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Effect of *Moringa oleifera* Leaves Extract, Whey Protein, and Their Combination on Growth, Carcass and Meat Quality of Broiler Chickens

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ABSTRACT

Restriction of synthetic antioxidants and prohibition of antibiotic growth promoters (AGP) have had an impact on impairing the growth rate of broiler chickens, and hence the alternatives for these additives are urgently needed by broiler farmers. The present study aimed to investigate the effect of *Moringa oleifera* leaves extract (MOLE), whey protein or their combination on the growth, carcass and meat quality of broilers. A total of 336 broiler chicks were arranged into four groups, including T0 (basal diet with no additive), T1 (basal diet with 1% MOLE), T2 (basal diet with 1% whey protein powder) and T3 (basal diet with 0.5% MOLE and 0.5% whey protein powder). Body weight and feed intake were recorded weekly. Internal organs and meats were obtained on day 42. Results showed that MOLE impaired body weight, body weight gain and feed conversion ratio of broilers ($p<0.05$). MOLE, whey protein and their blend decreased the abdominal fat content of broilers ($p<0.05$). Whey increased the moisture content of breast meats ($p<0.05$). Water holding capacity (WHC) was higher in T2 than in the other breast meats ($p<0.05$). Among the groups, pH value was highest in T2 breast meat ($p<0.05$). The lightness values of breast meat were lower in T2 than in the other groups ($p<0.05$), while the lowest yellowness values were found in T2 breast meat ($p<0.05$). The WHC was higher in T1 thigh meat than in T2 and T3 ($p<0.05$). The T1 thigh meat showed higher pH than the T2 group ($p<0.05$). The T1 thigh meat showed higher redness values than the other groups ($p<0.05$). In conclusion, MOLE reduced broiler growth and abdominal fat deposition. Whey reduced fat deposition and improved the meat quality of broilers.

Keywords: antioxidants; broiler chickens; Moringa oleifera; weight gain; whey protein

INTRODUCTION

Broiler chicken is a type of poultry that grows very fast, but because of their high metabolic rate and very intensive rearing system, they frequently undergo stress (Nawab *et al.,* 2018; Sugiharto, 2021). To alleviate the negative impact of stress that can inhibit the growth rate, broiler producers have long used synthetic antioxidants that are commercially available. Currently, there is a tendency to reduce the use of synthetic antioxidants and encourage the use of natural antioxidants that are safe for consumers' health. Among the natural sources of antioxidants is *Moringa oleifera* leaves (Moreno-Mendoza *et al.,* 2021). Various flavonoid compounds are found in *M. oleifera* leaves, which are very useful as a source of antioxidants (Makita *et al.,* 2016). Apart from being an antioxidant source, *M. oleifera* leaves also contain various active ingredients (e.g., carbohydrates, cardiac glycosides, saponins, terpenes, steroids, flavonoids and alkaloids) that can function as growth promoters and antibacterial agents (Tesfaye *et al.,* 2013; Alabi *et al.,* 2017). Thus, *M. oleifera* leaves can be used as an alternative to antibiotic growth promoters (AGP), which are currently banned in most parts of the world (Sugiharto, 2021). In broiler studies, *M. oleifera* leaves increased feed efficiency and body weight gain (Nkukwana *et al.,* 2014; Alabi *et al.,* 2017). In addition, *M. oleifera* leaves improved carcass traits and the quality of broiler meats (Mickdam *et al.,* 2022).

Whey, which is a by-product of the cheese-making process, has widely been established to have antioxidant capacity due to the presence of peptides derived from *β*-lactoglobulin and *α*-lactalbumin (Corrochano *et al.,* 2018). Likewise, the presence of some free amino acids, including Trp, Phe, Tyr, Cys, and His may contribute to the antioxidant potential of whey (Corrochano *et al.,* 2018). Besides having a complete amino acid content, whey contains a high concentration of branched-chain amino acids (BCAAs; including leucine, isoleucine and valine), which can act as growth promoters (Anthony *et al.,* 2001) for broiler chickens. In the *in vivo* study, Afkhami *et al.* (2020) reported that the inclusion of whey in feed improved the growth performance and antioxidant status of broiler chickens. In line with the above study, Ashour *et al.* (2019) reported that in addition to improving growth rate, the use of whey protein in feed increased the carcass and breast percentages as well as improved the meat quality of broiler chickens.

A study showed that whey protein can increase the bioavailability of the active components contained in an ingredient (Bortlik *et al.,* 2009). Based on this property, whey, in addition to being an antioxidant source and growth promoter, may also be used as a carrier, coating (encapsulant) or be mixed with other ingredients to increase the bioavailability and effectiveness (growth-promoting effect) of the ingredient for broiler chickens (Bortlik *et al.,* 2009). In this study, *M. oleifera* leaves extract (MOLE) was combined with whey protein powder to exert the synergistic effect between both ingredients as well as to maximize the potential of MOLE as a feed additive in improving the growth rate and meat quality of broiler chickens. To the best of our knowledge, such a combination of MOLE and whey protein has never been published to date. Therefore, The present study aimed to investigate the effect of MOLE, whey protein or their combination on broilers' growth, carcass and meat quality.

MATERIALS AND METHODS

Ethical Approval

The Committee of Animal Ethics of the Faculty of Animal and Agricultural Sciences, Universitas Diponegoro, approved the present experiment (58-04d/A-6/KEP-FPP, year 2022).

In Vivo Study

A total of 336 broiler chicks with an average body weight of 45.75±0.96 g were used in the current trial. The experiment was arranged based on a completely randomized design with four treatment groups and seven replicates (pens) consisting of 12 chicks in each replicate. The treatments included T0 (basal diet with no additive), T1 (basal diet with 1% MOLE), T2 (basal diet with 1% whey protein powder) and T3 (basal diet with 0.5% MOLE and 0.5% whey protein powder). The treatments were applied from days 1 to 42 of age. The MOLE and whey protein powder were added at the expense of the basal diets. The MOLE and whey protein powder were added ("on top") to the basal feeds at the end of the mixing process and then thoroughly mixed. From arrival until day 7, the chicks were reared on commercial pre-starter feed containing 23% crude protein, 5% crude fat, 5% crude fibre and 7% ash (according to the feed label). The chicks were offered formulated starter and finisher feeds (Table 1) from day 8 onward. The MOLE was purchased from the local pharmaceutical industry (PT. Borobudur, Semarang, Central Java, Indonesia [the extraction procedure of *M. oleifera* leaves is concealed by the company]). The MOLE contains a total flavonoid of 14.295% and tannins 59.909%. Whey protein powder was bought from Davisco (80% crude protein; Davisco Foods International, Le Sueur, Minnesota, USA). The birds were raised in an open-sided poultry house with rice husks as bedding throughout the experiment. Drinking water and feed were given *ad libitum* using a manual feeder and drinker in each pen. Continuous lighting was applied all day long, and during the trial, the airflow inside the broiler house was controlled using an installed tarp and blowers. The chicks were vaccinated against Newcastle Disease (ND) and Infectious Bronchitis (IB) using Medivac ND-IB vaccines through eye drops at day 4 of age. The chicks were also vaccinated with Medivac Avian Influenza (AI) vaccine through subcutaneous injection on day 11 of age. The Medivac Gumboro A vaccine was also given (through drinking water) to chicks on day 11.

Weekly records were carried out on feed consumption, body weight gain and feed conversion ratio (FCR) of broilers. By dividing the feed intake by body weight gain, the FCR was obtained. Income over feed cost (IOFC) was calculated based on total revenue minus total feed cost at the time of the experiment. One male chick (representing the average body weight of the chickens in pen) per pen was slaughtered (according to Islamic law), de-feathered and eviscerated following 12-hour fast at the end of the trial (day 42). Internal organs were removed and weighed (empty condition). Meat samples (breast and thigh meats) were also collected for meat quality determination. As a proportion of the live body weight, the eviscerated carcass was calculated. The commercial cuts were calculated as the percentages of the eviscerated weight.

Table 1. Ingredients and nutrient contents of experimental basal diets for broiler chickens

Items	Starter (day 8-21)	Finisher (day 22-42)
Ingredients		
Yellow maize (%)	57.8	61.0
Soybean meal (%)	34.9	32.0
Palm oil (%)	2.52	2.95
DL-methionine (%)	0.19	0.19
Bentonite (%)	1.00	0.75
Limestone (%)	1.34	1.00
Monocalcium phosphate (%)	1.51	1.30
Premix (%)[1]	0.27	0.34
Chlorine chloride (%)	0.07	0.07
Salt (%)	0.40	0.40
Analysed nutritional compositions:		
Metabolizable energy (kcal/kg)[2]	3,386	3,433
Crude protein (%)	20.8	18.1
Crude fibre (%)	3.53	3.24
Crude fat (%)	2.39	3.26
Ash (%)	6.96	6.99

Note: [1]Premix contained (per kg of diet) of Vitamin A 7750 IU, Vitamin D3 1550 IU, Vitamin E 1.88 mg, Vitamin B1 1.25 mg, Vitamin B2 3.13 mg, Vitamin B6 1.88 mg, Vitamin B12 0.01 mg, Vitamin C 25 mg, folic acid 1.50 mg, Ca-d-pantothenate 7.5 mg, niacin 1.88 mg, biotin 0.13 mg, Co 0.20 mg, Cu 4.35 mg, Fe 54 mg, I 0.45 mg, Mn 130 mg, Zn 86.5 mg, Se 0.25 mg, L-lysine 80 mg, choline chloride 500 mg, DL-methionine 900 mg, $CaCO_3$ 641.5 mg and dicalcium phosphate 1500 mg.
[2]Metabolizable energy was calculated according to Bolton (1967) formula: 40.81 {0.87 [crude protein + 2.25 crude fat + nitrogen-free extract] + 2.5}.

Laboratory Analysis

A digital pH meter (Hanna Instruments, Woonsocket, Rhode Island) was used to measure the pH after homogenizing the meats (breast or thigh) from each sample in 9 mL of distilled water. With a digital colour meter configured to CIE Lab, the colour of the breast or thigh meat was measured in Mac OS X. The L* (lightness), a* (redness) and b* (yellowness) values were used to depict the colour of the breast and thigh meats. Standard proximate analysis (AOAC, 2007) was used to determine the chemical quality of the meat utilizing the oven method for water content, the Kjeldahl method for protein content, the Soxhlet method for fat content and the dry ash method for ash content. Using filter paper, the press method was used to assess the water-holding capacity (WHC) of meats (Grau & Hamm, 1953). Cooking loss was determined as the difference in weight between the raw and cooked meat samples. The raw meat samples were stored at 4 °C for 48 hours, removed, and the water on the surface was wiped off with bibulous paper before being weighed (W_1). The meat samples were placed in zip-sealed polyethylene bags and heated in a water bath at 85 °C for 20 minutes. The meat samples were then cooled at room temperature, dried and reweighed (W_2). The cooking loss was calculated as (%) = $(W_1-W_2)/W_1 \times 100\%$.

Statistical Analysis

Data collected from this study were statistically analyzed based on analysis of variance (ANOVA; Steel & Torrie, 1997). Duncan's multiple-range test was conducted when significant differences ($p<0.05$) were found among the treatment groups.

RESULTS

Growth Performance of Broiler Chickens

Data on the growth performance of broiler chickens fed MOLE, whey protein or a combination of both are presented in Table 2. Final body weight, body weight gain, and IOFC were lower in T1 than in T0 and T2 but did not differ from the T3 groups ($p<0.05$). The FCR tended to be higher in T1 than in the other groups of broilers ($p=0.057$). Accumulative feed intake did not vary across the groups of treatments ($p>0.05$).

Internal Organ Weight of Broiler Chickens

The relative weight of the small intestine was higher in T1 as compared to that in other groups of broiler chickens ($p<0.05$). The chicks in T1, T2, and T3 had lower abdominal fat deposition than that in the T0 group ($p<0.05$). The relative weight of the heart, liver, proventriculus, pancreas and caecum did not considerably vary among the broiler chickens (Table 3).

Carcass Traits of Broiler Chickens

The data on the carcass proportion of broilers are presented in Table 4. Treatments had no significant effect on the eviscerated carcass of broilers ($p>0.05$). Whereas the proportions of breast, wings, drumsticks and back did not differ among the treatment groups, the proportion of thigh was lower in T1 than in T0 ($p<0.05$) but did not differ from those in T2 and T3 groups.

Meat Characteristics of Broiler Chickens

The data on the chemical and physical characteristics of broiler breast and thigh meats are shown in Table 5. Observation of breast meat showed that moisture content was higher in T2 than those in the other groups ($p<0.05$). The T3 had higher ash content than the T2 ($p<0.05$), but the difference was not observed when compared with the T0 and T1 groups. The WHC was higher in T2 than in the other breast meats ($p<0.05$). The T2 had the highest pH values among the breast meats ($p<0.05$). The L* values were lower in T2 than in the other groups, while the lowest b* values were observed in the T2 group ($p<0.05$). Data on thigh meats showed that WHC was higher in T1 than in T2 and T3 ($p<0.05$) but did not differ from that in the T0 group. The T1 showed higher pH than the T2 group ($p<0.05$) but did not vary from those in the T0 and T3 groups. The T1 thigh meat showed higher a* values than those of the other groups ($p<0.05$).

DISCUSSION

Growth Performance of Broiler Chickens

In contrast to most of the studies (Mahfuz & Piao, 2019; Ullah *et al.*, 2022), the use of MOLE resulted in a lower final body weight and a higher FCR compared to

Table 2. Growth performance of broiler chickens administrated with MOLE, whey protein, or their combination

Variables	Treatments				SEM	p-value
	T0	T1	T2	T3		
Final BW (g/bird)	1,949^a	1,787^b	1,964^a	1,861^{ab}	22.4	0.009
BWG (g/bird)	1,627^a	1,495^b	1,639^a	1,551^{ab}	21.0	0.039
Accumulative FI (g/bird)	3,273	3,259	3,329	3,156	25.1	0.094
FCR	1.68	1.83	1.69	1.70	0.02	0.057
IOFC (IDR/bird)	10,071^a	6,332^b	9,942^a	8,975^{ab}	529	0.033

Note: Means in the same row with different superscripts differ significantly ($p<0.05$). BW= body weight; BWG= body weight gain; FI= feed intake; FCR= feed conversion ratio; IOFC= income over feed cost; IDR= Indonesian Rupiah (Indonesian currency); T0= basal diet with no additive; T1= basal diet with 1% *Moringa oleifera* leaves extract (MOLE); T2= basal diet with 1% whey powder; T3= basal diet with 0.5% MOLE and 0.5% whey powder; SEM= standard error of means.

Table 3. Internal organ weight over live-body weight of broiler chickens administrated with MOLE, whey protein, or their combination

Variables (%)	Treatments				SEM	p-value
	T0	T1	T2	T3		
Heart	0.48	0.50	0.46	0.54	0.03	0.794
Liver	2.38	2.67	2.35	2.27	0.07	0.183
Proventriculus	0.52	0.50	0.47	0.52	0.01	0.437
Gizzard	1.62	1.83	1.72	1.59	0.04	0.133
Pancreas	0.24	0.23	0.27	0.25	0.01	0.658
Small intestine	2.20[b]	2.88[a]	2.14[b]	2.39[b]	0.08	0.001
Caecum	0.36	0.37	0.33	0.38	0.02	0.763
Abdominal fat pad	1.61[a]	1.01[b]	1.20[b]	1.11[b]	0.07	0.005

Note: Means in the same row with different superscripts differ significantly (p<0.05). T0= basal diet with no additive, T1= basal diet with 1% *Moringa oleifera* leaves extract (MOLE), T2= basal diet with 1% whey powder, T3= basal diet with 0.5% MOLE and 0.5% whey powder, SEM= standard error of the means.

Table 4. Carcass traits weight of broiler chickens administrated with MOLE, whey protein, or their combination

Variables (%)	Treatments				SEM	p-value
	T0	T1	T2	T3		
Over live-BW						
Eviscerated carcass	71.0	70.4	73.7	75.3	0.82	0.109
Over eviscerated carcass						
Breast	34.2	34.3	34.5	34.6	0.31	0.967
Wings	10.7	11.6	11.0	11.1	0.15	0.306
Thigh	18.0[a]	16.1[b]	17.0[ab]	16.9[ab]	0.23	0.021
Drumsticks	15.1	15.4	15.1	15.2	0.18	0.965
Back	21.9	22.7	22.3	22.3	0.31	0.871

Note: Means in the same row with different superscripts differ significantly (p<0.05). BW= body weight; T0= basal diet with no additive; T1= basal diet with 1% *Moringa oleifera* leaves extract (MOLE); T2= basal diet with 1% whey powder; T3= basal diet with 0.5% MOLE and 0.5% whey powder; SEM= standard error of means.

the control and the other treatment groups. So far, it is unknown why MOLE negatively impacts the growth and FCR of broiler chickens. Yet, it was very likely that the content of anti-nutritional substances in MOLE, especially tannins (Wahyuni *et al.*, 2020), could impair the growth rate of broilers. In such cases, the presence of anti-nutritional components may impede nutrient digestion, resulting in nutrient insufficiency. This nutritional imbalance may slow down metabolism and, as a result, hinder the growth of broilers. As mentioned above, the MOLE used in the current study contained 59.909% tannins. Hence, according to calculations, the feed applied in the T1 group had a tannin content of around 5.99 g/kg. Indeed, Jamroz *et al*. (2009) revealed that at 5 g/kg of feed, tannins compromised the growth performance of broiler chickens. Considering the facts mentioned above, the variation in nutritional contents of MOLE may determine the efficacy of such an additive in promoting the growth rate of broilers. Another possibility for the reduced growth rate of broilers in the T1 group was that the administration of MOLE reduced the proportion of abdominal fat, thereby decreasing the chickens' overall body weight gain. Compared to T0 chickens, the MOLE reduced the abdominal fat content by 37.27% in T1 chickens. Fouad & El-Senousey (2014) revealed that modern broilers contain 15% to 20% fat in their body. Considering that abdominal fat is directly correlated with total body fat content in broilers (Fouad & El-Senousey, 2014), the decreased abdominal fat content on T1 chicks seemed to correlate with the decrease in total body fat and broiler body weight. The latter inference should, however, be interpreted with caution, as in most cases, an increase in body weight in chicken will result in a concomitant increase in abdominal fat and *vice versa* (Santoso & Sartini, 2001). In other words, the decrease in abdominal fat is the result of a decrease in the body weight of the chicken and *vice versa*. Yet, the inference mentioned above might be disproved by the current observation that an increase in abdominal fat did not accompany the increase in body weight in the T2 group, as was the case in the T0 group.

Unlike Pineda-Quiroga *et al*. (2018), Alwaleed *et al*. (2020) and Afkhami *et al*. (2020), the dietary inclusion of whey protein powder in this study had no impact on the growth rate and efficiency of feed used by broiler chickens. Nonetheless, the results in this study were in line with those reported by Ashour *et al*. (2019), who found no whey protein effect on broiler chickens' final body weight at 42 days of age. The exact reason why whey protein did not affect broiler chickens' growth is still unclear. However, it was very possible that the lactose content in whey protein powder may lead to small intestinal disorders due to the purgative properties of lactose (Majewska *et al.*, 2009). The latter condition may therefore attenuate the growth-promoting effect of whey on broiler chickens. Note that poultry is lactose intoler-

Table 5. Chemical and physical characteristics of meats of broiler chickens administrated with MOLE, whey protein, or their combination

Variables	Treatments				SEM	p-value
	T0	T1	T2	T3		
Breast meat						
Moisture (%)	75.2[b]	74.9[b]	75.8[a]	75.1[b]	0.09	0.005
Crude protein (%)	21.8	21.5	21.1	21.4	0.11	0.190
Lipid (%)	0.83	0.78	0.86	0.80	0.21	0.495
Ash (%)	0.99[ab]	1.00[ab]	0.92[b]	1.13[a]	0.03	0.018
WHC (%)	35.2[b]	35.0[b]	36.9[a]	34.8[b]	0.20	0.000
Cooking loss (%)	30.1	31.2	31.0	30.3	0.19	0.124
pH	5.98[c]	6.03[ab]	6.05[a]	6.02[b]	<0.01	0.002
L*	52.9[a]	52.9[a]	51.6[b]	52.7[a]	0.18	0.020
a*	0.27	0.27	0.21	0.12	0.17	0.427
b*	5.91[b]	5.86[b]	5.35[c]	6.52[a]	0.09	0.001
Thigh meat						
Moisture (%)	76.3	76.3	75.9	75.8	0.10	0.131
Crude protein (%)	19.4	19.8	19.3	19.7	0.11	0.398
Lipid (%)	1.59	1.29	1.58	1.40	0.08	0.540
Ash (%)	1.02	1.00	1.04	1.03	0.01	0.909
WHC (%)	29.5[ab]	30.7[a]	29.4[b]	27.4[b]	0.41	0.032
Cooking loss (%)	36.5	36.3	37.0	37.5	0.18	0.087
pH	6.14[ab]	6.16[a]	6.13[b]	6.14[ab]	0.01	0.022
L*	51.3	51.1	52.1	51.4	0.30	0.686
a*	3.34[b]	6.18[a]	4.24[b]	4.47[b]	0.30	0.008
b*	6.39	7.77	7.62	6.73	0.26	0.171

Note: Means in the same row with different superscripts differ significantly (p<0.05). WHC= water holding capacity; L*= lightness value; a*= redness value; b*= yellowness value; T0= basal diet with no additive; T1= basal diet with 1% *Moringa oleifera* leaves extract (MOLE); T2= basal diet with 1% whey powder; T3= basal diet with 0.5% MOLE and 0.5% whey powder; SEM= standard error of means.

ant due to a lack of the lactase enzyme. In accordance with the study above, Hamilton & Card (1924) noted that feeding lactose to chickens over 2 g/day resulted in digestive problems and diarrhoea. Our current study did not specify the precise amount of lactose in the whey protein powder (not specifically mentioned on the label). Yet, according to the label, 20% of whey protein powder is made up of lactose in addition to minerals, carbohydrates and lipids, while 80% of whey is protein. The efficacy of whey protein powder in promoting the growth rate of broiler chickens was also very likely to depend on the protein quality of the whey (in terms of the type, amount and digestibility of each amino acid) (Ashour *et al.,* 2019). Unfortunately, the type, quantity and digestibility of the amino acids contained in the whey protein powder used in the current study were not identified and measured.

This study did not demonstrate the synergistic effect of MOLE and whey protein in enhancing the growth rate and feed efficiency of broilers. The cause of this condition is yet unknown, although it was possible that the lactose content in whey and the fat-lowering properties and the antinutrient contents of MOLE rendered the effect of their combination on weight gain unnoticeable. Another possibility was that the doses of MOLE (0.5% of feed) and whey protein powder (0.5% of feed) applied in T3 feed might not be sufficient to exert the complementary effect between both additives in improving the growth rate of broiler chickens.

Internal Organ Weight of Broiler Chickens

Dietary use of MOLE, whey protein or a combination of both decreased the abdominal fat content of broilers in the present study. This finding agreed with Cui *et al*. (2018) reporting the lower abdominal fat content in broilers with feeding *M. oleifera* leaf meal. In this regard, Cui *et al*. (2018) reported that several active compounds, such as flavonoids, phenols and polyunsaturated fatty acids (PUFA), are responsible for lipid metabolism so that they have an impact on reducing fat deposition in the abdomen of broiler chickens. Indeed, flavonoids and phenols (Tan *et al.,* 2022) and PUFA (Cui *et al*., 2018) can activate the fatty acid β-oxidation resulting in reduced abdominal fat content of broilers. However, this inference must be interpreted with caution since, referring to Santoso & Sartini (2001), as discussed above, low abdominal fat content in MOLE-fed broilers was very likely related to the low growth rate in these chickens. Regarding whey protein, our present finding agreed with Ibrahim *et al*. (2015), showing the reduced abdominal fat pad in broilers receiving whey protein concentrate. The latter authors suggested that the content of essential amino acids in whey plays an important role in lipid metabolism, especially in promoting fat burning. In line with this, Fouad & El-Senousey (2014) revealed that certain amino acids (methionine, lysine and arginine) could reduce the activity of fatty acid synthase (FAS; lipogenesis) and increase the activity of hormone-sensitive lipase (HSL) (lipolysis). These had an impact on reducing fat deposition in the abdomen of broiler chickens. It was apparent in the present study that the relative weight of the small intestine was higher in broilers receiving 1% MOLE in feed as compared to the other broilers. The exact explanation for the relatively high small intestine values in chicks that received MOLE was not known until recently. Indeed, the data on small intestine weight in this investigation was expressed as a ratio to the live weight of broiler chickens (chicken live weight was the denominator in the calculation). Given these circumstances, it was most likely that the low body weight of the chickens given MOLE was responsible for the elevated relative weight of the small intestine in this current study.

Carcass Traits of Broiler Chickens

Our present data showed no effect of treatments with MOLE, whey protein or a combination of both on the eviscerated carcass of broilers. This result was following that reported by Alwaleed *et al*. (2020), in which the use of *M. oleifera* leaves meal had no impact on the dressing percentage for broiler chickens. The same finding was also reported by Nkukwana *et al*. (2014), where the administration of *M. oleifera* leaves meal did not significantly affect broiler chicken carcasses. Besides the absence of MOLE on the eviscerated carcass, dietary treatment of MOLE reduced the proportion of broiler thighs in this study. So far, it was not known for certain about these conditions. Ncube *et al*. (2017) previously revealed that the use of leaf-based ingredients in feed greatly affected the proportions of the breast and thigh, possibly due to the higher nutritional requirements (especially protein) for the development of the breasts and thigh. They further pointed out that such reduced commercial cuts yield in broilers indicated that the birds could not attain the desired muscle tissue growth from the diet. Taken all these together, the presence of anti-nutritional components in the leaf-based ingredients (Wahyuni *et al.,* 2020) may reduce nutrient availability, especially protein, thus impacting inferior thigh yield in the present study.

Meat Characteristics of Broiler Chickens

This study showed that adding whey protein powder to feed increased the moisture content of broiler breast meats. Mir *et al*. (2017) revealed that moisture content and WHC in broiler chicken meats are positively correlated. Accordingly, our study demonstrated that the higher WHC accompanied the higher moisture content in whey-fed broiler breast meat in the corresponding meats. Indeed, our present results differed from that reported by Ashour *et al*. (2019) where supplementation of broiler chickens with whey protein concentrate had no impact on WHC and cooking loss of broiler breast meats. Currently, studies on the relationship between dietary whey protein and moisture content and WHC in broiler chicken meat are still scarce. In general, a rise in the moisture and WHC of chicken meat is accompanied by a rise in protein and a decline in the fat content of the meats (Mir *et al.,* 2017). Hence, we speculated that dietary whey administration might be able to raise the meat's protein content, which would affect raising the meat's moisture content and WHC. However, this inference must be viewed with caution, considering that the protein content in meat was not significantly different among treatment groups in this study. Different circumstances were observed in the thigh meats, where there was no difference in the moisture content among the treatment groups. Thigh meat from chickens receiving MOLE (1% of feed) actually had a greater WHC value than that of chickens provided with whey (1% of feed) or a combination of MOLE (0.5%) and whey powder (0.5%). Despite the fact that this condition remains unknown, it was most likely that the ability of MOLE to reduce fat in the thigh meat was positively connected with a rise in WHC in the thigh meat. As a note, the low-fat content (possessing hydrophobic trait) may be attributed to the increase in the WHC of meats (Mir *et al.,* 2017). However, this speculation must be interpreted with caution because even though the fat content in the MOLE-thigh meat was numerically lower compared to the other groups, the value did not reach a statistically significant level.

A study showed that chicken meat with pH ≥6.0 undergoes minimal protein denaturation, and conversely, chicken meat with pH <6.0 will undergo more protein denaturation (Mir *et al.,* 2017). This study showed that the pH values of breast meat from broilers receiving either MOLE, whey protein or a combination of both had a value >6.0, while breast meat in the control group had a pH <6.0. It was unknown why breast meat in treated chickens had higher pH values than the control. However, the ability of MOLE, whey and the combination of MOLE and whey to reduce the fat content in chicken meats made it possible for the meats to have a higher pH value than the control. According to Mir *et al*. (2018), there is a correlation between the pH value of broiler chicken meat and the fat content in the meats, which is relevant to this condition. The latter investigators showed that decreases in the fat contents in broiler breast and thigh meats were followed by an increase in the pH value of the meat. This presumption, however, needs to be interpreted very carefully because, despite the fact that MOLE, whey and their combination reduced the amount of abdominal fat in the broiler breast meat, the treatment could not significantly lower the amount of fat in the broiler meats. Unlike breast meat, thigh meats of broilers fed MOLE, whey, or a combination did not significantly differ from the control's pH. It can be assumed that since the pH in all treatment groups was >6.0, the thigh meats had minimal protein denaturation.

Based on the L* values, de Carvalho *et al*. (2018) distinguished broiler chicken meats into normal (44<L*<53) and Pale, Soft, Exudative (PSE; L*≥53). In this regard, all of the meats in this trial were included in the normal category. This study showed that the L* value was lower in the breast meat of broilers that were given whey in the feed when compared to the L* value in the other chicken breast meats. According to Mir *et al*. (2017), there is an inverse relationship between the L* value and pH in chicken meat. The same condition was also found in this study where the low L* value was accompanied by a high pH in whey-treated chicken meat. It was apparent in this study that b* values were lower in whey-treated chicken breasts compared to breast meat in the other chickens. A previous study by Abdurrahman *et al*. (2016) documented that there is a relationship between the b* value and the fat content in chicken meat. In this case, the fat-lowering effect of whey was very likely to reduce the fat content so that it has an impact on decreasing the b* value of the meat. However, such an interpretation must be carefully made because there was no substantial difference in fat content in broiler breast meat among treatment groups in this study. In thigh meat, the a* value was higher in the group of chickens given MOLE than the other chickens. Indeed, the high a* value implies low protein denaturation in broiler meat and positively corresponds with a high pH value (Mir *et al.,* 2017). Accordingly, the higher a* value was in line with the higher pH value in broiler thigh meat in this study. In agreement with this study, Cui *et al*. (2018) reported that using *M. oleifera* leaves meal increased the a* value of broiler meats. They further explained that the administration of *M. oleifera* leaves meal could improve oxidative stability so that myoglobin (a type of protein responsible for the colour of meat) in the meat was not much oxidized.

CONCLUSION

Dietary MOLE reduced the growth rate and abdominal fat deposition of broiler chickens. Whey protein powder reduced fat deposition and improved meat's chemical and physical qualities of broilers. Combining MOLE and whey protein did not exert a synergistic effect on the growth, carcass and meat quality of broilers.

CONFLICT OF INTEREST

The authors declared that they had no conflict of interest.

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