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Fish Meal Substitution with Black Soldier Fly Larva on Growth Performance of Female Grower Quail

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ABSTRACT

The study aimed to evaluate fish meal substitution with black soldier fly larvae supplemented with 2% *Trichoderma* culture on female grower performance and carcass traits quail. The materials used were 200 two-week-old female quails with an average body weight of 51.5 ± 7.3 grams. The study used a completely randomized design with five treatments and four replications. The treatments were: BSF0 (100% fish meal), BSF25 (75% fish meal + 25% BSF meal), BSF50 (50% fish meal + 50% BSF meal), BSF75 (25% fish meal + 75% BSF meal)), BSF100 (100% BSF meal). The BSF meal used was added with 2% *Trichoderma* culture. The rations were iso-calorie and iso-protein, formulated from yellow corn, soybean meal, fish meal, BSF meal, mineral mix, and *Trichoderma* culture, with metabolic energy of 2,900 kcal/kg and 24% protein. The data were analyzed using variance analysis to determine if it had a significant effect, then continued with Duncan's Multiple Range Test. The results showed that the treatments were insignificant to the FCR, percentages carcass, and ovaries. Nevertheless, significant to ($p<0.05$) feed intake, live weight, average daily gain and abdominal fat. The substitution of fish meal with BSF + 2% *Trichoderma* culture was more than 75% decreased feed consumption which caused a decrease in quail's life weight. According to the carcass traits, the BSF + *Trichoderma* sp 2% could be used up to 100%. However, based on quail performance, it can be concluded that BSF + 2% *Trichoderma* sp can be used up to 75% in quail rations without harmful effects.

Keywords: abdominal fat, carcass, *Hermetia*, ovarium, *Trichoderma*

INTRODUCTION

Feed costs can form up to 70% of the total cost of poultry production. Animal protein sources are preferred over plant protein sources because animal proteins contain a good balance of essential amino acids and excellent vitamins (Saima et al., 2008; Mahmood et al., 2014). Fish meal is a high-quality protein source used mainly in poultry diets. However, the high price of fish meals has been searched for alternative protein sources. Some substitution studies of fishmeal with black soldier fly (BSF)/ *Hermetia illucens* larvae/prepupae have been carried out. However, the results cannot reach 100% depending on the larval/prepupa age.

The utilization of BSF larvae at various ages gives different results. For example, the 10-day BSF maggot cultivated in the coconut cake can be used up to 25%, replacing the fish meal or 11.25% in broiler rations to dry matter and protein digestibility (Rambet et al., 2016). Dengah et al. (2016) showed that the use of larvae aged ten days with a level of 0, 25, 50, 75, and 100% in broiler rations (15% fish meal content) showed that in starter period substitution, more than 50% its reduced feed consumption, but no effect on body weight gain and feed efficiency. More than 50% substitution decreased feed consumption and body weight gain during the finisher period.

Widjastuti et al. (2014) stated that substituting fish meal (10% in rations) with BSF meal to a limit of 50% could support the performance of quail production. One obstacle to using BSF is chitin as an exoskeleton component of insects, where poultry cannot digest chitin. The chitin content in a fish meal is 0%; on the contrary, the BSF was 9.6%. The chitin content of BSF larvae is lower than that of pupae. The physiological phase of the larvae influences chitin content. The older, the higher the chitin level. Chitin contributes to nitrogen instead of protein and contributes 1-7% of all body N (Finke, 2007). The chitin content of BSF larvae is estimated to be 5.4% (Finke, 2012). High chitin levels in insects harm feed intake and disrupt protein use (Longvah et al., 2011), reducing nutrient utility availability and affecting performance growth (Kroeckel et al., 2012). Marono et al. (2015) studied that in vitro chitin reduces protein digestibility. They solve chitin that cannot be digested, and low palatability is the key to developing BSF prepupa as a feed resource. *Trichoderma produces chitinase, β-1,3-glucanase, and protease* (Elad et al., 1982; Sandhya et al., 2004).

Chitinase enzymes can break down chitin polymers into *N-acetyl glucosamine* monomer units (Ulhoa and Peberdy, 1991; Isahak et al., 2014)

Therefore, this research now aims to evaluate the effects of different levels of administration of BSF larvae supplemented with 2% *Trichoderma* for the fish meal on the female quail grower period.

MATERIALS AND METHODS

Animal and diets

This study used 200 two-week female quails (*Coturnix coturnix* japonica) with an average body weight of 51.5±7,3 g. They were randomly allocated to 20 pens with five treatments and four replicates each. Each treatment was composed of 10 quails. The experiment used a completely randomized (CRD) design with 5 x 4. Five diets are formulated with processed BSF meal (+2% *Trichoderma* culture) to replace protein fish meal at levels of 0%, 25%, 50%, 75%, and 100% designated as BSF0, BSF25, BSF50, BSF75, and BSF100, respectively. Adding 2% Trichoderma, based on Mulyono et al. (2019) studies, showed that adding 2% Trichoderma spp increased the digestibility of BSF larvae meal. The diets were formulated to be isonitrogenous and isoenergy. The BSF larvae of various ages were obtained from the manure of laying hens. The collected larvae are then cleaned and dried in the sun to dry. Finally, the dried larvae are ground into flour and ready for research. The chemical composition of the feed ingredients and nutritional contents are listed in Table 1. The composition of feed ingredients and nutrient content of the experimental ration are listed in Table 2.

Table 1. The nutrient content of the feedstuff (%)

Feedstuff	DM	Ash	CP	EE	CF	Ca	P total
Yellow corn	86.68	1.23	8.56	2.89	3.70	0.020	0.28
BSF larvae	92.39	18.78	39.57	18.15	11.84	5.000	1.50
Soybean meal	88.37	6.22	43.12	1.35	7.13	0.250	0.630
Fish meal	91.38	22.68	41.96	15.02	11.38	5.000	2.50

DM: Dry matter, CP: Crude protein, EE: Ether extract, CF: crude fibre

Table 2. Feed ingredients and nutritional content.

Ingredient	Treatments				
	BSF0	BSF25	BSFT50	BSF75	BSF100
Yellow corn (%)	51.70	51.45	51.16	51.04	50.82
BSF+2% *Trichoderma sp* (%)	0.00	2.12	4.24	6.36	8.48
Fish meal (%)	8.00	6.00	4.00	2.00	0.00
Soybean meal (%)	38.60	38.73	38.90	38.90	39.00
Premix[1] (%)	1	1	1	1	1
$CaCO_3$ (%)	0.70	0.70	0.70	0.70	0.70
total	100.00	100.00	100.00	100.00	100.00
Calculated nutrition					
Dry matter (%)	87.9	87.9	87.9	88.0	88.0
Ash (%)	5.5	5.4	5.4	5.3	5.3
Crude protein (%)	24.4	24.5	24.5	24.5	24.5
Ether extract (%)	3.22	3.30	3.37	3.45	3.53
Crude fibre (%)	5.6	5.6	5.6	5.6	5.7
Ca (%)	0.8	0.8	0.8	0.8	0.8
P total (%)	0.6	0.6	0.6	0.5	0.5
Metabolizable energy (kcal/kg)[2]	2,935	2,940	2,945	2,951	2,957
ME/CP ratio	120	120	120	120	121

[1] Mineral and vitamin contents per kg of the diet: 12500 IU vitamin A, 2500 IU vitamin D3, 7.5 IU vitamin E, 2 mg vitamin K, 2.5 mg vitamin B1, 4 mg vitamin B2, 1 mg vitamin B6, 0.012 mg vitamin B12; 0,2 mg biotin, 0,5 mg folic acid, 50 mg vitamin C, 50 mg Nicotinate acid, 4 mg Ca-D Pantothenate, 15 mg choline chloride, 5 mg copper, 25 mg iron, 0,2 mg iodine, 60 mg manganese, 0,2 mg selenium, 70 mg zinc, 0.2 mg cobalt, 21 mg Zinc bacitracin, 160 Lysine, 50 mg DL-methionine, 4 mg Threonine

[2] Calculated value by Bolton formula (Bolton, 1967): 40.81 ×{0.87 (crude protein + 2.25 crude fat + nitrogen-free extract) +2.5)

The experiment was conducted at a quails farm in the Kalisidi village, West of Ungaran subdistrict, Semarang Regency, Central Java, Indonesia. The chemical composition of BSF larvae of various ages obtained from the manure of laying hens can be found in Table 1. Birds' weight was recorded at the beginning and weekly on a 1000 ± 0.1 g scale. Feed in the form of the mash was given *ad libitum* during the study (28 days), and drinking water was given *ad libitum*.

Parameters measured

Quails' performance and carcass traits

Feed consumption was recorded daily by weighing the feed given reduced feed remains (gram/day). Live body weight (LW) was measured at the beginning of the study and once a week (g/bird). Average daily weight gain (ADG) was calculated by body weight gain (g)/ the number of days (grams/birds/day). The feed conversion ratio (FCR) was determined by feed intake (g)/weight gain (g).

Recording deaths that occurred, mortality rates were calculated based on deaths during the study period. One bird in each treatment was randomly selected for carcass measurements. Quails were fasted for approximately four hours and then individually weighed and sacrificed (Genchev and Mihaylov, 2008). After sacrificing, each bird's head, neck, feathers, feet, and internal organs were removed, and carcass weights were recorded. The yield of the carcass was calculated as a percentage of live weight.

Furthermore, the abdominal fat tissue adjacent to the cloaca was removed and weighed. The abdominal fat content was calculated as a percentage of live weight. The reproductive organs are the ovaries and oviducts, measured in percentages.

Statistical analysis

Data analysis used variance analysis procedures. Duncan's multiple range test examines the differences between treatment means at the significance level was $p<0.05$ (Steel et al., 1997).

RESULT AND DISCUSSION

Growth performance

Performance parameters are shown in Table 3. The treatment effects were significant ($p<0.05$) on feed intake, live weight, and average daily gain, yet no significant effect ($p> 0.05$) on feed ration conversion. In addition, mortality occurred in BSF0, BSF25, and BSF75 treatments, each one tail in the sixth week, whereas in BSF25 and BSF100 treatments, there was no death.

Table 3. The effect of diets on the performance of Japanese quail

Variable	Treatment						
	BSF 0	BSF 25	BSF 50	BSF 75	BSF 100	SEM	p
Feed intake (g/day)	16.5^{a}	16.2^{a}	16.0^{a}	15.9^{a}	15.1^{b}	0.22	0.006
Life weight (g/bird)	157.4^{a}	154.6^{a}	156.6^{a}	152.8^{ab}	148.9^{b}	1.61	0.015
ADG (g/d)	3.8^{a}	3.7^{a}	3.7^{a}	3.6^{ab}	3.5^{b}	0.06	0.017
FCR	4.8	4.7	4.8	4.6	4.7	0.009	0.874
Mortality (tails)	1	0	1	1	0		

ADG=average daily gain, FCR = feed conversion ratio

$^{a-d}$ Means in a row with different superscripts differ at $p<0.05$

Feed intake

Quail feed intake per day of each treatment ranged from 15.1 to 16.5. The results showed that the treatments significantly affected feed intake. The treatments BSF 0, BSF 25, BSF 50, and BSF 75 were insignificant; however, the BSF 0, BSF 25, and BSF 50 were significant to BSF 100 ($p <0.05$) BSF 75 was not significant with BSF 100 treatment. The results showed that substituting a fish meal with BSF larvae supplemented with 2% Trichoderma fungi could be used up to 75%, but 100% substitution reduced feed intake. Although the metabolic energy content of feed in this study was equivalent, there was a decrease in feed consumption in the BSF 100 treatment. It was suspected that the low palatability of BSF larvae meal compared with fish meal. Low palatability causes the consumption of rations to decrease.

The present study results are higher than Dengah et al. (2016), who stated that substitution above 75% reduced ration consumption in the broiler stater phase and above 50% in the finisher phase. Widjastuti et al. (2014), feed consumption declined by more than 50% in the quail layer period. According to Awoniyi et al. (2003), replacing fish meal (4% in ration) with maggot flour was at a level greater than 50% in the decrease in broiler feed consumption.

Live Weight and Average Daily Gain

The results showed that substituting a fish meal with BSF larvae supplemented with 2% Trichoderma could be used up to 75% without reduced body weight and quail weight gain. However, 100% substitution decreases body weight and quail weight gain. This result results from Teguia et al. (2002) that replacing fish meal (4.5% in ration) with magot to 15% in the broiler starter phase increases ration consumption. Still, fish meal replacement (2% in ration) grower-finisher to 100% reduce ration consumption.

Based on the final body weight parameters and daily weight gain, using up to 75% to replace fish meals was recommended. Bodyweight and quail weight gain was strongly influenced by ration consumption. The results of this study appear to be the final body weight caused by feed consumption rather than feed efficiency. It was seen that the FCR between treatments is not significantly different. The regression analysis results with the dependent variable body weight gain and the independent variable ration consumption showed the equation $Y = 72.3 + 0.183X$ with the value of $R\text{-sq} = 0.546$, $r = 0.739$ and $p = 0.000$. This equation shows that every 1-gram increase in consumption will increase the quail bodyweight by 0.183 grams. R-sq value explains that 54.6% of quail body weight is caused by ration consumption. The higher feed consumption leads to more incredible energy and nutrient consumption, and vice versa; low consumption of rations causes energy consumption, and nutrients are also low. Energy and nutrients convert for growth and maintenance needs, and low feed consumption at BSF100 results in low body weight.

Substitution of fish meal with BSF meal supplemented with 2% *Trichoderma* did not affect FCR. It shows that the rations BSF 0, BSF 25, BSF 50, BSF 75, and BSF 100 have equal quality, and quails have the same ability to convert rations. Feed intake and body weight gain directly affect feed conversion; the better the quality of the feed, the smaller the value of conversions. There was indicated that the substitution flour BSF larvae supplemented with 2% of *Trichoderma* can be used up to 100% without affecting FCR.

Carcass, abdominal fat, and ovary

The results showed that the treatment did not affect the weight of carcass and carcass percentage, ovarian weight, and ovarian percentage. However, it affected ($p < 0.05$) the weight and percentage of abdominal fat.

Table 4. Effect of treatment on the carcass, abdominal fat, and ovary of female quails age six weeks

Variables	Treatments						
	BSF 0	BSF 25	BSF 50	BSF 75	BSF 100	SEM	p
Carcass (g)	96.8	95.2	96.3	94.7	93.1	1.004	0.141
Percent carcass (%)	62.0	61.4	61.6	61.6	61.8	0.368	0.838
AL (g)	2.73^{a}	2.73^{a}	2.81^{a}	2.18^{b}	1.55^{c}	0.106	0.000
Per cent AL (%)	1.75^{a}	1.76^{a}	1.79^{a}	1.41^{b}	1.03^{c}	0.062	0.000
Ovarium (g)	4.35	4.35	4.20	4.25	4.20	0.247	0.983
Percent ovarium(%)	2.78	2.80	2.69	2.76	2.78	0.146	0.982

AL = abdominal fat

^{ac} Means in a *row with different superscripts differ at p<0.05*

The factors that affect carcass weight are sex, age, activity, breeds, quantity and quality of feed, body fat, body weight and feed consumption. High final body weight was expected to produce a high percentage of carcasses and relatively lesser byproducts. Therefore, the weight and percentage of the carcass were produced by feed consumption and final quail body weight.

The percentage of carcasses in this study ranged from 61.4 to 62.0%, higher than the research of Tugiyanti and Herijanto (2018), precisely 55.0 – 59.2%, and Oguz et al. (1999) 67.69%. Factors affecting carcass production are consuming protein and energy to produce meat. The carcass weight and the percentage of carcass do not correspond to the quail slaughter weight, where the slaughter weight in BSF 0, BSF 25, and BSF 50 treatment differed from BSF 75 and BSF 100. It was presumably because the carcass will be relatively constant when the adult body has been reached. The food consumed will be diverted for reproduction and not for meat formation so that the live weight and percentage of the carcass do not differ. With age, the growth rate of carcass tissue will remain in line with the growth of body tissue. Quail body adult age is when the quail has reached six weeks (Mizutani, 2003).

Abdominal fat weights and percentages are listed in Table 4. Average abdominal fat weights range from 1.6 to 2.8 g, more significant than the study of Narinc et al. (2014) of 1.19 g. The average percentage of abdominal fat ranges from 1.04 to 1.09%; this result follows the study of Kianfar et al. (2013)), which ranges from 0.99-1.41%.

The treatment of fish meal replacement with BSF flour supplemented with 2% *Trichoderma* affected ($p < 0.05$) the weight and percentage of abdominal fat. Treatment BSF 0, BSF 25, and BSF 50 were not different; however were different ($p < 0.05$) with BSF 75 and BSF 100, and BSF 75 treatments were different from BSF 100. These results indicate that BSF 100 treatment has the lowest abdominal fat content.

Abdominal fat is an extensive deposition of fat in poultry and correlates with total carcass fat (Lotfi et al., 2011). Abdominal fat is related to body weight and quail age. Bodyweight gain is caused by abdominal fat accumulation (Narinc et al., 2014). The accumulation of fat is related to the consumption of rations. More elevated consumption causes an increase in energy and protein consumption which were used for quail growth. The excess energy was deposited in the form of abdominal fat.

Conversely, the lower consumption of rations leads to decreased nutrients and energy. It causes a lower fat that was seen in low abdominal fat. Abdominal fat weight was higher in quails that consume higher metabolic energy than lower metabolic energy consumption. It was consistent with previous research that broilers fed with high EM abdominal fat are greater than low EM (Deaton et al., 1983; Deaton and Lott, 1985; Fouad and El-Senousey, 2014).

The ovarian weight and relative weight of the ovary

The treatment of fish meal replacement with BSF flour supplemented with 2% *Trichoderma* culture did not affect ovarian weight and relative ovarian weight (%). These results indicate that the treatment of replacing fish meal with BSF flour added with 2% *Trichoderma* mould does not interfere with ovarian growth. The growth and development of ovaries are related to age and quail weight; following Arora and Samples (2011) and Elkomy et al. (2019), the greater the quail's weight, the greater the ovarian weight.

The average ovarian weight ranges from 4.2 to 4.4 g; the present study is under Hakim et al. (2014) research that the weight of a quail ovary at six weeks with a body weight of 158 grams fed commercial feed was 4.6 g. Thus, the relative weight of the ovary ranges from 2.69 to 2.78%, smaller than the study of Elkomy et al. (2019), that 98 days of quail has a relative ovarian weight of 3.43 - 4.33%. Adult female quail sex is characterized by first laying eggs (Elkomy et al., 2019); quails reach average adult sex at six weeks (Wiradimadja et al., 2007). However, there are younger ones who are also older than that age. This situation is caused by health, management, genetic factors (Sezer et al., 2006), lighting (Elkomy et al., 2019; (Hakim et al., 2014); body weight (Sezer et al., 2006), food (Ocak and Erener, 2005) also influence adulthood.

CONCLUSION

Substitution of fish meal (8% in the diet) using BSF + 2% larval culture of *Trichoderma* can be used up to 100% and does not negatively affect the FCR, carcass and ovarian weights. Nevertheless, decreased feed intake and quail growth above 75%.

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