

COMPARATIVE EFFICACY OF NEEM LEAVES, PINEAPPLE LEAVES AND LEVAMISOLE AGAINST GASTROINTESTINAL NEMATODIASIS IN SHEEP

Ashis Chandra Shaha¹, Md. Anwar Hossian¹, Md. Siddiquil Islam¹, Md Rashedunnabi Akanda^{1*}, Md. Mahfujur Rahman², Mir Md. Iqbal Hasan³

¹ Department of Pharmacology and Toxicology, Sylhet Agricultural University, Bangladesh

² Department of Medicine, Sylhet Agricultural University, Bangladesh

³ Department of Physiology, Sylhet Agricultural University, Bangladesh

Corresponding author: - rashed.mvd@gmail.com

The study was conducted to evaluate the effects of neem leaves, pineapple leaves and levamisole HCl against gastrointestinal nematodiasis in sheep. A total 32 sheep were randomly selected on the basis of their egg count by McMaster method and divided into 4 equal groups. T₀ served as control group, whereas groups T₁, T₂ and T₃ were treated with neem leaves, pineapple leaves (10% water extract of leaves @ 100 ml/sheep) and levamisole HCl (Levavet[®] @ 8mg/kg body weight), respectively. A significant (p<0.01) reduction of EPG count was found in all treated groups. The body weight and total erythrocyte count (TEC) was significantly (p<0.01) increased on treated groups. On the other hand, total leukocyte count (TLC) was decreased significantly (p< 0.01). The hemoglobin (Hb) content was increased significantly (p<0.05) and packed cell volume (PCV) was increased significantly (p<0.01). We propose that 10% water extract (w/v) of both neem and pineapple leaves have some potential effects on gastrointestinal nematodiasis in sheep.

Key word: Levamisole HCl, neem leaves, nematodiasis, pineapple leaves, sheep

Among the small ruminants sheep are important in Bangladesh, providing meat and wool. During the last 12 years sheep population increased 2.5 times, with annual growth rate of 5% (BBS, 2008). Bangladesh is home to 2.7 million sheep (BBS, 2008), however the immense potential this huge population represents is yet to be realized due to multitude factors. In terms of meat and milk production sheep ranks third in position (Khan, 2000). The productivity of

this small ruminant is, however, hampered by several factors amongst, which is ectoparasitism (James-Rugu and Iwuala, 2000). Ectoparasites including ticks, lice, fleas and mange mites incriminated to cause a wide range of health problems as mechanical tissue damage, irritation, inflammation, hypersensitivity, abscesses, and lameness; and when present in large numbers may cause anemia and reduced productivity of small ruminants including sheep and goat (Fthenakis et al. 2001; Nyangiwe and Horak 2007; De Matos et al. 2009). Furthermore, ticks, lice, fleas, and mange mites are reported to cause preslaughter defects responsible for downgrading and rejection of goat skins. Heavy louse infestations may cause pruritus, alopecia, excoriation and self-wounding (Wall and Shearer, 1997).

Considering these grave consequences, the magnitude, epidemiology and control measure of ectoparasitic infestation in sheep has been have been partially documented in Bangladesh by a number of authors (Barmon et al. 2010, Hanif et al, 2005) but no one reported the effects of herbal drugs against ectoparasitism which would be cost-effective and reduce the parasitic load as well. Medicinal plants are one of the most important natural resources of a country. Bangladesh is also enriched by this resource. There are many native medicinal herbal plants which have potent anthelmintic action against both ecto and endoparasites (Mannan et al., 1997). Pharmacological action of these herbal plants may be studied experimentally, which have

proven worthy of therapeutic value. Pineapple leaf extract has therapeutic value as anti-inflammatory, anti-bacterial and anthelmintic agents (Amin et al., 2009).

Imported anthelmintic have long been considered the only effective way of controlling parasitic infestation. However, as these are very expensive and unavailable to farmers in rural areas, livestock producers are not interested to use these anthelmintic. For these reasons, interest in the screening of medicinal plants for their anthelmintic activity has remained of great scientific interest despite extensive use of synthetic chemicals in modern clinical practices all over the world (Akhtar et al., 2000). Plant anthelmintic has been in the forefront of this growing awareness (Hammond et al., 1997). A reason for this could be that they fall into the category of readily applicable elements of ethno veterinary medicine in livestock development (McCorkle and Mathias-Mundy, 1992). Studying herbal medicine can serve to validate and enhance existing local uses and can give clues to remedies with further potential. On the other hand, herbal medicine which are equally active but compatible to the economic status of our people as they are produced from the plants grown in our country by native technology very cheaply. Thus present study was undertaken with the objective to find out the effects of two commonly available leaves extract; neem leaves extract and pineapple leaves extract against ectoparasite infestation in goat.

MATERIALS AND METHODS

Study area and duration

The study was conducted on sheep which was taken care under the department of Pharmacology and Toxicology. Duration of the research work is May to June, 2014. The collected samples were examined in the Laboratory of Department of Pharmacology and Toxicology, Sylhet Agricultural University, Sylhet.

Study animals

A total of 32 sheep were selected randomly, the age of the sheep were 1 to 1.5 years. During collection of samples, the age, sex,

body condition/nutritional status, breed, and level of husbandry of sheep were recorded. Total 32 sheep were selected for this experiment having more or less similar age and randomly divided into four equal groups (T₀, T₁, T₂ and T₃) each group consisting of 8 sheep. Sheep of the group T₀ was kept an infected control group. Rest groups (T₁, T₂ and T₃) of sheep were treated with neem leaves extract, pineapple leaves extract and patent drug levamisole HCl (Levavet®), respectively.

Collection and preservation of faecal and blood samples

About 15-20gm of fresh feces was collected directly from rectum of the sheep. Fresh fecal samples were also collected from the top of the voided fecal mass on the ground when the animals were found in the act of defecation. 5-8ml blood samples were collected from jugular vein of sheep added anticoagulant (EDTA).

Extract preparation

After collection and bringing plants to the laboratory, all fresh leaves of plants were washed in running tap water and cut into small pieces. These small pieces of plant materials were grinded separately by pestle and mortar in the laboratory. After grinding little amount of distilled water was added to make paste of plant materials. A 10% water extract (w/v) of both neem and pineapple leaves were prepared from previously produced paste. For this 10gm of paste were added to 90 ml fresh drinking water in separate container and then made 100ml by adding additional water.

Administration of extracts and patent drug

A patent drug, levamisole HCl (Levavet®, Acme Ltd.) was used in this research work. The drug was used to compare the anthelmintic efficacy of different plant extracts in sheep. Group T₁ was treated with 10% water extract of neem leaves @ 100 ml/sheep administered orally for single dose. Group T₂ was treated with 10% water extract of pineapple leaves @ 100ml/sheep administered orally for single dose. Group T₃ was treated with patent drug levamisole HCl (Levavet® @ 8mg/kg body weight administered orally for single dose.

Clinical and para-clinical examinations

The fecal sample from all groups were examined by McMaster egg counting technique as described by Soulsby (1986) on pre-treatment (day 0) and at 7th, 14th, 21st and 28th day of post-treatment. Eggs per gram (EPG) of fecal sample were recorded. The body weight of each sheep was measured by using digital weight machine. Total erythrocytes count (TEC), total leucocytes count (TLC), hemoglobin (Hb) and packed cell volume (PCV) was done by following method described by Lamberg and Rothstein (1977).

Statistical analysis

The data obtained from this research work were entered in Microsoft Excel worksheet, organized and processed for analysis. Mean, standard deviation (SD) and level of significant were estimated with the help of the statistical package programme (SAS) software, version 9.4 (SAS, 1998).

RESULTS AND DISCUSSION

EPG count were found significantly decreased ($p < 0.01$) in treated group and significantly ($p < 0.01$) increased in control

group during the whole experimental period. Mean EPG count was increased up to 15.21% on 28th day (table 1). Reduction of EPG on 28th day after treatment with levamisole HCl was 67.27%. More or less similar results were also described by Godara et al. (2011) and Byaruhanga and Okwee-Acai (2013). These results indicate that the activities of this drug were constant. Although the efficacy was gradually decreased from days 7 to 28, the level of decreased was negligible. Reduction of EPG on 28th day after treatment with neem was 44.00%. Similar findings also observed by Ahmed et al. (1994) and Arunachal et al. (2002) in sheep. These findings support the earlier works of Rahman (2002) in goat. Adlibitum feeding of fresh neem leaves produced 82% reduction in worm eggs of animals (Chandrawathani et al. 2000) and a further trial on a limited number of sheep showed that neem produced a significant reduction in worm burdens (Chandrawathani et al. 2002). These results suggest that neem extracts may be useful in the control of gastro-intestinal nematodes of small ruminants. Reduction of EPG on 28th day

Table 1: Effects of neem leaves, pineapple leaves and levamisole on EPG of gastrointestinal nematodiasis in goat

Group	Treatment	Pre-treatment	Post-treatment							
		'0' day	7 th day		14 th day		21 st day		28 th day	
		EPG Mean \pm SD	EPG Mean \pm SD	EPG Increase/Decrease (%)	EPG Mean \pm SD	EPG Increase/Decrease (%)	EPG Mean \pm SD	EPG Increase/Decrease (%)	EPG Mean \pm SD	EPG Increase/Decrease (%)
T0	Control	337.27 \pm 60.23	344.60 \pm 49.18*	2.17 (+)	351.93 \pm 41.79*	4.34 (+)	366.60 \pm 25.92*	8.69 (+)	388.59 \pm 41.79**	15.21 (+)
T1	Neem	366.60 \pm 57.96	183.30 \pm 57.96*	50.08 (-)	190.63 \pm 47.79*	48.00 (-)	197.96 \pm 41.79*	46.00 (-)	205.29 \pm 55.59**	44.0 (-)
T2	Pineapple	329.94 \pm 44.89	197.96 \pm 61.34*	40.00 (-)	205.29 \pm 49.18*	37.77 (-)	212.62 \pm 47.79*	35.55 (-)	227.29 \pm 54.37**	31.11 (-)
T3	Levamisole	403.26 \pm 68.58	109.98 \pm 68.58*	72.72 (-)	117.31 \pm 60.23*	70.90 (-)	124.64 \pm 61.34*	69.09 (-)	131.97 \pm 66.59**	67.27 (-)

** = Significant at 1 per cent level ($P < 0.01$), (+) = increase, (-) = decrease

Table 2: Effects of neem leaves, pineapple leaves and levamisole on body weight of gastro-intestinal nematodiasis in sheep

Group	Treatment	Pre-treatment	Post-treatment			
		'0' day	7 th day	14 th day	21 st day	28 th day
T0	Control	15.23±0.44	15.08±0.41**	14.97±0.44*	14.88±0.42*	14.91±0.54**
T1	Neem	14.96±0.79	15.10±0.77**	15.28±0.72*	15.44±0.70*	15.38±0.63**
T2	Pineapple	15.22±0.68	15.43±0.73**	15.54±0.71*	15.66±0.71*	15.59±0.59**
T3	Levamisole	15.31±0.84	15.52±0.81**	15.64±0.78*	15.73±0.76*	15.78±0.64**

** = Significant at 1 per cent level (P<0.01)

* = Significant at 5 per cent level (P<0.05)

Table 3: Effects of neem leaves, pineapple leaves and levamisole on TEC (million/cu.mm) of gastro-intestinal nematodiasis in sheep

Group	Treatment	Pre-treatment	Post-treatment			
		'0' day	7 th day	14 th day	21 st day	28 th day
T0	Control	8.34±0.35	8.03±0.27*	7.88±0.26**	7.79±0.28**	7.55±0.27*
T1	Neem	8.16±0.31	8.78±0.17*	8.70±0.18**	8.62±0.18**	8.54±0.25*
T2	Pineapple	8.27±0.23	8.64±0.25*	8.56±0.21**	8.49±0.22**	8.47±0.28*
T3	Levamisole	8.05±0.33	8.85±0.26*	8.74±0.21**	8.66±0.33**	8.64±0.19*

** = Significant at 1 per cent level (P<0.01)

* = Significant at 5 per cent level (P<0.05)

after treatment with pineapple was 31.11%. Pineapple had also moderate (40.00%) anthelmintic activity. Similar findings were also reported by Mostofa et al. (1983) and Khatun et al. (1995).

The body weight was increased significantly (p<0.01 and p<0.05) after neem, pineapple and levamisole treatment

in group. On the other hand, body weight was decreased significantly (p<0.01 and p<0.05) in controlled group (table 2). These consequences were agreeable with the results of Ahmed *et al.* (1994) for neem of sheep. These results were also agreeable with the findings of Khatun *et al.* (1995) for pineapple of goat. Similarly, Amin *et al.*

Table 4: Effects of neem leaves, pineapple leaves and levamisole on TLC (thousand/cu.mm) of gastro-intestinal nematodiasis in sheep

Group	Treatment	Pre-treatment	Post-treatment			
		'0' day	7 th day	14 th day	21 st day	28 th day
T0	Control	7.97±0.18	8.13±0.14	8.17±0.14*	8.27±0.11**	8.37±0.10
T1	Neem	8.29±0.21	7.74±0.09	7.81±0.06*	7.83±0.06**	7.86±0.06
T2	Pineapple	8.45±0.23	7.87±0.15	7.90±0.15*	7.94±0.16**	7.99±0.16
T3	Levamisole	8.10±0.32	7.63±0.22	7.66±0.21*	7.68±0.22**	7.70±0.21

** = Significant at 1 per cent level (P<0.01)

* = Significant at 5 per cent level (P<0.05)

Table 5: Effects of neem leaves, pineapple leaves and levamisole on Hb (gm %) of gastro-intestinal nematodiasis in sheep

Group	Treatment	Pre-treatment	Post-treatment			
		'0' day	7 th day	14 th day	21 st day	28 th day
T0	Control	7.84±0.26	7.60±0.31	7.36±0.16*	7.28±0.10	7.16±0.16*
T1	Neem	7.32±0.30	7.56±0.38	7.88±0.33*	7.72±0.41	7.56±0.16*
T2	Pineapple	7.68±0.22	7.96±0.32	8.00±0.37*	7.76±0.32	7.68±0.22*
T3	Levamisole	7.60±0.20	7.88±0.30	7.92±0.43*	7.64±0.35	7.64±0.26*

* = Significant at 5 per cent level (P<0.05)

(2008) reported that body weight was increased significantly in neem treated cattle and decreased in untreated cattle. Hossain *et al.* (1996) also observed neem leaves and neem seed kernels increased body weight of cattle. Khalid *et al.* (2005) the body weight was increased significantly in neem and pineapple treated sheep. The parasitic infestation might be responsible for the stunted growth. The body weight was increased may be due to removal of parasitic load might have had facilitate the weight regain through proper digestion,

absorption and metabolism of feed nutrient in the parasite free gastrointestinal tract.

TEC was increased significantly (p<0.01 and p<0.05) after treatment with neem leaves, pineapple leaves and levamisole HCl at onward day in post-treated sheep. Conversely, TEC was decreased significantly (p<0.01 and p<0.05) up to the last day of experimental period in control group (table 3). Rob, (2004) stated that water extracts of neem leaves increased TEC in sheep on 28 day post-treatment. Likewise,

Table 6: Effects of neem leaves, pineapple leaves and levamisole on PCV (%) of gastrointestinal nematodiasis in sheep

Group	Treatment	Pre-treatment	Post-treatment			
		'0' day	7 th day	14 th day	21 st day	28 th day
T0	Control	31.00±2.82	28.60±2.70*	27.80±2.16**	27.20±1.64**	26.60±1.14**
T1	Neem	27.80±2.77	29.60±2.30*	31.40±1.34**	30.00±1.22**	29.60±1.14**
T2	Pineapple	28.40±2.07	30.20±1.92*	31.00±1.22**	30.20±1.09**	30.00±1.00**
T3	Levamisole	29.00±2.91	30.00±2.44*	31.80±1.30**	30.60±1.14**	30.40±0.54**

** = Significant at 1 per cent level (P<0.01)

Rahman (2002) observed water extract of neem leaves increased TEC on 21st day of post-treatment in goat. Similarly, Amin et al. (2008) reported that neem (10% water extract of leaves) increased TEC in cattle on 28 day post-treatment. Likewise, Khatun et al. (1995) reported that pineapple elevated TEC in goat. The results of the effect of neem leaves, pineapple leaves and levamisole HCl on TLC in sheep (table 4). TLC was decreased significantly (p<0.01 and p<0.05) at 7th, 14th, 21st and 28th day post-treatment. Conversely, TLC was increased significantly (p<0.01 and p<0.05) in control group. The result of the present study is similar to the report of Rahman (2002). More or less similar findings also observed by Khalid et al. (2004) in sheep. The increase in total leucocytes count (TLC) is attributable to the animals' immune response to infection or sensitization. Hb was increased significantly (p<0.01 and p<0.05) at 7th, 14th, 21st and 28th day post-treatment in sheep and decreased significantly (p<0.01 and p<0.05) in control group. Rob, (2004) stated that water extracts of neem leaves increased Hb in sheep on 28 day post-treatment. Likewise, Rahman (2002) observed water extract of neem leaves increased Hb on 21st day of post-treatment in goat. Similarly, Amin et al. (2008) reported that neem (10% water extract of leaves) increased Hb in cattle on 28 day post-treatment. Likewise, Khatun et

al. (1995) reported that pineapple increased Hb content in goat which supported the present findings. Hossain et al. (1996) also reported that neem leaves increased Hb content in cattle. The increase in Hb may be due to the increase of TEC. PCV was increased significantly (p<0.01 and p<0.05) at 7th, 14th, 21st and 28th day post-treatment in sheep and reduced significantly (p<0.01 and p<0.05) in control group. Rob, (2004) stated that water extracts of neem leaves increased PCV in sheep on 28 day post-treatment. Likewise, Rahman (2002) observed water extract of neem leaves increased PCV on 21st day of post-treatment in goat. Similarly, Amin et al. (2008) reported that neem (10% water extract of leaves) increased PCV in cattle on 28 day post-treatment. Hematological changes in all treated groups as observed over four weeks treatment period did not show any adverse or suppressive effects when compared with control group. Interestingly, plants extracts including patent drug rather have shown to improve hematological profiles. However, reports on the improvement of hematological values, body conditions and body weight gains due to modern anthelmintic treatment of gastrointestinal nematodes infected sheep. Levamisole HCl was more effective drug than neem and pineapple extract against gastrointestinal nematodiasis in sheep. On the other hand relatively neem extract was more effective

than pineapple extract against gastrointestinal nematodiasis. Therefore from the reflection of this study, it is obvious that plant materials have potential effects on gastrointestinal nematodes in sheep.

CONCLUSION

Watery extracts of neem and pineapple leaves have potential effect and may be used as an alternative drugs against gastrointestinal nematodiasis in sheep. The present study is a preliminary work and confirms the folkloric claims regarding the anthelmintic efficacy of neem and pineapple leaves against intestinal helminthes in sheep. Further studies will however be necessary to reveal the species specific effects of the extracts and largely use in veterinary field.

REFERENCES

1. Ahmed, N.U., Mostofa M., Awal M.A. & Alam M.N. (1994). Comparative efficacy of modern anthelmintics with that of neem seeds against gastro-intestinal nematodiasis in sheep. *Bangladesh Veterinary Journal*, 28, 21-23.
2. Akhtar, M.S., Iqbal, Z., Khan, M.N. & Lateef, M. (2000). Anthelmintic activity of medicinal plants with particular reference to their use in animals in the Indo-Pakistan subcontinent. *Small Ruminant Research*, 38, 99-107.
3. Akter M.K., Aziz, F.B., Islam, M.N., Rashid, M.B. & Hasan, M. (2015). Studies on prevalence of ascariasis in indigenous chickens in Gaibandha district and treatment by pineapple leaves extract. *International Journal of Natural and Social Sciences*, 2(2), 37-42.
4. Amin, M.R., Mostofa, M., Hoque, M.E. & Sayed, M.A. (2009). In vitro anthelmintic efficacy of some indigenous medicinal plants against gastrointestinal nematodes of cattle. *Journal of Bangladesh Agricultural University*, 7(1), 57-61.
5. Amin, M.R., Mostofa, M., Awal, M.A. & Sultana, M.A. (2008). Effects of Garlic, Turmeric and Betel Leaf against Gastrointestinal Nematodes in Cattle. *Bangladesh Journal of Veterinary Medicine*, 6(1), 115-119.
6. Arunachal, P.K., Karunanithi, K. & Narendrababu R. (2002). Comparative study on anthelmintic efficacy of neem products and praziquantel in sheep. *Indian Journal of Small Ruminant*, 8, 131-132.
7. BBS (2008). Bangladesh Bureau of Statistics. Planning Division, Ministry of Planning. Government of the People's Republic of Bangladesh, Dhaka, Bangladesh.
8. Byaruhanga, C. & Acai J.O. (2013). Efficacy of albendazole, levamisole and ivermectin against gastrointestinal nematodes in naturally infected goats at the National Semi-arid Resources Research Institute, Serere, Uganda. *Veterinary Parasitology*, 195(1-2), 183-186.
9. Chandrawathani, P., Adnan, M. & Zaini C.M. (2000). Preliminary studies on Neem (*Azadirachta indica*) as an alternative anthelmintic for sheep. *Proc. 12th Vet. Assoc. Malaysia Scientific Congress*, Malaysia.
10. Chandrawathani, P., Berlin, D., Fasahah, S.N., Adnan, M., Jamnah, O., Soni, A., Hoagland, J. & Waller, P.J. (2002). Evaluation of the Neem tree (*Azadirachta indica*) as a herbal anthelmintic for nematode parasite to control in small ruminants in Malaysia. *Tropical Biomedicine*, 19(1&2), 41-42.
11. De Matos, M., Siteo, C., Neves, L., Nöthling, J.O., Horak, I.G. (2009). The comparative prevalence of five ixodid tick species infesting cattle and goats in Maputo Province, Mozambique. *Onderstepoort Journal of Veterinary Research*, 76, 201 - 208.
12. Fthenakis, G.C., Karagiannidis, A., Alexopoulos, C., Brozos, C. & Papadopoulos, E. (2001) Effects of sarcoptic mange on the reproductive performance of ewes

- and transmission of *Sarcoptes scabiei* to newborn lambs. *Veterinary Parasitology*, 95, 63 – 71.
13. Godara, R., Sharma R.L., & Sodhi, S.S. (2011). Efficacy of fenbendazole, levamisole and ivermectin against gastrointestinal nematodes in Jamunapari goats. *Journal of Parasitic Diseases*, 35(2), 219–221.
 14. Hammond, J.A., Fielding D. & Bishop S.C. (1997). Prospects for plant anthelmintics in tropical veterinary medicine. *Veterinary Research Communications*, 21, 13–28.
 15. Hossain, S.A., Mostofa, M., Alam, M.N., Awal, M.A. & Ahmed, N.U. (1996). Comparative efficacy of modern anthelmintics and Neem (leaves and seeds) in the treatment of bovine nematodiasis. *Progressive Agriculture, Bangladesh*, 7 (1), 29–33.
 16. James-Rugu, N. N. & Iwuala, M. O. E. (2000). Ectoparasites of some domestic animals on the Jos Plateau, Nigeria. *ScienceForum*, 5, 146 – 156.
 17. Khalid S.M.A., Amin, M.R., Mostofa, M., Choudhury, M.E. & Uddin, B. (2005). Effects of Indigenous Medicinal Plants (Neem and Pineapple) against Gastro-intestinal Nematodiasis in Sheep. *International Journal of Pharmacology*, 1(2), 185-189.
 18. Khan, F. H. (2000). *Geology of Bangladesh*, The University Press Limited, Red Crescent Building, 114, Motijheel Commercial Area, Dhaka-1000, Bangladesh.
 19. Khalid, S.M.A., Amin, M.R., Mostofa, M., Hossain, M.J. & Azad M.A.K. (2004). Effects of Vermic® against Gastro-intestinal Nematodiasis in sheep. *Journal of Biological Sciences*, 4(6), 720-724.
 20. Khatun, M., Awal, M.A., Mostofa, M. & Rashid, M.S.H. (1995). Comparative efficacy of Pineapple leaves with Fenbendazole against gastro-intestinal nematodes in goats. *Bangladesh Veterinary Journal, Bangladesh*, 29, 75-78.
 21. Lamberg, S.L. & Rothstein R. (1977). *Laboratory Manual of Hematology and Urinalysis*. Avi. Publishing Company, Inc, West Port Connecticut, U.S.S.R.
 22. Mannan, MA., Rafiq, K., Mostofa, M. & Hason, Q. (1997). Comparative efficacy of Ivomec pour on, Neguvon Ointment and Neem-tobacco herbal preparation against naturally occurring hampsores Lesion in cattle. *Bangladesh Veterinary Journal*, 31 (3-4), 119-122.
 23. McCorkle, C.M. & Mundy E.M. (1992). Ethnoveterinary medicine in Africa. *Africa International African Institute*, 62: 59–93.
 24. Mostofa, M., Hasan, Q. & Sobhan, M.A. (1983). Efficacy of indigenous medicinal plants against gastro-intestinal nematodiasis in cattle and their comparative activities with that of Nemafox. *Bangladesh Veterinary Journal, Bangladesh*, 17(1-4): 1-4.
 25. Nyangiwe, N. & Horak, I.G. (2007) Goats as alternative hosts of cattle ticks. Onderstepoort. *Journal of Veterinary Research*, 74, 1– 7.
 26. Rahman, M.H. & Mondal, M.M.H. (1985). Tick Fauna of Bangladesh. *Journal of Parasitology*, 9(2), 145-49.
 27. Rahman, M. (2002). In vitro and in vivo anthelmintic effects of some plants against gastro-intestinal nematodes of goats. M.S. Thesis, Bangladesh Agricultural University, Mymensingh, Bangladesh.
 28. Rob, S. (2004). Efficacy of albendazole (Endokil) and neem against gastrointestinal nematodiasis in sheep. MS Thesis, Department of Pharmacology, Faculty of Veterinary Science, BAU, Mymensingh.
 29. Samad, M.A. (2000). An overview of livestock research reports

- published during the twentieth century in Bangladesh. Bangladesh Veterinary Journal, 34:53-149.
30. SAS (1998). SAS User's guide. SAS Institute Inc., Cary, North Carolina, USA.
31. Soulsby, E.J.L. (1986). Helminth, Arthropod and Protozoa of Domesticated Animals. 7th Edn. Bailliere and Tindall, London, 763-766.
32. Wall, R. & Shearer, D. (1997). Veterinary Entomology, 1st edition, Chapman and Hall. London, UK. P.265 and 290