

EFFECT OF DIETARY BLACK SOLDIER FLY, *Hermetia illucens* (Linnaeus) LARVAE MEAL AND POULTRY MEAL ON PRODUCTION PERFORMANCE, EGG QUALITY, AND NUTRIENT DIGESTIBILITY IN POST-PEAK CHICKEN LAYERS

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ABSTRACT

This study was conducted to determine the effects of feeding black soldier fly larvae meal (BSFLM) and poultry meal (PM) on production performance, acceptability of eggs through sensory evaluation, digestibility of dietary energy, fat, and protein in post-peak Babcock white layer chickens. A total of 192 54-week-old Babcock white layer hens were randomly allocated into one of four dietary treatments (T1 – control, T2 – 3% poultry meal (PM), T3 – 1.5% PM + 1.5% BSFLM, T4 – 3% BSFLM) with 16 replicates each, using randomized complete block design and with location of cages as blocking factor. There were no significant differences observed on hen-day egg production but the average egg weight was significantly greater ($P < 0.05$) in birds fed with 3% BSFLM and control than T2 and T3. Significant improvements ($P < 0.0001$) in albumen height, egg yolk color, and Haugh unit were observed with 3% BSFLM inclusion compared to T1 and T2. Inclusion of BSFLM in diets significantly increased the feed cost per bird but had no effect on the cost of feed per kilogram of eggs produced per bird. BSFLM supplementation at 3% did not have any significant effects on albumen texture, yolk color, yolk flavor, and overall acceptability of eggs. The results suggest that BSFLM can be included in the diet without any negative effect on overall acceptability at 3% inclusion. Inclusion of dietary BSFLM did not have a significant effect on the digestibility of dietary gross energy but significantly increased the digestibility of protein ($P < 0.0114$) and fat ($P < 0.0001$).

Key words: Stratiomyidae, Diptera, alternative protein source, supplemental animal protein, sensory evaluation

INTRODUCTION

Food insecurity is one of the fundamental challenges of today and, if left unaddressed, will continue to be a problem in the future. With the projection that the global human population will reach almost 10 billion by 2050, the demand for agricultural output is also expected to increase (Vos 2015). Such projections emphasize the need to consider the sustainability of various agricultural inputs. Poultry by-product meal or poultry meal (PM) is favored over fish meal due to the latter's effect on the taste of yolks and the problem in the overexploitation and the negative impact of the El Niño cycle led to a drastic decrease in the supply of fish meal from roughly eight million metric tons in the year 2000 to six million metric tons as of 2015 (Rabobank 2015). PM similar to Fish meal has a high crude protein

(CP) content which ranges from 58-68% on as fed basis, the relative abundance of amino acids, and high digestibility compared to soya and other sources. PM from poultry processing wastes provides a very sustainable source of protein for poultry; however, the price of PM is still high (Samli et al. 2006) but the cost is also high.

Insects are vastly present in any place on the globe, comprising as high as 80 million in diversity but with just roughly one million identified species (Stork et al. 2015). Its enormous biomass and ability to mass-produce biowaste with a high conversion rate make them a viable natural source of protein for animals, especially for poultry and fishes (Ravindran 2013). One of the most commonly mass-produced insects is the black soldier fly (*Hermetia illucens*) or BSF. The ability to efficiently convert various organic wastes, from kitchen wastes to human and animal manure, into large insect biomass in a short period of time makes BSF the most promising compared to other studied insects such as the common housefly, *M. domestica* (Pretorius 2011), meal worm, *T. molitor*, (Veldkamp et al. 2012), and locusts (Van Huis, et al. 2012). Approximately 42% crude protein and 29% crude fat render the BSF comparable to soya, and when defatted, the crude protein content can go as high as 60% crude protein which is close to the fish meal's protein content (Spranghers et al. 2017).

Insect meal can stabilize the demand for high-quality protein source which can result to much lower prices of raw materials and higher economic impact especially when reared by the farmers themselves (Onsongo et al. 2018). This lowers the input and maximizes the income of farmers, especially the smallholders. The overexploitation of fish for fish meal and fish oil production can also be minimized. Furthermore, the greenhouse gas emissions can also be reduced by shifting to insect production instead of converting rain forests into soya plantations. Black Soldier Fly farming GHG emission is 47% lower than of windrow composting (Mertenat et al. 2019.) Insects as decomposers can close the loop on the food chain by converting wastes into high quality protein source for animals, and possibly, for human consumption.

The study sought to determine the effect of the black soldier fly larvae meal (BSFLM) on production performance, egg quality, acceptability through sensory evaluation, cost of production, and nutrient digestibility of layer diets.

MATERIALS AND METHODS

Animals and experimental design. A total of 192 54-week-old Babcock white layers with an initial average weight of 1.58 kg were used in an 8-week production performance study. Layers were blocked by location and were randomly divided into four treatment groups. There were 48 birds per treatment, composed of 16 replicates of three birds per cage on a completely randomized block design. The layers were housed in A-type cages in an open-sided housing with a floor space of one square foot per hen. A total of 110 grams of feed per day were allocated to each bird with ad libitum access to water, under 16:8 (day:night) photoperiod. The study was carried out at the University Animal Farm of the University of the Philippines Los Baños located in Barangay Tuntungin-Putho, Los Baños, Laguna.

Experimental diets. Four experimental treatments were utilized in this study: 1) typical corn-soy based diet without BSFLM and poultry meal, 2) basal diet with 3% poultry meal (PM), 3) basal diet with 1.5% BSFLM and 1.5% PM, and 4) basal diet with 3% BSFLM. The BSFLM used was reared from a corn-coffee ground substrate, oven-dried and its particle size reduced with a mechanical grinder without any further defatting process. Poultry Meal (PM) used came from local supplier of poultry by-product meal. All dietary treatments were prepared in mashed form. The same diets were used in the digestibility study. The ingredient composition, calculated nutrient composition and analyzed nutrition composition of the experimental diets are shown in Tables 1, 2, and 3, respectively.

Table 1. Ingredient composition (as fed) of the experimental diets of post-peak chicken layers.

Ingredients %	Diet ¹			
	T1 Control	T2 3% (PM)	T3 1.5% PM + 1.5% BSFLM	T4 3% BSFLM Control
Yellow corn	48.4	52.2	51.1	48.7
US soya high protein	22.2	17.6	18.44	19.2
Rice bran D1	15.0	15.0	15.0	16.2
Palm oil	2.2	0.9	1.0	1.3
Monocalcium phosphate	1.2	0.7	1.0	1.2
Limestone	10.0	9.7	9.5	9.4
Salt, iodized	0.2	0.2	0.2	0.3
DL-Methionine	0.2	0.2	0.2	0.2
L-Threonine	0.1	0.1	0.1	0.1
L-Lysine	0.1	0.02	0.03	0.03
Choline chloride, 60%	0.1	0.1	0.1	0.1
Vitamin Premix	0.1	0.1	0.1	0.1
Mineral Premix	0.1	0.1	0.1	0.10
Toxin binder	0.05	0.05	0.05	0.05
Antioxidant	0.05	0.05	0.05	0.05
Poultry meal	-	3	1.5	-
BSF larvae meal	-	-	1.5	3.00
Total	100	100	100	100

¹BSFLM – Black soldier fly larvae meal.

²PM- Poultry Meal

Table 2. Calculated nutrient composition (as fed) of the experimental diets.

Item	Diet			
	T1 Control	T2 3% (PM)	T3 1.5% PM + 1.5% BSFLM	T4 3% BSFLM
ME, kcal/kg	2700	2700	2753	2700
Crude protein, %	16.5	16.5	16.5	16.5
Crude fiber, %	3.4	3.37	3.4	3.5
Crude fat, %	6.2	5.57	6.8	6.2
Linoleic acid, %	1.7	1.71	1.7	1.7
Digestible Amino Acid, %				
Lysine	0.8	0.84	0.8	0.8
Threonine	0.7	0.7	0.7	0.7
Methionine	0.4	0.4	0.4	0.5
Methionine+Cystine	0.7	0.7	0.7	0.7
Tryptophan	0.2	0.2	0.3	0.2
Mineral composition, %				
Ca	4.0	4.0	4.0	4.0
P, available	0.4	0.4	0.4	0.4

Table 3. Analyzed nutrient composition (as fed) of the experimental diets.

Item	Diet			
	T1 Control	T2 3% Poultry Meal (PM)	T3 1.5% PM + 1.5% BSFLM	T4 3% BSFLM Control
Dry matter, %	90.5	90	90.5	91.1
Crude protein, %	16.1	16.3	16.2	16.0
Crude fat, %	3.3	4.4	3.2	4.2
Crude fiber, %	3.5	4.5	3.3	3.8
Ash, %	11.1	11.3	11.0	12.9
Gross energy, kcal/kg	3485.5	3464.4	3489.6	3466.19

Production performance. Eggs were harvested, counted, and weighed daily for evaluation of production performance and egg quality. Total feed intake and feed refusal were recorded on a weekly basis.

Hen-Day Egg Production (HDEP). Collected eggs were counted for each treatment to determine HDEP. HDEP was computed by dividing the number of eggs laid by the number of birds per treatment, then multiplying the result by 100.

$$HDEP = \frac{\text{Total number of eggs laid}}{\text{Total number of birds}} \times 100$$

Average Egg Weight (AEW). All eggs upon collection were weighed and recorded. For each treatment, the total egg weight per cage was divided by the total number of eggs per cage to determine the AEW.

$$AEW = \frac{\text{Total egg weight}}{\text{Total number of eggs weighed}}$$

Economic parameter. The cost of feed consumed per bird for each treatment was calculated by multiplying the total feed intake (in kg) by the price per kg of feed.

$$\text{Feed cost per layer chicken (PHP)} = \text{Total feed consumed} \times \text{Price per kg of feed}$$

For each treatment, feed cost per kg of eggs produced was computed by dividing the total feed cost per chicken layer by the weight (in kg) of the eggs laid.

$$\text{Feed cost per kg of eggs produced (PHP)} = \frac{\text{Total feed cost}}{\text{Total kg of eggs produced}}$$

Feed efficiency. Feed efficiency was determined by dividing the total amount of feed consumed per bird by the total weight of laid eggs multiplied by 100.

$$\text{Feed Efficiency} = \frac{\text{kg of feed consumed}}{\text{kg of egg produced}} \times 100$$

Egg quality. Egg weight was determined using a digital scale and a digital (Mitutoyo Digital Vernier Caliper 500-196-30/197-30/173 MM/Inch Electronic Micrometer Gauge 0-150/200/300mm/0.01mm) caliper for albumen height and eggshell thickness. Egg yolk color was determined using a color (DSM YolkFan™) fan.

Haugh unit. Haugh unit was computed using the following formula:

$$HU: 100 \log (H + 7.5 - 1.7W^{0.37})$$

Where:

H = albumen height

W = egg weight in grams

Eggshell thickness. Eggshell thickness was determined by getting the average thickness of the top, mid, and bottom parts of the eggshell upon removal of the eggshell membrane.

Sensory evaluation. A total of 11 trained panelists participated in the sensory evaluation of egg characteristics. The test was conducted at the Sensory Evaluation Laboratory of the Animal Products Science and Technology Division, Institute of Animal Science, College of Agriculture and Food Science, University of the Philippines Los Baños, Laguna, Philippines. The panelists were placed in individual cubicles to avoid peer influence on the ratings. Evaluation was done in three separate sessions for three consecutive days. A total of 66 eggs were evaluated in terms of yolk color, albumen texture, yolk flavor, yolk off-flavor, and overall acceptability. Six eggs per treatment were randomly selected from the final week of the experimental period. Eggs were collected at the start of Week 8 at around 11:00 o'clock in the morning. All eggs were stored in the refrigerator to maintain freshness prior to cooking the following day. All egg samples were placed in a pan with boiling water (4L) for ten minutes. Boiled eggs were cooled with running tap water and shells were removed. Each egg was cut into halves and then quartered. Cut egg samples arranged in pre-coded tray using a 3-digit random numbers and served on a plate to the panelists in individual cubicles or stations for immediate evaluation. Samples from each treatment were served randomly to the panelists. Cold water was also served to the panelists for cleansing their palate in between tasting of samples. Yolk color, albumen texture, yolk flavor, yolk off-flavor, and overall acceptability were rated from zero to 100 using a 100-mm horizontal line sheet.

Digestibility of layer diets. A total of 96 layers were used for the digestibility study. Three birds per cage with eight replicates from four experimental diets were randomly selected for the trial using a completely randomized design. Layers were housed in metabolic cages with installed waterers and feeders. Aluminum trays were placed directly under the cages for excreta collection. Marker-guided (using titanium dioxide) total collection method was used in the study.

Digestibility calculations. The apparent total tract digestibility (ATTD, %) of crude protein (CP), crude fat (CFat), and gross energy (GE) were calculated using the following equation:

$$\text{ATTD of CP/GE/CFat, \%} = \frac{(\text{Feed intake} \times \text{CP/GE/CFat feed}) - (\text{Excreta output} \times \text{CP/GE/CFat excreta})}{(\text{Feed intake} \times \text{CP/GE/CFat feed})} \times 100$$

Feeding and excreta collection. Titanium dioxide was mixed in all diets at 0.40% of the total diet to indicate the start and end of the five-day total collection period. Excreta samples were weighed, labelled, dried, homogenized (after drying), and stored at -20°C until analyses. Impurities such as feathers and broken eggs seen on the excreta were removed to avoid contamination and overestimation of CP, GE, and CFat contents.

Chemical analysis. BSFLM, Poultry Meal and dietary treatment samples were analyzed for moisture, CP, CFat, crude fiber (CF), ash, and nitrogen-free extract (NFE), as well as GE, following the guidelines and procedures established by AOAC (2007). Amino and fatty acid profile were analyzed using high-performance liquid chromatography (HPLC) at the Upscience Labs Solutions, Vietnam. The pooled and stored excreta samples were further oven-dried to constant weight at 70°C. Dried samples were finely ground and analyzed for GE, CP, and CFat following the guidelines and protocols from AOAC (2007). All samples were analyzed in triplicates.

Statistical analysis. MIXED procedure (SAS Institute Inc., Cary, NC) of SAS was used in analyzing data with cage as experimental unit and diet and block as the fixed and random effects part of the model. Tukey-Kramer test was used as mean separation for the least square means for each independent variable. Orthogonal contrasts were used to compare the effect of each treatment on all production performance parameters and egg quality characteristics, sensory evaluation parameters, and digestibility of GE, CP, and CFAT. Significant level for each test was set at $P < 0.05$ to detect statistical significance.

RESULTS AND DISCUSSION

Production performance. Table 4 shows the analyzed nutrient composition of BSFLM and the locally sourced poultry meal. The CP content of BSFLM analyzed in this study (44.25 % CP) is similar to 44.2% CP for BSFLM reared on abattoir waste (Lalander 2019) and 42-44% CP reared on swine manure (St. Hilaire et al. 2007), but slightly higher than the 42.1% CP content published by Feedipedia.com.

Table 4. Nutrient composition of Black Soldier Fly Larvae Meal (BSFLM) and Poultry Meal (PM).

Component	Nutrient composition	
	BSFLM	PM
Dry matter, %	90.5	91.6
Crude protein, %	44.3	64.7
Crude fiber, %	7.8	1.8
Crude fat, %	38.8	14.9
Ash, %	6.4	14.8
Gross energy, kcal/kg	6328.3	4840.2

The observed CP content of BSFLM which was fed with a corn-coffee diet was different compared with previous studies. This may be attributed to nutrient composition of substrate used in mass producing the larvae (St. Hilaire et al. 2007). The high CP content corresponded to high quality amino acids present in BSFLM as shown in Table 5. The results of the amino acid profiling from this study and the high GE value can be attributed to the high CFat content of BSFLM at 38.82%, which agrees with the values reported by Barragan-Fonseca (2017); Choi (2013), Mutafela (2015), Mywaniki (2018), and St. Hilaire (2007.).

The Cfat,CP, and GE of both ingredients influenced the different levels of inclusion of corn and palm oil across dietary treatments to meet the layer requirement.

Table 5. Amino acid profile of Black Soldier Fly Larvae Meal (BSFLM)

AMINO ACID	Amount of AA(mg)/ 100g BSFLM
Alanine	3.1
Arginine	1.9
Aspartic acid	3.8
Cysteine	0.3
Glutamic acid	4.4
Glycine	2.2
Histidine	1.1
Isoleucine	1.8
Leucine	3.0
Lysine	2.4
Methionine	0.9
Phenylalanine	1.1
Proline	2.6
Serine	1.5
Threonine	1.6
Tryptophan	0.6
Tyrosine	2.1
Valine	2.4
Total Protein	36.8

Majority of the fatty acid present in BSFLM were the short chain fatty acid, lauric acid (Table 6). BSFLM is composed of 70% saturated fatty acid, 13.6% omega-9, and 13.4% omega-6 fatty acids. The variations of the nutrient composition of BSFLM and nature of its amino and fatty acid profile showed the ability of BSFL to adopt the nutrient profile of substrates used as feed (Shumo et al. 2019.) The higher the protein and fat content of the substrates are, the more likely to produce BSFLM with high CP and CFat content (Choi et al. 2013, Mutafella et al. 2015, and Mywaniki et al., 2018). The corn used in the study to feed the larvae had CP content of 8% but due to its high energy content, the rearing period of the BSF larvae ranged from 15-16 days but with a small to medium size larvae. The BSF larvae reared on pure coffee yielded a very small size of larvae over a longer rearing period of 30-40 days. The combination of coffee and corn increased the rearing duration to 21-25 days that allowed the larvae to accumulate more protein that translated to a higher CP content and fat (Tschirner and Simon 2015).

Table 6. Fatty acid profile of Black Soldier Fly Larvae Meal (BSFLM).

Fatty acid (FA)	% Relativity	Amount of FA (mg) /100g BSFLM
Total Saturated Fatty Acid	71.0	26554.2
C10:0 Capric	1.0	376.4
C12:0 Lauric	43.8	16621.0
C14:0 Myristic	7.6	2817.7
C15:0 Pentadecanoic	0.1	40.5

Fatty acid (FA)		% Relativity	Amount of FA (mg) /100g BSFLM
C16:0	Palmitic acid	15.4	5609.4
C17:0	Margaric	0.7	238.9
C18:0	Stearic acid	2.0	730.1
C20:0	Arachidic acid	0.2	60.5
C22:0	Behenic acid	0.1	28.2
C24:0	Lignoceric acid	0.1	31.5
Total Mono Unsaturated Fatty Acid			
C14:1 n-5	Myristoleic acid	0.1	40.5
C16:1 n-7	Palmitoleic acid	1.1	387.2
C17:1 n-8	9-cis-heptadecenoic acid	0.0	14.4
C18:1	Oleic acid	13.8	4921.2
C20:1 n-12	Cis-8-eicosenoic acid	0.1	38.9
Total PolyUnsaturated Fatty Acid			
C16:2	Hexadecadienoic acid	0.0	14.4
C18:2 n-6	Linoleic acid	13.5	4769.3
C18:3 n-3	alpha-linoleic acid	0.4	144.3
C18:4	Octadecatetraenoic acid	0.1	45.4
C20:5 n-3	Cis-5, 8,11,14,17- eicosapentaenoic acid (EPA)	0.0	13.8
SUMMARY			
Sum of omega-3	n-3	0.5	158.1
Sum of omega-6	n-6	13.4	4740.9
Sum of omega-9	n-9	13.6	4853.3
Sum of Trans fat		0.1	28.6
Saturated fatty acid		70.9	26569.5
Monounsaturated fatty acids (MUFA)		15.1	5383.7
Polyunsaturated fatty acids (PUFA)		14.1	4987.2
Total fatty acids		127.7	36940.4

Characteristics such as egg weight, albumen height, Haugh unit, egg yolk color, and eggshell thickness were measured to determine the quality of eggs produced by birds fed with experimental diets. Table 7 shows the hen-day egg production (HDEP) of birds fed with experimental diets formulated in this trial. It can be noted that no significant effect was observed for all experimental diets on the HDEP of birds. This is contrary to the result published by Al-Qazzaz et al. (2016) of significantly higher HDEP and house-hen egg production (HHEP) at 5% inclusion of BSFLM in the experimental diet. The increase in HDEP and HHEP from the above report was not realized in this study perhaps due to the lower inclusion of BSFLM in the diet at only 3%. The isocaloric and isonitrogenous formulated diets met the nutrient requirements for egg production and the inclusion of 3% BSFLM was, perhaps, inadequate to increase HDEP of layers used in this study.

It was generally observed that T1- and T4-fed birds expressed significantly higher AEW compared to T2- and T3-fed birds, with T2-fed birds having the lowest AEW. The increased egg weight observed in T4-fed birds was contrary to the results reported by Al-Qazzaz et al. (2016) and Mwaniki et al. (2018) where the lowest egg weight was observed from birds fed with diets including 5% BSFLM.

The score for egg yolk color in this study was higher ($P < 0.0001$) in T4 with 3% BSFLM compared to the rest of the treatments (Table 7). High score on egg yolk color is important due to the consumer preference towards eggs with golden yellow to orange yolk colors. Yolk color improvement in treatments with BSFLM can be explained by the elevated carotenoid concentration found in the BSFLM (Secci et al. 2018).

The highest albumen height was at T4 with 10.24 mm ($P < 0.0001$). Albumen height was observed to be higher in T4 ($P < 0.0001$) with 3% BSFLM, followed by T3 ($P = 0.0001$) with 1.5% BSFLM and 1.5% PM (Table 7). Average albumen height from the study ranged from 9.57 - 9.99 mm, which is higher than the results from Kawasaki et al. (2019) at 7.16 - 8.08 mm and from Ruhnke et al. (2018) at 8.91 - 9.18 mm.

None of the experimental diets in this study yielded significant effect on eggshell thickness. The average thickness of eggshell ranged from 0.32 - 0.33 mm, lower than the results from Ruhnke et al., (2018) at 0.44 - 0.46 mm, and within the range of the results from Park et al. (2017) at 0.33 – 0.44 mm. The thinner eggshell was due to the age difference of the birds since older birds produce bigger eggs with thinner shells.

Table 7. Effects of Black Soldier Fly Larvae Meal (BSFLM) and poultry meal on production performance of laying hens.

Item	Diet				SEM	P-value
	T1	T2	T3	T4		
	Control	3% PM	1.5% PM + 1.5% BSFLM	3% BSFLM		
Hen-day egg production, eggs	19.3	19.7	19.5	19.6	0.210	0.482
Average egg weight, g	64.1 _a	61.7 _b	62.9 _{ab}	63.9 _a	0.528	0.008
Albumen height, mm	9.6 _{bc}	9.4 _c	9.7 _b	10.0 _a	0.063	<0.0001
Haugh Unit	97.3 _{bc}	96.5 _c	97.7 _b	99.0 _a	0.296	<0.0001
Egg yolk color	5.6 _b	5.3 _b	6.6 _a	7.1 _a	0.216	<0.0001
Eggshell thickness, mm	0.3	0.3	0.3	0.3	0.005	0.749

¹ Least square means of 16 replicates per treatment with 3 birds per replicate

Haugh unit (HU) is dependent on the albumen height and egg weight and used as an indicator of egg freshness. It can be noted that the highest average significant HU value recorded was from T4 with 3% BSFLM at 99.3 ($P = 0.0004$) on week 8 of the trial. The HU in the study ranged from 96.5 (T1) to 99.03 (T4) at p-value of <0.0001. Higher albumen height corresponds to superior egg quality (Selim

et al. 2018). Results from this trial on HU are within the range set by USDA standards. Categories set by USDA on the quality of eggs are as follows: “AA” for HU value of > 72, “A” for HU values 60-72, “B” for HU values 31-60, and “C” for values < 31. Therefore, eggs from this study fell under AA category, indicating better quality eggs.

There were significant differences on the average feed cost per bird among the dietary treatments (Table 8). Feed costs per bird on diets containing BSFLM (T3 and T4) were significantly greater than in T1 and T2, with T4 having the highest feed cost per bird. This study showed that increasing the level of BSFLM in the diet will result in higher average feed cost per bird due to the present high cost of production of the BSFLM. However, this is compensated by the higher AEW resulting in T1 and T4 (P=0.008)

Table 8. Economic analysis on the inclusion of Black Soldier Fly Larvae Meal (BSFLM) and poultry meal in the experimental diets.

Item	Diet				SEM	P-value
	T1 Control	T2 3% PM	T3 1.5% PM + 1.5% BSFLM	T4 3% BSFLM		
Feed cost, Php per Kg	25.1	25.3	25.4	25.6		
Average total feed intake, Kg (56 days)	6.1	6.2	6.1	6.1	0.010	0.240
Average total egg weight per bird, Kg	3.3	3.2	3.3	3.32	0.041	0.369
Average feed cost per layer, Php	154.3 _c	155.4 _b	155.9 _b	165.8 _a	0.264	<0.0001
Average feed cost per kilogram of eggs (Php/kg)	47.0	48.3	48.2	47.3	0.633	0.384

¹ Least square means of 16 replicates per treatment with 3 birds per replicate

² Cost of BSFLM/kg used in the study is Php55.00

³ Cost of PM/kg used in the study is Php54.00

Sensory evaluation. Albumen texture, yolk color, yolk flavor, yolk off-flavor, and overall acceptance were not significantly affected by experimental diets with BSFLM as shown in Table 9. The result of the study was not consistent with Al-Qazzaz et al. (2018) who reported that increasing levels of BSFLM in the diet improved the appearance, texture, taste, and overall acceptance of the eggs. The level of glutamic acid content in eggs has shown interaction with human perception of taste (Yoshida et al. 1998). The enhancement on the flavor was seen to come from eggs treated with increasing level of BSFLM. Though BSFLM in this study has high levels of glutamic acid, the inclusion of 3% in the diet was probably inadequate to improve the taste of the eggs.

Table 9. Sensory evaluation of egg quality fed with Black Soldier Fly Larvae Meal (BSFLM) and poultry meal.

Item	Diet				SEM	P-value
	T1	T2	T3	T4		
	Control	3% PM ²	1.5% PM + 1.5% BSFLM	3% BSFLM ¹		
Albumen texture	46.8	46.8	45.7	40.5	3.266	0.252
Yolk color	49.6	52.0	47.5	49.3	2.863	0.496
Yolk flavor	68.2	66.0	67.4	68.3	0.512	0.548
Yolk off-flavor	0.8	1.6	1.1	0.9	0.566	0.512
Overall Acceptability	71.7	72.7	72.1	70.6	2.223	0.704

¹BSFLM – Black soldier fly larvae meal.

²PM- Poultry Meal

Digestibility of layer diets. Inclusion of BSFLM did not have a significant effect on the ATTD of GE (Table 10). Reports for ATTD of GE for layers fed with BSFLM are limited, hence, comparison with the result of the present study could not be made. However, ATTD of GE for broilers fed with BSFLM was reported to be lower than that of this study at 64% for *Tenebrio molitor* and 69% for BSFLM (De Marco et al. 2015). The difference in ATTD values can be explained by the differences in utilization of energy due to the development of gastrointestinal tract in broilers in contrast to layers. Significantly higher ($P<0.0001$) ATTD of CFat was observed in diets containing BSFLM than in the control. There was also a significant ($P<0.0114$) difference on ATTD of CP. Treatments supplemented with BSFLM had higher ATTD of CP than the control diet. This suggests that up to 3% BSFLM’s improved dietary protein and fat utilization without negatively affecting laying performance.

Table 10. Apparent total tract digestibility of crude fat, crude protein, and gross energy.

Item	Diet				SEM	P-value
	T1	T2	T3	T4		
	Control	3% Poultry Meal (PM)	1.5% PM + 1.5% BSFLM	3% BSFLM		
Crude fat, %	83.8c	89.1a	87.2ab	85.3bc	0.57	<0.0001
Crude protein, %	55.7b	61.6a	62.5a	61.6a	1.614	<0.0114
Gross Energy unit, %	79.8a	79.9a	81.6a	79.3a	0.808	0.1633

¹ Least square means of 8 replicates per treatment with 3 birds per replicate

This was supported by the results published by Marono et al. (2015) where chitin lowered the digestibility of protein *in vitro*. This is contrary to the study by De Marco et al. (2015) which reported that chitin from the BSFL exoskeleton had negative influence on the ATTD of nutrients in broiler chickens, with 25% inclusion of BSFLM in the diet.

SUMMARY AND CONCLUSIONS

Inclusion of 1.5% and 3% BSFLM and PM in the diets did not have a negative effect on hen-day egg production. The weight and size of eggs produced by BSFLM-fed birds were superior to those produced by PM-fed birds and comparable to the control. The inclusion of combined 1.5% BSFLM with 1.5% PM and 3% BSFLM significantly improved the egg quality in terms of albumen height, Haugh unit, and yolk color. The cost of feed per bird was higher with increasing level of BSFLM in the diet but the cost of feed per kilogram of egg produced was not affected due to the higher number and bigger sizes of eggs produced. BSFLM supplementation at 1.5% and 3% did not have a negative effect on albumen texture, yolk color, and yolk flavor. The overall acceptability of eggs produced by BSFLM-fed birds was similar to those produced by birds given other dietary treatments. The inclusion of 1.5% and 3% BSFLM in the diet increased the ATTD of crude protein and fat but did not have a negative effect on the ATTD of energy.

Therefore, the black soldier fly larvae meal can be used as an alternative protein source in the diet without affecting negatively production performance, egg quality in terms of higher average egg weight, albumen height, Haugh unit, egg yolk color, and digestibility of fat, protein, and energy in post-peak chicken layers.

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REFERENCES CITED

- Al-Qazzaz, M., D. Ismail, H. Akit, and L. Hakim Idris. 2016. Effect of using insect larvae meal as a complete protein source on quality and productivity characteristics of laying hens. *Rev. Bras. de Zootec.* 45: 518-523.
- Barragan-Fonseca, K. Dicke, and M. Van Loon, J. 2017. Nutritional value of the black soldier fly (*Hermetia illucens* L.) and its suitability as animal feed. *J. Insects as Food and Feed.* 3: 105-120.
- Choi, Y., K. Park, S. Nam, B. Jang, J. Kim, D. Kim, and D. Yu. 2013. The effect on growth performance of chicken meat in broiler chicks by dietary supplementation of black soldier fly larvae, *Hermetia illucens* (Diptera : Stratmyidae). *J. Sericultural and Entomol. Sci.* 51.1:30-35.
- De Marco, M., S. Martínez, F. Hernandez, J. Madrid, F. Gai., L. Rotolo, Belforti, M. Belforti, D. Bergero, H. Katz, S. Dabbou, A. Kovitvadh, I. Zoccarato, L. Gasco, and A. Schiavone. 2015. Nutritional value of two insect larval meals (*Tenebrio molitor* and *Hermetia illucens*) for broiler chickens: apparent nutrient digestibility, apparent ileal amino acid digestibility and apparent metabolizable energy. *Anim. Feed. Sci. Technol.* 209: 211-218.
- Lalander, C., S. Diener, C. Zurbrügg, and B. Vinneras. 2019. Effects of feedstock on larval development and process efficiency in waste treatment with black soldier fly (*Hermetia illucens*), *J. Cleaner Prod.*, 208: 211-219
- Mertenat, A., S. Diener, and C. Zurbrügg. 2019. Black Soldier Fly biowaste treatment – Assessment of global warming potential, *Waste Manag.* 84: 173-181
- Mutafela, R. N. 2015. High value organic waste treatment via Black Soldier Fly bioconversion. Onsite Pilot Study Dissertation. KTH Royal Institute of Technology, Stockholm, Sweden.

- Mwaniki, Z. and E. Kiarie. 2018. Standardized ileal digestible amino acids and apparent metabolizable energy content in defatted black soldier fly larvae meal fed to broiler chickens. *Can. J. Anim. Sci.* 99(2): 211-217.
- Mwaniki, Z., M. Neijat, and E. Kiarie. 2018. Egg production and quality responses of adding up to 7.5% defatted black soldier fly larvae meal in a corn–soybean meal diet fed to Shaver White Leghorns from wk 19 to 27 of age. *Poult. Sci.* 97:2829-2835
- Onsongo, V., I. Osuga, C. Gachuri, A. Wachira, D. Miano, C. Tanga, S. Ekesi, D. Nakimbugwe, and K. Fiaboe. 2018. Insects for income generation through animal feed: Effect of dietary replacement of soybean and fish meal with black soldier fly meal on broiler growth and economic performance. *J. Econ. Entomol.* 111:1966-1973
- Pretorius, Q. 2011. The evaluation of larvae of *Musca domestica* (common house fly) as protein source for broiler production. Ph.D. Dissertation, Stellenbosch University, Stellenbosch, South Africa.
- Rabobank. 2015. Rabobank World Seafood Trade Map 2015. Rabobank Industry Note. 486:1-9
- Ravindran, V. 2013; Poultry feed availability and nutrition in developing countries: Main ingredients used in poultry feed formulation. *Poult. Dev. Rev.* 60-63.
- Samli, H., N. Senkoylu, M. Ozduven, H. Akyurek, and A. Agma. 2006. Effects of poultry by product meal on laying performance egg quality and storage stability. *Pak. J. Nut.* 5:06-09
- Secci, G., F. Bovera, S. Nizza, N. Baronti, L. Gasco, G. Conte, A. Serra, A. Bonelli, and G. Parisi. 2018. Quality of eggs from Lohmann Brown classic laying hens fed black soldier fly meal as substitute for soya bean. *Animal.* 10:2191-2197
- Shumo, M., I.M. Osuga, F.M. Khamis, C.M. Tanga, K.K.M. Fiaboe, S. Subramanian, A. van Huis, and C. Borgemeister. 2019. The nutritive value of black soldier fly larvae reared on common organic waste streams in Kenya. *Sci. Rep. Ser. No. 9*, 10110. p.
- Spranghers, T., M. Ottoboni, C. Klootwijk, A. Oryn, S. Deboosere, B. De Meulenaer, J. Michiels, M. Eeckhout, P. De Clercq, and S. De Smet. 2018. Nutritional composition of black soldier fly (*Hermetia illucens*) prepupae reared on different organic waste substrates. *J. Sci. Food Agric.* 97: 2594–2600.
- Stork, N., J. Mcbroom, C. Gely, and A. Hamilton. 2015. New approaches narrow global species estimates for beetles, insects, and terrestrial arthropods. *Proc Natl Acad Sci USA.* 112(24):7519-23
- Tschirner, M. and A. Simon. 2015. Influence of different growing substrates and processing on the nutrient composition of black soldier fly larvae destined for animal feed. *J. Insects as Food and Feed.* 1 (4):249–259
- Van Huis, A., J. Van Itterbeeck, H. Klunder, E. Mertens, A. Halloran, G. Muir, and P. Vantomme. 2013. Edible insects: future prospects for food and feed security. *FAO For. Pap. No.* 171.
- Veldkamp, T., G. Van Duinkerken, A. Van Huis, C.M.M. Lakemond, E. Ottevanger, G. Bosch, and M.A.J.S. Van Boekel. 2012. Insects as a sustainable feed ingredient in pig and poultry diets – a feasibility study. In: Rapport 638. Wageningen Livestock Research, Wageningen, Netherlands
- Vos, R. 2015. Thought for Food: Strengthening global governance of food security. *CDP Background Paper No.* 29. 26 pp.