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Supplementation with *Moringa oleifera* Polysaccharides Improves Growth Performance, Carcass Traits, and Meat Quality in Broiler Chickens

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ABSTRACT

Growth promoting antibiotics in poultry feed are banned and this has resulted in a rise in the search for efficient and safe alternatives. *Moringa oleifera* polysaccharides (MOLPs) are bioactive carbohydrate polymers isolated mainly from leaves, seeds of *Moringa oleifera* tree with antioxidant, anti-inflammatory and immunomodulatory activities. However, effects of MOLPs in broiler chickens are less studied. To evaluate the effects of dietary MOLPs on growth performance, serum biochemistry, antioxidant capacity, immune function, intestinal morphology, and cecal microbiota in Guangxi Sanhuang chickens. A total of 600 one day old chicks were randomly assigned to five groups (8 replicates of 15 birds each): control (basal diet) and four treatment groups supplemented with 0.8, 1.2, 1.6, and 2.0 g/kg MOLPs for 42 days. Growth performance was measured; serum and liver samples were analyzed for biochemical and antioxidant parameters; immune organ indices and cytokine levels were determined; intestinal villus morphology was assessed; and cecal microbiota were profiled by 16S rRNA sequencing. Compared with the control, the 1.2 and 1.6 g/kg MOLPs groups showed significantly lower feed conversion ratio (FCR) across all feeding stages ($P < 0.05$). The 1.2 g/kg group had higher final body weight, average daily gain, and average daily feed intake ($P < 0.05$). MOLPs at 1.6 g/kg increased liver glutathione peroxidase (GSH-Px) and total antioxidant capacity (T-AOC), and decreased malondialdehyde (MDA) ($P < 0.05$). Immune organ development was enhanced; thymus weight increased in all MOLPs groups, and bursa weight increased with 1.2 and 1.6 g/kg MOLPs ($P < 0.05$). Serum and liver pro-inflammatory cytokines (IL-1β, IL-6, TNF-α) decreased, while anti-inflammatory IL-4 increased ($P < 0.05$). MOLPs at 1.2 g/kg upregulated hepatic IL-4 mRNA and downregulated IL-6 mRNA ($P < 0.05$). Intestinal villus height and the villus height to crypt depth ratio improved in duodenum, jejunum, and ileum. Cecal microbiota alpha diversity (Chao1 and Shannon indices) increased with 1.2 g/kg MOLPs, and the relative abundances of beneficial bacteria such as Parabacteroides and Roseburia were elevated ($P < 0.05$). Dietary MOLPs at 1.2–1.6 g/kg enhance growth performance, antioxidant status, immune function, intestinal morphology, and cecal microbial diversity in Guangxi Sanhuang chickens. MOLPs represent a promising plant derived feed additive for antibiotic-free poultry production.

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Key words
***Moringa oleifera* polysaccharides, Chicken growth performance, Immune function, Gut microbiota, antioxidant**

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INTRODUCTION

Broilers are now the world's second largest livestock industry because they grow fast, have a short feeding cycle and a high feed conversion rate (FCR). But the excessive use of antibiotics in poultry production has seriously disordered the intestinal microbial balance of the animals, which seriously threatens human health and causes great ecological damage. Plant polysaccharides could be a promising alternative (Wang *et al*., 2021). Growth performance has a direct effect on economic returns in animal production. Plant polysaccharides increase growth by increasing digestion and absorption of nutrients. For instance, *Astragalus* polysaccharide enhances average daily gain and decreases feed-to-weight ratio in broilers by promoting beneficial gut bacteria and digestive enzyme activity (Wang *et al*., 2021). Dandelion polysaccharide has a positive effect on protein digestion, calcium and

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phosphorus deposition and bone development, and improves feed palatability and intake. Furthermore, plant polysaccharides can reduce stress responses *Astragalus* and ginseng polysaccharides can relieve immune stress induced by lipopolysaccharide (LPS) and improve intestinal barrier function, while *Hericium erinaceus* polysaccharide can alleviate heat stress-induced metabolic disorders (Wang *et al*., 2020).

Dietary plant polysaccharides also boost immunity. Lentinan (50–150 mg/kg) increased average daily gain, feed conversion, and immune organ development in yellow-feathered broilers, and increased serum IL-2 and stimulated spleen cell proliferation through the cAMP signaling pathway (Chen *et al*., 2003). Inflammatory cytokines play an important role in poultry immunity (Al-Sadi *et al*., 2016). *Astragalus* polysaccharides together with taurine reinforce the integrity of the mucosal barrier, which also benefits intestinal health, increased ZO-1 mRNA levels and decreased intestinal permeability. In general, plant polysaccharides have been proven to improve the antioxidant capacity, immune function, intestinal development, digestive enzyme activity and economic benefits of poultry production (Zhou *et al*., 2020; Long *et al*., 2020). *Moringa oleifera* Lam. (Moringa), the "miracle tree" is native to northern India and widely cultivated in southern China. Its leaves, seeds and roots are rich in vitamins, minerals and vital nutrients, making them valuable as human food and animal feed (Saini *et al*., 2016; Mansour *et al*., 2018). *Moringa oleifera* polysaccharides (MOLP) have various bioactivities and are a promising feed additive (He *et al*., 2018). Moringa improves growth performance, intestinal health and meat quality in monogastrics. *Moringa* leaf meal (5%) improves average daily gain reduces lipid peroxidation and improves the unsaturated fatty acids content in chicken meat (Fida *et al*., 2021; Nkukwana *et al*., 2014; Sebola *et al*., 2019). *Moringa* leaf powder (1.2%) improves intestinal health by increasing the weight of small intestine and cecum, villus height, surface area and acid mucin production (Baloch *et al*., 2021; Khan *et al*., 2017). Egg yolk colour, protein absorption, eggshell strength and antioxidant enzyme activity in laying hens was improved by 5% Moringa leaf powder (Lu *et al*., 2016; Shen *et al*., 2021). In our previous studies, we found that fermented *Moringa* and *Moringa* leaf extract enhanced the development of tibia, intestinal villi, slaughter performance, immune function and beneficial gut microbiota in broilers.

Broilers under intensive farming are susceptible to stress-induced intestinal disorders, reduced immunity and decreased performance, thus increasing production costs. Nutritional intervention is an effective strategy to mitigate these adverse effects (Chauhan *et al*., 2021; Liu *et al*., 2019). Considering the increasing interest in plant polysaccharides as functional and non-residue feed additives (Kouakou *et al*., 2013), this study has explored the positive effects of dietary MOLP supplementation on growth performance, immune function and intestinal microflora of Guangxi Sanhuang chickens, which can be used as a reference for non-antibiotic breeding. Objectives of this study were (1). Evaluating growth performance (BW, ADG, ADFI and F/G) of broiler fed 0.8–2.0 g/kg MOLP from 1–42 days; (2) Evaluate slaughter performance (dressing percentage, eviscerated rates, abdominal fat, breast/leg muscle yields, organ indices) at 42 days, (3) Measure meat quality (pH, color L/a/b*, shear force) of breast and leg muscles and determine the optimal level of supplementation (1.2–1.6 g/kg) to improve production traits without negative effects.

MATERIALS AND METHODS

Chicken and experimental design

A total of 600 one day old Guangxi Sanhuang broiler chicks with an average initial body weight of 45.6 ± 2.3 g (mean ± SD) were randomly assigned to five experimental groups, with 8 replicates of 15 birds each (n = 120 birds/group). The control group (CK) was fed a basal diet without any supplement. The basal diet with *Moringa oleifera* polysaccharide (MOLP) at levels of 0.8, 1.2, 1.6 and 2.0 g/kg of feed was supplied to T1, T2, T3 and T4 respectively. The feeding trial time was of 42 days, which consisted of the starter phase and finisher phase Table I. All birds were provided feed and water *ad libitum* throughout the duration of the experiment and were handled according to standard commercial broiler production practices. MOLP extract with 50.18% polysaccharide content was provided by Xi'an Tetra Leaf Biological Company.

On the 1st day, 21st day and 42nd day of the experiment, Sanhuang chickens in each group were weighed in replicates, fasted for 12 h before weighing, and drank freely during the experiment. During the experiment, feed intake and residual feed amount of the chickens were recorded in replicates in detail. At the end of the experiment, feed/weight ratio (F/G), average daily feed intake (ADFI) and average daily gain (ADG) were calculated, as follow. Average daily feed intake (g/d)(ADFI)= total feed intake/(number of test chickens × test days), Average daily weight gain (g/d)(ADG) = (final weight-initial weight)/(number of test chickens × test days), Feed to weight ratio (F/G)= average daily feed intake/average daily weight gain.

Evaluation of carcass traits

On the 42 day of the experiment, one test chicken was randomly selected from each replicate, weighed empty stomach and slaughtered. At the end of the trial, three birds

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per replicate were slaughtered by jugular exsanguination. Carcass weight was recorded after feather removal, and dressing percentage was calculated. Semi-eviscerated and fully eviscerated carcass weights were obtained after sequential removal of internal organs. Heart, liver, pancreas, gizzard, and proventriculus were weighed to calculate organ indices.

Abdominal fat, left pectoralis major, and left thigh and drumstick were weighed to determine abdominal fat rate, breast muscle rate, and leg muscle rate, respectively.

Table I. Basic diet composition and nutrient levels (air-dry basis).

Diet componenets	1–21 days	22–42 days
Ingredients composition (%)		
Corn	55.7	59.5
Soybean meal	26.3	22.35
Fish meal	2.5	2.5
Expanded full fat soybeans	8.2	6.45
Soybean oil	1	3.3
NaCl (Salt)	0.4	0.4
Limestone	1.6	1.4
$CaHPO_4$ (Calcium hydrogen phosphate)	2.3	2.1
Premix[1]	2	2
Total	100	100
Nutrient levels[2]		
Metabolizable energy (ME), MJ/kg	12.38	12.87
Crude protein (CP), %	20.32	19
Ether extract (EE), %	3.45	4.23
Calcium (Ca), %	0.97	0.85
Total phosphorus (P), %	0.69	0.64
Lysine (Lys), %	1.15	0.98
Methionine (Met), %	0.48	0.42

[1]The supplied premix (per kg diet) VB 1 3.0 mg, VB 2 7.25 mg, VB 6 4.75 mg, VE 4.5 mg, VK 28.5 mg, VA 10500 IU, VD 3 3200 IU, choline 550 mg, pantothenic acid 25 mg, biotin 0.30 mg, niacin 32 mg, folic acid 1.20 mg, Cu 8.8 mg, Mn 95 mg, Zn 80 mg, Se 0.25 mg, Fe 75 mg, Co 0.15 mg. Crude protein and total phosphorus are measured nutrient values; all other nutrients are calculated. The experiment lasted for 42 days from September to November in 2022, broilers were randomly assigned to five dietary treatment groups and were provided *ad libitum* access to feed and water. The health status of chickens was monitored daily.

Assessment of meat quality

At the end of the 42-days experimental period, three birds per replicate were randomly selected and slaughtered. The left and right pectoralis major (breast) muscles and thigh (leg) muscles were carefully dissected and collected for the determination of pH, meat color, and shear force. The pH of the left breast muscle and left leg muscle was measured using a portable pH meter (model, manufacturer, city, country) calibrated with standard buffer solutions (pH 4.0 and 7.0). The probe was inserted directly into the muscle at 45 min postmortem. Each sample was measured in triplicate, and the average value was recorded. Color parameters (L* for lightness, a* for redness, and b* for yellowness) of the breast and leg muscles were measured using a NR20XE colorimeter (Shenzhen, China). Measurements were taken at the same location on each muscle after 30 min of blooming at room temperature. Each sample was read in triplicate, and the mean values were calculated. Shear force of the breast and leg muscles was determined using a tenderness meter. Core samples (1.0 cm diameter) were cut parallel to the muscle fiber direction, and the maximum shear force (N) was recorded. Each sample was measured in triplicate.

Data processing and analysis

All experimental data were initially summarized in Microsoft Excel 2019 (Microsoft Corp., Redmond, WA, USA). Statistical analyses were performed using one-way analysis of variance (ANOVA) with SPSS version 25.0 (IBM Corp., Armonk, NY, USA). If significant differences were found among treatment means ($P < 0.05$), Duncan's multiple range test was used to compare group means. All data were expressed as mean ± standard deviation (SD). The graphs were drawn using GraphPad Prism 9.0 (GraphPad Software, San Diego, CA, USA).

RESULTS

Growth performance of chicken

From Table II, it can be seen that there was no significant difference in body weight (BW) among the treatment groups at 1 day of age. However, the F/G ratio of the MOLP groups (T2: 1.2 g/kg; T3: 1.6 g/kg) was significantly lower than that of the control group (T1) ($P < 0.05$). From 22 to 42 days of age, the 42-day body weight of broilers was significantly increased by adding 1.2 g/kg MOLP ($P < 0.05$). Furthermore, the F/G ratio of the polysaccharide groups (T2 and T3) was significantly lower than that of the control group (T1). Compared with the control group, the average daily gain (ADG) increased significantly ($P < 0.05$), and the F/G ratio decreased significantly ($P < 0.05$) in the 1.2 g/kg and 1.6 g/kg MOLP groups. Additionally, the average daily feed intake (ADFI) increased significantly ($P < 0.05$) in the 1.6 g/kg MOLP group.

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Table II. Effect of MOLPs on growth performance of Guangxi Sanhuang chickens (1–42 days).

Item	CK	T1 (0.8 g/kg)	T2 (1.2 g/kg)	T3 (1.6 g/kg)	T4 (2.0 g/kg)	P-value
1–21 days						
Initial BW (g)	29.02 ± 0.52	28.98 ± 0.40	28.97 ± 0.49	29.01 ± 0.56	29.03 ± 0.62	0.173
21 d BW (g)	229.77 ± 25.79	232.43 ± 25.88	236.36 ± 24.65	237.32 ± 26.83	233.66 ± 25.46	0.253
ADG (g/d)	9.54 ± 1.23	9.65 ± 1.23	9.85 ± 1.18	9.89 ± 1.28	9.73 ± 1.21	0.243
ADFI (g/d)	17.43 ± 1.54	17.15 ± 1.39	17.63 ± 1.47	17.65 ± 1.55	17.53 ± 1.73	0.76
F/G	1.85 ± 0.05a	1.81 ± 0.04ab	1.77 ± 0.03b	1.79 ± 0.05b	1.82 ± 0.07ab	0.012
22–42 days						
42 d BW (g)	516.79 ± 29.58b	520.86 ± 29.78ab	532.26 ± 28.92a	524.63 ± 26.53ab	525.31 ± 31.37ab	0.02
ADG (g/d)	13.67 ± 0.69	13.73 ± 0.56	13.92 ± 1.62	14.04 ± 1.77	13.89 ± 0.35	0.167
ADFI (g/d)	35.88 ± 1.68	36.06 ± 2.28	37.11 ± 2.01	36.02 ± 1.87	35.93 ± 1.67	0.225
F/G	2.71 ± 0.09a	2.67 ± 0.10ab	2.58 ± 0.08b	2.56 ± 0.05b	2.68 ± 0.05ab	0.013
1–42 days						
ADG (g/d)	11.60 ± 0.70b	11.70 ± 0.71ab	11.88 ± 0.64a	11.97 ± 0.69a	11.81 ± 0.75ab	0.03
ADFI (g/d)	26.02 ± 2.01b	26.66 ± 2.71ab	27.00 ± 3.55ab	28.74 ± 1.75a	26.97 ± 3.59ab	0.039
F/G	2.20 ± 0.03a	2.20 ± 0.02ab	2.14 ± 0.03b	2.11 ± 0.03b	2.18 ± 0.03ab	0.002

ADG, average daily gain; ADFI, average daily feed intake; F/G ratio feed/ weight ratio. Different lowercase letters in the shoulder of the same line data indicate significant difference ($P < 0.05$), the same letter or no letter in the shoulder indicates no significant difference ($P > 0.05$), Compared with CK, the 1.2 and 1.6 g/kg MOLPs groups had significantly lower F/G during all feeding stages ($P < 0.05$). The 1.2 g/kg MOLPs group had significantly higher 42-day body weight, 1–42 d ADG, and 1–42 d ADFI ($P < 0.05$).

Table III. Effect of MOLPs on slaughter performance of Guangxi Sanhuang chickens (42 days).

Item	CK	T1 (0.8 g/kg)	T2 (1.2 g/kg)	T3 (1.6 g/kg)	T4 (2.0 g/kg)	P-value
Slaughter rate (%)	87.83 ± 2.33	88.84 ± 1.43	88.20 ± 2.04	89.07 ± 2.46	87.90 ± 1.57	0.669
Half-eviscerated rate (%)	80.40 ± 2.90	81.76 ± 3.15	81.34 ± 2.29	83.16 ± 3.08	82.13 ± 1.85	0.075
Full-eviscerated rate (%)	62.08 ± 3.38b	63.98 ± 4.36ab	63.34 ± 1.79ab	67.67 ± 5.78a	64.66 ± 3.55ab	0.027
Abdominal fat rate (%)	1.52 ± 0.16a	1.36 ± 0.25ab	1.27 ± 0.05b	1.41 ± 0.12ab	1.37 ± 0.26ab	0.04
Breast muscle rate (%)	13.78 ± 1.23	13.53 ± 1.63	14.19 ± 1.84	13.99 ± 1.39	14.08 ± 1.34	0.327
Leg muscle rate (%)	16.42 ± 1.94	16.53 ± 2.01	16.89 ± 3.93	17.43 ± 1.24	16.17 ± 2.76	0.682
Heart index	5.04 ± 0.16b	5.17 ± 0.22b	5.42 ± 0.13a	5.23 ± 0.11ab	5.15 ± 0.16b	0.002
Liver index	20.83 ± 1.19b	21.71 ± 1.05ab	22.05 ± 0.84ab	22.72 ± 0.85a	21.91 ± 1.13ab	0.042
Gizzard index	5.99 ± 0.62	6.20 ± 1.04	6.16 ± 0.45	6.89 ± 0.90	6.23 ± 1.67	0.392
Glandular stomach index	21.59 ± 1.98	22.17 ± 3.24	22.28 ± 2.87	22.04 ± 1.64	21.96 ± 1.65	0.854
Pancreatic index	1.71 ± 0.28	2.09 ± 0.44	2.08 ± 0.40	1.91 ± 0.32	1.88 ± 0.22	0.113

Full eviscerated rate was significantly increased in the 1.6 g/kg MOLPs group compared with CK ($P < 0.05$). Abdominal fat rate was significantly decreased in the 1.2 g/kg MOLPs group ($P < 0.05$). Heart index was significantly increased in the 1.2 g/kg MOLPs group ($P < 0.05$). Liver index was significantly increased in the 1.6 g/kg MOLPs group ($P < 0.05$).

Slaughter performance of chicken

From Table III, it can be seen that the clean bore rate of MOLP group at 1.6 g/kg is significantly higher than that of CK group ($P < 0.05$). The abdominal fat percentage of MOLP group decreased significantly ($P < 0.05$); The heart index of *Moringa oleifera* L. polysaccharide group was significantly higher than CK, 0.8 g/kg and 2.0 g/kg *Moringa oleifera* L. polysaccharide groups ($P < 0.05$); At the same time, the liver index of Guangxi Sanhuang chickens was significantly increased by adding *Moringa oleifera* L. polysaccharide group ($P < 0.05$).

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Table IV. Effect of MOLPs on meat quality of breast and thigh muscles (42 days).

Item	CK	T1 (0.8 g/kg)	T2 (1.2 g/kg)	T3 (1.6 g/kg)	T4 (2.0 g/kg)	P-value
Thigh muscle						
Lightness (L*)	46.96 ± 1.68a	46.40 ± 2.46ab	45.53 ± 1.34ab	45.92 ± 0.87ab	45.06 ± 1.30b	0.028
Redness (a*)	7.43 ± 1.20	7.56 ± 1.20	7.73 ± 1.73	7.34 ± 1.16	7.70 ± 1.62	0.119
Yellowness (b*)	8.33 ± 0.71	8.12 ± 0.94	8.31 ± 0.83	8.06 ± 0.62	8.49 ± 0.94	0.493
pH45min	6.05 ± 0.10	6.06 ± 0.17	6.12 ± 0.42	6.04 ± 0.24	6.08 ± 0.39	0.721
Shear force (N)	23.21 ± 2.67a	22.62 ± 2.35ab	20.81 ± 1.05b	21.87 ± 1.51ab	22.89 ± 2.56ab	0.013
Breast muscle						
Lightness (L*)	48.31 ± 1.58	47.16 ± 1.99	48.03 ± 3.73	46.95 ± 3.41	46.72 ± 2.20	0.417
Redness (a*)	6.41 ± 0.88	6.22 ± 0.66	5.94 ± 0.55	6.51 ± 0.86	6.61 ± 0.62	0.182
Yellowness (b*)	7.39 ± 0.55	7.58 ± 0.39	7.75 ± 0.46	7.33 ± 0.92	7.26 ± 0.95	0.404
pH45min	5.94 ± 0.11b	6.08 ± 0.17ab	6.10 ± 0.09a	6.05 ± 0.16ab	6.03 ± 0.13ab	0.01
Shear force (N)	16.78 ± 2.41a	13.48 ± 3.08b	13.94 ± 1.71b	14.40 ± 1.71ab	15.27 ± 2.58ab	0.042

Shear force of both breast and thigh muscles was significantly reduced in the 0.8 and 1.2 g/kg MOLPs groups ($P < 0.05$). Breast muscle pH (45 min) was significantly increased in the 1.2 g/kg MOLPs group ($P < 0.05$). Thigh muscle L* (lightness) was significantly decreased in the 2.0 g/kg MOLPs group ($P < 0.05$). These results demonstrate that dietary MOLPs at 1.2–1.6 g/kg improve growth performance (lower F/G, higher ADG and final BW), enhance slaughter performance (higher full-eviscerated rate, lower abdominal fat), and improve meat quality (lower shear force, higher pH).

Meat quality of chicken

As can be seen from Table IV, the leg muscle color brightness (L*) of the MOLP group at 2.0 g/kg was significantly lower than that of the CK group ($P < 0.05$), but there was no significant difference in red (a*) and yellow (b*) of meat color ($P > 0.05$). Compared with CK group, the pH value of pectoral muscle was significantly increased and the shear force of leg muscle was significantly decreased in MOLP group (1.2g/kg), ($P < 0.05$). At the same time, 0.8 g/kg and 1.2 g/kg MOLP groups also significantly decreased the shear force of pectoral muscle. In addition, MOLP supplementation had no significant effect on breast muscle color brightness (L*), red color (a*) and yellow color (b*) of Guangxi three-yellow chicken. The effect of MOLP on the slaughter performance of Guangxi Sanhuang chickens.

DISCUSSION

Moringa is rich in nutrients, especially rich in protein, vitamins and mineral elements, and can be used as a source of animal protein feed. *Moringa* leaf crude extract to Tianfu broilers, which could significantly increase the average daily gain of broilers and reduce the feed weight ratio. *Moringa* leaf powder was used to replace alfalfa powder. When the addition amount of *Moringa* leaf powder was 20%, it could significantly improve the average daily weight gain and feed conversion rate of meat rabbits, and could significantly improve spleen index, and promote the growth performance and development of immune organs of meat rabbits to some extent (Sun *et al*., 2018), adding 0.2% Moringa crude flavonoids to duckling feed could improve feed conversion rate and increase serum immunoglobulin content. In this study, 42-day feeding experiment was conducted on Guangxi Sanhuang chickens. The results showed that compared with CK group, adding 1.2 g/kg and 1.6 g/kg MOLP in diet significantly decreased F/G of Guangxi Sanhuang chickens at different feeding stages; meanwhile, adding 1.6 g/kg MOLP in diet significantly increased BW at 42 days and ADFI and ADG at 1-42 days. The results showed that MOLP could promote the growth and development of Guangxi Sanhuang chickens, increase the weight of broilers, increase the utilization rate of feed and promote the intake of feed, which was basically consistent with the above results. Plant polysaccharides have a very strong antioxidant effect. They can remove oxidative free radicals from animals and reduce the stress response suffered by animals during feeding. Moreover, plant polysaccharides can not only stimulate the growth of beneficial bacteria in the intestine, optimize the microbial community in the intestine, but also have a positive effect on the development of the intestine.

This effect can further promote the digestion and absorption of nutrients in broilers. The polysaccharide extracted from the plant can promote the development of the intestines of poultry, facilitate the entry of nutrients into the body to improve the digestion, absorption and metabolism, and reduce the feed weight ratio, so as to promote the growth of poultry (Liu *et al*., 2021). In summary, MOLP can improve the growth performance

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by improving the digestion and absorption capacity of nutrients and anti-stress ability of Guangxi Sanhuang chicken. Slaughter performance is an important index to measure ketone body quality and economic benefits of broilers. It has been found that, when the abdominal fat rate of broilers is too high, it will affect the processing of meat products and reduce consumers 'desire to buy (Zhang et al., 2017). In this study, 0.8 g/kg and 1.2 g/kg MOLP significantly decreased abdominal fat percentage of Guangxi Sanhuang chicken, 1.6 g/kg MOLP significantly increased total carcass percentage, carcass percentage, half-carcass weight, breast muscle percentage and leg muscle percentage, but the difference was not great. Similarly, some research results show that the addition of Moringa extract has no significant effect on the slaughter rate, half-bore weight and full-bore weight of Guangxi partridge, but significantly reduces the abdominal fat rate of partridge. *Moringa* leaf meal in different levels in the diet of meat geese aged 22-70 days. The results indicated that the slaughter rate, semi-clean bore rate and leg muscle rate of meat geese were not influenced by Moringa leaf meal, but the breast muscle rate increased linearly with the addition amount of *Moringa* leaf. When the addition amount was 100mg/kg, the abdominal fat rate was the lowest. Organ index can reflect the development of animal internal organs to a certain extent (Zhai *et al*., 2020). The growth and development of animal organs can affect the physiological functions of the body. The heart plays a vital role in maintaining normal metabolism of the body. It can provide oxygen and nutrients to various organs and tissues of the body by transporting blood. The liver is the largest detoxification organ of the body and is vulnerable to a number of stress sources, thus affecting the metabolic stability of the liver and the body system. The liver is important for digestion and absorption and has an important function in the process of lipid and lipoprotein uptake, synthesis and secretion (Trefts *et al*., 2017). The pancreas is an important digestive organ, secreting various digestive enzymes. The digestive enzymes play an important role in the digestion of nutrients such as proteins and carbohydrates in food (Chen *et al*., 2020). In this study, compared with CK group, dietary supplementation of MOLP at 1.2 g/kg significantly increased heart index, and dietary supplementation of MOLP at 1.6 g/kg significantly increased liver index. The increase of liver index may be due to the antioxidant activity of polysaccharide can reduce lipid peroxidation in liver and improve antioxidant enzyme antioxidant effect (Abdel *et al*., 2020). *Moringa* and some active compounds in *Moringa* extract have protective effects on the heart, reducing heart damage due to oxidative stress (Mabrouki *et al*., 2020), and *Moringa* can also improve heart damage, protect heart function and promote heart development through anti-inflammatory effects (Li *et al*., 2020), which may be the reason for the increase in cardiac index. To sum up, MOLP can improve the carcass performance of Guangxi Sanhuang chicken by increasing the total carcass rate, reducing the abdominal fat rate, and improving the physiological function of the body by promoting the development of tissues and organs.

Meat color, pH and shear force are important indicators reflecting the freshness, taste and tenderness of livestock and poultry muscle. Meat color reflects the freshness of muscle, usually measured by brightness (L*), redness (a*) and yellowness (b*), redness is mainly affected by myoglobin and hemoglobin in muscle (Zahedi *et al*., 2016); brightness is related to muscle aging, the greater the brightness value, the older the meat quality (McKenna *et al*., 2005); Yellowing is mainly associated with the yellow pigment content in muscle fat. Yellowness value pH is an important factor affecting meat flavor The higher the fat content (D'Alessandro, *et al*., 2012). Too fast a decrease in pH after slaughter will seriously affect the tenderness of meat and the muscle water retention will be poor. Muscle tenderness can be evaluated by shear force. The smaller the shear force, the greater the muscle tenderness (Li *et al*., 2019). In addition, meat quality indicators can directly reflect the oxidation level of muscle to a certain extent, the more More oxidation damages the muscle biofilm and structural functions, the muscle cells lose water and the muscles shrink, resulting in poor meat color, stale meat, and shortened shelf life. Studies have suggested that plant polysaccharides can decrease excessive oxidative damage and improve the quality of muscle (Mu *et al*., 2021). When different doses of *Artemisia annua* polysaccharide were added to AA broilers diet, L* value and a* value of leg muscle of AA broilers increased linearly, shear force decreased linearly, and pH value of leg muscle had no significant difference at 45min and 24h after slaughter; 0.5% and 1.0% *Lycium barbarum* polysaccharide added to diet could significantly reduce pH value of broilers 45min after slaughter and significantly increase muscle redness (a*) value, compared with the control group, adding alfalfa polysaccharide at 1000 mg/kg and 2000 mg/kg in the diet could significantly reduce the shear force of hen leg muscle, adding alfalfa polysaccharide at 1000 mg/kg could significantly increase the pH value of cock breast muscle at 45 min, and significantly increase the pH value of hen leg muscle at 24 h. Compared with the control group, the shear force of each leg muscle of broiler chest muscle in the *Lycium barbarum* polysaccharide group was significantly reduced, and the b* value of chest muscle and leg muscle in the *Lycium barbarum* polysaccharide group with 500 mg/kg was significantly lower than that in the control group, while the L* value and a* value had no significant

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difference (Chang *et al*., 2023). In this study, the L* value, a* value and b* value of chest muscle and leg muscle in the five different treatment groups were all positive, indicating that the meat color was bright; Compared with CK group, L * value of breast muscle and leg muscle of Guangxi Sanhuang chicken decreased significantly in MOLP group (2.0 g/kg), and a* value of breast muscle and leg muscle increased in different degrees; shear force of breast muscle and leg muscle decreased significantly in MOLP group (1.2 g/kg), and that of breast muscle decreased significantly in MOLP group (0.8g/kg) and MOLP group (1.2 g/kg); pH value of breast muscle decreased significantly in MOLP group (1.2 g/kg). The increase of a* value in this study may be related to the synthesis of myoglobin and the accelerated deposition of fat in pectoral muscle and leg muscle (Li *et al*., 2020); The larger the L* value, the older the meat quality. In this study, MOLP decreased the L* value in pectoral muscle and leg muscle, which may be due to MOLP's antioxidant effect, reducing the oxidation rate of free radicals to muscle and protecting the integrity of muscle cell membrane. Cell membrane phospholipids can weaken lipid peroxidation damage, thus prolonging muscle freshness and improving muscle mass (Raja *et al*., 2016).

CONCLUSION

Dietary supplementation of *Moringa* polysaccharides (MOLPs) at 1.2–1.6 g/kg obviously promoted the growth performance, slaughter performance and meat quality of Guangxi Sanhuang chickens. MOLPs, specifically, decreased feed conversion ratio, increased final body weight and average daily gain, increased full-eviscerated rate and heart/liver indices, decreased abdominal fat rate, decreased shear force, but increased breast muscle pH. No untoward effects were seen. The optimum level of supplementation was 1.2–1.6 g/kg, supporting MOLPs as a promising antibiotic-free feed additive in poultry production.

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IRB approval

This study involved only broiler chickens and did not include human subjects. All animal experiments were conducted in accordance with the guidelines of the Animal Ethics Committee of SAU, Sindh, Pakistan. The study protocol was approved by the same committee under approval number 2019-821.

Ethical statement

The study protocol was approved by the Animal Ethics Committee of Sindh Agriculture University (SAU), Sindh, Pakistan, under approval number 2019-821.

Generative AI and AI-assisted technology statement

The authors declare that no generative artificial intelligence (AI) tools or AI-assisted technologies were used in data analysis, preparation and writing of this manuscript.

Statement of conflict of interest

The authors have declared no conflict of interest.

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