



## Research Paper

# The Effects of Dietary Lemongrass Oil (*Cymbopogon nardus*) and Selenium on Production Performances, Internal Organs, and Immune Organ Weights in Broiler Chickens



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How to cite this article

Darmawan A, Sumiati S, Mutia R, Latifah A, Nainggolan DK, Prana JI, et al. The Effects of Dietary Lemongrass Oil (*Cymbopogon nardus*) and Selenium on Production Performances, Internal Organs, and Immune Organ Weights in Broiler Chickens. *Archives of Razi Institute Journal*. 2025; 80(6):1591-1602. <https://doi.org/10.32598/ARI.80.6.3455>

doi

<https://doi.org/10.32598/ARI.80.6.3455>

### Article info:

Received: 12 Feb 2025

Accepted: 15 Apr 2025

Published: 01 Nov 2025

### ABSTRACT

**Introduction:** Raising broiler chickens in tropical countries faces challenges, primarily heat stress, which reduces feed intake, weakens immunity, increases infections and mortality, and impairs growth, ultimately leading to economic losses. This study evaluated the effect of dietary lemongrass oil and selenium on broiler production performance and internal organ weight.

**Materials & Methods:** A total of 288 Cobb strain broilers were allocated into 6 treatments with 6 replicates, using a completely randomized factorial design. Treatment included lemongrass oil (0 mL/kg, 2 mL/kg, 4 mL/kg) and selenomethionine (0 ppm, 0.4 ppm). Parameters measured were broiler production performances, internal organs, and immune organ weights.

**Results:** The inclusion of 4 mL/kg lemongrass oil significantly reduced ( $P<0.05$ ) feed intake, body weight, and body weight gain. The inclusion of 0.4 ppm selenomethionine significantly increased ( $P<0.05$ ) the body weight and reduced the FCR of broiler chickens. The inclusion of 4 mL/kg lemongrass oil significantly reduced ( $P<0.05$ ) the feed intake, body weight gain, and body weight, while significantly increasing ( $P<0.05$ ) proventriculus weight. The combination of 4 mL/kg lemongrass oil and 0.4 ppm selenomethionine significantly reduced ( $P<0.05$ ) the weights of the ileum and gallbladder. The treatment did not affect ( $P>0.05$ ) the weight of the ventriculus, heart, liver, duodenum, jejunum, cecum, colon, abdominal fat, thymus, lymph, and bursa of Fabricius.

**Conclusion:** The conclusion is that the inclusion of 0.4 ppm selenomethionine in the diet increases the body weight and feed efficiency, whereas inclusion of up to 4 mL/kg lemongrass oil in the diet reduces the broiler's production performances and the weight of the ileum and gallbladder.

### Keywords:

Body weight, Essential oil, Feed intake, Lemongrass, Selenomethionine

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## 1. Introduction

**B**roiler chicken rearing in tropical countries faces many obstacles that can reduce chicken productivity. The most critical constraint is heat stress due to extreme temperature and humidity outside the broiler's comfort zone, resulting in decreased feed intake and immune system, increased pathogenic bacterial infections and mortality, as well as impaired growth, and economic losses [1]. Recently, the use of natural ingredients to reduce the adverse effects of heat stress has gained growing attention, particularly essential oils. Essential oils are safe as antioxidants, antibiotic substitutes due to their antimicrobial properties, and digestive stimulants [2-4]. One of the potential essential oils is lemongrass oil (*Cymbopogon nardus*), which contains citral, citronellal, geraniol, and citronellol, exhibits antibacterial, anti-fungal, and anti-free radical properties [5]. The provision of lemongrass oil in feed can increase broiler weight gain [6], while its inclusion in drinking water has been reported to increase growth and reduce feed conversion ratio and consumption [7].

Essential oils can also be combined with antioxidant minerals such as organic selenium. Selenium plays a crucial role in minimizing oxidative stress in poultry by enhancing the activity of antioxidant enzymes, including glutathione peroxidase (GPx) and superoxide dismutase [SOD] [8]. Selenium is particularly important due to its involvement in immunity, thyroid hormone activity, and heat stress gene expression [9]. Selenium supplementation in feed has been shown to increase antioxidant activity, body weight, sperm motility, feed efficiency, villi height, and villi surface area in the broiler intestine [8, 10]. The combination of lemongrass oil and selenium is expected to have a better effect than a single administration, as they have complementary mechanisms of action. Selenium increases resistance to oxidative stress, while lemongrass oil improves digestive health and nutrient digestibility. This synergy has the potential to increase feed efficiency, support optimal growth, and improve immune response without causing negative effects on internal organs. However, no previous study has investigated the effect of their combination in broiler diets. Therefore, this study aimed to evaluate the effects of the dietary lemongrass oil and selenium on production performance, internal organs, and the immune organs weight in broiler chickens.

## 2. Materials and Methods

### 2.1. Animal and diet

A total of 288 one-day-old unsexed Cobb broiler chicks were allocated to partitioned cages using a completely randomized factorial design with 6 treatments and 6 replicates, and raised for 33 days. A total of 36 cages measuring  $1 \times 1 \times 0.5$  m, each equipped with 1 feeder and 1 drinker, were used in this study. Ambient temperature was maintained at 33 °C for the first two days and then gradually reduced by 1 °C per day until it reached 20 °C during the finisher phase. A crumbled-form diet was prepared based on the standard nutritional requirements of Cobb broiler chickens for the starter phase with a crude protein content of 22% and a metabolizable energy of 2,900 kcal/kg, and the growth phase with a crude protein content of 20% and a metabolizable energy of 2,950 kcal/kg. Diet ingredients included soybean meal, meat and bone meal, yellow corn, rice bran, crude palm oil, L-lysine, DL-methionine, salt, CaCO<sub>3</sub>, premix, selenomethionine, and lemongrass oil. The composition and nutrient content of diet treatments are presented in Table 1. Diet and drinking water were provided ad libitum.

Treatments were:

T1: Control diet (no lemongrass oil or selenomethionine)

T2: Diet containing 0.4 ppm selenomethionine without lemongrass oil

T3: Diet containing 2 mL/kg lemongrass oil without selenomethionine

T4: Diet containing 4 mL/kg lemongrass oil without selenomethionine

T5: Diet containing 2 mL/kg lemongrass oil and 0.4 ppm selenomethionine

T6: Diet containing 4 mL/kg lemongrass oil and 0.4 ppm selenomethionine

### 2.2. Production performance measurements

At the beginning of rearing, broilers were weighed to obtain initial body weight. Feed intake was calculated as the difference between the initial feed and the residual feed at the end of the week. The feed conversion ratio was derived by dividing feed intake by the body weight gain. Body weight gain was determined as the difference

**Table 1.** Composition and nutrient content of diet treatments (as fed)

Variables	Starter	Grower
Yellow corn	58	61
Rice bran	3	3
Soybean meal	26.5	26
Meat and bone meal	9	5
Crude palm oil	2	3
Raw materials (%)		
CaCO <sub>3</sub>	0.15	0.7
NaCl	0.2	0.2
Premix*	0.5	0.5
DL-methionine	0.35	0.3
L-lysine	0.3	0.3
Total	100	100
Crude protein	22.45	20.47
Metabolizable energy (Kcal/kg)	2,920.25	2,960.5
Crude fat	3.45	2.7
Crude fiber	2.7	2.72
Nutrient content (%)		
Lysine	1.34	1.23
Methionine	0.62	0.56
Cystine	0.28	0.26
Methionine+cystine	0.9	0.81
Calcium	1	0.83
Available phosphorus	0.65	0.44

\*Premix composition (per kg premix): Vitamin A 200,000 IU, vitamin D3 200,000, vitamin E 800 IU, vitamin K3 200 mg, vitamin B2 500 mg, Vitamin B6 50 mg, vitamin B12 1,200 mg, vitamin C 2,500 mg, calcium-D-pantothenate 600 mg, niacin 400 mg, choline chloride 1,000 mg, manganese 1,200, iron 2,000 mg, iodine 20 mg, zinc 10,000 mg, cobalt 20 mg, copper 400 mg.

between the final body weight and the body weight measured in the previous week.

### 2.3. Internal and immune organs weight measurements

On day 33, chickens were fed overnight with access to drinking water. Then all chickens in each replication were weighed, and 2 chickens were slaughtered with a weight close to the average weight in each replication. Slaughter was performed by cutting the trachea, jugular vein, carotid artery, and esophagus simultaneously at the front of the neck to ensure complete bleeding and animal

welfare. The chickens were immediately de-feathered, and the internal organs were removed. The weight of the proventriculus, ventriculus, heart, liver, abdominal fat, gallbladder, pancreas, intestinal, thymus, lymph, and bursa of Fabricius was measured using a digital balance with 0.1 g precision. The weight percentage of internal and immune organs was calculated by dividing organ weight by live weight, multiplied by 100.

### 2.4. Data analysis

Data were analyzed using analysis of variance (ANOVA) with the IBM SPSS Statistics software, version 22.

**Table 2.** Impact of lemongrass oil and selenomethionine inclusion in the diet on body weight (g/bird) of broiler chickens

Period (d)	Lemongrass Oil (mL)	Selenomethionine		Mean
		0 ppm	0.4 ppm	
1-14	0	300.55±26.13	297.9±13.15	299.23±19.55
	2	282.75±16.69	292.75±10.92	287.75±14.3
	4	286.35±17.99	271.65±10.84	279±16
	Mean	289.88±4.34	287.43±4.34	
15-33	0	1270.9±92.7	1307.11±114.22	1289±99.91
	2	1186.94±84.82	1244.1±99.226	1215.52±92.11
	4	1127.37±24.41	1158.01±56.64	1142.69±44.18
	Mean	1195.07±21.73	1236.41±21.73	
1-33	0	1653.9±114.74	1745.52±159.65	1699.71±139.68
	2	1569±91.22	1604.7±145.86	1586.85±116.23
	4	1468.62±17.82	1541.2±64.75	1504.91±58.89
	Mean	1563.84±28.43	1630.47±85.44	

A significance level of  $P<0.05$  was applied, and Duncan's multiple range test was conducted for further comparison.

### 3. Results

#### 3.1 Production performances

The inclusion of selenomethionine in broiler chickens' diet significantly increased ( $P<0.05$ ) body weight and reduced ( $P<0.05$ ) feed conversion ratio (Tables 2 and 3). Conversely, the inclusion of up to 4 mL/kg lemongrass oil significantly reduced ( $P<0.05$ ) body weight, body weight gain, and feed intake. Meanwhile, dietary supplementation with 0.4 ppm selenomethionine did not affect body weight gain (Tables 2, 4, and 5).

#### 3.2 Internal and immune organs

The inclusion of 4 mL/kg lemongrass oil significantly increased ( $P<0.05$ ) proventriculus weight, however, the combination of 4 mL/kg lemongrass oil and 0.4 ppm selenomethionine significantly reduced ( $P<0.05$ ) the weight of the ileum and gallbladder (Table 6). The treatments did not affect the weight of the heart, liver, abdominal fat, gallbladder, pancreas, duodenum, jejunum, cecum, colon, thymus, lymph, bursa of Fabricius (Table 7).

### 4. Discussion

#### 4.1. Production performances

In the current study, the inclusion of selenomethionine in broiler chickens' diet significantly improved body weight and reduced the feed conversion ratio. Conversely, lemongrass oil inclusion at levels up to 4 mL/kg decreased body weight and feed intake. Vieira et al. [11] previously reported that incorporating selenomethionine into corn-soybean meal diets significantly enhanced broiler growth performance, carcass, and breast meat yields. Indeed, Selenomethionine exhibits high bioavailability since its structure resembles an amino acid that can be easily absorbed through the same mechanism as methionine in the chicken digestive tract [12]. This organic structure allows it to be directly utilized in various metabolic processes or stored in body tissues as a selenium reserve, which is not the case with inorganic selenium [11]. This makes selenomethionine more efficient in supporting broiler health and productivity.

Selenium is an essential mineral that improves feed efficiency and promotes body weight gain in broilers through its antioxidant mechanisms. It contributes to the formation of GPx, which protects cells from oxidative damage caused by free radicals, thereby maintaining tissue health, enhancing metabolic function, and sup-

**Table 3.** Impact of lemongrass oil and selenomethionine inclusion in the diet on the feed conversion ratio of broiler chickens

Period (d)	Lemongrass Oil (mL)	Selenomethionine		Mean
		0 ppm	0.4 ppm	
1-14	0	1.38±0.09	1.45±0.05	1.41±0.08
	2	1.51±0.03 <sup>c</sup>	1.41±0.07	1.46±0.08
	4	1.5±0.11 <sup>c</sup>	1.54±0.07 <sup>c</sup>	1.52±0.09 <sup>c</sup>
	Mean	1.46±0.02	1.46±0.02	
15-33	0	1.77±0.19	1.64±0.21	1.71±0.2
	2	1.73±0.24	1.62±0.12	1.67±0.19
	4	1.93±0.24	1.63±0.06	1.78±0.23
	Mean	1.81±0.05	1.63±0.05	
1-33	0	1.67±0.16	1.59±0.17	1.63±0.16
	2	1.68±0.19	1.56±0.07	1.62±0.15
	4	1.83±0.21	1.61±0.03	1.72±0.18
	Mean	1.73±0.04	1.59±0.04	

**Table 4.** Impact of lemongrass oil and selenomethionine inclusion in the diet on feed intake (g/bird) of broiler chickens

Period (d)	Lemongrass Oil	Selenomethionine		Mean
		0 ppm	0.4 ppm	
1-14	0	515.52±39.2	536.54±27.84	526.03±33.92
	2	519.58±38.8	506.88±35.74	513.23±35.8
	4	521.44±41.43	511.26±28.58	516.35±33.98
	Mean	518.85±9.21	518.23±9.21	
15-33	0	2172.72±182.55	2160.88±220.01	2166.8±190.69
	2	2019.4±159.76	1925.44±111.11	1972.42±138.86
	4	2069.26±250.64	1894.58±95.69	1981.92±201.16
	Mean	2087.13±46.14	1993.63±46.14	
1-33	0	2688.24±220.44	2697.42±239.84	2692.83±217.22
	2	2538.98±126.83	2432.32±143.7	2485.65±139.6
	4	2590.7±268.13	2405.84±110.34	2498.27±216.46
	Mean	2605.97±207.66	2511.86±210.68	

**Table 5.** Impact of lemongrass oil and selenomethionine inclusion in the diet on body weight gain (g/bird) of broiler chickens

Period (d)	Lemongrass Oil (mL)	Selenomethionine		Mean
		0 ppm	0.4 ppm	
1-14	0	374.82±37.08	370.76±21.8	372.79±28.75
	2	344.42±23.07	360.64±18.2	352.53±21.38
	4	349.24±30.84	333.42±15.95	341.33±24.6
	Mean	356.16±6.6	354.94±6.6	
15-33	0	1232.5±83.26	1327.92±141.01	1280.21±120.2
	2	1178±81.64	1196.6±132.86	1187.3±104.42
	4	1072.22±37.6	1162.9±53.5	1117.56±64.69
	Mean	1160.91±24.81	1229.14±24.81	
1-33	0	1607.32±114.02	1698.68±159.6	1653±139.35
	2	1522.42±90.82	1557.24±145.09	1539.83±115.58
	4	1421.46±18.34	1496.32±64.39	1458.89±59.57
	Mean	1517.07±111.14	1584.08±148.91	

porting growth [13]. Selenium also supports immune function, helping prevent stress and disease that impair feed efficiency [8]. In addition, selenium plays a role in thyroid function by facilitating the conversion of T4 to T3 hormones, which regulate energy and protein metabolism and accelerate muscle tissue formation [14]. These mechanisms enable broilers to utilize nutrients more effectively for growth and production. Contrary to initial expectations, lemongrass oil inclusion reduced feed intake and body weight. These findings align with Nogueira et al. [15], who reported that lemongrass oil supplementation decreased body weight, feed intake, and feed efficiency. The effects of essential oils are dose-dependent; high dietary inclusion levels can become toxic and impair growth [16]. Excessive essential oils may irritate the intestinal mucosa, induce inflammation, and reduce nutrient absorption [17]. Previous studies also reported that citronella and  $\beta$ -citronellol essential oils decreased appetite by increasing sympathetic nerve activity [18]. Essential oil components such as citral, limonene,  $\beta$ -citronellol, and thymol have been shown to reduce appetite by altering mRNA expression of cocaine- and amphetamine-regulated transcript and proopiomelanocortin, modulating leptin release, and influencing sympathetic and parasympathetic nerve activity [19]. Volatile compounds such as limonene and  $\beta$ -citronellol may also provide direct signals to the brain's emotion centers (e.g. amygdala and hippocampus), reducing feed

cravings [20]. Furthermore, some essential oils affect digestive hormone release, including cholecystokinin and glucagon-like peptide-1, which promote satiety [21].

Selenium is an essential mineral that contributes significantly to improving feed efficiency and promoting body weight gain in broiler chickens through mechanisms as an antioxidant. Selenium contributes to the formation of the enzyme GPx, which protects cells from oxidative damage due to free radicals, thus maintaining tissue health, improving metabolic function, and supporting broiler growth [13]. Selenium is reported to support immune function, which helps prevent stress and diseases that contribute to decreased feed efficiency [8]. In addition, selenium also plays a role in thyroid function, supporting the conversion of T4 to T3 hormones that regulate energy and protein metabolism, and accelerating the formation of muscle tissue [14]. Thus, broilers can utilize nutrients in feed more effectively for growth and production.

The inclusion of lemongrass oil in this study did not conform to the initial prediction that lemongrass oil would have a positive effect on broiler performance. On the contrary, the inclusion of lemongrass oil reduced the feed intake and body weight of broilers. These results were in line with the study of Nogueira et al [15] that the utilization of lemongrass oil in the diet reduced body

**Table 6.** Impact of lemongrass oil and selenomethionine inclusion in the diet on the internal organs and abdominal fat weight of broiler chickens

Parameter (%)	Lemongrass Oil (mL)	Selenomethionine		Mean
		0 ppm	0.4 ppm	
Ventriculus	0	1.42±0.26	1.32±0.08	1.37±0.18
	2	1.42±0.04	1.58±0.42	1.5±0.29
	4	1.44±0.18	1.46±0.21	1.45±0.18
	Mean	1.43±0.06	1.45±0.06	
Proventriculus	0	0.37±0.03	0.34±0.07	0.35±0.05 <sup>a</sup>
	2	0.43±0.07	0.43±0.1	0.43±0.08 <sup>ab</sup>
	4	0.51±0.1	0.43±0.06	0.47±0.08 <sup>b</sup>
	Mean	0.44±0.02	0.4±0.02	
Liver	0	2.12±0.94	2.09±0.74	2.11±0.08
	2	2.01±0.23	2.23±0.37	2.12±0.31
	4	2.02±0.2	2.03±0.16	2.02±0.16
	Mean	2.05±0.06	2.11±0.06	
Heart	0	0.45±0.78	0.46±0.06	0.45±0.06
	2	0.45±0.2	0.49±0.09	0.46±0.06
	4	0.45±0.06	0.45±0.05	0.44±0.05
	Mean	0.44±0.01	0.46±0.01	
Gallbladder	0	0.06±0.01 <sup>a</sup>	0.07±0.02 <sup>ab</sup>	0.06±0.01
	2	0.07±0.02 <sup>ab</sup>	0.07±0.01 <sup>ab</sup>	0.07±0.01
	4	0.12±0.06 <sup>b</sup>	0.04±0.01 <sup>a</sup>	0.07±0.05
	Mean	0.08±0.01	0.05±0.01	
Pancreas	0	0.24±0.04	0.27±0.02	0.25±0.03
	2	0.25±0.05	0.28±0.05	0.26±0.05
	4	0.22±0.04	0.28±0.07	0.24±0.06
	Mean	0.23±0.01	0.27±0.01	
Duodenum	0	0.54±0.07	0.48±0.05	0.51±0.06
	2	0.52±0.14	0.48±0.1	0.5±0.11
	4	0.57±0.26	0.51±0.06	0.54±0.18
	Mean	0.54±0.04	0.49±0.04	

Parameter (%)	Lemongrass Oil (mL)	Selenomethionine		Mean
		0 ppm	0.4 ppm	
Jejunum	0	0.99±0.14	0.91±0.11	0.95±0.12
	2	0.97±0.2	1.02±0.21	0.99±0.19
	4	1.02±0.22	1.08±0.38	1.05±0.29
	Mean	0.99±0.06	1.01±0.06	
Ileum	0	0.95±0.09 <sup>b</sup>	0.82±0.03 <sup>ab</sup>	0.88±0.09
	2	0.78±0.11 <sup>ab</sup>	1.01±0.19 <sup>b</sup>	0.89±0.19
	4	0.88±0.23 <sup>b</sup>	0.59±0.21 <sup>a</sup>	0.74±0.25
	Mean	0.87±0.04	0.81±0.04	
Cecum	0	0.38±0.02	0.32±0.04	0.35±0.04
	2	0.35±0.14	0.29±0.02	0.32±0.1
	4	0.31±0.01	0.33±0.07	0.32±0.04
	Mean	0.34±0.02	0.31±0.02	
Colon	0	0.11±0.01	0.11±0.02	0.11±0.02
	2	0.11±0.05	0.1±0.01	0.11±0.03
	4	0.12±0.04	0.09±0.05	0.1±0.05
	Mean	0.11±0.01	0.1±0.01	
Abdominal fat	0	1.43±0.26	1.68±0.26	1.55±0.28
	2	1.4±0.71	1.53±0.5	1.46±0.57
	4	1.6±0.16	1.42±0.15	1.51±0.17
	Mean	1.47±0.1	1.54±0.1	

weight, feed intake, and feed efficiency. Indeed, the effects of essential oils depended on the dose administered, where inclusion at high doses in the diet became toxic and impaired chicken growth [16]. Furthermore, high levels of essential oils could irritate the intestinal mucosal layer and induce inflammation, eventually reducing nutrient absorption [17]. Moreover, previous studies reported that the utilization of citronella and  $\beta$ -citronellol essential oils decreased appetite. Inhalation of these oils increased sympathetic nerve activity associated with reduced feed intake [18]. Essential oil components such as citral, limonene,  $\beta$ -citronellol, and thymol decreased appetite through changes in cocaine and proopiomelanocortin or amphetamine-regulated transcript mRNA expression, which promote satiety, modulation of leptin release, and sympathetic and parasympathetic nerve activity [19]. Reportedly, volatile compounds such as

limonene and  $\beta$ -citronellol provide direct signals to the brain's emotion centers (e.g. amygdala and hippocampus), which may reduce feed cravings [20]. In addition, some essential oils may affect the release of digestive hormones such as cholecystokinin and glucagon-like peptide-1, which promote satiety [21].

#### 4.2. Internal and immune organs weight

The inclusion of 4 mL/kg lemongrass oil significantly increased the proventriculus weight, however, the combination of 4 mL/kg lemongrass oil and 0.4 ppm selenomethionine significantly decreased the weight of the ileum and gallbladder. The treatments did not impact the weight of the immune organs (bursa of Fabricius, thymus, lymph). The inclusion of 4 mL/kg lemongrass oil in the diet increased the weight of the proventriculus,

**Table 7.** Impact of lemongrass oil and selenomethionine inclusion in the diet on immune organ weight of broiler chickens

Parameter (%)	Lemongrass Oil (mL)	Selenomethionine		Mean
		0 ppm	0.4 ppm	
Thymus	0	0.18±0.14	0.29±0.02	0.23±0.11
	2	0.28±0.12	0.29±0.1	0.28±0.1
	4	0.27±0.09	0.28±0.11	0.27±0.09
	Mean	0.23±0.03	0.28±0.03	
Lymph	0	0.13±0.03	0.11±0.04	0.11±0.03
	2	0.15±0.04	0.16±0.06	0.15±0.05
	4	0.1±0.01	0.1±0.05	0.09±0.03
	Mean	0.12±0.01	0.12±0.01	
Bursa of Fabricius	0	0.16±0.06	0.18±0.02	0.17±0.04
	2	0.15±0.07	0.19±0.02	0.16±0.05
	4	0.13±0.03	0.16±0.05	0.14±0.04
	Mean	0.14±0.01	0.17±0.01	

possibly due to the bioactive compounds content, such as citral and geraniol, which have stimulating properties on the digestive tract. In the proventriculus, these compounds probably stimulate the secretion of hydrochloric acid and pepsinogen, which have also been proven to stimulate the secretion of protease, amylase, and lipase in the intestinal tract of poultry [22].

The combination of lemongrass oil inclusion and selenomethionine in the diet decreased ileal weight, indicating physiological changes reflecting reduced digestive activity attributed in part to lower feed intake in the lemongrass oil inclusion treatment. The decrease in feed intake reduced the amount of substrate passing through the ileum, thereby reducing the workload and growth of ileal tissue. Similarly, a previous study reported a decrease in the intestinal weight of broiler chickens fed lemongrass oil and essential oil blends due to reduced intestinal wall thickness [23]. On the other hand, selenium acts as an antioxidant through selenoprotein enzymes such as GPx, which protects cells from oxidative stress and improves the efficiency of digestion and nutrient absorption [8]. This positive effect may reduce the workload of the ileum and lead to a lower weight. It can be attributed to the increased feed efficiency with selenomethionine addition in this study.

The combination of lemongrass oil and selenomethionine decreased gallbladder weight in broilers due to the synergistic effect of these two ingredients in optimizing lipid metabolism and liver function. Selenium improved metabolic efficiency through its role as a component of selenoproteins, which function as antioxidants and regulators of lipid metabolism in the liver [24]. On the other hand, lemongrass oil's bioactive compounds have hypocholesterolemic properties, which can inhibit cholesterol synthesis in the liver [25], ultimately reducing the need for bile secretion. This combination results in decreased liver workload and reduced bile volume, thereby lowering gallbladder weight.

According to our study, the inclusion of 0.4 ppm selenomethionine in the diet increases body weight and feed efficiency without impairing the immune organs' weight (thymus, lymph, bursa of Fabricius). The inclusion of up to 4 mL/kg lemongrass oil in the diet reduces broiler performance and the weight of the ileum and gallbladder.

#### 4.2. Internal and immune organ weights

The inclusion of 4 mL/kg lemongrass oil significantly increased proventriculus weight, whereas the combination of 4 mL/kg lemongrass oil and 0.4 ppm selenomethionine significantly decreased ileum and gallbladder

weights. No significant effects were observed on immune organs (bursa of Fabricius, thymus, lymph nodes).

The increase in proventriculus weight may be attributed to bioactive compounds such as citral and geraniol, which stimulate digestive tract activity. In the proventriculus, these compounds likely enhance secretion of hydrochloric acid and pepsinogen, and have been shown to stimulate protease, amylase, and lipase secretion in the poultry intestine [22].

The reduction in ileal weight observed with the combination treatment suggests physiological changes reflecting reduced digestive activity, partly due to lower feed intake. Reduced substrate passage through the ileum decreases tissue workload and growth. Similar findings were reported by previous studies, where lemongrass oil and essential oil blends reduced intestinal weight due to decreased wall thickness [23]. Selenium, acting as an antioxidant via selenoproteins such as GPx, protects cells from oxidative stress and improves digestion and nutrient absorption efficiency [8]. This effect may reduce ileal workload, contributing to lower tissue weight.

The decrease in gallbladder weight with combined lemongrass oil and selenomethionine supplementation may reflect synergistic effects on lipid metabolism and liver function. Selenium improves metabolic efficiency through its role in selenoproteins, which act as antioxidants and regulators of hepatic lipid metabolism [24]. Meanwhile, lemongrass oil bioactive compounds possess hypocholesterolemic properties, inhibiting cholesterol synthesis in the liver [25]. Reduced cholesterol synthesis lowers bile secretion requirements, decreasing gallbladder workload and weight.

## 5. Conclusion

Overall, the inclusion of 0.4 ppm selenomethionine improved body weight and feed efficiency without impairing immune organ weights. In contrast, inclusion of up to 4 mL/kg lemongrass oil reduced broiler performance and decreased ileum and gallbladder weights.

## Ethical Considerations

### Compliance with ethical guidelines

The authors declare that all ethical standards were adhered to during the animal experiments and manuscript preparation.

### Data availability

Data are available from the corresponding author upon reasonable request.

### Funding

This study was funded by the [IPB University](#), Bogor, Indonesia, through the Young Lecturer Research Scheme (Code: 23409/IT3/PT.01.03/P/B/2024).

### Authors' contributions

Conceptualization, validation, data analysis, and writing the original draft: Arif Darmawan, Sumiati, and Rita Mutia; Investigation and data collection: Adinda Najwa Humaira, Alya Latifah, Dodi Kurniawan Nainggolan, and Juniasa Isnanti Prana; Analysis: Alya Latifah, Dodi Kurniawan Nainggolan, and Juniasa Isnanti Prana; Visualization: Adinda Najwa Humaira; Review and editing: Adinda Najwa Humaira, Arif Darmawan, Sumiati, and Rita Mutia; Final approval: All authors.

### Conflict of interest

The authors declared no conflict of interest.

### Acknowledgements

The authors acknowledge the Poultry Nutrition Laboratory for providing research facilities.

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