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The effect of adding synbiotics between *Moringa* and Yeast (MYS) in feed on growth and feed efficiency in starter phase broilers

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Abstract

In recent years, the use of antibiotics in feed has been prohibited, due to residues in poultry products and the effect of resistance on certain bacteria. An alternative to antibiotics is a combination of probiotics and prebiotics, known as synbiotics. This research aims to examine the effect of synbiotics between *Moringa* and Yeast (MYS) in feed on growth and feed efficiency in starter phase broilers. A total of 160 day-old-chick (DOD) broiler chickens of the MB 202 strain were randomly divided into 4 treatment groups, namely: broiler group fed with 0.2% MYS (S1); 0.4% MYS (S2); and 0.6% MYS (S3) supplementation, respectively. Control group chickens were fed without MYS (S0). Each treatment had 5 repetitions using 40 birds. The results showed that the response of broilers to MYS administration from 1-14 days of age did not show any significant difference (P>0.05) compared to controls (without MYS). The positive response of broilers to MYS administration was seen to be significant (P<0.05) starting at 15-21 days of age, namely a significant decrease in feed consumption (P<0.05) in the S2 and S3 broiler groups. The highest feed efficiency was found in the S2 broiler group. It was concluded that the addition of synbiotics between *Moringa* and Yeast (MYS) in feed at the level of 0.2% to broiler feed only saw an impact starting at 15-21 days of age, seen from a decrease in feed consumption and an increase in feed efficiency.

Keywords: Broiler; Feed efficiency; Prebiotics; Probiotics; Synbiotics

1. Introduction

Nowadays, broilers are one of the top priority birds in providing meat and also as a source of animal protein for the community. As a meat producer and provider of animal protein for the community, broilers have a fairly short rearing time. Broilers at the beginning of their life (starter phase) require intensive and careful care, are very sensitive to changes in environmental temperature, nutrition, and are easily infected with disease. One disease that easily infects broilers is Colibacillosis which is caused by the pathogenic bacteria Escherichia coli.

In recent years, the use of antibiotics in feed has been prohibited, due to residues in poultry products and the effect of resistance on certain bacteria [1,2]. An alternative to antibiotics is a combination of probiotics and prebiotics, known as synbiotics. *Moringa* leaves (*Moringa oleifera*) can act as a prebiotic because they contain high crude fiber [3]. Besides that, *Moringa* leaves contain phytochemical compounds which have antimicrobial activity against pathogens and are able to stimulate nutrient absorption [4,5] and contain high levels of β -carotene which functions as a carcass colorant [6]. The benefit of prebiotics is to improve the health of the digestive tract of poultry through their ability to change the composition or activity of the microbiota in the digestive tract.

Probiotics are live microorganisms that are consumed alive in sufficient quantities and are able to reproduce in the digestive tract of poultry and bring health benefits, maintaining the balance of the digestive tract microflora, so that nutrition can be absorbed properly[7]. Yeast culture can act as a probiotic in poultry [8,9]. Giving probiotics to poultry

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should be at the beginning of the growth phase, so that probiotic microbes in the digestive tract can grow dominantly. Several researchers report that probiotic supplementation in feed can improve performance and make feed use more efficient [10,11].

Synbiotics between *Moringa* and yeast in the chicken's digestive tract can cause the pH value in the jejunum to be optimal, which ranges between 5.80-6.90 [2], so that absorption of food substances can be maximized [12] which can cause feed efficiency in broilers to increase. This increase appears to be caused by the antibacterial activity of probiotics and phytochemical compounds in *Moringa* leaves which can reduce the population of pathogenic bacteria in the intestine [13,14,15], so that nutrient absorption can be maximized. The combination of prebiotics sourced from herbal leaves and probiotics in feed significantly increases nutrient absorption, feed efficiency and health status of quail [16].

The aim of this research was to determine the effect of synbiotic supplementation between *Moringa* flour (*Moringa oleifera*) and Yeast in starter phase broilers on growth and feed efficiency.

2. Material and methods

2.1. Material

This *in vitro* research was carried out in the Microbiology Laboratory, Faculty of Animal Husbandry, while the feeding trial research was carried out in the poultry house, Research Farm, Faculty of Animal Husbandry, Udayana University in Jl. Raya Sesetan, Denpasar, Bali, Indonesia. The equipment used consisted of analytical scales and a heating stove, petri dishes, potato dextrose agar (PDA) media, colony counter. The chicken used is a broiler day-old-chick (DOD) strain MB 202 produced by PT. Japfa Comfeed Indonesia Tbk., Surabaya, Indonesia. The feed provided was commercial complete feed 511 HI-PRO-VITE in crumble form produced by PT. Charoen Pokphand Indonesia, Tbk., in Jl. Raya Surabaya-Mojokerto Km. 26, East Java, Indonesia.

2.2. Methods

A total of 160 day-old-chick (DOD) broiler chickens of the MB 202 strain were produced by PT. Japfa Comfeed Indonesia Tbk., Surabaya, Indonesia. All chickens were divided randomly into 4 treatment groups, namely: broiler group fed with 0.2% MYS supplementation (S1); 0.4% MYS (S2); and 0.6% MYS (S3), respectively. Control group chickens were fed without MYS (S0). Each treatment had 5 repetitions using 40 birds. All chickens were kept in battery colony cages made of wire and bamboo slats. The dimensions of each cage plot are: length 200 cm, width 150 cm and height 40 cm. Each plot contains 8 DOC broilers with homogeneous body weight. All chickens were given food and water *ad libitum*. The feed container is made of PVC pipe and the drinking water container is made of plastic with a volume of 5 liters.

2.3. Synbiotic between Moringa and Yeast

Synbiotics are a combination of probiotics and prebiotics. Yeast (*Saccharomyces spp.*) functions as a probiotic, while *Moringa* leaves function as a prebiotic. Synbiotics are expected to occur in the chicken's digestive tract. *Moringa* leaves were obtained from local *Moringa* plantations in the Tabanan area, Bali Province, Indonesia. The *Moringa* leaves used are old *Moringa* leaves, then dried in the sun for two days, then crushed into flour and filtered using a flour sieve. The yeast used is yeast for making tape tape (*Saccharomyces spp.*) obtained from the local public market. Determining the ratio between *Moringa* leaf flour and yeast to obtain optimal sybiotics based on the *Saccharomyces spp.* population was carried out *in vitro* in the Microbiology laboratory. *In vitro* research shows that the synbiotic between *Moringa* and Yeast in a ratio of 1:1 (g/g) gives the highest results in the number of colonies of *Saccharomyces spp.*, namely: 1.70x10⁹ cfu/ml. According to [17] that the minimum probiotic dose that can be beneficial is 10⁵cfu/ml, while 10⁶ cfu/ml is the general dose used to be set as an industry standard.

2.4. Observed Variables

The variables observed were initial body weight, final body weight (FBW), live weight gain (LWG) and feed consumption (FC) which were measured every week. Feed conversion ratio (FCR) was calculated every week, namely the comparison between FC and LWG. The data obtained were analyzed using one-way analysis of variance (ANOVA), if there was a significant difference (P<0.05) between treatments, it was continued with Duncan's multiple range test.

3. Results and discussion

The effect of providing synbiotics between *Moringa* and yeast on broiler performance from 1-21 days of age is presented in Table 1. The addition of 0.2-0.6% MYS to the feed apparently did not have a significant effect (P>0.05) on growth and feed efficiency in broilers from 1-7 days and 8-14 days of age.

Table 1 The effect of adding synbiotics between *Moringa* and Yeast (MYS) in feed on growth and feed efficiency in starter phase broilers

Variable MYS addition level in feed (%)					SEM
	0	0.2	0.4	0.6	
Age 1-7 days					
Initial body weight, g	47.00	47.40	46.70	46.90	0.26
FC, g/7 days	197.3	191.75	189.95	189.00	2.65
FBW, g	221.10	220.85	217.50	211.65	5.50
LWG, g/7days	174.10	173.45	170.80	164.75	5.53
FCR	1.14	1.11	1.11	1.15	0.09
Age 8-14 days					
Initial body weight, g	221.10	220.85	217.50	211.65	5.50
FC, g/7 days	435.95	458.80	444.30	455.35	15.11
FBW, g	541.90	562.90	534.30	534.45	17.44
WG, g/7days	320.80	342.05	316.80	32.80	13.24
FCR	1.36	1.34	1.40	1.42	0.03
Age 15-21 days					
Initial body weight, g	541.90	538.10	534.30	534.45	21.39
FC, g/7 days	1847.50a	1807.50a	1619.00b	1701.50b	31.27
FBW, g	1005.55	1013.30	981.90	996.05	24.65
WG, g/7days	463.65	475.20	447.60	461.60	18.51
FCR	1.57ª	1.46 ^b	1.55ª	1.53ª	0.031

Note: The mean with superscript (^{a,b}) was significantly different ($P \le 0.05$); FC = feed consumption; FBW= final body weight; LWG = live weight gain; FCR = feed conversion ratio (FC:WG)

The positive impact of giving MYS began to be seen in broilers aged 15-21 days. The addition of 0.4-0.6% MYS to the feed significantly (P<0.05) reduced feed consumption. However, it did not have a significant effect (P>0.05) on final body weight and live weight gains. The chicken group given 0.2% MYS had the lowest FCR value compared to other chicken groups (groups S0, S3, and S4). The addition of 0.2% MYS to the feed (broiler group S1) had an FCR value: 7.01% significantly (P<0.05) lower than the control (without MYS administration). The FCR value in broiler groups S2 and S3 was not significantly different (P>0.05) compared to group S0, however the FCR value was significantly (P<0.05) higher than group S1. More details are presented in Table 1.

Based on the results of observations, it turns out that giving MYS at the beginning of the chicken's age (1-14) has not shown a significant response. Feed consumption, body weight gain and feed efficiency were still the same for all treatments. The same results were caused by giving MYS at a low percentage compared to feed consumption, so that the digestive tract microbes were still adjusting. As reported by [18], giving prebiotic herbal leaf flour and probiotics in the early phase of chicken growth still showed the same response compared to controls.

The impact of giving MYS can only be seen in broilers aged 15-21 days, or after two weeks of giving MYS. Increasing the level of MYS in the feed (broiler groups S2 and S3), significantly reduced feed consumption and had no significant effect

on FBW and LWG. The decrease in feed consumption due to MYS administration is in accordance with what was reported by [19] who found that feed consumption decreased significantly in chickens that were given herbal supplements compared to chickens that were not given herbal supplements. Herbal leaf flour can function as a prebiotic because it contains oligosaccharides which cannot be digested by the host animal, but has a beneficial effect on the host by stimulating the growth of digestive tract microflora [20].

Giving MYS at a level of 0.2% provides the highest feed efficiency compared to other treatments. This is closely related to *Moringa's* role as a prebiotic in the chicken's digestive tract which can stimulate optimal growth of intestinal villi. According to [21], prebiotics are a place for pathogenic bacteria to attach, so they do not attach directly to and infect the surface of the intestinal villi. Higher intestinal villi indicate a more mature epithelium and increased absorptive function. A similar thing was reported by [22] that supplementation of prebiotics, probiotics and synbiotics in broiler feed can improve the histology of the chicken intestine, so that nutrient absorption can increase.*Moringa* leaves (*Moringa oleifera*) are a potential source of prebiotics given to broilers. As a prebiotic, *Moringa* leaves are classified as a fiber that is difficult to dissolve in the digestive tract of chickens [23] which is feed for probiotic yeast tape. Tirajoh et al. [24] stated that adding 5% *Moringa* leaf flour to native chicken feed can improve growth, feed consumption and feed efficiency. On the other hand, Yunus [23] stated that the addition of *Moringa* leaf flour up to 4% in broiler feed had no effect on growth, but reduced feed efficiency.

Ragi tape is one of the probiotic alternatives widely available on the market. Yeast tape was chosen because it contains the microbe *Saccharomyces cerevisiae* (dominant yeast) which can act as a probiotic. Tape yeast is usually consumed by humans in making fermented foods, so it is safe for livestock. Several studies have confirmed the effect of yeast tape 3 g/kg in the diet increasing beneficial microbes and suppressing pathogenic bacteria [22]. Bidura's research results [10] state that supplementation of 0.10-0.30% probiotic *Saccharomyces spp*.N-2 (tape yeast isolate) in the ration of laying hens can increase the digestibility of dry matter, organic matter and protein. On the other hand, it reduces the number of *Choliform* and *E.coli* bacteria, as well as reducing the ammonia gas content in chicken excreta.

The existence of synbiotics between probiotics and prebiotics which have a synergistic effect, will provide more optimal health effects [25]. Ngatirah [26] stated that synbiotics can have a beneficial effect on the host, because the fermentation of fructo oligosaccharides (FOS) by probiotic bacteria will produce acidic conditions which will reduce intestinal pH. This can get rid of pathogenic bacteria, improve the survival and implantation or attachment of microbes in the intestinal tract, and stimulate the growth activity of beneficial microbes. Raheem [22] stated that synbiotics have the potential to improve growth performance, histomorphology and broiler health.

Hartono et al. [27] stated that the addition of 2% natural synbiotics could increase the number of lactic acid bacteria in the duodenum, jejunum and ileum, and reduce *Escherichia coli* bacteria in the ileum, while the use of 4% commercial synbiotics could increase the height of the duodenal intestinal villi, jejunum and ileum in male chickens. Raksasiri et al. [28] stated that synbiotics can improve FCR values and reduce ammonia concentrations in the digestive tract. Probiotic and prebiotic synbiotics can lower the pH in the intestine, making them effective in reducing cholesterol concentrations and improving broiler health [29]. The research results of [30] reported that the addition of probiotic *Bacillus subtilis* or Yeast culture to the rations of weaned piglets showed the same health effects compared to the addition of antibiotics (zeng bacitracin).

Increased feed efficiency in group S1 broilers, because MYS contains the probiotic *Saccharomyces spp*. According to [8], *Saccharomyces spp*. apart from being a crude fiber degrader, it can also increase feed digestibility. Probiotics in the digestive tract of chickens can increase nutrient digestibility, so that growth and feed efficiency can be optimal [31]. It was reported by [32] that probiotics can effectively improve broiler growth performance through modulating the beneficial microbiota in the caecum. Prebiotics Herbal leaves have a promising ability to maintain growth performance, by increasing nitrogen digestibility and modulating intestinal bacterial populations in broiler chickens [33]. Similar things were also reported by [34,35] that supplementation of herbal leaves in feed resulted in increased nutrient digestibility in broilers.

4. Conclusion

Based on the research results, it can be concluded that the addition of synbiotics between *Moringa* and Yeast (MYS) in feed at a level of 0.2% to the feed only shows its impact when the broilers are 15-21 days old, seen from a decrease in feed consumption and an increase in feed efficiency.

Compliance with ethical standards

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Disclosure of conflict of interest

No conflict of interest to be disclosed.

Statement of ethical approval

The Research Ethics Commission of the Faculty of Veterinary Medicine, Udayana University, Indonesia has approved this research.

References

- [1] Babaei S, Rahimi S, Karimi Torshizi MA, Tahmasebi G, Khaleghi Miran SN. Effects of propolis, royal jelly, honey and bee pollen on growth performance and immune system of Japanese quails. Vet. Res. Forum., 2016; 7: 13-20
- [2] Purbarani SA, Wahyuni HI and Suthama N. Dahlia inulin and *Lactobacillus sp.* in step down protein diet on villi development and growth of kub chickens. Tropical Animal Science Journal, 2019; 42(1): 19-24. https://doi.org/10.5398/tasj.2019.42.1.19
- [3] Gomes GS, Bidura IGNG, Budaarsa IK, and Partama IBG. 2019. 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
- [4] Dalukdeniya DACK, DeSilva KLSR, 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. doi: http://dx.doi.org/10.20546/ijcmas.2016.507.078
- [5] Pliego AB, Tavakoli M, Khusro A, Seidavi A, Elghandour MMY, and 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
- [6] Chukwuebuka E. *Moringa* oleifera "The Mother's Best Friend". International Journal of Nutrition and Food Sciences 2015; 4(6): 624-630
- [7] Priastoto D, Kurtini T, and Sumardi. 2016. The effect of providing probiotics from local microbes on the performance of laying hens. Jurnal Ilmiah Peternakan Terpadu, 2016; 4(1): 80-85
- [8] Bidura IGNG, Suyadnya IP, Mahardika IG, Partama IBG, Oka IGL, 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 Science Research Journal, 2012; 2 (9): 486-492
- [9] Soccol CR, Vandenberghe LP, Spier M., Medeiros AB, Yamaguishi CT, Lindner JD, Pandey A, Thomaz-Soccol V. The potential of probiotics: A review. Food Technology and Biotechnology, 2010; 48: 413-434.
- [10] Bidura IGNG. The effect of probiotic *Saccharomyces spp*. in the diet on feed digestibility and ammonia gas content in chicken excreta. Majalah Ilmiah Peternakan, 2020; 23(2): 84-90.
- [11] Hasan SAJ, Lokman IH, Naji SA, Zuki ABZ, and Kassim AB. Effects of dietary supplementation of wet fermented feed with probiotic on the production performance of Akar Putra chicken. Asian Journal of Poultry Science, 2016; 10(2): 72-77.
- [12] Hidayat R, Yunianto VD, Sukamto B and Sugiharto S. Effect of dietary supplementation of probiotic, phytobiotics or their combination on performance, blood indices and jejunal morphology of laying hens during post peak production. Online J. Anim. Feed Res., 2012; 11(1): 08-12. DOI: https://dx.doi.org/10.51227/ojafr.2021.2
- [13] Hedayati M and Manafi M. Evaluation of an herbal compound, a commercial probiotic, and an antibiotic growth promoter on the performance, intestinal bacterial population, antibody titers, and morphology of the jejunum

and ileum of broilers. Brazilian Journal of Poultry Science, 2018; 20(2): 305-316. https://doi.org/10.1590/1806-9061-2017-0639

- [14] Hussein E, Ahmed SH, Abudabos AM, Aljumaah MR, Alkhlulaifi MM, Nassan MA, Suliman GM, Naiel M, and Swelum AA. Effect of antibiotic, phytobiotic and probiotic supplementation on growth, blood indices and intestine health in broiler chicks challenged with *Clostridium perfringens*. Animals, 2020; 10(3): 507. https://doi.org/10.3390/ani10030507
- [15] 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, https://doi.org/10.1007/s11481-011-9286-3
- [16] Lokapirnasari WP, Al Arif MA, Maslachah L, Kirana ALP, Suryandari A, Yulianto AB, and A. Sherasiya. The potency of *Lactobacillus acidophillus* and *L. lactis* probiotics and *Guazuma ulmifolia* 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 https://doi.org/10.22358/jafs/156018/2022
- [17] Plessas S, Bosnea L, and alexopoulos A. Potential effects of probiotics in cheese and yogurt production: A review. Eng. life Sci., 2012; 12(4): 1-9.
- [18] Bidura IGNG. Herbal Leaves and Probiotics. Alternative to Antibiotic Growth Promoters (AGPs) in Livestock. First Ed. Penerbit Swasta Nulus, Jl. Tukad Batanghari, Denpasar, Indonesia, 2020.
- [19] Hammershoj M, Kidmose U, Steenfeldt 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.
- [20] Abdurrahman ZH, and Yanti Y. General description of the influence of probiotics and prebiotics on chicken meat quality. Jurnal Ternak Tropika, 2018; 19(2): 95-104 doi: 10.21776/ub.jtapro.2018.019.02.4
- [21] Macfarlane G, Steed H, and Macfarlane S. Bacterial metabolism and health-related effects of galactooligosaccharides and other prebiotic. Journal of Applied Microbiology, 2007; 104(2): 305-344. doi:https://doi.org/10.1111/j.1365-2672.2007.03520.x
- [22] Raheem SMA, Abd-Allah SMS, and Hassanein KMA. The effects of prebiotic, Probiotic and synbiotic supplementation on intestinal microbial ecology and histomorphology of broiler chickens. IJAVMS, 2012; 6(4): 277-289.
- [23] Yunus M. Response of broiler chickens to the provision of *Moringa* leaf flour (*Moringa* oleifera) in feed. Thesis of the Animal Husbandry Science and Technology study program, Faculty of Animal Husbandry, Hasanudin University, Makasar, Indonesia, 2016
- [24] Tirajoh S, Tiro BMW, Palobo F, and Lestari RHS. Utilization of *Moringa* oleifera leaves on the growth quality of superior village chickens from Balitbangtan in Jayapura Papua. Jurnal Ilmu Peternakan dan Veteriner Tropis, 2020; 10(2): 119-127. doi:10.46549/jipvet.v10i2.113
- [25] Kaur I., Chapra K, and Saini A. Probiotics "Potential Pharmaceutical Applications". Eur. J. Pharm. Sci., 2002; 15: 287-293.
- [26] Ngatirah. Probiotics, prebiotics and synbiotics. Scientific Review. Agroteknose, 2009; IV (2): 13-18
- [27] Hartono EF, Iriyanti N, and Suhermiyati S. (2016). The effect of using synbiotics on the condition of the intestinal microflora and histology of male Sentul chickens. Agripet : Vol (16) No. 2 : 97-105.
- [28] Raksasiri BV, Paengkowum S, and Poonsuk K. The effect of supplementation of synbiotic in broiler diets on production performance, intestinal histomorphology and carcass quality. International J. of Agricultural Technology, 2018; 14(7): 1743-1754.
- [29] Ashayerizadeh AN, Dabiri KH, and Mirzadeh. Effect of dietary supplementation of probiotic and prebiotic on growth indices and serum biochemical parameters of broiler chickens. Journal of Cell and Animal Biology, 2021; 5(8): 152-156, August 2011Available online at http://www.academicjournals.org/JCAB
- [30] Cui YM, Wang J, Lu W, Zhang HJ, Wu SG, and Qi GH. Effect of dietary supplementation with *Moringa* oleifera leaf on performance, meat quality, and oxidative stability of meat in broilers. Poult. Sci., 2018; 97: 2836-2844, https://doi.org/10.3382/ps/pey122

- [31] Jannah SL, Lamid M, Sukmanadi M, Arif MAA, Chusniati S, Hamid IS, and Solfaine R. Potential of giving probiotics to increase body weight, consumption, and feed conversion of laying hens in the pre layer phase. Media Kedokteran Hewan, 2022; 33(2): 96-104.
- [32] Li Y, Xu ., Huang Z, Lv L, Liu ., Yin C, Yan H, and Yuan J. Effect of *Bacillus subtilis* CGMCC 1.1086 on the growth performance and intestinal microbiota of broilers. J. Appl. Microbiol., 2016; 120: 195-204, https://doi.org/10.1111/jam.12972
- [33] Hoque MR, and Kim IH. Effect of *Achyranthes japonika* Nakai extract on growth performance, apparent nutrient digestibility, excreta microbial count and gas emission in broilers fed different protein diets. Journal of Animal and Feed Sciences, 2023; 32(1): 34-42 https://doi.org/10.22358/jafs/155126/2022
- [34] 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. Poul. Sci., 2020; 99: 463-470, https://doi.org/10.3382/ps/pez533
- [35] 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. Poul. Sci., 2020; 99: 5728-5735, https://doi.org/10.1016/j.psj.2020.07.023