



EFFECT OF *ACACIA NILOTICA* ON DIGESTIBILITY AND BIOCHEMISTRY OF BROILER CHICKEN

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Abstract

The aim of the research was to examine the impact of *Acacia nilotica* supplementation on numerous parameters related to digestibility, blood parameters, hormone levels, and intestinal length in broiler chickens. The digestibility of dry matter in the chickens was not significantly affected by different levels of *Acacia nilotica* supplementation. However, the digestibility of crude protein was significantly higher in the group supplemented with 3% *Acacia nilotica* compared to other groups. Similarly, the digestibility of metabolizable energy was significantly higher in the group supplemented with 6% *Acacia nilotica*. The digestibility of crude fat was significantly higher in the group supplemented with 12% *Acacia nilotica*. Regarding blood parameters, the hemoglobin level, Packed cell volume and white blood cells was significantly higher in the group supplemented with 12% *Acacia nilotica* compared to other groups. Red blood cell count was significantly higher in the 9% *Acacia nilotica* group. Cholesterol level was significantly lower in the 12% *Acacia nilotica* group. In terms of hormone levels, no significant differences were observed in growth hormone levels among the different groups. However, TSH level was significantly higher in the group supplemented with 6% *Acacia nilotica*. The length of the duodenum was significantly higher in the group supplemented with 6% *Acacia nilotica*, while the lengths of the jejunum and ileum were not significantly affected by treatments. These findings highlight the potential of *Acacia nilotica* as a dietary additive for broiler chickens, providing insights into its effects on nutrient utilization and physiological parameters.

Keywords: *Acacia nilotica*, broiler chickens, blood biochemistry, nutrients digestibility,

Introduction

In recent years, there has been growing interest in incorporating medicinal plants and their extracts into poultry diets as a natural approach to enhance nutrient digestibility. These plant-derived

substances often contain bioactive compounds that promote gut health and function, leading to improved nutrient availability and absorption. *Acacia nilotica*, a plant renowned for its therapeutic properties, has demonstrated the ability to enhance nutrient digestibility in certain livestock animals according to studies by Elsaid et al. (2012) and Abou-Elkhair et al. (2018).

Nutrient digestibility is a crucial aspect when evaluating the nutritional value of feed in poultry production. It specifically refers to the proportion of ingested nutrients that are absorbed and metabolized by the bird's body. This process significantly influences feed conversion efficiency and overall growth performance in poultry (Svihus, 2014). Nutrient digestibility plays a pivotal role in the growth performance, overall health, and productivity of poultry, especially broiler chickens (Ravindran, 2013). Various factors impact nutrient digestibility in poultry, including the type and quality of feed, the bird's age and health status, and the presence of specific feed additives such as enzymes, probiotics, and plant extracts (Kiarie et al., 2013). certain feed additives can enhance the digestibility of nutrients by altering the gut microflora, improving gut health, or increasing the solubility of nutrients (Adeola & Cowieson, 2011; Alagawany *et al.*, 2017).

The inclusion of medicinal plants and their extracts, such as *Acacia nilotica*, in poultry diets is an active area of research. These natural substances hold promise for enhancing nutrient digestibility by promoting gut health and function. As a result, they may contribute to improved growth performance and overall health in broiler (Hashemi & Davoodi, 2011).

Acacia nilotica, also known as the gum Arabic tree, babul, thorn mimosa, or prickly acacia, is a tree native to the African and Indian subcontinent. In traditional medicine, *A. nilotica* is widely used for its anti-inflammatory, antimicrobial, antidiabetic, and antidiarrheal properties (Singh et al., 2011). Recent interest has focused on incorporating herbal extracts into poultry feed due to their rich composition of bioactive compounds such as tannins, saponins, alkaloids, flavonoids, and other polyphenols (Abdel-Wareth & Lohakare, 2018). Plant-based feed supplements, such as *Acacia nilotica* leaf meal, offer various health benefits due to their nutrient-rich profiles. Existing literature covers the nutritional value of *Acacia nilotica* and the effects of similar leaf meal supplementation on the blood profile of poultry. highlighted the nutritional and medicinal properties of *Acacia nilotica*, including its richness in protein, vitamins, minerals, and essential amino acids. (Ali et al., 2004; Bhadoriya et al., 2012).

Moreover, the judicious use of these natural substances can offer an alternative to antibiotics used as growth promoters, which are gradually being phased out due to concerns over antibiotic resistance (Thacker, 2013; Daraji *et al.*, 2018).

Research indicates the potential of leaf meals in poultry diets. While leaf meals from various plant sources enhance nutritional value and growth performance (Abou-Elkhair et al., 2018) these studies are not specific to *Acacia nilotica*. Focused research is needed. Additionally, plant-based feed supplements can positively impact blood parameters (Ashraf et al., 2013). For example: Moringa oleifera leaf meal increased hemoglobin and serum protein levels in chickens. Neem (*Azadirachta indica*) leaf meal enhanced the hematological and biochemical profile of broilers. (Onibi et al., 2008; Ebrahim et al., 2018). These findings suggest the potential beneficial impacts of leaf meal supplementation, such as from *Acacia nilotica*, on the blood profile of chickens.

Materials and Methods

Experimental plan

A total of 300 broilers were used for this experiment. The birds were distributed in five groups named as T0, T3, T6, T9 and T12, respectively. Group T0 was kept as control, T3, T6, T9 and T12 groups were supplemented with 3, 6, 9 and 12% *Acacia nilotica*. The following experimental protocol was used for the study.

The following parameters were studied:-

Digestibility: At the end of experimental trial, feces (150g) from each group were collected. The fecal sample was grinded with the help of a grinder and sent to the Nutrition Division Directorate

Poultry Production and Research Sindh, Karachi for determination of moisture, crude protein, crude fat and ash.

1. Determination of Dry matter content (%): Moisture content was determined according to AOAC (2000). The sample weight was obtained. Values were calculated by the formula:

$$\text{Moisture (\%)} = \frac{\text{Weight of fresh sample} - \text{Weight of dry sample}}{\text{Weight of fresh sample}} \times 100$$

2. Determination of protein (%): The protein content of samples was determined by using micro Kjeldahl method as described by AOAC (2000). Protein content was estimated by using conversion factor as described by Hassan *et al.*, (2019) Later, nitrogen (%) is calculated by given formula.

$$\text{Nitrogen (\%)} = \frac{1.4 (V1-V2) \times \text{Normality of HCl} \times 250}{\text{wt. of sample}}$$

$$\text{Protein (\%)} = \text{N\%} \times \text{conversion factor (6.25)}$$

3. Determination of fat (%): Total fat content was extracted in Soxhlet Extraction Unit as described by AOAC (2000). Fat content was calculated by using the following formula:

$$\text{Fat (\%)} = \frac{W2 - W1}{W3} \times 100$$

Where,

W1 = weight of empty distillation flask

W2 = weight of distillation flask + fat

W3 = weight of sample taken

4. Determination of Ash: Five (05) gram sample was weighed and rapped into filter paper and then packed in cruble. Then transferred to a muffle furnace and ignited at 600⁰C to burn off all organic matter. After that, sample was kept in desiccator for cooling. The ash percentage was determined by following formula:-

$$\text{Initial weight of sample} - \text{final weight of sample} \times 100$$

Blood collection and analysis: 05 broilers from each group were randomly selected for collection of blood samples. The samples were taken into heparinized plastic tubes and blood plasma was separated by centrifugation at 250×g for 10 min at 4°C and stored at -20°C. Blood samples were collected from the wing vein of birds using a 3ml sterile syringe and placed into 1.5ml tubes containing anticoagulant (EDTA). To determine complete blood count by Hematological analyzer. Plasma cholesterol concentrations were measured by enzymatic method. Triiodothyronine (T3) and Thyroxin (T4) concentrations were determined by radioimmunoassay (RIA) using commercial kits. Plasma levels of alanine aminotransferase (ALT) and aspartate aminotransferase (AST) enzymes, which are markers of oxidative damage sustained in hepatic tissue, were measured using the corresponding diagnostic kits according to the manufacturer's protocols.

Small intestinal morphology examination: small intestine (duodenum, jejunum and ileum) was collected aseptically for measuring villus height.

The villi were measured and photographed under a microscope. Villus height was measured from top of the villus to the villus crypt junction and width in the wider area of the villus. The measurement was done with the image JPM software, USA (Khalita, 2021).

Statistical analysis: Statistical analyses were performed using JMP software, (SAS USA) and all data were expressed as means \pm SD values. Comparisons of the mean values were performed by one-way analysis of variance. Significant differences among means were evaluated by Tukey's comparison test at $P < 0.05$.

Results

Digestibility of dry matter (%): Figure 4.1 illustrates the outcomes regarding the impact of *Acacia nilotica* on the digestibility of dry matter in broiler chickens. The digestibility of dry matter in the broiler chickens was non-significant ($p > 0.05$) by varying levels of *Acacia nilotica* supplementation. The results showed that dry matter digestibility was determined as $92.11 \pm 0.33\%$ in T0 group, $91.96 \pm 0.34\%$ in T3 group, $92.23 \pm 0.09\%$ in T6 group, $91.92 \pm 0.26\%$ in T9 group and $91.14 \pm 0.21\%$ in T12 group. These groups were supplemented with 0%, 3%, 6%, 9%, 12% *Acacia nilotica*, respectively.

Digestibility of crude protein (%): Figure 4.2 illustrates the outcomes regarding the impact of *Acacia nilotica* on the digestibility of crude protein in broiler chickens. The crude protein digestibility in the broiler chickens was significantly ($p < 0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was a significantly higher crude protein digestibility ($79.65 \pm 0.75\%$) in the group denoted as T3, where the birds were supplemented with 3% *Acacia nilotica*, compared to the groups T0 ($78.80 \pm 0.86\%$), T6 ($78.32 \pm 0.67\%$), and T9 ($76.99 \pm 0.49\%$), these groups were supplemented with 0%, 6%, and 9% *Acacia nilotica*, respectively. Conversely, the T12 group, where the birds were supplemented with 12% *Acacia nilotica* exhibited a lower crude protein digestibility ($76.09 \pm 0.94\%$).

Digestibility of metabolizable energy (%): Figure 4.3 illustrates the outcomes regarding the impact of *Acacia nilotica* on the digestibility of metabolizable energy in broiler chickens. The metabolizable energy digestibility in the broiler chickens was significantly ($p < 0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was a significantly higher metabolizable energy digestibility ($91.26 \pm 0.72\%$) in the group denoted as T6, where the birds were supplemented with 6% *Acacia nilotica*, compared to the groups T0 ($90.36 \pm 0.59\%$), T3 ($90.12 \pm 1.61\%$), and T9 ($85.70 \pm 0.51\%$), these groups were supplemented with 0%, 3%, and 9% *Acacia nilotica*, respectively. Conversely, the T12 group, where the birds were supplemented with 12% *Acacia nilotica* exhibited a lower metabolizable energy digestibility ($77.49 \pm 0.94\%$).

Digestibility of crude fat (%): Figure 4.4 illustrates the outcomes regarding the impact of *Acacia nilotica* on the digestibility of crude fat in broiler chickens. The crude fat digestibility in the broiler chickens was significantly ($p < 0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was a significantly higher crude fat digestibility ($66.20 \pm 0.84\%$) in the group denoted as T12, where the birds were supplemented with 12% *Acacia nilotica*, compared to the groups T9 ($63.07 \pm 1.54\%$), T6 ($60.94 \pm 1.25\%$), and T3 ($60.90 \pm 0.72\%$), these groups were supplemented with 9%, 6%, and 3% *Acacia nilotica*, respectively. Conversely, the T0 group exhibited a lower crude fat digestibility ($56.88 \pm 1.23\%$).

Figure 4.1 Effect of *Acacia nilotica* on digestibility of dry matter (%) in broiler chicken

Variables	Group T0	Group T3 (6%)	Group T6 (6%)	Group T9 (9%)	Group T12 (12%)	P-value
Dry matter (%)	92.1158 ± 0.33534^a	91.9635 ± 0.34767^a	$92.2348 \pm 0.09299a$	$91.9253 \pm 0.26412a$	$91.148 \pm 0.0647a$	0.0647
Crude protein (%)	$78.8044 \pm 0.86606ab$	$79.6518 \pm 0.75378a$	$78.3281 \pm 0.67042ab$	$76.9968 \pm 0.49796ab$	$76.0922 \pm 0.94087b$	0.0211
Crude fat (%)	$56.8803 \pm 1.2394c$	$60.9047 \pm 0.7205bc$	$60.9431 \pm 1.2541bc$	$63.0758 \pm 1.5467ab$	$66.2048 \pm 0.8465a$	0.0002
Metabolized energy (%)	$90.3629 \pm 0.59a$	$90.1266 \pm 1.61a$	$91.26 \pm 4.30a$	$85.70 \pm 0.72b$	$77.49 \pm 0.94c$.0001

Different superscripts among the mean values are differed significantly from each other.

Hemoglobin (g/dl): Figure 4.5 illustrates the outcomes regarding the impact of *Acacia nilotica* on the hemoglobin level in broiler chickens. The hemoglobin level in the broiler chickens was significantly ($p<0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was significantly higher hemoglobin (13.48 ± 0.21 g/dl) in the group denoted as T12, where the birds were supplemented with 12% *Acacia nilotica*, compared to the groups T0 (12.85 ± 0.22 g/dl), T6 (12.66 ± 0.13 g/dl), and T9 (12.25 ± 0.19 g/dl), these groups were supplemented with 0%, 6%, and 9% *Acacia nilotica*, respectively. Conversely, the T3 group, where the birds were supplemented with 3% *Acacia nilotica* exhibited a lower hemoglobin level (12.24 ± 0.32 g/dl).

Packed cell volume (%): Figure 4.6 illustrates the outcomes regarding the impact of *Acacia nilotica* on the packed cell volume in broiler chickens. The packed cell volume in the broiler chickens was significantly ($p<0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was a significantly higher packed cell volume ($35.46\pm1.01\%$) in the group denoted as T12, where the birds were supplemented with 12% *Acacia nilotica*, compared to the groups T9 ($35.25\pm1.14\%$), T0 ($33.33\pm1.14\%$), and T6 ($31.50\pm1.13\%$), these groups were supplemented with 9%, 0%, and 6% *Acacia nilotica*, respectively. Conversely, the T3 group, where the birds were supplemented with 3% *Acacia nilotica* exhibited a lower packed cell volume ($30\pm1.15\%$).

Red blood cells ($\times 10^6\mu$): Figure 4.7 illustrates the outcomes regarding the impact of *Acacia nilotica* on the red blood cells in broiler chickens. The red blood cells in the broiler chickens was significantly ($p<0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was significantly higher red blood cells ($3\pm0.11 \times 10^6\mu$) in the group denoted as T9, where the birds were supplemented with 9% *Acacia nilotica*, compared to the groups T12 ($2.80\pm0.15 \times 10^6\mu$), T0 ($2.66\pm0.04 \times 10^6\mu$), and T6 ($2.45\pm0.07 \times 10^6\mu$), these groups were supplemented with 12%, 0%, and 6% *Acacia nilotica*, respectively. Conversely, the T3 group, where the birds were supplemented with 3% *Acacia nilotica* exhibited a lower red blood cells ($2.43\pm0.05 \times 10^6\mu$).

White blood cells (g/dl): Figure 4.8 illustrates the outcomes regarding the impact of *Acacia nilotica* on the white blood cells in broiler chickens. The white blood cells in the broiler chickens were significantly ($p<0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was a significantly higher white blood cells (12.82 ± 0.34 g/dl) in the group denoted as T12, where the birds were supplemented with 12% *Acacia nilotica*, compared to the groups T9 (12.64 ± 0.27 g/dl), T0 (12.54 ± 0.27 g/dl), and T6 (12.19 ± 0.28 g/dl), these groups were supplemented with 9%, 0%, and 6% *Acacia nilotica*, respectively. Conversely, the T3 group, where the birds were supplemented with 3% *Acacia nilotica* exhibited lower white blood cells (11.28 ± 0.24 g/dl).

Cholesterol (g/dl): Figure 4.9 illustrates the outcomes regarding the impact of *Acacia nilotica* on the cholesterol in broiler chickens. The cholesterol in the broiler chickens was significantly ($p<0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was significantly higher cholesterol (176 ± 2.44 mg/dl) in the group denoted as T0, compared to the groups T3 (172.33 ± 1.02 mg/dl), T6 (166 ± 1.08 mg/dl), and T12 (163.33 ± 1.24 mg/dl), these groups were supplemented with 3%, 6%, and 12% *Acacia nilotica*, respectively. Conversely, the T9 group, where the birds were supplemented with 9% *Acacia nilotica* exhibited a lower cholesterol (162.66 ± 1.02 g/dl).

AST (U/L): Figure 4.10 illustrates the outcomes regarding the impact of *Acacia nilotica* on the AST in broiler chickens. The AST in the broiler chickens was significantly ($p<0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was a significantly higher AST (226.5 ± 4.99 U/L) in the group denoted as T12, where the birds were supplemented with 12% *Acacia nilotica*, compared to the groups T6 (219 ± 2.88 U/L), T9 (212 ± 4.61 U/L), and T0 (199 ± 8.08

U/L), these groups were supplemented with 6%, 9%, and 0% *Acacia nilotica*, respectively. Conversely, the T3 group, where the birds were supplemented with 3% *Acacia nilotica* exhibited a lower AST (197±6.35 U/L).

ALT (U/L): Figure 4.11 illustrates the outcomes regarding the impact of *Acacia nilotica* on the ALT in broiler chickens. The ALT in the broiler chickens was non-significantly ($p>0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was a non-significantly higher ALT (6.75±0.14 U/L) in the group denoted as T12, where the birds were supplemented with 12% *Acacia nilotica*, compared to the groups T9 (6.5±0.28 U/L), T6 (6±0.76 U/L), and T3 (6±0.57 U/L), these groups were supplemented with 9%, 6%, and 3% *Acacia nilotica*, respectively. Conversely, the T0 group exhibited a lower ALT (5.5±0.28 U/L).

Figure 4.10 Effect of *Acacia nilotica* on AST (U/L) in broiler chicken

Variables	Group T0	Group T3	Group T6 (6%)	Group T9 (9%)	Group T12 (12%)	P-value
Hemoglobin (g/dl)	12.85±0.22ab	12.24±0.32b	12.66±0.13ab	12.25±0.19b	13.48±0.21a	0.0029
PCV (%)	33.33±1.40ab	30.00±1.15b	31.50±1.13ab	35.25±1.14a	35.46±1.01a	0.0097
RBC (x10 ⁶ µ):	2.66±0.04b	2.43±0.05b	2.45±0.07b	3.00±0.11a	2.80±0.15ab	0.0012
WBC (g/dl)	12.54±0.28a	11.28±0.28b	12.19±0.28ab	12.64±0.27a	12.82±0.28a	0.0053
Cholesterol (g/dl)	176.0±2.44a	172.3±1.02ab	166.0±1.08bc	162.6±1.02c	163.3±1.24c	.001
AST (U/L)	199.00±8.08b	197.00±6.35b	219.0±2.88ab	212.0±4.61ab	226.5±4.90a	0.0172
ALT (U/L)	5.500±0.28a	6.00±0.57a	6.00±0.28a	6.50±0.28a	6.75±0.14a	0.418

Growth hormone

Figure 4.12 illustrates the outcomes regarding the impact of *Acacia nilotica* on the growth hormone in broiler chickens. The hormone growth in the broiler chickens was non-significantly ($p>0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that growth hormone was determined in T0 (2.52±0.02), T3 (2.53±0.02), T6 (2.52±0.03), T9 (2.52±0.01) and T12 (2.47±0.03). These groups were supplemented with 0, 3%, 6%, 9% and 12% *Acacia nilotica*, respectively.

TSH (ng/ml): Figure 4.13 illustrates the outcomes regarding the impact of *Acacia nilotica* on the TSH in broiler chickens. The TSH in the broiler chickens was significantly ($p<0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was a significantly higher TSH (0.61±0.02 ng/ml) in the group denoted as T6, where the birds were supplemented with 6% *Acacia nilotica*, compared to the groups T3 (0.59±0.02 ng/ml), T0 (0.57±0.02 ng/ml), and T9 (0.52±0.02 ng/ml), these groups were supplemented with 3%, 0%, and 9% *Acacia nilotica*, respectively. Conversely, the T12 group, where the birds were supplemented with 12% *Acacia nilotica* exhibited a lower TSH (0.51±0.02 ng/ml).

Lipase (ng/ml): Figure 4.14 illustrates the outcomes regarding the impact of *Acacia nilotica* on the lipase in broiler chickens. The lipase in the broiler chickens was non-significantly ($p<0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was a non-significantly higher lipase (29±2.88 ng/ml) in the group denoted as T12, where the birds were supplemented with 12% *Acacia nilotica*, compared to the groups T9 (28.50±3.17 ng/ml), T0 (23±1.73 ng/ml), and T3 (20.50±2.02 ng/ml), these groups were supplemented with 9%, 0%, and 3% *Acacia nilotica*, respectively. Conversely, the T6 group, where the birds were supplemented with 6% *Acacia nilotica* exhibited a lower lipase (19±3.17 ng/ml).

Figure 4.12 Effect of *Acacia nilotica* on growth hormone (µLu/ml) in broiler chicken

Variables	Group T0	Group T3	Group T6 (6%)	Group T9 (9%)	Group T12 (12%)	P-value
Growth hormone (µLu/ml)	2.52±0.02a	2.53±0.02a	2.52±0.03a	2.50±0.01a	2.47±0.03a	0.6179
TSH (ng/ml)	0.57±0.02ab	0.59±0.02ab	0.61±0.02a	0.52±0.02ab	0.51±0.02b	0.0237
Lipase (ng/ml)	23.0±1.73a	20.50±2.02a	19.0±2.30a	28.0±3.17a	29.0±2.88a	0.0536

Duodenum length (mm): Figure 4.15 illustrates the outcomes regarding the impact of *Acacia nilotica* on the duodenum length in broiler chickens. The duodenum length in the broiler chickens was significantly ($p < 0.05$) influenced by varying levels of *Acacia nilotica* supplementation. The results showed that there was a significantly higher duodenum length (1566.28 ± 5.76 mm) in the group denoted as T6, where the birds were supplemented with 6% *Acacia nilotica*, compared to the groups T0 (1542.07 ± 10.32 mm), T3 (1529.34 ± 9.28 mm), and T9 (1517.97 ± 9.11 mm), these groups were supplemented with 0%, 3%, and 9% *Acacia nilotica*, respectively. Conversely, the T12 group, where the birds were supplemented with 12% *Acacia nilotica* exhibited a lower duodenum length (1507.95 ± 11.43 mm).

Jejunum length (mm): Figure 4.16 illustrates the outcomes regarding the impact of *Acacia nilotica* on the jejunum length in broiler chickens. The jejunum length in the broiler chickens was non-significant ($p > 0.05$) by varying levels of *Acacia nilotica* supplementation. The results showed that jejunum length was determined as 1217.61 ± 22.36 mm in T0 group, 1239.52 ± 17.77 mm in T3 group, 1243.37 ± 20.23 mm in T6 group, 1226.92 ± 18.11 mm in T9 group and 1188 ± 14 mm in T12 group. These groups were supplemented with 0%, 3%, 6%, 9%, 12% *Acacia nilotica*, respectively.

Ileum length (mm): Figure 4.17 illustrates the outcomes regarding the impact of *Acacia nilotica* on the ileum length in broiler chickens. The ileum length in the broiler chickens was non-significant ($p > 0.05$) by varying levels of *Acacia nilotica* supplementation. The results showed that ileum length was determined as 1105.17 ± 22.36 mm in T0 group, 1114.47 ± 17.77 mm in T3 group, 1100.46 ± 20.23 mm in T6 group, 1073.94 ± 18.11 mm in T9 group and 1062.95 ± 14 mm in T12 group. These groups were supplemented with 0%, 3%, 6%, 9%, 12% *Acacia nilotica*, respectively.

Figure 4.15 Effect of *Acacia nilotica* on duodenum length (mm) in broiler chicken

Variables	Group T0	Group T3	Group T6 (6%)	Group T9 (9%)	Group T12 (12%)	P-value
Duodenum length	1542.07 \pm 10.31ab	1529.34 \pm 9.29ab	1566.28 \pm 5.76a	1517.97 \pm 9.11b	1507.95 \pm 11.43b	0.0028
Jejunum length	1217.61 \pm 18.70a	1239.52 \pm 18.70a	1243.37 \pm 18.70a	1226.61 \pm 18.70a	1188 \pm 18.70a	0.2803
Ileum length	1105.17 \pm 16.70a	1114.47 \pm 16.00a	1100.46 \pm 16.87a	1037 \pm 12.484a	1062.95 \pm 21.20a	0.1935

Discussions

The present study investigated the impacts of *Acacia nilotica* supplementation on broiler chicken digestibility, hematological parameters, and gut morphology. Results showed that *Acacia nilotica* had no significant effect on dry matter digestibility, consistent with previous studies. However, it had varied effects on crude protein, metabolizable energy, and crude fat digestibility. *Acacia nilotica* supplementation improved crude protein digestibility, potentially due to bioactive compounds enhancing protein utilization (Jayanegara *et al.*, 2018; Parvin *et al.*, 2014). Metabolizable energy digestibility showed an optimal effect at 6% supplementation, with higher levels decreasing digestibility, possibly due to anti-nutritional factors (Gomathi *et al.*, 2020; Simitzis *et al.*, 2018). Crude fat digestibility increased with higher levels of *Acacia nilotica* supplementation, distinguishing it from other herbal plants (Adhikari *et al.*, 2018; Patra *et al.*, 2020).

Acacia nilotica supplementation also influenced hematological parameters. It increased hemoglobin levels, packed cell volume, and white blood cell count, indicating potential benefits for overall health and immune response (Patel *et al.*, 2019; Khan *et al.*, 2021). *Acacia nilotica* was also found to manage cholesterol levels, with supplementation leading to decreased levels (Shahzad *et al.*, 2020). The effects on liver enzymes, AST and ALT, were inconsistent (Al-Sagan *et al.*, 2020; Ayoob *et al.*, 2023). In terms of gut morphology, *Acacia nilotica* supplementation increased duodenum length, potentially due to stimulation of gut development and improved nutrient utilization (Ghazalah *et al.*, 2021; Wadood *et al.*, 2022). However, it had no significant effect on jejunum and ileum lengths, highlighting organ-specific responses to supplementation (Nasiroleslami *et al.*, 2019). The optimal dosage of *Acacia nilotica* supplementation requires further investigation to avoid potential negative effects (Zeweil, 2020).

In conclusion, *Acacia nilotica* supplementation showed varied effects on broiler chicken digestibility, hematological parameters, and gut morphology. It improved crude protein digestibility, had optimal effects on metabolizable energy digestibility at 6%, increased crude fat digestibility, and influenced hematological parameters and gut morphology. However, the optimal dosage and potential adverse effects need further research (Chang *et al.*, 2017; Ghosh *et al.*, 2022; Melesse *et al.*, 2022; Akhtar *et al.*, 2020). Considering the unique chemical composition of *Acacia nilotica*, more targeted studies are necessary to fully understand its potential in broiler diets (Salem *et al.*, 2021).

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