



Potential Value of Using Insect Meal As an Alternative Protein Source for Japanese Quail Diet

■ Author(s)

Hatab MH¹  <https://orcid.org/0000-0001-8418-3841>
Ibrahim NS¹  <https://orcid.org/0000-0003-4907-2905>
Sayed WA¹  <https://orcid.org/0000-0003-1933-4994>
Sabic EM¹  <https://orcid.org/0000-0003-4945-9982>

¹ Biological Applications Department, Nuclear Research Center, Atomic Energy Authority, Inshas, Egypt.

■ Mail Address

Corresponding author e-mail address
Nashat Saeid Ibrahim
Biological Applic. Dep., Nuclear Res. Center,
Atomic Energy Authority, Inshas, Egypt,
P.O, Box 13759.
Phone: (0202) 01119992240
Email: nashaat1977@yahoo.com

■ Keywords

Insect meal, quail chicks, performance, hematological, biochemical indices.



Submitted: 02/January/2018
Approved: 10/November/2019

ABSTRACT

A feeding trial was conducted to determine the potential value of replacing 100 % of meat and bone meal (MBM) ingredient as the major animal protein source in Japanese quail diets by 50% and 100% of insect meal derived from *S. littoralis* Larvae. A total of 360, 7 days-old quail chicks, were randomly divided into three equal groups (120 chicks) with three replicates each. They were offered three different diets 1, 2 and 3, respectively. The Diet 1 contained MBM only as a major source of animal protein, diet 2 had both MBM (50%) and insect meal (50%) while diet 3 contained insect meal (100%). Results: Partial or total replacement of *S. littoralis* larvae with MBM increased body weight gain and improved feed conversion ratio ($p \leq 0.05$). Moreover, treated groups with insect meal were economically much more profitable than conventional MBM. On the other hand, serum total protein, albumin, globulin, triglycerides, cholesterol, total antioxidant, thyroxin, estradiol-17B, and testosterone hormones level were significantly higher ($p \leq 0.05$) in treated groups than in the control one. While, serum alkaline phosphates, alanine aminotransferase and aspartate aminotransferase activities in all treated groups were within equal range and did not differ among them. The inclusion of insect meal protein as a replacement for MBM up to 100% replacement has no adverse effect on growth performance, carcass characteristics, hematological and serum biochemical indices of growing Japanese quail chicks. Moreover, it can be used as one of a protein source for lowering the production costs of poultry diets.

INTRODUCTION

Protein is a very important nutrient source of poultry diets and is useful in maintaining and repairing tissues for organisms to enable proper growth and development (Sleman *et al.*, 2015). The high cost of animal protein meal and their involvement in the transmission of diseases has added additional constraint to poultry production worldwide (Womani *et al.*, 2012). It is, therefore, necessary to search for safe, locally available and low cost feedstuffs to replace protein sources. Insects have been addressed as a possible alternative feed for poultry, because of their rich nutrient content and extremely low environmental impact (Bovera *et al.*, 2016). The FAO strongly recommends the inclusion of insect protein in poultry diets to lower the cost of feeds and improved growth and production parameters (FAO, 2014). Preimaginal stages of *Spodoptera littoralis* was successfully used as a replacement for fish meal in rainbow trout diets (Danieli *et al.*, 2011). In this perspective, cotton leafworm *Spodoptera littoralis* is regarded as one of the most important agricultural pest in the Middle East. It is a very harmful potential pest to many crops including cotton, alfalfa, peanut and potato (Mohamed,



2003). It can convert the low-nutritive waste products as feed into valuable nutrients very efficiently, making *S. littoralis* meal a promising safe, healthy and economic source of alternative protein. Larvae are rich in nutrients, depending on the rearing substrate and the quantity of fat and protein that they can store in their body during the larval stage (Makkar *et al.*, 2014). According to Basiouny *et al.* (2016) *S. littoralis* meal is known to have high nutritional value and may be an important source of protein, carbohydrate, fat, vitamin, minerals, etc. Until now, literature data about the potential application of *Spodoptera littoralis* larvae as a poultry feed ingredient are scarce. Therefore, the present research studied for the first time the potential application of insect meal protein derived from *S. littoralis* larvae, in Japanese quail diets. The effects on productive performance (growth performance and carcass characteristics), animal welfare (evaluated by the blood traits) and economic cost were considered in this study.

MATERIALS AND METHODS

Animal ethics

All of the experiments were carried out according to the guidelines for animal experiments at the National Institute of Animal Health.

Rearing of *Spodoptera littoralis* larvae and preparation of insect meal

The experiments were conducted on cotton leafworm *S. littoralis* taken from the insect farm of the Department of Biological Application, Nuclear Research Center, Egypt. The larvae were reared on castor bean *Ricinus communis* leaves. The rearing conditions were 25 ± 2 °C, $60 \pm 5\%$ relative humidity (RH), with a photoperiod of as 14 L: 10 D hours. The last instar larvae were collected and ground, then freeze dried until using to formulate diets along with other ingredients, as insect meal.

Chemical analysis

MBM were purchased from Arabian Milling and Food Industries co. Egypt. Both MBM and insect meal were analyzed for chemical composition Table (1). Crude protein, crude fat, crude fiber and ash contents of MBM and *S. littoralis* were analysed according to AOAC methods (2002). The amino acid profile of MBM and *S. littoralis* full fat meal were analysed by High-Performance Liquid Chromatography (HPLC) following the method by Alikwe *et al.* (2010). Carbohydrate

content was estimated following Ammar *et al.* (2013) method. Gross caloric content was determined by using Oxygen Bomb Calorimeter (Instrumentation India Co.). Calcium content was determined by Atomic absorption Spectrophotometry (Varian Tectron AA575 series). Inorganic phosphorus was determined colorimetrically using the commercial kit produced by Stanbio Company, USA by computerized spectrophotometer model Milton Roy 1201.

Table 1 – Chemical analysis of MBM¹ and Insect meal² used in this study.

Composition (g/100g. dry matter)	MBM ¹	Insect meal ²
Crude protein	50.4	54
Crude fiber	2.8	4.4
Carbohydrate	2	4
Crude fat	8.47	9.3
Ash	31.6	15.8
Calcium	10.3	6
Total phosphorus	5.1	1.5
Essential amino acids (% of crude protein)		
Arginine	7.4	6.6
Histidine	3.96	3.98
Isoleucine	3.54	3.96
Leucine	6.28	6.22
Lysine	6.61	9.22
Methionine	3.69	3.34
Phenyl-alanine	3.81	5.57
Threonine	3.7	4
Valine	7.3	5.9
Total essential amino acids (% of crude protein)	46.3	48.8
Non-essential amino acids (% of crude protein)		
Aspartic acid	6.2	6.2
Cystine	3.69	3.42
Glutamic acid	11.75	10.2
Proline	3.55	3.58
Glycine	7.65	3.7
Serine	9.2	10.6
Alanine	6.27	6.9
Tyrosine	5.4	6.5
Total non-essential amino acids (% of crude protein)	53.7	51.2
Gross caloric content (kcal/kg)	2150	3400

MBM¹: Meat and bone meal Insect meal²: *Spodoptera littoralis*. larva meal

Birds and husbandry

The feeding trial was conducted on Japanese quail chicks maintained at the Poultry experimental farm of the biological Application Department, Nuclear Research Center (Egypt). The poultry farm was 10 m wide X 40 m long X 5 m high, equipped with waterproof floor and walls, covered completely by tiles and with an automatic ventilation system. A total of 360 Japanese 7 day-old quail chicks weighing 20 g on average were randomly allotted to three dietary



treatments (120 chicks in each group) each consisting of three pens as replicates with 20 female and 20 male per pen. The experimental period lasted for 6 weeks. Each pen was 1.0 m wide X 1.2 m long and was equipped with a feeder and an automatic drinker. All groups were farmed in electrically heat-controlled batteries: in the first week, the temperature was 35 °C and then the quails were kept at 28±2 °C until the end of the experiment. Relative humidity was 50±5% and photoperiod was 14 L: 10 D hours. Mash feeds and water were available *ad libitum* throughout the experimental period. At hatching, the chicks received vaccination against Newcastle disease.

Diets

The trial was carried out to evaluate the effects of a partial or total replacement of MBM with insect meal protein on Japanese quail diets using two levels of inclusion. A basal diet based on corn meal, soybean meal and MBM (Table 2) was formulated and served as control group (T1) and the 50 and 100% replacement of MBM with insect meal formed the two treatment groups (T2 and T3 group, respectively). All diets were formulated to meet the nutrient requirements of Japanese quail chicks (NRC,1994). Feed and water were provided *ad libitum* throughout the trial. The calculated chemical composition of the basal and experimental diets are given in Table (2).

Growth performances and feed cost

Clinical signs and mortality were monitored daily during the whole experimental period. Body weight gain (BWG) and feed consumption were recorded weekly during the experiment period, feed intake (FI), daily weight gain (DWG) and feed conversion ratio (FCR) were determined for the overall experimental period (1– 42 days). All measurements were performed on the pen basis using a high precision electronic scale. The processing cost of insect meal in this study (at, 2016) was 2.5 Egyptian pound (LE)/ kg, while MBM was purchased at 5 LE/ kg. The cost of feed was based on the current market prices.

Carcass traits and blood analysis

At the end of the experimental period (6 weeks), 18 quail birds (6 birds/pen) from each feeding group (chosen on the basis of pen average final body weight) (3 female and 3 male) per pen were weighed and slaughtered for carcass analysis. Carcass, liver, heart, proventriculus, spleen, and bursa of Fabricius for each slaughtered bird were calculated as a relative percentage of live body weight. Blood samples were collected from slaughtered birds and placed in two tubes, one with lithium heparin to determine hematological parameters and the other without anticoagulant and left to clot then centrifuged at (4000 rpm) for 10

Table 2 – Ingredients and composition of experimental diets.

Ingredients (g/kg) as-fed	Experimental diets		
	T1 Control	T2 50% insect meal	T3 100% insect meal
Yellow corn	530	530	530
Soybean meal	280	280	280
Meat and bone meal	100	50	-
Insect meal	-	50	100
Vegetable oil	35	35	35
DL-methionine	1.2	1.2	1.2
Choline chloride	1.8	1.8	1.8
L-Lysine	3	3	3
Dicalcium phosphate	16	16	16
Calcium carbonate	27	27	27
Sodium chloride	3	3	3
Vitamin and min. premix*	3	3	3
Calculated composition (g/kg) as-fed except for Metabolizable Energy			
Metabolizable Energy(Kcal /kg)	2872	2883	2894
Analysed composition(g/kg)			
Dry matter	898	895	890
Crude protein	232	232	233
Crude fat	85.5	86	87
Crude fibre	67	60	58
Ash	60	64	70

*Vitamin and mineral premix (contained per kgm):- vit A, 1200 IU; vit D 1100 IU; vit E, 12mg; vitB12, 0.02mg; vit B1, 1mg; choline chloride, 0.16 mg; copper, 3mg; iron, 30 mg; manganese, 40mg; zinc, 45 mg; and selenium, 3mg.



minutes, and the resulting serum was stocked at -20 °C for hormonal and chemical analyses.

Serum total proteins, albumin, alkaline phosphates (ALP), alanine aminotransferase (ALT), aspartate aminotransferase (AST), total antioxidant, triglyceride, and total cholesterol were determined colorimetrically using commercial kits produced by Stanbio Company, USA by computerized spectrophotometer model Milton Roy 1201. Serum Globulin values were calculated by subtracting albumin values from their corresponding total proteins values of the same sample. Concerning, blood hematological parameters, red blood cell (RBCs) and white blood cell (WBCs) counts were determined using a hemocytometer according to Natt & Herrick (1952). Hemoglobin concentration (Hb) and packed cell volume (PCV %) were determined according to Dacie & Lewis (1991). Thyroxin (T4), Estradiol-17_β (E2) and Testosterone hormones were determined using radioimmunoassay (RIA) Commercial Kit produced by IZOTOP Company (INSTITUTE OF ISOTOPES Ltd.) Hungarian Company (<http://www.izotop.hu>), and samples were counted on Packard Gamma Counter.

Statistical analysis

One way analysis of variance was done using SAS (2002) software version 9.1.3, following the General Linear Model procedure with dietary treatment as fixed effect. Mean values assessed for significance using (Duncan, 1955) multiple range tests. The experimental unit was the pen. The model used was:

$$Y_{ij} = \mu + T_i + e_{ij}$$

Where: Y_{ij} = any value from the overall population.
 μ = the overall mean.

T_i = the effect of the i^{th} treatment (1 = 0 % of insect meal, 2 = 50 % of insect meal and 3 = 100 % of insect meal replacement). e_{ij} = the random error associated with the j^{th} individual.

RESULTS AND DISCUSSION

Nutritional potential value of *Spodoptera littoralis* larvae meal as feed

The nutrient composition of *S. littoralis* meal in comparison to MBM is presented in Table (1). Data showed that, *S. littoralis* meal numerically has higher levels of crude protein, crude fat, crude fiber, carbohydrate and gross caloric content than MBM. Furthermore, *S. littoralis* meal has higher total essential amino acid (in % of crude protein) than MBM. Lysine, phenyl-alanine, threonine and isoleucine were found to have higher levels in *S. littoralis* meal than MBM. The

methionine, arginine and valine levels were slightly lower than in MBM. Similarly, some insect species provide high amino acids concentrations, for example the caterpillars of *Saturniidae*, have a lysine content higher than 100 mg/100g of CP. (Bukkens, 2005). Elwert *et al.* (2010) compared *Hermetia illucens* meal to fishmeal and found higher levels of threonine, valine, isoleucine, leucine and histidine in the insect meal relative to fishmeal. Calcium and phosphorus concentration of the *S. littoralis* meal are lower than in MBM. Previous studies on the chemical composition of variety of insects showed their high nutritive value. For instance, high crude protein content ranged from (40 to 70%) of dry matter, lipid content (9 to 25% on a dry basis), highest unsaturated to saturated fatty acid ratio (5.8 - 8.2%) crude fiber (0.93-11%), (Al-Qazzaz *et al.*, 2016; Jozefiak *et al.*, 2016). The actual nutrient content of insects depends highly on the given diet, the life stage and the rearing conditions (Makkar *et al.*, 2014). Because of this, different compositions of insect meal are found in different studies. The present study is the first one testing the *S. littoralis* larvae meal as a partial or total replacement of MBM in the diet for growing quails. The first study which assessed the nutritional value of *S. littoralis* larval meal for growing quails reported that the *S. littoralis* larvae meal can be considered a valuable source of energy, amino acids and fat thus being a potential feed ingredient for poultry diets in the near future.

Dietary potential value of *Spodoptera littoralis* larvae meal on productive performance

Productive performance and feed cost data of growing Japanese quail chicks were summarized in Table (3). Partial or total replacement of insect meal derived from *S. littoralis* larva with MBM increased body weight gain and improved feed conversion ratio ($p \leq 0.05$). Chicks fed total replacement of insect meal with MBM had the best feed conversion ratio and greater BW gain, but lower feed intake than birds fed the MBM diet ($p \leq 0.05$). Moreover, the chicks remained healthy (absence of clinical signs) throughout the study and no mortality occurred during the experimental period. This response could be due to the nutritional content of insect meal of high biological value and may positively change body weight or other traits without adverse consequences of the growth parameters and chick's health and therefore beneficial. Most of the experiments published to date have been carried out with broiler chickens fed *Hermetia illucens* larvae



Table 3 – Live body weight, feed intake (g/bird), feed performance and feed cost as affected by insect meal replacement in Japanese quails chicks (Mean \pm SE).

Live Body Weight (g)	Experimental groups		
	Control	50% insect meal	100% insect meal
Initial	21.9 \pm 0.5	21.6 \pm 0.4	21.9 \pm 0.5
1 st week	34.6 \pm 1.3	35.8 \pm 1.2	35.9 \pm 1.3
2 nd week	61.5 \pm 1.9 ^c	65.2 \pm 1.6 ^b	69.5 \pm 1.7 ^a
3 rd week	90.2 \pm 1.9 ^c	96.1 \pm 1.7 ^b	100 \pm 1.8 ^a
4 th week	135.2 \pm 1.6 ^b	140.2 \pm 1.7 ^b	144.7 \pm 1.7 ^a
5 th week	165.5 \pm 1.8 ^c	170.3 \pm 1.7 ^b	177.5 \pm 1.5 ^a
Final (6 th week)	185.6 \pm 1.4 ^c	192.5 \pm 1.5 ^b	197.6 \pm 1.5 ^a
Feed Intake (g/bird/day)			
1 st week	5.7 \pm 0.5 ^a	5.5 \pm 0.4 ^{ab}	5.0 \pm 0.5 ^c
2 nd week	14.1 \pm 1.6 ^a	13.7 \pm 1.2 ^b	13.1 \pm 1.2 ^c
3 rd week	22.4 \pm 1.9 ^a	21.2 \pm 1.6 ^b	20.2 \pm 1.7 ^c
4 th week	29.8 \pm 1.9 ^a	28.5 \pm 1.6 ^b	27.3 \pm 1.3 ^c
5 th week	35.7 \pm 1.7 ^a	34.1 \pm 1.6 ^b	31.1 \pm 1.5 ^c
6 th week	39.9 \pm 3.7 ^a	37.5 \pm 2.5 ^b	33.8 \pm 3.3 ^c
Feed Performance			
Daily weight gain (g)	15.5 \pm 1.5 ^c	16.2 \pm 1.4 ^b	16.7 \pm 0.85 ^a
Daily Feed intake (g/bird)	24.6 \pm 1.2 ^a	23.4 \pm 1.3 ^b	21.7 \pm 0.95 ^c
Feed consumption(g/bird) for 42 days	1033.2 \pm 2.3 ^a	983.5 \pm 2.3 ^b	913.5 \pm 2.3 ^c
Feed conversion ratio(feed: gain)	1.6 \pm 0.1 ^a	1.4 \pm 0.09 ^b	1.3 \pm 0.09 ^c
Feed Cost			
Cost of diet / kg of feed, LE ¹	3	2.875	2.75
Cost of MBM ² / kg of feed, LE ¹	0.5	0.25	0
Cost of insect meal / kg of feed, LE ¹	0	0.125	0.25
Cost of diet /Feed consumed (g) /bird, LE ¹	3.1	2.83	2.51

^{a,b,c} means with different superscripts within the same row are significantly different at ($p < 0.05$).

LE¹: Egyptian pound MBM²: meat and bone meal

meal and were also tested as an alternative protein source to soybean meal and revealed positive results in terms of productive performance (body weight gain, feed intake, feed conversion ratio and mortality rate) Schiavone *et al.* (2017). Al-Qazzaz *et al.* (2016) showed the highest feed conversion ratio of Laying quails with inclusion of maggot larvae meal up to 15% in feed. Widjastuti *et al.* (2014) showed that the substitution of fish meal by maggot meal up to 5% level can support a good result on quail production performance: better feed conversion ratio, reduced feed consumption. Also, the feeding of black soldier fly larvae as a substitute for soybean meal resulted in a similar weight gain but a lower feed intake as compared to the control indicating an improved feed conversion, Makkar *et al.* (2014). Additional experiments by Schiavone *et al.* (2014) showed that mealworm meal can be included at maximum dietary concentrations of 25% without causing growth depression.

Oyogoke *et al.* (2006) showed that the performance and growth rate of broiler chicks fed entirely or partly with insect meal derived from caterpillars (50% replacement of fishmeal) did not differ significantly from those fed with conventional fish meal. Ijaiya

and Eko (2009) gradually substituted fishmeal with silkworm, caterpillar meal in broiler chicken feed without negative effects on feed intake, weight and feed efficiency.

Economic assessment of using insect meal as feed is presented in Table (3) and compared to the traditional diet of the control. Numerically, the cost of harvesting and processing one kg of insect meal is 50% cheaper than that of 1 kg of MBM. Economically, in this study the cost of diet 2 and diet 3 had the lowest cost of production (2.87 and 2.75 LE/ kg of feed) respectively, as compared to the cost of the control diet 1 (3 LE/ kg of feed). Moreover, treated groups showed lowest cost of 1kg live weight (2.83 and 2.51 LE/ kg of feed/ bird) as compared to the control group (3.1 LE/ kg of feed consumed/ bird), and consequently, the economic benefit in MBM ingredient replacement in Japanese quail diets (by 50% and 100%) with insect meal derived from *S. littoralis* larvae, in treated groups were higher by (9 and 39 %) respectively, rather than the control group. Therefore, the groups treated with insect meal were economically much more profitable than the ones treated with conventional MBM. The results relating to cost of production in this study are



also in line with those of Adeniji (2007), who narrated that insect meal based diets were cheaper than MBM. Also, Bombata & Balogun (1997) reported that one kg of insect meal derived from maggot was 20% cheaper to harvest and process than 1 kg of fish meal. Economic evaluation of insect meal as protein sources for quail chicks indicated that cost and profit indices of these protein sources were better than for MBM-based diets.

Dietary potential value of *Spodoptera littoralis* larvae meal on carcass traits and relative organ weights

The results of relative weight of carcass, liver, spleen and bursa of Fabricius Table (4) were significantly ($P \leq 0.05$) affected by a partial/total replacement of MBM with *S. littoralis* larvae meal. These findings are in agreement with Bovera, *et al.* (2016) who showed

the highest weight of carcass and lymphoid organs in broilers fed *Tenebrio molitor* larvae meal as a protein source. In addition, the highest weight of lymphoid organs such as spleen or bursa of Fabricius in growing quail chicks fed *S. littoralis* larvae meal as a protein source can be attributed to an increase in the activity of the immune system. In this regard, van Huis (2013) observed that feeding insects to chickens may decrease the use of antibiotics in the poultry industry. The significant increase in the relative weight of carcass, liver, spleen and bursa of Fabricius in the present study could be due to the *S. littoralis* larvae meal having a higher content of protein, essential amino acid (Lysine, phenyl-alanine, threonine and isoleucine), fat and energy than MBM. Based on these considerations, the inclusion of *S. littoralis* larvae meal in quail diets is effective in promoting growth and improving feed conversion ratio.

Table 4 – Effects of insect meal replacement on relative weight of carcass and some organs of quail chicks (Mean \pm SE).

Relative weight of carcass and some organs (%)	Experimental groups		
	Control	50% insect meal	100% insect meal
Carcass	66.2 \pm 0.6 ^c	68.4 \pm 0.6 ^b	69.3 \pm 0.8 ^a
Liver	2.51 \pm 0.1 ^c	2.76 \pm 0.01 ^b	2.86 \pm 0.04 ^a
Heart	0.76 \pm 0.01	0.78 \pm 0.05	0.79 \pm 0.03
Proventriculus	0.65 \pm 0.03	0.67 \pm 0.03	0.68 \pm 0.05
gizzard	2.48 \pm 0.19	2.49 \pm 0.09	2.53 \pm 0.2
Spleen	0.09 \pm 0.01 ^b	0.14 \pm 0.01 ^a	0.15 \pm 0.01 ^a
Bursa	0.09 \pm 0.01 ^b	0.12 \pm 0.02 ^{ab}	0.13 \pm 0.01 ^a

^{a,b} means with different superscripts within the same row are statistically different at ($p \leq 0.05$).

Dietary potential value of *Spodoptera littoralis* larvae meal on the hematological responses

The results of the hematological responses, Table (5), showed no significant effect ($p \leq 0.05$) among the treatments in red and white blood cell counts, hemoglobin level and hematocrit values. The values obtained were in consonance with the findings of earlier studies by Schiavone *et al.* (2017) who stated that the blood traits were not affected by dietary black soldier fly larvae inclusion in chicken diets.

The results of hematological traits represent strong indicators of assessing clinical and nutritional health status of chicks in feeding trial Schiavone *et al.* (2017). The values obtained for treated groups were good indication that insect meal protein was well utilized by chicks attributed to the nutritional adequacy, composition. Moreover, these observations showed no physiological stressful condition introduced in quails by feeding insect meal protein (Bovera *et al.*, 2016).

Dietary potential value of *Spodoptera littoralis* larvae meal on blood biochemical and hormonal levels

Regarding blood parameters, Table 5, there was a significant increase ($p \leq 0.05$) in serum total protein, albumin, globulin, total antioxidant, triglycerides and cholesterol. Moreover, thyroxin, estradiol-17_B, and testosterone hormones levels were significantly higher ($p \leq 0.05$) in the treated groups than in the control group. While, serum ALP, ALT and AST activities showed no significant differences among treatment groups. Since total proteins, albumen and globulin are generally influenced by total protein intake. Bovera *et al.* (2016) observed that the albumin:globulin ratio decreased when broilers were fed an insect meal diet. The values obtained in this study indicate nutritional adequacy of the dietary proteins. Abnormal serum albumin usually indicates an alteration of normal systemic protein utilization. Total serum protein may be used as an indirect measurement of dietary protein quality, Alikwe *et al.* (2010). In addition, a low albumin:globulin



Table 5 – Blood hematology, serum constituents and hormonal levels as affected by insect meal replacement in Japanese quails at 42 days of age (Mean \pm SE)

Parameters	Experimental groups		
	Control	50% insect meal	100% insect meal
RBCs ¹ (count $\times 10^6$)	5.0 \pm 0.01	5.14 \pm 0.03	5.25 \pm 0.03
Hb ² (g/dl)	23.96 \pm 0.55	24.43 \pm 0.86	25.19 \pm 0.38
PCV ³ %	51.3 \pm 0.8	51 \pm 0.82	52.3 \pm 1.05
WBCs ⁴ (count $\times 10^3$)	282 \pm 6.53	283 \pm 7.14	284 \pm 7.89
Total protein(g/dl)	3.3 \pm 0.05 ^b	3.4 \pm 0.07 ^b	3.6 \pm 0.05 ^a
Albumin(g/dl)	1.75 \pm 0.05 ^b	1.8 \pm 0.04 ^a	1.85 \pm 0.07 ^a
Globulin(g/dl)	1.55 \pm 0.08 ^b	1.6 \pm 0.16 ^b	1.75 \pm 0.05 ^a
Albumin/Globulin ratio	1.13 \pm 0.05	1.12 \pm 0.04	1.05 \pm 0.05
Total antioxidant (mM / L)	1.4 \pm 0.08 ^b	1.5 \pm 0.03 ^{ab}	1.6 \pm 0.04 ^a
Alkaline Phosphatase (IU/L)	84.4 \pm 3.2	85.4 \pm 3.9	84.51 \pm 2.7
ALT ⁵ (U/ml)	38.7 \pm 0.02	38.4 \pm 0.4	37.73 \pm 0.4
AST ⁶ (U/ml)	33.8 \pm 0.3	33.0 \pm 0.3	33.20 \pm 0.3
Triglyceride(mg/dl)	178.7 \pm 1.87 ^b	179.8 \pm 1.87 ^b	189.2 \pm 1.87 ^a
Cholesterol(mg/dl)	190.3 \pm 4.35 ^b	197.5 \pm 3.55 ^b	211.5 \pm 1.86 ^a
Estradiol-17 _β (E2) (pg/ml)	210 \pm 5.78 ^b	241.2 \pm 5.37 ^a	244.2 \pm 7.91 ^a
Testosterone (ng/ml)	5.16 \pm 0.55 ^c	9.74 \pm 0.37 ^a	7.17 \pm 0.73 ^b
T ₄ ⁷ (nmol/L)	60.9 \pm 0.75 ^{ab}	59.5 \pm 1.56 ^b	63.15 \pm 1 ^a

^{a,b,c} means with different superscripts within the same row are statistically different at ($p \leq 0.05$).

RBCs1: red blood cell Hb2: Hemoglobin concentration PCV3: packed cell volume WBCs4: white blood cell ALT5: alanine aminotransferase AST6: aspartate aminotransferase T47: Thyroxin

ratio indicates better disease resistance and immune response of birds. Bovera *et al.* (2016), observed that the albumin:globulin ratio decreased when broilers were fed an insect meal diet. The values obtained for growing Japanese quail on tested diets were in consonance with the findings of Duwa *et al.* (2014) who showed significant effect ($p \leq 0.05$) among treatments in serum globulin, triglycerides and cholesterol values of rabbits fed maggot meal as a replacement for fish meal. In addition, the significant increase ($p \leq 0.05$) in serum total antioxidant in the present study seems to suggest that there were no heavy metals and toxins in the insect meal that negatively affected nutrient metabolism. In this regard, van Huis (2013) observed that feeding insects to chickens may decrease the use of antibiotics in the poultry industry. Therefore, research efforts should be carried out in order to assess antibacterial, antifungal, anti-nutritional and toxic factors of *Spodoptera littoralis* larvae.

Concerning liver function, our results revealed that there were no significant differences in serum ALP, ALT and AST activities among treatment groups. They fell within the normal range in all treated groups. The activities of serum ALP, ALT and AST enzymes could be important in the diagnosis of diseases as well as in the investigation and thorough assessment of feed, drugs and extracts used in the treatment as these could give indications of progressive toxicity long before the actual manifestation of the toxic effects. Moreover,

increased serum ALP, ALT and AST activities have all been associated with physiological stressful condition (Bovera *et al.*, 2016).

In general, the results of serum ALT and AST indicated that the tested diets did not precipitate any significant harmful effect on the health status of the growing quail chicks, this is good indication that dietary protein was well utilized by chicks.

The potential importance of dietary composition of *S. littoralis* larvae meal in regulating the circulating concentrations of serum testosterone, estradiol-17_β and thyroxin (T₄) is presented in Table (5). This study showed a marked increase in plasma (T₄) level in the treated group with 100% insect meal compared to the one treated with 50% insect meal. The thyroid gland and its secretions appear to play a role in the endocrine regulation of growth in birds (Darras *et al.* 1994). In addition, feeding patterns play an important role in the regulation of plasma thyroid hormones concentration. Thus, the present results are similar to those of Rahimi & Hassanzadeh (2007), who fed a diet containing low protein (protein-deficient) to growing chickens which resulted in a reduction in serum thyroxin hormones concentrations, while the higher protein content of the diet, significantly ($p \leq 0.05$) affected plasma thyroxin hormone levels and higher plasma growth hormone levels. The dietary protein was associated with higher circulating plasma thyroid hormones levels. The



significant increase ($p \leq 0.05$) in serum hormones levels in the treated groups is a result of insect meal protein replacement indicating the quality, rich in essential amino acids and higher protein content of the feed. The observed significant increase ($p \leq 0.05$) in the T_4 in the treated groups as comparison with the control group in this study is logic since it is necessary for most body functions. The significant increase ($p \leq 0.05$) in serum testosterone and estradiol-17B levels in the treated groups in comparison to the control group are in agreement with those finding by Longcope *et al.* (2000) who showed in animal studies a high protein diet increased sex hormone level. They also, indicated that protein intake can be a controlling factor of sex hormone concentration. Thus, diets high in protein content may lead to increased serum concentrations of testosterone or estradiol bioactivity.

The present study indicates that, the Dietary potential value of *S. littoralis* larvae meal protein on the levels of thyroid, estradiol-17 β , and testosterone hormones in the blood serum of Japanese quail chicks at 6 weeks of age and provides new interesting data about a possible causal relationship between the growth promoting effect of insect meal and thyroid hormone.

CONCLUSION

The inclusion of *S. littoralis larvae meal* as a replacement of MBM in the diets of growing quail chicks, resulted in better effects on growth, feed performance parameters, carcass characteristics, hematological and biochemical indices than in the control group; and economically, lowering the production costs. Thus, suggesting that *S. littoralis* larvae could be considered a promising new feed ingredient for poultry.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

ACKNOWLEDGMENT

The authors are grateful to Dr. Wakwak, Mohamed Magda, who assisted and provided advice in the production of this article.

REFERENCES

Adeniji AA. Effect of replacing groundnut cake with maggot meal in the diet of broilers. *International Journal of Poultry Science* 2007;6(11):822-825.

Alikwe PCN, Faremi AY, Egwaikhide PA. Biochemical evaluation of serum metabolites, enzymes and hematological indices of broiler chicks fed with varying levels of rumen epithelial scraps in place of fish proteins. *Research Journal of Poultry Science* 2010;3(2):27-30.

Al-Qazzaz MFA, Ismail D, Akit H, Idris LH. Effect of using insect larvae meal as a complete protein source on quality and productivity characteristics of laying hens. *Brazilian Journal of Animal Science* 2016;45:518-523.

Ammar A, Albalasmeh, Berhe AA, Teamrat A, Ghezzehei. A new method for rapid determination of carbohydrate and total carbon concentrations using UV spectrophotometer. *Carbohydrate Polymers* 2013;97:253-261.

AOAC - Association of Analytical Chemists. Official methods of analysis of official chemists. 17th ed. Washington; 2002.

Basiouny A, Ghoneim K, Tanani M, Hamadah Kh, Waheeb H. Disturbed protein content in Egyptian cotton leafworm *Spodoptera littoralis* (Boisd.) (Lepidoptera:Noctuidae) by some novel chitin synthesis inhibitors. *International Journal Advanced Research Biological Science* 2016;3(3):1-12

Bombata FHA, Balogun O. The effect of partial or total replacement of fish meal with maggot meal in the diet of tilapia (*Oreochromis niloticus*) fry. *Journal of Prospects in Science* 1997;1:178-181.

Bovera F, Loponte R, Marono S, Piccolo G, Parisi G, Iaconisi V, et al. Use of tenebrio molitor larvae meal as protein source in broiler diet: effect on growth performance, nutrient digestibility, and carcass and meat traits. *Journal of Animal Science* 2016;94:639-647.

Bukkens SGF. Insects in the human diet: nutritional aspects. *In: Paoletti MG, editor. Ecological implications of minilivestock; role of rodents, frogs, snails, and insects for sustainable development.* New Hampshire: Science Publishers; 2005. p.545-577.

Dacie J, Lewis S. Practical hematology. 7th ed. Londres: Churchill Livingstone; 1991. p.37-48.

Danieli PP, Ronchi B, Speranza S. Alternative animal protein sources for aquaculture: a preliminary study on nutritional traits of Mediterranean brocade (*Spodoptera littoralis* Boisduval) larvae. *Italian Journal of Animal Science* 2011;10:109.

Darras VM, Kühn ER, Decuyper E. Comparative aspects of maturation and control of thyroid hormone deiodination during development. *Journal of Zoology* 1994;40:383-400.

Duncan DB. Multiple range and multiple F tests. *Biometrics* 1995;11:1-42.

Duwa, H, Saleh B, Igwebuike JU. The replacement of fish meal with maggot meal on the performance, carcass characteristic, hematological and serum biochemical indices of growing rabbits. *Global Journal of Bio-Science and Biotechnology* 2014;3(2):215-220.

Elwert C, Knips I, Katz P. A novel protein source: maggot meal of the black soldier fly (*Hermetia illucens*) in broiler feed. *In: Gierus, M, Kluth H, Bulang M, Kluge H, editors. Tagung schweine- und geflügelernährung, Wittenberg: Institut für Agrarund Ernährungswissenschaften, Universität Halle-, Lutherstadt Wittenberg; 2010. p.140-142.*

FAO - Food and Agriculture Organization of the United Nations. Insects to feed the World . *In: Proceedings of the 1st International Conference; 2014 May 14-17; Wageningen. The Netherlands; 2014.*

Ijaiya AT, Eko EO. Effect of replacing dietary fish meal with silkworm (*Anaphe infrecta*) caterpillar meal on performance, carcass characteristics and hematological parameters of finishing broiler chicken. *Pakistan Journal of Nutrition* 2009;8:850-855.

Jozefiak D, Jozefiak A, Kieronczyk B, Rawski M, Swiatkiewicz S, Dlugosz J, et al. Insects – a natural nutrient source for poultry – a review. *Annals of Animal Science* 2016;16:297-313.



- Longcope C, Feldman HA, Mckinlay JB, Araujo AB. Diet and sex hormone-binding globulin. *The Journal of Clinical Endocrinology and Metabolism* 2000;85 (1):293–296.
- Makkar HPS, Tran G, Heuze V, Ankers P. State-of-the-art on use of insects as animal feed. *Animal Feed Science and Technology* 2014;197:1–33.
- Mohamed HM. Comparative study of host plants on growth, development and fecundity of the cotton leaf worm *Spodoptera littoralis* (Boisduval), Noctuid: Lepidoptera. *Journal of the Egyptian German Society of Zoology* 2003;42E:167-183.
- Natt MP, Herrick CA. A new blood count diluents for counting the erythrocytes and leucocytes of chicken. *Poultry Science* 1952;31:735-738.
- NRC - National Research Council. Nutrient requirements of poultry. 9th rev ed. Washington: National Academy Press; 1994.
- Oyegoke OO, Akintola AJ, Fasoranti JO. Dietary potentials of the edible larvae of *Cirina forda* (westwood) as a poultry feed. *African Journal of Biotechnology* 2006;5:1799-1802.
- Rahimi G, Hassanzadeh M. Effects of different protein and energy contents of the diet on growth performance and hormonal parameters in two commercial broiler strains. *Internationa Journal of Poultry Science* 2011;6(3):195-200.
- SAS - Statistical Analysis System. User's guide. Release 9.1. 3rd ed. Cary: SAS Institute; 2002.
- 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. *Italian Journal of Animal Science* 2017;6(1):93–100.
- Robert AB, Paul AS, Iji. Specialized protein products in broiler chicken nutrition: a review. *Animal Nutrition* 2015;1(2):47-53.
- Van Huis A. Potential of Insects as Food and Feed in Assuring Food Security. *Annual Review of Entomology* 2013;58(58):563-583.
- Widjastuti T, Wiradimadja R, Rusmana D. The effect of substitution of fish meal by Black Soldier Fly (*Hermetia illucens*) maggot meal in the diet on production performance of quail (*Coturnix coturnix japonica*). *Animal Science* 2014;57:125-129.
- Womeni HM, Tiencheu B, Linder M, Nabayo EMC, Tenyang NFT, Mbiapo P, et al. Nutritional value and effect of cooking, drying and storage process on some functional properties of *Rhynchophorus phoenicis*. *International Journal Life Science Pharma Research* 2012;2(3):203-219.

