

Supplementation of Selenium-enriched Black Soldier Fly (*Hermetia illucens*) Larvae Meal on Growth Performance, Blood Parameters, and Immune Function in Broiler Ducks

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ABSTRAK

Kurniawan D, Widodo E, Susilo A, Sjojfan O. 2024. Suplementasi tepung larva lalat tentara hitam (*Hermetia illucens*) yang diperkaya selenium terhadap performa pertumbuhan, parameter darah, dan fungsi imun pada itik pedaging. JITV 29(4):227-235. DOI: <http://dx.doi.org/jitv.v29i4.3452>.

Penelitian ini bertujuan untuk mengevaluasi pengaruh dari tepung larva *Hermetia illucens* yang diperkaya dengan Se (Se-BSF) terhadap penampilan produksi, biokimia darah, dan status kekebalan tubuh itik pedaging. Sebanyak 200 ekor itik pedaging persilangan berumur satu hari tanpa dibedakan jenis kelamin dibagi secara acak ke dalam empat kelompok perlakuan, masing-masing dengan lima ulangan yang terdiri dari 10 ekor itik. Itik pedaging diberi pakan dengan pakan kontrol, pakan dengan 5% dan 7,5% Se-BSF, serta pakan kontrol positif dengan 10 mg/kg Se-Yeast dengan lama pemeliharaan selama 49 hari. Hasil penelitian menunjukkan bahwa performa produksi, karakteristik karkas, dan organ dalam itik pedaging dipengaruhi secara signifikan ($P<0,001$) oleh suplementasi Se-BSF. Pemberian pakan yang mengandung 7,5% Se-BSF menunjukkan tren penurunan konsumsi pakan, bobot badan akhir, pertambahan bobot badan, dan efisiensi pakan. Pengaruh pemberian Se-BSF berbeda nyata ($P<0,001$) antara semua perlakuan pada bobot hidup, bobot karkas, persentase karkas, daging dada, daging paha, hati dan limpa. Penambahan Se-BSF pada pakan itik pedaging tidak menunjukkan perbedaan yang signifikan antara semua perlakuan pada organ jantung, ampela, lemak abdominal, dan bursa fabricius. Pemberian pakan yang mengandung 5% sampai 7,5% Se-BSF berpengaruh secara signifikan ($P<0,001$) terhadap kadar HDL, LDL, Kolesterol, Trigliserida, Ig-A, SOD, dan IL-6 dalam serum dibandingkan dengan kontrol dan 10 mg/kg Se-yeast. Hasil ini menunjukkan bahwa pemberian Se-BSF dalam pakan meningkatkan parameter biokimia darah dan fungsi kekebalan tubuh itik pedaging, yang mengindikasikan potensi manfaat dari penggunaan Se-BSF sebagai aditif pakan untuk unggas.

Kata Kunci: Tepung Larva BSF, Penampilan Produksi, Imunitas, Selenium Organik

ABSTRACT

Kurniawan D, Widodo E, Susilo A, Sjojfan O. 2024. Supplementation of selenium-enriched black soldier fly (*Hermetia illucens*) larvae meal on growth performance, blood parameters, and immune function in broiler ducks. JITV 29(4):227-235. DOI: <http://dx.doi.org/jitv.v29i4.3452>.

This study aimed to evaluate the effects of Se-enriched *Hermetia illucens* larvae meal (Se-BSF) on production performance, blood biochemistry, and immune status of broiler ducks. Two hundred one-day-old hybrid broiler ducks without sex were randomly allocated into four groups, each with five replications of 10 ducklings. Broiler ducks were fed diets with a controlled diet, an experimental diet with 5% and 7.5% Se-BSF, and positive control with 10 mg/kg Se-Yeast, respectively, daily for 49 days with drinking water. The results showed that the performance production on the carcass traits and visceral organs of broiler ducks was significantly affected ($P<0.001$) by Se-BSF supplementation. Dietary 7.5% Se-BSF showed trends in decreasing the feed intake, final body weight, average daily gain, and feed efficiency ratio. The effect of dietary Se-BSF was significant differences ($P<0.001$) between all treatments in live body weight, carcass weight, dressing, breast meat, thigh meat, liver, and spleen. The addition of Se-BSF to broiler ducks' diet showed no significant differences between all heart, gizzard, abdominal fat, and bursa treatments. The effects of dietary 5% to 7.5% Se-BSF had significantly affected ($P<0.001$) HDL, LDL, Cholesterol, Triglyceride, Ig-A, SOD, and IL-6 levels in serum compared with control and 10 mg/kg yeast-Se. These results indicate that dietary Se-BSF improves broiler ducks' blood biochemistry and immune function, suggesting potential benefits from using Se-BSF as a feed additive for poultry.

Key Words: BSF Larvae Meal, Growth Performance, Immunity, Organic Selenium

INTRODUCTION

Poultry meat and eggs are among the most popular animal-derived consumables worldwide, regardless of

culture, tradition, or religion. Duck meat is highly nutritional because it contains all required amino acids and has a healthy fatty acid composition (Ismoyowati & Sumarmono 2019). The Food and Agriculture

Organization (2022) reported that in 2020, the worldwide population of ducks (*Anas spp.*) reached 1.15 billion and 1.0 billion (89 per cent) were in Asia. Duck meat production in Indonesia has averaged 37,878 tons/year over the last 5 years, contributing less than 1% of the meat production of all livestock species nationally (Directorate General of Animal Husbandry and Animal Health, 2022). The livestock industry is expected to rise to the challenge of developing innovative and sustainable methods in the future. Meeting the need for safe and resource-efficient livestock products, resulting in quality products that can compete globally (Barragan-Fonseca et al. 2017), the use of natural feed additives as an alternative to antibiotics (Ismita et al. 2022), the use of alternative sources of animal protein are environmentally friendly and sustainable that do not compete with humans (Cullere et al. 2019) and increase the nutritional value of poultry meat products produced (Jachimowicz et al. 2022).

Selenium (Se) is one of the indispensable nutrients for human health and livestock growth that plays a role in various physiological functions, such as antioxidant activity, immune response, and metabolism (Ferro et al. 2021). Se in the form of selenoproteins contains 21 amino acids and has 25 forms in the livestock body (Dalgaard et al. 2018). Se can improve antioxidant status through the enzymatic redox system by forming Se-Cysteine at the cellular level (Gu & Gao 2022). Se also plays a role in DNA formation, immune response to oxidative stress, maintenance of endoplasmic reticulum integrity, and nutrient metabolism (Silva et al. 2019). In nonruminant livestock such as poultry and pigs, Se in the form of selenoproteins regulates oxidative stress, redox mechanisms, and other critical cellular processes involved in innate and adaptive immune responses (Qiu et al. 2021).

There are two types of Se additions in animal feed: organic and inorganic. Organic Se (selenoprotein and Se-yeast) is more effective than inorganic Se (sodium selenite) in enhancing production performance, antioxidant status, and Se concentration deposited in animal body tissues, and plays a role in improving livestock meat quality (Ferrari et al. 2022). It has been observed that using organic sources of Se in broiler diets improves the physical and chemical properties of meat while also extending shelf life (Silva et al. 2019). However, some Se sources exhibit shortcomings that include a low content of organic Se, a long production period, and a high cost (Zhang et al. 2014). As a result, a novel Se-enriched technique that enables a high yield of organic Se in a brief production time and at a reasonable cost is still needed (Dong et al. 2021). Selenium conjugated insect protein (SCIP) is a new type of organic Se source obtained through two biotransformation steps of microbial fermentation and synthesis of insect larvae to possibly guarantee high bioavailability, biosafety, and high biological value. SCIP has been shown to benefit

the health of laying hens and improve the safety of Se-fortified eggs (Qiu et al. 2021).

Black soldier fly /BSF (*Hermetia illucen* L.) has been researched for their capacity to transform organic waste into high-quality protein, suppress some dangerous bacteria and insect pests, supply possible chemical precursors to produce biodiesel, and be fed to a variety of livestock (Barragan-Fonseca et al. 2017). BSF larvae contain high levels of protein (37-63%), dry matter, and other macro and micronutrients essential for animal feed. Existing research on BSF larvae in poultry, pigs, and fish feed shows that BSF larvae can only partially replace conventional feed ingredients (Cullere et al. 2019). (Ferrari et al. 2022) investigated the enrichment of Se in BSF larvae and found that the average total Se content obtained was produced more than five times that of the control group. Efforts are needed to explore non-toxic sources of Se as feed additives to reduce the stressors effect on poultry during production. Our study's objective was to investigate the potential of Se-enriched black soldier fly larvae meal (Se-BSF) as feed ingredients on production performance, blood biochemistry, and immune system of broiler ducks.

MATERIALS AND METHODS

Ethical approval

The Research Ethics and Animal Care Committee at the Institute of Biosciences, Brawijaya University, Indonesia, approved the experimental methodology (No 004-KEP-UB-2024).

Animals, diets, and experimental design

Two hundred one-day-old Hybrid broiler ducks were obtained from a commercial hatchery (Blitar, East Java, Indonesia). The ducklings were randomly grouped into four feeding treatments with 5 replicates and 10 birds in each group and housed in communal cages measuring 100 cm wide × 200 cm long × 60 cm high. The diet consisted of a control diet, 5% and 7.5% Se-BSF, and positive control with 10 mg/kg Se-Yeast. Dried and bio-synthesized selenium-enriched BSF larvae (Se-BSF) meals were supplied from the Department of Poultry Product Processing, Community College State of Putra Sang Fajar Blitar, Indonesia. The procedure for producing Se-BSF meal through two-step biotransformation using sodium selenite refers to the study of (Qiu et al. 2021). First, under optimal conditions, BSF larval growth media fermentation enriched with sodium selenite using yeast *Saccharomyces cerevisiae*. Bran and soybean meal are added as raw materials, and 400 mg/kg sodium selenite

Table 1. Composition (%) of the experimental broiler duck diets containing different levels of Se-BSF

Item	Starter (0-21 days)				Finisher (21-42 days)			
	Contro l	Se-BSF 5%	Se-BSF 7.5%	Se- Yeast	Contr ol	Se-BSF 5%	Se-BSF 7.5%	Se- Yeast
Corn	10.05	10.05	9.67	10.05	19.03	19.03	18.65	19.03
Soybean meal	29.05	29.05	26.93	29.05	20.07	20.07	17.95	20.07
Rice bran	45.00	45.00	45.00	45.00	45.00	45.00	45.00	45.00
Fish meal	7.00	7.00	7.00	7.00	7.00	7.00	7.00	7.00
Palm oil	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Premix	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50
Salt	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
L-Lysine	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
DL-Methionine	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
BSF meal	5.00	-	-	5.00	5.00	-	-	5.00
Se-BSF meal	-	5.00	7.50	-	-	5.00	7.50	-
Calculated composition								
Metabolizable energy (kcal/kg)	2,759	2,759	2,758	2,759	2,860	2,860	2,859	2,860
Crude Protein (%)	23.00	23.00	23.00	23.00	20.00	20.00	20.00	20.00
Crude Fat (%)	7.80	8.02	8.85	7.80	7.96	8.17	9.00	7.96
Crude Fiber (%)	3.36	3.36	3.23	3.36	3.00	3.00	2.87	3.00
Calcium (%)	0.62	0.62	0.61	0.62	0.60	0.60	0.59	0.60
Phosphorus (%)	0.38	0.38	0.36	0.38	0.33	0.33	0.31	0.33
Lysine (%)	1.82	1.82	1.82	1.82	1.58	1.58	1.57	1.58
Methionine (%)	0.69	0.69	0.70	0.69	0.65	0.65	0.66	0.65

Se-BSF= selenium-enriched black soldier fly

is added (Kurniawan et al. 2024). Second, fermented growth media (Se-rich protein yeast) is given as BSF larval feed. Finally, selenium-enriched BSF larvae are dried and ground into a Se-BSF meal, then stored in a dry place until the following process. The experimental diets were formulated using ground yellow maize, rice brand, and a soybean meal (Table 1) in two phases: the starter phase on days 1 to 21 and the finisher phase on days 21 to 49. All birds were in the same condition.

Growth performance and relative weights of organs

Weight recording was conducted weekly to determine average body weight gain (BWG) along with recording feed intake (FI) and calculating feed conversion ratio (FCR). At the age of 49 days, representative ducks were slaughtered and randomly selected from each replicate that had previously been fed

for 12 hours. Slaughter was carried out by manual slaughtering using a sharp knife, scalded in a hot water bath, and mechanically plucked feathers. Then the weight of the carcass, breast and thighs, abdominal fat, and internal organs such as liver, heart, gizzard, bursa Fabricius, and spleen were determined by weighing and calculating the relative weight as a percentage of the live body weight to determine the carcass traits of broiler ducks.

Biochemical analysis of blood serum

At the end of the finishing period (7 weeks), the representative ducks from each replicate were collected as blood samples. Blood samples were drawn into sterile, non-heparinized tubes to separate the serum and centrifuged for 15 minutes at 4°C at 3000 rpm. After collected, the serum was transferred to Eppendorf tubes

and kept cold (-20°C) for further biochemical examination. The amount of cholesterol, triglycerides, high-density lipoprotein (HDL), and low-density lipoprotein (LDL) in serum was determined using a commercial kit on a spectrophotometer. Superoxide dismutase (SOD) assay was determined with a commercial kit (Khan et al. 2023). IgA ELISA quantification kits explicitly designed for chickens were used to measure the levels of plasma IgA (Abdel-Moneim et al. 2022).

Intestinal immunohistochemistry

Intestinal immunohistochemistry was performed using the same slaughtered broiler ducks. The immunohistochemistry protocol refers to the study reported by (Khan et al. 2020). An automated tissue processor was used to process the pre-fixed ileum samples for a night. The processed ileum samples were embedded in paraffin blocks using an embedding apparatus. We sectioned ileum specimens into 4 mm thick pieces using a rotary microtome. After that, the portions were placed on highly frosted glass slides. They were run through distilled water, xylene, and ethanol at varying concentrations to de-paraffinize the paraffinized sections. They were microwave-heated in citrate buffer for 10 minutes to prime the sections for effective antigen retrieval. For immunohistochemistry, we used a DAB Detection kit. IL-6 was employed as the main antibody for the proinflammatory cytokines. Primary antibodies were first applied to the sections and added to the DAB substrate. Lastly, Harris Haematoxylin counterstaining was applied to the sections to examine them under a light microscope. Calculating the number of expression antibodies is performed using the ImageJ program to analyze the stained preparation immunohistochemistry (IHC) image to see the amount of expression-specific antibodies that can be downloaded online. The expression of IL-6 antibodies in the intestine can be calculated by describing the brown colour expressed in the preparation. Results of IL-6 expression calculation using ImageJ of the form percentage. Expression assessment was carried out on 5 fields, a different view. This observation was performed with a light microscope (Nikon Eclipse type Ei) and an Optilab Microscope Camera connected to a computer.

Statistical analysis

All data were presented as mean \pm standard deviation and analyzed using one-way ANOVA with a completely randomized design in SPSS software. To compare the means, differences between means were determined using the least significant difference (LSD) tests (Mattjik and Sumertajaya 2013). For all tests, P-values <0.05 indicated significant differences.

RESULTS AND DISCUSSION

Effect of Se-BSF on the production performance

The effects of feeding Se-BSF compared to Se-yeast on the production performance of broiler ducks are presented in Table 2. There was no mortality during the experiment. From 1 to 49 d, the FBW, ADG, FI, FCR, FER, PI, PER, EI, and EFR of broiler ducks were influenced by Se-BSF supplementation ($P<0.001$). Dietary Se-BSF 7.5% showed trends in decreasing the FI, FBW, ADG, and FER and increasing the FCR. These data revealed that dietary Se-BSF 5% did not negatively influence broiler ducks' performance production. The effect of feeding Se-BSF on broiler ducks' carcass and internal organ traits showed significant differences ($P<0.001$) in Dressing, BM, TM, Liver and Spleen. Adding Se-BSF to the broiler duck diet resulted in no significant differences in heart, gizzard, abdominal fat, or bursa.

Growth performance characteristics are utilized to assess the financial advantages of additives, such as Se-BSF supplementation. Compared to Se-yeast, the current study showed that dietary Se-BSF significantly impacted the growth performance of broiler ducks. Feeding selenium improved body weight, feed consumption, and feed conversion ratio. According to (Al-Quwaie 2023) the addition of *Bacillus subtilis* selenium nanoparticles (BseNPs) to the feed considerably raised the chicks' final body weight, reduced their feed intake, and improved their FCR value. Similarly, (Rehman et al. 2022) discovered that adding SeNP-MOS to the broiler diet enhanced feed consumption, FCR, weekly BWG, and final body weight of broilers that were ready for the market. The SeNP supplementation had greater body weight, weight gain, and performance indicators after 38 days of the feeding trial. Some research showed that selenium had no significant effect on growth and performance.

Bami et al. (2022) reported that different levels of green synthesized nano selenium (GNS) had no significant effect on broiler chicken growth. The discrepancies in results could be attributed to variances in Se levels, experimental setups, or chicken breeds used. The dietary Se-BSF showed significant differences in CW, BM, TM, Liver, and Spleen. Still, there was no significant difference among all treatments in dressing, heart, gizzard, abdominal fat, and bursa. The previous findings (Al-Quwaie 2023) demonstrated that feeding BSeNPs to broiler diets significantly decreased fat content and increased breast muscle. Still, there were no significant differences in the carcass properties. (Elkhateeb et al. 2022) reported that broilers fed diets fortified with different sources of selenium had higher percentages of dressing and abdomen fat but no effect on internal organs among all treatments. (Bakhshalinejad et

al. 2019) reported similar findings, demonstrating that Se source, Se inclusion rate, and their interaction did not affect carcass traits.

control group. In terms of oxidative stress characteristics, ducks treated with 5% and 7.5% Se-BSF exhibited significantly lower serum SOD levels than ducks in the control group.

Effect of Se-BSF on the blood biochemistry

The effect of Se-BSF feeding on blood biochemical parameters and the immune status of broiler ducks are presented in Table 4. After 49 days, the serum levels of HDL, LDL, Cholesterol, Triglycerides, Ig-A, SOD, and IL-6 were significantly (P<0.001) affected in ducks with 5% to 7.5% Se-BSF compared to controls and Se-yeast. In addition, compared to controls, ducks in Se-BSF 7.5% had significantly decreased HDL, LDL, triglycerides, and cholesterol levels. Regarding immunological status parameters, ducks fed between 5% and 7.5% Se-BSF showed significantly greater serum Ig-A levels than the

Ileum immunohistochemistry of broiler ducks

The immunohistopathology of the ileum of broiler ducks showed immunostaining for IL-6 (Figure 1). The results of the semi-quantitative analysis of immunostaining for different proinflammatory cytokines are summarized in Table 4. Dietary Se-BSF significantly increased interleukin 6 (IL-6) levels in broiler ducks compared to the control group (P<0.001).

Expression IL-6 antibodies in the intestine can be calculated through the description brown colour expressed in the preparation (Arrow). Results of IL-6

Table 2. Effect of dietary Se-BSF on the growth performance, carcass traits, and visceral organ of broiler ducks from 0 to 49 d of age

Parameters	Control	Se-BSF (%)		Se-Yeast	SEM	P-value
		5	7.5			
Growth performance						
IBW (g)	47.00±1.22	46.88±1.21	46.58±0.83	46.6±0.82	0.1907	0.942
FBW (g)	2,052±93 ^b	1,999±74 ^b	1,666±107 ^a	1,690±82 ^a	80.83	<0.001
ADG (g)	2,005±94 ^b	1,952±74 ^b	1,620±107 ^a	1,656±87 ^a	78.85	<0.001
FI (g)	5,245±283 ^b	4,937±384 ^{ab}	4,680±236 ^a	4,66±93 ^a	155.28	<0.001
FCR	2.77±0.13 ^a	2.69 0.11 ^a	3.11±0.24 ^b	2.89±0.18 ^{ab}	0.082	<0.001
FER	36.20±1.64 ^b	37.21±1.50 ^b	32.33±2.56 ^a	34.70±2.07 ^{ab}	0.827	<0.001
PI	1,029±51 ^b	972±69 ^{ab}	918±42 ^a	914±17 ^a	32.70	<0.001
PER	1.96±0.09 ^{bc}	2.10±0.08 ^c	1.76±0.14 ^a	1.81±0.18 ^{ab}	0.046	<0.001
EI	15,522±791 ^b	14,662±1074 ^{ab}	13,908±660 ^a	13,872±317 ^a	488.91	<0.001
EFR (%)	7.71±0.36 ^{ab}	7.50±0.31 ^a	8.67±0.68 ^b	8.42±0.92 ^b	0.246	<0.001
Carcass traits						
Dressing (%)	63.37±2.86	65.04±1.29	63.18±2.83	62.74±4.63	0.669	0.019
BM (%)	18.93±1.16 ^b	18.58±2.48 ^b	16.95±1.00 ^a	16.31±1.08 ^a	0.369	<0.001
TM (%)	12.71±0.67 ^a	13.92±0.48 ^{ab}	14.42±1.08 ^b	14.13±0.95 ^b	0.236	0.011
Visceral organ						
Liver %	2.26±0.18 ^a	2.53±0.40 ^{ab}	2.78±0.52 ^{ab}	3.09±0.62 ^b	0.116	0.010
Heart %	0.53±0.03	0.55±0.11	0.57±0.03	0.54±0.03	0.012	0.387
Gizzard %	2.63±0.47	2.71±0.20	2.65±0.53	3.10±0.54	0.105	0.440
AF %	1.36±0.56	1.55±0.22	1.40±0.33	1.57±0.49	0.101	0.907
Spleen %	0.21±0.10 ^b	0.09±0.01 ^a	0.10±0.05 ^a	0.13±0.05 ^{ab}	0.013	0.040
Bursa %	0.11±0.02	0.15±0.04	0.11±0.04	0.15±0.03	0.007	0.175

Data are presented mean ± SD. P value ≤0.05 indicates significant difference. Lowercase letter (^{a-c}) in the same row indicates significant difference at P value ≤0.05. Se-BSF= selenium-enriched black soldier fly, IBW= Initial body weight, FBW= Final body weight, ADG: Average daily gain, FI= Feed intake, FCR= Feed conversion ratio, FER= Feed efficiency ratio, PI= Protein intake, PER= Protein efficiency ratio, EI= Energy intake, ERR= Energy efficiency ratio, Dressing, BM= Breast muscle, TM= Thigh muscle, AF= Abdominal fat

Table 3. Effect of dietary Se-BSF on the blood biochemistry and immune status of broiler ducks from 0 to 49 d of age

Parameters	Control	Se-BSF (%)		Se-Yeast	SEM	P-value
		5	7.5			
Blood biochemistry						
HDL (mg/dl)	147.96±0.21 ^d	93.01±0.50 ^b	100.90±1.57 ^c	89.40±0.40 ^a	7.10	<0.001
LDL (mg/dl)	152.89±1.47 ^d	89.78±0.84 ^a	93.94±1.74 ^b	107.72±2.54 ^c	7.56	<0.001
Cholesterol (mg/dl)	166.00±2.61 ^b	90.90±0.43 ^a	88.47±2.68 ^a	88.83±1.10 ^a	10.01	<0.001
Triglyceride (mg/dl)	36.93±0.13 ^c	36.83±0.66 ^c	35.32±1.11 ^b	33.58±0.14 ^a	0.44	<0.001
Blood immune						
Ig-A (mg/ml)	1.83±0.04 ^a	6.53±0.02 ^b	13.23±0.40 ^d	12.51±0.01 ^c	1.41	<0.001
SOD (U/ml)	37.99±0.66 ^a	39.07±0.95 ^a	52.20±0.79 ^b	82.39±0.66 ^c	5.41	<0.001
Immunohistochemistry						
IL-6 (%)	19.62±1.01 ^a	23.12 ± 2.16 ^a	27.89±3.44 ^b	33.96 ± 3.17 ^c	1.35	<0.001

Data are presented mean ± SD. P value ≤ 0.05 indicates significant difference. Lowercase letter (^{a-d}) in the same row indicates significant difference at P value ≤ 0.05. HDL: High-density lipoproteins, LDL: Low-density lipoproteins, Ig-A: Immunoglobulin A, SOD: Superoxide dismutase, IL-6: Interleukin 6.

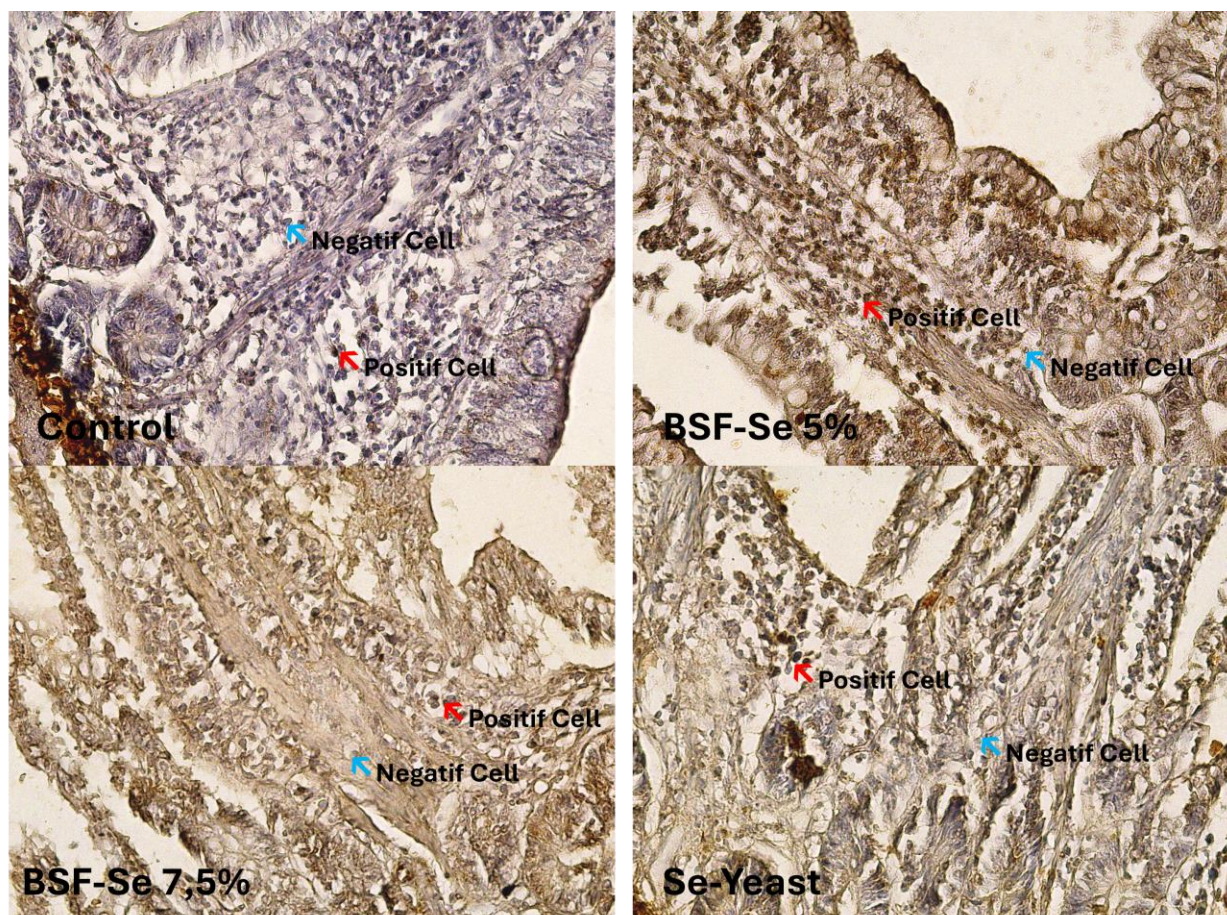


Figure 1. Immunohistochemistry of IL-6 in ileum of broiler duck treated with Se-BSF

expression calculation using ImageJ of the form percentage. Biochemical indicators are positively connected with the animals' nutritional status and can be used to evaluate the animals' general and physiological health. The dietary Se-BSF had significant differences ($P>0.001$) in serum HDL, LDL, Cholesterol, and Triglyceride compared with the control. This agrees with previous findings that feeding broilers with Se had significantly affected the plasma concentration of total cholesterol and triglyceride. (Abdel-Moneim et al. 2022) reported that birds fed diets enriched with *Spirulina platensis* (SP), selenium nanoparticles (SeNPs), or their mixtures significantly reduced blood cholesterol, triglycerides, and LDL-cholesterol. (Sun et al. 2020) Se-enriched earthworm powder (SEP) downregulated triglycerides, total cholesterol, glucose, and nitric oxide. Serum total cholesterol and triglycerides are two extensively used indices for assessing body lipid metabolism; an abnormal increase of either indicates a lipid metabolism disorder. SE-BSF reduced serum total cholesterol and triglycerides, suggesting that BSF meals enhanced with Se play an essential role in regulating body lipid metabolism and lowering blood fat of broiler duck.

In the current investigation, the dietary Se group had significantly decreased SOD level in serum compared to control. Previous research (Qiu et al. 2023) found that Se-BS supplementation improved the levels of superoxide dismutase (SOD), glutathione peroxidase (GPx), catalase (CAT), peroxidase (POD), and total antioxidant capacity (T-AOC) in plasma as compared to the broiler control group. (Sun et al. 2020) found that selenium-enriched earthworm powder with 1.0 mg Se per kg significantly improved the antioxidative enzyme activity of glutathione peroxidase and superoxide dismutase. Oxidative stress is an imbalance in the ratio of reactive oxidative species to antioxidants. The organism's antioxidant defence mechanism relies mainly on GSH-Px, SOD, and catalase to remove excess free radicals such as O_2 , H_2O_2 , and ROO_2 . Increased reactive oxygen species generation and high levels of oxidative stress on blood metabolites are associated with selenium deficiency (Liu et al. 2022).

B cell-produced antibodies and immunoglobulins are two key indications of humoral immunity. Besides its antimicrobial and antiviral properties, IgA is also involved in the "barrier" function of the body's mucosal immunity in the digestive and respiratory systems. (Dalia et al. 2018) Se supplementation from inorganic and bacterial organic sources boosted immunity in broiler chickens by increasing IgA, IgG, and IgM levels. In this investigation, the dietary Se-BSF had increasing Ig-A levels in the serum of the broiler duck at day 42. This could be explained by Se's role in protecting and activating B-lymphocyte cells, which produce immunoglobulin. Our investigation found that selenium might alter cytokine release in broiler duck ileum, which

is consistent with earlier findings. (Zhang et al. 2022) found that low selenium levels lowered IL-6 and IL-10 levels in mouse serum, but high selenium levels enhanced IL-6 content. The findings revealed that low and high selenium levels can modify immune cell composition via modulating cytokine release, creating inflammatory injury, poor immune response in the mouse spleen, and inflammatory damage to other organs.

CONCLUSION

The dietary Se-BSF 5% did not have a negative influence on the performance production of broiler ducks. These findings suggest that feeding broiler ducks dietary Se-BSF enhances their blood biochemistry and immune system, suggesting possible advantages for using Se-BSF as a feed supplement for poultry.

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REFERENCES

- Abdel-Moneim AME, Shehata AM, Selim DA, El-Saadony MT, Mesalam NM, Saleh AA. 2022. *Spirulina platensis* and biosynthesized selenium nanoparticles improve performance, antioxidant status, humoral immunity and dietary and ileal microbial populations of heat-stressed broilers. *J Therm Biol.* 104:103195. DOI:10.1016/j.jttherbio.2022.103195
- Al-Quwaie DA. 2023. The influence of bacterial selenium nanoparticles biosynthesized by *Bacillus subtilis* DA20 on blood constituents, growth performance, carcass traits, and gut microbiota of broiler chickens. *Poult Sci.* 102:102848. DOI:10.1016/j.psj.2023.102848
- Bakhshalinejad R, Hassanabadi A, Swick RA. 2019. Dietary sources and levels of selenium supplements affect growth performance, carcass yield, meat quality and tissue selenium deposition in broilers. *Anim Nutr.* 5:256–263. DOI:10.1016/j.aninu.2019.03.003
- Bami KM, Afsharmanesh M, Espahbodi M, Esmailzadeh E. 2022. Effects of dietary nano-selenium supplementation on broiler chicken performance, meat selenium content, intestinal microflora, intestinal morphology, and immune response. *J Trace Elem Med Biol.* 69:126897. DOI:10.1016/j.jtemb.2021.126897.
- Barragan-Fonseca KB, Dicke M, van Loon JJA. 2017. Nutritional value of the black soldier fly (*Hermetia illucens* L.) and its suitability as animal feed - a review. *J*

- Insects as Food Feed. 3:105–120. DOI: 10.3920/JIFF2016.0055.
- Cullere M, Schiavone A, Dabbou S, Gasco L, Zotte AD. 2019. Meat quality and sensory traits of finisher broiler chickens fed with black soldier fly (*Hermetia illucens* L.) larvae fat as alternative fat source. *Animals*. 9:1–15. DOI: 10.3390/ani9040140.
- Dalgaard TS, Briens M, Engberg RM, Lauridsen C. 2018. The influence of selenium and selenoproteins on immune responses of poultry and pigs. *Anim Feed Sci Technol*. 238:73–83. DOI:10.1016/j.anifeedsci.2018.01.020.
- Dalia AM, Loh TC, Sazili AQ, Jahromi MF, Samsudin AA. 2018. Effects of vitamin E, inorganic selenium, bacterial organic selenium, and their combinations on immunity response in broiler chickens. *BMC Vet Res*. 14:1–10. DOI: 10.1186/s12917-018-1578-x.
- Directorate General of Animal Husbandry and Animal Health. 2021. Livestock and animal health statistics 2021. Jakarta (Indones): Directorate General of Animal Husbandry and Animal Health, Ministry of Agriculture.
- Dong Z, Lin Y, Wu H, Zhang M. 2021. Selenium accumulation in protein fractions of *Tenebrio molitor* larvae and the antioxidant and immunoregulatory activity of protein hydrolysates. *Food Chem*. 334:127475. DOI:10.1016/j.foodchem.2020.127475.
- Elkhateeb F, Ghazalah AA, Abdel-Wareth AAA. 2022. Effect of selenium sources on growth performance, carcass criteria and physical meat quality of broiler chickens. *SVU-International J Agric Sci*. 4:207–217. DOI: 10.21608/svuijas.2023.186986.1265
- Ferrari L, Sele V, Silva M, Bonilauri P, De Filippo F, Selmin F, Ørnsrud R, Pinotti L, Ottoboni M. 2022. Biofortification of selenium in black soldier fly (*Hermetia illucens*) prepupae reared on seaweed or selenium enriched substrates. *J Insects as Food Feed*. 8:887–899. DOI:10.3920/JIFF2021.0153.
- Ferro C, Florindo HF, Santos HA. 2021. Selenium Nanoparticles for biomedical applications: From development and characterization to therapeutics. *Adv Healthc Mater*. 10:1–50. DOI:10.1002/adhm.202100598.
- [FAO] Food and Agriculture Organisation. 2022. Gateway to poultry production and products. 2022. [accessed year?-month?-date] <https://www.fao.org/poultry-production-products/production/poultry-species/ducks/en/>.
- Gu X, Gao C. 2022. New horizons for selenium in animal nutrition and functional foods. *Anim Nutr*. 11:80–86. DOI:10.1016/j.aninu.2022.06.013.
- Ismita J, Islam K, Mamun M, Debi M. 2022. Comparative efficacy of citric acid, *Spirulina platensis*, and their combination as alternatives to an antibiotic growth promoter on the performances of broilers. *J Adv Vet Anim Res*. 9:1. DOI: 10.5455/javar.2022.i562.
- Ismoyowati and Sumarmono. 2019. Duck Production for Food Security Duck Production for Food Security. 1st Anim Sci Food Technol Conf. DOI:10.1088/1755-1315/372/1/012070.
- Jachimowicz K, Winiarska-mieczan A, Tomaszewska E. 2022. The impact of herbal additives for poultry feed on the fatty acid profile of meat. *Animals*. 12:1054. DOI:10.3390/ani12091054.
- Khan HA, Ibrahim KE, Alrashood ST, Alamery S, Alrokayan SH, Al-harbi N, Al-mutary MG, Sobki SH, Khan I. 2020. Immunohistochemistry of IL-1 b , IL-6 and TNF- a in spleens of mice treated with gold nanoparticles. *Saudi J Biol Sci*. 27:1163–1168. DOI:10.1016/j.sjbs.2020.01.025.
- Khan MT, Niazi AS, Arslan M, Azhar M, Asad T, Raziq F, Gondal MA, Rauf M, Liaqat S, Naz S, et al. 2023. Effects of selenium supplementation on the growth performance, slaughter characteristics, and blood biochemistry of naked neck chicken. *Poult Sci*. 102:102420. DOI:10.1016/j.psj.2022.102420.
- Kurniawan D, Widodo E, Susilo A, Sjojfan O. 2024. Synthesis of selenium conjugated insects' protein in *Hermetia illucens* larvae as poultry feed. *BIO Web Conf*. 88. DOI:10.1051/bioconf/20248800004.
- Liu C, Li Y, Li H, Wang Y, Zhao K. 2022. Nano-selenium and macleaya cordata extracts improved immune functions of intrauterine growth retardation piglets under maternal oxidation stress. *Biol Trace Elem Res*. 200:3975–3982. DOI:10.1007/s12011-021-03009-1
- Qiu H, Gao S, Hou L, Li A, Zhu L qin, Dong J, Chen F. 2023. Selenium-enriched *Bacillus subtilis* improves growth performance, antioxidant capacity, immune status, and gut health of broiler chickens. *Biol Trace Elem Res*. 201:5756–5763. DOI:10.1007/s12011-023-03610-6
- Qiu K, Ma Y, Obianwuna UE, Wang J, Zhang H, Qi G, Wu S. 2021. Application of selenium conjugated to animal protein in laying hens' diet for the production of selenium-enriched eggs. *Foods*. 10. DOI:10.3390/foods10061224
- Rehman HF ur, Zaneb H, Masood S, Yousaf MS, Hayat K, Majeed KA, Zeeshan M, Ashraf S, Khan I, Khan A, Rehman H. 2022. Effect of selenium nanoparticles and mannan oligosaccharide supplementation on growth performance, stress indicators, and intestinal microarchitecture of broilers reared under high stocking density. *Anim*. 12. DOI: 10.3390/ani12212910.
- Silva VA, Clemente AHS, Nogueira BRF, De Carvalho AC, De Freitas LFVB, Ramos ADLS, Bertechini AG. 2019. Supplementation of selenomethionine at different ages and levels on meat quality, tissue deposition, and selenium retention in broiler chickens. *Poult Sci*. 98:2150–2159. DOI:10.3382/ps/pey569.
- Sun X, Yue S zhong, Qiao Y hui, Sun Z jun, Wang C, Li H fen. 2020. Dietary supplementation with selenium-enriched earthworm powder improves antioxidative ability and immunity of laying hens. *Poult Sci*. 99:5344–5349. DOI:10.1016/j.psj.2020.07.030.
- Zhang L, Wang YX, Zhou Y, Zheng L, Zhan XA, Pu QH. 2014. Different sources of maternal selenium affect selenium retention, antioxidant status, and meat quality of 56-day-old offspring of broiler breeders. *Poult Sci*. 93:2210–2219. DOI: 10.3382/ps.2013-03605.

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Zhang X, Zhang L, Xia K, Dai J, Huang J, Wang Y, Zhu G, Hu Z, Zeng Z, Jia Y. 2022. Effects of dietary selenium on

immune function of spleen in mice. J Funct Foods. 89:104914. DOI:10.1016/j.jff.2021.104914.