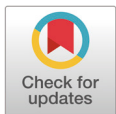


Evaluation of black soldier fly larvae oil as a dietary fat source in broiler chicken diets

Byeonghyeon Kim, Han Tae Bang, Ki Hyun Kim, Min Ji Kim, Jin Young Jeong, Ju Lan Chun and Sang Yun Ji*

National Institute of Animal Science, Rural Development Administration, Wanju 55365, Korea



Received: Nov 26, 2019
 Revised: Jan 3, 2020
 Accepted: Feb 17, 2020

*Corresponding author

Sang Yun Ji
 National Institute of Animal Science,
 Rural Development Administration,
 Wanju 55365, Korea.
 Tel: +82-63-238-7455
 E-mail: syjee@korea.kr

Copyright © 2020 Korean Society of Animal Sciences and Technology. This is an Open Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (<http://creativecommons.org/licenses/by-nc/4.0/>) which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

ORCID

Byeonghyeon Kim
<https://orcid.org/0000-0003-4651-6857>
 Han Tae Bang
<https://orcid.org/0000-0003-0441-3542>
 Ki Hyun Kim
<https://orcid.org/0000-0002-9834-2126>
 Min Ji Kim
<https://orcid.org/0000-0003-2106-1921>
 Jin Young Jeong
<https://orcid.org/0000-0002-8670-7036>
 Ju Lan Chun
<https://orcid.org/0000-0002-4618-586X>
 Sang Yun Ji
<https://orcid.org/0000-0001-7235-3655>

Competing interests

No potential conflict of interest relevant to this article was reported.

Funding sources

This study was carried out with the support of "Cooperative Research Program for Agriculture Science

Abstract

The present study was conducted to evaluate the effects of black soldier fly larvae oil (BSFLO) from the black soldier fly larvae (BSFL) as a partial or total replacement of soybean oil (SBO) on growth performance, fatty acid (FA) profile, and meat quality of broiler chickens from 1 to 5 wk of age. A total of 210 male broiler chickens (Ross 308) at one-day of age were randomly allotted to 3 dietary treatments (10 replicates and 7 birds/group): a basal control diet (CON), the basal diet in which the SBO was replaced by 50% (50 BSFLO) or 100% (100 BSFLO) of BSFLO. The growth performance, physical measurements and chemical traits of leg meat, and sensory analysis of breast meat were not influenced by diets. However, the relative weight (g/kg) of gizzard of CON was significantly higher (14.85, 12.52, and 13.02 for CON, 50 BSFLO, and 100 BSFLO; $p < 0.05$) than that of other treatments. As expected, the FA profile of breast meat was affected by BSFLO inclusion. The proportion (%) of saturated fatty acid (SFA) was increased (27.16, 27.58, and 28.72 for CON, 50 BSFLO, and 100 BSFLO; $p < 0.05$) by BSFLO inclusion and the percentage (%) of MUFA was also increased (43.36, 44.58, and 48.55 for CON, 50 BSFLO, and 100 BSFLO; $p < 0.01$). On the contrary, the proportion (%) of PUFA was decreased (29.49, 27.84, and 22.74 for CON, 50 BSFLO, and 100 BSFLO; $p < 0.01$). In conclusion, the present study suggests that the replacement of BSFLO did not show an adverse effect on growth performance and it could be an ingredient as a dietary fat source for a broiler diet.

Keywords: Black soldier fly larvae oil, Growth performance, Fatty acid profile, Meat quality

INTRODUCTION

Soybean oil (SBO) is the main dietary fat source for poultry diets because of its high energy content and digestibility [1]. However, the limited supply and the high price of the soybean are the critical aspects of poultry feed. The evaluation of the alternative ingredient as a dietary fat source in the broiler diet is therefore required.

Among the insect species, the black soldier fly (BSF, *Hermetia illucens*) is a promising species because of its ability to dissolve organic matter such as spoiled feed and manure [2,3]. The BSF larvae (BSFL) consume organic waste and store protein and fat in their bodies to supply the nutrients for the pupal period and adult stage [3]. Moreover, the nutritional component of BSFL depends on what they

and Technology Development (Project No. PJ01456002)" Rural Development Administration, Korea.

Acknowledgements

This study was supported by the 2019 RDA Fellowship Program of the National Institute of Animal Science, Rural Development Administration, Korea.

Availability of data and material

Upon reasonable request, the datasets of this study can be available from the corresponding author.

Authors' contributions

Conceptualization: Bang HT, Ji SY.

Data curation: Kim B.

Formal analysis: Kim B, Kim KH.

Methodology: Kim KH, Kim MJ, Jeong JY, Chun JL.

Software: Kim B, Kim MJ.

Validation: Kim B, Jeong JY.

Investigation: Bang HT, Ji SY.

Writing - original draft: Kim B, Bang HT.

Writing - review & editing: Jeong JY, Chun JL, Ji SY.

Ethics approval and consent to participate

This study was approved by IACUC of Rural Development Administration (No. NIAS-2019-1710).

were fed [4]. Although the contents of protein and fat in the larvae meal and oil can be modulated by their feed and defatting procedure, the BSFL contains 42% of protein and 35% fat and it has shown the possibility as a protein and fat source for poultry [2,3,5].

The use of the BSFL meal (BSFLM) and oil in the broiler diet have been reported [6–8]. The inclusion of BSFLM in broiler chicken diets up to 100 g/kg had no negative effects on growth performance and meat quality [8]. Furthermore, Dabbou et al. [9] also reported that low level (100 g/kg) inclusion of the BSFLM in a starter broiler diet may improve growth performance. It seems that low-level inclusion of BSFLM may be a more appropriate method as they showed a common result that increasing the level of BSFLM can cause a decrease in body weight [8,9]. However, the replacement of black soldier fly larvae oil (BSFLO) had no detrimental effects on growth performance, intestinal morphology, and health, even though the SBO in a diet was fully replaced [6,7]. All these studies suggested that the use of BSFLO in a broiler diet is suitable. Nevertheless, information about the inclusion of BSFLO in a broiler diet on the meat quality and fatty acid (FA) profile is scarce. For this reason, the objective of the present study was to evaluate the BSFLO on growth performance, meat quality, and FA profile of broiler chickens.

MATERIALS AND METHODS

Birds and diets

The present study was conducted in the poultry facility of the National Institute of Animal Science of Korea. A total of 210 male broiler chicks (Ross 308) at one day of age were randomly allotted to 3 dietary treatments (10 pens/treatment and 7 birds/pen). The poultry house was equipped with an automatic ventilation system. Each pen was equipped with a feeder and an automatic drinker. On day 21, chicks were randomly allocated to individual cages with a feeder and drinker.

The study was performed to evaluate the effects of a partial or total replacement of SBO with BSFLO on broiler chickens using two levels of inclusion. The diets were a control diet based on corn and soybean meal (SBM) and 50 and 100% replacement of SBO with BSFLO. The diets were fed in three phases: starter (day 1 to 7), grower (day 7 to 21), and finisher (day 21 to 35). All diets were formulated to meet or exceed the NRC [10] requirements (Table 1). Feed and water were provided *ad libitum* throughout the trial.

Growth performances

Health status and mortality were recorded daily during the whole experimental period. The initial body weight (IBW) and the final body weight (FBW) were recorded on days 1 and 35. Daily feed intake (DFI), average daily gain (ADG), and feed conversion ratio (FCR) were determined for the overall experimental period (1 to 35 days).

Slaughtering procedure

Birds were grown to the age of 35 days and then slaughtered. Before slaughter, 33 birds (11 birds/treatment) were chosen on the basis of average final live weight. Head, neck, and feet were removed and the length and weight of three sections of the small intestine were measured. The weight of the gizzard, liver, pancreas, spleen, bursa of Fabricius were also immediately weighed and recorded. A total of 33 chicken leg and breast meat samples were individually vacuum sealed and chilled at 4 °C to analyze physical measurements, chemical traits, oxidative parameters, and FA profile.

Physical measurements and chemical traits of chicken leg meat

Each 5 g of chicken leg meat samples (6 replicates) was homogenized with 15 mL distilled water

Table 1. Ingredients and chemical composition of the experimental diets

Items	Starter			Grower			Finisher		
	CON	50 BSFLO	100 BSFLO	CON	50 BSFLO	100 BSFLO	CON	50 BSFLO	100 BSFLO
Ingredients (%)									
Corn	53.05	53.05	53.05	59.00	59.00	59.00	61.80	61.80	61.80
Soybean meal (44%)	30.00	30.00	30.00	30.00	30.00	30.00	30.00	30.00	30.00
Corn gluten meal	6.35	6.35	6.35	4.00	4.00	4.00	1.60	1.60	1.60
Wheat bran	3.15	3.15	3.15	-	-	-	-	-	-
Soybean oil	3.00	1.50	-	3.00	1.50	-	3.00	1.50	-
Black soldier fly larvae oil	-	1.50	3.00	-	1.50	3.00	-	1.50	3.00
Dicalcium phosphate	1.75	1.75	1.75	1.50	1.50	1.50	1.45	1.45	1.45
Limestone	1.25	1.25	1.25	1.15	1.15	1.15	1.05	1.05	1.05
Vitamin-mineral premix ¹⁾	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
L-Lysine	0.40	0.40	0.40	0.25	0.25	0.25	0.10	0.10	0.10
DL-Methionine	0.20	0.20	0.20	0.20	0.20	0.20	0.15	0.15	0.15
Salt	0.35	0.35	0.35	0.40	0.40	0.40	0.35	0.35	0.35
Calculated composition									
ME (kcal/kg)	3,031	3,031	3,031	3,106	3,106	3,106	3,152	3,152	3,152
Lysine (%)	1.42	1.42	1.42	1.26	1.26	1.26	1.11	1.11	1.11
Methionine (%)	0.53	0.53	0.53	0.50	0.50	0.50	0.43	0.43	0.43
Ca (%)	0.96	0.96	0.96	0.91	0.91	0.91	0.86	0.86	0.86
Total P (%)	0.77	0.77	0.77	0.70	0.70	0.70	0.65	0.65	0.65
Available P (%)	0.46	0.46	0.46	0.41	0.41	0.41	0.36	0.36	0.36
Analyzed composition									
Crude protein (%)	24.63	24.48	23.28	22.15	22.77	23.14	22.71	22.03	21.37
Crude fat (%)	3.63	5.39	5.93	6.76	7.51	7.70	4.68	4.44	5.22
NDF (%)	8.95	10.51	10.27	11.00	12.49	14.43	11.83	16.39	12.55
ADF (%)	3.93	4.17	3.78	3.88	3.56	3.45	4.13	4.24	3.33
Ash (%)	8.22	7.60	7.06	7.67	7.13	7.24	7.78	7.16	7.89

¹⁾Supplied per kilogram of diet: vitamin A 1,600,000 IU; vitamin D₃ 300,000 IU; vitamin E 800 IU; vitamin K₃ 132 mg; vitamin B₁ 97 mg; vitamin B₂ 500 mg; vitamin B₆ 200 mg; vitamin B₁₂ 1.2 mg; nicotinic acid 2,000 mg; pantothenic acid 800 mg; folic acid 60 mg; choline chloride 35,000 mg; Mn 12,000 mg; Zn 9,000 mg; Fe 4,000 mg; Cu 500 mg; I 250 mg; Co 100 mg; Se 50 mg.

CON, control diet; 50 BSFLO, 50% black soldier fly larvae oil diet; 100 BSFLO, 100% black soldier fly larvae oil diet; ME, metabolizable energy.

and then homogenized with a homogenizer (T 25 digital ultra-turrax, IKA, Germany) at 10,000 ×g for 1 min. The pH values of the homogenates were determined using a pH meter (AM-7, Nihonseiki Kaisha, Tokyo, Japan). All determinations were performed in triplicate.

The leg meat samples were cooked in a polyethylene plastic bag and heated in a water bath operating at 80°C to an internal temperature of 75°C. After cooking and cooling, the samples were dried and weight to calculate the cooking loss (%). The samples were cut into 1.5 × 1.5 cm sized pieces. Shear force analysis was performed using a TA-HDi Texture Analyzer (Stable Macro System, London, UK). Each block was sheared at a constant speed of 1mm/s. The water holding capacity (WHC) was measured followed by the guide of Kauffman et al. [11]. The Samples were also analyzed for moisture, crude protein (CP), ether extract (EE), and ash [12].

Thiobarbituric acid-reactive substances (TBARS) and volatile basic nitrogen (VBN)

The thiobarbituric acid-reactive substances (TBARS) and volatile basic nitrogen (VBN) were also measured on d 0, 3, and 6 during the storage period as described by Witte et al. [13]. On each day,

10 g minced leg meat samples (5 replicates) were homogenized with 10 mL of trichloroacetic acid, and the sample volume was adjusted to 50 mL by adding distilled water. The homogenization was performed at 10,000 \times g for 5 min with a homogenizer (T 25 digital ultra-turrax, IKA, Germany). After centrifugation, the supernatant was filtered using a filter paper (Whatman No. 1) and 5 mL of the supernatant was added to 5 mL of 2-thiobarbituric acid (0.005 mM). The samples were then mixed and the mixture was heated. Next, the absorbance was measured at 532 nm using a UV/VIS spectrophotometer (OPTI-ZEN 2120UV, Mecasys, Daejeon, Korea). The result was expressed as mg malonaldehyde (MDA) equivalents/kg sample.

The VBN was analyzed according to Conway [14]. The 10 g of leg meat samples (5 replicates) was homogenized for 10 min with 90 mL of distilled water using homogenizer (T 25 digital ultra-turrax, IKA, Germany). The homogenate was centrifuged for 10 min at 800 \times g, and the supernatant was filtered using a filter paper (Whatman No. 1). Next, 1 mL of 0.01 N boric acid as a VBN absorber was placed in the inner section of a Conway micro diffusion cell. And then, 1 mL of filtrate and 1 mL of 50% potassium carbonate were added to the outer section of the Conway micro diffusion cell. The cell was incubated at 37°C for 120 min and titrated with 0.02N sulfuric acid. The blank test was conducted following the same process without adding 1 mL of 50% potassium carbonate.

Fatty acid profile of black soldier fly larvae oil and chicken breast meat

The lipid extraction was performed using chloroform:methanol (1:2) for breast meat samples (6 replicates). The samples were transmethylated using a methanolic solution of H₂SO₄ (4%) to determine fatty acid methyl esters (FAME). A biphasic separation was obtained by adding 0.5 mL of distilled water and 1.5 mL of N-Heptane to each sample. The FAME was determined by gas chromatography (Agilent 7890A series, Agilent Technologies, Wilmington, DE, USA), equipped with a Hewlett Packard HP-88 capillary column (60 m length, 0.52 mm internal diameter, and 0.20 μ m film thickness; J&W Scientific, USA) and a flame ionization detector. The carrier gas was helium and the detector temperature was 260°C with the split ration (30:1). The FAs were identified based on a standard FAME mixture (37-Component FAME Mix, Supelco, Bellefonte, PA, USA). The results were expressed as the percentage (%) of total detected FAME (Table 2).

Sensory analysis of chicken breast meat

Sensory evaluation was conducted by a panel consisting of 6 assessors at the National Institute of Animal Science. For the experiment, a total of 5 breasts per treatment were used and 3 days of analysis were scheduled (day 0, 3, and 6 during the storage period). The breast meats were removed from the refrigerator on each sampling day and used directly. After that, each sample was heated at 80°C for 8 min, until the core temperature reached 74°C. The samples were put on aluminum trays and served to the assessors. The descriptors were color, flavor, juiciness, firmness, and overall preference and the intensity of the sensory attributes was scored from 1 to 9, with 1 as low intensity and 9 as high intensity.

Statistical analysis

Data were analyzed using the PROC GLM of SAS (SAS Inst., Cary, NC, USA). The experimental unit was the pen for growth performance and the individual bird was used for slaughter traits, oxidative and meat quality parameters, and FA profile. Results are given as mean and standard error of the mean (SEM). Statistical significance and tendency were considered at $p < 0.05$ and $0.05 \leq p < 0.10$, respectively.

Table 2. Fatty acid profile (% of total FAME) of the black soldier fly larvae oil (BSFLO)

Fatty acids	BSFLO
C10:0	1.76
C12:0 (Lauric)	35.72
C14:0 (Myristic)	5.03
C16:0 (Palmitic)	13.78
C17:0	0.20
C18:0 (Stearic)	2.81
C16:1	2.12
C17:1	0.18
C18:1 n-9 (Oleic)	18.28
C22:2	0.18
C18:2 n-6 (Linoleic)	15.02
C18:3 n-3 (Linolenic)	1.95
C18:4 n-3	0.54
C20:3 n-3	0.32
C20:5 n-3 (EPA)	0.98

FAME, fatty acid methyl esters.

RESULTS AND DISCUSSION

In the present study, partial or total replacement of SBO with BSFLO for broiler chickens has no detrimental effect on growth performance. The inclusion of 100% BSFLO also did not lead to any negative effect on growth performance compared to 50% BSFLO inclusion. This result shows the possibility of total replacement of SBO by BSFLO. The IBW was the same in all groups and the FBW also did not show differences among dietary treatment groups (Table 3). The DFI, ADG, and FCR were not affected by the inclusion of BSFLO (Table 3). The mortality was not affected by the inclusion of BSFLO (not shown). The previous researches were also reported that the inclusion of BSFLO in a broiler diet did not affect the growth performance [6,7]. Through these results, it is possible to replace 100% of the SBO with the BSFLO in the broiler diet in terms of growth performance.

Similarly to the result of growth performance, the replacement of BSFLO did not affect the organ yields except the relative weight of the gizzard. The previous studies also have shown no significant differences in carcass traits by the replacement of BSFLO [6,7]. However, the inclusion

Table 3. Effect of the dietary black soldier fly larvae oil inclusion level on the growth performance of the broiler chickens¹⁾

Items	Dietary treatments			SEM	p-value
	CON	50 BSFLO	100 BSFLO		
IBW (g, d 1)	41.60	41.71	41.20	0.16	0.104
FBW (g, d 35)	1,739.82	1,730.88	1,743.50	43.92	0.978
DFI (g)	79.06	81.38	79.60	1.19	0.386
ADG (g)	49.71	49.45	49.81	1.25	0.978
FCR (g/g)	1.59	1.65	1.60	0.03	0.492

¹⁾Each value is the mean of 10 replicates (7 birds/group).

CON, control diet; 50 BSFLO, 50% black soldier fly larvae oil diet; 100 BSFLO, 100% black soldier fly larvae oil diet; SEM, standard error of the mean; IBW, initial body weight; FBW, final body weight; DFI, daily feed intake; ADG, average daily gain; FCR, feed conversion ratio.

(150 g/kg) of BSFLM for broiler chickens negatively affect the carcass weight [8]. Dabbou et al. [9] observed that the negative effects on growth performance and carcass trait by 15% inclusion level in the broiler diet were caused by reduced villus height: crypt depth. Interestingly, the relative gizzard weight was significantly higher ($p = 0.028$) in the control group than in other groups (Table 4). A similar result was reported that the weight of gizzard in a control group was heavier than that of other treatments where BSFLM was included in a broiler chicken diet [15]. Moreover, they compared several kinds of BSFLM which are differently defatted in terms of organ yields. The relative weight of the gizzard of broilers fed full-fat BSFLM was lower than those fed extruded BSFLM. However, the replacement of fish meal with a maggot in a broiler diet, as opposed to BSFLM and BSFLO, increased the weight of gizzard [16]. Overall, it seems that the inclusion of the BSFL by-product may affect the weight of the gizzard and further studies are required to better understand the mechanism.

The results of the physical and chemical traits of chicken leg meat were satisfactory and suggested that the use of the BSFLO in the broiler diet as an alternative of SBO. The inclusion of BSFLM in terms of carcass weight and dressing percentage had no adverse effect on chicken meat quality [17]. They also investigated the pH, color, and cooking loss of breast meat and the pH was decreased by the inclusion of BSFLM. The low pH value can cause a decrease of WHC and the increase of cooking loss when the value of pH is between 5.2 to 5.5 [17]. However, in the present study, the replacement of BSFLO did not show any adverse effects on the physical measurements of chicken leg meat in Table 5 and the pH value was in a normal range [18]. For this reason, the results of moisture, WHC, and cooking loss which are related to pH value had no significant differences among treatments. However, the more substitution level (24.8% of SBM and 100% of SBO) of BSFLM in a broiler diet increased the shear force and cooking loss compared to less substitution (16.1% of SBM and 28.4% of SBO) of BSFLM [17]. It seems that the high inclusion level of BSFLM may

Table 4. Effects of the dietary black soldier fly larvae oil inclusion level on the relative length (cm/kg) of the different sections of the digestive tract, relative weight (g/kg) of the digestive track and internal organs of broiler chickens at 35 d of age¹⁾

Items	Dietary treatments			SEM	p-value
	CON	50 BSFLO	100 BSFLO		
LW (kg)	1.99	1.98	2.08	0.04	0.229
Intestinal sections					
Duodenum length (cm/kg)	14.32	14.44	13.34	0.55	0.330
Jejunum length (cm/kg)	33.17	30.50	32.69	0.90	0.107
Ileum length (cm/kg)	33.56	30.00	32.09	1.14	0.111
Duodenum weight (g/kg)	6.63	6.26	6.27	0.30	0.628
Jejunum weight (g/kg)	10.91	10.41	10.41	0.45	0.665
Ileum weight (g/kg)	7.61	8.16	7.68	0.33	0.456
Internal organs					
Gizzard (g/kg)	14.85 ^a	12.52 ^b	13.02 ^b	0.59	0.028
Liver (g/kg)	21.67	19.94	21.19	0.85	0.349
Pancreas (g/kg)	2.28	2.20	2.31	0.13	0.849
Spleen (g/kg)	1.14	0.95	1.09	0.08	0.271
Bursa of Fabricius (g/kg)	1.80	1.66	1.42	0.29	0.655

¹⁾Each value is the mean of 11 replicates (11 birds/treatment).

^{a,b}Means with different letters within each variable differ ($p < 0.05$).

CON, control diet; 50 BSFLO, 50% black soldier fly larvae oil diet; 100 BSFLO, 100% black soldier fly larvae oil diet; SEM, standard error of the mean; LW, live weight.

Table 5. Physical measurements and chemical traits of chicken leg meat as affected by diets containing different levels of black soldier fly larvae oil (BSFLO)¹⁾

Parameters	Treatments			SEM	p-value
	CON	50 BSFLO	100 BSFLO		
pH	5.82	5.82	5.86	0.04	0.802
Shear force (kg/0.5 inch ²)	3.33	3.29	2.79	0.55	0.745
Cooking loss (%)	15.29	18.52	15.49	1.02	0.074
WHC (%)	61.66	61.14	59.64	0.77	0.196
Moisture (%)	75.47	76.11	75.83	0.38	0.516
Crude protein (%)	22.14	21.83	21.89	0.37	0.833
Ether extract (%)	1.36	1.18	1.17	0.19	0.754
Ash (%)	1.11	1.07	1.11	0.03	0.527

¹⁾Each value is the mean of 6 replicates (6 birds/treatment).

CON, control diet; 50 BSFLO, 50% black soldier fly larvae oil diet; 100 BSFLO, 100% black soldier fly larvae oil diet; SEM, standard error of the mean; WHC, water holding capacity.

affect the physical measurements of chicken breast meat. On the other hand, the total substitution of the BSFLO did not affect the shear force and cooking loss in the chicken leg meat compared to the partial substitution of the BSFLO in this study (Table 5). The chemical quality such as CP, EE, and ash of chicken leg meat was also not affected by treatment (Table 5). Several studies have also shown the same results that the total replacement of the SBO with BSFLO did not affect the chemical quality of chicken meat [6,8].

A major concern of using BSFLM and BSFLO is that the supplementation negatively affects the meat quality by a decrease of polyunsaturated fatty acid (PUFA), increase of saturated fatty acid (SFA) and monounsaturated fatty acid (MUFA) in the meat. The SFA is the main FA in BSF derived oil and lauric acid (C12:0), palmitic acid (C16:0), and oleic acid (C18:1) are predominant FAs [4]. Moreover, the BSFL contains a high level of lauric acid which has an antimicrobial effect on gut pathogens [19–21]. Even though the partial replacement of BSFLO has shown no negative effects on growth performance and gut health, the BSFLO has a defect that alters the FA profile of the meat [6–8]. As expected, in the present study, the replacement of the BSFLO altered the FA profile of the chicken breast meat (Table 6). The content of the SFA was increased (27.16%, 27.58%, and 28.72% in CON, 50 BSFLO, and 100 BSFLO, $p = 0.011$) by the addition of the BSFLO. However, contrary to the result of the SFA, the unsaturated fatty acid (UFA) was decreased (72.85%, 72.42%, and 71.29% in CON, 50 BSFLO, and 100 BSFLO, $p = 0.011$) by the total replacement of the BSFLO and the UFA/SFA was also significantly decreased (2.69, 2.63, and 2.48 in CON, 50 BSFLO, and 100 BSFLO, $p = 0.013$). The reason for the change of the FA profile is that the high level of SFA in the BSFLO influences the fraction of PUFA [22]. Schiavone et al. [6] reported that the MUFA was not affected by the BSFLO. On the contrary, in the present study, the fraction of MUFA was increased (43.36%, 44.58%, and 48.55% in CON, 50 BSFLO, and 100 BSFLO, $p < 0.001$) and this result stems from the desaturation and elongation of SFA such as C14:0, C16:0, and C18:0. The inclusion of BSFLM in a broiler and laying quails also changed the FA profile in the breast meat and eggs [8,23]. They commonly reported that the concentration of SFA and MUFA was increased and PUFA was decreased. The one thing that should be improved is the negative change of the FA composition of meat when the BLSFM and BSFLO were included. Even though 100 BSFLO showed increased UFA/SFA, there was no significant difference between CON and 50 BSFLO (Table 6). This result means that it seems appropriate to replace with the BSFLO less than 50% in terms of the meat quality.

Table 6. Fatty acid profile (% of total FAME) of chicken breast meat as affected by diets containing different levels of black soldier fly larvae oil (BSFLO)¹⁾

Parameters	Treatments			SEM	p-value
	CON	50 BSFLO	100 BSFLO		
C14:0 (Myristic)	0.55 ^c	1.07 ^b	1.74 ^a	0.08	<.001
C16:0 (Palmitic)	20.35 ^b	21.06 ^{ab}	21.90 ^a	0.28	0.005
C18:0 (Stearic)	6.26 ^a	5.45 ^b	5.08 ^b	0.22	0.007
SFA	27.16 ^b	27.58 ^b	28.72 ^a	0.32	0.011
C16:1 n-7 (Palmitoleic)	3.82 ^b	3.59 ^b	4.78 ^a	0.23	0.006
C18:1 n-9 (Oleic)	39.13 ^c	40.61 ^b	43.32 ^a	0.45	<.001
C20:1 n-9 (Eicosenoic)	0.41 ^b	0.38 ^b	0.45 ^a	0.01	0.003
MUFA	43.36 ^b	44.58 ^b	48.55 ^a	0.54	<.001
C18:2 n-6 (Linoleic)	26.88 ^a	25.48 ^a	20.90 ^b	0.49	<.001
C18:3 n-6 (γ-Linolenic)	0.24 ^a	0.19 ^b	0.17 ^b	0.01	0.005
C18:3 n-3 (Linolenic)	1.78 ^a	1.68 ^a	1.20 ^b	0.04	<.001
C20:4 n-6 (Arachidonic)	0.59	0.49	0.47	0.04	0.176
PUFA	29.49 ^a	27.84 ^b	22.74 ^c	0.53	<.001
UFA	72.85 ^a	72.42 ^a	71.29 ^b	0.32	0.011
UFA/SFA	2.69 ^a	2.63 ^a	2.48 ^b	0.04	0.013

¹⁾Each value is the mean of 6 replicates (6 birds/treatment).

^{a-c}Means with different letters within each variable differ ($p < 0.05$).

FAME, fatty acid methyl esters; CON, control diet; 50 BSFLO, 50% black soldier fly larvae oil diet; 100 BSFLO, 100% black soldier fly larvae oil diet; SEM, standard error of the mean; SFA, sum of saturated fatty acid; MUFA, sum of monounsaturated fatty acid; PUFA, sum of polyunsaturated fatty acid; UFA, sum of unsaturated fatty acid.

Table 7. Effects of the dietary black soldier fly larvae oil inclusion level on the thiobarbituric acid-reactive substances (TBARS) and volatile basic nitrogen (VBN) of chicken leg meat¹⁾

Items	Storage (d)			SEM	p-value
	0	3	6		
TBARS (mg MDA/kg)					
CON	0.24 ^{bx}	0.29 ^b	0.48 ^a	0.03	<.001
50 BSFLO	0.13 ^{cy}	0.35 ^b	0.54 ^a	0.04	<.001
100 BSFLO	0.12 ^{cy}	0.36 ^b	0.61 ^a	0.04	<.001
SEM	0.01	0.02	0.06	-	-
p-value	<.001	0.058	0.434	-	-
VBN (mg %)					
CON	9.53 ^c	10.18 ^b	10.85 ^a	0.17	<.001
50 BSFLO	9.33 ^c	10.15 ^b	10.93 ^a	0.23	0.001
100 BSFLO	9.42 ^b	9.94 ^b	10.86 ^a	0.25	0.006
SEM	0.14	0.28	0.22	-	-
p-value	0.629	0.819	0.959	-	-

¹⁾Each value is the mean of 5 replicates (5 birds/treatment).

^{a-c}Different letters within same row differ significantly ($p < 0.01$).

^{x-y}Different letters within same column differ significantly ($p < 0.01$).

SEM, standard error of the mean; CON, control diet; 50 BSFLO, 50% black soldier fly larvae oil diet; 100 BSFLO, 100% black soldier fly larvae oil diet.

The substitution of the BSFLO altered the oxidative status of the chicken leg meat resulting in a decrease of the TBARS value and this result seems to be affected by the decreased proportion of the PUFA. The TBARS and VBN values of all treatments were increased ($p < 0.01$) with longer storage periods (Table 7). However, the TBARS value of the control group was significantly higher ($p < 0.001$) than in other groups on d 0 (Table 7). Even though the FA profile of the leg meat was only measured in the present study, the previous studies reported that the replacement of BSFLO in the broiler diet changed the FA profile of both breast meat and leg meat and the composition of SFA was also increased [6,22]. Although the high composition of the PUFA is more valuable for the chicken meat quality, the SFA is less vulnerable to lipid peroxidation than the USFA and TBARS inhibition of SFA was greater than that of USFA [24,25]. On the contrary to our result, Cullere et al. [22] observed that the result of TBARS did not show a significant difference by the inclusion of BSFLO in breast meat.

The sensory traits of the meat are an important point of view to evaluate the BSFLO and the sensory profiles of the breast meat derived from the chicken fed the BSFLO showed the meaningful result in the aspect of using the BSFLO as an alternative of the SBO. The breast meat of

Table 8. Effects of the dietary black soldier fly larvae oil inclusion level on the sensory evaluation of chicken breast meat¹⁾

Items	Storage (d)		
	0	3	6
Color			
CON	4.77	4.22	4.44
50 BSFLO	4.33	4.22	4.72
100 BSFLO	4.44	4.27	4.22
SEM	0.27	0.26	0.25
Flavor			
CON	4.88	4.27	4.44
50 BSFLO	4.83	4.66	4.50
100 BSFLO	4.88	4.38	4.50
SEM	0.25	0.17	0.33
Juiciness			
CON	4.16	4.22	3.88
50 BSFLO	3.93	4.33	4.22
100 BSFLO	4.46	4.11	4.44
SEM	0.18	0.15	0.17
Firmness			
CON	5.16	4.83	5.05
50 BSFLO	4.44	5.00	5.00
100 BSFLO	5.16	4.66	4.94
SEM	0.26	0.46	0.46
Overall preference			
CON	5.00	4.69	4.44
50 BSFLO	4.44	4.63	4.50
100 BSFLO	5.11	4.44	4.61
SEM	0.25	0.26	0.30

¹⁾Each value is the mean of 5 replicates (5 birds/treatment).

CON, control diet; 50 BSFLO, 50% black soldier fly larvae oil diet; 100 BSFLO, 100% black soldier fly larvae oil diet; SEM, standard error of the mean.

chickens fed the BSFLO showed similar sensory profiles compared to those fed CON (Table 8). The sensory traits of the chicken meat when the insect meal was included in a diet had also no detrimental effect on sensory analysis of eggs from layers fed BSFLM and of fish fed insect meal [5,22].

In conclusion, the present study showed that the partial or total replacement of SBO in the broiler chicken diets by the BSFLO showed comparable results on growth performance, meat quality, and sensory properties to the CON and the BSFLO could be a promising substitute as a dietary fat source in the broiler diet. However, further researches are necessary to improve the drawback that lowering the proportion of the PUFA by improving the FA profiles in the BSFLO as modulating their feed and defatting procedure.

REFERENCES

1. Dei HK. Soybean as a feed ingredient for livestock and poultry. In: Krezhova D, editor. Recent trends enhancing diversity quality of soybean products. Rijeka: IntechOpen; 2011.
2. Sheppard DC, Newton GL, Thompson SA, Savage S. A value added manure management system using the black soldier fly. *Bioresour Technol.* 1994;50:275-9.
3. Newton L, Sheppard C, Watson DW, Burtle G, Dove R. Using the black soldier fly, *Hermetia illucens*, as a value-added tool for the management of swine manure. Raleigh, NC: North Carolina State University; 2005. p. 1-17.
4. Surendra KC, Olivier R, Tomberlin JK, Jha R, Khanal SK. Bioconversion of organic wastes into biodiesel and animal feed via insect farming. *Renew Energy.* 2016;98:197-202.
5. Lock ER, Arsiwalla T, Waagbø R. Insect larvae meal as an alternative source of nutrients in the diet of Atlantic salmon (*Salmo salar*) postsmolt. *Aquac Nutr.* 2016;22:1202-13.
6. Schiavone A, Cullere M, De Marco M, Meneguz M, Biasato I, Bergagna S, et al. Partial or total replacement of soybean oil by black soldier fly larvae (*Hermetia illucens* L.) fat in broiler diets: effect on growth performances, feed-choice, blood traits, carcass characteristics and meat quality. *Ital J Anim Sci.* 2017;16:93-100.
7. Schiavone A, Dabbou S, De Marco M, Cullere M, Biasato I, Biasibetti E, et al. Black soldier fly larva fat inclusion in finisher broiler chicken diet as an alternative fat source. *Animal.* 2018;12:2032-39.
8. Schiavone A, Dabbou S, Petracci M, Zampiga M, Sirri F, Biasato I, et al. Black soldier fly defatted meal as a dietary protein source for broiler chickens: effects on carcass traits, breast meat quality and safety. *Animal.* 2019;13:2397-405.
9. Dabbou S, Gai F, Biasato I, Capucchio MT, Biasibetti E, Dezzutto D, et al. Black soldier fly defatted meal as a dietary protein source for broiler chickens: effects on growth performance, blood traits, gut morphology and histological features. *J Anim Sci Biotechnol.* 2018;9:49.
10. NRC [National Research Council]. Nutrient requirements of poultry. 9th ed. Washington, DC: National Academies Press; 1994.
11. Kauffman RG, Eikelenboom G, van der Wal PG, Engel B, Zaar M. A comparison of methods to estimate water-holding capacity in post-rigor porcine muscle. *Meat Sci.* 1986;18:307-22.
12. AOAC [Association of Official Analytical Chemists] International. Official methods of analysis of AOAC International. 16th ed. Arlington, VA: AOAC International; 1995.
13. Witte VC, Krause GF, Bailey ME. A new extraction method for determining 2-thiobarbituric acid values of pork and beef during storage. *J Food Sci.* 1970;35:582-5.
14. Conway EJ. Microdiffusion analysis and volumetric error. 3rd ed. London: Crosby Lockwood and Son; 1950.
15. Calderon A, Coles S, Davis D, Lanier J, Oliveira A. How to hear the shape of a billiard table

- [Internet]. 2018. <http://arxiv.org/abs/1806.09644>. Accessed 27 Dec 2019.
16. Okah U, Onwujiariri EB. Performance of finisher broiler chickens fed maggot meal as a replacement for fish meal. *J Agric Technol*. 2012;8:471-7.
 17. Cullere M, Tasoniero G, Giaccone V, Miotti-Scapin R, Claeys E, De Smet S, et al. Black soldier fly as dietary protein source for broiler quails: apparent digestibility, excreta microbial load, feed choice, performance, carcass and meat traits. *Animal*. 2016;10:1923-30.
 18. Tavaniello S, Maiorano G, Siwek M, Knaga S, Witkowski A, Di Memmo D, et al. Growth performance, meat quality traits, and genetic mapping of quantitative trait loci in 3 generations of Japanese quail populations (*Coturnix japonica*). *Poult Sci*. 2014;93:2129-40.
 19. Zeitz JO, Fennhoff J, Kluge H, Stangl GI, Eder K. Effects of dietary fats rich in lauric and myristic acid on performance, intestinal morphology, gut microbes, and meat quality in broilers. *Poult Sci*. 2015;94:2404-13.
 20. Kim SA, Rhee MS. Highly enhanced bactericidal effects of medium chain fatty acids (caprylic, capric, and lauric acid) combined with edible plant essential oils (carvacrol, eugenol, β -resorcylic acid, trans-cinnamaldehyde, thymol, and vanillin) against *Escherichia coli* O157: H7. *Food Control*. 2016;60:447-54.
 21. Belghit I, Waagbø R, Lock EJ, Liland NS. Insect-based diets high in lauric acid reduce liver lipids in freshwater Atlantic salmon. *Aquac Nutr*. 2019;25:343-57.
 22. Cullere M, Schiavone A, Dabbou S, Gasco L, Zotte AD. Meat quality and sensory traits of finisher broiler chickens fed with black soldier fly (*Hermetia illucens* L.) larvae fat as alternative fat source. *Animals*. 2019;9:140.
 23. Zotte AD, Singh Y, Michiels J, Cullere M. Black soldier fly (*Hermetia illucens*) as dietary source for laying quails: live performance, and egg physico-chemical quality, sensory profile and storage stability. *Animals*. 2019;9:115.
 24. Rael LT, Thomas GW, Craun ML, Curtis CG, Bar-Or R, Bar-Or D. Lipid peroxidation and the thiobarbituric acid assay: standardization of the assay when using saturated and unsaturated fatty acids. *J Biochem Mol Biol*. 2004;37:749-52.
 25. Øverland M, Borge GI, Vogt G, Schøyen HF, Skrede A. Oxidative stability and sensory quality of meat from broiler chickens fed a bacterial meal produced on natural gas. *Poult Sci*. 2011;90:201-10.