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Effect of *Moringa oleifera* Leaf Powder as an Alternative to Antibiotic Growth Promoters on Growth Performance, Meat Value, Haemato-Biochemical Indices for Sustainable Sonali Chicken (RIR × Fayoumi) Production

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Abstract

This study investigated the effects of supplementing *Moringa oleifera* leaf powder (MLP) on the growth performance, meat quality and health status in Sonali chicken. A 60-day feeding trial was conducted using 144 day-old male Sonali chicks, which were randomly allocated into three dietary treatments with eight replicates per treatment and six birds per replicate. The treatments consisted of: (i) control group fed a basal diet, (ii) antibiotic group fed the basal diet supplemented with chlortetracycline at 100 mg/kg feed, and (iii) MLP group fed the basal diet supplemented with MLP at 500 mg/kg feed. The MLP-supplemented group showed higher ($P < 0.05$) final body weight (FBW) and average daily weight gain (ADG) in the whole experimental period (1-60 days) with improved feed conversion ratio. Analysis of the meat quality revealed that dietary supplementation with MLP significantly improved the meat quality of Sonali chickens by increasing postmortem muscle pH at 45 min and reducing drip loss at 24 h and 8 days compared with the control diet ($P < 0.05$). Hemoglobin (12.4 g/dL in MLP vs. 11.9 g/dL in control), red cell distribution width-standard deviation (56.4 fL in MLP vs. 44 fL in control), and total white blood cell count (11,700/µL in MLP vs. 9,300/µL in control) were all significantly higher *($P < 0.05$)* according to hematological parameters, indicating the enhanced immunity of Sonali chickens. MLP supplementation improved serum calcium, albumin, and lipid profiles by lowering total cholesterol and triglycerides and significantly decreased ($P < 0.05$) serum glutamic pyruvic transaminase levels, indicating improved liver function. These results prove that MLP is a powerful natural feed additive that enhances Sonali chicken's growth, meat quality and health, providing a suitable substitute for antibiotic growth promoters in sustainable poultry production.

Introduction

The poultry sector is essential for maintaining food security and economic growth in Bangladesh. Chicken meat is popular and suitable for all age groups due to its low-fat level, and its appeal is not limited to any particular demographic. However, the

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sector encounters difficulties like growing production costs and reliance on antibiotic growth promoters (AGPs). According to recent estimates, antibiotic resistance may cause roughly 10 million deaths worldwide by 2050 (De Kraker *et al*., 2016). However, on January 1, 2006, European nations outlawed the use of in-feed antibiotics as growth promoters due to public health and antibiotic resistance concerns (Chakma *et al*., 2020), and on January 28, 2010, Bangladesh followed suit (Bangladesh Parliament, 2010). Researchers have therefore stepped up their efforts to find natural alternatives to AGPs, with a special emphasis on phytobiotics as sustainable replacements.

Moringa oleifera is one of the potential natural plant alternatives to pawpaw, neem, bamboo, and turmeric that are being researched (Ogbuewu *et al*., 2022; Oloruntola *et al.*, 2019). According to Zvinorova *et al*. (2015), this drought-tolerant plant is referred to as the "Miracle Tree" and has shown promise as a treatment for more than 300 illnesses (Afrianto & Metananda, 2023). Moringa is native to the Indian subcontinent and is a member of the Moringaceae family, and it is referred to locally as Sajna or Sajina in Bangladesh (Islam *et al*., 2021). Due to their remarkable nutritional profile and possible therapeutic advantages for both people and animals, the leaves are very beneficial in poultry diets (Soundararajan *et al.,* 2023). Moringa leaves have been found to be abundant in essential amino acids, high-quality protein, minerals (calcium, iron, potassium, and phosphorus), vitamins (A, D, and E), and energy sources (Islam *et al.,* 2021; Liang *et al*., 2019). The amino acid content of moringa leaf was presented from our previous work (Akib *et al.*, 2024). The results indicated that *M. oleifera* leaves contain essential amino acids including leucine (1.75 mg/100 g), phenylalanine (1.43 mg/100 g), arginine (1.07 mg/100 g), threonine (1.07 mg/100 g), lysine (1.03 mg/100 g), isoleucine (0.95 mg/100 g), valine (0.89 mg/100 g), and methionine (0.34 mg/100 g). Moreover, these leaves contain bioactive substances, including rutin, alkaloids, kaempferol, quercetin, catechin, and chlorogenic acid that possess antimicrobial and antioxidant values (Abdalla *et al*., 2022; Prabakaran *et al.*, 2018). These substances have expressed effectiveness in lowering inflammation, preventing damage from free radicals, and improving the general health of birds (Vergara-Jimenez *et al*., 2017). According to recent research, *Moringa oleifera* has a wide range of medicinal benefits, including defence against liver damage, diabetes, cancer, inflammation, and hypertension (Bidura *et al.*, 2020; Chiș *et al*., 2023). One of the most notable compounds in Moringa is 'Pterygospermin', which has significant antifungal and antibacterial activities and breaks down into benzyl isothiocyanate, indicating that it may be used as a natural health stimulant in chickens (Fahey, 2005). Research has demonstrated that supplementing poultry with Moringa oleifera improves their growth performance, blood profile, antioxidant status, and meat quality while lowering gut infections (Mahfuz & Piao, 2019; Soundararajan et al., 2023). Additionally, Ahmed and El-Rayes (2019) noted that Japanese quail fed diets with 7 g/kg of Moringa leaf powder performed better and had better nutrient digestibility. Moringa oleifera may also be used as a treatment for cardiovascular disorders due to its hypolipidemic and hypocholesterolemic characteristics (Abu Hafsa et al., 2020; Gasmalbari et al., 2020). Consumers can benefit from higher levels of polyunsaturated fatty acids (PUFA) in chicken meat when MLP is added to poultry diets (Cui et al., 2018; Selim et al., 2021). Among the poultry, Sonali chickens, a hybrid variation that is popular in South Asia, have drawn notice due to their excellent meat quality, ability to produce more eggs, and easily adapt in local environments (Abbas, 2013 ; David *et al.,* 2015). Demand for Sonali, a crossbreed of Fayoumi hens and Rhode Island Red cocks has increased, rising from 20% in July 2018 to 45% in July 2019 (Jaman *et al.*, 2024).

Despite the promising potential of *Moringa oleifera*, wide uses in rations and precise mechanisms of action in poultry remain unclear (Mahfuz & Piao, 2019). Research has shown that inclusion rates exceeding 25 g Moringa leaf meal/kg diet may adversely affect broiler chicken growth (Nkukwana *et al.,* 2014). Previous research has been conducted on *Moringa oleifera* leaf powder in poultry diets using doses of 0.5% (500 mg/kg), 1% (1 g/kg), 3% (3 g/kg) and 7% (7 g/kg) by Abu Hafsa *et al.* (2020), Alwaleed *et al.* (2020), Ahmed and El-Rayes (2019) respectively. Considering the findings of previous investigations and economic aspects, a single lower dosage of *Moringa oleifera* leaf powder at 500 mg/kg diet (0.5%) was selected as the main treatment in this experiment. However, there has been no investigation into the effect of MLP supplementation on Sonali chickens. Therefore, this study aims to investigate the effects of low-concentration MLP supplementation on growth performance, meat quality, hemato-biochemical parameters, and physiological aspects in Sonali chickens, addressing the current knowledge gaps in optimal utilization of this promising natural feed additive.

Materials and Methods

Design, Nutrition and Supervision of Experiments

The Animal Care and Use Committee of Sylhet Agricultural University, Sylhet, Bangladesh, authorized and supervised the study (No. AUP202326), which was conducted at the poultry research shed using a cage rearing system. A total of 144 one-day-old male Sonali chicks, with an average

body weight of 28.96 g, were randomly allotted to three dietary treatments: (i) control group fed a basal diet, (ii) antibiotic group fed the basal diet supplemented with chlortetracycline at 100 mg/kg feed, and (iii) MLP group fed the basal diet supplemented with MLP at 500 mg/kg feed. Each treatment consisted of eight replicates, with six birds per replicate. For management purposes, each replicate was housed in two cages (pens), each containing three birds. Each cage measured 80 cm × 47 cm × 45 cm (length × width × height). All birds had free access to feed and clean drinking water throughout the 60-day experimental period. The nutritional requirements of Sonali chickens were met through phase feeding by dividing the experimental period into two dietary phases: starter (1–30 days) and grower (31–60 days) (Table 1). A home with a controlled temperature was used to grow chickens. The Sonali chicken shed's temperature mainly was maintained at 37°C for the purposes of brooding, but until it was set at 25°C, it gradually dropped by 3°C each week. To aid in brooding in the shed, 50% relative humidity was first placed on day 1 and progressively grew by 5% each week until it stood at 70%. In order to avoid direct sunlight, the research shelter was a case-system house oriented east-west. On day three, the Sonali chickens received the Baby Chick Ranikhet Disease Vaccine (BCRDV), and on day twenty-three, they received the Ranikhet Disease Vaccine (RDV); on day nine, they received the Infectious Bursal Disease Vaccine (IBDV), and on day seventeen, they received a booster dose.

Table 1: Nutritional composition and ingredients of basal diets[1]

Ingredients (%)	Starter (1-30 day)	Grower (31-60 day)
Maize	59.37	60.31
Soybean meal	11.69	9.39
Rice polish	1.1	1.5
Animal protein concentrate	6	6.9
Full Fat Soya	18.2	18.6
Di-calcium phosphate[2]	1.49	1.57
Limestone	1.2	1.18
Salt	0.3	0.3
Lysine-L[2] (98%)	0.01	0.01
Methionine [2] (98%	0.13	0.13
Threonine[2] (98%)	0.01	0.01
Premix[3]	0.5	0.1
Analyzed value (%)		
Moisture	12	12
Crude Protein	19.13	17.62
Ash	5.25	5.26
Crude fat	3.98	3.98
Calcium	0.95	0.95
Av. Phosphorus	0.45	0.45
Calculated Value (%, unless mentioned)		
ME (Kcal/kg)	2810	2960
Methionine	0.45	0.45
Lysine	1	1
Threonine	0.72	0.72

[1]Each basal meal was well combined with a premix containing 500 mg/kg of MLP and 100 mg/kg of CTC, an antibiotic.
[2]Available in commercially.
[3]Composition per kg: Vitamins: Vitamin A, 50000 IU; Vitamin D3, 10000 IU; Vitamin E, 60 mg; Vitamin C, 300 mg; Vitamin K3, 3 mg; Vitamin B1, 5 mg; Vitamin B2, 10 mg; Vitamin B4, 5 mg; Vitamin B12, 300 mcg; Minerals: Copper sulphate, 7 mg; Cobalt sulphate, 60 mg; Ferrous sulphate, 50 mg; Magnesium sulphate, 35 mg; Zinc sulphate, 60 mg; Sodium Chloride, 15 gm; Potassium iodide, 200 mg; Manganese, 35 mg; Calcium carbonate, 100 gm; Di calcium phosphate, 100 gm; Amino acids: L-Lysine, 2.5 gm; DL Methionine, 10 gm; Choline Chloride, 25 mg; Glycine, 20 mg; Enzymes: Cellulase, 15000 IU; Amylase, 20000 IU; Protease, 15000 IU; Xylanase, 10000 IU; Prebiotic: Dextran oligosaccharides, 200 mg; Probiotic and Yeast: Lactobacillus acidophilus, 25 billion CFU; Bacillus subtilis, 50 billion CFU; Saccharomyces cerevisiae, 5 billion CFU; Selenium yeast, 0.10 mg; Fatty acid: 15% (Vety Faster, Vetymed Agro Pharma, Dhaka, Bangladesh).

Manufacturing and Chemical Evaluation of Powdered Moringa Leaves

From the nearby area, *Moringa oleifera* leaves in various states of maturity were gathered, sorted, and thoroughly cleaned. To reduce moisture, the collected leaves were spread out in the shade and turned frequently for three days. To make the semi-dried leaves brittle and suitable for grinding, the leaves were oven-dried for 30 min at a constant temperature of 30 °C. A blender was used to grind the leaf into powder. In accordance with the standards established by the Association of Official Analytical Chemists (AOAC), the MLP specimens were examined in triplicate. Crude protein (CP) was measured by

employing the Kjeldahl technique; the crude fat (EE) content of petroleum ether samples was extracted by applying a Soxhlet system, and the amount of ash was assessed by heating the sample to 600°C for six hours. Inductively coupled plasma (ICP), optical emission spectroscopy, and phosphorus and calcium estimates were employed. Dry matter is subtracted from 100 to determine the moisture content. To compute organic matter (OM), the anticipated ash value and moisture were subtracted from 100. The chemical composition of Moringa leaf powder was presented in Table 2.

Equation 1

$$Organic\ Matter\ \% = 100 - (Moisture\ \% + Ash\ \%)$$

Equation 2

$$\text{Nitrogen} - \text{free extract} = \text{Dry matter} - (\text{Crude protein} + \text{Crude fiber} + \text{Ether extract} + \text{Ash})$$

Table 2: Chemical composition of *Moringa oleifera* leaves powder (g/100 g).

Proximate components	Amount
Dry Matter	91.26
Ash	9.20
Crude Protein (CP)	26.58
Crude Fiber (CF)	7.92
Crude Fat	4.51
Organic matter (OM)	82.06
Fatty acid	2.06
Nitrogen Free Extract (NFE)	43.05
Total Phosphorus	0.439
Calcium	0.072

Development and Growth of Sonali Chicken

The experimental Sonali chickens' average daily feed intake (ADFI), average daily body weight gain (ADG), and feed conversion ratio (FCR) were determined for each dietary group. ADFI is known as the difference between the amount of feed given and the summation of the leftover feed and feed waste, divided by the total number of birds and the total number of experimental days. ADG is calculated as the difference between the total weight of birds at the end of a given period and their total weight at the beginning of that period, divided by the total number of birds and the total duration (days). FCR is measured as the ratio between the average daily feed intake and the average daily gain.

Hemato-biochemical Parameters

On day sixty, blood samples were taken for hemato-biochemical examination in the evening. After selecting one Sonali chicken at random from each replicate (8 birds per treatment), two aliquots of blood specimens were taken from the vein in its wing. In accordance with standard hematological procedures, the initial sample was collected in a tube that included EDTA (an anticoagulant) for assessing hematological parameters including the number of red blood cells (RBCs), hemoglobin (Hb) levels, packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular hemoglobin concentration (MCHC), red cell distribution width-coefficient of variation (RDW-CV), total leukocyte counts (WBCs), and differential leukocyte counts (DLC). However, the second blood aliquot was left to clot after being drawn without the use of an anticoagulant. Following separation, the resultant serum was kept for subsequent examination at -20°C. Using commercial test kits, blood biochemical parameters (creatinine, total cholesterol, serum glutamic pyruvic transaminase, and triglycerides) were evaluated.

Quality Assessment of Meat

Twenty-four (24) birds were sacrificed (8 birds per treatment) at the age of 60 days. A probe equipped with an electrode from a portable pH meter (Orion model 301; Orion, Beverly, MA, USA) was applied to evaluate the pH values of the breast muscle at 45 minutes and 24 hours after slaughter. With the help of pH 4 and pH 7 calibration buffers, the probe was calibrated before measurement. Each sample's mean values were regarded as the final value once readings were obtained from at least three different sites.

To determine drip loss, the method described by Honikel (1998) was followed. Meat samples (5-8 g) were trimmed, weighed, and placed in inflated sample bottles, which were then hung at 4 °C for 72 h without contacting the sides or bottom of the bottles. After hanging, the samples were wiped with filter paper and reweighed, and the percentage of drip loss was calculated from the difference in weight before and after hanging.

A sample of 100 g of breast meat was cut and placed in a plastic bag to measure cooking loss. The sample's internal temperature afterwards ascended to 75°C for five minutes in a chamber filled with water. Water was emptied from the bag, and samples were taken out of the water bath after cooking. The specimens were then immediately cooled under running water to room temperature. Before the cooked sample was finally weighed, excess surface moisture was dried with paper towels. Using the following formula, cooking loss was determined (Vargas-Ramella *et al.*, 2022).

$$\text{Cooking loss \%} = \frac{weight\ of\ fresh\ meat - weight\ of\ cooked\ meat}{weight\ of\ fresh\ meat} \times 100$$

Statistical Analysis

The data were initially tested for normality of residuals (Shapiro-Wilk) and homogeneity of variances (Bartlett). Data were analyzed by one-way analysis of variance (ANOVA) using SPSS software (Version 26.0; IBM Corp., Armonk, NY, USA).

When significant differences were detected, Duncan's multiple range test was applied for mean separation. Statistical significance was declared at $P < 0.05$.

Results

Growth Performance

Table 3 delineates the influence of MLP on the growth efficacy of Sonali chicken. The MLP-fed group revealed a significantly higher ($P < 0.05$) FBW when compared with the control group, though the IBW had no significant difference among dietary groups. In addition to the entire research period (day 1–60) and the various trial days (day 1-15, day 16-30, day 31-45 and day 46-60), there was no significant difference ($P > 0.05$) in ADFI among the different dietary groups. ADG was noted substantially higher ($P < 0.05$) in the MLP-supplemented group in the whole experimental period (day 1-60) than the other two groups. FCR was significantly lower in the MLP group than the control during days 46-60, and over the entire experimental period (days 1-60), both the MLP and antibiotic groups exhibited lower FCR than the control ($P < 0.05$).

Meat Quality

The Impact of MLP on the flesh quality of Sonali chicken was demonstrated in Table 4. Muscle pH at 45 min postmortem was significantly higher ($P < 0.05$) in both the MLP and antibiotic groups compared with the control, whereas pH at 24 h showed no significant differences ($P > 0.05$). Drip loss at 24 h and 8 days was significantly lower ($P < 0.05$) in the MLP-supplemented group than in the other groups, while cooking loss did not differ significantly among the dietary treatments.

Hemato-biochemical Parameters

Table 5 illustrates the dietary significance of MLP on the Hemato-biochemical parameters of Sonali chicken. Hb, RDW-SD, RDW-CV and total WBC were noted to be greater ($P < 0.05$) in the MLP-fed group compared to the control and antibiotic groups. Interestingly, other blood parameters showed no significant variation ($P > 0.05$) among the dietary groups.

Table 3: Effects of experimental treatment on growth performance of Sonali chicken.

Measures	Age(days)	Control	Antibiotic	MLP	SEM	p-value
IBW	1st	28.43	29.23	29.23	0.54	0.686
FBW	60th	628.33^{b}	662.77^{ab}	752.20^{a}	24.5	0.041
ADG (g/day/bird)	1-15	3.36	2.96	3.25	0.09	0.251
	16-30	7.95	7.43	8.02	0.32	0.757
	31-45	8.05	13.03	9.78	1.10	0.185
	46-60	20.63	18.82	27.15	1.80	0.131
	1-60	10.00^{b}	10.56^{b}	12.05^{a}	0.4	0.041
ADFI (g/day/bird)	1-15	10.01	10.13	10.22	0.12	0.839
	16-30	26.03	28.64	29.04	1.09	0.538
	31-45	35.96	29.71	34.40	1.42	0.184
	46-60	66.02	51.73	57.11	3.344	0.228
	1-60	34.51	30.06	32.69	0.83	0.063
FCR	1-15	3.00	3.43	3.18	0.12	0.414
	16-30	3.32	3.96	3.68	0.26	0.678
	31-45	4.50	2.32	3.51	0.54	0.132
	46-60	3.22^{a}	2.75^{ab}	2.15^{b}	0.19	0.040
	1-60	3.46^{a}	2.87^{b}	2.71^{b}	0.13	0.018

^{a,b,c}show that the difference is significant at $P < 0.05$; SEM stands for pooled standard error of the means; IBW, Initial Body Weight; FBW, Final Body Weight; ADG: Average Daily Weight Gain; ADFI, Average Daily Feed Intake; FCR, Feed Conversion Ratio; Antibiotic, chlortetracycline, at 100 mg/kg; *Moringa oleifera* leaf powder (MLP) at 500 mg/kg basal diet.

Table 4: Effects of experimental treatment on meat quality of Sonali chicken.

Items		Control	Antibiotic	MLP	SEM	*P*-value
pH	45 mins	5.96^{b}	6.12^{a}	6.05^{a}	0.07	0.039
	24 hrs	5.69	5.77	5.84	0.03	0.193
Drip loss%	24hrs	2.57^{a}	2.82^{a}	1.65^{b}	0.22	0.01
	8th day	4.49^{a}	4.64^{a}	2.85^{b}	0.33	0.022
Cooking Loss %		22.30	23.26	20.31	1.48	0.746

n=8 samples in each treatment. SEM stands for pooled standard error of the means; ^{a,b} implies a significant difference at $P < 0.05$. Antibiotic, chlortetracycline, at 100 mg/kg; *Moringa oleifera* leaf powder (MLP) at 500 mg/kg basal diet.

Serum Metabolic Profile

The role of MLP supplementation on the serum metabolic profile of Sonali chicken was displayed in Table 6. Higher ($P < 0.05$) serum calcium and serum albumin were noted in the MLP-fed group than in the other groups. Alongside, SGPT was identified as significantly lower ($P < 0.05$) in the MLP-fed group compared to the control and antibiotic groups. However, serum urea -N was not affected by the dietary groups.

Table 5: Effects of experimental treatment on hematological parameters of Sonali chicken.

Test parameters		Control	Antibiotic	MLP	SEM	*P*-value
Hb (g/dL)		11.9[b]	11.7[b]	12.4[a]	0.62	0.021
RBC (Million/uL)		2.65	2.7	2.6	0.05	0.651
PCV (%)		33.9	34.2	33.6	2.3	0.331
MCV (fL)		128	130	130	8.2	0.559
MCH (pg)		47.6	46.3	48.1	4.6	0.572
MCHC (gm/dL)		36	36	37	1.98	0.773
RDW-SD (fL)		44[b]	46[b]	56.4[a]	3.33	0.03
RDW-CV (%)		9.2[b]	9.6[b]	12[a]	0.231	0.045
Total platelet count (k/cumm)		198	215	220	23.11	0.734
Total WBC ($\times 10^3/\mu L$)		9300[c]	10200[b]	11700[a]	24.27	0.044
	Heterophil %	28	27	28	1.23	0.77
Differential	Lymphocyte %	67	68	66	1.3	0.643
leucocyte count	Monocyte %	3	3	3	0.07	0.887
(DLC)	Eosinophils%	2	2	3	0.01	0.912
	Heterophil/Lymphocyte	0.42	0.40	0.42	0.06	0.732

n=24 samples in each treatment. SEM stands for pooled standard error of the means; [a,b,c] imply a significant difference at $P < 0.05$.; Hb, Hemoglobin; RBC, Red Blood cells; PCV, Packed cell volume; MCH, Mean corpuscular hemoglobin; MCV, Mean corpuscular volume; MCHC, Mean corpuscular hemoglobin concentration); RDW-SD, Red cell distribution width-standard deviation; RDW-CV, Red cell distribution width-coefficient of variation; WBC, White blood cells; Antibiotic, chlortetracycline at 100 mg/kg; *Moringa oleifera* leaf powder (MLP) at 500 mg/kg basal diet.

Table 6: Effects of investigational treatment on the serum metabolic profile of Sonali chicken.

Parameters	Control	Antibiotic	MLP	SEM	*P*-value
Serum calcium (mg/dL)	8.9[b]	9.0[b]	9.9[a]	0.43	0.031
Serum albumin (mg/dL)	3.2[b]	3.5[b]	4.4[a]	0.22	0.022
Serum Urea -N (mg/dL)	18	18	19	3.15	0.642
SGPT/ALT (U/L)	29[a]	32[a]	18[b]	3.46	0.001

n=8 samples in each treatment; SEM stands for pooled standard error of the means; [a,b] implies a significant difference at $P < 0.05$; SGPT (Serum glutamic pyruvic transaminase); Antibiotic, chlortetracycline at 100 mg/kg; *Moringa oleifera* leaf powder (MLP) at 500 mg/kg basal diet.

Lipid Profile

Table 7 elucidates the MLP feeding effect on the lipid profile of Sonali chicken. TC, LDLC, and TG were significantly lower ($P < 0.05$) in the MLP-fed group when compared to the control and antibiotic groups. Conversely, no significant alteration ($P > 0.05$) was demonstrated in HDLC among the groups.

Table 7: Effects of experimental treatment on the lipid profile of Sonali chicken.

Parameters	Control	Antibiotic	MLP	SEM	*P*-value
TC, mg/dL	182[a]	184[a]	178[b]	6.32	0.033
HDL-C, mg/dL	42	41	38	2.21	0.088
LDL-C, mg/dL	101[a]	104[a]	91[b]	3.34	0.044
TG, mg/dL	189[a]	191[a]	177[b]	3.55	0.027

n=8 samples in each treatment; SEM stands for pooled standard error of the means; [a,b] implies a significant difference at $P < 0.05$; TC, Total cholesterol; LDL-C, Low-Density lipoprotein cholesterol; HDL-C, High-density lipoprotein cholesterol; TG, Triglyceride; Antibiotic, chlortetracycline at 100 mg/kg; *Moringa oleifera* leaf powder (MLP) at 500 mg/kg basal diet.

Discussion

Sustainable poultry production is essential for combating world hunger and advancing health to supply high-quality protein and micronutrients through meat and eggs (Khan *et al.*, 2023; Khan *et al.*, 2020). This study investigates how supplementing *Moringa oleifera* leaf powder (MLP) affects the growth performance, meat quality, hemato-biochemical characteristics, and lipid profile of Sonali chickens. The average daily feed intake did not differ substantially between dietary groups, which was in line with the findings of Khan *et al*. (2017) and Lu *et al.* (2016). Alabi *et al.* (2017) reported the opposite, indicating that MLP supplementation led to a decline in feed intake. On the other hand, Akib *et al*. (2024) noted that broilers fed MLP consumed more feed, which they attributed to bioactive substances such as moringinine and flavanol glycosides (Mbikay, 2012) that improved appetite, glucose homeostasis, and the generation of digestive enzymes. The MLP-fed group exhibited a significantly higher average daily gain (ADG) compared with the control and antibiotic groups, and a higher final body weight than the control group.

Similar results were observed by Abu Hafsa *et al*. (2020) and Alwaleed *et al.* (2020) with different MLP inclusion levels (0.5%, 1%, 3%). The MLP-fed group exhibited the lowest feed conversion ratio (FCR) during days 46-60, and over the entire experimental period (days 1-60), FCR was lower in both the MLP-fed and antibiotic groups compared with the control group, consistent with the findings of Agashe *et al*. (2017). Bioactive substances that promote nutrition utilization are probably the cause of improved production performance (Khan *et al.,* 2024). The improved BWG and reduced FCR in the MLP-supplemented group may be due to its highly digestible proteins and essential amino acids, bioactive phytochemicals such as moringinine and flavonoids that enhance digestive enzyme activity, and antimicrobial polyphenols that improve gut health and nutrient absorption.

MLP supplementation had a good impact on meat quality, with a considerably higher initial breast muscle pH (45 minutes post-mortem) than the control, supporting the findings of Rehman *et al*. (2018). Moringa's high alkaline mineral content (Ca, Mg, and K) and alkaloid chemicals like moringinine are probably the cause of this higher pH (Mbikay, 2012; Sultana, 2020). According to our research, MLP-supplemented meat had a much-reduced drip loss percentage. This could be because antioxidants such as ascorbic acid, phenolics, flavonoids, and carotenoids improved the meat's ability to hold water (Mahfuz & Piao, 2019; Peñalver *et al.,* 2022). However, Cui *et al*. (2018) found no positive impacts of supplementing with Moringa on cooking or drip loss.

According to Oyeagu *et al.* (2019), the hemato-biochemical profile is a reliable indicator of broiler health and offers information on the effectiveness of feed additives (Matshogo *et al*., 2020). Hemoglobin (Hb) levels were considerably boosted in this study with MLP supplementation. These results are consistent with those of Abbas *et al.* (2018) and Akib *et al*. (2024), who found that broilers fed with Moringa leaf meal had higher Hb and RBC levels. MLP's high iron (Fe) content could improve oxygen delivery and storage by playing a crucial role in hemoglobin and myoglobin synthesis (Elbashier & Ahmed, 2016). The MLP-fed group had higher red cell distribution width-standard deviation (RDW-SD) and total white blood cell (WBC) count, which may correlate with erythropoiesis and immunological function. This observation aligns with Zelin (2018) but differs from Marupanthorn *et al.* (2024), who reported that the total WBC was lower in the group fed MLP. The immunomodulatory potential of MLP is shown by the presence of normal levels of neutrophils, eosinophils, basophils, monocytes, and lymphocytes that indicate a balanced immunological response and normal physiology of Sonali chickens.

The considerably higher serum calcium levels in the MLP-fed group when compared to the control and antibiotic groups suggest that *Moringa oleifera* may also have a positive effect on calcium metabolism. The high mineral content of Moringa leaves, which are abundant in accessible nutrients, is probably the cause of rising Ca in the respective group of birds (Islam *et al*., 2021; Mbikay, 2012). Our study demonstrated a reduction in serum glutamic pyruvic transaminase (SGPT) levels in the MLP-fed group, indicating improved liver health and reduced hepatic stress (Jassawalla, 2024). This is consistent with previous studies by Verma *et al*. (2021) that have shown the bioactive compounds, flavonoids (quercetin) and carotenoids (β-carotene), positively affected and reduced the levels of SGPT in the serum, which showed improved liver performance. The flavonoids, polyphenols, and antioxidants in *Moringa oleifera* probably decreased oxidative stress that ensured the improved liver function, while elevated SGPT values are frequently suggestive of hepatic stress or injury (Dalei *et al*., 2016). The lower SGPT levels suggest that MLP can mitigate liver damage more effectively than antibiotics, which can be a significant finding for poultry health management. Enhanced protein metabolism is indicated by significantly greater blood albumin concentrations in the MLP-fed group. We hypothesized that dietary addition of MLP may enhance the albumin concentration in the serum of experimental Sonali chickens. The high-quality proteins and vital amino acids in Moringa leaves can improve protein synthesis (Dalei *et al*., 2016; Islam *et al*., 2021).

In this study, Sonali chickens fed with MLP showed lower total cholesterol (TC) and serum triglycerides (TG). This result is consistent with that of Abu Hafsa *et al*. (2020), who found that broilers fed Moringa leaves at 5% had significantly lower plasma TC and TG levels. Similarly, Essien *et al*. (2022) observed comparable results when Moringa leaf meal was added to finisher broiler chickens' diets. The high concentration of bioactive substances in MLP, such as flavonoids, polyphenols, sterols, and alkaloids, which are known to promote bile acid excretion and block lipogenic enzymes, is responsible for the lipid-lowering function (Ajantha *et al.,* 2020; Sharmin *et al*., 2021). Furthermore, MLP supplementation dramatically lowered the low-density lipoprotein and triglyceride levels in this investigation. According to Patil *et al*. (2010) and Sharmin *et al*. (2021), alkaloids may be involved in lowering cholesterol and encouraging the excretion of bile acids.

Conclusion

Moringa oleifera leaf powder (MLP) exhibits great promise as a natural feed supplement for Sonali chickens. This study ensured that dietary supplementation of Moringa leaf powder could

improve growth performances, including body weight, FCR, and overall health of Sonali chicken. MLP reduced serum cholesterol, triglycerides, and liver enzyme levels, suggesting improved metabolic and liver functions of Sonali chicken. Additionally, enhanced hemoglobin levels and improved blood biochemistry indicate better immunity and overall health. This study can open the door for the use of MLP as an efficient and sustainable feed additive to improve the growth performance and health of Sonali chicken.

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