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Effects of *Moringa oleifera* on Growth Biomarkers, Carcass Characteristics and Hematological Parameters of Broiler Chicken

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ABSTRACT

Antibiotics are frequently employed for promoting growth and preventing diseases, yet their application entails drawbacks including residual effects, drug toxicity, and the rise of bacterial resistance. The objective of the present study was to examine the impact of *Moringa oleifera* leaf mash (MOLM) on various parameters, including feed intake, carcass characteristics, final weight, weight gain, feed conversion ratio, blood profile, and the growth of broiler chicks. A six-week trial was conducted on day-old broiler chicks (Hubbard), with a total of 350 chicks distributed evenly into five groups. The first group, designated as the Control group, was fed a basal feed common to the other groups without MOLM. The remaining groups, namely MOLM2, MOLM4, MOLM6, and MOLM8, received 2, 4, 6, and 8g/Kg of MOLM, respectively, in their common basal feed. The results indicated that broiler chicks fed with 4% MOLM exhibited significantly higher growth performance compared to other treatment groups. The chicks fed with 4% Moringa oleifera-supplemented feed showed a significant increase in body weights compared to the control group. A notable upward trend in live and dressed body weights was observed compared to other treatment groups. There was no significant change in feed intake across all treatment groups. Hematological analysis revealed that, in comparison to the control group, broiler chicks fed with 2%, 4%, and 6% MOLM-supplemented feed showed significantly increased values of hemoglobin and packed cell volume. The same trend was observed for white blood cells, with significantly increased values in all treatment groups compared to the control group. While there was a non-significant difference in red blood cells, a significant increase in white blood cells was observed across all treatment groups compared to the control group. In conclusion, the addition of 4% MOLM to the feed resulted in increased values for growth performance and carcass traits in broiler chicks.

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SU and SU designed the study. SU and FR collected the sample. SU and MSK wrote and submitted the manuscript. FR, MSK and SU edited and reviewed the manuscript.

Key words
***Moringa oleifera*, Growth, Carcass traits, Blood profile, Broiler**

INTRODUCTION

Poultry production stands as a crucial foundation for the advancement of food safety measures. Moreover, it serves as a pivotal driver in the socio-economic and cultural development of diverse nations (Chuang *et al.*, 2007). The significance of poultry production extends beyond mere sustenance, positioning itself as both a source of livelihood and a valuable reservoir of protein. This dual role not only contributes to immediate income generation but also ensures a reliable protein source for communities (Khan *et al.*, 2023a; Ahmad *et al.*, 2020). However, despite its numerous benefits, emerging economies grapple with challenges, particularly the escalating ratio of high feed to gain (Ahmad *et al.*, 2020). Furthermore, the soaring costs of feed ingredients pose a continuous hurdle (Chuang *et al.*, 2007). These challenges underscore the need for strategic interventions and sustainable practices to enhance the resilience and efficiency of poultry production in the face of evolving economic and environmental dynamics (Hafeez *et al.*, 2020a).

Numerous initiatives have been undertaken to address the challenges associated with poultry production, and one notable approach involves administering antibiotics through nutrition. These antibiotics serve dual purposes: As growth promoters and as preventive measures against diseases (Hafeez *et al.*, 2020b). While the utilization of antibiotics as growth enhancers has shown positive outcomes, it is not without its drawbacks. Persistent issues include residual effects, potential drug toxicity, and the alarming emergence of bacterial resistance (Khan *et al.*, 2020). Recognizing these concerns, the European Union has taken a significant step by prohibiting the use of antibiotics as growth promoters. The drawbacks associated

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with antibiotic use have prompted a growing awareness of the need for natural and safe alternatives, such as herbal remedies, to mitigate the reliance on antibiotics in poultry production (Rehman *et al*., 2020; Khan *et al*., 2023a). This shift in focus reflects a broader commitment to sustainable and environmentally friendly practices within the industry. As the poultry sector continues to navigate these challenges, the exploration of herbal alternatives not only promotes the health and well-being of the poultry but also aligns with a global effort to foster responsible and ethical agricultural practices. Consequently, the increasing adoption of natural alternatives signals a positive step toward creating a more resilient and sustainable future for poultry production (Murslain *et al*., 2014; Sallam *et al*., 2018; Hafeez *et al*., 2020b; Khan *et al*., 2023b).

Moringa oleifera, a widely acclaimed and easily cultivable plant, is recognized for its diverse applications and nutritional richness. The *M. oleifera* tree is native to South Asia, specifically in countries such as Pakistan, Bangladesh, India, Afghanistan, Sri Lanka, as well as in regions of Africa, Arabia, and Madagascar (Babiker *et al*., 2022). With its abundance of essential phytochemicals, the plant's various components serve as valuable sources of nutrition for human consumption and are integrated into traditional foods across different parts of the world. The nutritional potency of *M. oleifera* makes it a sought-after resource in both therapeutic and feed industries (Fahey, 2005; Murslain *et al*., 2014; Naz *et al*., 2022). The plant's leaves, seeds, pods, and flowers are particularly esteemed for their multifaceted benefits in human nutrition and herbal medicine (Alazzouni *et al*., 2021). This widespread utilization of different parts of the moringa plant reflects its versatility and effectiveness in contributing to both dietary needs and holistic healthcare practices across diverse global regions. As awareness of its nutritional and therapeutic properties continues to grow, *M. oleifera* stands out as a valuable and sustainable resource in promoting health and well-being (Murslain *et al*., 2014; Khan *et al*., 2023c; Huang *et al*., 2023).

The available data on the impact of *M. oleifera* leaf meal (MOLM) extract in broiler feed on both growth and meat quality yields conflicting and incomplete results. Khan *et al*. (2023d) concluded that while there was a reduction in ingestion, there was an improvement in the feed conversion ratio (FCR). On the contrary, Ogbunugafor *et al*. (2011) reported a decrease in the growth performance of broiler chickens when the concentration of MoLM exceeded 5% in the feed. Interestingly, findings from Ogbunugafor *et al*. (2011) and Luqman *et al*. (2012) collectively indicated that incorporating MOLM up to a 10% level did not have adverse effects on the reproductive performance of broiler chickens. However, concentrations higher than 10% led to detrimental outcomes. This variation in results may be attributed to factors such as impure extraction of leaf meal, variations in preparation methods such as cooking or tincturing as a leaf extract (Moustafa *et al*. 2015; Hamza, 2016). Moreover, Abbas (2013) suggested that the reduced productivity at elevated constituent levels could be due to an increase in anti-nutritional factors, poor digestibility of energy fiber, the presence of dust, and the occurrence of proteins in raw leaf form. These factors collectively contribute to a nuanced understanding of the complexities involved in incorporating MOLM extract into broiler feed, emphasizing the importance of precision in preparation methods and concentration levels to ensure optimal growth and meat quality. Further research and standardization of practices are essential to unlock the full potential of *M. oleifera* as a valuable component in broiler nutrition (Ghaffar *et al*., 2018; Babiker *et al*., 2022). The current study was planned to investigate how *M. oleifera* leaf mash (MOLM) affects a range of parameters, encompassing feed intake, carcass characteristics, final weight, weight gain, feed conversion ratio, blood profile, and the growth of broiler chicks.

MATERIALS AND METHODS

Test ingredients and preparation of the extracts

Fresh *M. oleifera* leaves were sourced from local farmers, meticulously washed, and air-dried on a polythene sheet in a shaded area. Subsequently, the dried leaves underwent grinding into a fine powder using an electric grinder. To prepare the aqueous extract, 100 g of the powder were soaked in 1 L of fresh water for 24 h with regular agitation. Afterward, the solution was meticulously filtered through a muslin cloth to eliminate any remaining particles, and the resulting extract was stored in a clean, airtight container at 4°C for future use. Various concentrations were derived from the *M. oleifera* aqueous extract, specifically 30mL, 60mL, 90mL, and 120mL. These concentrations were employed in the experimental setup. To monitor the impact of the *M. oleifera* extract, the initial body weight of the subjects was measured at the commencement of the experiment. Subsequent measurements were recorded weekly, encompassing both body weight and feed intake. The calculation of the FCR was carried out based on the relationship between feed consumption and body weight gain. This meticulous process ensured a comprehensive evaluation of the effects of *M. oleifera* on the growth and dietary patterns of the subjects throughout the experiment.

Housing management

The birds were maintained in thoroughly clean and hygienic environment. At the onset of second week, the birds were provided with Super Starter Mash, which continued throughout the third and fourth week. Upon reaching the fifth week of age, the diet transitioned to finisher feed pellets, extending through the sixth week. The respective feeds were made available *ad libitum*, and the transition from one feed type to the next was executed gradually to prevent any potential digestive issues.

Experimental design

The 350 birds were distributed into five distinct groups: the control group (C) received conventional feed exclusively, serving as the baseline, and the four groups viz. MOLM2, MOLM4, MOLM6 and MOLM8 were provided with conventional feed supplemented with *M. oleifera* leaf mash at a rate of 2, 4, 6 and 8g/kg, respectively.

Carcass traits and blood analysis

At the end of the experiment, a representative sample of 5 birds was randomly selected from each experimental group. These selected birds underwent an overnight fasting period before being humanely slaughtered. The dressing percentage, calculated as the ratio of the live body weight to the dressed weight after removal of viscera, feet, and skull, was determined for each bird. Additionally, a total of 3ml of blood was collected from each bird using EDTA-coated vacutainers for subsequent analysis. The complete blood count (CBC) was done utilizing a hematology analyzer, providing comprehensive information on various blood parameters.

Statistical analysis

The data was analyzed via one-way ANOVA using the SPSS version-21 statistical package, with statistical significance set at $P<0.05$. Means were compared using Tukey's test.

RESULTS

Birds from various treatment groups (C, MOLM2, MOLM4, MOLM6, and MOLM8) consumed 4301g, 4200g, 4305g, 4253g, 4281g, and 4220g feed per bird, respectively. Table I summarizes the final body weight, total weight gain, and FCR for the different treatment groups. Interestingly, birds fed a diet consisting of 4% Moringa mash exhibited the highest body weight compared to other groups, while those fed with 8% MOLM had the lowest final body weight. The same pattern was observed for daily weight gain, with MOLM4 showing higher values than other groups. Notably, the FCR was significantly better in the MOLM4 group, displaying the lowest ratio compared to other treatment groups, including the control group. Furthermore, a significant increase in dressing weight and percentage was observed in the MOLM4 group (Table I). While no significant difference was noted in dressing percentage among the treatment groups, a significant decline in the relative weight of the proventriculus was observed. Conversely, non-significant differences were noted in the relative weights of the heart, gizzard, and spleen in birds fed on MOLM.

Table I. Growth performance and carcass characteristics of broilers at different experimental diets.

Item	C	$MOLM_2$	$MOLM_4$	$MOLM_6$	$MOLM_8$	P value
Growth performance						
Initial body weight (g)	50.13±0.512	49.87±0.76	51.21±0.74	50.26±0.62	49.78±0.86	0.125
Final body weight (g)	2272±9.21	2345±9.62	2478±10.56	2248±13.56	2135±14.96	0.001
Weight gain (g)	2223±9.42	2295±9.32	2427±9.88	2198±14.06	2085.22	0.01
Feed intake	4983.53	4834.03	4762.22	4533.91	4487.21	0.0451
FCR	2.24±0.001	2.10±0.002	1.96±0.001	2.06±006	2.15±0.005	0.000
Carcass characteristics						
Live body weight	2272±9.21	2345±9.62	2478±10.55	2248±13.56	21.35±14.96	0.036
Dressed weight	1797±10.51	1972±9.16	1964±10.25	1929±12.92	1445±14.67	0.000
Dressing %	79.09±0.52	82.17±2.87	79.25±1.62	72.46±1.26	67.68±0.89	0.002

C, control; $MOLM_{2, 4,6,8}$, conventional feed supplemented with *M. oleifera* leaf mash at a rate of 2, 4, 6 and 8g/kg, respectively.

Table II. Hematology profile of different groups of broiler fed on Moringa mixed feed.

Parameter	C	$MOLM_2$	$MOLM_4$	$MOLM_6$	$MOLM_8$
Hb (g/dL)	11.67^a	11.47^a	11.29^a	11.16^a	10.82^b
RBC ($\times 10^{12}$/L)	2.19^a	2.11^a	2.08^a	2.03^a	2.01^a
WBC($\times 10^9$/L)	5.32^a	5.74^a	5.78^a	5.93^a	6.36^a
PCV (%)	38.93^a	37.59^a	37.18^a	35.42^a	31.65^b
MCV (fL)	143.73^a	139.44^b	143.17^a	144.21^a	144.94^a
MCH (pg)	40.28^a	40.05^a	40.36^a	40.48^a	40.57^a
MCHC (g/dL)	28.33^a	28.94^a	27.91^a	28.36^a	29.16^a

*The mean in rows having different superscripts are differ to each other significantly ($P<0.05$).
Hb, hemoglobin; RBC, red blood cells; WBC, white blood cells; PCV, packed cell volume; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration.

Table II presents the impact of MOLM feed on various blood parameters within the treatment groups. The blood profile serves as a biomarker, reflecting the nutritional, physiological, and pathological status of the animals. In comparison to the control group, broiler chicks fed on 2%, 4%, 6%, and 8% MOLM displayed significantly increased values of hemoglobin and packed cell volume. This trend was similarly observed for white blood cells, where all treatment groups showed significantly higher values compared to the control group. Conversely, a non-significant difference was noted for red blood cells across the treatment groups.

DISCUSSION

In alignment with the United Nations' sustainable development goals, sustainable poultry production plays a pivotal role in addressing hunger and promoting good health and well-being by supplying essential high-quality protein and micronutrients through the provision of meat and eggs (Khan *et al*., 2020, 2023b). To achieve this, broiler producers worldwide are prioritizing optimal carcass yield and meat quality traits through the maintenance of high broiler welfare standards, enhancement of gut health, and reduction of locomotion disorders (Hafeez *et al*., 2020b; Babiker *et al*., 2021). This focus has led to the adoption of alternatives such as prebiotics, probiotics, synbiotics, and phytogenics, replacing prophylactic and growth-promoting antibiotics. Probiotics, consisting of live beneficial bacteria cultures, are utilized in poultry production to establish a balanced and diverse gut microbial environment, specifically targeting pathogens (Fahey *et al*., 2005; Murslain *et al*., 2014). Proper probiotic dosages offer nutritional and physiological benefits, including enhanced growth performance, intestinal flora, immune response, and meat quality (Haroon *et al*., 2022; Alazzouni *et al*., 2021). The increasing demand for natural and environmentally friendly alternatives has propelled the market for phytogenic products. Phytochemicals, biologically active compounds in plants, induce physiological changes in farm animals, with *M. oleifera* emerging as a recently utilized phytogenic plant in poultry nutrition (Murslain *et al*., 2014; Sallam *et al*., 2018; Zubair *et al*., 2020; Hafeez *et al*., 2020b; Khan *et al*., 2023b).

M. oleifera stands out for its rich composition of minerals, vitamins, and proteins, notably featuring all eight essential amino acids (Khan *et al*., 2023d). The leaves of this plant exhibit exceptional digestibility and are devoid of any factors that might impede feed intake. Consequently, in the present study, there was a notable absence of significant differences in feed intake among the experimental groups. Furthermore, the research demonstrated an improved FCR and weight gain, aligning with similar observations reported by various researchers such as Sammad *et al.* (2022) . The enhanced growth performance observed in the current study can be attributed to the high digestibility and nutritional content of Moringa, contributing to a more favorable FCR. This improved body weight and FCR may be linked to the presence of bioactive compounds in the plant leaves, enhancing nutrient utilization in the birds (Khan *et al*., 2024). However, a noteworthy observation in the current study was that higher doses of *M. oleifera* had an adverse impact on the weight gain of broiler chickens, consistent with findings by Maheshwari *et al.* (2014). This phenomenon could be attributed to the higher fiber content in the leaves, potentially exerting a negative effect on the availability of energy and protein, as reported by Zubair *et al*. (2020).

The outcomes of the present study indicate that the supplementation of *M. oleifera* leaves leads to an increase in dressing weight. These results are consistent with reports from several other studies (Murslain *et al*., 2014; Sallam *et al*., 2018). In summary, it can be concluded that the findings from the current study align with previous research reports, confirming that the inclusion of *M. oleifera* leaf supplement in broiler feed enhances the growth and production of broilers. This positive impact is likely attributed to the presence of specific phytochemicals and nutrients (Cheng *et al*., 2019).

CONCLUSION

In conclusion, the present study highlights that the inclusion of 4% MOLM in the feed significantly enhances the growth performance and carcass traits of broiler chicks. The observed increase in body weights, both live and dressed, signifies a positive impact on overall growth. Additionally, hematological analyses indicate favorable changes in blood parameters with MOLM supplementation. Future recommendations include further exploration of optimal dosage levels, long-term effects on broiler health, and potential applications of MOLM in diverse poultry production systems for sustainable and improved outcomes.

DECLARATIONS

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Statement of conflict of interest

The authors have declared no conflict of interest.

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