

Effect of Earthworm (*Eisenia fetida*) Supplementation on Production Performance of Layer Chickens

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Abstract: The present study aimed to investigate the effect of earthworm supplementation on growth and egg production performance of layer chickens. A total of 96 layers were distributed randomly into four treatment groups with three replications using a completely randomized design. Feeding trial was started when layer were reach at 24 weeks ages. The experimental diets that containing 0 (Control), 0.66, 1.32 and 1.98 % of earthworm (EW) were fed to birds for the duration of twelve weeks. Eggs were collected daily and weighed in every week and then the egg production was calculated as %. Though, body weight measurement was taken at the commencement and at the end of the experimental period; whereas egg quality was evaluated at the middle and then in the last week of experimental period. Feed intake had not statistical difference among treatment groups. Chickens fed with both diets containing 1.32 and 1.98% of EW showed an increment in bodyweight change and egg production. The hen day egg production (HDEP) and hen housed egg production (HHEP) were significantly ($P < 0.05$) higher for hens in T_4 than others, but there was no significant ($P > 0.05$) difference between T_2 and T_3 . There was no measurable effect on egg quality due to feeding diets, containing earthworm. Supplementation of 1.98% earthworm in layer diets trigger egg production performances without affecting the quality eggs that starting from 24 week. It is concluded that earthworm supplementation has the potential to improve the egg production performance of laying hens.

Keywords: Earthworm, Egg Production, Layer Chicken, Supplementation

1. Introduction

The poultry industry is one of the most vibrant sectors in the livestock farming. Feed cost is extremely important in the production of poultry, specifically in intensive agriculture. A important world concern remains the demand for high-quality protein sources from an enhancing world population and the parallel loss of land suitable for agricultural crop cultivation [1]. The production of meat and egg is challenged by both quality and quantity of feeds. To enhance the quality and quantity of poultry meat and eggs, poultry nutritionist uses several sources of high-protein feed components, such as soybean meal, fishmeal, or meat meal, to increase the value of the diets [2]. Matching available feed resources to animal requirements could increase productivity, reduce environmental impacts, and lower feed costs.

The thought of utilizing insects in animal nutrition is a recent phenomenon that inspiring and promising novelty among the alternative feed resource and receiving much attention lately [3]. In recent years, there has been a shift in the source of proteins used in animal nutrition. These should not be of vertebrate animal origin and have a better composition than soya products, one such alternative is earthworm [2, 4, 5]. Earthworm are known by different names of such as nightcrawlers, worms, lobworms, granddaddy earthworms, and dew worms. About 4000 species earthworms are existed on this planet. Rare species of earthworms are used for vermicomposting as they have certain features like resistance to a varied of environmental conditions, good composting rate and high reproductive rate, short life cycles. Earthworm species used for vermicomposting are, *Eisenia fetida*, *Perionyx excavates*, *Eudrilus eugeniae*, *Dendrobaena* and *Eisenia Andrei Veneta*

[6]. Worms can be used as a bio-converting tool of organic wastes into animal protein sources in environmentally friendly manner of climate smart agriculture. Recently, the major benefit of earthworms is to generate vermin compost used as a source of protein for ruminant animals, fish, and poultry farming [7]. Earthworms are consumable and roasted cooked, fried, sautéed, and put sides with meat dishes over the global. Earthworm is very crucial for vermicomposting (organic fertilizer) which increase soil fertility. Farmers utilize this earthworm dually for composting and as chicken feed. The nutritional profile of earthworm meal is comparable to that of other protein sources that currently used in poultry feeds, especially that of fish meal [8-10]. It has been proven to be even better, in terms of a higher percentage of essential amino acids as well as adequate amounts of fatty acids than fishmeal [5], and is considered a significant source of lysine to meet the dietary requirements for this amino acid in poultry ration [11]. On a dry matter basis, earthworms contain 60-70% protein, 6-11% fat, 5-21% carbohydrates, and 2-3% minerals and a range of vitamins, including niacin [6].

Use of worms as chicken feed is not in direct competition with the human population as a food resource for people. Alternative way of reducing a major gap exists between the demand and supply of conventional feed resources for feeding chicken is exploiting the use of non-conventional feed resources in poultry farming. Recently animal protein sources have become increasingly scarce in the Ethiopia countries and new alternative protein sources need to be developed. As a result, animal farmers are looking for less expensive ways to provide nutritional dietary supplements that can partially or entirely replace poultry feed ingredients. Previous research has demonstrated that earthworms are rich in protein and various amino acids, and can be used as replacement of fish meal in chickens, fish and pig feeds [12]. So far, many researches have tested earthworms in broiler diets [13] with positive effect on broiler chicken performance. Indigenous chickens with access to outdoor area pick up insects at all life stages and eat them with great pleasure, which indicates that they are evolutionary adapted to insects as natural part of their ration menu. Feeding chicken diets encouraged with earthworms has been shown to positively effect on the growth, immunity and meat quality. Though, there is little information regarding the effects of diets, containing earthworms on egg production and egg quality characteristics in Ethiopia. Therefore, the objective of the current study was to explore the effect of different levels of earthworm (*Eisenia fetida*) supplementation on the production performance of laying hens.

2. Materials and Methods

2.1. Experimental Design and Treatment Arrangements

The experiment was undertaken in Debreziet Agricultural Research Center. A completely randomized design (CRD) was used with four treatments, which were replicated three

times and Bovan Browns laying hens were randomly assigned to treatment groups. The treatments were: T₁ (100% commercial diets as a control), T₂ (99.34% commercial diets + (0.66% of EW), T₃ (98.68% commercial diets + (1.32 % of EW) and T₄ (98.02 % commercial diets + (1.98 % of EW). Each experimental diet was prepared to fulfill the chickens' nutrient requirements (NRC, 1994). Feed ingredients used in the formulation of the experimental rations for the study were wheat middling, nougseed cake, maize, soybean meal, layer concentrated premix 2.5%, salt, limestone, lysine, methionine and fresh earthworms. Duration of experimentation was elapsed for twelve weeks.

2.2. Production and Preparation of Earthworm as Chicken Feed

An initial earthworm breeding stock was supplied by the natural resource research process of DZARC. The Cattle dung and vegetable wastes were used to cultivate the earthworms (*Eisenia fetida*). The process of culturing was done in a wooden box with 1 x 1.5 x 1 m³ size and additionally in five plastic containers with 0.5 m³. *Teff* straw (*Eragrostis teff*) was used as the substrate for the earthworm bedding materials, and water was added to moistening of the bedding materials while it was being cultured. Initially, matured earthworms (*Eisenia fetida*) that could lay eggs were stocked in the culture media setup. After two months, fresh matured earthworms retrieved manually and cleaned using tap water. Practical production of earthworm meal includes the use of simple technology for large volume production. This technology is used to isolates the worms from organic waste materials where they are produced, that's why hampering the use of earthworm meal in developed countries of the globe. Though, there is great potential for cultivating earthworm meal in developed countries because employment expanse are not as much of developed countries. So far, special attention is required when we produce earthworms for the use of animal feeds. The other issue is that contaminants and heavy metals present in the worms can be moved into the birds' digestive tract by consumption the earthworm meal [14]. Thus, it is imperative that what materials are used in the rearing of earthworm.

The proximate composition (dry matter, crude protein, crude ash, calcium, phosphorus and energy) of fresh earthworm reared on cow-dung and vegetables waste substrates was analyzed using the method reported by AOAC at National Veterinary Institute and Holetta Agricultural Research Center, Ethiopia [15]. Proximate composition of the dried earthworm (*Eisenia fetida*) meal cultured with cow dung and vegetable waste substrates are presented in (Table 1).

Table 1. Average proximate composition of earthworms (*Eisenia foetida*) meal produced on teff straw, vegetables and cow dungs.

Parameter	Composition (%)
Dry matter	91.70
Crude protein	64.75
Ash	12..12

Parameter	Composition (%)
Crude fiber	NA
Ca%	1.82
Phosphorus	1.15

2.3. Management of Experimental Birds

The experimental house was cleaned and disinfected very well before the introduction of the experiment birds. Partitioned pens with disinfected teff straw (*Eragrostis teff*), as the bedding materials were used. Before the beginning of the actual experiment at 24 weeks age, the experimental pens, watering and feeding troughs, and laying nests were thoroughly cleaned, disinfected, and sprayed against external parasites. Birds were adapted to treatment diets for a week. All needed vaccinations were given to experimental laying hens properly. Earthworms were offered to the birds along with vermi-hums, which are sources of humic acid that facilitate nutrient absorption and maintain gut health. It was provided using a flat plastic tray during study periods. Water was provided ad libitum throughout the experimental period.

2.4. Feed Intake and Production Performance of Laying Hens

The experimental period elapsed for 12 weeks during which the amount of feed offered to and refused from birds per pen was recorded daily. Feed intake was calculated as the difference between feed offered and feed remaining. Laying hens were weighed at the start and at the end of the experiment. Mean daily weight gain was calculated as a difference of the two consecutive weightings and then divided for the total experimental days. Eggs, from every pen were manually collected and recorded three times a day. The average egg weight was determined after weighing the daily collected eggs per pen. Hen day egg production (%), and hen housed egg production (%) were calculated for each replicate during the experiment according to the following formula [16]: Hen Day egg production percentage was calculated as the total number of eggs produced on daily basis divided by number of hens available in the flock on that day times 100 whereas hen housed egg production percentage was calculated as the total number of eggs produced divided by the number of hens originally housed times 100.

2.5. External and Internal Egg Quality Measurements

Three eggs of each replicate were collected at the beginning, middle and at the end of the experimental period and these eggs were individually opened; yolks were separated from the albumen and then weighed. Shell thickness (with shell membrane) was measured, using a caliper, at three locations on the eggs at broad end, center, and at the point-end. Eggshell weight was taken by sensitive balance without washing and drying. Egg width and length was measured using caliper. Egg yolk color was determined by comparison with the Roche color scale, where the lightest color is 1 and the darkest color is 15. Micrometer was used to measure albumen and yolk height. The egg width (W) and

length (L) were used to measure egg shape index with the formula $ESI = W/L \times 100$ [17]. The Haugh Unit score was calculated for individual eggs by using the following formula: $HU = 100 \log_{10} (H + 7.5 - 1.7W^{0.37})$ Where: H, W = recorded height of the albumen in mm and weight of egg in grams, respectively [18].

2.6. Statistical Data Analysis

SAS version 9.0 was used to analyses the data using the GLM procedures of the Statistical Analysis System [19]. The LSD test (Least Significance Difference) separation approach was used to compare means with significant differences at $P = 0.05$. A statistical model used was $Y_{ij} = \mu + T_j + e_{ij}$.

3. Results and Discussions

3.1. Nutritional Contents of Earthworm Meal

The key limiting issues in feeding chicken are the protein and energy contents of the diets. Mostly, soybean and fish meal are used as protein sources in chicken diets in present conditions. Though, the increases in the prices of fish meal and soybean meal due to several reasons similarly because some increases in the prices of the rations of chickens [20]. During analysis of nutrient compositions, it was observed that worms contain dry matter, crude protein, and minerals that are essential for chickens at large levels (Table 1). As indicated in Table 1, the crude protein content of earthworm used in the current study was 64.75. The crude protein content of earthworm meal recorded from the current study was lower than that reported from earthworm meal (68.88%) by [23]. Contrary to the result of the current study, [13] reported that earthworm meal contains 47.43 and 59.01% of crude proteins, respectively. This difference is might be attributed to factors such as feeding materials, culturing methods, stage of harvesting and drying techniques. The ash content of earthworm (*Eisenia fetida*) evaluated from current study was 12.12% which was greater than results of others [21-23]. who reported 11.39, 5.15 and 9.88%, respectively. It is also greater than ash content of soybean and fish meal [24]. Based on the present laboratory results, earthworm has potential as source of protein feed.

3.2. Feed Intake and Growth Performance

The effect of feeding fresh earthworms at different levels on feed intake and bodyweight of Bovan Browns hens are presented in Table 2. Supplementation of fresh earthworms did not significantly ($P > 0.05$) affect the feed intake. The result of the present study was in opposition with that of Finke et al. (2010), who claimed that supplementing layers diet with various levels of earthworm meal led to a considerable increase in feed intake. It is estimated that supplementation of earthworm did not affect the feed palatability; hence it did not affect the feed consumption. Feed consumption is affected by environmental temperature, also the quality and quantity of ration [25]. The mean final body weight of hens fed on diet T_1 , T_2 , T_3 and T_4 were

1463.75, 1616.67, 1656.67 and 1689.17 grams per bird, respectively. Hens in T4 had significantly higher final bodyweight ($P<0.05$) than hens in T1, T2, and T3. In broiler chickens, [26] reported an increase in body weight and a decrease in feed intake, which is consistent with the current study in layer chickens.

The results indicated that supplementation of fresh earthworm in the diets at increased levels of earthworm produced body weight values that were superior to those produced by basal diet. This might due high crude protein content of earthworms.

Table 2. Effects of fresh earthworm supplementation on the feed intake, final body weight and body weight gain of Bovans Brown layers.

Parameter	Treatments			
	T1	T2	T3	T4
AFI/hen/day	111.06±1.96	114.46±2.56	116.69±0.27	112.84±1.46
Initial BW/g/bird	1377±14.92	1392.83±47.54	1374.29±14.98	1384.50±51.88
Final BW/ g/bird	1463.75±13.76 ^c	1616.67±15.62 ^b	1656.67±18.72 ^{ab}	1689.17±30.68 ^a
BWG/g/bird	1.02±0.11 ^b	2.66±.64 ^{ab}	3.36±.47 ^a	3.62±0.56 ^a

a, b & c Means within a row with different superscripts are significantly different. AFI (Average feed intake), BW (bodyweight), BWG (Bodyweight gain), g = gram, T1= (100 % basal diets, T2= (99.34 % basal diets + 0.66% fresh EW), T3= (98.68% basal diets + 1.32 % fresh EW) and T4= (98.02 % + 1.98 % fresh EW).

3.3. Egg Production Performance

Results of egg production and other parameters are shown in Table 3. Feeding birds with fresh earthworms at 1.98% level in the hen's diet of the present study resulted in higher ($P<0.05$) in total egg production and HDEP than the rest of the treatments. Birds fed on T2 and T3 shown significant difference ($p<0.05$) in most of egg production parameters than birds in T1. According to [27]., supplementing layer chicken feed with earthworm meal increased egg production, supporting the current finding that shows improvements in egg production with the supplementation of fresh earthworms. The hen day egg production (HDEP) and hen housed egg production (HHEP) were higher ($P<0.05$) for hens in T4 than the others, but there was no significant ($P>0.05$) difference between T2 and T3 in their HDEP and HHEP. Results of present study indicated that the average daily egg production significantly ($P<0.05$) increased when increasing levels of

fresh earthworm supplementation. Trends in improved egg production with higher level of fresh earthworm supplementation might be attributed to high crude protein content of the diets. The lowest HDEP and HHEP percentage were recorded for T1. During this experimental period there is no recorded mortality due to experimental diets. Diet that containing earthworm meal is a source of good protein and can be positively used in the ration of laying hens. Various scholars have reported the valuable effect of the dietary inclusion of earthworm meals on the production performance of layers. Supplementation of ration containing earthworm meal varied from 0.2 to 0.6% in layers feed improves layer performance, and better egg quality especially the ratio on-3 fatty acids and egg yolks [28]. which is in line with present study. Unlike the current, the inclusion of 0.6% of earthworm meals in the chicken diet did not affect egg and meat safety [28].

Table 3. Effects of fresh earthworm supplementation on average egg production parameters of Bovans Brown layer chickens.

Parameters	Treatments			
	T1	T2	T3	T4
Egg numbers/ hen/month	18.43± 0.34 ^c	19.44± 0.58 ^b	20.97±0.43 ^b	22.95±0.35 ^a
Egg numbers /hen/total periods	55.29±8.35 ^c	59.83±1.74 ^b	62.92±1.3 ^b	68.87±1.07 ^a
Hen day egg production (%)	65.23±1.65 ^c	71.23±2.08 ^b	74.90±1.55 ^b	81.95±1.28 ^a
Hen housed egg production (%)	65.23±1.65 ^c	71.23±2.08 ^b	74.90±1.55 ^b	81.95±1.28 ^a

a, b&c. Means with in a row with different superscripts are significantly different. gm= gram, T1= 100 % basal diets, T2= 99.34 % basal diets + 0.66% EW, T3= 98.68% basal diets + 1.32 % EW and T4= 98.02 % + 1.98 % EW = earthworm worm, %=Percentage.

3.4. External and Internal Egg Quality Analysis

Egg quality measuring values are shown in Table 4. In this current trial, most of the egg quality measuring parameters didn't show a significant difference among the treatments. Egg weight was higher ($P<0.05$) in T3 than T1, T2 and T4. Egg weight is correlated with body weight of laying hens. Relatively, heavier birds lay heavier egg than light birds [29]. reported that supplementation of different level of earthworm

meal increases egg weight. This showed the slight increment of egg weight in the current trial. Yolk weight was higher ($P<0.05$) in T4 than the rest of the treatments. Hens in T4 had the highest yolk length compared to the rest of treatments. The lack of changes in yolk color due to the tested diets was expected since there was no detectable presence of carotenoids in earthworms. The utilization of fresh earthworms 5% in the ration based on Dry matter, can maintain the production performance of the laying hens. Like the present study, Egg quality is not affected by the use of

earthworm meal [30].

Table 4. The effect of supplementing different level of fresh earthworm on internal and external egg quality characteristics.

Quality Parameters	Treatments			
	T1	T2	T3	T4
Egg weight (g)	52.63±0.08 ^b	53.51±1.27 ^b	57.67±1.74 ^a	53.23±1.15 ^b
Egg length (mm)	53.01±0.62	53.78±0.71	51.28±2.24	53.34±0.22
Egg width (mm)	41.17±0.52	41.50±0.30	41.70±0.70	41.22±0.19
Shape index	77.69±0.26	77.190±0.51	82.12±2.98	77.300±.24
Shell weight (g)	7.31±0.19	7.720±0.40	7.98±0.70	7.82±0.21
Albumen height ((mm)	8.750±0.20	8.160±0.12	8.5±20.37	8.36±0.15
Haugh unit	88.05±0.55	85.91±0.75	86.74±0.97	48.62±0.36
Albumen width (mm)	46.29±0.77	44.78±0.08	44.211±.50	44.640±.83
Yolk weight (g)	12.840±.13 ^{ab}	12.73±0.18 ^b	12.24±0.16 ^b	13.62±0.71 ^a
Yolk length (mm)	37.13±0.98 ^b	36.36±0.28 ^{ab}	37.12±0.68	39.61 ^a ±2±0.13
Yolk width (mm)	34.67±0.32	34.73±1.08	34.740±.34	34.08±0.24
Yolk height (mm)	18.44±0.22	17.50±0.33	17.16±0.30	17.70±0.25
Yolk colour	7.44±0.22	7.72±0.40	7.720±0.11	7.44±0.50
Shell thickness	0.370±.06	0.36±0.01	0.34±0.02	0.330±0.05

^{ab} Means with in a row with different superscripts are significantly different. g= gram, T1= 100 % basal diets, T2= 99.34 % basal diets + 0.66% fresh EW, T3= 98.68% basal diets + 1.32 % fresh EW and T4= 98.02 % + 1.98 % fresh EW, = earthworm, mm (millimeter).

4. Conclusion and Recommendations

Supplementation of layers with highest level of fresh EW (1.98%) of this current study improved bodyweight change, final body weight, egg production and some egg quality parameters. Most of the egg quality parameters were not negatively or positively affected by supplementation of fresh earthworms. It is concluded that supplementation of fresh earthworms has the potential to improve the egg production performance of laying hens. However, in order to draw the optimum level of supplementation, further trial should be conducted involving increasing levels of fresh earthworms under framer management condition, and earthworm meal for intensive production.

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Conflicts of Interest

The authors declare no conflict of interest.

References

- [1] FAO. 2017. The future of food and agriculture: Trends and challenges. Rome.
- [2] Bahadori, Z., Esmaylzadeh, L., & Karimi-Torshizi, M. A. S. 2015. The effect of earthworm (*Eisenia foetida*) and vermi-humus meal in diet on broiler chicken efficiency and carcass components. *Biological Forum*, 7, 998–1005.
- [3] Van Huis, A. 2020. Insects as food and feed, a new emerging agricultural sector: a review. *Journal of Insects as Food and Feed*; 6(1): 27-44.
- [4] Rezaeipour V, Nejad OA and Miri HY, 2014. Growth performance, blood metabolites and jejunum morphology of broiler chickens fed diets containing earthworm (*Eisenia fetida*) meal as source of protein. *International Journal of Advanced Biological and Biomedical Research*, 2(8): 2483-2494.
- [5] Bahadori, Z., Esmailzadeh, L., Karimi-Torshizi, M. A., Seidavi, A., Olivares, J., Rojas, S., Salem, A., Khuro, A., & López, S. (2017). The effect of earthworm (*Eisenia foetida*) meal with vermi-humus on growth performance, hematology, immunity, intestinal microbiota, carcass characteristics, and meat quality of broiler chickens. *Livestock Science*, 202, 74–81.
- [6] Domínguez, J. and C. A. Edwards (2011). Biology and ecology of earthworm species used for vermicomposting. In: Edwards CA, Arancon NQ and Sherman RL (eds) *Vermiculture Technology: Earthworms, Organic Waste and Environmental Management*. Boca Raton, FL: CRC Press, pp. 25–37.
- [7] Deane (2020). Eat the weeds website. Feedipedia Accessed at <http://www.eattheweeds.com/barnyard-grass>.
- [8] Hasanuzzaman, A., Hossian Sk., and Das, M., 2010. Nutritional potentiality of earthworm (*Perionyx excavatus*) for substituting fishmeal used in local feed company in Bangladesh. *Mesopot. J. Mar. Sci.* 25(2): 25-30.
- [9] Sogbesan, O. and Ugwumba, A. 2008. Nutritional values of some non-conventional animal protein feedstuffs used as fishmeal supplement in aquaculture practices in Nigeria. *Turkish Journal of Fisheries and Aquatic Sciences* 8: 159-164.
- [10] Kostecka, J. and Pączka, G., 2006. Possible use of earthworm *Eisenia fetida* (Sav.) biomass for breeding aquarium fish. *European Journal of Soil Biology*, 42, pp. S231-S233.
- [11] Istiqomah, L., Sofyan, A., Damayanti, E., & Julendra, H. 2009. Amino acid profile of earthworm and earthworm meal (*Lumbricus rubellus*) for animal feedstuff. *Journal of the Indonesian Tropical Animal Agriculture*, 34, 253–257.
- [12] Harwood, M. 2006. Recovery of protein from poultry waste by earthworms. In: *Proceedings of the 1st Australian Poul. Stockfeed Conv.*, Melbourne 9: 138-143.

- [13] Loh, T. C., Fong, L. Y., Foo, H. L., Thanh, N. T., & Sheikh-Omar, A. R. 2009. Utilization of earthworm meal in partial replacement of soybean and fish meals in diets of broilers. *Journal of Applied Animal Research*, 36, 29–32.
- [14] Sharma S, Pradhan K, Staya S, Vasudevan P (2005) Potentially of earthworms for waste management and in other uses- a review. *J Am Sci* 1: 4–16.
- [15] AOAC. 1995. Association of Official Analytical Chemists. Official Methods of Analysis Association of Official Analytical Chemists. 15th edition. Ed. Helrich, K. Association Official Analytical Chemists, Inc., Suite, 400, Arlington, Virginia. 3. 685-1298.
- [16] Hunton, P., 1995. Egg production, processing and marketing. *World Poultry Science*, Elsevier, Tokyo, pp. 457-480.
- [17] Anderson, K., Tharrington, P., Curtis, F., and Jones, T., 2004. Shell characteristics of eggs from historic strains of single comb white leghorn chickens and relationship of egg shape to shell strength. *International Journal of Poultry Science* 3, 17-19.
- [18] Haugh, R., 1937. The Haugh unit for measuring egg quality. *US Egg. Poult. Mag.* 43, 572-573.
- [19] SAS. (2002) Statistical analysis systems. SAS Institute Inc., Cary, NC, USA.
- [20] Tacon A G J, Metian M (2008). Global overview on the use of fish meal and fish oil in industrially compounded aqua feeds. *Aquaculture* 285: 146-158.
- [21] Ganguly, A. Anand, and P. Haldar, 2010. Potential value of earthworms as high protein supplement for poultry feed. *International Journal of Poultry Science* 7: 722-725.
- [22] Guerrero, R. D. 2010. The culture and use of *Eisenia fetida* as protein resource in the Philippines. In: J. E. Satchell (Ed.), *Earthworm Ecology*, Chapman and Hall, London 5: 309-319.
- [23] Kasye, M, 2016. Evaluation of Earthworm (*Eisenia fetida*) for Chicken Production in Haramaya University Poultry Farm (Doctoral dissertation, Haramaya University).
- [24] Ntukuyoh, A., Udiong, D. S., Ikpe, E and Akpakpan, E. 2012. Evaluation of Nutritional Value of earthworm (*Eisenia fetida*): Soldiers, Workers, and Queen in the Niger Delta Region of Nigeria. *International Journal of Food Nutrition and Safety* 1: 60- 65.
- [25] Leeson S, and Summers J, 2005 *Commercial Poultry Nutrition*, 3rd edn. Univ. Books, Guelph.
- [26] Sofyan, A., Julendra, A., Julendra, H., Damayanti, E., Sutrisno, B., & Wibowo, M. H., 2010. Performa dan histopatologi ayam broiler yang diinfeksi dengan *Salmonella pullorum* setelah pemberian imbuhan pakan mengandung tepung cacing tanah (*Lumbricus rubellus*). *Media Peternakan*, 33, 31-35.
- [27] Khatun, A., Carlos, A., Luois, A. and Hebel, P., 2012. Nutritional and toxicological evaluation of earthworm (*Eisenia foetida*) meal as protein source for animal feed. *Animal Feed Science and Technology* 42: 165- 172.
- [28] Son, (2009). The study on treatment of poultry waste by earthworms, and the effect of feeding earthworms' meal on the performance of broilers and laying hens, and safety of meat and egg. *Korean J. Organic Agric.*, 17 (1): 63-82.
- [29] Khalil, D. Hohler, and H. Henkel. 2005. Utilization of rice bran, earthworm meal and peanut meal in broilers. Characterization of the feed efficiency of rice bran/peanut meal diet. *Arch. Gefluegelkd.* 61: 88-94.
- [30] Mekada, H., N. Hayashi, H. Yokota, J. Okumura (1979). Performance of growing and laying chickens fed diets containing earthworms (*Eisenia foetida*). *Japan. Poult. Sci.*, 16: 293- 297.