EFFECT OF NEEM LEAF POWDER SUPPLEMENTED IN BASAL DIET ON GROWTH PERFORMANCE AND APPARENT ILEAC DIGESTIBILITY IN BROILER CHICKEN

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Table of contents

List of tables

List of figures

List of abbreviations

FCR	Feed conversion ratio
DM	Dry matter
OM	Organic matter
CP	Crude protein
EE	Ether extract

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Abstract

The aim of this study was to investigate the effects of feeding *Azadirachtaindica* leaf powder to broilers on their growth performance and apparent ileal digestibility in broilers. It may be because neem leaves have a variety of effects on intestinal micro flora, which protects it from stressful situations, that broiler chicks' body weight and weight gain increased when neem leaf powder was introduced to their diets. Both the control and neem leaf-fed broiler chicken groups consume considerably different amounts of feed. Its appetite- and digestion-stimulating, antibacterial, and hepatoprotective qualities, which assist to lower the microbial burden on birds and increase feed consumption, may account for the increased feed intake. The body weight, body weight gain, feed intake, and FCR across the treatment groups were found significantly higher ($P< 0.05$) compare to control. Thus, Neem leaf powder can be used in commercial broiler chicken diets as a natural, cost-effective feed addition to improve overall performance. It is concluded that adding 0.5% NLM to broiler diets will promote optimal performance and financial gain.

Keywords: Neem leaf powder, Growth performance, FCR, Apparent ileal Digestibility

Chapter 1: Introduction

In developing countries where daily protein consumption is significantly below the recommended levels, poultry raising is a key contributor to closing the protein gap. The cost of feed, which makes up 70–80% of the cost of production, is one of the major obstacles to poultry production, particularly in developing nations (Siahan*et al.*, 2021; Zacaria and Ampode, 2021). Public health is also at risk when artificial antibiotics are added to chicken diets to improve performance and meet the demand for animal protein (Lagua and Ampode, 2021). Due to the persistence of antibiotic residues in the tissues of the birds, synthetic antibiotics used as growth promoters are costly and have a negative impact on consumer health (Ezzatet al., 2018).

Traditional medicine has made use of neem (Azadirachtaindica) (Saleemet al., 2018). Neem leaves produce a variety of liminoids (nimbin and its derivatives), as well as quercetin and nimbosterol, according to Elangovan et al. (2000).Neem leaf meal's approximate composition was reported by Esonu et al. (2006) to be 92.40% dry matter, 7.58% moisture, 20.68% crude protein, 16.60% crude fibre, 4.13% ether extract, 7.10% ash, and 43.91% nitrogen free extract. However, because its leaves have high levels of dietary fibre (Esonuet al., 2005; Bonsuet al., 2012), adding 2.5% to 7.5% of them to the meal has a negative impact on chickens' ability to grow (Udedibie and Opara, 1998; Bonsuet al., 2012; Ubuaet al., 2019). However, a dried extract of neem leaves can be added to chicken feed without having a deleterious impact on the animals' ability to grow or their serum biochemistry (Nnenna and Okey, 2013). Additionally, due to the high content of phenolic chemicals, including gallic acids and ferulic acids (Singh et al., 2005; Shewale and Rathod, 2018), neem leaves show significant antioxidant properties (Prakash et al., 2007; Heymanet al., 2017).In their trial, Laboni et al. (2007) shown that providing broiler chickens with NLM up to a 20 g/kg dietary level reduces mortality and morbidity while also enhancing their general health.

Neem leaves have been used in animal production in numerous feeding trials, particularly as an anti-helminth agent. The majority of research, however, utilised neem seeds as a source of protein in animal feed (Aruwayoet al., 2011). According to Ubua et al. (2019), the plant's antifungal, antiviral, antibacterial, and growth-promoting therapeutic properties demonstrated their relevance without having any negative impacts on chickens. Moreover, the anti-nutritional components

sodium nimbolide, gallic acid, azadirachtin, and nimbidin in neem leaf meal reduce the ability of plants to use nutrients (Kharde and Soujanya, 2014). However, there are limited work on hybrid neem leaf powder on growth performance and apparent ileal digestibility in broiler. Therefore, the present research has been conducted to evaluate the effect of neem leaf on body weight gain, FCR and ileal digestibility in broiler.

Specific objectives

1) To assess the effectiveness of neem powder as an antimicrobial growth promoter.

2) To observe the growth performance of broiler.

3) To determine the digestibility of feed sample.

Chapter 2: Materials and Methods

2.1 Study area:

The whole examination was run at the Chattogram Veterinary and Animal Sciences University (CVASU), which is situated in Khulshi, Chattogram, Bangladesh. Animal experiments were conducted at the poultry research shed, and all laboratory work was conducted in the department's post-graduate (PG) and undergraduate animal nutrition laboratory.

2.2 Preparation of Neem powder:

Fresh hybrid neem leaves were taken from different area of chattogram and properly cleaned with clean water. The leaf was then exposed to the sun for seven days to dry. Finally, using an electric grinder (Panasonic MX-AC555), it was ground into powder and kept at 4°C until it was blended with the feed.

Fig 2.1: Sun dried neem powder

Fig 2.2: Ground neem powder

2.3 Experimental unit collection:

From Kazi farms limited in Chattogram, a total of 63 day-old chicks were collected. Following unloading, all of the chicks were inspected for any abnormalities and their weights were recorded to preserve uniform size across all treatment groups. They were split into three groups, each with three replicates and seven birds in each replicate (T0 = control, T1 = 0.5% neem, and T2 = 1% neem).

Table 2.1. Layout of the experiment

*Dietary treatment	Ratio of herb	Replication	Group-Replication	No. of birds	Total no. of birds per treatment	Total
To	No herb	3	ToR1 ToR2 ToR3	7 7 7	21	63
T1	0.5 % neem powder	3	ToR1 ToR2 ToR3	7 7 7	21	
T2	1% neem powder	3	ToR1 ToR2 ToR3	7 7 7	21	

2.4 Preparation of shed:

2.4.1 Cleaning the shed and sanitation:

The surfaces of the broiler rearing cages and the floor of the poultry research shed were initially cleaned with bleaching powder and running water. After a thorough cleaning and washing, the shed was left to dry for a minimum of four to five days. To ensure appropriate ventilation while the clothing was drying, all windows were left open. A 1:2 mixture of formaldehyde (CH2O) and potassium permanganate (KMNO4) was used to disinfect the chicken coop. To disperse the chicks evenly, all of the cages were constructed based on treatments without any prejudice. For the first

two weeks of their lives, all the birds were kept in wire cages with paper bedding. For every seven birds, there was a cage measuring (3.5 ft. × 1.63 ft.).A separate drinker and a round feeder were set up for the birds' water needs during the starter period. A feeding dish was then placed in front of each cage for feed supply.

Every biosecurity rule was strictly observed while the bird was being grown in the poultry research shed. At the entrance to the shed, there was a footbath filled with potassium permanganate (KMNO4). All drinkers and feeders were completely cleaned before being used to supply water or food, respectively, using soap and clean water on a regular basis. As the bird's feces produce more ammonia gas and smell, the waste was removed every day and the exhaust fan was turned up for proper ventilation.

2.5 Management of birds:

2.5.1 Brooding:

The floor of the brooder cages was covered with clean, dry newspaper, which served as safe bedding for healthy brooding. The paper sheet was regularly replaced during the day. During the period of brooding, hardboard was covering the cages' exterior walls. The chicks were brooded at 95°F, 90°F, and 85°F for the first, second, and third weeks, respectively. Each cage featured a 60-watt bulb for adequate lighting. A room thermometer was used to determine the room's temperature. The entire paper sheet and hardboard were removed following the brooding time.

2.5.2 Feeding and watering:

The control group (TO) had simply a basic diet in accordance with the experiment's design. In the treatment groups T1 and T2, neem powder was added to the basal diet at a ratio of 0.5% and 1%, respectively. Clove powder contained 0.04% and neem powder contained 0.5% in treatment group T3. The entire feed ration was created using DM. The birds received broiler starter ration during the first two weeks and broiler finisher ration during the final three weeks. Water was made available to the birds at all times**.**

2.5.3 Vaccination:

Throughout the trial, the birds were vaccinated against Newcastle and infectious bursal diseases. The Newcastle disease vaccine (BCRDV) was injected into one eye of each bird on the fifth day, and a booster shot was administered on the fifth day. The next day, the 11th infectious bursal disease vaccine (Gumboro) was administered, and on the 16th, a booster dose.

2.6 Ration for the experiment:

Starter and Grower, two distinct rations, were meticulously prepared, and all feed components were bought at the neighborhood store. The feed components were then combined and prepared for the rations in the laboratory. The meals for the corresponding treatment groups included ground neem powder. The Association of Official Analytical Chemists' (AOAC, 2005) method was used to determine the proximate analysis of the starter and grower diets. The basal diets' components and predicted chemical compositions are displayed in Table.

Table 2.2. Ingredient composition of starter ration

	Starter Diet		
Ingredients	**T0**	**T1**	**T2**
Maize	53.3	53.4	53
Rice polish	3.5	3.2	3.1
Palm oil	4	4	4
Soybean meal	32.9	32.9	32.7
Neem leaf	**0**	**0.5**	**1**
Fishmeal	3.25	3.15	3.25
Lime stone	1.4	1.2	1.5
Dicalcium phosphate	0.45	0.45	0.25
Common salt	0.25	0.25	0.25
Vitamin-mineral premix	0.25	0.25	0.25
Toxin binder	0.1	0.1	0.1
Coccidiostat	0.1	0.1	0.1
Methionine	0.2	0.2	0.2
Lysine	0.2	0.2	0.2
Antioxidant	0.1	0.1	0.1
Total	100	100	100

Table 2.3. Ingredient composition of starter ration

	Finisher Diet		
Ingredients	**T1**	**T6**	**T7**
Maize	60	59.86	60.01
Rice polish	5.5	5.22	4.58
Palm oil	4	4	4
Neem leaf meal	**0**	**0.5**	**1**
Soybean meal	22.7	22.56	22.66
Fishmeal	5.4	5.45	5.35
Lime stone	1.02	1.03	1.02
Dicalcium phosphate	0.38	0.38	0.38
Common salt	0.25	0.25	0.25
Vitamin-mineral premix	0.25	0.25	0.25
Toxin binder	0.1	0.1	0.1
Coccidiostat	0.1	0.1	0.1
Methionine	0.1	0.1	0.1
Lysine	0.1	0.1	0.1
Antioxidant	0.1	0.1	0.1
Total	100	100	100

2.7 Data collection:

Every week for the entire experiment, bird feed consumption, weight gain, and FCR were recorded. The weight increase was estimated by dividing the ultimate body weight of the birds by their initial body weight. The amount of feed consumed was calculated by subtracting any remaining food from the total amount of food provided to the birds. The feed conversion ratio (FCR) was calculated by dividing feed intake by bird weight increase.

2.8 Proximate analysis:

Proximate analysis techniques were utilized to determine the sample's moisture Dry matter, crude protein, ash, and crude fiber contents in accordance with AOAC (2006).

The study was conducted at Chattogram Veterinary and Animal Sciences University's Department of Animal Science and Nutrition laboratory.

Fig 2.3: Determination of Ash

Fig 2.4: Determination of CP

2.9 Nutrient Digestibility

Three days before slaughter, titanium oxide (TiO2) was added to the finisher meal as an indigestible marker at a concentration of 3g/1000g. After the birds were killed, the ileal digesta was collected and refrigerated until testing. The association of official analytical chemist technique (AOAC, 2005) was used to measure the nutrients (DM, CP, EE, and total ash) in the frozen ileal

digesta after it had been pulverized and dried in a hot air oven (POL-34035, POL-EKO APARATURA). The TiO2 content of the feed and digesta was measured using the techniques described by Khatun et al. (2008). This test was first conducted with three solutions prepared.

Solution1: Standard TiO2 solution

Solution2: 7.4M H2SO4 solution

Solution3: 30% H2O2 solution

To test the TiO2 absorbance in standard solution, eleven volumetric flasks (100 ml) were used. Solution 1, 2, and 3 were added together with distilled water in accordance with the instructions in Table 2.4.

Table 2.4.

N0. of volumetric flask(100ml)	1	2	3	4	5	6	7	8	9	10	11
TiO2 solution (ml)	0	1	2	3	4	5	6	7	8	9	10
7.4M H2SO4 solution (ml)	10	9	8	7	6	5	4	3	2	1	0
30% H2O2 solution (ml)	20	20	20	20	20	20	20	20	20	20	20
Distilled water (ml)	70	70	70	70	70	70	70	70	70	70	70

The feed and ingesta sample were prepared according to the table's technique for measuring TiO2 absorbance:

a 0.1 g dry ground sample was put in a porcelain crucible and preheated for 13 hours at 580 o C in a muffle furnace. The crucible was then heated for 60 minutes while receiving 10 ml of 7.4M

H_2SO_4. The aforementioned solution was added to a beaker along with 25ml of distilled water after cooling. The solution was then filtered into a 100ml volumetric flask using Whatman filter paper. Then, 20ml of H_2O_2 solution was poured to a volumetric flask, and 100ml of distilled water was added to make 100ml. Last, the solution's absorbance was measured at 410 nm with a spectrophotometer (UV-2600, Shimadzu, UV-VIS Spectrophotometer).

According to Hashemi et al. (2014), ileal content and titanium marker ratios in the diet were used to calculate apparent ileal digestibility (AID) of DM, CP, EE, and ash:

AID of nutrient = 100- [(% TiO_2 in feed / %TiO_2 in ileal content) * (% of nutrient in ileal content / %of nutrient in feed * 100].

Fig 2.5: collection of digesta from ileum

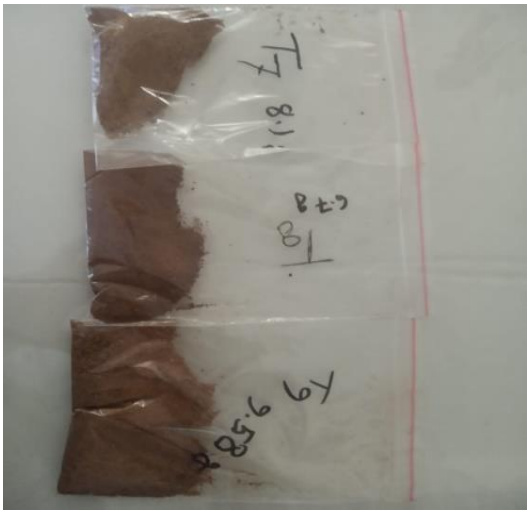

Fig 2.6: Dried digesta sample

Fig 2.7: Determination of the digestibility of feed

Chapter3: Result and Discussion

The findings of the dietary effect of neem powder on broiler growth (Body weight, body weight gain, FCR) performance, digestibility are described in this chapter.

3.1 Body weight: The impact of feeding neem powder of broiler has great effect on growth performance. The body weight on different week among the three groups are shown in Table 3.1. The table shows that there is no effect of neem powder on initial body weight and 1st week age. At 2nd and 3rd weeks it is significant means intake of neem powder increase the body weight where highest value in T1, lowest in To. Similarly, at 4thand 5thweek age it is highly significant where rapidly increase body weight and which is highest in T1 and lowest in To.

Table 3.1. Effect of neem on body weight on broiler in every week

parameter	treatment				
	To	T1	T2	P value	Level of significance
Initial body wt	44.32000^{ab}	44.13333^{b}	44.42000^{a}	0.0689	NS
Body wt at 1st week	134.62^{a}	135.00^{a}	116.62^{a}	0.1980	NS
Body wt at 2nd week	292.339^{ab}	312.933^{a}	276.639^{b}	0.0186	*
Body wt at 3rd week	464.92^{b}	557.78^{a}	507.53^{ab}	0.0118	*
Body wt at 4th week	798.58^{c}	959.31^{a}	878.89^{b}	<0.0001	***
Body wt at 5th week	1219.42^{c}	1421.96^{a}	1367.89^{b}	0.0002	***

Note: [a,b,c] in a row with no shared superscripts deviate significantly ($p<0.05$). The data is presented as an average of three replicate groups of seven birds each. (n=21). * TO =Basal diet, T1 = Basal diet+0.5% neem powder, T2=Basal diet+1% neem powder ** = Significant, NS = Not Significant.

3.2 Body weight gain:

Feeding of neem powder has also great effect on body weight gain which are represent in table 2. Here at 1st and 2nd week of age there has no significant difference of feeding neem powder in body weight gain. In 3rd week age it was significant. Similarly, at 4th and 5th week age it was highly significant which means neem powder has great effect on body weight gain.

Table 3.2. Effect of neem on body weight gain

parameter	treatment			P value	Level of significance
	To	T1	T2		
Body wt gain at 1st week	12.900[a]	12.981[a]	10.314[a]	0.1910	NS
Body wt gain at 2nd week	22.531[a]	25.419[a]	22.860[a]	0.2023	NS
Body wt gain at 3rd week	24.654[b]	34.979[a]	32.984[a]	0.0263	*
Body wt gain at 4th week	47.667[b]	57.361[a]	53.052[a]	0.0069	**
Body wt gain at 5th week	60.119[b]	66.092[a]	69.856[a]	0.0062	**

Note: [a,b,] in a row with no shared superscripts deviate significantly ($p<0.05$). The data is presented as an average of three replicate groups of seven birds each. (n=21). * TO =Basal diet,T1 = Basal diet+0.5% neem powder, T2=Basal diet+1% neem powder ** = Significant, NS = Not Significant.

3.3 Feed intake: There has no significant effect of feeding neem powder on feed intake which are shown in table 3. At 1st and 2nd week feed intake is less in T1 and T2 than To. At 3rd week age feed intake is less in To than T1 and T2. But at 4th and 5th week age feed intake is more less in T1 and T2 compare to To.

Table 3.3. Weekly feed intake

parameter	treatment			P value	Level of significance
	To	T1	T2		
feed intake at 1st week	18.374[a]	17.177[a]	16.898[a]	0.3717	NS
feed intake at 2nd week	45.768[a]	36.012[b]	40.419[ab]	0.0581	NS
feed intake at 3rd week	70.540[a]	75.839[a]	78.063[a]	0.7420	NS
feed intake at 4th week	93.514[a]	79.492[b]	78.032[b]	0.0515	NS
feed intake at 5th week	111.940[a]	95.384[a]	99.163[a]	0.2802	NS

Note:[a,b] in a row with no shared superscripts deviate significantly ($p<0.05$). The data is presented as an average of three replicate groups of seven birds each. (n=21). * TO =Basal diet, T1 = Basal diet+0.5% neem powder, T2=Basal diet+1% neem powder ** = Significant, NS = Not Significant.

3.4 FCR: Effect of neem powder on FCR value are shown in table 4. At 1st and 3rdweek age there have no significant effect of neem powder on FCR. At 2nd and 4th week age there have highly significant effect of neem powder on FCR where T1 and T2 is less than To. Similarly, at 5th week age the value is significant.

Table3.4. Weekly FCR (feed conversion ratio)

parameter	treatment			P value	Level of significance
	To	T1	T2		
FCR at 1st week	1.4496[a]	1.3297[a]	1.6517[a]	0.1774	NS
FCR at 2nd week	2.02722[a]	1.41780[c]	1.77167[b]	0.0012	**
FCR at 3rd week	2.8651[a]	2.1725[b]	2.3658[ab]	0.0544	NS
FCR at 4th week	1.9627[a]	1.3895[b]	1.4711[b]	0.0058	**
FCR at 5th week	1.8634[a]	1.4423[b]	1.4197[b]	0.0307	*

Note:[a,b,c] in a row with no shared superscripts deviate significantly ($p<0.05$). The data is presented as an average of three replicate groups of seven birds each. (n=21). * TO =Basal diet, T1 = Basal diet+0.5% neem powder, T2=Basal diet+1% neem powder ** = Significant, NS = Not Significant

3.5 Overall growth performance: In table 5 shows the overall growth performance of broiler till 35 days of trial from day 1. The result revealed that neem powder has no significant effect on initial body weight and feed intake. But the diet has significant effect on final weight, weight gain

and FCR. Significantly higher ($p<0.05$) body weight gain and FCR were observed in neem leaf powder supplemented group compare to control.

Table 3.5. Overall growth performance of broiler

parameter	treatment				
	To	T1	T2	P value	Level of significance
Initial body wt	44.1867^{a}	44.1333^{a}	44.3267^{a}	0.4284	NS
Final body wt	1219.42^{b}	1421.96^{a}	1427.50^{a}	0.0040	**
Total body wt gain	1175.23^{b}	1377.82^{a}	1383.17^{a}	0.0040	**
Total feed intake	2381.0^{a}	2127.3^{a}	2188.0^{a}	0.1098	NS
Overall FCR	2.02737^{a}	1.54296^{b}	1.58304^{b}	0.0012	**

Note: ^{a,b,c} in a row with no shared superscripts deviate significantly ($p<0.05$). The data is presented as an average of three replicate groups of seven birds each. (n=21). * TO =Basal diet, T1 = Basal diet+0.5% neem powder, T2=Basal diet+1% neem powder ** = Significant, NS = Not Significant

3.6 Apparent nutrient digestibility of digesta:

The digestibility of ileal digesta shows table 3.6. Ingestion of neem powder with fed significantly influenced the percentage of ash and crude protein, although neem powder had no significant effect on the dry matter, organic matter and ether extract digestibility percentage. Here highest CP digestibility value was observed in T2 group and highest ash value in T1 group compared with control group To.

Table 3.6. Apparent nutrient digestibility of Digesta sample

parameter	treatment			P value	Level of significance
	To	T1	T2		
Dry matter	96.6475[a]	97.7264[a]	97.2453[a]	0.3013	NS
OM	83.117[a]	87.949[a]	89.227[a]	0.1129	NS
Ash	58.881[b]	73.992[a]	80.944[a]	0.0306	*
Crude Protein	73.010[b]	86.351[a]	80.699[ab]	0.0406	*
Ether extract	81.085[b]	91.695[a]	87.669[ab]	0.0665	NS

Note: [a,b,c] in a row with no shared superscripts deviate significantly ($p<0.05$). The data is presented as an average of three replicate groups of seven birds each. (n=21). * TO =Basal diet, T1 = Basal diet+0.5% neem powder, T2=Basal diet+1% neem powder ** = Significant, NS = Not Significant

Broilers growth performance was greatly improved ($p< 0.05$) in the current study by adding graded levels of NLP to their diet. Neem leaf powder can be used as a natural feed supplement at a reasonable price to commercial broiler chicken diets to improve overall performance. The FCR shows that the efficiency with which birds convert feed to live weight increases with decreasing value (Ampodeet al., 2020).Broiler chickens fed with 1% neem powder (T2) had the lowest (better) cumulative feed conversion ratio (FCR) as compared to other treatments. It should be emphasized that FCR assesses how effectively livestock and poultry transform animal feed into the desired output. We also observe that body weight gain are better in T1 and T2 group than To group. The overall growth performance is excellent in T1 and T2 group than To. Neem leaves have anti-protozoal and immunostimulatory qualities that may contribute to improvements in weight increases and broiler chicken performance by lowering the microbial load (Wankaret al., 2009; Kharde& S. Soujanya, 2014).

DM, OM, CP, EE, and Ash digestibility in chicken feed diets supplemented with probiotics and combinations of probiotic and phytogenic feed may be due to these agents' ability to increase the amount of nutrients available for absorption and animal growth by inhibiting the growth and metabolic activities of the gut microflora as well as altering intestinal growth, morphology, and function, such as decreasing intestinal epithelium thickness (Barton, 2000; Miles et al., 2006).

Conclusion

Neem can be thought of as a therapeutic plant based on earlier research and the findings of this study. The addition of 0.5% to 1% neem powder to broiler diets improves performance and provides a frame with financial gain. Thus antibiotics should not be used in boilers; Neem leaf meal might be used instead.

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