

Effects of Dietary Supplementation of Vitamin E and Selenium Yeast on Laying Performance, Hatching Performance, and Vitamin E and Selenium Deposition in Eggs of Yellow-Feathered Broiler Breeder Chickens: Postprint

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Abstract

This experiment aimed to investigate the effects of dietary vitamin E and selenium yeast supplementation on laying performance, hatching performance, and vitamin E and selenium (Se) deposition in eggs of yellow-feathered broiler breeder chickens. The experiment adopted a 3×3 two-factor design, with three vitamin E levels (0, 20, 40 IU/kg) and three Se levels (0, 0.15, 0.30 mg/kg). A total of 864 32-week-old fast-growing Lingnan yellow-feathered broiler parent stock breeder hens were selected and randomly divided into 9 groups based on the principle of consistency in laying rate and body weight, namely VE0Se0, VE0Se0.15, VE0Se0.30, VE20Se0, VE20Se0.15, VE20Se0.30, VE40Se0, VE40Se0.15, and VE40Se0.30 groups, with 6 replicates per group and 16 hens per replicate. The experimental period lasted 8 weeks. The results showed: 1) Dietary vitamin E and Se levels had no significant effects on body weight gain, average daily egg production, feed-to-egg ratio, average egg weight, laying rate, broken egg rate, and unqualified egg rate of yellow-feathered broiler breeder chickens ($P>0.05$). 2) Dietary vitamin E and Se levels had no significant effects on egg length, egg width, albumen height, yolk color, Haugh unit, yolk weight, shell weight, and shell thickness of yellow-feathered broiler breeder chickens ($P>0.05$). 3) Dietary vitamin E and Se levels had no significant effects on healthy chick hatch weight, hatchability, weak chick rate, and fertilization rate of yellow-feathered broiler breeder chickens ($P>0.05$). 4) Dietary vitamin E level had a significant effect on plasma MDA content of yellow-feathered broiler breeder chickens ($P<0.05$), with plasma MDA content showing a decreasing trend as dietary vitamin E level increased, where plasma MDA content at 40 IU/kg vitamin E was significantly lower than that at 0 IU/kg ($P<0.05$); dietary vitamin E and Se levels

had a significant interaction effect on plasma MDA content ($P < 0.05$), where plasma MDA content in the VE40Se0 group was significantly lower than that in the VE0Se0.15 and VE0Se0.30 groups ($P < 0.05$). 5) With increasing dietary vitamin E and Se levels, vitamin E and Se contents in eggs of yellow-feathered broiler breeder chickens increased significantly ($P < 0.05$); dietary vitamin E and Se levels had a significant interaction effect on vitamin E and Se contents in eggs ($P < 0.05$). The results suggest that vitamin E can improve the antioxidant capacity of yellow-feathered broiler breeder chickens, and dietary vitamin E and Se levels can significantly affect vitamin E and Se contents in eggs; it is recommended that under conditions without any stress factors and without using feed ingredients from Se-deficient regions, vitamin E and Se need not be additionally supplemented in corn soybean meal diets for yellow-feathered broiler breeder chickens.

Full Text

Effects of Dietary Vitamin E and Selenium Yeast on Laying Performance, Hatching Performance, and Vitamin E and Selenium Deposition in Eggs of Yellow-Feathered Broiler Breeders

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Abstract: This study investigated the effects of dietary vitamin E (VE) and selenium yeast on laying performance, hatching performance, and vitamin E and selenium (Se) deposition in eggs of yellow-feathered broiler breeders. A 3×3 factorial design was employed with three vitamin E levels (0, 20, 40 IU/kg) and three Se levels (0, 0.15, 0.30 mg/kg). Eight hundred sixty-four 32-week-old fast-growing Lingnan yellow-feathered parental broiler breeder hens were randomly divided into nine groups (VE0Se0, VE0Se0.15, VE0Se0.30, VE20Se0, VE20Se0.15, VE20Se0.30, VE40Se0, VE40Se0.15, and VE40Se0.30) with six replicates per group and sixteen hens per replicate, based on similar laying rate and body weight. The experimental period lasted 8 weeks.

The results showed: (1) Dietary vitamin E and Se levels had no significant effects on body weight gain, average daily egg mass, feed-to-egg ratio, average egg weight, laying rate, broken egg rate, or unqualified egg rate of yellow-feathered broiler breeders ($P > 0.05$). (2) Dietary vitamin E and Se levels had no significant effects on egg long diameter, egg short diameter, albumen height, yolk color, Haugh unit, yolk weight, eggshell weight, or eggshell thickness ($P > 0.05$). (3) Di-

etary vitamin E and Se levels had no significant effects on healthy chick hatching weight, hatchability, weak chick rate, or fertilization rate ($P>0.05$). (4) Dietary vitamin E level significantly affected plasma malondialdehyde (MDA) content ($P<0.05$). Plasma MDA content showed a decreasing trend with increasing dietary vitamin E level, with the 40 IU/kg group being significantly lower than the 0 IU/kg group ($P<0.05$). Dietary vitamin E and Se levels showed a significant interaction effect on plasma MDA content ($P<0.05$), with the VE40Se0 group being significantly lower than the VE0Se0.15 and VE0Se0.30 groups ($P<0.05$). (5) Egg vitamin E and Se contents increased significantly with increasing dietary vitamin E and Se levels ($P<0.05$), and dietary vitamin E and Se levels showed a significant interaction effect on egg vitamin E and Se contents ($P<0.05$). In conclusion, vitamin E can improve the antioxidant capacity of yellow-feathered broiler breeders, and dietary vitamin E and Se levels significantly affect egg vitamin E and Se contents. It is recommended that corn-soybean meal diets for yellow-feathered broiler breeders do not require additional vitamin E or Se supplementation when no stress factors are present and feed ingredients from Se-deficient regions are not used.

Keywords: yellow-feathered broiler breeder; vitamin E; selenium yeast; egg hatching

Introduction

Vitamin E (VE) and the trace element selenium (Se) are two essential micronutrients for chickens. Vitamin E plays roles in alleviating poultry stress, improving growth performance, and enhancing reproductive performance [1-3]. Selenium is a functional component of various selenoproteins in animal bodies, capable of scavenging oxidative substances and exerting antioxidant and immune-enhancing effects [4-5]. Deficiency in either of these micronutrients can cause severe stress responses in breeding chickens [6]. Currently, research on the dietary requirements of vitamin E and Se for breeding hens is limited. The NRC (1994) proposed minimum requirements of 10 IU/kg for vitamin E and 0.05 mg/kg for Se in diets for laying breeder hens. Zhang [6] suggested that 4 mg/kg of dietary vitamin E could meet normal production and reproductive performance needs in laying breeder hens, and that adding 0.35 mg/kg of organic Se could improve egg production. The *Feeding Standard of Chickens* (NY/T 33–2004) recommends vitamin E requirements of 18, 9, 9, and 27 IU/kg for pre-laying hens (1-6 weeks), pre-laying hens (7-18 weeks), onset of lay (19 weeks to lay), and laying hens, respectively, with a Se requirement of 0.27 mg/kg for all stages. Different breeds, age stages, and rearing conditions result in varying requirements for dietary vitamin E and Se. Our research laboratory has long studied the nutritional requirements of yellow-feathered broiler breeders, having previously investigated requirements for protein and energy [7], calcium [8], phosphorus [9], zinc [10], lysine [11], methionine [12], isoleucine [13], and vitamin A [14]. Building on these studies, this experiment investigated the re-

quirements for vitamin E and Se during the laying period of yellow-feathered broiler breeders to fill research gaps and provide scientific basis for establishing nutritional standards and practical production guidelines.

1. Materials and Methods

1.1 Experimental Animals and Design A 3×3 factorial design was employed with three vitamin E levels (0, 20, 40 IU/kg) and three Se levels (0, 0.15, 0.30 mg/kg). Eight hundred sixty-four 32-week-old fast-growing Lingnan yellow-feathered parental broiler breeder hens were randomly divided into nine groups (VE0Se0, VE0Se0.15, VE0Se0.30, VE20Se0, VE20Se0.15, VE20Se0.30, VE40Se0, VE40Se0.15, and VE40Se0.30) with six replicates per group and sixteen hens per replicate, based on similar laying rate and body weight.

1.2 Experimental Diets A corn-soybean meal basal diet was used. Dietary vitamin E and Se levels in each group were formulated according to the experimental design, with other nutrients referenced to the *Feeding Standard of Chickens* (NY/T 33–2004) and formulated based on the Chinese Feed Composition and Nutritional Value Table (26th edition) and measured values. The basal diet composition and nutrient levels are shown in Table 1. The basal diet contained 3.19 mg/kg vitamin E and 0.12 mg/kg Se. During diet preparation, vitamin E and Se were added at different levels to equally replace carrier corn-cob meal in the premix. Except for vitamin E and Se levels, all dietary nutrient levels were identical across groups. Selenium yeast was provided by Angel Yeast Co., Ltd. (Se content: 2,000 mg/kg). Vitamin E was vitamin E acetate with 50% active content.

1.3 Management Experimental hens were housed in the breeder experimental facility of the Animal Nutrition Research Laboratory at Guangdong Academy of Agricultural Sciences. A three-tier step cage system was used with two hens per cage. Hens were fed once daily at a rate of 130 g per hen per day and provided ad libitum access to water via nipple drinkers. During the experiment, lighting was maintained at 16 h per day, with natural ventilation and clean housing conditions. Daily maximum and minimum temperatures at 06:00, 12:00, and 18:00, humidity, and weather conditions were recorded. Hen health status was observed daily, with culling and mortality recorded. Hens were managed and immunized according to conventional procedures. The pre-trial period lasted 7 days, followed by an 8-week formal experimental period. Prior to the experiment, hens were fed diets meeting the nutrient levels of the *Feeding Standard of Chickens* (NY/T 33–2004). During the final two weeks, artificial insemination was performed at 16:00 every two days at a dose of 30 L per hen, with a rooster-to-hen ratio of 1:30.

1.4 Measurements

1.4.1 Laying Performance At the start and end of the experiment, feed was withdrawn at 20:00 while water was provided, and hens were weighed by replicate at 08:00 the following day to calculate body weight gain and feed-to-egg ratio. Feed-to-egg ratio (%) = (feed intake during experiment / egg mass during experiment) \times 100. During the experiment, daily records were kept for egg number, daily egg mass, broken eggs, and unqualified eggs (including cracked, soft-shelled, sand-shelled, misshapen, double-yolk, and shell-less eggs). Laying performance was calculated as follows:

- Laying rate (%) = (number of eggs / number of hens) \times 100
- Average daily egg mass (g) = laying rate \times average egg weight
- Average egg weight (g) = total egg mass / total number of eggs
- Broken egg rate (%) = (number of broken eggs / number of eggs) \times 100
- Unqualified egg rate (%) = (number of unqualified eggs / number of eggs) \times 100

1.4.2 Egg Quality At the end of weeks 4 and 8, four eggs were randomly selected from each replicate for egg quality analysis. Eggshell thickness was measured at three points (blunt end, middle, and pointed end) after membrane removal using a micrometer, and the average was calculated. Eggs were then broken, contents removed, and shells wiped clean before weighing. Albumen height and yolk color were measured using a multifunctional egg quality analyzer (ORKA, Israel). Yolks were separated using a yolk separator and weighed. Haugh unit was calculated as: Haugh unit = $100 \times \log(H - 1.7W^{0.37} + 7.6)$, where H is albumen height and W is egg weight.

1.4.3 Hatching Performance Starting from week 7, 50 eggs were collected from each replicate for hatching. Clear eggs, hatched chicks, healthy chicks, and chick hatching weight were recorded. Hatching performance was calculated as:

- Fertilization rate (%) = [(number of incubated eggs - number of clear eggs) / number of incubated eggs] \times 100
- Hatchability (%) = (number of hatched chicks / number of incubated eggs) \times 100
- Weak chick rate (%) = (number of weak chicks / number of incubated eggs) \times 100

1.4.4 Plasma Biochemical Indices Based on laying performance, two hens per replicate with laying rates close to the average were selected at the end of the experiment. Blood (6 mL) was collected from the wing vein, and plasma was separated and stored at -20°C. Plasma malondialdehyde (MDA) content and total superoxide dismutase (T-SOD) activity were measured using a Beckman automatic biochemical analyzer (CX5, Beckman Instruments, USA) and corresponding kits following the manufacturer's instructions. Kits were purchased from Nanjing Jiancheng Bioengineering Institute.

1.4.5 Vitamin E and Se Content in Eggs After the experiment, hens continued to be fed experimental diets for one additional day. One egg per replicate was collected from the VE0Se0, VE40Se0, VE0Se0.30, and VE40Se0.30 groups for vitamin E and Se analysis. Se content in diets and eggs was determined by hydride-atomic fluorescence spectrometry (GB 5009–2010), and vitamin E content was determined by high-performance liquid chromatography (GB 5009–2003).

1.5 Statistical Analysis Data were analyzed using the GLM procedure in SPSS 17.0 software for two-way ANOVA. When significant effects were detected, Duncan's multiple comparison test was performed. The statistical model included vitamin E level, Se level, and their interaction. Data are expressed as "mean \pm standard error (mean \pm SE)" with $P < 0.05$ as the significance threshold.

2. Results

2.1 Effects of Dietary Vitamin E and Se Levels on Laying Performance

As shown in Table 2, dietary vitamin E and Se levels had no significant effects on body weight gain, average daily egg mass, feed-to-egg ratio, average egg weight, laying rate, broken egg rate, or unqualified egg rate of yellow-feathered broiler breeders ($P > 0.05$). No significant interaction between dietary vitamin E and Se levels was observed for laying performance ($P > 0.05$).

2.2 Effects of Dietary Vitamin E and Se Levels on Egg Quality

As shown in Tables 3 and 4, at the end of weeks 4 and 8, dietary vitamin E and Se levels had no significant effects on egg long diameter, egg short diameter, albumen height, yolk color, Haugh unit, yolk weight, eggshell weight, or eggshell thickness ($P > 0.05$). No significant interaction between dietary vitamin E and Se levels was observed for egg quality ($P > 0.05$).

2.3 Effects of Dietary Vitamin E and Se Levels on Hatching Performance

As shown in Table 5, dietary vitamin E and Se levels had no significant effects on healthy chick hatching weight, hatchability, weak chick rate, or fertilization rate ($P > 0.05$). No significant interaction between dietary vitamin E and Se levels was observed for hatching performance ($P > 0.05$).

2.4 Effects of Dietary Vitamin E and Se Levels on Plasma Biochemical Indices

As shown in Table 6, dietary vitamin E level significantly affected plasma MDA content ($P < 0.05$). Plasma MDA content showed a decreasing trend with increasing dietary vitamin E level, with the 40 IU/kg group being significantly lower than the 0 IU/kg group ($P < 0.05$). Dietary vitamin E and Se levels showed a significant interaction effect on plasma MDA content ($P < 0.05$), with the VE40Se0 group being significantly lower than the VE0Se0.15 and VE0Se0.30 groups ($P < 0.05$).

2.5 Effects of Dietary Vitamin E and Se Levels on Vitamin E and Se Content in Eggs As shown in Table 7, egg vitamin E and Se contents increased significantly with increasing dietary vitamin E and Se levels ($P < 0.05$). Dietary vitamin E and Se levels showed a significant interaction effect on egg vitamin E and Se contents ($P < 0.05$).

3. Discussion

3.1 Effects of Dietary Vitamin E and Se Levels on Laying Performance Cai et al. [15] found that feeding Hy-Line Brown laying hens diets with different selenium yeast levels had no significant effects on laying rate, average egg weight, or feed-to-egg ratio. Similarly, feeding Beijing You chickens and Roman \times White Leghorn crossbred layers diets containing 0.3-5.0 mg/kg selenium yeast showed no significant effects on laying performance [16]. Studies on Daheng breeder hens and Leghorn chickens also found no significant effects of dietary Se level on laying performance [17-18]. These results indicate that dietary Se level has no significant effect on laying performance of breeder hens, consistent with our findings. Guo et al. [19] reported that dietary vitamin E supplementation at 4, 14, and 24 mg/kg had no significant effects on laying performance of breeder hens. Zhang et al. [20] found that feeding Hy-Line Brown breeder hens diets with 0-320 IU/kg vitamin E significantly improved laying rate at 160 and 320 IU/kg, while 0-80 IU/kg had no significant effects. These results suggest that high-dose vitamin E can improve laying rate, while low-dose vitamin E has no significant effect. Our study showed no significant effects of different vitamin E levels on laying performance of yellow-feathered broiler breeders, possibly because our vitamin E supplementation levels were similar to the recommendation in the *Feeding Standard of Chickens* (NY/T 33-2004) (27 IU/kg), which may not be high enough to demonstrate the performance-enhancing effects of vitamin E.

3.2 Effects of Dietary Vitamin E and Se Levels on Egg Quality and Vitamin E and Se Deposition Our results showed that different dietary vitamin E and Se levels had no significant effects on egg quality but significantly affected vitamin E and Se deposition in eggs, with egg contents increasing as dietary levels increased. Tan et al. [21] found that dietary supplementation with 1.0-2.0 mg/kg Se and 83.32-166.64 IU/kg vitamin E had no significant effects on egg weight but significantly increased Se and vitamin E deposition in eggs. Other studies also reported that dietary selenium yeast supplementation had no significant effects on egg quality but significantly increased yolk Se content [22-25]. It has been suggested that dietary vitamin E supplementation has no significant effects on yolk color, Haugh unit, eggshell thickness, or eggshell weight but can increase vitamin E deposition in yolk [26-27]. Our results indicate that increasing dietary vitamin E and Se levels can enhance their contents in eggs.

3.3 Effects of Dietary Vitamin E and Se Levels on Hatching Performance Studies have shown that dietary vitamin E supplementation at 0, 20,

40, 80, and 160 IU/kg had no significant effects on hatchability, fertilization rate, or progeny hatching weight, while 320 IU/kg significantly improved fertilization rate and hatchability [20]. Supplementing broiler breeder hen diets with 100 IU/kg vitamin E increased chick hatching weight [28]. Wahyuni et al. [29] reported that dietary vitamin E improved hatchability of breeder eggs. However, other research suggested that 30 mg/kg vitamin E improved fertilization rate but had no significant effects on hatchability or chick hatching weight [30]. These results indicate that high-dose vitamin E can improve hatching performance, while our lower supplementation levels showed no significant effects, similar to the laying performance results. In our study, different Se levels had no significant effects on hatching performance. Zhang et al. [31] found that 0.5 mg/kg organic Se in Guizhou Yellow breeder hen diets had no significant effects on fertilization rate or hatchability. However, other studies reported that total dietary Se of 0.9-1.3 mg/kg significantly improved fertilization rate and hatchability [32], while hatchability decreased when methionine-Se exceeded 0.25 mg/kg [17]. Overall, inconsistent results regarding Se effects on hatching performance may be related to breed, age, rearing conditions, and Se source.

3.4 Effects of Dietary Vitamin E and Se Levels on Plasma Biochemical Indices

Vitamin E can enhance antioxidant enzyme activity, protect cell membranes from free radical damage, and effectively improve antioxidant function in poultry [33-34]. Selenium not only serves as a component of antioxidant enzyme systems, improving glutathione peroxidase (GSH-Px) and catalase (CAT) activities to exert antioxidant effects, but also directly scavenges free radicals in blood to resist free radical damage [35-37]. Our results showed that dietary vitamin E level significantly affected plasma MDA content, which decreased with increasing dietary vitamin E level, and that dietary vitamin E and Se levels showed a significant interaction effect on plasma MDA content. Studies have reported that dietary vitamin E supplementation can increase T-SOD and GSH-Px activities and decrease MDA content [38-39]. Other research suggests that vitamin E supplementation is particularly effective in alleviating stress under cold or immune stress conditions [6,40]. Our results indicate that increasing dietary vitamin E level can reduce plasma MDA content and alleviate oxidative stress in yellow-feathered broiler breeders.

3.5 Discussion on Vitamin E and Se Requirements of Yellow-Feathered Broiler Breeders

Our results showed that dietary supplementation with 0, 20, or 40 IU/kg vitamin E had no significant effects on laying performance, egg quality, or hatching performance, indicating no negative effects from omitting vitamin E supplementation. Current research on vitamin E in breeder or laying hens mostly focuses on effects of excess supplementation, with few studies on complete omission. Zhang et al. [20] found that 0, 20, and 40 mg/kg vitamin E had no significant effects on laying performance, and 0-160 mg/kg had no significant effects on hatching performance. Han [41] suggested that vitamin E requirements for laying hens are very low without stress factors,

and that mild, short-term immune stress only causes temporary feed intake reduction without significantly affecting laying performance. Possible reasons include: (1) Vitamin E is fat-soluble and can be stored in large amounts in the body with slow excretion. Before our experiment, hens were fed diets meeting the *Feeding Standard of Chickens* (NY/T 33–2004) with adequate vitamin E, resulting in body stores. (2) Vitamin E primarily functions in antioxidant activity and promoting reproductive organ development. As breeder age increases, disease resistance improves and reproductive organs mature, diminishing the role of vitamin E in enhancing immunity and improving laying performance. Studies show that vitamin E stress-alleviating effects are significant only when hens are under stress [42-43].

Our results also showed that dietary supplementation with 0, 0.15, or 0.30 mg/kg Se had no significant effects on laying performance, egg quality, or hatching performance, indicating no negative effects from omitting Se supplementation. The NRC (1994) established a minimum Se requirement of 0.05 mg/kg for broiler breeder hens, while our basal diet contained 0.12 mg/kg Se, indicating that diets formulated with feed ingredients from non-Se-deficient regions already meet the Se requirement of yellow-feathered broiler breeders. In practice, Se deficiency mainly occurs in growing broilers, rarely in laying breeders, suggesting that normal corn-soybean meal diets (using non-deficient ingredients) can satisfy Se requirements under normal physiological conditions.

Dietary trace elements consist of two parts: those from feed ingredients and those added as supplements. However, in practice, trace elements from feed ingredients are often overlooked, leading to excessive supplementation, which wastes resources, increases feed costs, and pollutes the environment. Many studies have begun addressing excessive trace element supplementation and baseline levels in basal diets [44-46]. Our basal diet contained 3.19 mg/kg vitamin E and 0.12 mg/kg Se, with no significant differences in laying performance, egg quality, or hatching performance compared to supplemented groups. Therefore, under conditions without stress factors and without using feed ingredients from Se-deficient regions, the vitamin E and Se contents in corn-soybean meal basal diets already meet the requirements of yellow-feathered broiler breeders.

In summary, vitamin E can improve the antioxidant capacity of yellow-feathered broiler breeders, and dietary vitamin E and Se levels significantly affect egg vitamin E and Se contents. It is recommended that corn-soybean meal diets for yellow-feathered broiler breeders do not require additional vitamin E or Se supplementation when no stress factors are present and feed ingredients from Se-deficient regions are not used.

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