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Effects of Dietary Selenium Supplementation Levels on Laying Performance, Egg Quality, Serum Antioxidant Indices, and Dynamic Changes in Egg Selenium Content in Linwu Ducks During the Peak Laying Period: Postprint

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Abstract

This study aimed to investigate the effects of dietary selenium supplementation levels on laying performance, egg quality, serum antioxidant indices, and dynamic changes in egg selenium content in peak-laying Linwu ducks. Two hundred 29-week-old peak-laying Linwu ducks with good health status and similar laying rates were randomly allocated into 5 groups with 5 replicates per group and 8 ducks per replicate. The control group was fed a basal diet (without additional selenium supplementation, measured selenium content of 0.15 mg/kg) for 45 d, while groups I, II, III, and IV were fed the basal diet supplemented with 0.10, 0.20, 0.40, and 0.80 mg/kg selenium as selenium yeast for 35 d, followed by the basal diet for 10 d. The results showed: 1) Dietary selenium supplementation levels had no significant effect on any indices of laying performance or egg quality in laying ducks ($P>0.05$). 2) Compared with the control group and group I, supplementation with 0.40 and 0.80 mg/kg selenium significantly increased serum selenium and glutathione (GSH) contents as well as superoxide dismutase (SOD) and glutathione peroxidase (GSH-PX) activities ($P<0.05$), and significantly decreased serum malondialdehyde (MDA) content ($P<0.05$). 3) With increasing experimental days, egg selenium content showed no significant changes in the control group and group I ($P>0.05$), while groups II, III, and IV exhibited a trend of increasing first and then decreasing, all reaching peak values on day 9 of the experiment. On days 7, 9, 13, 17, 21, 28, and 35 of the experiment, egg selenium content in groups II, III, and IV was significantly higher than that in the control group and group I ($P<0.05$); on days 1–4 after withdrawal of the selenium-supplemented diet, egg selenium content in groups II, III, and IV remained significantly higher than that in the control group (P<0.05); on days 5-8 after withdrawal, egg selenium content in groups III and IV was significantly higher than that in the control group (P<0.05); on day 9 after withdrawal, egg selenium content in all treatment groups had attenuated to the same level as the control group (P>0.05). These results indicate that supplementation with different levels of selenium yeast in the diet had no adverse effects on laying performance or egg quality of peak-laying Linwu ducks. Supplementation with 0.40 and 0.80 mg/kg selenium as selenium yeast increased egg selenium content, serum selenium content, and antioxidant capacity, and prolonged the retention duration of egg selenium. Taking all factors into consideration, the optimal supplementation level of selenium (as selenium yeast) in diets for peak-laying Linwu ducks is 0.40 mg/kg.

Full Text

Effects of Dietary Selenium Supplemental Level on Laying Performance, Egg Quality, Serum Antioxidant Indices, and Dynamic Changes in Egg Selenium Content of Linwu Ducks During Peak Laying Period

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Abstract

This experiment was conducted to investigate the effects of dietary selenium supplementation on laying performance, egg quality, serum antioxidant indices, and the dynamic changes in egg selenium content of Linwu ducks during peak production. Two hundred 29-week-old healthy Linwu ducks with similar laying rates were randomly assigned to five groups, each consisting of five replicates with eight ducks per replicate. The control group was fed a basal diet without supplemental selenium (measured selenium content: 0.15 mg/kg) for 45 days. Groups I, II, III, and IV received the basal diet supplemented with 0.10, 0.20, 0.40, and 0.80 mg/kg selenium in the form of selenium yeast for 35 days, followed by the basal diet alone for an additional 10 days. The results showed: (1) Dietary selenium level had no significant effects on any indices of laying performance or egg quality (P>0.05). (2) Compared with the control and Group I, supplementation with 0.40 and 0.80 mg/kg selenium significantly increased serum selenium and glutathione (GSH) contents, as well as superoxide dismutase (SOD) and glutathione peroxidase (GSH-Px) activities ($P<0.05$), while significantly decreasing serum malondialdehyde (MDA) content ($P<0.05$). (3) As the experiment progressed, egg selenium content remained unchanged in the control and Group I ($P>0.05$), whereas Groups II, III, and IV exhibited a trend of initial increase followed by decrease, with all three groups reaching peak values on day 9. On days 7, 9, 13, 17, 21, 28, and 35, egg selenium content in Groups II, III, and IV was significantly higher than in the control and Group I ($P<0.05$). During days 1-4 after withdrawal of selenium supplementation, egg selenium content in Groups II, III, and IV remained significantly higher than the control ($P<0.05$). During days 5-8 post-withdrawal, Groups III and IV maintained significantly higher egg selenium content than the control ($P<0.05$). By day 9 after withdrawal, egg selenium content in all treatment groups had declined to levels comparable to the control ($P>0.05$). These findings indicate that dietary supplementation with different levels of selenium yeast does not adversely affect the laying performance or egg quality of peak-laying Linwu ducks. Supplementation with 0.40 and 0.80 mg/kg selenium yeast can enhance egg selenium content, serum selenium levels, and antioxidant capacity while prolonging selenium retention in eggs. Considering all factors comprehensively, the appropriate dietary selenium supplemental level for peak-laying Linwu ducks is 0.40 mg/kg as selenium yeast.

Keywords: selenium yeast; antioxidant capacity; egg selenium content; Linwu duck

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Introduction

Selenium is an essential trace element in animals that plays crucial roles in antioxidant defense, stress resistance, immune function, and thyroid activity [1]. Previous studies indicate that the optimal selenium supplementation range in poultry diets is 0.1-0.5 mg/kg. Specifically, the selenium requirement for meat-type ducks during laying is 0.3 mg/kg [2], while recommended levels are 0.28–0.35 mg/kg for 1–70 day-old goslings [3], 0.36–0.38 mg/kg for 1–4 week-old Taihe black-bone chickens [4], 0.4 mg/kg for laying pigeons [5], and 0.10–0.50 mg/kg for laying hens [6-7]. Additionally, Chen et al. [8] found that dietary selenium levels of 0.27 and 0.20 mg/kg optimized production performance in laying ducks during early and peak laying phases, respectively, while 0.36 mg/kg was optimal for antioxidant capacity during peak laying. He et al. [9] reported that supplementing green-shell laying hen diets with 0.25 mg/kg selenium yeast and 400 mg/kg tea polyphenols improved production performance and egg quality. These studies primarily examined the effects of continuous selenium supplementation on poultry performance and serum antioxidant indices. However, research on the dynamic changes and retention duration of egg selenium content after discontinuing selenium supplementation remains scarce, and studies on optimal selenium levels for laying ducks, particularly Linwu ducks, are limited. Therefore, this experiment investigated the effects of varying dietary selenium levels on laying performance, egg quality, and serum antioxidant indices in peak-laying Linwu ducks, and analyzed the dynamic changes in egg selenium content following withdrawal of selenium supplementation to determine the optimal dietary selenium level.

Materials and Methods

1.1 Experimental Material Selenium yeast was purchased from Hunan Novozymes Biological Technology Co., Ltd., with a selenium content of 2,000 mg/kg.

1.2 Experimental Design and Diets Two hundred 29-week-old healthy Linwu ducks with similar laying rates were randomly divided into five groups, each comprising five replicates of eight ducks. The 45-day trial consisted of a 35-day supplementation period (days 1–35) followed by a 10-day withdrawal period (days 36–45) when all groups received the basal diet. The control group was fed a basal diet without supplemental selenium (measured selenium content: 0.15 mg/kg). Groups I, II, III, and IV received the basal diet supplemented with 0.10, 0.20, 0.40, and 0.80 mg/kg selenium as selenium yeast, respectively. The composition and nutrient levels of the basal diet are presented in Table 1 .

Table 1 Composition and nutrient levels of the basal diet (air-dry basis) %

Ingredients	Content	Nutrient levels[2]	Content
Corn	61.00	Metabolizable energy (MJ/kg)	11.12
Soybean meal	22.00	Crude protein	17.50
Corn gluten powder	3.00	Calcium	3.20
Wheat middling	5.00	Total phosphorus	0.60
Wheat bran	3.00	Available phosphorus	0.35
Limestone	8.50	Lysine	0.85
L-Lys (78.5%)	0.10	Methionine	0.38
DL-Met (98.5%)	0.15	Methionine + Cysteine	0.65
$CaHPO_4$	1.20	Selenium (mg/kg)	0.15
NaCl	0.30		
Premix[1]	0.75		
Total	**100.00**		

[1]The premix provided per kilogram of diet: VA 5,000 IU, VB_1 2 mg, VB_2 15 mg, VB_6 4 mg, VB_{12} 0.02 mg, VD_3 800 IU, VE 20 IU, VK_3 0.5 mg, biotin 0.2 mg, folic acid 0.6 mg, D-pantothenic acid 60 mg, nicotinic acid 60 mg, choline 1,500 mg, antioxidant 100 mg, Cu (as copper sulfate) 8 mg, Fe (as ferrous sulfate) 80 mg, Mn (as manganese sulfate) 50 mg, Zn (as zinc sulfate) 60 mg, I (as

potassium iodide) 0.40 mg.
[2]Selenium was measured; other nutrient levels were calculated.

1.3 Management The feeding trial was conducted at the Waterfowl Experimental Farm of Hunan Institute of Animal Science and Veterinary Medicine. Ducks were housed individually in double-tier metal cages in a closed duck house with ad libitum access to feed and water. Conventional feeding management and immunization protocols were followed throughout the experimental period.

1.4 Sampling and Measurements **1.4.1 Laying Performance**
Eggs were collected daily at 09:00, and total egg weight and daily egg number were recorded per replicate. Feed intake and residual feed were recorded weekly to calculate average daily feed intake (ADFI), laying rate, average egg weight, daily egg yield, and feed-to-egg ratio.

1.4.2 Egg Quality
On day 42, 15 eggs per group (3 per replicate) with weights close to the group average were collected and stored at 4°C. Within 24 hours, egg quality parameters were measured: yolk ratio, shell weight percentage, eggshell thickness (measured with an eggshell thickness gauge, ORKA, Israel), egg shape index (measured with vernier calipers), yolk color, and albumen height (measured with an albumen height gauge, ORKA, Israel). Haugh units were calculated using the formula:
$HU = 100 \times \log(H - 1.7W^{0 \cdot 37} + 7.57)$, where HU is Haugh unit, H is albumen height (mm), and W is egg weight (g).

1.4.3 Serum Biochemical Indices
On day 42, two ducks per replicate with similar body weights were selected after 12 hours of fasting, and 5 mL blood samples were collected from the wing vein. Serum was separated after centrifugation at 3,000 rpm for 15 minutes following 30 minutes of coagulation and stored at -20°C. Serum glutathione peroxidase (GSH-Px) and superoxide dismutase (SOD) activities, and glutathione (GSH) and malondialdehyde (MDA) contents were measured using colorimetric assay kits (Nanjing Jiancheng Bioengineering Institute). Serum selenium content was determined by hydride generation atomic fluorescence spectrometry (GB/T 13883-2008).

1.4.4 Selenium Content in Eggs and Diets
On days 1, 3, 5, 7, 9, 13, 17, 21, 28, and 35 of the trial, and on days 1–10 after selenium withdrawal (days 36–45), 15 eggs per group (3 per replicate) with weights close to the average were collected. Egg samples were processed according to Pan et al. [10] before analysis. Selenium content in both eggs and diets was determined by hydride generation atomic fluorescence spectrometry (GB/T 13883-2008).

1.5 Statistical Analysis Data were analyzed using one-way ANOVA with SPSS 18.0 software, followed by Duncan' s multiple comparison test. Results are expressed as mean ± standard deviation (SD). Differences were considered significant at P<0.05 and highly significant at P<0.01.

Results

2.1 Effects of Dietary Selenium Level on Laying Performance As shown in Table 2 , dietary selenium supplementation level had no significant effects on any indices of laying performance (P>0.05).

Table 2 Effects of selenium supplemental level on laying performance of laying ducks

Items	Control	Group I	Group II	Group III	Group IV	P-value
ADFI (g/d)	147.78±0.63	148.45±0.81	147.81±1.17	147.74±0.72	148.09±0.34	0.641
Layingrate(	±0.39	82.24±1.5				

In the same row, values with no letter or the same letter superscripts indicate no significant difference (P>0.05), while different lowercase letters indicate significant difference (P<0.05). The same applies below.

2.2 Effects of Dietary Selenium Level on Egg Quality As shown in Table 3 , dietary selenium level had no significant effects on any egg quality parameters (P>0.05), though Group III exhibited the highest values for yolk ratio, yolk color, and albumen height.

Table 3 Effects of selenium supplemental level on egg quality of laying ducks

Items	Control	Group I	Group II	Group III	Group IV	P-value
Egg shape index	1.33±0.06	[illegible]±0.02	1.36±0.05	1.39±0.03	1.34±0.04	0.312
Eggshellthickness(*mm*)	0.39±0.03	0.39				

2.3 Effects of Dietary Selenium Level on Serum Antioxidant Indices As shown in Table 4 , serum selenium content increased linearly with dietary selenium level (P<0.05), with Groups III and IV showing significantly higher levels than the control and Group I (P<0.05). Compared with the control and Group I, supplementation with 0.40 and 0.80 mg/kg selenium significantly increased serum GSH content and SOD and GSH-Px activities (P<0.05) while significantly decreasing serum MDA content (P<0.05).

Table 4 Effects of selenium supplemental level on serum antioxidant indices of laying ducks

<table>
<tr><th>Items</th><th>Control</th><th>Group I</th><th>Group II</th><th>Group III</th><th>Group IV</th><th>P-value</th></tr>
<tr><td>Selenium (mg/kg)</td><td colspan="6">0.07\$±0.02^{c}<0.001|0.10±0.02^{bc}|0.14±0.03^{ab}|0.18±0.04^{a}|0.19±0.03^{a}| < 0.001||$MDA(nmol/mL)$|8.91±1.75^{a}|8.42±0.83^{a}|8.12±1.80^{a}|5.79±0.44^{b}|5.76±0.40^{b}|0.003||$SOD(U/m$ 0.001||$GSH-Px(U/mL)$|992.37±115.70^{b}|954.12±64.90^{b}|1, 404.22±312.84^{a}|1, 456.88±237.68^{a}|1, 574.38±\$99.44</td></tr>
</table>

2.5 Effects of Dietary Selenium Level on Dynamic Changes in Egg Selenium Content As shown in Table 5 , egg selenium content remained unchanged in the control and Group I throughout the experiment (P>0.05), while Groups II, III, and IV exhibited an initial increase followed by a decrease, with all three groups reaching peak values on day 9. On day 3, no significant differences were observed among groups (P>0.05). On day 5, Groups III and IV showed significantly higher egg selenium content than the control and Group I (P<0.05), while Group II was significantly higher than the control (P<0.05) but not different from other groups (P>0.05). On days 7, 9, 13, 17, 21, 28, and 35, Groups II, III, and IV had significantly higher egg selenium content than the control and Group I (P<0.05). During days 1-4 after selenium withdrawal (days 36-39), Groups II, III, and IV maintained significantly higher egg selenium content than the control (P<0.05). During days 5-8 post-withdrawal (days 40-43), Groups III and IV showed significantly higher egg selenium content than the control (P<0.05). By day 9 after withdrawal (day 44), egg selenium content in all treatment groups had declined to levels comparable to the control (P>0.05).

Table 5 Effects of selenium supplemental level on dynamic change of egg selenium content (mg/kg)

Time	Control	Group I	Group II	Group III	Group IV	P-value
Day 1	0.31±0.01	[illegible]±0.01	0.30±0.03	0.31±0.03	0.31±0.02	0.823
Day 3	0.30±0.01	0.31±0.01	0.32±0.03	[illegible]	[illegible]	< 0.001
Day 7	$0.29±0.01^b$	$0.32±0.01^b$	$0.38±0.01^a$	$0.40±0.01^a$	$0.40±0.03^a$	< 0.001
Day 9	$0.30±0.01^b$	$0.31±0.03^b$	$0.38±0.03^a$	$0.44±0.01^a$	$0.43±0.03^a$	< 0.001
Day 13	$0.29±0.01^c$	$0.31±0.02^c$	$0.37±0.01^b$	$0.42±0.02^a$	$0.42±0.01^a$	< 0.001
Day 17	$0.29±0.01^b$	$0.31±0.04^b$	$0.38±0.01^a$	$0.42±0.01^a$	$0.43±0.01^a$	< 0.001
Day 21	$0.28±0.01^c$	$0.30±0.02^c$	$0.39±0.01^b$	$0.42±0.01^{ab}$	$0.43±0.01^a$	< 0.001
Day 28	$0.29±0.01^b$	$0.32±0.01^b$	$0.39±0.02^a$	$0.42±0.03^a$	$0.43±0.03^a$	< 0.001
Day 35	$0.29±0.01^c$	$0.31±0.01^c$	$0.39±0.01^b$	$0.42±0.01^{ab}$	$0.43±0.02^a$	< 0.001
Day 36	$0.28±0.01^c$	$0.30±0.01^c$	$0.37±0.01^b$	$0.41±0.01^a$	$0.41±0.01^a$	< 0.001
Day 37	$0.30±0.01^b$	$0.29±0.01^b$	$0.37±0.01^a$	$0.40±0.03^a$	$0.40±0.01^a$	< 0.001
Day 38	$0.28±0.01^c$	$0.31±0.02^{bc}$	$0.39±0.01^a$	$0.40±0.04^a$	$0.38±0.04^{ab}$	< 0.001
Day 39	$0.29±0.02^b$	$0.32±0.02^{ab}$	$0.37±0.04^a$	$0.42±0.04^a$	$0.39±0.06^a$	< 0.001
Day 40	$0.28±0.01^b$	$0.30±0.01^b$	$0.33±0.01^{ab}$	$0.38±0.04^a$	$0.37±0.04^a$	< 0.001
Day 41	$0.26±0.01^b$	$0.31±0.01^{ab}$	$0.31±0.01^{ab}$	$0.36±0.01^a$	$0.36±0.04^a$	< 0.001
Day 42	$0.25±0.01^b$	$0.29±0.01^{ab}$	$0.30±0.01^{ab}$	$0.33±0.01^a$	$0.33±0.04^a$	< 0.001
Day 43	$0.28±0.01^b$	$0.28±0.01^b$	$0.30±0.01^{ab}$	$0.34±0.02^a$	$0.33±0.01^a$	< 0.001
Day 44	0.30±0.01	0.30±0.03	0.25±0.05	0.32±0.05	0.31±0.01	0.123
Day 45	0.28±0.06	0.30±0.04	[illegible]	[illegible]	[illegible]	[illegible]

Discussion

3.1 Effects of Dietary Selenium Level on Laying Performance and Egg Quality As an essential trace element, selenium participates in thyroid hormone synthesis, which regulates growth hormone and insulin secretion, thereby promoting animal growth and development. Additionally, selenium is a vital component of antioxidant enzymes that scavenge free radicals and peroxides [11]. Laika et al. [12] reported that dietary supplementation with 0.20–0.40 mg/kg organic selenium significantly improved laying hen performance and extended egg shelf life. Zhao et al. [13] found that compared with unsupplemented diets, 0.20–0.50 mg/kg selenium-enriched probiotics significantly increased laying rate and reduced feed-to-egg ratio without affecting egg quality. Yan et al. [14] observed that dietary selenium level did not significantly affect laying performance in Cherry Valley breeder ducks but that high selenium (0.25 mg/kg) improved eggshell strength. Variations in poultry species, production stage, selenium level, and selenium source may explain these differing results. In the current study, supplementation with 0.20–0.80 mg/kg selenium yeast did not significantly affect laying performance or egg quality of Linwu ducks, consistent with findings in laying hens by Utterback et al. [15], Chantiratikul et al. [16], and Payne et al. [17].

3.2 Effects of Dietary Selenium Level on Serum Antioxidant Indices After absorption in the duodenum, selenium binds to α- and β-globulins in blood for transport to tissues [18]. Studies show that increasing dietary selenium elevates selenium reserves in animal tissues [19]. As an essential component of GSH-Px, selenium deficiency reduces GSH-Px activity, increases MDA and peroxide levels, and consequently affects SOD activity [20]. Thus, dietary selenium directly and indirectly influences antioxidant capacity. The optimal selenium range is relatively broad; supplementation with as little as 0.10 mg/kg improves antioxidant capacity in rats [21], while 0.31–0.36 mg/kg is optimal for goslings [3], and 0.30 mg/kg significantly increases serum GSH-Px activity and reduces MDA in Gushi chickens [22]. Chen et al. [8] reported that 0.18 mg/kg selenium met production needs in laying ducks, but 0.37 mg/kg was required for optimal antioxidant capacity. In this study, serum selenium increased with dietary selenium level but plateaued when supplementation reached 0.40 mg/kg, suggesting serum selenium homeostasis at this level. Supplementation with 0.40 and 0.80 mg/kg selenium significantly increased serum GSH content and SOD and GSH-Px activities while decreasing MDA content, consistent with previous reports. However, higher selenium levels are not necessarily better; 0.40 mg/kg provided optimal antioxidant capacity in this study.

3.3 Effects of Dietary Selenium Level on Dynamic Changes in Egg Selenium Content With socioeconomic development, consumer demands for egg products have expanded beyond quantity to include quality, particularly selenium-enriched eggs. Research demonstrates that increasing dietary selenium produces selenium-enriched eggs [23], with egg selenium content rising as dietary selenium increases [24]. Payne et al. [17] reported that supplementing a basal diet (0.10 mg/kg selenium) with 0, 0.15, 0.30, 0.60, and 3.00 mg/kg selenium yeast for 28 days yielded egg selenium contents of 0.249, 0.366, 0.495, 0.670, and 2.207 mg/kg, respectively. Li et al. [25] found that egg selenium peaked on day 14 with 0.5 mg/kg selenium yeast supplementation, consistent with He et al. [26]. He et al. [9] reported that 0.25 and 0.50 mg/kg supplemental selenium increased yolk selenium by 43.81% and 103.14% on day 14, and by 48.70% and 107.81% on day 28, respectively. Pan [27] and Hu et al. [28] observed that egg selenium increased significantly with supplementation duration. These studies focused on continuous supplementation, but the dynamics of selenium deposition and retention after withdrawal remain unreported. In this study, 0.20–0.80 mg/kg selenium supplementation significantly increased egg selenium content, which peaked on day 9 in Groups II, III, and IV. During days 1–4 post-withdrawal, Groups II, III, and IV maintained significantly higher egg selenium than the control. During days 5–8 post-withdrawal, Groups III and IV remained significantly elevated. By day 9 post-withdrawal, all groups returned to control levels. These results indicate that 0.40 mg/kg dietary selenium achieves steady-state egg selenium content. Compared with 0.20 mg/kg, 0.40–0.80 mg/kg selenium extended egg selenium retention by 4–5 days after withdrawal. The prolonged retention in high-selenium groups may result from elevated selenium storage in blood, muscle, and liver, which continues to be transferred to eggs after selenium withdrawal, though the specific mechanism requires further investigation.

Conclusion

1. Supplementing a basal diet containing 0.15 mg/kg selenium with 0.20–0.80 mg/kg selenium yeast did not significantly affect laying performance or egg quality of peak-laying Linwu ducks.
2. Supplementation with 0.40 and 0.80 mg/kg selenium yeast increased egg selenium content, serum selenium levels, and antioxidant capacity while prolonging selenium retention in eggs.
3. Considering all factors comprehensively, the optimal dietary selenium supplemental level for peak-laying Linwu ducks is 0.40 mg/kg as selenium yeast.

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Note: Figure translations are in progress. See original paper for figures.

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