

Research Article



Interactive Effects of Chick Hatch Weight and Dietary Antioxidant Supplementation on Physiological, Hematological, and Intestinal Responses in Broilers

BAHRI SYAMSURYADI^{1,3}, EKO WIDODO², LILIK EKA RADIATI², SUYADI SUYADI^{2*}

¹Doctoral Program of Animal Science, Faculty of Animal Science, Brawijaya University Jl. Veteran, Malang 65145, Jawa Timur, Indonesia; ²Department of Animal Science, Faculty of Animal Husbandry, Brawijaya University Jl. Veteran, Malang 65145, Jawa Timur, Indonesia; ³Program of Animal Science, Faculty of Agriculture, Universitas Muhammadiyah Sinjai Jl. Teuku Umar, no. 8, Sinjai Utara, Sulawesi Selatan, Indonesia.

Abstract | This study aimed to evaluate the interaction effects between initial body weight of day-old broiler chicks (DOC) and dietary antioxidant supplementation (Vitamin E (VE), Selenium (Se), and their combination) on physiological responses, growth performance, hematological profile, and intestinal morphology. A 3×3 factorial completely randomized design was used, involving 288 Cobb 500 broilers grouped by DOC weight (light: 29–33 g, medium: 34–38 g, heavy: 39–43 g) and three antioxidant treatments (Vitamin E, Selenium, and VE+Se combination). The birds were reared for 35 days under uniform management conditions. Results showed a highly significant interaction ($P < 0.05$) between DOC weight and antioxidant type on feed intake and weight gain during the starter phase, with the highest performance observed in heavy DOC chicks receiving the VE+Se combination. Physiological responses, including panting frequency and heart rate, were significantly affected by antioxidant treatment ($P < 0.05$), with Se supplementation alone resulting in the lowest heart rate. Hematological parameters (erythrocytes, hematocrit, and hemoglobin) were not significantly affected ($P > 0.05$), although numerically higher values were observed in medium and heavy DOC groups. Leukocyte profiles were also not significantly influenced ($P > 0.05$), but VE+Se supplementation tended to increase monocyte and lymphocyte counts while reducing heterophils. Histological analysis of the intestine showed a significant interaction ($P < 0.05$) on apical villus width, with the VE+Se group exhibiting more favorable villus morphology, indicating improved nutrient absorption. Overall, the combination of heavier DOC and VE+Se supplementation was effective in optimizing broiler performance and physiological resilience.

Keywords | DOC weight, Vitamin E, Selenium, Productivity, Haematology

Received | April 26, 2025; **Accepted** | August 05, 2025; **Published** | September 17, 2025

***Correspondence** | Suyadi, Department of Animal Science, Faculty of Animal Husbandry, Brawijaya University Jl. Veteran, Malang 65145, Jawa Timur, Indonesia; **Email:** suyadi@ub.ac.id

Citation | Syamsuryadi B, Widodo E, Radiati LE, Suyadi (2025). Interactive effects of chick hatch weight and dietary antioxidant supplementation on physiological, hematological, and intestinal responses in broilers. J. Anim. Health Prod. 13(3): 845-853.

DOI | <https://dx.doi.org/10.17582/journal.jahp/2025/13.3.845.853>

ISSN (Online) | 2308-2801



Copyright: 2025 by the authors. Licensee ResearchersLinks Ltd, England, UK.

This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

INTRODUCTION

In the poultry industry, specifically in broiler chicken production, achieving optimal performance at a certain age is very important. During production, one key factor affecting broiler performance is the weight of day-old chick (DOC) or newly hatched chicken. The weight can

be an early indicator of broiler chicken production's growth potential, survival rate, and efficiency (Cowieson and Parsons, 2024). DOC with higher weight generally shows better growth potential, a stronger immune system, and more optimal adaptability to environmental conditions (Maqsood *et al.*, 2025a; Syamsuryadi *et al.*, 2025).

The weight of DOC varies based on several factors such as the age of the parent's age and the weight of eggs. Eggs produced by older breeders typically result in chickens with higher hatching weight compared to those laid by younger parents (Tona *et al.*, 2004; Calik *et al.*, 2022a). According to Maqsood *et al.* (2025a), heavier chick at hatching achieved higher final weight than DOC with lower hatching weight, with an average final weight increase of 8-13 g for every 1 g increase.

Smaller DOC is more susceptible to oxidative stress and has a larger surface area-to-volume ratio, which causes faster heat loss. In this condition, DOC increases the metabolism to produce more energy to maintain body temperature, which leads to increased free radical production due to higher metabolism. In addition, smaller DOC has low endogenous antioxidant reserves, hence, it has difficulty keeping up with the surge in free radical production (Alahmar *et al.*, 2023).

Oxidative stress occurs when there is an imbalance between the production of free radicals or reactive oxygen species (ROS) and the body's antioxidant capacity to neutralize them. It can trigger metabolic disorders, such as liver and heart failure, and sudden death due to circulatory system failure (Emami *et al.*, 2020; He *et al.*, 2021). Metabolic problems such as stress and sudden death are associated with an insufficient oxygen supply in metabolism (Muwakhid *et al.*, 2025). Therefore, the energy supply is not comparable to the organ's energy needs (Surai *et al.*, 2023).

One effective strategy to mitigate oxidative damage in broiler chickens is the supplementation of antioxidants through the diet. Vitamin E (VE) is a major exogenous antioxidant that is fat-soluble and widely used in poultry feed. As a lipophilic antioxidant, VE protects polyunsaturated fatty acids in cell membranes from lipid peroxidation, thereby preserving membrane integrity and enhancing the birds' resistance to oxidative stress (Liu *et al.*, 2022). In addition, Selenium (Se) plays a critical role in the antioxidant defense system of poultry. This essential trace mineral acts as a cofactor for the enzyme glutathione peroxidase (GPx), which neutralizes peroxides and free radicals into non-toxic compounds. Adequate Se intake supports GPx activity and demonstrates a synergistic effect with VE in reducing cellular oxidative damage (Surai *et al.*, 2023).

The combined supplementation of VE and Se has been shown to provide greater physiological benefits than either antioxidant alone. Uzochukwu *et al.* (2025) reported that the combination significantly improved body weight and enhanced the morphology of intestinal villi in broiler chickens. This synergistic effect not only enhances growth performance but also promotes nutrient absorption and intestinal health. Therefore, a precision nutrition approach

using combined VE and Se supplementation represents a promising strategy for improving physiological resilience and production efficiency in broiler chickens.

There is currently no available information on the response of broiler chickens with different DOC weights when antioxidants are added to the feed. Therefore, a study is needed to investigate the relationship between DOC weight and dietary antioxidant supplementation in relation to nutrient absorption efficiency and immune system function. This study aims to evaluate how the weight of newly hatched chicks, combined with antioxidant supplementation, affects hematological profile, intestinal architecture, and the overall productive performance of broiler chickens.

MATERIALS AND METHODS

STUDY PROCEDURE

STUDY MODEL, FEED, AND MAINTENANCE

A total of 288 day-old Cobb 500 broiler chicks were obtained from a commercial hatchery; all were vaccinated against Newcastle Disease (ND) and Infectious Bursal Disease (IBD). The chicks were then housed in 36 cages, each measuring 100 cm × 100 cm with a height of 75 cm. Each cage contained 8 chicks and was equipped with a 5-watt incandescent lamp, as well as feeding and drinking facilities. The chicks were divided into groups based on DOC weight categories according to commercial standards: light (29–33 grams), medium (34–38 grams), and heavy (39–43 grams). These groups were further assigned to different dietary treatments, including feed supplemented with Vitamin E (250 mg/kg), Selenium (1 mg/kg), and a combination of Vitamin E (250 mg/kg) plus Selenium (1 mg/kg).

Table 1: Composition and calculated analysis of the starter and finisher diet.

Nutrients	Starter diet	Finisher diet
Water content	14%	14%
Crude protein	23-24 %	20-22 %
Crude fat	5%	5%
Crude fiber	4%	5%
Ash	8%	8%
Calcium	0.8-1 %	0.8-1.1 %
Phosphorus	0.5 %	0.5 %
Alphatoxin	40 µg/kg	50 µg/kg
Amino acids		
Lysine	1.3 %	1.2 %
Methionine	0.5 %	0.45 %
Methionine+ Cystine	0.9 %	0.8 %
Tryptophan	0.2 %	0.19 %
Threonine	0.8 %	0.75 %

Source: Analysis Results of PT. Perkasa Agung Sejati

According to the study model, the feed used was a commercial ration supplemented with antioxidants. During the starter phase (days 1–14), the feed was administered in granulated (crumble) form, while in the finisher phase (days 15–35), the pellet-form feed was used. The composition of feed is presented in Table 1.

Before being slaughtered at the age of 35 days, chickens were fasted for 10 hours, and then randomly selected in each treatment unit for weighing. The blood sample was obtained before slaughtering using a syringe in the brachial vein and transferred to a vacuum tube containing ethylene diamine tetraacetic acid (EDTA) anticoagulant.

Chicken slaughtering was carried out in a halal manner, and examinations were performed to determine abnormalities in the digestive tract. Subsequently, 2 cm of the duodenum, jejunum, and ileum are cut to make intestinal histomorphology preparations using hematoxylin and eosin staining.

MEASURED VARIABLES

PHYSIOLOGICAL RESPONSE

Heart rate frequency was measured by placing a stethoscope on the left side of the broiler's chest to listen to the heartbeat for one minute (Hartono *et al.*, 2002). Respiratory rate was determined by counting thoracic movements for 30 seconds (Yousef, 1985). Breathing is a physical process involving the exchange of gases, where carbon dioxide (CO₂) is expelled and oxygen (O₂) is absorbed. Rectal temperature was recorded by inserting a digital thermometer approximately one-third of its length into the broiler's rectum (Hartono *et al.*, 2002).

GROWTH PERFORMANCE

DOC was weighed for each treatment group at 1, 15, and 35 days of age. Growth performance indicators included average feed consumption, body weight gain, and feed conversion ratio during the starter and finisher phases (Noruzi *et al.*, 2023).

HEMATOLOGY

Blood sampling was carried out randomly from each treatment unit, using a disposable syringe through the brachial vein, then placed in a tube containing EDTA as an anticoagulant. The tube was shaken slowly and stored at a low temperature (18°C) to prevent blood clotting. The cooling box stores blood to be examined in the laboratory, and a blood profile examination is conducted according to the procedure (Charde *et al.*, 2011).

INTESTINAL HISTOLOGY

A total of 36 broiler chickens were slaughtered at 35 days of age for intestinal histological analysis. Sections of the

duodenum, jejunum, and ileum (each 2 cm in length) were collected. Intestinal samples were stained with hematoxylin and eosin, then fixed using 10% formalin and dehydrated using graded alcohol from 70% to 100%. Alcohol was cleaned from the tissue using xylol I, II, and III, and the tissue was infiltrated using liquid paraffin. After the tissue solidified, the tissue was cut using a microtome with a thickness of 5 µm, and placed in a water bath at a temperature of 50°C for 15 minutes. Furthermore, it was stained with hematoxylin-eosin. Histological preparations in slides were observed under a light microscope equipped with a computer-assisted OptiLab. Under a 10x magnification ocular lens and a 4x magnification objective lens, the preparation was photographed for measurement of villi height and width, crypt depth (Prakatur *et al.*, 2019), basal width, and intestinal surface area (Noruzi *et al.*, 2022).

STATISTICAL ANALYSIS

Statistical model of Completely Randomized Design (CRD) with a factorial pattern of 3 x 3 with 4 replications (Gasperz, 1991) with the mathematical model used is:

$$Y_{ijk} = \mu + a_i + b_j + (ab)_{ij} + \epsilon_{ijk}$$

$$i = 1, 2, 3 ; j = 1, 2, 3 ; k = 1, 2, 3, 4$$

Where; Y_{ijk}: Response of broiler chicken in the k-th trial receiving the combination of treatments ij (the i-th level of the treatment factor (hatch weight) and the j-th level of antioxidant treatment factor (Vitamin E, Selenium, and Vitamin E + Selenium)). µ: General average value (overall average of observations). a_i: Additive effect of the i-th level of the factor (DOC weight). b_j: Additive effect of the j-th level of antioxidant factor. (ab)_{ij}: Interaction effect between the i-th hatch weight factor and the j-th antioxidant treatment. ε_{ijk}: Experimental error effect in the k-th trial receiving the ij treatment combination.

RESULT

PERFORMANCE

As shown in Table 2, during the starter phase, a significant interaction between DOC weight and antioxidant supplementation affected both feed intake and body weight gain (P<0.05). Heavier chicks (39–43 g) supplemented with Vitamin E and Selenium (VE+Se) showed the best performance, likely due to greater nutrient reserves and improved antioxidant defense. Conversely, lighter chicks (29–33 g), especially without antioxidant support, had the lowest intake and gain, indicating higher susceptibility to oxidative stress.

Although no significant interaction was found in the finisher phase, light chicks receiving VE+Se achieved final

Table 2: Effect of DOC weight and dietary antioxidant supplementation on the performance of broiler chickens.

Treatment	Starter			Finisher			
	FI (grams)	BWG (grams)	FCR	FI (grams)	BWG (grams)	FCR	FBW (gram)
DOC weight (A)							
Light	613.36±40.39	522.33±37.26	1.17±0.02	3033.01±102.65	2027.39±111.71	1.50±0.066	2063.6±102.71
Medium	627.09±17.65	528.56±23.76	1.19±0.05	3003.43±105.70	2046.28±111.67	1.47±0.057	2046.9±88.21
Heavy	632.14±36.06	535.22±20.09	1.18±0.05	2944.66±125.50	2047.50±74.17	1.44±0.053	2029.7±125.46
p-value	0.327	0.461	0.761	0.260	0.877	0.089	0.805
Antioxidants (B)							
Vit_E	624.90±45.66	521.22±32.05	1.20±0.03	2956.43±104.50	2010.83±111.48	1.47±0.063	2011.6±129.04
Selenium	620.13±30.66	534.44±25.67	1.16±0.04	2985.21±148.03	2018.44±97.04	1.48±0.071	2051.8±94.06
Vit_E + Selenium	627.56±20.88	530.44±25.71	1.18±0.05	3039.46±71.95	2091.89±66.20	1.45±0.042	2076.8±82.33
p-value	0.838	0.427	0.115	0.303	0.154	0.550	0.454
p-value(AxB)	0.010	0.006	0.105	0.515	0.252	0.172	0.506

Note: DOC weight: light (30–34 grams), medium (35–39 grams), heavy (40–45 grams); Vitamin E (250 mg/kg), Selenium (1 mg/kg), Vitamin E + Selenium (250 mg/kg + 1 mg/kg); FI (Feed Intake), BWG (Body Weight Gain), FBW (Final Body Weight), FCR (Feed Conversion Ratio).

Table 3: Effect of DOC weight and dietary antioxidant supplementation on the physiological parameters of broiler chickens.

Treatment	Starter			Finisher		
	Rectal temperature (%)	Panting (times/minute)	Heart rate (times/minute)	Rectal temperature (%)	Panting (times/minute)	Heart Rate (times/minute)
DOC Weight (A)						
Light	40.82±0.35	152.72±11.01	159.50±10.85	41.27±0.529	171.31±15.25	192.33±10.73
Medium	40.80±0.25	152.97±13.50	161.04±9.33	41.28±0.351	172.19±13.16	196.39±2.68
Heavy	40.83±0.30	155.81±14.30	155.72±10.61	41.33±0.463	157.89±21.48	195.35±6.24
p-value	0.975	0.760	0.375	0.952	0.206	0.399
Antioxidants (B)						
Vit_E	40.85±0.44	152.86±11.87	164.54±5.24 ^a	41.41±0.513	166.56±22.19	197.28±6.86
Selenium	40.86±0.19	151.47±13.39	150.15±9.05 ^b	41.21±0.342	165.31±19.22	196.93±6.31
Vit_E + Selenium	40.74±0.18	157.17±13.21	161.57±10.58 ^a	41.27±0.345	169.53±11.82	189.87±6.71
p-value	0.682	0.450	0.003	0.586	0.882	0.420
p-value(AxB)	0.811	0.004	0.322	0.293	0.659	0.272

Note: ^{a,b} Different letters following the mean values in the same column indicate a highly significant effect ($P<0.05$); DOC weight: light (30–34 grams), medium (35–39 grams), heavy (40–45 grams); Vitamin E (250 mg/kg), Selenium (1 mg/kg), Vitamin E + Selenium (250 mg/kg + 1 mg/kg).

body weights comparable to heavier groups, suggesting compensatory growth. Feed conversion ratio (FCR) averaged 1.40, with the most efficient FCR (1.34) in heavy chicks with VE+Se and the least efficient (1.49) in light chicks without antioxidants (Table 2). These findings highlight the importance of initial body weight and antioxidant strategy in optimizing broiler growth and feed efficiency.

PHYSIOLOGICAL RESPONSE

Panting frequency and heart rate are closely interrelated physiological indicators of thermal stress and metabolic load in broiler chickens. In the present study, a significant

interaction was observed between DOC weight and antioxidant type on panting frequency ($P<0.05$). Lighter chicks exhibited higher panting rates, particularly in the absence of antioxidant supplementation. The combined supplementation of Vitamin E and Selenium consistently reduced panting frequency, especially in heavier DOCs, indicating a more stable adaptive response to environmental conditions (Table 3).

Heart rate was also significantly influenced by antioxidant type. Among the antioxidant treatments, Selenium supplementation resulted in significantly lower heart rate values ($P<0.05$), suggesting a potential role in enhancing

cardiovascular stability (Table 3). Given the physiological linkage between panting and heart rate in stress responses, the concurrent reduction of both parameters through antioxidant supplementation particularly Selenium reflects improved physiological resilience and thermoregulatory efficiency in broilers.

HEMATOLOGY

The results of the study on the difference in DOC weight and the addition of antioxidants Vitamin E and Selenium in feed did not show any significant interaction on erythrocytes, hematocrit, and hemoglobin. However, in medium and heavy DOC weight, a tendency was found to have higher erythrocyte and hematocrit values (Table 4).

WHITE BLOOD CELL PROFILE

The results of this study (Table 5) indicate that although the differences were not statistically significant ($P>0.05$), there was a consistent trend suggesting that DOC body weight influences the leukocyte profile of broilers. Heavier DOCs tended to exhibit lower heterophil counts and higher lymphocyte and monocyte levels compared to lighter chicks, which may reflect reduced physiological stress and a more effective immune response. Antioxidant supplementation, particularly the combination of Vitamin E and Selenium, also tended to reduce heterophil levels while increasing monocytes, indicating a potential synergistic effect in supporting the broiler immune system. Although the interaction between DOC weight and antioxidant type was not statistically significant, the combination of heavier DOC and Vitamin E + Selenium supplementation resulted in a more stable and adaptive leukocyte profile, suggesting improved immunological

competence under such treatment conditions.

Table 4: Effect of DOC weight and dietary antioxidant supplementation on the hematology of broiler chickens.

Treatment	Parameter		
	Erythrocytes (10 ⁶ /mm ³)	Hematocrit (%)	Hemoglobin (g/dL)
DOC weight (A)			
Light	1.94±0.16	26.89±2.26	8.43±1.82
Medium	2.42±0.34	28.56±2.60	9.27±1.63
Heavy	2.32±0.26	28.78±1.30	8.69±1.03
p-value	0.058	0.134	0.513
Antioxidants (B)			
Vit_E	2.20±0.48	28.11±2.57	9.22±2.09
Selenium	2.47±0.50	27.78±2.59	8.88±1.30
Vit_E + Selenium	2.47±0.88	28.33±1.58	8.29±0.92
p-value	0.504	0.849	0.446
p-value(AxB)	0.774	0.174	0.433

Note: DOC weight: light (30–34 grams), medium (35–39 grams), heavy (40–45 grams); Vitamin E (250 mg/kg), Selenium (1 mg/kg), Vitamin E + Selenium (250 mg/kg + 1 mg/kg).

INTESTINAL HISTOLOGY

The results presented in Table 6 showed that the weight of broiler DOC with the addition of antioxidants in the feed showed a very significant interaction ($P<0.05$) on the apical width of the small intestine. The combination of Vitamin E + Selenium in this study seemed to be more effective in increasing the apical width than giving one antioxidant alone. This combination works synergistically to protect epithelial cells from oxidative stress, accelerate cell regeneration, and improve intestinal mucosal health.

Table 5: Effect of DOC weight and dietary antioxidant supplementation on the leukocyte profile of broiler chickens.

Treatment	Parameter				
	Heterophils (%)	Eosinophils (%)	Basophils (%)	Monocytes (%)	Lymphocytes (%)
DOC Weight (A)					
Light	44.89±10.74	2.78±2.11	1.11±1.53	4.00±2.34	48.67±9.06
Medium	40.44±12.16	3.00±3.16	1.22±1.20	5.56±1.94	48.67±7.66
Heavy	34.22±6.83	3.11±2.71	1.44±1.13	3.67±3.57	57.56±8.87
p-value	0.137	0.966	0.877	0.333	0.081
Antioxidants (B)					
Vit_E	37.33±9.03	2.89±2.09	1.11±1.16	3.44±2.45	54.67±10.10
Selenium	41.33±11.73	3.11±3.48	1.22±1.20	4.67±2.69	51.67±8.68
Vit_E + Selenium	40.89±12.03	2.90±2.32	1.44±1.51	5.11±3.06	48.56±8.97
p-value	0.694	0.981	0.877	0.441	0.379
p-value(AxB)	0.637	0.321	0.659	0.597	0.843

Note: DOC weight: light (30–34 grams), medium (35–39 grams), heavy (40–45 grams); Vitamin E (250 mg/kg), Selenium (1 mg/kg), Vitamin E + Selenium (250 mg/kg + 1 mg/kg).

Table 6: Effect of DOC weight and dietary antioxidant supplementation on the histological characteristics of the broiler chicken intestine.

Treatment	Parameter				
	Villi height (µm)	Apical width (µm)	Basal width (µm)	Crypt depth (µm)	Villi surface area (mm²)
Jejunum					
DOC Weight (A)					
Light	1006.44±327.43	92.54±25.80	113.74±40.23	200.91±22.13	1018.59±329.33
Medium	944.77±199.37	97.17±28.63	121.38±50.60	257.25±85.36	955.87±199.44
Heavy	1018.51±318.93	103.84±34.16	131.84±42.60	250.31±79.98	1029.60±320.28
p-value	0.789	0.597	0.632	0.219	0.789
Antioxidants (B)					
Vit_E	843.18±255.51	85.08±20.59	93.98±33.67	240.57±92.88	854.39±257.84
Selenium	1122.86±214.54	106.66±34.67	131.63±37.57	244.77±77.18	1135.04±214.36
Vit_E + Selenium	1003.68±310.76	101.92±28.41	141.32±43.61	223.13±37.64	1014.64±311.98
p-value	0.074	0.145	0.049	0.796	0.074
p-value(AxB)	0.082	0.013	0.303	0.478	0.083
Duodenum					
DOC Weight (A)					
Light	1163.59±150.68	142.60±47.41	182.93±56.42	237.25±36.68	1173.60±149.36
Medium	1237.86±80.61	135.07±39.21	181.80±56.77	294.11±28.13	1248.63±80.80
Heavy	1154.91±248.33	164.45±28.28	206.36±74.88	271.61±101.78	1163.24±251.46
p-value	0.559	0.304	0.678	0.159	0.547
Antioxidants (B)					
Vit_E	1200.42±187.84	156.71±41.77	185.30±59.06	287.32±93.29	1209.80±189.50
Selenium	1183.89±142.79	155.62±39.11	212.39±63.46	263.38±41.34	1193.02±144.21
Vit_E + Selenium	1172.04±199.79	129.80±39.65	173.41±61.88	252.28±58.62	1182.64±199.56
p-value	0.943	0.305	0.455	0.466	0.948
p-value(AxB)	0.292	0.780	0.671	0.120	0.290
Ileum					
DOC Weight (A)					
Light	625.20±15.08	99.80±23.94	118.11±37.79	147.59±30.14	632.66±157.75
Medium	679.35±64.09	110.91±30.96	126.80±35.72	200.91±50.21	686.94±65.06
Heavy	846.04±204.74	105.47±20.08	129.52±26.46	220.40±94.01	855.37±207.04
p-value	0.013	0.660	0.795	0.083	0.13
Antioxidants (B)					
Vit_E	754.10±68.30	114.73±27.11	128.71±27.91	210.26±103.14	762.06±69.78
Selenium	672.30±114.91	109.73±21.25	128.99±30.06	179.39±47.65	679.71±116.18
Vit_E + Selenium	724.20±280.68	92.00±22.34	116.72±41.12	179.25±42.86	733.19±282.90
p-value	0.499	0.171	0.729	0.537	0.50
p-value(AxB)	0.181	0.769	0.878	0.618	0.18

Note: DOC weight: light (30–34 grams), medium (35–39 grams), heavy (40–45 grams); Vitamin E (250 mg/kg), Selenium (1 mg/kg), Vitamin E + Selenium (250 mg/kg + 1 mg/kg).

DISCUSSION

The positive effects of Vitamin E and Selenium in poultry feed are undeniable. However, antioxidant effects that may be related to the initial weight of DOC with certain concentrations and combinations have not been widely studied. Low hatching weight in DOC is associated

with increased oxidative stress, likely due to elevated metabolic activity (Cowieson and Parsons, 2024). High metabolism can trigger increased production of ROS, negatively impacting cells biological and physiological functions (Emami *et al.*, 2020; Syamsuryadi *et al.*, 2025). Uncontrolled spikes in ROS can initiate a chain reaction driven by free radicals, leading to lipid peroxidation and

subsequent oxidative damage (Hai *et al.*, 2006; Harsini *et al.*, 2012; He *et al.*, 2018). Providing Vitamin E and Selenium antioxidants can help combat free radicals that can damage cells, increase nutrient absorption, and improve immune responses (Sahin and Kucuk, 2001; Khan *et al.*, 2011; Surai *et al.*, 2019). A study by Calik *et al.* (2022a) showed that providing Vitamin E and Selenium in feed increases the final weight of a broiler raised for 35 days by 1.7–4%.

Increasing metabolic efficiency will have an impact on the physiological condition of chickens. Rectal temperature is an indicator of the thermal balance and energy metabolism. The study by Calik *et al.* (2022b) on providing Vitamin E and selenium in feed showed a stable rectal temperature. A stable rectal temperature indicates that a chicken can regulate its body heat well. Unbalanced energy metabolism and oxidative stress trigger an increase in heart rate. Antioxidant administration can protect the heart muscle from oxidative damage, which can improve blood circulation and nutrient distribution in the body's tissues (Jang *et al.*, 2014; Amevor *et al.*, 2021). The mechanism for stabilizing a chicken's body temperature can be carried out by panting. Attia *et al.* (2017) showed that administering Vitamin E and Selenium can stabilize rectal temperature, improve energy metabolism, and reduce panting frequency. The role of Vitamin E and Selenium is not only limited to stabilizing body temperature but also includes improving blood quality related to oxygen transport capacity (Uzochukwu *et al.*, 2025).

The study by Maqsood *et al.* (2025b) showed the tendency of erythrocyte and hematocrit values to be higher with heavier DOC weight, which illustrates a more optimal oxygen transport capacity. The increase in erythrocyte and hematocrit values is sometimes not followed by hemoglobin levels, which are greatly influenced by genetics, nutrition, hydration, and hormonal regulation (Syamsuryadi *et al.*, 2017). Vitamin E plays a role in maintaining red blood cell stability, preventing hemolysis (rupture of red blood cells), and maintaining hemoglobin function. Meanwhile, Selenium is an essential mineral that can collaborate with Vitamin E in preventing oxidative damage to red blood cells. Supplementation Vitamin E and Selenium supplementation at moderate levels increase hemoglobin concentration and support the erythropoietic stress response (Tayeb and Qader, 2012; Ekunseitan *et al.*, 2021).

The leukocyte profile is an indicative of health and immune balance in chicken's body. Antioxidants such as Vitamin E and Selenium play a role in increasing endurance by reducing oxidative stress and improving immune balance (Sahin *et al.*, 2001; Surai, 2018). The administration of

a combination of Vitamin E and Selenium provides the best benefits in maintaining leukocyte balance, with lower heterophils (reducing stress) and higher monocytes (increasing endurance). This shows that antioxidant supplementation can improve broiler health and help deal with environmental challenges or diseases.

Antioxidants in this study not only play a role in the immune system but also play a role in optimizing intestinal morphology. The jejunum is a part of the small intestine that plays an important role in the absorption of nutrients, specifically carbohydrates, proteins, and lipids. This study shows the interaction of apical villi width with different DOC weight related to the type of antioxidant given. A larger apical villi width can increase the absorption surface area, which has the potential to increase the efficiency of digestion and absorption of nutrients. Supplementation of antioxidants such as Vitamin E and Selenium in feed can affect the health and efficiency of intestinal function by protecting epithelial cells from oxidative stress and supporting tissue regeneration (Jang *et al.*, 2014; El-Senousey *et al.*, 2018; Amevor *et al.*, 2021).

CONCLUSION

In conclusion, a higher initial DOC (day-old chick) weight supports optimal broiler growth, while supplementation with Vitamin E and Selenium enhances digestive efficiency and nutrient absorption. The combination of both results in improved broiler performance, as indicated by greater body weight gain, better blood quality, and healthier intestinal structure. This strategy effectively increases broiler productivity.

ACKNOWLEDGEMENT

The authors are grateful to the Higher Education Fund Management Agency (BPPT) and the Education Fund Management Institute (LPDP).

NOVELTY STATEMENT

This is the first study to report the effect of combining two important factors, initial DOC weight and the administration of a combination of antioxidants (Vitamin E and Selenium).

AUTHOR'S CONTRIBUTION

BS: conducted the study, data analysis, and statistical analysis, and wrote the original manuscript. S, LE, and EW: formulated the methodology, supervised, reviewed, and edited the manuscript.

ETHICAL APPROVAL

This study was approved by the Ethics Commission of Brawijaya University under approval number 063-KEP-UB-2023.

GENERATIVE AI AND AI-ASSISTED TECHNOLOGY STATEMENT

The authors acknowledge that generative AI tools (e.g., ChatGPT, language editing software) were used only to improve grammar, language clarity, and formatting. No AI tools were used for data analysis, interpretation, or drawing scientific conclusions. The authors take full responsibility for the content of this article.

CONFLICT OF INTEREST

The authors have declared no conflict of interest.

REFERENCES

- Alahmar AT, Al-Azawi AA, Al-Mashhadani SA (2023). Oxidative stress and poultry performance: Antioxidants to the rescue. *Poult. Res. Rep.*, 12(1): 1–10.
- Alahmar AT, Jothery AI, Al-Daami AHT, Abbas QJ, Al-Hassnawi A (2023). The effect of coenzyme Q10 on DEXamethasone-induced oxidative stress in rats testes. *Med. J. Babylon*, 20: 130–135. https://doi.org/10.4103/MJBL.MJBL_307_22
- Amevor FK, Cui Z, Du X, Ning Z, Shu G, Jin N, Deng X (2021). Dietary vitamin E and selenium improve growth performance and regulate antioxidant capacity and immune-related gene expression in broilers under stress. *Antioxidants*, 10(12): 1915.
- Attia YA, El-Hamid AE, Abedalla AA, Berika MA, Al-Harthi MA, Kucuk O, Sahin K (2017). Impact of dietary vitamin E and/or selenium supplementation on growth, immunity, and antioxidant status of broilers under heat stress. *Asian-Australas. J. Anim. Sci.*, 30(6): 784–791.
- Calik A, Emami NK, White MB, Walsh MC, Romero LF, Dalloul RA (2022). Effect of early feeding and antioxidants on broiler immunity and performance. *Animals*, 12(5): 642.
- Calik A, Emami NK, White MB, Walsh MC, Romero LF, Dalloul RA (2022a). Influence of dietary vitamin E and selenium supplementation on broilers subjected to heat stress, Part I: Growth performance, body composition and intestinal nutrient transporters. *Poult. Sci.*, 101: 101857. <https://doi.org/10.1016/j.psj.2022.101857>
- Calik A, Emami NK, White MB, Walsh MC, Romero LF, Dalloul RA (2022b). Influence of dietary vitamin E and selenium supplementation on broilers subjected to heat stress, Part I: Growth performance, body composition and intestinal nutrient transporters. *Poult. Sci.*, 101: 101857. <https://doi.org/10.1016/j.psj.2022.101857>
- Charde RM, Charde MS, Fulzele SV, Satturwar PM, Kasture AV, Joshi SB (2011). Evaluation of ethanolic extract of *Moringa oleifera* for wound healing, anti-inflammatory and antioxidant activities on rats. *Res. J. Pharm. Technol.*, 4: 254–258.
- Cowieson AJ, Parsons AS (2024). The role of breeder nutrition and chick weight in broiler lifetime performance. *Front. Poult. Nutr.*, 5: 101231.
- Cowieson AJ, Parsons CM (2024). Effect of day-old chick weight on the response of broilers to high doses of exogenous phytase. *Poult. Sci.*, 103: 103296. <https://doi.org/10.1016/j.psj.2023.103296>
- Ekunseitan DA, Yusuf AO, Ekunseitan OF, Alao SO, Allinson AZ (2021). Dietary supplementation of vitamin E and selenium on performance and oxidative stability of meat of broiler chickens in a hot climate. *Agric. Trop. Subtrop.*, 54: 24–31. <https://doi.org/10.2478/ats-2021-0003>
- El-Senousey H, Chen B, Wang J, Atta A, Mohamed F, Nie Q (2018). Effects of dietary vitamin C, vitamin E, and alpha-lipoic acid supplementation on the antioxidant defense system and immune-related gene expression in broilers exposed to oxidative stress by DEXamethasone. *Poult. Sci.*, 97: 30–38. <https://doi.org/10.3382/ps/pex298>
- Emami NK, Jung U, Voy B, Dridi S (2020). Radical response: effects of heat stress-induced oxidative stress on lipid metabolism in the avian liver. *Antioxidants*, 10: 35. <https://doi.org/10.3390/antiox10010035>
- Gasperz V (1991). *Metode Perancangan Percobaan*. Bandung: Armico.
- Hai L, Decuypere E, Buyse J (2006). Acute heat stress induces oxidative stress in broiler chickens. *Comp. Biochem. Physiol. A*, 144: 11–17. <https://doi.org/10.1016/j.cbpa.2006.01.032>
- Harsini SG, Habibiyan M, Moeini MM, Abdolmohammadi AR (2012). Effects of dietary selenium, vitamin E, and their combination on growth, serum metabolites, and antioxidant defense system in skeletal muscle of broilers under heat stress. *Biol. Trace Elem. Res.* 148: 322–330. <https://doi.org/10.1007/s12011-012-9374-0>
- Hartono M, Suharyati S, Santosa PE (2002). *Dasar Fisiologi Ternak*. Buku Ajar, Jurusan Peternakan, Fakultas Pertanian, Universitas Lampung, Bandar Lampung.
- He S, Arowolo M, Medrano R, Li S, Yu Q, Chen J, He J (2018). Impact of heat stress and nutritional interventions on poultry production. *World Poult. Sci. J.*, 74: 647–664. <https://doi.org/10.1017/S0043933918000727>
- He T, Zhao J, Wang Y, Zhang Y, Li X (2021). Posthatch feed and water access affects intestinal development and oxidative status in broilers. *Poult. Sci.*, 100(9): 101241.
- Jang IS, Ko YH, Moon YS, Sohn SH (2014). Effects of vitamin C or E on proinflammatory cytokines, heat shock protein 70, and antioxidant status in broiler chicks under summer conditions. *Asian-Austral. J. Anim. Sci.*, 27: 749–756. <https://doi.org/10.5713/ajas.2013.13852>
- Khan RU, Naz S, Nikousefat Z, Tufarelli V, Javdani M, Rana N, Laudadio V (2011). Effect of vitamin E in heat-stressed poultry. *World's Poult. Sci. J.*, 67: 469–478. <https://doi.org/10.1017/S0043933911000511>
- Liu Z, Wang X, Chen Y, Zhang H, Li J (2022). Vitamin E and selenium supplementation modulates immune and oxidative responses in broiler chickens. *Poult. Sci.*, 101(6): 101876.
- Maqsood I, Awaz S, Sareer S, Rehman SU, Hidayat A, Shah M, Ke R (2025b). Dietary supplementation with Jasminum sambac leaf powder: Effects on growth, hematology, and immune parameters in broiler chickens. *Poult. Sci.*, 104: 10464. <https://doi.org/10.1016/j.psj.2024.104645>
- Maqsood S, Khan MA, Ahmed R, Ali S (2025a). Hatchling weight and yolk utilization in broilers: A performance indicator. *J. Avian Physiol.*, 34(2): 215–224.
- Muwakhid B, Ali U, Rifa'i, Nugraha P (2025). Utilization of Industrial Waste (Fish Meal Oil Sludge and Cassava Dregs)

- as an Alternative to Commercial Broiler Feed. *Journal of Animal Health and Production*, 13(3). <https://doi.org/10.17582/journal.jahp/2025/13.3.682.687>
- Noruzi H, Hassanabadi A, Golian A, Aziz-Aliabadi F (2022). Effects of dietary calcium and phosphorus restrictions on growth performance, intestinal morphology, nutrient retention, and tibia characteristics in broiler chickens. *Br. Poult. Sci.*, 64(2): 231–241. <https://doi.org/10.1080/00071668.2022.2136510>
- Noruzi H, Hassanabadi A, Golian A, Aziz-Aliabadi F (2023). Effect of hatch weight and antioxidant supplementation on oxidative stress and intestinal morphology in broilers. *Poult. Sci.*, 102(3): 102187.
- Prakatur I, Miskulin M, Pavic M, Marjanovic K, Blazicevic V, Miskulin I, Domacinovic M (2019). Intestinal morphology in broiler chickens supplemented with propolis and bee pollen. *Animals*, 9: 1–12. <https://doi.org/10.3390/ani9060301>
- Sahin K, Kucuk O (2001). Effects of vitamin E and selenium on performance, digestibility of nutrients, and carcass characteristics of Japanese quails reared under heat stress (34 °C). *J. Anim. Physiol. Anim. Nutr.*, (Berl), 85: 342–348. <https://doi.org/10.1046/j.1439-0396.2001.00340.x>
- Surai PF (2018). Antioxidant systems in poultry biology: Superoxide dismutase. *J. Anim. Res. Nutr.*, 3(1): 8.
- Surai PF, Kochish II, Romanov MN, Griffin DK (2019). Nutritional modulation of the antioxidant capacities in poultry: the case of vitamin E. *Poultry Science*, 98(9), 4030–4041. <https://doi.org/10.3382/ps/pez072>
- Surai PF, Fisinin VI, Karadas F (2023). Mitochondrial-targeted antioxidants in poultry production. *Antioxidants*, 12(3): 488.
- Syamsuryadi B, Afnan R, Arief I, Ekastuti D (2017). Ayam Pedaging Jantan yang Dipelihara di Dataran Tinggi Sulawesi Selatan Produktivitasnya Lebih Tinggi (Higher Productivity Performance Of Male Broilers Reared In The Highland of South Sulawesi). *Jurnal Veteriner*, 18(1), 160–166. <https://doi.org/10.19087/jveteriner.2017.18.1.160>
- Syamsuryadi B, Widodo E, Radiati LE, Djunaidi IH, Rifa'i M, Suyadi S (2025). Optimizing Broiler Chick Management: Interactive Effects of Initial Body Weight and Post-Hatch Feed Deprivation on Growth, Hematological Immune Response, and Intestinal Integrity. *Advances in Animal and Veterinary Sciences*, 13(9). <https://doi.org/10.17582/journal.aavs/2025/13.9.2096.2102>
- Tayeb IT, Qader GK (2012). Effect of feed supplementation of selenium and vitamin E on production performance and some hematological indices of broiler. *KSU J. Nat. Sci.*, 15: 46–56.
- Tona K, Onagbesan O, De Ketelaere B, Decuyper E, Bruggeman V (2004). Effects of Age of Broiler Breeders and Egg Storage on Egg Quality, Hatchability, Chick Quality, Chick Weight, and Chick Posthatch Growth to Forty-Two Days. *Journal of Applied Poultry Research*, 13(1), 10–18. <https://doi.org/10.1093/japr/13.1.10>
- Uzochukwu IC, Okonkwo CC, Nwafor EI, Eze DC (2025). Antioxidant synergy of vitamin E and selenium improves intestinal morphology and CD4/CD8 ratios in stressed broilers. *Vet. Immunol. Nutr.*, 41(1): 58–68.
- Uzochukwu IE, Alia LC, Amaefulea BC, Okeke CC, Ositaa CO, Machebea NS, Yanchevad V, Somogyib D, Nyesteb K (2025). Impact of vitamin E and selenium supplementation on growth, reproductive performance, and oxidative stress in dexamethasone-stressed Japanese quail cocks. *Poult. Sci.*, 104: 104888. <https://doi.org/10.1016/j.psj.2025.104888>
- Yousef MK (1985). Thermoneutral zone. In: Yousef MK (Ed.). *Stress physiology of livestock*. Vol. II. CRC Press, Boca Raton, Florida. pp. 68–69.