

The effect of administering aqueous extract of fermented *Moringa oleifera* leaves via drinking water on performance and carcass characteristics of broilers

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World Journal of Biology Pharmacy and Health Sciences, 2024, 20(01), 566–572

Publication history: Received on 19 September 2024; revised on 25 October 2024; accepted on 27 October 2024

Article DOI: <https://doi.org/10.30574/wjbphs.2024.20.1.0838>

Abstract

The need to adopt natural herbal plants as feed additives in poultry production has received attention among poultry nutrition and production experts in recent years. Herbal plants have few or no side effects, unlike synthetic additives. The aim of the research was to examine the effect of fermented *Moringa oleifera* leaf aqueous extract (FMLAE) on growth performance and broiler carcass characteristics. A total of 240 healthy day-old-chick broilers with homogeneous average body weight were randomly divided into 4 treatments and 6 replications with 10 heads per cage in a completely randomized design. Fermented *Moringa* leaf aqueous extract (FMLAE) treatment was given through drinking water at levels: 0% as control (A); 2% (B), 4% (C), and 6% (D), respectively. The results showed that FMLAE supplementation in drinking water had a significant increase ($P < 0.05$) in body weight gain, carcass weight, carcass percentage, feed consumption and feed efficiency. It can be concluded that the addition of 2-6% fermented *Moringa* leaf aqueous extract (FMLAE) to the drinking water of broilers aged 0-4 weeks significantly increases growth, carcass and feed efficiency.

Keywords: Broiler; Carcass characteristics; Fermented *Moringa* leaf; Performance

1. Introduction

Recently, the use of synthetic feed additives in poultry farming, especially broilers, has become a big concern, both for animal nutritionists and the end user, namely humans. The ban on the use of antibiotics as growth promoters, due to the negative and dangerous effects on livestock and humans who consume these products and their by-products, requires the search for alternative natural feed ingredients in animal diets [1].

Phytogetic feed additives in poultry production have been studied extensively for their impact on health, gut histology and gastrointestinal function, as well as their implications for growth and food safety of poultry products. This plant-based herbal feed, which contains bioactive compounds, can improve the digestive tract health of poultry [2] and has attracted a lot of interest from researchers, because of its possible use as an alternative to antibiotics as a growth promoter [1].

Medicinal plants are widely used in the development of herbal supplements because of their high profit margins, safety, cost-effectiveness, environmental friendliness and easy availability. This medicinal plant has a rich source of phytochemicals, such as flavonoids, phenolics, tannins and other compounds. This plant may provide beneficial effects, such as inhibiting radical scavenging activity, known as an antioxidant [3].

Individual plant extracts contain complex mixtures of bioactive compounds and various combinations of plant extracts, possibly providing interacting effects on antioxidant activity in the broiler gastrointestinal tract [3]. The interaction

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between herbs and probiotic microbes in the chicken's digestive tract can provide a synergistic impact between herbal phytochemicals and probiotics. This event is caused by the gut microbial community playing an important role in the overall health and digestion of broiler chickens [4]. This is proven by the research of [5] that adding fermented liquid feed to the ration can increase the levels of acetic acid, propionic acid and butyric acid in pig manure and improve broiler performance [6]. Probiotic fermented feed has been widely used in poultry production [7,8].

Generally, during the fermentation process, various anti-nutritional factors in animal feed are reduced, and the nutritional quality and bioavailability of nutrients are increased, along with the increase in beneficial metabolites, enzymes and probiotics, such as *Lactobacillus* and *Bacillus* [4]. The bacterial strains used to initiate fermentation can promote the degradation of complex carbohydrates, consisting of cellulose and hemicellulose, into short chain organic acids, thereby causing a lower pH, and can also create an anaerobic microecological environment in the intestine [9,10,11]. The probiotic activity of these fermented feeds has the potential to not only improve production performance but also modulate gastrointestinal ecology and metabolic behavior [4]. Supplementation of fermented feed products significantly increases the villus height (VH) of the jejunum and the ratio of villus height to crypt depth (VH/CD) of the ileum, which has an impact on increasing the absorption area, resulting in increased feed efficiency [10].

The aim of the research was to examine the effect of fermented *Moringa* leaf aqueous extract on growth performance and broiler carcass characteristics.

2. Material and methods

The research was carried out at the Research Station of the Faculty of Animal Husbandry, Udayana University on Jl. Raya Sesetan, Denpasar. The broiler used is the day-old-chick broiler strain CP 707 produced by PT. Charoen Pokphand Indonesia, Sidoarjo, East Java, Indonesia with homogeneous body weight. The cages used were colony cages which are made of wire and steel frames. The dimensions of each cage plot were: length 220 cm, width 150 cm and height 50 cm. Each cage contains 10 day-old-chick broilers with homogeneous body weight.

2.1. Experimental design

A total of 24 cages, each holding 10 chickens, were used for the experiment. Each cage was used as an experimental unit. The trial of providing drinking water had four treatments and six replications in total. Each group of chickens was randomly given fermented *Moringa* leaf aqueous extract (0, 2, 4, and 6 cc/100 cc of drinking water). Feed and water were provided *ad libitum*. All chickens were exposed to light for 24 hours per day during the growth and finishing phases.

2.2. *Moringa oleifera* Leaves Fermentation Process

Mature *Moringa* leaves harvested by hand from a *Moringa* plantation in Penebel Village, Tabanan, Bali, Indonesia. These leaves were harvested with minimal plant disturbance, allowing shoots to regrow. The leaf stalks were removed, then the leaves were cut into pieces measuring 1 to 3 cm using a shredder. The chopped leaves were fermented with yeast culture (*Saccharomyces spp.*) and molasses to increase the fermentation rate. *Saccharomyces spp.* was inoculated on *Moringa* leaves with a concentration of 2.9×10^7 cfu/g. Molasses was added at a rate of 5 g/100 g. Next, put it in a plastic bag filled with small holes (facultative anaerobe) and incubate for three days at room temperature (30°C). Then 1 kg of fermented leaves were taken and put into 1 liter of clean water, then blended and left for 24 hours (cold maceration) filtered through a satin cloth in duplicate to get the water extract (fermented *Moringa* leaf aqueous extract was considered 100%). This water extract was then mixed into the chicken's drinking water according to the treatment concentration (0, 2, 4, and 6 cc/100 cc of drinking water).

2.3. Observed Variables

The variables observed were initial body weight, final body weight (BW), live weight gain (WG) and feed intake (FI) which was measured every week. The feed conversion ratio (FCR) was calculated every week, i.e. comparison between FI and WG. At the end of the study, two chickens were slaughtered in each cage to observe carcass weight and carcass percentage (carcass weight/slaughter weight).

3. Results and discussion

3.1. Growth performance

The effect of providing drinking water with different FMLAE inclusion levels on the growth performance of broiler chickens is shown in Table 1. Total feed intake (FI) during the growth phase was not significantly different ($p>0.05$) for chickens fed with FMLAE. Final body weight (BW) and body weight gain (WG) were higher in chickens given drinking water containing FMLAE compared to chickens given control (drinking water without FMLAE). Body weight gain increased linearly ($p<0.05$) with the level of FMLAE inclusion in chicken drinking water. There was no significant ($P>0.05$) relationship between FI and drinking water consumption at inclusion level of FMLAE in drinking water of broiler.

Table 1 Effect of providing drinking water graded with aqueous extract of fermented *Moringa* leaves on broiler growth performance

Variable	FMLAE inclusion level in drinking water (cc/100 cc)				SEM
	0	2	4	6	
Initial body weight (g)	57.60a	57.56a	57.55a	57.65a	1.095
Final body weight (g)	1558.55b	1634.05a	1619.50a	1632.50a	22.721
Live weight gains (g/4 weeks)	1500.95b	1576.49a	1561.95a	1574.85a	21.986
Feed intake (g/ekor/4 weeks)	2598.80a	2630.60a	2601.90a	2637.95a	42.927
Drinking water (ml/ekor/4 weeks)	7843.34a	7816.45a	7804.73a	7824.51a	51.083
Feed Conversion Ratio(FI/WG)	1.73b	1.67a	1.67a	1.68a	0.013

Note: The different superscript within the same row are significantly different ($p<0.05$); SEM= Standard Error of the Treatment Means

The lowest FCR value or highest feed efficiency was in chickens given drinking water containing 2 and 4% FMLAE and the lowest feed efficiency was in the control (drinking water without FMLAE). At the final stage, feed efficiency was highest ($p<0.05$) in chickens given drinking water with FMLAE 2-4% (6cc/100cc drinking water). The FCR value decreased linearly ($p<0.05$) with the level of FMLAE inclusion in broiler drinking water.

3.2. Carcass characteristics.

The effect of providing drinking water with different FMLAE inclusion levels on the carcass characteristic of broiler is shown in Table 2. Slaughter weight and carcass weight were higher in chickens given drinking water containing FMLAE compared to chickens given control (drinking water without FMLAE). Slaughter weight and carcass weight linearly ($p<0.05$) with the level of FMLAE inclusion in broiler drinking water. There was no significant ($P>0.05$) in drumstick and thigh at inclusion level of FMLAE in drinking water of broiler.

Table 2 Characteristics of broiler carcasses given aqueous extract of fermented *Moringa* leaves in drinking water from 0-4 weeks of age

Variable	FMLAE inclusion level in drinking water (cc/100 cc)				SEM
	0	2	4	6	
Slaughter weight (g)	1524.6b	1641.8a	1640.6a	1641.2a	35.182
Carcass weight (g)	1083.4b	1173.8a	1172.4a	1172.2a	27.075
Carcass percentage (%)	71.04b	71.50a	71.46a	71.42a	0.059
Percentage of breast (% carcass weight)	40.93b	41.65a	41.67a	41.63a	0.194
Drumstick(% carcass weight)	14.42a	14.55a	14.51a	14.49a	0.058
Thigh (% carcass weight)	13.15a	12.87a	13.02a	12.99a	0.051

Note: The different superscript within the same row are significantly different ($p<0.05$); SEM= Standard Error of the Treatment Means

Inclusion of FMLAE in broiler drinking water significantly ($P < 0.05$) increased breast weight and broiler carcass percentage. Breast meat increases linearly with increasing FMLAE concentration in drinking water.

The results of the research found that the inclusion of 2-6% water extract of *Moringa* leaves fermented by tape yeast (*Saccharomyces spp.*) in the drinking water of broilers from 0-4 weeks of age could increase the final body weight and weight gain of broilers. This increase is due to *Moringa* leaves containing phytochemical compounds, such as saponins, flavonoids and tannins and several other phenolic compounds which have antimicrobial activity, for example saponins have been proven to have antimicrobial properties [12], and can increase WG and feed efficiency [13].

Supplementation of phytogenic compounds containing phytochemical compounds in drinking water can cause improvements in growth, stimulate digestion, nutrient utilization, immune response, antimicrobial pathogens, and reduce gas emissions in poultry feces [14,15]. The concept of phytogenic feed additives refers to natural medicinal products of herbal origin used in poultry nutrition to improve poultry performance and health [16].

Generally, phytogenic feed additives are used to improve the overall health of poultry, aid the digestive process and help detoxify the body [17]. However, phytogenic additives can also have much more specific effects, namely inhibiting the development of pathogenic microorganisms, regulating the composition and abundance of the gastrointestinal microbiome, promoting the regeneration of intestinal epithelium and villi, as well as exerting antioxidant, immune stimulating, anti-inflammatory, antimicrobial and neuroprotective effects [18]. The phytochemical compounds contained in *Moringa* leaves are: saponins, flavonoids and tannins [19], and contain beta-carotene, which is the active substance color to improve carcass and yolk color [20,21]. In broiler chickens, herbal leaf supplementation causes increased body weight gain, nutrient digestibility, and reduces NH_3 gas emissions [22,23]. Fermented products will also be able to increase the vitamin content that broilers really need to stimulate weight gain and broiler weight gain.

According to [24], during the fermentation process, the biosynthesis of vitamins, essential amino acids and protein occurs which can increase the nutritional content, resulting in an increase in the quality and digestibility of protein. Apart from that, according to [25], the enzyme activity produced by micro-organisms during the fermentation process causes chemical changes in the organic substrate. Probiotics in the chicken's digestive tract can increase nutrient digestibility, so that growth and feed efficiency can be optimal. As reported by [11] that supplementation with probiotic *Saccharomyces spp.* into the ration significantly increases egg production, egg mass, shell thickness, Mg and Ca content in egg shells.

Feed consumption (FI) in this study did not show any significant differences. There is no difference in FI because the metabolizable energy content of the rations is the same. Chickens consume food to meet their energy needs. Providing fermented *Moringa* leaf water extract in drinking water has not had a significant effect on FI. Research by [26] reported that feed consumption decreased significantly in chickens given herbal supplements compared to chickens not given herbal supplements. These results are similar to those reported by [11,27] that the addition of fermentation products by tape yeast (*Saccharomyces spp.*) in feed has no effect on feed consumption in ducks. Siti and Bidura [28] reported that giving herbal leaves to poultry significantly increased feed consumption. The results of this study are the same as those reported by [29] that the use of herbal leaves in feed has no effect on feed intake.

Feed efficiency in broiler groups B, C, and D was better than the control (group A). As with other herbal leaves, *Moringa* leaves contain high levels of beta-carotene. Beta-carotene or pro-vitamin A in *Moringa* leaves is converted into vitamin A which plays a role in epithelial cell differentiation and maintaining digestive organs, thus affecting digestibility and feed efficiency. Increasing feed efficiency and feed digestibility can influence protein synthesis and calcium intake, which can influence the high and low levels of protein mass and calcium mass in meat [30]. Restiayanti et al. [13] reported that giving herbal extracts to broilers can significantly increase body weight gain and feed efficiency. The increase in feed efficiency in treatments B, C, and D cannot be separated from the role of the yeast *Saccharomyces spp.* *Saccharomyces spp.* used in the fermentation process of *Moringa* leaves can act as a probiotic in the chicken's digestive tract, thereby increasing enzymatic activity and absorption of food substances [27,31]. This result is proven in research [11] which reports that the use of probiotics in feed can increase the digestibility of dry matter and organic feed, as well as improve the nutritional quality of feed.

Slaughter weight, carcass weight, carcass percentage, breast percentage of broiler carcasses increased with the provision of fermented *Moringa* leaf aqueous extract in broiler drinking water from 0-4 weeks of age. Carcass percentage is an important variable in determining the performance of the broiler carcass produced. The higher the carcass percentage value, the more economic value of poultry farming. Slaughter weight has a significant effect on carcass weight and carcass parts consisting of meat, bones and fat [32]. The difference in carcass percentage obtained is greatly influenced by age at slaughter, strain and quality of feed provided. Wahyono et al. [33] reported that

differences in breed and slaughter weight resulted in differences in carcass weight. However, differences in strain and slaughter weight did not affect the carcass percentage. *Moringa* leaf water extract contains phytochemical compounds which can reduce the number of pathogenic bacteria in the intestine, resulting in optimal nutrient absorption for the body and synthesis of meat (breast meat). Supplementation with *Moringa* leaf herbal extract will improve the morphology of the small intestine using different mechanisms [34], so that nutrient absorption can be optimal.

Moringa leaves contain secondary plant compounds, such as condensed tannins, which may inhibit their use in poultry [35,36], so their administration in feed needs to be combined with probiotic microbes. Probiotics as feed additives have been reported to improve nutrient digestibility, growth performance, ceecal microflora balance, plasma immunoglobulins, and chicken immunity [37]. This is proven by research by [38] which combines phytogenic additives with probiotics in feed, significantly increases nutritional intake, feed efficiency, quail health, and reduces production costs.

4. Conclusion

From the research results it can be concluded that FMLAE can be a nutritional supplement for chicken. Inclusion levels of 2-4cc/100cc of drinking water can be used in broiler chickens during the growth phase. Our findings also showed that the inclusion of FMLAE in chicken drinking water had no effect on feed consumption. Future research on the profile of phytochemical compounds in FMLAE is needed to assess its ability to suppress pathogenic bacteria in the broiler intestine.

Compliance with ethical standards

Acknowledgments

The author would like to thank the Head of the Research Station, Faculty of Animal Husbandry, Udayana University, Denpasar, Indonesia for his research room and laboratory facilities

Disclosure of conflict of interest

We declare that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

Statement of ethical approval

The experimental animals in this study were approved by the Animal Ethics Committee, Faculty of Veterinary Medicine, Udayana University, Denpasar, Indonesia.

References

- [1] Ampode KMB. Oregano leaf meal as phytoegenic feed supplements: A potential antibiotic alternative in broiler chicken diets. *Adv. Anim. Vet. Sci.*, 2024; 12(7):1362-1370.
- [2] Abdelli N, Solà-Oriol D, Pérez JF. Phytogenic feed additives in poultry: Achievements, prospective and challenges. *Animals*, 2021; 11(12): 3471.
- [3] Rahim NFA, Muhammad N, Putra TNMT, Rashidi WNASWM, Talip BA, Abdullah N and Bakar MFA. Interaction Effect of Polyherbal Formulations with Antioxidant Activity. *Eurasian Journal of Analytical Chemistry*, 2018; 13(6): 613-618
- [4] Zhu X, Tao L, Liu H and Yang G. Effects of fermented feed on growth performance, immune organ indices, serum biochemical parameters, ceecal odorous compound production, and the microbiota community in broilers. *Poultry Science*, 2023; 102: 102629
- [5] Demeckova V, Kelly D, Coutts AGP, Brooks PH and Campbell A. The effects of fermented liquid on the faecal microbiology and colostrums quality of farrowing sows. *Int. J. Food Microbiol.*, 2002; 79:85-97.
- [6] Candrawati DPMA, Mahardika IG, Bidura IGNG and Siti NW. Effect of prebiotics-probiotics synbiotic supplementation in feed on performance, serum cholesterol, and pathogenic bacteria in broilers intestines. *Advances in Animal and Veterinary Sciences*, 2024; 12(9): 1752-1758; <https://dx.doi.org/10.17582/journal.aavs/2024/12.9.1752.1758>

- [7] Li L, Li WF, Liu SZ and Wang HH. Probiotic fermented feed improved the production, health and nutrient utilization of yellow-feathered broilers reared in high altitude in Tibet. *Br. Poult. Sci.*, 2020; 61:746-753. <https://doi.org/10.1080/00071668.2020.1801988>
- [8] Puspani E, Warmadewi DA and Bidura IGNG. The combined effect of herbal leaves and yeast in feed as a growth promoter and biocontrol of pathogenic bacteria in the intestines of native chickens. *World Journal of Biology Pharmacy and Health Sciences*, 2023;15(02), 085-090
- [9] Hui Y, Tamez-Hidalgo P, Cieplak T, Satessa GD, Kot W, Kjaerulff S, Neilsen MO, Nielsen DS and Krych L. Supplementation of a lacto-fermented rapeseed-seaweed blend promotes gut microbial- and gut immune-modulation in weaner piglets. *Journal Anim. Sci. Biotechnol.*, 2021; 12: 85.
- [10] Lv J, Guo L, Chen B, Hao K, Ma H, Liu Y and Min Y. Effects of different probiotic fermented feeds on production performance and intestinal health of laying hens. *Poult. Sci.*, 2022; 101: 101570. <https://doi.org/10.1016/j.psj.2021.101570>
- [11] Bidura, IGNG, Siti NW and Partama IBG. 2019. Effect of probiotics, *Saccharomyces* spp. Kb-5 and Kb-8, in diets on growth performance and cholesterol levels in ducks. *South African Journal of Animal Science*, 2019; 49(2): 219-226 <http://dx.doi.org/10.4314/sajas.v49i2.2>
- [12] Bukar A, Uba TI and Oyeyi. Antimicrobial Profile of *Moringa oleifera* Lam. extracts against some food-borne microorganism. *Bayero Journal of Pure and Applied Sciences*, 2010; 3(1): 43-48.
- [13] Restiayanti L, Bidura IGNG, and Sumardani NLG. The effect of giving *Moringa* leaf extract (*Moringa oleifera* Lam) and garlic leaves (*Allium sativum*) through drinking water on the distribution of body fat and cholesterol levels in broilers aged 2-6 weeks. *E-jurnal Peternakan Tropika*, 2014; 2(3): 402.
- [14] Dalukdeniya DACK, De Silva KLASR and Rathnayaka RMUSK. Antimicrobial activity of different extracts of leaves bark and roots of *Moringa oleifera* (Lam). *Int. J. Curr. Microbiol. App. Sci.*, 2016; 5(7): 687-691.
- [15] Oso AO, Suganthi RU, Manjunatha Reddy GB, Malik PK, Thirumalaisamy G, Awachat VB, Selvaraju S, Arangasamy A and Bhatta R. Effect of dietary supplementation with phyto-genic blend on growth performance, apparent ileal digestibility of nutrients, intestinal morphology, and cecal microflora of broiler chickens. *Poult. Sci.*, 2019; 98: 4755-4766, <https://doi.org/10.3382/ps/pez191>
- [16] Pliego AB, Tavakoli M, Khusro A, Seidavi A, Elghandour MMY, Salem AZM. Beneficial and adverse effects of medicinal plants as feed supplements in poultry nutrition: a review. *Anim. Biotechnol.*, 2020; 33: 369-391, <https://doi.org/10.1080/10495398.2020.1798973>
- [17] Krauze M. Phytobiotics, a natural growth promoter for poultry. In: L. Babinszky, J. Oliveira EM, Santos (Editors). *Promoter for Poultry, Advanced Studies in the 21st Century Animal Nutrition*. Intech Open Ltd. London (UK), 2021; pp.1-22, <https://doi.org/10.5772/intechopen.99030>
- [18] Khasnavis S, and Pahan K. Sodium benzoate, a metabolite of cinnamon and a food additive, upregulates neuroprotective Parkinson disease protein DJ-1 in astrocytes and neurons. *J. Neuroimmune Pharmacol.*, 2012; 7: 424-435.
- [19] Gomes GS, Bidura IGNG, Budaarsa IK, and Partama IBG. Supplementation of *Moringa oleifera* leaf flour in diet to Increase growth performance and reduce cholesterol content in meat of East Timor local pigs. *J. Biol. Chem. Research.*, 2019;36(2): 61-68
- [20] Ayssiwede SB, Dieng A, Bello H, Chrysostome CAAM, Hane MB, Mankor A, Dahouda M, Houinato MR, Hornick JL and Missohou A. Effects of *Moringa oleifera* (Lam.) leaves meal incorporation in diets on growth performances, carcass characteristics and economics results of growing indigenous Senegal chickens. *Pakistan Journal of Nutrition*, 2011; 10(12): 1132-1145
- [21] Chukwuebuka E. *Moringa oleifera* "The Mother's Best Friend". *International Journal of Nutrition and Food Sciences* 2015; 4(6): 624-630
- [22] Park JH and Kim IH. Effects of dietary *Achyranthes japonica* extract supplementation on the growth performance, total tract digestibility, cecal microflora, excreta noxious gas emission, and meat quality of broiler chickens. *Poult. Sci.*, 2020; 99: 463-470, <https://doi.org/10.3382/ps/pez533>
- [23] Sun HY, Kim YM, and Kim IH. Evaluation of *Achyranthes japonica* Nakai extract on growth performance, nutrient utilization, cecal microbiota, excreta noxious gas emission, and meat quality in broilers fed corn-wheat-soybean meal diet. *Poult. Sci.*, 2020; 99: 5728-5735, <https://doi.org/10.1016/j.psj.2020.07.023>

- [24] Oboh G and Elusiyan CA. Changes in the nutrient and antinutrient content of microfungi fermented cassava flour produced from low and medium cyanide variety of cassava tuber. *African J. Biotech.*, 2007; 6(18): 2150-2157; <https://doi.org/10.5897/AJB2007.000-2336>
- [25] Suprihatin. *Fermentation Technology*. UNESA Press. Surabaya, Indonesia 2010.
- [26] Hammershoj M, Kidmose U, and Steinfeldt S. Deposition of carotenoids in egg yolk by short-term supplement of coloured carrot (*Daucus carota*) varieties as forage material for egg-laying hens. *Journal of the Science of Food and Agriculture*. 2010; 90: 1163-1171.
- [27] Bidura IGNG, Mahardika IG, Suyadnya IP, Partama IBG, Oka IGL, Candrawati DPMA and Aryani IGAI. The implementation of *Saccharomyces spp.n-2* isolate culture (isolation from traditional yeast culture) for improving feed quality and performance of male Bali duckling. *Agricultural Sci. Research J.*, 2012;2(9): 486-492
- [28] Siti NW and Bidura IGNG. Effects of carrot leaves on digestibility of feed, and cholesterol and β -carotene content of egg yolks. *South African Journal of Animal Science*, 2022; 51(6): 786-792
- [29] Siti NW, Partama IBG, Wibawa AAPP and Bidura IGNG. Combination effect of water extract herbal leaves on drinking water to stimulate the growth and bio-control of pathogenic bacteria in broiler. 2021. *Plant Archives* 21 (1):1644-1649
- [30] Prabowo, Mahfudz LD and Atmomarsono U. Calcium and protein meat mass due to the use of waste product of Carrot powder in ration. *Jurnal Sain Peternakan Indonesia*, 2019; 14(2): 201-207. DOI: <https://doi.org/10.31186/jspi.id.14.2.201-207>
- [31] Zurmiati ME, Mahata MH, Abbas and Wizna. The application of probiotic on duck. *JurnalPeternakan Indonesia*. 2014; 16(2): 134-144
- [32] Royadi R, Nur H and Malik B. The effect of giving boiled betel leaves (*Pipper betlelinn*) in drinking water on slaughter weight and carcass percentage of local rabbits. *Jurnal Peternakan Nusantara*, 2016; 2(2): 73-78. <https://doi.org/10.30997/jpnu.v2i2.742>
- [33] Wahyono T, Sadarman, Handayani T, Trinugraha AC and Priyoatmojo. Carcass performance evaluation of Local and New Zealand white bucks from different slaughter weight. *Jurnal Peternakan*, 2021; 18(1): 51-60.
- [34] Karukarach T, Rakangthong C, Bunchasak C and T Poeikhampha. Comparative effects of blended herbal extracts and mixed prebiotics on growth performance, carcass yields and intestinal morphology of broiler chickens. *International Journal of Poultry Science*, 2016; 15: 264-270.
- [35] Mangka W, Sebola NA, Mokoboki HK, Manyeula F and Mabelebele M. Growth performance and blood profiles of weaned New Zealand rabbits (*Oryctolagus cuniculus*) supplemented with *Moringa oleifera* leaf meal. *Journal of Animal and Feed Sciences*, 2022; 31 (2): 152–160 <https://doi.org/10.22358/jafs/148079/2022>
- [36] El-Desoky AMI, Alazab AM, Bakr ELO, Elseady YA. Effect of adding *Moringa* leave meal to rabbit diets on some productive and reproductive performance traits. *Egypt. J. Rabbit Sci.*, 2018; 28: 263-286, <https://doi.org/10.21608/ejrs.2018.44115>
- [37] Alaqil AA, Abbas AO, El-Beltagi HS, El-Atty HKA, Mehaisen GM, Moustafa ES. Dietary supplementation of probiotic *Lactobacillus acidophilus* modulates cholesterol levels, immune response, and productive performance of laying hens. *Animals*, 2020; 10: 1588, <https://doi.org/10.3390/ani10091588>
- [38] Lokapirnasari WP, Al-Arif MA, Maslachah L, Kirana ALP, Suryandari A, Yulianto AB and Sherasiya A.2023. The potency of *Lactobacillus acidophillus* and *L. lactis* probiotics and *Guazumaulmifolia* Lam. extract as feed additives with different application times to improve nutrient intake and feed efficiency in *Coturnix coturnix japonica* females. *Journal of Animal and Feed Sciences*, 2023; 32(1): 59-67