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

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ABSTRACT We examined egg production and quality responses of adding up to 7.5% defatted black soldier fly larvae meal (**BSFLM**) in a corn–soybean meal diet fed to pullets (19 to 27 wk of age). The concentration of CP and crude fat in BSFLM sample was 59.3 and 7.0% DM, respectively. A corn–soybean meal diet was formulated with 0 or 5.0 or 7.5% BSFLM and fed ($n = 6$) to a total of 108, 19-wk-old Shaver White pullets placed in conventional cages (6 birds/cage). The birds had free access to feed and water. Hen-day egg production (**HDEP**) and average egg weight were monitored daily and feed intake (**FI**) weekly. Egg quality parameters were assessed on individual eggs collected on the 5th d of wk 22, 24, and 26 and included individual EW (**IEW**), albumen height (**HU**), yolk color (**YC**), egg shell-breaking strength (**SBS**) and thickness (**ST**). A quadratic response ($P < 0.02$) was observed for HDEP, EW and egg mass. Specifically, birds fed 0 and 7.5% BSFLM diets had similar ($P > 0.05$) values for these parameters with birds fed 5.0% BSFLM showing lower ($P < 0.05$) HDEP than 0 or 7.5% BSFLM fed birds. The HDEP was 89.4, 84.8, and 87.8 for 0, 5.0, and 7.5% BSFLM, respectively. Feeding BSFLM linearly ($P < 0.01$) increased FI and feed conversion ratio (FCR) (FI/egg mass). There was no diet effect ($P > 0.05$) on IEW and HU, however, BSFLM linearly ($P = 0.02$) reduced CV of IEW. The IEW was 53.7, 52.3, and 53.0 g for 0, 5.0, and 7.5% BSFLM-fed birds, respectively and corresponding CV values of IEW were 7.9, 5.2, and 5.1%. Feeding BSFLM linearly ($P < 0.01$) increased YC, SBS, and ST. In conclusion, birds fed 7.5% BSFLM had similar HDEP and egg mass but poor FCR relative to corn–soybean meal diet without BSFLM. The effects of BSFLM on egg quality characteristics warrant further investigations.

Key words: black fly soldier fly meal, egg production, egg quality, feed conversion

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INTRODUCTION

Feed cost accounts for more than 65% of variable cost of producing poultry products, and energy and amino acids account for more than 90% of this cost (Kiarie et al., 2013). In the recent past, the global feed industry has seen soaring and volatile prices of traditional feedstuffs commonly used in livestock and poultry diets due to competition with the food and ethanol industries (Woyengo et al., 2014). Moreover, in the context of anticipated human population growth, the current animal protein production will need to increase 60% or more by 2050 (FAO, 2011). This increase in animal protein demand will need enormous resources, the feed being the most challenging because of the limited availability of natural resources, climate change pressure and food–feed–fuel competition (FAO, 2011). This trend has clearly demonstrated the danger of relying on a limited pool of ingredients to formulate feeds and underscored the need to characterize nutritive value of other feedstuffs with potential to serve as alternatives to or can complement traditional feedstuffs.

It has been estimated that more than 1.3 billion tons of organic waste are produced on a global scale resulting in enormous environmental, social, and economic costs (Makkar, 2017). In Canada, $27 billion worth of food ends up in landfills or composters each year (Parizeau et al., 2015). The nutrients in the organic waste could be recycled back for animal feeding through insect rearing (Rumpold and Schluter, 2013b; Makkar, 2017). Using insects as feedstuff can contribute to global food security via feed or as a direct food source for humans (Schader et al., 2015). Insects contain high amounts of energy, amino acids, fatty acids and micronutrients (Rumpold and Schluter, 2013a; Makkar et al., 2014).

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The insect species with the highest potential for large-scale production are the black soldier fly (**BSF**) (*Hermetia illucens*), common housefly (*Musca domestica*), and yellow mealworm (*Tenebrio molitor*). Specifically, BSF larvae achieve high growth rate and excellent conversion of organic waste to produce a meal (**BSFLM**) with consistent amino acid concentration when raised on diverse substrates (Diener et al., 2009; Nguyen et al., 2015; Spranghers et al., 2017).

The use of BSFLM as a component of diet has been reported for poultry (De Marco et al., 2015; Marono et al., 2017; Secci et al., 2018), swine (Newton et al., 1977), and for several commercial fish species (St-Hilaire et al., 2007). Although the feeding value of BSFLM in commercial poultry diets has been reported, this field is its infancy. Moreover, majority of insect meal research has focused on whole insect meal and manufacturers have started defatting the meal mechanically or chemically to increase protein fraction, improve meal keeping quality, and to create fat stream for other value-added applications (Fasakin et al., 2003; Schiavone et al., 2017). The typical crude fat content in whole BSFLM is 15 to 35% DM (Makkar et al., 2014) and defatting processes produce meal with crude fat content of as low as 5% DM depending on fat extraction procedures (Fasakin et al., 2003; Schiavone et al., 2017). Defatting has been shown to increase crude protein from 40 to 44% DM in whole BSFLM (Makkar et al., 2014; Spranghers et al., 2017) to a high of 65.5% DM (Schiavone et al., 2017). Moreover, defatted BSFLM was shown to have higher or comparable digestible amino acids concentration to typical animal and plant protein sources used in poultry feed (Schiavone et al., 2017). Defatting is also seen as critical control point for BSFLM as fat component has been shown to be the most variable component in larva grown on diverse substrates (Spranghers et al., 2017). To our knowledge, limited studies have been reported on feeding defatted BSFLM to poultry particularly the egg-laying strains. Therefore, the objective of the present study was to evaluate effects of 0, 5.0, and 7.5% inclusion of defatted BSFLM in practical corn–soybean diet fed to laying hens.

METHODS AND METHODS

The use of animals was approved by the University of Guelph Animal Ethics Committee and complied with the Canadian Code of Practice for the Care and Use of Animals for Scientific Purposes (CCAC, 2009).

Insect Meal and Diets

Defatted BSFLM (approximately 6% crude fat as fed) was procured from a commercial manufacturer and vendor (Enterra feed Corp., Vancouver, BC, Canada). The meal is a dry, powder product derived from larvae of the BSF (*Hermetia illucens*) reared on pre-consumer recycled food collected from local farms, food processors, and grocery stores. The meal is approved by the Canadian Food Inspection Agency for feeding poultry. A standard corn–soybean meal (0% BSFLM) diet was formulated to meet the nutrient requirements for 19 wk of age pullets according to Shaver White commercial management guidelines. The BSFLM was included at 5.0 and 7.5% to maintain iso-caloric and iso-nitrogenous specification (Table 2). All diets were prepared in crumble form at Arkell research station feed mill, University of Guelph.

Birds, Housing and Experimental Procedures

One hundred and eight, 19-wk-old pullets (Shaver White Leghorns) were placed in cages (6 birds per cage) and allocated to experimental diets based on BW in a completely randomized design to give 6 replicates per diet. The diets were fed from wk 19 to 27. The birds had free access to feed and water throughout the experimental period. Hen-day egg production (**HDEP**, number of eggs laid per d/number of hens) and average egg weight (**AEW**) per cage were recorded on daily basis. Feed intake was determined on weekly basis and BW at the end of wk 21, 23, 25, and 27. All eggs collected on the 5th d of wk 22, 24, and 26 were submitted for egg quality analyses on the same day.

Egg Quality Measurements The individual egg weight (**IEW**), height of albumin (haugh units, **HU**), and yolk color were determined by egg Analyzer (ORKA Food Technology Ltd, Ramat HaSharon, Israel). The system detect, calculate, and report values for yolk color (1 to 15 colors scale based on DSM/Roche yolk color fan), HU and egg weight (g). Prior to measurements, the unit was calibrated as per manufacturer recommendations. The egg shell thickness (**ST**) was measured using a high-resolution non-destructive device that measures ST without breaking using precision ultrasound (ESTG-1, ORKA Food Technology Ltd.). Briefly, gel was applied on the egg followed by placement of the egg on cradle to read ST in mm. Shell-breaking strength (**SBS**, kgf) was measured by Force Reader (ORKA Food Technology Ltd.), the unit measures accurately the breaking point of the egg shell by applying mechanical force on vertically placed egg on the cradle.

Chemical Analyses Samples of BSFLM and diets were finely ground in a coffee grinder and thoroughly mixed for analyses. Samples were analyzed for DM, CP, gross energy, crude fat, starch, ethanol soluble carbohydrates, neutral detergent fiber (**NDF**), and minerals. Dry matter determination was carried out according to standard procedures method 930.15 (AOAC, 2005). Nitrogen was determined by combustion method 968.06 (AOAC, 2005) using a CNS-2000 carbon, N, and sulfur analyzer (Leco Corporation, St. Joseph, MI). The CP values were derived by multiplying the assayed N values by a factor of 6.25. Gross energy was determined using a bomb calorimeter (IKA Calorimeter System C 5000; IKA Works, Wilmington, NC). The NDF content was determined according to Van Soest et al. (1991) using Ankom 200 Fiber Analyzer (Ankom Technology, Fairport, NY). Crude fat content was determined using ANKOM XT 20 Extractor (Ankom Technology, Fairport, NY). Samples for AA analysis were prepared by acid hydrolysis according to the method of AOAC (2005, method 982.30), and as modified by Mills et al. (1989). Briefly, about 100 mg of each sample was digested in 4 mL of 6 N HCl for 24 h at 110°C, followed by neutralization with 4 mL of 25% (wt/vol) NaOH and cooled to room temperature. The mixture was then equalized to 50 mL volume with sodium citrate buffer (pH 2.2) and analyzed using an AA Analyzer (Sykam, Germany). Samples for analysis of sulfur containing AA (Met and Cys) were subjected to performic acid oxidation prior to acid hydrolysis. Tryptophan was not determined. The samples were wet acid digested with nitric and perchloric acid mixture (AOAC, 2005; method 990.08) and concentrations of minerals (Ca, P, K, Mg, and Na) read on an inductively coupled plasma mass spectrometer (Varian Inc., Palo Alto, CA). Ethanol soluble carbohydrates and starch were analyzed in a commercial laboratory (SGS Canada Inc., Guelph, ON, Canada).

Table 1. Chemical composition of defatted black soldier fly larva meal, as fed basis.

Item	Amount
DM (%)	97.5
CP (%)	56.1
Gross energy (kcal/kg)	4973
Fat (%)	6.84
Starch (%)	5.97
Ethanol soluble carbohydrates (%)	4.55
Ca (%)	1.21
P (%)	0.95
K (%)	1.58
Mg (%)	0.39
Na (%)	0.20
Amino acids (%) (% of CP)	
Indispensable	
Arg	2.72 (4.8)
His	5.51 (10.1)
Ile	2.38 (4.2)
Leu	3.81 (6.8)
Lys	3.22 (5.7)
Met	0.9 (1.6)
Met + Cys	1.3 (2.3)
Phe	2.11 (3.8)
Thr	2.26 (4.0)
Val	3.38 (6.0)
Dispensable	
Ala	3.8 (6.8)
Asp	5.13 (9.1)
Cys	0.40 (0.7)
Glu	6.67 (11.9)
Gly	2.99 (5.3)
Pro	3.34 (6.0)
Ser	2.5 (4.5)
Tyr	2.76 (4.9)

Statistical Analyses

Data were analyzed using GLM procedures (SAS Inst. Inc., Cary, NC). The model had fixed effects of diet and wk and interaction. The cage was the experimental unit. Contrast coefficients from unequally spaced BSFLM were generated using the interactive matrix language procedure of SAS. An α level of $P \leq 0.05$ was used as the criterion for statistical significance.

RESULTS AND DISCUSSION

The analyzed chemical composition of BSFLM sample and experimental diets are shown in Tables 1 and 3, respectively. The crude fat concentration was lower than the values of 15 to 35% DM reported for non-defatted BFLSM (Makkar et al., 2014) but comparable to defatted BSFLM sample (Marono et al., 2017; Schiavone et al., 2017). The concentration of CP was higher than the values of 40 to 44% DM for whole BSFLM (Makkar et al., 2014; Spranghers et al., 2017) but within the range of 47.6 to 65.5% DM for defatted BSFLM (Marono et al., 2017; Schiavone et al., 2017). Crude protein variations in BSFLM are indications of variable fat and chitin concentrations as well as growth substrates (Liu et al., 2012). The concentrations of Lys, His, and Val were higher than the values reported for defatted BSFLM (~crude fat 4.7% DM) (Schiavone et al., 2017). However, the concentrations of other AA were comparable to defatted sample (Schiavone et al., 2017). The concentration of Ca was somewhat lower (5 to 8% DM) whereas concentration of P comparable (0.6 to 1.5% DM) to literature values (Makkar et al., 2014). Black soldier fly larvae are converters of organic waste into edible biomass, of which the composition of the meal depends on the substrate (Diener et al., 2009; Nguyen et al., 2015). However, in a recent study, it was demonstrated that the concentration of CP and AA is very consistent, and fat was variable in a meal from larvae grown on diverse substrates (chicken feed, vegetable waste, biogas digestate, and restaurant waste) (Spranghers et al., 2017). This suggested a defatted BSFLM could be a very attractive protein feed ingredient for poultry diets. The diet with 5% BSFLM assayed slightly lower CP relative to other diets but was above formulation target of 17% (Table 2). However, the AME and AA concentrations were comparable across all diets.

The effects of BSFLM inclusion on HDEP are shown in Table 4. There was a quadratic effect ($P < 0.01$) on HDEP with birds fed 5% BSFLM showing lower HDEP than birds fed 0 or 7.5% BSFLM. Similarly, BSFLM inclusion had quadratic ($P < 0.021$) response on AEW and egg mass with 5% BSFLM-fed birds showing lower EW relative to 0% BSFLM. Feed intake was linearly and quadratically increased ($P < 0.05$) by inclusion of BSFLM with birds fed 7.5% BSFLM showing the highest feed intake relative to the 0 or 5% BSFLM. As a result, a linear ($P = 0.003$) increase in feed

Table 2. Composition of experimental diets, *as fed*.

Item	Black fly soldier larvae meal (%)		
	0.0	5.0	7.5
Corn	43.3	46.7	48.2
Soybean meal	25.4	18.5	15.1
Wheat	15.0	15.0	15.0
Black fly soldier larvae meal	–	5.0	7.5
Soy oil	3.27	1.92	1.30
Limestone fine, <1 mm	7.29	7.21	7.17
Limestone coarse, 2 to 4 mm	2.19	2.16	2.15
Mono calcium phosphate	2.00	1.98	1.98
Vitamin-trace premix[1]	1.00	1.00	1.00
Salt	0.37	0.29	0.33
DL-methionine	0.2	0.21	0.22
Sodium bicarbonate	0.04	0.05	0.05
L-Lysine	0.01	–	–
L-Threonine	–	0.01	0.02
Calculated provisions			
AME (kcal/kg)	2800	2800	2800
Crude protein (%)	17.0	17.0	17.0
Crude fat (%)	5.14	4.32	3.96
SID Lys (%)	0.75	0.75	0.75
SID Met (%)	0.43	0.46	0.48
SID Met + Cys	0.67	0.67	0.67
SID Try (%)	0.20	0.17	0.16
SID Thr (%)	0.52	0.52	0.53
Ca (%)	4.10	4.10	4.10
Available P (%)	0.44	0.44	0.44
Na (%)	0.18	0.18	0.18
Cl (%)	0.25	0.20	0.23

[1] Vitamin mineral premix provided per kilogram of premix: vitamin A, 880,000 IU; vitamin D3, 330,000 IU; vitamin E, 4,000 IU; vitamin B12, 1,200 mcg; biotin, 22,000 mcg; menadione, 330 mg; thiamine, 400 mg; riboflavin, 800 mg; pantothenic acid, 1,500 mg; pyridoxine, 300 mg; niacin, 5,000 mg; folic acid, 100 mg; choline, 60,000 mg; iron, 6,000 mg; and copper, 1,000 mg.

Table 3. Analysed chemical composition of experimental diets, as fed basis.

Item	Black fly soldier larvae meal (%)		
	0	5	7.5
Dry matter (%)	89.3	88.8	89.2
Gross energy (kcal/kg)	3559	3533	3482
Crude protein (%)	18.5	17.3	18.1
Crude fat (%)	4.32	4.13	3.44
Starch (%)	37.9	37.8	37.7
Ethanol soluble carbohydrates (%)	3.79	3.53	3.15
NDF (%)	7.97	8.30	10.68
Ca (%)	3.73	3.66	4.08
P (%)	0.74	0.73	0.80
K (%)	0.77	0.73	0.77
Mg, %	0.18	0.18	0.20
Na (%)	0.16	0.16	0.17
Indispensable amino acids (%)			
Arg	1.08	1.00	1.03
His	0.54	0.67	0.77
Ile	0.66	0.70	0.72
Leu	1.46	1.35	1.41
Lys	0.93	0.91	0.96
Met	0.47	0.49	0.44
Met + Cys	0.74	0.74	0.65
Phe	0.89	0.71	0.75
Thr	0.67	0.64	0.67
Val	0.76	0.85	0.88
Dispensable amino acids (%)			
Ala	0.88	0.74	0.94
Asp	1.77	1.61	1.69
Cys	0.28	0.25	0.22
Glu	3.46	3.07	3.21
Gly	0.74	0.73	0.77
Pro	1.12	1.09	1.10
Ser	0.95	0.85	0.92
Tyr	0.67	0.56	0.67

conversion ratio (**FCR**) was observed with increasing level of BSFLM. Generally, HDEP, AEW, egg mass, and FI increased as expected from wk 19 to 27. However, interaction ($P < 0.05$) between diet and wk was observed for egg mass and FI.

Energy and AA intakes are the greatest driver of egg production and egg size (Leeson and Summers, 2005). The 5% BSFLM diet had comparable AME and AA with other diets, it is thus rather difficulty to explain why we observed reduced HDEP and EW in birds fed this diet. A total replacement of soybean meal with defatted BSFLM (17% BSFLM) in diets for Lohmann Brown classic (wk 24 to 45) indicated birds-fed BSFLM had lower HDEP, EW, egg mass, and FCR than birds-fed soybean meal (Marono et al., 2017). In contrast, egg production, feed intake, and FCR of Lohmann White Leghorn laying hens-fed (wk 64–74) diets in which defatted BSFLM replaced 100% of soybean cake (24% BSFLM) were unaffected by dietary treatments (Maurer et al., 2016). The differences in responses in the current study and others (Maurer et al., 2016; Marono et al., 2017) could be ascribed to the age and strains of birds (brown consume feed). Thus, feed intake increased with inclusion of BSFLM. The beginning of lay is physically a very challenging time for the young hens. As a result, negative nutrient balances can occur (Leeson and Summers, 2005). At the onset of lay, the bird is not only adjusting to her new environment, but she must consume enough energy and nutrients for her body weight development and to reach the high peak in egg production. It is imperative to increase their feed intake from the end of the growing period towards the peak of production in a short time. Increased feed consumption was also observed in laying quails fed up to 10% BSFLM in practical diets (Widjastuti et al., 2014). The high feed intake of birds-fed BSFLM might be due to higher fiber content in the form of chitin (Liu et al., 2012). Feed composition in terms of nutrient content and nutrient balance is an important determinant of voluntary feed intake in poultry. In general, when factors such as ingredient composition (e.g., fiber), health, and genotype are standardized, evidence suggests that, chickens offered feed *ad libitum* will consume feed to meet their requirement of the first limiting nutrient, which in most cases are energy yielding nutrients (Newcombe and Summers, 1984). In poultry, dietary fiber affects availability of energy and nutrients, and thus birds-fed fibrous diet will consume more of that diet. The implications for the current study were such that the birds fed 7.5% BSFLM consumed more feed to meet requirements. In this context, it is noteworthy that 7.5% BSFLM diet had slightly lower crude fat and gross energy concentration compared to other diets. Feeding BSFLM linearly increased BW in wk 23 and 27 (Table 5). In wk 27, the BW was 1.67, 1.71, and 1.72 kg for 0, 5.0, and 7.5% BSFLM, respectively. In contrast,

Table 4. Effects of inclusion of black soldier fly larva meal (BSFLM) in corn–soybean meal diet fed to laying pullets (19 to 27 wk of age) on egg production, average egg weight, egg mass, and FCR.

Item	Hen day egg production (%)	Average egg weight (g)	Egg mass (g/d)	Feed intake (g/bird/d)	FCR
Main effect of BSFLM inclusion (%)					
0.0	89.4^{a}	50.5^{a}	45.8^{a}	92.2^{b}	2.256^{b}
5.0	84.8^{b}	50.1^{b}	43.0^{b}	92.0^{b}	$2.385^{a,b}$
7.5	87.8^{a}	$50.4^{a,b}$	$44.8^{a,b}$	95.7^{a}	2.430^{a}
SEM	0.642	0.123	0.327	0.350	0.042
Main effect of age (wk)					
19	46.0^{d}	42.3^{g}	19.4^{g}	63.8^{f}	4.250^{a}
20	73.9^{c}	44.2^{f}	32.7^{f}	89.4^{d}	3.050^{b}
21	89.0^{b}	46.5^{e}	41.4^{e}	76.7^{e}	1.904^{c}
22	94.4^{a}	49.8^{d}	47.0^{d}	94.6^{c}	2.044^{c}
23	96.3^{a}	51.8^{c}	49.9^{c}	96.5^{c}	1.954^{c}
24	96.0^{a}	53.4^{b}	51.3^{b}	97.0^{c}	1.905^{c}
25	96.2^{a}	$54.4^{a,b}$	$52.3^{a,b}$	102.2^{b}	1.973^{c}
26	96.6^{a}	54.9^{a}	53.1^{a}	110.2^{a}	2.089^{c}
27	97.2^{a}	55.3^{a}	53.7^{a}	109.1^{a}	2.045^{c}
SEM	1.029	0.197	0.511	0.561	0.067
Probabilities					
BSFLM	<0.01	0.038	<0.01	<0.01	0.010
Week	<0.01	<0.01	<0.01	<0.01	<0.01
BSFLM*wk	0.089	0.237	0.031	<0.01	0.797
Response to BSFLM inclusion					
Linear	0.005	0.263	0.002	<0.01	0.003
Quadratic	<0.01	0.021	<0.01	<0.01	0.794

Note: Means assigned different letters (a–f) within a factor of analysis (BSFLM, wk) are significantly different, $P < 0.05$.

Table 5. Effects of inclusion of black soldier fly larva meal (BSFLM) in corn–soybean meal diet fed to laying pullets (19 to 27 wk of age) on body weight (kg).

	Black soldier fly larva meal inclusion (%)					BSFLM response	
Week	0	5.0	7.5	SEM	*P*-value	Linear	Quadratic
19[1]	1.376	1.382	1.394	0.018	0.782	–	–
21	1.377	1.425	1.419	0.016	0.097	0.052	0.335
23	1.409^{b}	1.544^{a}	1.453^{b}	0.020	<0.01	0.031	<0.01
25	1.650	1.683	1.693	0.015	0.141	0.052	0.798
27	1.668^{b}	1.709^{b}	1.724^{a}	0.014	0.034	0.011	0.853

[1] Intial body weight.

Note: Means assigned different letters (a, b) within a row are significantly different, $P < 0.05$.

Lohmann Brown classic (wk 24 to 45) hens fed 17% BSFLM had lower body weight because of depressed feed intake (Marono et al., 2017). Specific studies in application of BSFLM in poultry feeding have focused on growing poultry and limited studies exist in layers. As a component of a complete diet, BSFLM was reported to increase quails body weight gain driven by increased FI (Widjastuti et al., 2014).

The data for individual eggs collected on wk 22, 24, and 26 are shown in Table 6. There was no diet effect ($P > 0.05$) on IEW and HU, however, BSFLM linearly ($P = 0.02$) reduced CV of IEW. The IEW values were 53.7, 52.3, and 53.0 g for 0, 5.0, and 7.5% BSFLM, respectively and corresponding CV values were 7.9, 5.2, and 5.1%. This observation suggested that feeding BSFLM improved uniformity of egg size an important metric for egg producers. There was no wk and interaction ($P > 0.05$) on mean and CV of IEW, HU, and YC. Generally, the mean and CV of HU and YC increased from wk 22 to 26. Feeding BSFLM linearly ($P < 0.01$) increased yolk color, SBS, and ST. The yolk color improvement suggested the meal had pigments that increased intensity of yolk color. Indeed, recent report demonstrated that feeding laying hens BSFLM increased concentration of γ-tocopherol, lutein, β-carotene, and total carotenoids compared with egg yolks from birds-fed soybean meal (Secci et al., 2018). The improved egg shell characteristics were indicative of either improved Ca absorption in the gut and improved Ca metabolism or both. Indeed, although egg shell quality was not reported, feeding laying hens (wk 24 to 45) 17% BSFLM increased circulating serum Ca levels relative to the control (0% BSFLM) despite the 2 diets having similar Ca concentration (Marono et al., 2017). Edible insects contain a significant amount of fiber in the form of chitin (Finke, 2007; Liu et al., 2012). Egg shell is 99% calcium carbonate and daily egg shell formation equate to a removal of 2 to 3 g of Ca equivalent to 10% of the hen body Ca reserve (Gilbert, 1983; Etches, 1987). About 60 to 75% of Ca in egg shell is derived from diet and 25 to 40% was from the skeletal stores (Comar and Driggers, 1949).

Table 6. Effects of inclusion of black soldier fly larva meal (BSFLM) in corn–soybean meal diet fed to laying pullets (19 to 27 wk of age) on egg quality characteristics.[1]

	Individual egg weight		Haugh units		Yolk color		Shell breaking strength		Shell thickness	
Item	Mean (g)	CV (%)	Mean (mm)	CV (%)	Mean	CV (%)	Mean (kgf)	CV (%)	Mean (mm)	CV (%)
Main effects of BSFLM inclusion (%)										
0.0	53.7	7.92^{a}	64.6	19.1	4.32^{b}	20.5	4.70^{b}	15.0	0.404^{b}	11.2
5.0	52.3	5.15^{b}	67.0	19.9	4.68^{a}	11.7	5.23^{a}	14.7	0.427^{a}	9.11
7.5	53.0	5.08^{b}	68.1	21.8	4.83^{a}	11.3	4.95^{b}	13.7	0.431^{a}	7.57
SEM	0.40	0.916	2.02	2.43	0.091	3.61	0.083	1.69	0.006	1.620
Main effects of age (wk)										
22	50.2^{c}	5.80	58.9^{b}	24.7^{a}	4.22^{b}	23.4^{a}	5.10^{a}	16.2	0.406^{b}	11.8
24	53.2^{b}	6.33	72.7^{a}	16.1^{b}	4.87^{a}	10.4^{b}	5.02^{a,b}	14.2	0.434^{a}	7.98
26	55.6^{a}	6.01	68.1^{a}	20.1^{a,b}	4.73^{a}	9.63^{b}	4.75^{b}	13.1	0.421^{a,b}	8.10
SEM	0.40	0.915	2.02	2.43	0.091	3.62	0.082	1.69	0.006	1.620
Probabilities										
BSFLM	0.070	0.053	0.464	0.737	<0.01	0.136	<0.01	0.857	0.003	0.294
Week	<0.01	0.916	<0.01	0.053	<0.01	0.015	0.010	0.417	0.005	0.179
BSFLM*wk	0.133	0.441	0.893	0.825	0.191	0.595	0.038	0.283	0.047	0.255
Response to BSFLM inclusion										
Linear	0.123	0.021	0.218	0.482	<0.01	0.057	0.007	0.616	<0.01	0.122
Quadratic	0.081	0.448	0.978	0.742	0.858	0.557	<0.01	0.813	0.515	0.870

Note: Means assigned different letters (a, b) within a factor of analysis (BSFLM, wk) are significantly different, $P < 0.05$.

Calcium homeostasis is created through a balance between intestinal absorption, renal excretion, and bone mineral metabolism to meet the bird's requirements (Elaroussi et al., 1994). Although we did not quantify chitin in the present study, dietary NDF content increased with addition of BSFLM (Table 3). It is plausible that the high fiber may have increased ceca fermentation (Kiarie et al., 2014). Increased hindgut fermentation has been shown to increase mineral absorption (Metzler-Zebeli et al., 2010) and thus better egg shell quality in hens-fed BSFLM. Indeed, total replacement of soybean meal with BSFLM in laying hens diet from 24 to 45 wk of age resulted in a higher caecal production of butyric acid (Cutrignelli et al., 2017). It is also possible other mechanisms related to feeding BSFLM may have influence strong egg shell characteristics.

Characterizing nutritive and functional value of insect meal, risks, and potential economic benefits when formulated correctly in practical diets will be pivotal for the feed industry uptake. Our data shows that feeding up to 7.5% BSFLM supported similar performance to corn–soybean meal diet in early phase of egg production. However, the quadratic response on egg production and poor FCR warrant further investigations. Stronger egg shell might be indicative of improved Ca metabolism in birds-fed BSFLM.

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