Indian Journal of Animal Sciences **82** (8): 818–821, August 2012

Exploration of resistance/resilience against gastrointestinal nematode infection in Garole sheep

J D GHOSH[1], RUMA JAS[2] and GAUTAM BORDOLOI[3]

West Bengal University of Animal and Fishery Sciences 37, K B Sarani, Kolkata, West Bengal 700 037 India

Received: 20 May 2011; Accepted: 16 January 2012

ABSTRACT

Possible existence of resistance and/ resilience against gastrointestinal nematode (GIN) infection in 160 Garole sheep was explored under field condition. Monthly qualitative and quantitative faecal examinations and coprocultural examination of all the Garole sheep were performed for 1 year. Based on the monthly faecal egg count (EPG), after 1 year the sheep were divided into group (Gr) 1 showing consistently lowest EPG, Gr 2 with consistently moderate EPG and Gr 3 with consistently highest EPG. Thereafter blood sample was collected from representative number (6) of sheep in each group on 4 occasions at 1-month-interval for estimation of haemoglobin concentration (Hb) and packed cell volume (PCV). The overall prevalence of GIN in the sheep under study was 75.41%. *Strongyle* nematodes constituted 72.92% to which the highest contribution was of *Haemonchus contortus* (70.17%) followed by *Oesophagostomum* (15.08%), *Trichostrongylus* (9.75%), *Strongyloides* (2.75%) and hookworms (2.17%). The overall intensity of infection in terms of mean EPG was 422.37 with the highest in September (950.26) and the lowest (231.02) in March. The mean overall EPG of Gr1 (128.00) was significantly (P>0.05) lower than Gr 2 (434.38) and Gr 3 (704.73). The Hb and PCV values did not differ significantly due to the difference in intensity of infection (EPG) in the 3 groups of sheep.

Key words: Garole sheep, Gastrointestinal nematode, Nematode resistance, Sheep

The home tract of Garole sheep is South 24–Parganas district of West Bengal, especially the saline belt of Sundarban Delta (Sharma *et al*. 1999). Dominated by a variety of helminths the gastrointestinal parasites (GIP) are responsible for causing significant economic losses attributable to reduced weight gain (Jas and Ghosh 2009), retarded growth and impaired reproductive efficiency (Lloyd and Soulsby 1978, Sykes 1994). In sheep, GIP constitutes the nematode parasites as the commonest and major component and it also occurs with high prevalence rate in the native tract of Garole sheep (AINP on GIP Annual Progress Reports-2007–08, WBUAFS, Kolkata). Systematic study on GIP in Garole sheep is lacking and it continues to be a major limiting factor for their optimal productivity. This sheep has also exhibited resistance to helminth infections (Nimbkar 2003), however there have been little efforts through systematic studies to elucidate this promising trait. This *in situ* study was therefore imperative as a first step towards exploitation of this promising trait in Garole sheep wherein faecal egg output (EPG), haemoglobin (Hb) content and packed cell volume (PCV) in relation to GI nematode infection served as the markers for resistance to the infection.

Present address: [1]Professor and Head (jdg_wbuafs@yahoo.co.in), [2]Research Associate (ruma_k007@yahoo.co.in), [3]Ph. D. student (gauti_022@rediffmail.com), Department of Veterinary Parasitology.

MATERIALS AND METHODS

The home tract of Garole sheep is located between 24°32´ to 22°40´ North latitude and 88°05´to 89°00´ East longitude comprising parts of North and South 24–Parganas districts of West Bengal. Four villages, where Garole sheep is the major livestock species reared under semi-intensive system, under Joynagar -II and Mathurapur-II blocks of South 24-Parganas district, were selected for 18-month study.

Garole sheep of either sex (160: 40 sheep/village), 5 to 12– month-old, were selected. Per-rectal faecal samples from individual animals were collected in plastic bottles containing 10% formalin, at monthly interval for 1 year. Pooled sample comprising the mixture of faecal samples of all the sheep were also collected without preservative for coprocultural examination and determination of species composition of different nematode larvae prevalent in those animals.

Examination of faecal sample: A part of each sample was subjected to qualitative faecal examination by standard sedimentation technique for the presence of trematode eggs and salt flotation technique for cestode and nematode eggs (Soulsby 1982). Modified McMaster's technique (Soulsby 1982) was employed for quantitative faecal examination of

the remaining part of the samples. Monthly coprocultural examination of the faeces pooled from all the sheep under study was also performed by honey-jar coproculture technique (Soulsby 1982) concomitantly with the qualitative and quantitative faecal examinations.

Determination of species composition of third stage larvae (L_3): The species composition of nematodes (comprising strongyles and *Strongyloides*) in the larval samples obtained by coproculture was determined following the standard guidelines of Anonymous (1971). The L_3 were examined under a compound microscope and the characters given in the keys (Anonynuom 1971) were used to identify a minimum of 200 larvae. The per cent composition of different nematode species in the L_3 suspension was calculated accordingly.

Grouping of animals and parameters studied: After performing quantitative faecal examination of all the selected sheep at monthly interval for the initial period of 12 months, the sheep were divided into 3 groups according to their overall faecal egg counts. Group (Gr)1 comprised 18 sheep with consistently lower EPG, Gr 2 of 82 sheep having consistently medium EPG and Gr 3 of 60 sheep with consistently higher EPG.

Blood was collected from representative number (6) of sheep in each group on 4 occasions at 1-month-interval following the standard methods. Faecal samples also were collected concomitantly with collection of blood and quantitative faecal examination (Soulsby 1982) was performed. Haematological parameters studied were haemoglobin concentration (Hb) and PCV. Sahlis' method as described by Schalm *et al.* (1986) was employed for estimation of Hb (gm/dl) in blood, and PCV as percentage (%) of total volume was estimated by Wintrobe's haematocrit method (Schalm *et al.* 1986).

Statistical analysis: Values for all the parameters estimated on different occasions within each group were compared (Analyze-Compare Means) and the mean values along with standard error (SE) were obtained. They were then separately analyzed between the groups and the days of estimation following the Duncan's method (one-way ANOVA) and the significance (P value) was recorded at 5% ($P<0.05$) and 1% ($P<0.01$) levels, using the statistical package for Social Scientist (SPSS), Windows Version 10.0.

RESULTS AND DISCUSSION

The overall prevalence of GIP in Garole sheep was 75.41% with the highest prevalence (85%) in the month of September. Strongyle nematodes recorded the highest contribution (72.92%) to the overall prevalence of GIP (Table 1), which also included *Trichuris* (20.44%), *Moniezia* (10.52%) and paramphistomes (20.44%). Gastrointestinal parasitism commonly occurs with moderate to high prevalence in all the Indian breeds of sheep. Garole sheep, which is commonly believed to be less susceptible to the diseases, is no exception to this as revealed by the results of this study.

Agro-climatic conditions of the areas where this study was undertaken favour the occurrence of gastrointestinal parasites (GIP) in all species of ruminant livestock and a moderate to high prevalence has been recorded in the earlier studies (AINP on GIP Annual Progress Reports-2007–08, WBUAFS, Kolkata). The high prevalence of GIP including the GIN in the Garole sheep as observed in this study was not unreasonable. Although prevalence of GIP in sheep was reported from different parts of India (Nesreen *et al.* 2005, Singh *et al*. 2005, Khillare *et al.* 2006, Saravanan *et al*. 2006), studies on this aspect in Garole sheep have largely been restricted in the state of West Bengal only. Such studies are scanty involving abattoir survey of trematode infections (Purohit 2002, Patra 2002). However, the prevalence of paramphistome infection in their study was lower (24.66%) as compared to the observed prevalence in this study (20.44%) and a high rate of prevalence (9.04%) of *Fasciola*

Table 1. Overall prevalence of naturally occurring gastrointestinal parasites in Garole sheep

Month	Overall prevalence (%)	Strongyle (%)	*Strongyloides* (%)	*Trichuris* (%)	Paramphistome (%)	*Moniezia* (%)
June	76.25	73.75	13.12	10.00	20.62	10.62
July	77.50	74.37	14.37	11.25	20.62	10.62
August	78.12	75.63	14.37	12.50	28.12	10.62
September	85.0	82.50	15.62	10.00	25.62	11.87
October	80.00	77.50	13.75	10.62	23.75	11.87
November	76.25	73.75	12.50	9.37	23.12	10.62
December	73.75	71.25	13.12	8.75	21.25	9.37
January	71.87	68.12	14.37	8.75	20.62	10.62
February	70.62	67.50	13.75	8.75	18.75	10.62
March	71.25	70.00	11.25	8.12	17.50	9.37
April	72.50	70.62	10.00	9.37	13.75	10.62
May	71.75	70.00	8.75	8.12	11.25	9.37
Overall	75.41	72.92	12.92	9.63	20.44	10.52

Table 2. Species composition of infective larvae of gastrointestinal nematode in Garole sheep

Month	Per cent (%) composition of infective nematode larvae				
	Haemonchus	*Oesophagostomum*	*Trichostrongylus*	*Hook worm*	*Strongyloides*
July	70	15	9	1	5
August	71	14	8	2	5
September	70	16	9	2	3
October	73	14	10	1	2
November	69	15	11	1	4
December	70	16	10	2	2
January	72	14	9	3	2
February	69	17	10	2	2
March	68	15	11	4	2
April	69	16	9	2	2
May	71	14	10	3	3
June	70	15	11	2	1
Overall	70.17	15.08	9.75	2.17	2.75

infection was also recorded. The earlier studies recorded the prevalence in respect of presence or absence of the flukes without discrimination between the immature and the mature ones, whereas detection of the faecal eggs obviating the exclusive presence of immature flukes was the basis of determining the prevalence in this study. Hence this disparity could be reasonable.

Translation and transmission of the free-living stages of strongyle nematodes is successfully effected by favorable micro- and macro-climates, which in the state of West Bengal are quite prevalent in September. The highest prevalence of strongyle nematodes in September obviously substantiated this fact.

Coproculture examination revealed *Haemonchus contortus* (70.17%) as the predominant nematode species along with *Oesophagostomum* (15.08%), *Trichostrongylus* (9.75%), *Strongyloides* (2.75%) and hookworm (2.17%) infecting Garole sheep (Table 2). This finding was favourably comparable with the previous report in other breeds of sheep in India (Chaudhari 2004). However nematodes besides those recorded in other species of this study were also reported from parts of the country with different geo-climatic conditions (Jithendran, 1998, Nesreen 2005). Strongyle nematodes combined together, predominated by *H. contortus* (70.17%), constituted the maximum (75.41%) of the prevalence of GIP in Garole sheep as observed in this study. It could therefore be reasonably stated that Garole sheep in a random population were not resistant to *H. contortus* and this was in contrast to the earlier report in this regard (ACIAR Project Overview 2007).

Overall mean EPG of strongyle nematodes irrespective of the group was 422.37 with the highest intensity (EPG=950.26) in September. Overall mean EPG in Gr1, 2 and 3, irrespective of the month, were 128.00, 434.38 and 704.73, respectively, and interestingly, the highest EPG in all the 3 groups was also in September (Table 3). Therefore,

Table 3. Overall and group-wise intensity of strongyle group of nematode infection in Garole sheep

Months	Gr.1	Gr. 2	Gr. 3	Overall
June	114.7	450.82	720.28	354.81
July	143.45	479.31	860.32	380.47
August	123.05	543.39	850.49	570.62
September	161.94	623.73	1200.62	950.26
October	134.73	531.74	950.31	680.00
November	120.84	463.54	760.39	417.78
December	115.05	410.79	550.79	389.81
January	123.89	380.82	480.62	316.24
February	127.14	340.76	470.69	258.02
March	124.98	332.32	450.72	231.02
April	128.63	366.67	660.79	266.53
May	117.64	288.53	500.69	252.86
Overall	128.00	434.38	704.73	422.37

both prevalence and intensity (EPG) of strongyle nematode infection was highest in September. It was also evident from Tables 1 and 3 that the monsoon season (Aug-Oct) was most favourable for the translation and transmission of strongyle nematodes, which was in accordance with our previous findings (Jas and Ghosh 2007).

Significant decline in Hb concentration and PCV is generally considered as the major pathological effect of GIN with *H. contortus* as the predominant species (Jas *et al*. 2008). Nevertheless, Hb and PCV in all the three groups of sheep did not differ significantly during the entire tenure of this study (Table 4). As stated above Garole sheep failed to resist the establishment of the GIN, which was revealed by their moderate to high prevalence, and the EPG. This study therefore sufficiently indicated that Garole sheep generally has susceptibility to GIN that is comparable to other breeds. However, they have fairly high degree of resilience against the infection.

The common but yet to be established belief that Garole

Table 4. Haemoglobin concentration and packed cell volume in different groups of Garole sheep

Hb (g/dl)				PCV (%)				Mean faecal egg count (EPG)			
Occasions	Gr.1 (n = 6)	Gr. 2 (n = 6)	Gr. 3 (n = 6)	Occasions	Gr.1 (n = 6)	Gr. 2 (n = 6)	Gr. 3 (n = 6)	Occasions	Gr.1 (n = 6)	Gr. 2 (n = 6)	Gr. 3 (n = 6)
1st	9.46 ± 0.12	9.18 ± 0.09	9.16 ± 0.15	1st	33.75 ± 1.42	32.67 ± 0.57	32.83 ± 2.36	1st	138.32	450.35	760.64
2nd	9.23 ± 0.06	9.37 ± 0.05	9.23 ± 0.20	2nd	35.8 ± 0.69	33.17 ± 0.74	32.17 ± 2.15	2nd	142.64	564.78	800.36
3rd	9.18 ± 0.08	9.26 ± 0.09	9.09 ± 0.07	3rd	35.33 ± 0.92	32.17 ± 0.42	34.42 ± 2.38	3rd	179.45	620.39	950.29
4th	9.56 ± 0.10	9.43 ± 0.08	9.33 ± 0.18	4th	36.42 ± 0.99	33.50 ± 0.62	33.58 ± 2.08	4th	170.32	600.38	1000.46
								Overall	157.68	558.98	877.94

sheep are resistant/ less susceptible to parasitic infections has stimulated sporadic works in this regard. Lambs sired by Garole sheep were considerably more resistant to *H. contortus* than lambs of Deccani rams (Nimbkar 2003).

Interestingly, 18 sheep (Gr1) had consistently lowest EPG (mean = 128) compared to the mean EPG of all the sheep in this study, which was indicative of some degree of resistance in these sheep against the GIN infections. There are reports of existence of breed differences in sheep for resistance to *Haemonchus contortus* infection where higher packed cell volume, lower faecal egg count and worm burdens and higher body weight gains were considered as the criteria for resistance (Vanimisetti 2004a, b). Further studies involving experimental infection and genetic analyses of these selected sheep would throw light on the background factor(s) behind such persistently lower worm load (EPG).

ACKNOWLEDGEMENTS

The authors thankfully acknowledge the financial assistance of the Indian Council of Agricultural Research, New Delhi in conducting this study under the research project entitled 'All India Network Programme on Gastrointestinal Parasitism'.

REFERENCES

Anonymous 1971. *Manual of Veterinary Parasitological Laboratory Techniques*.Technical Bulletin No. 18, Her Majesty's Stationery Office, Ministry of Agriculture, Fisheries and Food, London, U K.

Chaudhari S S. 2004. Epidemiology of parasitic diseases in small ruminants. *Proceedinsg of XV National Congress of Veterinary Parasitology and National Symposium on 'Application of Molecular Biology in Parasitic Diseases for Rural Development'*. Pp.6. October, Pantnagar.

Jas R and Ghosh J D. 2007. Seasonal qualitative and quantitative variation in environment contamination with gastrointestinal nematodes of goats. *Environment and Ecology* **25S**(4): 1142–45.

Jas R, Datta S and Ghosh J D. 2008. Haemato-biochemical impact of gastrointestinal nematodosis in Bengal goat. *Journal of Veterinary Parasitology* **22** (1): 21–26.

Jas R and Ghosh J D. 2009. Economic impact of gastrointestinal nematodosis in sheep: enhanced meat production by anthelmintic treatment. *Indian Journal of Animal Sciences* **79**(8): 3–5.

Jithendran K P. 1998. Epidemiology of gastrointestinal nematodes in migratory sheep and goats in north-west humid Himalayan region. *Indian Journal of Animal Sciences* **68**(8): 894–96.

Khillare B S, Narladkar B W, Deshpande A R and Deshpande P D. 2006. Prevalence of coccidiosis in goats (*Capra hircus*) of Marthwada region. *Proceedings of XVII National Congress of Veterinary Parasitology and National Symposium on 'Strength, Challenges and Opportunities in Veterinary Parasitology'* November, Pp. 59. Puducherry.

Nesreen S, Jeelani S G and Hakeem M. 2005. Incidence of gastrointestinal nematodes in sheep in Kashmir Valley. *Journal of Veterinary Parasitology* **19**(1): 27–29.

Nimbkar C, Ghalasi P M, Swan A A, Walkden-Brown S W and Kahn L P. 2003. Evaluation of growth rates and resistance to nematodes of Deccani and Bannur lambs and their crosses with Garole. *Animal Science* **76**: 503–15.

Patra N C. 2002. 'Pathology of hepatic disorder in Garole sheep with special reference to LFT, histopathological and cytological study.' M.V.Sc. thesis, West Bengal University of Animal and Fishery Sciences, Kolkata, West Bengal.

Purohit K. 2002. 'Spontaneous amphistomiasis in garole sheep: Incedence, Haematobiochemical and pathological changes.' M.V.Sc. thesis, West Bengal University of Animal and Fishery Sciences, Kolkata, West Bengal.

Saravanan B C, Pandey B, Kataktalware M A, Bandyopadhyay S, Pourouchottamane R and Bora M. 2006. Studies on prevalence of parasitic infection in organized sheep farm at Arunachal Pradesh. *Proceedings of XVII National Congress of Veterinary Parasitology and National Symposium on 'Strength, Challenges and Opportunities in Veterinary Parasitology'*. November, Pp. 55 Puducherry.

Schalm O W, Jain N C and Carroll E J. 1986. *Veterinary Haematology.* 4th edn. Lea and Febiger, Philadelphia.

Sharma R C, Arora A L, Narula H K and Singh R N. 1999. Characteristics of Garole sheep in India.*AGRI* **26**: 56–64.

Singh H, Rai H S, Singh N K and Kaur A. 2005. Prevalence of helminthic infection in sheep in Ludhiana. *Journal of Veterinary Parasitology* **19**(2): 97–101.

Soulsby E J L. 1982. *Helminths, Arthropods and Protozoa of Domesticated Animals*. 7th edn. The English Language Book Society and Bailliere, Tindall, London.

Vanimisetti H B, Andrew S L, Zajac A M and Notter D R. 2004a. Inheritance of faecal egg count and packed cell volume and their relationship with production traits in sheep infected with *Haemonchus contortus. Journal of Animal Science* **82**: 1602–11.

Vanimisetti H B, Greiner S P, Zajac A M, Notter D R. 2004b. Performance of hair sheep composite breeds: Resistance of lambs to *Haemonchus contortus*. *Journal of Animal Science* **82**(2):595–604.