratio of Group 3 (0.86) was significantly (P< 0.01) lower than Group 1 (1.08) and Group 2 (1.04). The result of serum enzyme activity is presented in Table 13 and the statistical analysis of the result is presented in Table 14. There was a sharp increase in the alkaline phosphatase activity of Group 3 (158.14 IU/L) than Group 1 (132.36 IU/L) and Group 2 (134.72 IU/L) and the increase in the value of Group 3 was significant (P< 0.01) than both Group 1 and Group 2.

The result showed that Group 3 (66.11 IU/L) had significantly (P< 0.01) higher activity of AST than Group 1 and Group 2. There was slight decrease in the activity of AST in Group 2 (60.89 IU/L) than Group 1 (61.22 IU/L) but the variation was statistically non-significant. The result of ALT showed that Group 3 (33.19 IU/L) had higher ALT activity than Group 1 (31.38 IU/L) and Group 2 (31.94 IU/L) but the changes within groups were non-significant.

Haemato- biochemical study

Haematological study

The gastrointestinal nematode infections are generally known to cause serious impact on the major haematological parameters like Hb concentration, PCV and TEC resulting in anaemia. In the present investigation, it was evident that the goats having EPG level higher than 1200 showed significantly lower level of Hb, PCV and TEC. This result was corroborated with the observation of Borah (1982), Siddiqua *et al.*, (1990), Mottelib *et al.*, (1993), Nguyen *et al.*, (1998) and Nsoso *et*

al., (2001) in natural infection with GI nematodes. Similar observation was also reported by Rahman and Collins (1990), Sharma *et al.,* (2000), Fakae *et al.,* (2004) in experimental basis.

The erythrocytopenia and anaemia as observed in the present study might be due to direct blood sucking activity of some of the nematodes in addition to causing leakage of blood from the site of attachment resulting in decrease in TEC and PCV. Marked blood loss resulting as haemorrhagic anaemia due to Haemonchosis was also reported by Chakrabarti (1994). Moreover, some nematodes might have cause excessive loss of plasma protein into the GI tract causing mild nutritional anaemia. Trichostrongylus infection causes anaemia due to impaired erythropoiesis reducing the life span of erythrocytes. In the present study TLC and DLC values did not showed any significant changes. However, Siddiqua *et al.,* (1990) and Mottelib *et al.,* (1993) reported an increase in these values. Under natural condition repeated exposure to nematode infection in the goat might have failed to stimulate the immune system as a result leucocytosis was not marked in the present study.

Serum biochemical study

The hypoproteinaemia with decreased level of TSP and serum albumin is an important consequent of gastrointestinal parasitism associated with protein loosing enteropathy. The serum albumin level showed a significant decrease with high level of EPG in Black Bengal goat. Similar observation was also reported earlier by Siddiqua *et al.,* (1989), Rahman and Collins (1990) and Kar *et al.,* (2007). The protein loss in GI parasitism might be due to selective loss of albumin having smaller size and osmotic sensitivity to fluid movement. Hypoalbuminaemia in the present study obviously has been aggravated by increased catabolism of albumin and protein malabsorption through the damaged intestinal mucosa (Tanwar and Mishra, 2001). Serum globulin did not reveal any significant change as reported earlier by Rahman and Collins (1990). On the contrary Ahmad *et al.,* (1990), Chakraborty and Lodh (1994) and Kar *et al.,* (2007) recorded increased level of serum globulin which might be due to the severity of the infection. As a result, serum albumin and serum globulin ratio also showed a corresponding decrease as observed by Ahmad *et al.,* (1990).

The activities of serum AP and AST were elevated in the goats with high level of EPG (>1200). The elevated level of these enzymes is considered as good indicator of hepatic damage (Kaneko *et al.,* 1997). However, elevated level of these enzymes has also been reported in GI nematode infection (Chakraborty and Lodh, 1994 and Sharma *et al.,* 2001) and also in heavy infection with Monieziosis (Kar *et al.,* 2007). The increased level of AST activity might be associated with damage to the intestinal mucosa by blood sucking parasites and also by severe blood loss. It might also be due to the immature stages of *Fasciola* sp. which damage the liver tissue. Moreover, toxic metabolites of the parasites and from the tissue damage caused by the parasites might be absorbed and detoxified in the liver as liver is the detoxifying organ resulting in increased activity of these enzymes.

The haemoglobin liberated due to breakdown of RBC, as evident from the result might be partly taken up, catabolized in the spleen with release of bilirubin in the blood. This excess bilirubin was utilized by liver resulting in increased activities of these marker enzymes. The increased serum bilirubin level was associated with elevated alkaline phosphatase activity (Kaneko *et al.,* 1997).

Table.1 Analysis of variance of haematological values of Black Bengal goat in different level of EPG

Haematological parameters	Source	df	Sum of Squares	Mean Square	F
HB	Between groups	2	11.22	5.61	11.74**
	Within groups	115	54.96	0.48	
	Total	117	66.19		
PCV	Between groups	2	135.9	67.95	8.39**
	Within groups	115	931.82	8.1	
	Total	117	1067.73		
TEC	Between groups	2	52.53	26.27	32.06**
	Within groups	115	94.22	0.82	
	Total	117	146.76		
TLC	Between groups	2	0.84	0.42	0.42
	Within groups	115	114.21	0.99	
	Total	117	115.04		
Lympho	Between groups	2	24.54	12.27	0.77
	Within groups	115	1823.98	15.86	
	Total	117	1848.52		
Neutro	Between groups	2	14.17	7.09	0.49
	Within groups	115	1670.62	14.53	
	Total	117	1684.79		
Eosino	Between groups	2	0.91	0.46	0.64
	Within groups	115	81.64	0.71	
	Total	117	82.55		
Baso	Between groups	2	0.25	0.13	0.75
	Within groups	115	19.45	0.17	
	Total	117	19.7		
Mono	Between groups	2	0.02	0.01	0.03
	Within groups	115	39.47	0.34	
	Total	117	39.49		

* $P < 0.05$ ** $P < 0.01$

Table.2 Serum biochemical values of Black Bengal goat in different level of EPG count (Mean ± SEM)

Biochemical parameters	Group 1 (N= 49)	Group 2 (N= 58)	Group 3 (N= 11)
Total Protein (g/dl)	6.68 ± 0.07 [a]	69.66 ± 0.06 [a]	**5.99 ± 0.08** [b]
Serum Albumin (g/dl)	3.46 ± 0.03 [a]	3.39 ± 0.03 [a]	**2.76 ± 0.02** [b]
Serum Globulin (g/dl)	3.23 ± 0.05	3.27 ± 0.04	**3.23 ± 0.07**
SA:SG ratio	1.08 ± 0.02 [a]	1.04 ± 0.01 [a]	**0.86 ± 0.01** [b]
Alkaline Phosphatase (IU/L)	132.36 ± 1.37 [a]	134.72 ± 1.22 [a]	**158.14 ± 5.17** [b]
AST (IU/L)	61.22 ± 0.64 [a]	60.89 ± 0.56 [a]	**66.11 ± 2.16** [b]
ALT (IU/L)	**31.38 ± 0.47**	**31.94 ± 0.43**	**33.19 ± 0.69**

Values bearing same or no superscript within a row do not differ significantly (P< 0.01)

Table.3 Analysis of variance of serum biochemical values of Black Bengal goat in different level of EPG count

Serum biochemical parameters	Source	df	Sum of Squares	Mean Square	F
TSP	Between groups	2	4.59	2.30	11.04**
	Within groups	115	23.91	0.21	
	Total	117	28.50		
SA	Between groups	2	4.43	2.22	59.43**
	Within groups	115	4.29	0.04	
	Total	117	8.72		
SG	Between groups	2	0.06	0.03	0.28
	Within groups	115	11.66	0.10	
	Total	117	11.71		
SA: SG	Between groups	2	0.46	0.23	20.19**
	Within groups	115	1.31	0.01	
	Total	117	1.78		
AP	Between groups	2	6136.52	3068.26	28.75**
	Within groups	115	12272.76	106.72	
	Total	117	18409.28		
AST	Between groups	2	259.15	129.57	5.99**
	Within groups	115	2487.08	21.63	
	Total	117	2746.22		
ALT	Between groups	2	30.95	15.48	1.49
	Within groups	115	1193.38	10.38	
	Total	117	1224.33		

* P < 0.05 ** P < 0.01

There was no significant change in the activity of serum ALT which was corroborated with the findings of Ahmed and Ansari (1989). However, Chakraborty and Lodh (1994), Sharma *et al.*, (2001) and Kar *et al.,* (2007) reported significant increase in the ALT value which might be associated with severity of infection. The does of base stock having EPG count more than 1200 (Group 3) showed anaemia and erythrocytopenia with significant decrease in PCV and TEC and hypoproteinaemia with reduction in TSP and SA. The Group 3 animals also showed significant increase in alkaline phosphatase and AST activity. Though the affected goats do not show clinical illness but EPG count more than 1200 can lead to anaemia, erythrocytopenia, hypoproteinaemia and increased activity of serum AST and alkaline phosphatase enzymes.

So it may be suggested that apart from other selection criteria like growth, biometry, reproductive performance of ancestors, selection of GI nematode resistant Black Bengal goats on the basis of faecal egg count of nematodes and its indicator trait i.e. PCV may be considered as an important selection tool for improvement of Black Bengal goat in the farmers' house towards economic goat rearing and hence this breeding practice has a potential for improving the livelihood of the poor farmers.

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