# Effect of replacement concentrate by hybrid *Azadirachta* indica (Neem) leaves on growth performance and anthelminthic properties of indigenous sheep.



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# **Abstract**

This study was conducted to evaluate the effect of hybrid *Azadirachta indica* (neem) leaves as a substitute to concentrate on growth performance and anthelmintic characteristics of indigenous sheep. After completing the quarantine period twelve indigenous male sheep with an initial mean body weight of 12 ±1.0 kg (means ± SD) were allotted through Completely Randomized Design to 4 treatments and 3 replicates. The supplement feed was offered at 300 g DM/ (animal•d). The experimental treatments were 300gm concentrate mixture (CM), 250gm CM + 50gm neem leaf meal (NLM), 200gm CM + 100gm NLM, 150gm CM + 150gm NLM supplement. The live weight of sheep was determined by digital weighing balance weekly up to 8 weeks. Fecal egg count was conducted by McMaster egg count technique by using 3gm feces sample weekly 2 times up to 8 weeks. There was found higher average daily body weight gain & the FCR value in neem fed group compared to the control group. Besides, there was found drastic reduction in fecal egg count on T4 treatment that was fed higher neem leaf compare to other treatments.

**Keywords:** Neem leaves, Growth performance, Anthelmintic, Indigenous sheep.

# **CHAPTER 1: INTRODUCTION**

Livestock is a significant aspect of a country's wealth since it supplies meat, milk, and manure to the great majority of its citizens in addition to draft power and leather. The economy of a country largely depends on livestock sectors. As the livestock number is increasing, the economy of a country is boosted.

Bangladesh is a developing country. In Bangladesh livestock are generally cattle, buffalo, goats, and sheep. Livestock production has been an important part of Bangladesh's agriculture for many years. Many rural people in Bangladesh largely depend on livestock animals for their livelihood (Rahman, 1989).

Nowadays, sheep production drew the attention of people, and it has since grown popular among livestock producers, particularly small-scale livestock farmers in Bangladesh. The population of sheep (2019-20) is 36.07 lakh & in (2020-21) the number is 36.79 (DLS 2021). As the Govt. of Bangladesh notice the importance and potentiality of sheep production, they have granted the second phase of the developmental project 'Conservation and Improvement of Native Sheep through Community Farming and Commercial Farming' (CINSCFCF) (Islam et al.,2016). Although the livestock rearing tendency to people is increasing day by day but parasitic illnesses continue to be a serious barrier to livestock production. Infections with worms are the most common causes of sluggish development, poor reproductive function, and mortality. Several sheep health issues remain a big danger to Bangladesh's effective sheep production. Infectious illnesses and parasitism are severe hazards to the successful sheep business in Bangladesh. Loss of production, reduced fertility, worse feed conversion efficiency, greater production costs, increased risk of zoonotic illnesses, and public health concerns are all consequences of the diseases.

The climatic condition of Bangladesh is helpful for the optimal ecological survival of most of the parasites and the intermediate hosts. Gastrointestinal nematodes such as Haemonchus spp., Trichostrongylus spp., Cooperia spp., Oesophagostomum spp., Trichuris spp., and Strongyloides spp. are the most frequent parasitic infections in Bangladesh and these nematodes cause both health deterioration of animals and economical loss of farmers (Amin *et al.*, 2008). To reduce the spread of worm infections in animals, pharmaceutical industries have released several anthelminthic drugs and the excessive use of these drugs has led to the development of helminth resistance (Sarker *et al.*, 2016). In addition to toxicity problems of these drug residues (Amin *et al.*, 2008). This is shocking news for the livestock farmers.

A higher variety of plants with particular or wide spectrum anthelmintic activity are naturally accessible. This is believed that the neem plant is one of the most widely studied tree species in the world due to its wide range of uses (Girish and Shankara 2008). The neem tree (Azadirachta indica) is a non-leguminous multi-purpose tree that belongs to the Meliaceae family. Most of the studies on neem in animal production are especially focused on its medicinal uses mostly as an anthelminthic agent (Adjorlolo *et al.*, 2016). In Bangladesh lack of green grass is a major constraint for livestock farmers. So the farmers largely depend on commercial concentrate feed for the production & reproduction performance of livestock.

In recent times the price of commercial concentrated feed is increasing day by day. In this incident, a farmer is unable to meet the daily requirements of concentrated feed for their animals. So, the animals lost both production & reproduction performance.

Evidence suggests that protein supplements are available at a very high price in developing countries so this allowed the use of non-protein nitrogen sources to improve the nitrogen deficiency in fibrous feeds, thus enhancing their digestibility, intake and nutrient availability through optimization of rumen fermentation (Makkar *et al.*, 2007). The leaves of *Leucaena leucocephala* and *Azadirachta indica* are potential nitrogen supplements. These tree forages not only provide nitrogen, energy, and micro-nutrients but have also many other advantages like easy accessibility to farmers, their laxative influence on the alimentary system, low degradability of nitrogen in the rumen and provision of variety in the diet (Dida *et al.*, 2019).

- The present study was designed with the following objectives:
- i) To introduce *Azadirachta indica* (Neem) leaf as a protein supplements and anthelmintic of small ruminants.
- ii) To investigate the optimum proportion of *Azadirachta indica* (Neem) leaf and concentrate mixture for maximum weight gain of small ruminants.

# **CHAPTER 2: REVIEW OF LITERATURE**

# 2.1 Chemical composition of neem leaves

The neem plant is one of the most prominent medical plants and it has been used for a variety of therapeutic reasons since ancient times. Many physiologically active substances were found in the extracted chemical contents of various portions of the neem tree, including triterpenoids, alkaloids, phenolic compounds, flavonoids, carotenoids, ketones, and steroids. It is utilized in many traditional treatments due to presence of biologically active components. Such as Nimbidin, Nimbin, Nimbolide, Gedunin, Azadirachtin, Mahmoodin, Cyclic trisulphide, and other chemical constituents are used as antipyretic, anti-inflammatory, antibacterial, antigastric, ulcer, antiarthritic, spermicidal antifungal, antimalarial, hypoglycemic, immunomodulatory, diuretic, and antitumor agents (Eid *et al.*, 2017). As far as having many other potentialities of neem further research is going on to explore it.

# 2.2 The nutrient profile of neem leaves

Neem (*Azadirachta indica*) possess a vast array of biologically active compounds which are structurally complex and chemically diverse (Koul *et al.*, 2003). Generally in neem leaves the crude protein (CP) is high (Adjorlolo *et al.*, 2016). The concentrations of crude protein between 17.5% and 18.7% have been reported (Bais *et al.*, 2002). Some authors reported the value of crude protein is higher or lower than this. For example, CP content of neem leaves 9.7% according to (Ramana *et al.*, 2000) on the other hand the CP value of neem leaves 20.9% reported by (Ogbuewu *et al.*, 2011). Varietal differences in the neem plant may be a cause of variation in crude protein levels.

According to available reports, it is indicated that the neem leaves have low fiber content. It has been reported that the neutral detergent fiber (NDF) & acid detergent fiber (ADF) were at the level of 38.0% and 27% respectively (Ramana *et al.*, 2000). The value of crude fiber (CF) of 11.3% & nitrogen-free extract (NFE) level of 53.9% was reported by (Bhowmik *et al.*, 2008). There are few reports on the mineral content of neem leaves.

Some available data on minerals which published are summarized in the table

Table 1: Mineral contents of neem leaves

Macro	o-mineral	s (% DM)	Micro minerals (ppm in DM)				References		
Ca	P	Mg	Cu	Fe	Mn	Zn	Co	Cr	
1.48	0.11	1.26	5.24	-	30.4	47.7	-	-	Bhowmik et al., 2008.
0.71	0.28	0.75	34.0	745	60.0	18.0	10.0	0.80	Ansari et al.,2012
1.53	0.25	-	8.90	566	23.5	-	-	-	Niranjan et al., 2008.

# 2.3 Anti-nutritional factors of Neem leaves

Some anti-nutritional factors such as triterpinoid derivatives (Azadirachtin, nimbidin) (Dida *et al.*, 2019) tannins, phenolic compounds and oxalates have been identified in neem leaves and this limit their use as animal fodder (Adjorlolo *et al.*, 2016). Tanin concentrations in neem leaves is lower than ipil ipil which will depress feed intake (Niranjan *et al.*, 2008).

Table 2: Some anti-nutritional compounds identified in Neem leaves

Anti-nutritional factors	Concentration (%)	Source
Condensed tannins	9.38	Ramana et al 2000
	11.4	Ngamsaeng et al 2006
Crude saponins	2.80	Ngamsaeng et al 2006
Oxalate	0.63	Niranjan et al 2008
Lignin	10.2	Ramana et al 2000
Azadirachtin	0.024	Radhakrishnan <i>et al</i> 2007
	0.002	Ghimeray et al 2009
Total phenolics	6.53	Ramana et al 2000

There are not found available information about the effects of anti-nutritional factors of neem leaves in ruminants. Some reports suggest that there is not found any adverse effect of neem as feed on ruminants, and poultry even in monogastric (Adjorlolo *et al.*, 2016).

# 2.4 Acceptability of neem leaves by a livestock farmer

The production of neem biomass is about 0.35 tons per year per mature neem tree (Panhwar, 2005) & 5 to 50 tons /ha (Girish & Sankara, 2008). So neem leaves would be a source of potential feed for small ruminant holder producers.

However, there is a widely held belief that ruminants do not accept neem leaves because of its bitter taste. The presence of triterpenoids particularly azadirachtin in neem is responsible for bitter taste. However, ruminants can tolerate bitter taste as having ability to detoxify secondary plant compounds through alleolochemical-type reactions that take place within them (Adjorlolo *et al.*, 2016). But Neem leaves are reported palatable to sheep (Chandrawathani *et al.*, 2006) and also to goats (Seresinhe and Marapana 2011). In this study, it was observed that as the sheep were hungry and the neem leaves powder when mixed with the concentrate mixture then it was easily accepted by the sheep.

#### 2.5 Anthelmintic properties of Neem leaves

Natural products like Neem plays a vital role in the pharmaceutical industry. Therefore 50% of modern clinical drugs are made from natural ingredients (Kingston *et al.*, 2011). The leaves, seeds, bark, roots, fruits, and oil of neem have been used medicinally to treat various diseases, particularly in Indian Ayurvedic medicine, Homeopathic medicine (Tiwary *et al.*, 2014).

The most important gastro-intestinal helminth parasites belong to three different classes, trematode, and cestodes (flatworm), nematode (roundworm). Small ruminants are severely harmed by GI nematodes, some nematodes are bloodsuckers and induce anemia, while many others impact the body's physiological, metabolic, and immunological systems, leading to major financial losses in the production of meat, milk, and wool as well as in reproduction (Dehuri *et al.*, 2021).

The in vitro activity of methanolic extract of neem against *H.contortus* larva indicated a 40% mortality at a concentration of 4 mg/ml (Rahman *et al.*, 2011). The aqueous extracts of neem leaves and bark inhibited nematode egg hatch (Yakubu *et al.*, 2006).

Neem leaves effective against strongyle nematode after day 7 post treatment (jamra *et al.*, 2015). Neem seeds and leaves have been tested against gastro-intestinal nematodes of ruminants in Bangladesh (Rob *et al.*, 2004, Khalid *et al.*, 2005). The presence of active substances in neem leaves showed parasiticide activities against different worms (Amin *et al.*,

2008). As most of the research shows that the neem leaves mainly effective against nematodes but the potentiality of neem leaves as natural anthelmintic should be admitted.

# 2.6 Effect of Neem leaves on growth and production of Animals

The proteins that are derived from the plant source are termed plant proteins. It is the cheapest and most abundant source of protein.

Neem leaf is one of the sources of plant protein that can be used in animal feed. Feed costs are reduced and net return is increased when solitary neem leaf is added in place of commercial concentrate mix (Patil *et al.*, 2021). Neem leaf & Pigeon Pea mixture at different levels replaced the highly prized commercial concentrate feeds without impairing the growth and productive performance of goats (Dida *et al.*, 2019). In ruminant diets, neem leaves may replace 50% of soya bean meals without hampering the feed intake, dry matter and fiber digestibility as well as body weight gain (Paengkoum *et al.*, 2010). Neem leaf feeding improved ruminant performance may be due to the effects of the bioactive chemicals in the leaves effects on intestinal worms. In another study, 30% of mustard straw was replaced by neem leaves as intakes of dry matter and crude protein were raised to comparable amounts, and synthesis of volatile fatty acids also increased at the same time. A sole diet of neem leaves is offered to goats and founded a high voluntary intake of 3.12% of body weight (Bais *et al.*, 2002).

So in conclusion, Neem leaves may be fed as a supplement to animals to boost feed intake and diet quality.

# **CHAPTER 3: MATERIALS & METHODS**

# 3.1 Study Area & Period

The experiment was conducted at Chittagong Veterinary and Animal Sciences University from April –June 2022.

# 3.2 Preparation of Neem leaf meal

#### 3.2.1 Collection of Neem leaves

Hybrid neem leaves have been harvested mostly from 1 to 3 years old trees. *Azadirachta indica* were collected from different locations of Chittagong. This selection point was based on the activities being carried out within and around the sampling point.

Samples collected were wrapped in a black polythene bag and properly labeled before transporting to the laboratory for further analysis.



Figure 1: Neem leaves



Figure 2: Neem foliage

#### 3.2.2 Grinding & Storage of leaves

After collection of leaf, the fresh samples were dried in sunlight and then ground into powder using a laboratory grinder to reduce particle size and then kept in plastic bags for Laboratory analysis and further use as Animal feed in the experimental diet.



Figure 3: Ground leaves Figure



4: Ground leaves foliage

# 3.2.3 Chemical analysis of Neem leaves

The proximate analysis of the samples (neem Leaves) for moisture, total ash, crude fiber, crude protein was carried out in triplicate using methods described by AOAC (AOAC, 2005).

# 3.3 Experimental Design

The study was designed in Completely Randomized Design (CRD). The sheep were randomly allocated to four dietary treatments with three replications. Here  $T_1$  was the control group of this study.

Table 3: Treatment arrangement

Treatments	Rhodes	Feed offered				
grass hay		Concentrate Mixture (Wheat bran, Rice polish, Soybean meal or till oil cake) (gm/day)	Neem Leaf Meal (gm/day)	Total Supplement(gm/day)		
T <sub>1</sub> (control)	Ad libitum	300	-	300		
T <sub>2</sub>	Ad libitum	250	50	300		
T <sub>3</sub>	Ad libitum	200	100	300		
T <sub>4</sub>	Ad libitum	150	150	300		

# 3.4 Preparation of Shed

The Sheep & Goat shed of Chattogram Veterinary and Animal Sciences University was used for this study. The shed was thoroughly cleaned and disinfected before arrival of the experimental sheep.

# 3.5 Collection of experimental Sheep

A total of 12 indigenous sheep about 6-month-old were bought from the CDA market in Chattogram. The average live body weight of those sheep was  $12 \pm 1.0$  kg (means  $\pm$  SD). The animals were quarantined for seven days to get used to their new environment and to observe their health condition. During this quarantine period the sheep was offered grass and small amount of concentrate feed. At the end of the quarantine period, the sheep were randomly allocated to four dietary treatments.

# 3.6 Management of experimental sheep

#### 3.6.1 Feeding & Watering

The supplement feed was offered at 300 g DM/ (animal•d). Before the experiment started, samples of supplement ingredients and Rhodes grass were analyzed for the chemical composition of DM content. Based on the chemical composition the supplement ratios were formulated. The experiment dietary treatment will be as follows (in % of DM offered). The experimental treatments were 300gm concentrate mixture (CM), 250gm CM + 50gm neem leaf meal (NLM), 200gm CM + 100gm NLM, 150gm CM + 150gm NLM supplement. The experimental feed was offered daily once at noon. The animals were allowed to graze from 10 am-1 pm daily. In the afternoon, fresh grass was supplied to the animals. All animals were free to access water. The supplied water was changed two times daily.

#### 3.6.2 *Housing*

The Sheep and Goat Shed of CVASU was used for this study. The floor was made of wood & the roof of the shed was made of tin. The length and width of the shed were 17 feet & 12 feet. The height was about 12 feet from the ground. The floor was 2.5feet above the ground for better drainage of urine. The shed was divided into 4 blocks and the area each block was about 36 square feet. Each block had 3 animals. The feeder and drinker were separately supplied in each block.



Figure 5: Experimental animal shed



Figure 6: Single block of shed

# 3.7 Determination of growth performance

# 3.7.1 Determination of live weight

In order to evaluate the growth performance live body weight of sheep was determined by digital weighing balance 1 day weekly up to 8 weeks.

# 3.7.2 Determination of average daily live weight gain

The average daily live weight gain indicates the average amount of weight that an animal gains each day for a given period of time.

Calculation of average daily live weight gain

#### 3.7.3 Determination of FCR

FCR (Feed Conversion Ratio) is a value that shows the efficiency with which the animal is capable of transforming feed consumed into body mass (meat).

# Calculation of FCR

# 3.8 Laboratory Analysis for fecal egg count

Daily fecal samples (2 days/week) were collected from each animal in the morning and this was continued for 8 weeks. These samples were subjected to the McMaster fecal egg counting technique, using 3 g individual fecal samples (Chandrawathani *et al.*, 2019). The feces sample was firstly checked with direct smear, sedimentation, and floatation techniques to confirm eggs present in the sample before going to the McMaster egg counting technique. Direct smear, flotation and sedimentation methods are described by (Urquhart *et al.*, 1996).



 ${\it Figure~7: Preparation~of~fecal~sample}$ 



Figure 8: Fecal egg count on microscope

#### 3.8.1 Direct smear

A small amount of feces sample and a few drops of water took in a microscopic slide and thoroughly mixed it. Then tilted the slide and allowed the lighter eggs free from heavier debris. After that, the debris is discarded from the slide and a cover slip is placed on the fluid part and then examined on the microscope.

#### 3.8.2 Sedimentation

At first, made a homogenous mixture with feces and water & passed the mixture through a coarse strainer. Then transferred the filtrate into a test tube. After that allowed to sediment for 10 minutes & remove the supernatant and collected a few drops of the sediment by Pasteur pipette & put it into a slide then examined microscopically. This technique is used for heavier eggs (trematodes egg).

# 3.8.3 Simple Tube Floatation

The floatation method is used for nematode and cestode eggs. In this method flotation fluid like saturated salt, or sugar solution is used generally.

At first, took a small amount of feces sample (approximately 2gm) and added 10 ml floatation fluid (saturated sugar solution) into a container and thoroughly mixed it. After that, the suspension is passed into a tea strainer and poured into a test tube up to the top leaving a convex surface and carefully put a cover slip on the surface. Then held the test tube for 10-15 minutes in a test tube rack. Carefully lifted off the cover slip from the tube, together with the drop of fluid adhering to it, and immediately placed the coverslip on a slide and examined the slide microscopically.

# 3.8.4 *McMaster egg counting method (simple floatation)*

This method is developed as a quantitative test of fecal sample. The other techniques previously described are unable to count the eggs present in fecal sample. But by this method we can count the eggs and express in a unit called EPG (egg per gram).

At first, weighed 3gm sample and took it into a measuring container. Added saturated sugar solution up to the 45 ml mark (1:15 dilution). Mixed the contents and filtered through a tea strainer. After that mixed the filtrate and fill up both the counting chamber of McMaster's slide. Then counted the eggs present within the ruled areas of both the chambers.

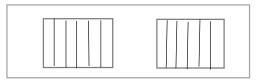


Figure 9: McMaster Counting Chamber

#### Calculation

Egg per gram feces (EPG) = 
$$\frac{\text{(Chamber 1+ Chamber 2)}}{2} \times 100$$

# 3.9 Statistical analysis

The analysis of variance was done on the data on feed intake, growth performance, and proximate analysis using the one-way ANOVA in the SAS 9.2 software (2007). The differences in the means will be compared by Duncan's multiple range tests at 5% level (P<0.05).

# **CHAPTER 4: RESULTS**

# 4.1 Chemical composition different parts of Azadirachta indica (Neem)

The Chemical composition different parts of Azadirachta indica (Neem) leaf are presented in Table 4. In our study DM% of neem foliage, leaf, stem is 34.79%, 32.42%, 37.87% respectively. It is observed that neem stem contains more dry matter than leaf and foliage. Moisture contents are respectively 65.21%, 67.58%, 62.13% which shows neem leaf has more moisture content than foliage and stem. Ash content of neem is 8.815% in foliage, 9.82% in leaf, 6.33% in stem which means leaf content more ash. CP% of foliage is 12.75%, leaf is 14.07%, stem is 5.28%. Leaf content more protein than other part of plant. CF% of foliage is 19.33%, leaf is 12.50%, stem is 42.95%. Here stem contains more fiber.

Table 4: Chemical composition of different parts of Azadirachta indica (Neem) (Mean  $\pm SE$ ; n=3)

Variables	Neem tree			P value	Level of significance
	Foliage	Leaf	Stem		significance
DM (%)	34.79 <sup>b</sup>	32.42 <sup>b</sup>	37.87 <sup>a</sup>	0.01	*
Moisture (%)	65.21 <sup>a</sup>	67.58 <sup>a</sup>	62.13 <sup>b</sup>	0.01	*
Ash (%)	8.81 <sup>b</sup>	9.82ª	6.33c	0.001	**
CP (%)	12.75 <sup>b</sup>	14.07 <sup>a</sup>	5.28 <sup>c</sup>	0.001	**
CF (%)	19.33 <sup>b</sup>	12.50°	42.95 <sup>a</sup>	0.001	**
OM (%)	91.19 <sup>b</sup>	90.17 <sup>c</sup>	93.67 <sup>a</sup>	0.001	**

a,b,c Means in the same row and in each treatment with different superscripts differ significantly among cutting interval at P<0.05 level; n= observation numbers.

# 4.2 Effect of dietary Neem foliage on growth performances of sheep

The effect of neem foliage on growth performance of sheep is represented in table 5. From that table it was observed that in case of body final weight of sheep no significant (P> 0.05) difference was found among the treatments. But there was found significant difference in average daily live weight gain (g  $d^{-1}$ ) among the treatments and also found strong significant difference on FCR among the treatments. The average daily weight gain was higher and the FCR value was lower at  $T_3$  which indicated high positive effect of neem leaves on this treatment compared to others.

Table 5: Growth performance data

	Treatments				P value	Level of
*Variables	T <sub>1</sub>	T <sub>2</sub>	<b>T</b> 3	T <sub>4</sub>		significance
	(control)					
Initial BW	12.10	12.17	12.50	12.33	0.75	NS
(kg)						
Final BW (kg)	16.30	16.73	17.17	16.80	0.28	NS
Average daily	70.00 <sup>b</sup>	76.11 <sup>a</sup>	77.78 <sup>a</sup>	74.44 <sup>ab</sup>	0.01	*
live weight						
gain $(g d^{-1})$						
FCR	8.57 <sup>a</sup>	7.89 <sup>b</sup>	7.72 <sup>b</sup>	8.06 <sup>ab</sup>	0.008	*

a.b.c Means in the same row and in each treatment with different superscripts differ significantly among cutting interval at P<0.05 level; n= observation numbers. FCR= Feed conversion ratio,  $*T_1$  = 300g concentrate mixture,  $T_2$  = 250 g concentrate mixture+ 50g Dry neem foliage,  $T_3$  = 200 g concentrate mixture+ 100g Dry neem foliage,  $T_4$ = 150g concentrate mixture+ 150g Dry neem foliage.

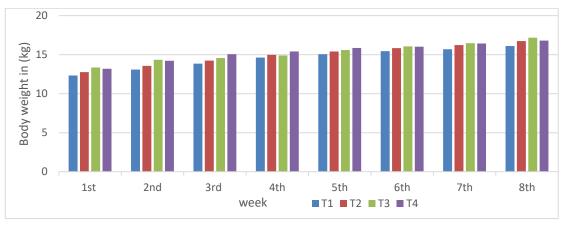


Figure 10: Growth performance chart

# 4.3 Effect of dietary Neem foliage on fecal egg counts of sheep

The effect of dietary neem foliage on fecal egg counts of sheep is represented on figure 11. In this figure, it was founded that the fecal egg count (FEC) among the treatments was almost similar up to the middle of  $2^{nd}$  week. Then, the egg count drastically lowered at the  $T_4$  treatments but the  $T_3$  &  $T_4$  treatments were similar up to the  $3^{rd}$  week. After that, the  $T_2$  &  $T_3$  were dropped down simultaneously but at  $T_1$  it was suddenly raised up to  $3^{rd}$  week & it was slowing down up to the  $7^{th}$  week again at  $8^{th}$  week the FEC was raised.

So it was observed from the study that the study the sheep that were supplied higher neem foliage (150gm) in feed the fecal egg count is quite lower than the other treatments. Besides those Sheep that were not supplied neem foliage had higher fecal egg count compared to the other treatments.

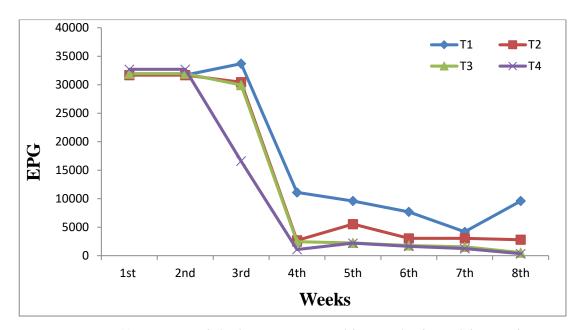


Figure 11: Mean nematode fecal egg counts (e.p.g.) of the Control and Neem fed (Treated) group.

Here in this figure, the blue line indicates the fecal egg count of  $T_1$  treatment which was considered the control group for this study. The red line, green line, and purple line indicate the fecal egg counts of  $T_2$ ,  $T_3$ , and  $T_4$  respectively.

 $*T_1 = 300g$  concentrate mixture,  $T_2 = 250$  g concentrate mixture+ 50g Dry neem foliage,  $T_3 = 200$  g concentrate mixture+ 100g Dry neem foliage,  $T_4 = 150g$  concentrate mixture+ 150g Dry neem foliage.

# **CHAPTER 5: DISCUSSION**

The purpose of this study to evaluate the potentiality of neem leaves on the growth performance of sheep as a replacement for concentrate feed and assessed the potency of neem leaves as a natural anthelmintic for sheep.

In a study Moisture, Ash, CP, and CF of neem plant shows result of  $14.30\pm0.02\%$ ,  $4.03\pm0.10\%$ ,  $1.22\pm0.22\%$ ,  $10.86\pm0.11\%$  (A.Otache. *et al.*, 2017) which are much lower than the value we found. These values are also lower than the values of 13.42% protein, 11.93% ash and 5.7% fiber recorded in other study (Atangwho *et al.*, 2009)

In this current study here, we found no significant difference in case of final body weight gain among the treatments which corresponds to earlier study by (Sarkar *et al.*, 2016), reported that in case of body weight of indigenous cows no significant difference was found after feeding of neem and ata leaves. On the other hand, in this study the average daily live weight gain and FCR value showed significant difference among the treatments. Here in the T<sub>3</sub> & T<sub>4</sub> treatments the average daily weight gain of animals quite higher compared to other two treatments. So, from this study we found positive effect of neem leaves on growth performance of sheep.

Previous studies by (Khalid *et al.*, 2005), reported that the body weight of neem leaves treated sheep was increased significantly. (Hossain *et al.*, 1996), found neem leaves and neem seeds kernels increased the body weight of cattle. Similarly, (Amin *et al.*, 2008), reported that the neem leaves significantly increased the body weight of cattle. In another report by (Dida *et al.*, 2019), found that supplementation of 300gm neem leaf had a higher gain of body weight of goats and was economically feasible.

In case of fecal egg count it showed the load of parasite was higher for a week among the treatments. But as experimental feed continued the egg count drastically reduced on those group supplied with neem leaf beside in the control group  $(T_1)$  the egg count also lowered this might be due to hatching of egg into larva. But after 7 week the curve became upward direction which indicate higher fecal egg present in  $T_1$  treatment.

In some earlier studies (Khalid *et al.*, 2005), reported that the neem (10% water extract of leaves) reduced fecal egg count of sheep significantly. (Rob *et al.*, 2004) observed that water extracts of neem were 53.72% effective against hemonchosis in sheep. Another study by (Arunachal *et al.*, 2002) found that neem leaves, seeds and bark were 53%, 49% and 38% infective against the gastrointestinal helminthes in sheep.

On the other hand, in some studies (Chandrawathani *et al.*, 2006) found that there was no statistical difference of fecal egg count on sheep but reported that the number of worms were less in animals fed with fresh neem leaves. This study similar to (Khadijah *et al.*, 2005) and (Wong *et al.*, 2005), in both studies found that fresh neem and pelleted neem showed no significant difference in fecal egg counts, compared with control sheep, although the mean fecal egg counts was higher in control group.

# Limitations

The study period was short, and this might be a cause of finding not significant result on differences between initial and final body weight among the treatments. Sometimes there might be a problem of finding trematodes eggs on fecal egg count by using McMaster egg counting method (simple floatation).

# **CONCLUSION**

The addition of neem leaf on diet of sheep found positive effect on growth performance. In addition to, it was observed that neem leaf also has positive effect on reducing parasitic load of sheep. The average daily weight gain was higher in neem fed group compared to control group. FCR value also indicated positive impact on neem fed group. But in case of differences of body weight of sheep there was not found significant result. However further study is recommended with large number of sheep to make final conclusion on neem leaves feeding.

# REFERENCES

- Adjorlolo, L.K., Timpong-Jones, E.C., Boadu, S. and Adogla-Bessa, T., 2016. Potential contribution of neem (*Azadirachta indica*) leaves to dry season feeding of ruminants in West Africa. *Development*, 28(5).
- Arunachal, P.K., Karunanithi, K. and Narendrababu, R., 2002. Comparative study on anthelmintic efficacy of Neem products and Prazlplus in sheep. *The Indian Journal of Small Ruminants*, 8(2), pp.131-132.
- Amin, M.R., Mostofa, M., Awal, M.A. and Hossain, M.A., 2008. Effects of neem (*Azadirachta indica*) leaves against gastrointestinal nematodes in cattle. *Journal of the Bangladesh Agricultural University*, 6(452-2018-4018), pp.87-92.
- Ansari, J., Khan, S.H., ul Haq, A. and Yousaf, M., 2012. Effect of the levels of *Azadirachta indica* dried leaf meal as phytogenic feed additive on the growth performance and haemato-biochemical parameters in broiler chicks. *Journal of Applied Animal Research*, 40(4), pp.336-345.
- AOAC (2005) Official Methods of Analysis. 18th Edition, Association Official Analytical Chemists, Gaitherburg.
- Atangwho, I.J., Ebong, P.E., Eyong, E.U., Williams, I.O., Eten, M.U. and Egbung, G.E., 2009. Comparative chemical composition of leaves of some antidiabetic medicinal plants: *Azadirachta indica*, Vernonia amygdalina and Gongronema latifolium. *African Journal of Biotechnology*, 8(18).
- Bais, B., Purohit, G.R., Dhuria, R.K. and Pannu, U., 2002. Nutritive value of sares and neem leaves in Marwari goats. *Indian Journal of Animal Nutrition*, 19(3), pp.266-268.
- Bhowmik, S., Chowdhury, S.D., Kabir, M.H. and Ali, M.A., 2008. Chemical composition of some medicinal plant products of indigenous origin. Bangladesh Veterinarian, 25(1), pp.32-39.
- Chandrawathani, P., Chang, K.W., Nurulaini, R., Waller, P.J., Adnan, M., Zaini, C.M., Jamnah, O., Khadijah, S. and Vincent, N., 2006. Daily feeding of fresh Neem leaves (*Azadirachta indica*) for worm control in sheep. *Tropical biomedicine*, 23(1), pp.23-30.
- Chandrawathani, P., Premaalatha, B., Omar, J. and Mamat, Z.C., 2019. Manual on Parasitology.

- Dehuri, M., Palai, S., Mohanty, B. and Malangmei, L., 2021. Anti-helminthic Activity of Plant Extracts against Gastrointestinal Nematodes in Small Ruminants-A Review. *Pharmacognosy Reviews*, 15(30).
- Dida, M.F., Challi, D.G. and Gangasahay, K.Y., 2019. Effect of feeding different proportions of pigeon pea (*Cajanus cajan*) and neem (*Azadirachta indica*) leaves on feed intake, digestibility, body weight gain and carcass characteristics of goats. Veterinary and Animal Science, 8, p.100079.
- DLS.(2021). Department of Livestock Services. Ministry of Fisheries and Livestock. Government of the Peoples' Republic of Bangladesh.
- Eid, A., Jaradat, N. and Elmarzugi, N., 2017. A Review of chemical constituents and traditional usage of Neem plant (*Azadirachta Indica*). *Palestinian Medical and Pharmaceutical Journal (Pal. Med. Pharm. J.*), 2(2), pp.75-81.
- Ghimeray, A.K., Jin, C.W., Ghimire, B.K. and Cho, D.H., 2009. Antioxidant activity and quantitative estimation of azadirachtin and nimbin in *Azadirachta indica* A. Juss grown in foothills of Nepal. *African Journal of Biotechnology*, 8(13).
- Girish, K. and Shankara, B.S., 2008. Neem–a green treasure. *Electronic Journal of Biology*, 4(3), pp.102-111
- Islam, K.S., Ershaduzzaman, M., Munsi, M.N., Kabir, M.H., Das, S. and Kabir, M.H.B., 2016. Establishment of health management package for native sheep of Bangladesh. Research in Agriculture Livestock and Fisheries, 3(1), pp.151-155.
- Jamra, N., Das, G., Singh, P. and Haque, M., 2015. Anthelmintic efficacy of crude neem (*Azadirachta indica*) leaf powder against bovine strongylosis. *Journal of parasitic diseases*, 39(4), pp.786-788.
- Khalid, S.M.A., Amin, M.R., Mostofa, M., Choudhury, M.E. and Uddin, B., 2005. Effects of indigenous medicinal plants (neem and pineapple) against gastrointestinal nematodiasis in sheep. *International Journal of Pharmacology*, *1*(2), pp.185-189.
- Kingston, D.G., 2011. Modern natural products drug discovery and its relevance to biodiversity conservation. *Journal of natural products*, 74(3), pp.496-511.
- Koul, O., Multani, J.S., Singh, G., Daniewski, W.M. and Berlozecki, S., 2003. 6β-hydroxygedunin from *Azadirachta indica*. Its potentiation effects with some non-azadirachtin limonoids in neem against lepidopteran larvae. Journal of agricultural and food chemistry, 51(10), pp.2937-2942.

- Makkar, H.P., 2007. Feed supplementation block technology–past, present and future. Urea-Molasses Multinutrient Blocks: Simple and Effective Feed Supplement Technology for Ruminant Agriculture, pp.1-12.
- Ngamsaeng, A., Wanapat, M. and Khampa, S., 2006. Evaluation of local tropical plants by in vitro rumen fermentation and their effects on fermentation end-products. *Pakistan Journal of Nutrition*, 5(5), pp.414-418.
- Niranjan, P.S., Singh, J. and Verma, D.N., 2008. Mineral status and antinutritional factors of common tree leaves. Indian veterinary journal, 85(10), pp.1067-1069.
- Ogbuewu, I.P., Odoemenam, V.U., Obikaonu, H.O., Opara, M.N., Emenalom, O.O., Uchegbu, M.C., Okoli, I.C., Esonu, B.O. and Iloeje, M.U., 2011. The growing importance of neem (*Azadirachta indica* A. Juss) in agriculture, industry, medicine and environment: a review. *Research Journal of Medicinal Plant*, *5*(3), pp.230-245.
- Otache, M.A. and Agbajor, G.K., 2017. Proximate and mineral composition of leaves of *Azadirachta indica. Int. J. Curr. Res. Chem. Pharm. Sci*, 4(11), pp.50-54.
- Paengkoum, P., 2010. Effects of neem (*Azadirachta indica*) and leucaena (*Leucaena leucocephala*) fodders on digestibility, rumen fermentation and nitrogen balance of goats fed corn silage. *Journal of Animal and Veterinary Advances*, 9(5), pp.883-886.
- Panhwar, F., 2005. The neem tree (*Azadirachin-Indica* A. Juss), a natural pesticide practice in Pakistan. *Digitalverlag GmbH: Germany*.
- Patil, P.V., Gendley, M.K. and Patil, M.K., 2021. Leaf protein: New protein source for ruminants.
- Radhakrishnan, L., Gomathinayagam, S. and Balakrishnan, V., 2010. Evaluation of anthelmintic effect of neem (*Azadirachta indica*) leaves on Haemonchus contortus in goats. *Research Journal of parasitology*, *5*(*4*), pp.292-297.
- Raghuvansi, S.K.S., Prasad, R., Mishra, A.S., Chaturvedi, O.H., Tripathi, M.K., Misra, A.K., Saraswat, B.L. and Jakhmola, R.C., 2007. Effect of inclusion of tree leaves in feed on nutrient utilization and rumen fermentation in sheep. Bioresource technology, 98(3), pp.511-517.
- Rahman, M.M., 1989. Sheep production and development in Bangladesh. In Proceedings of the workshop on sheep production in Asia, PACCARD, Los Banos, Philippines (pp. 81-95).
- Rahman, W.A., Lee, R. and Sulaiman, S.F., 2011. In vitro anthelmintic activity of Neem plant (*Azadirachta indica*) extract against third-stage *Haemonchus contortus* larvae from goats. Global Veterinaria, 7(1), pp.22-26.

- Ramana, D.B.V., Singh, S., Solanki, K.R. and Negi, A.S., 2000. Nutritive evaluation of some nitrogen and non-nitrogen fixing multipurpose tree species. Animal feed science and technology, 88(1-2), pp.103-111.
- Rob, S., Mostofa, M., Awal, M.A., Shahiduzzaman, M. and Sardar, S.A., 2004. Comparative efficacy of albendazole (Endokil) and neem (*Azadirachta indica*) leaves extract against hemonchosis in sheep. *Progressive Agriculture*, 15(2), pp.33-39.
- Sarker, M.A.H., Khan, M.A.S., Rashid, M.H. and Islam, M.A., 2016. Effect of *Azadirachta indica* and *Annona reticulate* leaf as natural anthelmintics and their effects on performances of zebu cow under subsistence farming condition in Bangladesh. *International Journal of Pharmaceutical, Chemical & Biological Sciences*, 6(1).
- Seresinhe, T. and Marapana, R.A.U.J., 2011. Goat farming systems in the southern province of Sri Lanka: Feeding and management strategies. World J. Agric. Sci, 7(4), pp.383-390.
- Tiwari, R., Verma, A.K., Chakraborty, S., Dhama, K. and Singh, S.V., 2014. Neem (*Azadirachta indica*) and its potential for safeguarding health of animals and humans: A review. *Journal of Biological Sciences*, 14(2), p.110.
- Urquhart, G.M., Armour, J., Duncan, J.L., Dunn, A.M. and Jennings, F.W., 1996. Veterinary parasitology. (2nd edn), Black Well Science. *The University of Glaskow, Scotland, UK*, pp.181-188.
- Yakubu, S., Saleh, U.A. and Abdullahi, G., 2006. In-vitro anthelmintic efficacy of crude aqueous extracts of neem (*Azadirachta indica*) leaf, stem and root on nematode. *Animal Research International*, *3*(3), pp.549-552.

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