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Effects of Vitamin E and Selenium on Growth Performance, Antioxidant Capacity, and Metabolic Parameters in Growing Furring Blue Foxes (*Alopex lagopus*)

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Abstract
The objective of this study was to determine whether different dietary vitamin E (VE) and selenium (Se) levels affect the nutrient digestibility, production performance, and antioxidant abilities of growing furring blue foxes. A 4 × 2 factorial arrangement that included 4 levels of VE (0, 100, 200, or 400 mg/kg diet from α-tocopherol acetate) and 2 levels of Se (0 or 0.2 mg/kg diet from glycine selenium) was performed from mid-September to pelting. A metabolism study was conducted for four days starting at the 30th day of the trial. Serum samples were collected at the last day of the study. The results showed that supplementation of growing furring blue fox diets with VE and Se significantly affected the average daily gain (ADG), average daily feed intake, and feed conversion ratio (F:G) ($P < 0.05$). Dietary Se supplementation enhanced protein and fat digestibility of male blue foxes. There were significant effects of different VE and Se levels in diets on serum antioxidant parameters and metabolic parameters of blue foxes ($P < 0.05$). In conclusion, this research indicated that dietary supplementation with VE improved ADG and F:G of blue foxes. Addition of VE and Se to blue fox diets increased the antioxidant capacity of blue foxes. The diet with high VE and Se supplementation reduced glucose and triglycerides concentrations in serum. The present study found that growing furring blue foxes had increased growth performance and antioxidant abilities when fed diets with 200 mg VE/kg and nearly 0.1 mg Se/kg.

Keywords Blue foxes · Selenium · Vitamin E · Growth performance · Antioxidant capacity · Metabolic parameters

Introduction

Vitamin E (VE) and selenium (Se) have been recognized for several years as nutrients critical for optimal livestock growth, productivity, and health [1]. In animal physiology, VE and Se work together. VE is recognized as the first line of defense against lipid peroxidation, protecting polyunsaturated fatty acids (PUFA) in cell membranes by quenching free radical activity in biomembranes at the early stage of free radical attack [2, 3]. The first line of antioxidant defense, which is composed of VE, other fat-soluble antioxidants and metal-binding proteins in the cell, is not sufficient to completely prevent free radical formation [4]. Therefore, the second level of defense consisting of chain-breaking antioxidants, e.g., glutathione (GSH) and selenoproteins, plays an important role [5, 6].

However, the exact interrelationship between VE and Se is unknown and may vary between species. Many researchers have focused on the beneficial effects of VE and Se on the immune responses of domestic animals. Ebeid et al. [7] reported that supplemental dietary VE and Se enhanced growth performance, serum antioxidative status, and immune responsiveness in growing rabbits. In chickens, VE and Se deficiencies cause multiple alterations in the antioxidant system and adversely affect the redox state of the superficial pectoralis muscle [8]. Supplementation with Se at 3 and 30 days of life increased dairy calve neutrophil function and glutathione peroxidase activity and reduced the incidence of health disorders, indicating that dietary Se at adequate levels supports the antioxidant system of the body [9]. Comparing the effects of different dietary Se sources, Falk et al. [10] found that organic Se positively influences the immune response and antioxidant capacity of pigs.

Because of the special physiological characteristics of fur-bearing animals, the feed resources contain high levels of

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PUFA [11]. However, when fox or mink diets contain rancid fats or are high in PUFA, the animals are subjected to yellow fat disease [11]. Minks fed unsaturated fatty acid diets require an adequate supply of VE, which effectively protects the growing mink and prevents yellow fat disease [12, 13]. VE deficiency may also lead to yellow fat disease of foxes [14]. Previous studies have indicated that dietary Se supplementation may prevent lesions caused by VE deficiency in mink [15] and in other carnivorous animals, especially during growth [16]. Müller et al. [17] found an essential role of Se in the prevention of lipid and protein oxidation in rabbits, especially during periods of VE deficiency.

Several recent studies have shown that VE or Se may have an adverse effect on the body. Miller et al. [18] performed a meta-analysis and found that high dosage VE supplementation may increase all-cause mortality. Aburto and Britton [19] found that high levels of VE in chicken diets negatively affect the utilization of vitamin D_3 (VD_3) when VD_3 is present at a marginal level. Moreover, Kim and Mahan [20] suggested that dietary Se from inorganic or organic sources is toxic at ≥ 5 ppm Se in growing finishing pigs. Nolan et al. [21] indicated that high doses of VE and Se may be deleterious to liver function, especially in the presence of high-fat diets in rats. In vitro studies on lymphocytes from sheep [22], cattle [23], and mice [24] showed that high levels of Se and VE added to these cells inhibit their responses and have a toxic effect.

In conclusion, VE and Se are essential for domestic animals, including fur-bearing animals, but excessive addition of VE and Se might be harmful to these organisms. There are no reports on the requirement of VE and Se in blue fox diets. Hence, it is necessary to explore the appropriate concentrations of VE and Se in blue fox diets. The objectives of the present research were to study the effects of VE and Se on the growth performance, blood antioxidant capacity, and blood metabolism abilities of blue foxes as well as to elucidate the VE and Se nutritional requirements of blue foxes.

Materials and Methods

Animal Diets, Management, and Experimental Design

In the present study, there were 160 blue foxes at 120 ± 5 days of age randomly assigned to eight groups with an equal number of males and females in each group. A 4 × 2 factorial experiment with four supplemental levels of VE (0, 100, 200, or 400 mg/kg dry matter) from α-tocopherol acetate (effective substance concentration of 50%) and two supplemental levels of Se (0 or 0.2 mg/kg) from glycine selenium with 1% Se concentrations (measured concentration of 0.86 ± 0.07 g/kg) was conducted. All diets met the nutrient requirements of growing foxes [11]. The composition and chemical analysis of the basal and the other trial diets are shown in Table 1. The experimental period was from mid-September to pelting in December. The first 7 days comprised the adaptation period, during which animals were allowed to be accustomed to the experimental diets based on dry and powdery components mixed with water. All foxes were individually housed in conventional cages (1 m × 0.8 m × 0.8 m) in two row sheds. The diets were provided twice a day namely at 7:00 and 15:00, and water was provided ad libitum. The use of animals in the present study followed guidelines similar to those provided by the Canadian Council on Animal Care [25].

Table 1 Ingredient and chemical composition of the basal diet (g/kg as dry matter)

	Contents
Ingredients	
Extruded corn	400
Soybean meal	239.6
Corn gluten meal	100
Fish meal	60.0
Meat and bone meal	88.0
Chicken oil	100
Met	2.4
Premix	10
Total	1000
Nutrient levels	
Crude protein	280
Crude fat	117.1
Ca	12.6
Total P	7.8
Ca/P	1.62
Se (mg/kg dry matter)	0.096
VE	ND

Ca, calcium; *Total P*, total phosphorus; *Ca/P*, calcium/total phosphorus; *Se*, selenium; *VE*, vitamin E; *ND*, not detected

Premix provided following per kilogram of diet: Fe (as ferrous sulfate) 80 mg; Zn (as zinc sulfate) 80 mg; Mn (as manganese sulfate) 15 mg; Cu (as copper sulfate) 10 mg; I (as calcium iodate) 0.5 mg; Co (as cobalt sulfate) 0.3 mg; VA 12500 IU; VD_3 2500 IU VK_3 2 mg; VB_1 20 mg; VB_2 10 mg; VB_6 10 mg; VB_{12} 0.1 mg; niacin 40 mg; pantothenic acid 20 mg; folic acid 1 mg; biotin 0.5 mg; choline 400 mg; VC 120 mg

The trial dietary supplementation with 0, 100, 200 and 400 mg VE/kg contained 0, 50, 100, and 200 mg VE/kg, respectively and with 0 and 0.2 mg Se/kg contained 0.096 and 0.268 mg Se/kg

Sample Collection and Preparation

The sample of the basal diet was randomly sampled by dietary sampler and thoroughly mixed. This sample and supplementations were shrunk by the quarter method. Three replicates

were measured for each sample. The amounts of feed given and that of leftover feed collected daily during the experimental period were weighed. The body weight (BW) of foxes was recorded in the morning before feeding on the 1st, 30th, and 60th day. After weighing on day 30 of the trial, 14 healthy blue foxes (7 females and 7 males) were randomly weighed before feeding and selected for digestibility measurements. Feces, feed refusals, and urine from the four consecutive days were collected daily. To feces, 10% sulfuric acid was added in accordance with 5% of the fresh weight and stored at − 20 °C. At the end of the experiment, all feces from one animal were mixed. Thereafter, 10% feces of the gross weight as well as feed refusals were dried to a constant weight at 60 ± 5 °C and then ground through a 40-mesh screen. The weight data of the whole feces and feed refusals were collected before and after drying. Urine was preserved with 10% sulfuric acid and 4 drops of toluene, and the urine volume of each animal was recorded. The mixed urine samples were filtered through filter paper, and 10 mL of each filtrate were stored at − 20 °C.

For final blood sampling, twelve healthy blue foxes (6 females and 6 males) were randomly selected after weighing on the day of pelting. After the blue fox was paralyzed with chlorinated succinylcholine, 5 mL blood was collected by heart puncture using serum separation tubes (BD-Pharmingen, USA). The blue foxes were then killed by an overdose of succinylcholine and pelted by skilled workers. Blood samples were centrifuged at 3500 r/min for 10 min, placed into 1.5-mL centrifuge tubes and stored at − 20 °C until analysis.

Analysis of Diets, Feces, and Urine Samples

The air-dried diets and feces were analyzed for dry matter (DM) and nitrogen (N) according to the procedures of AOAC [26]. Crude fat (CF) of feed and feces was determined by a diethyl ether extraction-submersion method [26]. The dietary calcium (Ca) and total phosphorus (total P) were estimated by the AOAC method [26].

The analysis of dietary and fecal Se was performed according to AOAC [27]. The Se standard solution was a selenite standard solution (1000 μg Se/mL in 1% HNO_3; commercially available from the National Standard Research Center of China), which was diluted to 10 μg Se/L with 0.1 M HCl. The selenite solution was further diluted to 0, 1, 2, 4, and 8 μg Se/L with 0.1 M HCl. Dietary and fecal samples (0.2 g each) were transferred into a sealed polytetrafluoroethylene beaker, and 8 mL of concentrated (99.99%) nitric acid and several glass beads were carefully added. The samples were digested using a microwave-assisted (Anton Paar, Austria) high-pressure digestion system until the samples were in solution and nitric oxide fumes subsided according to the instructions. The solutions were cooled at room temperature, and a constant volume of 50 mL was obtained by adding 1% nitric acid. The urine samples (5 mL each) were treated according to methods published by Błażewicz et al. [28]. Briefly, the liquid sample, 9.4 mL of deionized water and 0.1 mL of concentrated (99.99%) nitric acid were placed in a polytetrafluoroethylene beaker and subjected to the same procedures described above for the solid samples. The processed diet, feces, serum, and urine samples (10 mL) were analyzed using an atomic fluorescence spectrometer (AFS-9x, Jitian Instruments, Beijing, People's Republic of China) according to AOAC [27] using the supporting atomic fluorescence spectrometer software.

The concentrations of VE (α-tocopherol) in diets were measured according to the China Standard Press method (GB17812-2008). In detail, 10 g of the sample (accurately to 0.0001 g) and 50 mL of ascorbic acid ethanol solution were placed in a 250-mL round bottom flask, heated in a water bath until boiling point, washed with nitrogen and cooled slightly. Next, 10 mL of potassium hydroxide solution was added to the sample, mixed evenly, and boiled to obtain saponification under dinitrogen (N_2) flow for 30 min. The solution was washed with 5 mL of ethanol and 5 mL of water from the top of the condenser tube and then cooled to approximately 40 °C. The saponification liquids were transferred into a 500-mL funnel that contained 100 mL of anhydrous ether. The flask was rinsed 2 to 3 times with distilled water, shaken for 2 min, and allowed to rest in a static state. The water phase was transferred into a new separating funnel and extracted twice with ether. The ether phases were combined, dehydrated by adding anhydrous sodium sulfate, and transferred to a 250-mL brown volumetric flask. After dissolving, butylated hydroxytoluene (100 mg) was then added into the volumetric flask and fixed with ether to the scale. A certain volume of liquid was placed in a rotating evaporator and evaporated to dry in a water bath at approximately 50 °C. The residue was dissolved in methanol to 10 mL and filtered with a 0.45-μm membrane before analysis.

Analysis of Serum Samples

Prior to the measurement of the Se concentration, serum samples (0.5 mL each) were treated as described for urine samples. The serum VE concentrations were determined according to Alvarez and De [29]. In brief, a plasma sample (0.5 mL) and 400 μL of ethanol were placed in 15-mL polypropylene tubes and vortex mixed for 10 s. The mixture was extracted with 2 mL of *n*-hexane/dichloromethane (90/10, *v*/*v*) by vortex mixing for 60 s and centrifugation at 3000×*g* for 5 min. The *n*-hexane/dichloromethane was carefully transferred to a glass test tube and evaporated to dryness under a stream of N_2. The sample was dissolved in 250 μL of methanol and transferred to a micro-vial for the auto-sampler (Supelco, USA).

The α-tocopherol standard solution (1 mL of 1.0 mg/mL) was added into a 10-mL brown volumetric flask, evaporated to dryness under a steam of N_2 and then diluted to 1, 2, 4, 8, and 10 mg/L with methanol to generate a gradient of standard solutions. The

standard solution gradient and diet and serum samples were analyzed by high-performance liquid chromatography (Waters-2695, USA) using a C_{18} column (250 mm × 4.6 mm; 5.0 μm, Waters-XSELECT, USA). The initial mobile phase was methanol:water (95:5) and a flow rate of 1.0 mL/min. The injection volume was 20 μL, and the detector was a photodiode array detector. The concentration of VE was calculated according to the China Standard Press method (GB17812-2008).

The concentrations of serum glucose (GLU), triglyceride (TG), cholesterol (CHO), low-density lipoprotein cholesterol (LDL), highly density lipoprotein cholesterol (HDL), creatinine (CRE), total protein (TP), and albumin (ALB) (kits supplied by Zhongsheng Beikong Biotechnology LLC, Beijing, People's Republic of China) were analyzed by an automatic biochemistry analyzer (Hitachi 7020, Hitachi High Technologies, Inc., Ibaraki, Japan). The concentrations of blood urea nitrogen (BUN), maleic dialdehyde (MDA), and total glutathione (T-GSH) as well as the enzyme activities of total antioxidant capacity (T-AOC), superoxide dismutase (SOD), glutathione peroxidase (GSH-Px), glutathione S-transferase (GSH-ST), glutathione reductase (GR), thioredoxin reductase (TrxR), and thioredoxin peroxidase (TPX) were determined using respective diagnostic kits (Nanjing Jiancheng Bioengineering Institute, Nanjing, People's Republic of China).

Statistical Analysis

The average daily gain (ADG), average daily feed intake (ADFI), and feed conversion ratio (F:G) were calculated for each animal using the following formulas: ADG = (final weight−initial weight)/days; ADFI = sum of daily feed intake/days; and F:G = ADFI/ADG. The crude protein (CP) concentration in the diets, feces, and urine was calculated as N × 6.25.

The apparent digestibility (AD) coefficient of nutrients was calculated as follows: AD = $(a-b)/a \times 100\%$, where a is the nutrient intake from feed and b is the nutrient excretion in feces. The analysis of N metabolism used the following equations: N deposition = N intake−fecal N− urinary N; net protein utilization (NPU) (%) = N deposition/N intake × 100%; and biological value (BV) (%) = N deposition/(N intake−fecal N) × 100%. Se metabolism-related parameters were calculated with the following equations: total Se excretion = fecal Se excretion + urinary Se excretion; Se deposition = Se intake−total Se excretion; Se digestibility = Se deposition/Se intake × 100%.

The data were subjected to ANOVA in a completely randomized design with 4 × 2 factorial arrangement of treatments using the GLM procedure of SAS 9.2 (SAS Institute Inc., Cary, NC, USA). For all statistical analyses, significance was declared at $P < 0.05$. The following model was considered for analysis:

$$Y_{ijk} = \mu + A_i + B_j + (AB)_{ij} + \varepsilon_{ijk}$$

where Y_{ijk} is the characteristic that was measured, μ is the overall means, A_i is the effect of dietary VE, B_j is the effect of dietary Se, $(AB)_{ij}$ is the effect of the interaction between VE and Se, and ε_{ijk} is the random error term. When interactions were significant, the PDIFF option of LSMEANS in SAS was used to separate the means, and the effects of the main factors were not considered.

Results

Growth Performance and Nutrient Digestibility

The basal diet contained 0.096 ± 0.004 (mean ± variance) mg Se/kg, and VE was not detected, which is shown in Table 1. The trial diets were supplemented with VE 0, 100, 200, and 400 mg, which were contained 0, 50, 100, and 200 mg VE/kg, respectively, and added with 0 and 0.2 mg Se/kg, which included 0.096 and 0.268 mg Se/kg. Different levels of VE and Se in diets significantly affected the growth performance of blue foxes (Table 2). Dietary levels of Se markedly influenced BW of different groups of blue foxes at 60 days ($P < 0.05$), and an interaction of VE and Se was observed ($P < 0.05$). Moreover, BW of male blue foxes was significantly influenced by the concentrations of VE in diets ($P < 0.05$). ADG greatly increased with dietary Se levels in male blue foxes during the first 30 days ($P < 0.05$), but there was no significant effects on female blue foxes. From 30 to 60 days of the feed experiment, ADG of blue foxes was affected by the dietary amounts of VE and Se as well as the interaction of VE and Se levels ($P < 0.05$). Between days 30 and 60, ADFI was impacted by the interaction of VE and Se levels in female blue foxes ($P < 0.05$), and it increased with the amount of Se in male blue fox diets ($P < 0.05$). During the first 30 days, F:G was higher in male blue foxes fed diets supplemented with VE but with Se ($P < 0.05$). Regardless of gender Se supplementation and the interaction of dietary VE and Se influenced F:G in the period from day 30 until day 60 ($P < 0.05$).

There were significant effects of different VE and Se supplementation levels on AD and Se metabolism of growing furring blue foxes which are shown on Table 3. There were no significant effects of different VE and Se supplement levels in diets on female blue foxes AD ($P > 0.05$). Interestingly, adding Se to male blue fox diets enhanced the digestibility of DM, CP, and CF ($P < 0.05$). However, the concentrations of VE and the interaction of VE and Se in male blue fox diets did not influence AD in growing furring male blue foxes ($P > 0.05$). The Se excretions were impacted by the levels of Se in blue fox diets ($P < 0.05$). Fecal and total Se excretions from female blue foxes as well as the total Se excretion from male blue foxes were significantly influenced by the amounts of VE in the diets ($P < 0.05$). The total Se excretion in groups

Table 2 Effects of different vitamin E and selenium supplementation on growth performance of growing furring blue foxes

		VE and Se addition (mg/kg)								SEM	*P* value		
		0 and 0	100 and 0	200 and 0	400 and 0	0 and 0.2	100 and 0.2	200 and 0.2	400 and 0.2		VE	Se	VE × Se
BW (kg)													
1 day	♀	4.91	4.92	4.93	4.98	4.96	4.94	4.98	4.96	0.153	0.994	0.818	0.996
	♂	5.19	5.17	5.13	5.15	5.12	5.10	5.13	5.13	0.153	0.998	0.716	0.993
30 days	♀	5.88	5.63	5.69	5.86	5.71	5.74	5.90	5.89	0.149	0.663	0.681	0.624
	♂	6.23	5.98	5.85	6.10	6.29	6.23	6.28	6.35	0.177	0.636	0.055	0.796
60 days	♀	7.65^{a}	7.79^{a}	7.77^{a}	8.15^{a}	7.87^{a}	6.94^{b}	6.82^{b}	6.71^{b}	0.180	0.065	< 0.0001	0.0004
	♂	7.86^{bc}	8.11^{ab}	8.08^{ab}	8.28^{ab}	8.58^{a}	7.33^{c}	7.36^{c}	7.29^{c}	0.198	0.046	0.003	0.0003
ADG (g)													
1~30 days	♀	32.50	23.89	25.28	29.45	25.00	26.67	30.83	31.11	2.973	0.407	0.768	0.161
	♂	34.44^{abc}	26.94^{bc}	24.17^{c}	31.67^{abc}	39.17^{a}	37.50^{a}	38.34^{a}	40.56^{a}	3.409	0.281	0.0003	0.581
30~60 days	♀	58.89^{b}	71.95^{a}	69.17^{ab}	76.39^{a}	71.95^{a}	40.00^{c}	30.56^{cd}	27.22^{d}	3.686	0.001	< 0.0001	< 0.0001
	♂	54.45^{b}	71.11^{a}	74.45^{a}	72.78^{a}	76.11^{a}	36.94^{c}	36.11^{c}	31.39^{c}	3.545	0.003	< 0.0001	< 0.0001
ADFI (g)													
1~30 days	♀	247.92	261.67	253.13	263.13	257.08	241.04	248.54	246.46	8.292	0.965	0.171	0.284
	♂	269.79	270.21	262.71	273.33	272.92	258.54	263.13	258.13	8.155	0.747	0.318	0.616
30~60 days	♀	269.41^{ab}	293.95^{a}	256.17^{b}	270.60^{ab}	257.11^{b}	264.46^{b}	273.85^{ab}	264.73^{b}	8.008	0.203	0.193	0.043
	♂	266.39	262.40	269.96	288.84	292.62	285.40	278.27	292.01	10.555	0.371	0.049	0.644
F:G													
1~30 days	♀	7.91	12.26	10.60	9.15	11.10	9.52	8.80	8.22	1.220	0.353	0.512	0.094
	♂	8.05	11.05	11.16	9.27	7.66	7.06	7.33	6.61	1.000	0.376	0.0004	0.262
30~60 days	♀	4.66^{bc}	4.12^{c}	3.75^{c}	3.55^{c}	3.69^{c}	7.13^{b}	10.26^{a}	9.72^{a}	0.874	0.011	< 0.0001	0.0003
	♂	5.09^{b}	3.71^{b}	3.66^{b}	4.05^{b}	3.87^{b}	8.01^{a}	8.17^{a}	9.37^{a}	0.482	0.001	< 0.0001	< 0.0001

BW, body weight; *ADG*, average daily gain; *ADFI*, average daily feed intake; *F:G*, feed conversion ratio; *Se*, selenium; *VE*, vitamin E; *SEM*, standard mean error

VE × Se, the interaction between vitamin E and selenium supplementation

Means with different letters within the same line and the same item differ significantly ($P < 0.05$), data are expressed as least squares means with pooled SEM; $n = 10$ per treatment

supplemented with 400 mg VE/kg was significantly higher than that in other groups ($P < 0.05$), and groups supplemented with 200 mg VE/kg excreted more Se than groups fed diets not supplemented with VE ($P < 0.05$). In male and female foxes, Se deposition was significantly affected by the VE levels in diets ($P < 0.05$). In addition, a significant effect of dietary Se levels on Se deposition was found from male blue foxes ($P < 0.05$).

Serum Antioxidant Parameters

There were significant effects of different dietary levels of VE and Se on serum antioxidant parameters of blue foxes (Table 4). T-AOC was significantly influenced by the levels of Se in diets as well as the interaction of VE and Se addition ($P < 0.05$) in male but not in female. MDA concentrations were higher in female foxes receiving VE supplemented diets, whereas dietary Se influenced the MDA concentrations in male blue foxes ($P < 0.05$). The activities of SOD were increased in female serum of the group with 200 mg VE/kg and 0 mg Se/kg supplementation ($P < 0.05$). Regardless of gender, the SOD activities were significantly influenced in serum from foxes fed diets enriched with VE and Se ($P < 0.05$). The activities of GSH-PX were much lower in the groups without Se added in both female and male foxes ($P < 0.05$). In female foxes, the GSH-ST activities were significantly higher in the serum of groups with 100 or 200 mg VE/kg and 0 mg Se/kg in diet, but the GSH-ST activities were much lower in the serum of groups with 200 or 400 mg VE/kg and 0.2 mg Se/kg in diet ($P < 0.05$). The activities of GSH-ST were decreased with Se levels in male diets ($P < 0.05$), and the GSH-ST activities were affected by an interaction between VE and Se ($P < 0.05$). The activities of GR in female serum were not significantly affected by the levels of VE and Se ($P > 0.05$). However, the VE levels and the interaction between VE and Se supplementation significantly affected the activities of GR in males ($P < 0.05$). The TrxR-activity was

Table 3 Effects of different vitamin E and selenium supplementation on nutrients apparent digestibility as well as on the metabolism of nitrogen and selenium of growing furring blue foxes

		VE and Se addition (mg/kg)								SEM	*P* value		
		0 and 0	100 and 0	200 and 0	400 and 0	0 and 0.2	100 and 0.2	200 and 0.2	400 and 0.2		VE	Se	VE × Se
DM (%)	♀	76.35	75.93	75.20	75.13	75.19	74.41	73.71	76.40	1.816	0.871	0.576	0.845
	♂	71.83	72.70	70.37	73.26	74.43	75.14	73.80	74.41	1.233	0.424	0.008	0.832
CP (%)	♀	74.92	72.27	73.71	76.34	77.21	76.80	75.90	74.73	2.364	0.913	0.274	0.630
	♂	70.14	68.91	67.30	69.51	70.34	71.82	74.23	71.43	1.935	0.994	0.034	0.363
CF (%)	♀	90.29	90.82	89.61	91.64	92.00	90.51	90.07	92.06	1.008	0.253	0.427	0.791
	♂	89.32^{bcd}	90.10^{abcd}	88.12^{d}	88.86^{cd}	91.36^{abc}	90.88^{abc}	92.34^{a}	91.46^{ab}	0.800	0.978	< 0.0001	0.203
N deposition (g/d)	♀	5.18	5.49	4.83	6.00	5.39	5.50	5.28	4.40	0.497	0.844	0.507	0.168
	♂	5.26	3.33	4.00	5.46	4.71	4.55	4.74	3.86	0.610	0.378	0.902	0.101
NPU (%)	♀	43.73	42.50	40.69	50.33	46.48	47.80	44.13	37.53	4.203	0.904	0.913	0.128
	♂	44.49	29.28	32.44	42.56	35.55	36.63	39.17	30.85	4.718	0.522	0.624	0.090
BV of protein (%)	♀	57.90	58.88	54.58	65.13	59.77	62.58	58.07	50.35	4.858	0.831	0.679	0.181
	♂	64.09	41.98	47.84	61.49	50.53	50.86	51.87	43.78	6.406	0.386	0.316	0.117
Se fecal excretion (μg/day)	♀	6.50^{cd}	7.52^{cd}	4.84^{d}	9.06^{c}	18.77^{b}	23.42^{a}	23.05^{a}	25.12^{a}	1.163	0.005	< 0.0001	0.109
	♂	7.73^{c}	8.06^{c}	8.16^{c}	8.33^{c}	25.92^{b}	27.36^{b}	25.94^{ab}	29.85^{a}	1.045	0.158	< 0.0001	0.301
Se urinary excretion (μg/day)	♀	2.06^{bc}	2.21^{bc}	2.12^{bc}	1.93^{c}	2.66^{bc}	3.37^{ab}	3.13^{abc}	4.19^{a}	0.412	0.408	0.0002	0.243
	♂	1.01	2.05	1.39	1.64	1.69	2.92	2.79	2.61	0.538	0.223	0.017	0.923
Total Se excretion (μg/day)	♀	8.56^{d}	9.73^{cd}	6.96^{cd}	10.98^{c}	21.42^{b}	26.80^{a}	26.18^{a}	29.31^{a}	1.254	0.003	< 0.0001	0.083
	♂	8.74^{c}	10.11^{c}	9.55^{c}	9.97^{c}	27.61^{b}	30.28^{ab}	28.72^{b}	32.46^{a}	0.876	0.012	< 0.0001	0.182
Se deposition (μg/day)	♀	-0.16^{ab}	-0.29^{ab}	1.03^{ab}	-2.01^{b}	3.06^{a}	-2.64^{b}	-2.06^{b}	-3.19^{b}	1.291	0.030	0.360	0.094
	♂	-0.58^{ab}	-1.64^{ab}	-1.12^{ab}	-1.06^{ab}	0.34^{a}	-4.34^{bc}	-3.11^{ab}	-6.87^{c}	1.163	0.020	0.008	0.060
Se digestibility (%)	♀	−2.69	−4.09	11.97	−21.64	11.94	−11.01	−8.99	−12.18	9.189	0.111	0.885	0.227
	♂	−7.18	−19.13	−13.44	−13.34	0.96	−16.91	−13.56	−27.86	7.682	0.139	0.855	0.515

DM, dry matter; *CP*, crude protein; *CF*, crude fat; *N*, nitrogen; *NPU*, net protein utilization; *BV*, biological value of protein; *Se*, selenium; *VE*, vitamin E; *SEM*, standard mean error

VE × Se, the interaction between vitamin E and selenium supplementation

Means with different letters within the same line and the same item differ significantly ($P < 0.05$), data are expressed as least squares means with pooled SEM; $n = 7$ per treatment

Table 4 Effects of different vitamin E and selenium supplementation on serum antioxidant parameters of growing furring blue foxes

		VE and Se addition (mg/kg)								SEM	*P* value		
		0 and 0	100 and 0	200 and 0	400 and 0	0 and 0.2	100 and 0.2	200 and 0.2	400 and 0.2		VE	Se	VE × Se
T-AOC U/mL	♀	5.28	5.92	5.64	5.80	5.68	6.20	5.11	5.27	0.347	0.215	0.704	0.381
	♂	3.05^{d}	3.68^{cd}	4.50^{bc}	4.94^{b}	6.52^{a}	5.16^{b}	4.82^{b}	4.81^{b}	0.303	0.480	< 0.0001	< 0.0001
MDA mmol/mL	♀	1.05	1.36	1.37	1.52	1.09	1.44	1.47	1.41	0.340	0.019	0.783	0.847
	♂	1.07	1.14	1.12	1.35	1.53	1.35	1.36	1.31	0.132	0.898	0.024	0.314
SOD U/mL	♀	78.67^{b}	85.17^{b}	122.38^{a}	81.88^{b}	84.01^{b}	83.45^{b}	77.28^{b}	77.86^{b}	2.693	< 0.0001	< 0.0001	< 0.0001
	♂	68.96^{e}	70.95^{de}	119.59^{a}	64.96^{e}	86.57^{b}	81.34^{cb}	78.99^{cb}	77.08^{cd}	2.624	< 0.0001	0.950	< 0.0001
GSH-PX U/mL	♀	421.39^{c}	425.15^{c}	483.67^{bc}	477.33^{bc}	611.74^{a}	552.17^{ab}	550.71^{ab}	550.30^{ab}	25.100	0.624	< 0.0001	0.068
	♂	433.11^{c}	468.05^{bc}	533.34^{ab}	519.40^{ab}	555.83^{ab}	550.70^{ab}	592.33^{a}	537.62^{ab}	28.096	0.106	0.001	0.321
GSH-ST U/mL	♀	48.79^{b}	65.31^{a}	61.07^{a}	45.92^{b}	42.21^{bc}	34.63^{c}	24.47^{d}	23.17^{d}	2.789	< 0.0001	< 0.0001	< 0.0001
	♂	30.76^{c}	42.70^{ab}	49.28^{a}	40.40^{b}	40.27^{b}	22.58^{d}	19.28^{d}	21.99^{d}	2.295	0.274	< 0.0001	< 0.0001
GR U/mL	♀	15.34	15.53	15.82	15.47	16.28	20.15	15.47	16.13	1.909	0.618	0.285	0.589
	♂	13.47^{bc}	14.64^{bc}	12.67^{bc}	23.12^{a}	17.65^{b}	17.20^{b}	11.32^{c}	10.84^{c}	1.610	0.024	0.140	< 0.0001
TrxR U/mL	♀	28.14	29.48	30.32	33.18	34.47	34.49	34.59	35.68	2.793	0.712	0.029	0.920
	♂	31.41	33.75	33.78	34.86	44.26	34.68	34.59	34.53	3.159	0.611	0.121	0.144
TPX U/mL	♀	0.023^{c}	0.042^{b}	0.052^{a}	0.053^{a}	0.055^{a}	0.053^{a}	0.052^{a}	0.030^{c}	0.003	< 0.0001	0.012	< 0.0001
	♂	0.026^{d}	0.041^{bc}	0.042^{abc}	0.051^{a}	0.048^{ab}	0.048^{ab}	0.037^{c}	0.027^{d}	0.003	0.083	0.908	< 0.0001
T-GSH μmol/L	♀	2.54^{c}	4.15^{a}	3.63^{ab}	3.14^{bc}	3.01^{bc}	2.90^{bc}	2.86^{bc}	2.71^{c}	0.256	0.030	0.010	0.016
	♂	2.99^{bc}	3.63^{ab}	3.89^{a}	2.71^{c}	2.49^{c}	3.04^{abc}	3.09^{abc}	2.71^{c}	0.282	0.015	0.024	0.541

T-AOC, total antioxidant capacity; *MDA*, maleic dialdehyde; *SOD*, superoxide dismutase; *GSH-PX*, glutathione peroxidase; *GSH-ST*, glutathione S-transferase; *GR*, glutathione reductase; *TrxR*, thioredoxin reductase; *TPX*, thioredoxin peroxidase; *T-GSH*, total glutathione; *Se*, selenium; *VE*, vitamin E; *SEM*, standard mean error

VE × Se, the interaction between vitamin E and selenium supplementation

Means with different letters within the same line and the same item differ significantly ($P < 0.05$), data are expressed as least squares means with pooled SEM; $n = 6$ per treatment

decreased in female blue foxes receiving Se supplemented diets ($P < 0.05$), but not influenced by diets in males ($P > 0.05$). The TPX activity was lower in serum from female foxes in the group without VE and Se addition as well as in the group with 400 mg VE/kg and 0.2 mg Se/kg addition ($P < 0.05$). The highest TPX activities in male blue fox serum were found in the group with 400 mg VE/kg and 0 mg Se/kg supplementation ($P < 0.05$). Interestingly, the concentrations of T-GSH were highest in female serum in the group with 100 mg VE/kg and 0 mg Se/kg addition but lowest in the group without VE and Se addition ($P < 0.05$). The T-GSH concentrations in male blue fox serum were influenced by the concentrations of VE and Se in diets ($P < 0.05$), but no interaction of VE and Se supplementation was observed ($P > 0.05$).

Metabolism Parameters

The effects of different VE and Se levels in diets on blood metabolism parameters of blue foxes are shown in Table 5. The concentrations of TP in female fox serum were markedly influenced by the concentrations of Se in diets ($P < 0.05$), and they were significantly impacted by the interaction of VE and Se in male fox serum ($P < 0.05$). The effects of supplementation with VE and Se in female diets on the ALB concentrations were not significant ($P > 0.05$), but the interaction of VE and Se influenced the concentrations of ALB in male fox serum ($P < 0.05$). The GLB concentration in female blue foxes serum was lower in female blue foxed fed diets supplemented with Se ($P < 0.05$), but not influenced by diet in males ($P > 0.05$). The concentrations of GLU and TG in serum were significantly influenced by VE and Se supplementation, and the interaction of VE and Se was observed on the concentrations of GLU and TG in serum ($P < 0.05$). The GLU concentrations were significantly higher in the groups with 0.2 mg Se/kg and 0 mg VE/kg compared to the other groups ($P < 0.05$). However, the blue foxes fed diets enriched with 200 or 400 mg VE/kg and 0 mg Se/kg diet had higher serum TG levels compared with the other feeding groups ($P < 0.05$). The levels of serum CHO were not changed with supplementation of VE and Se in female foxes ($P > 0.05$), but serum CHO levels were significantly influenced by the amount of VE addition in male diets ($P < 0.05$). Moreover, the HDL concentrations were greatly impacted by the VE levels in diets ($P < 0.05$), and the lowest serum HDL concentrations were found in the group without VE and Se added in diets. However, there were no significant effects of different dietary VE and Se supplementation levels on serum LDL concentrations ($P > 0.05$). The BUN concentrations in serum from blue foxes were significantly affected by the interaction between dietary VE and Se levels ($P < 0.05$). There were no significant effects of VE and Se addition to diets on the concentrations of Se in female serum ($P > 0.05$), but dietary VE significantly influenced the concentrations of Se in male serum ($P < 0.05$). Furthermore, supplementation with VE affected the serum VE concentrations, but the effect was more pronounced in foxes fed diets supplemented with VE and Se ($P < 0.05$).

Discussion

The present study showed that the growth performance was better in foxes fed with suitable VE or Se addition compared with those receiving diets without VE and Se and those fed diets supplemented with high amounts of VE. Ebeid et al. [7] and Eiben et al. [30] demonstrated that the addition of VE or Se to diets results in higher BW and lower F:G in growing rabbits. Työppönen et al. [31] reported that the requirement of VE in growing mink is 60 to 80 mg/kg diets. When growing mink are fed high oxidizing sea fish, additional VE should be added to diets [14], because VE as a natural antioxidant reacts with reactive oxygen metabolites (ROM) to inactivate them, which destroys VE in the process. Foxes are carnivores, indicating that fat plays a more significant role in meeting their energy requirements. Unfortunately, higher concentrations of PUFA in fox diets may decrease their antioxidant capacity. Therefore, fox diets should be supplemented with sufficient VE levels. Chauhan et al. [32] found that dietary VE and Se supplementation increased ADFI and ADG of lambs during the 3-week finisher period. Se is widely distributed in the environment [33], and fur animals can obtain sufficient levels of Se from marine fish [11]. Due to inclusion of fishmeal, no clinical signs of Se deficiency were observed in the present study. Kiiskinen and Maekelae [34] reported that several Finnish mink diets contain 0.05–0.42 mg Se/kg in dry matter. The Se concentrations of basal diets were contained in the research of Kiiskinen and Maekelae [34], which is comparable to the Se concentrations in the basal diet used in our study.

Results in male blue foxes showed that Se concentrations in diets significantly enhanced the AD of DM, CP, and CF, but did not affect the nutrient AD in female blue foxes. Adkins and Ewan [35] and Nuijten et al. [36] found that supplementation with VE and Se in pig diets significantly increased AD of nutrients. In addition, dietary Se supplementation may partly increase the utilization of DM and CP in goats [37]. However, Tian et al. [38] observed that dietary levels of Se did not affect the nutrient AD in growing finishing pigs, which was similar to the results obtained for female foxes in the present study. This difference may be due to the different physiological digestive characteristics of male and female foxes, which should be studied in the future.

In the present study, the total excretion of Se of foxes increased with the level of Se supplementation in diets. Absorbed Se accumulates in the liver and metabolized and a large portion is then transferred into the serum [39]. Se that is not absorbed is expelled in feces as an insignificant source of environmental Se accumulation [40]. Dietary Se supplementation increased the urinary Se excretion in blue foxes, which

Table 5 Effects of different vitamin E and selenium supplementation on serum metabolism parameters of growing furring blue foxes

		VE and Se addition (mg/kg)								SEM	*P* value		
		0 and 0	100 and 0	200 and 0	400 and 0	0 and 0.2	100 and 0.2	200 and 0.2	400 and 0.2		VE	Se	VE × Se
TP g/L	♀	66.52^{bc}	67.80^{bc}	71.37^{ab}	73.95^{a}	66.65^{bc}	63.81^{c}	62.77^{c}	65.16^{c}	1.795	0.209	0.0003	0.061
	♂	60.89^{c}	60.70^{c}	64.23^{bc}	71.04^{a}	69.88^{ab}	63.19^{c}	63.35^{c}	64.37^{bc}	2.015	0.054	0.497	0.006
ALB g/L	♀	39.92	40.75	44.60	44.62	43.25	40.86	41.55	43.02	1.408	0.157	0.765	0.156
	♂	38.21^{c}	40.26^{bc}	45.54^{ab}	47.43^{a}	46.82^{a}	40.58^{bc}	40.55^{bc}	39.58^{c}	1.735	0.320	0.431	0.0004
GLB g/L	♀	26.60	27.06	26.77	29.34	23.40	22.94	21.22	22.14	1.742	0.799	0.0004	0.684
	♂	22.68	20.44	18.69	23.61	23.06	22.62	22.81	24.79	1.651	0.193	0.105	0.702
GLU mmol/L	♀	10.22^{e}	11.43^{cd}	11.38^{cd}	12.39^{cbc}	15.16^{a}	15.95^{a}	12.62^{b}	11.18^{de}	0.463	< 0.0001	< 0.0001	< 0.0001
	♂	10.50^{d}	10.81^{d}	12.88^{bc}	12.77^{bc}	14.04^{a}	13.67^{ab}	12.25^{c}	10.72^{d}	0.326	0.036	0.0003	< 0.0001
TG mmol/L	♀	1.19^{dc}	1.29^{c}	2.25^{a}	2.12^{a}	1.68^{b}	1.06^{de}	0.98^{e}	0.97^{e}	0.053	< 0.0001	< 0.0001	< 0.0001
	♂	0.91^{cd}	0.94^{c}	1.75^{a}	1.79^{a}	1.49^{b}	0.89^{cd}	0.82^{d}	0.84^{d}	0.032	< 0.0001	< 0.0001	< 0.0001
CHO mmol/L	♀	3.92	4.12	4.28	4.24	3.74	4.14	4.27	4.05	0.262	0.404	0.634	0.964
	♂	3.60	3.78	4.38	4.47	4.17	3.59	4.25	4.46	0.232	0.008	0.732	0.355
HDL mmol/L	♀	2.38	2.66	2.72	2.73	2.47	2.58	2.70	2.73	0.077	0.002	0.946	0.746
	♂	2.29^{c}	2.40^{bc}	2.74^{ab}	2.95^{a}	2.62^{abc}	2.54^{abc}	2.68^{abc}	2.61^{abc}	0.125	0.032	0.851	0.078
LDL mmol/L	♀	0.17	0.23	0.15	0.17	0.14	0.19	0.19	0.22	0.021	0.059	0.798	0.091
	♂	0.17	0.17	0.19	0.20	0.19	0.15	0.19	0.22	0.023	0.260	0.731	0.682
BUN mmol/L	♀	7.03^{a}	7.05^{a}	6.53^{ab}	5.99^{b}	6.22^{ab}	6.90^{a}	6.98^{a}	6.82^{a}	0.263	0.199	0.676	0.021
	♂	7.18^{d}	8.16^{c}	8.25^{c}	9.52^{a}	9.26^{ab}	8.58^{bc}	8.37^{bc}	8.21^{c}	0.308	0.175	0.145	< 0.0001
CRE μmol/L	♀	118.34	112.91	111.74	117.66	108.19	119.29	112.62	125.56	4.029	0.115	0.664	0.132
	♂	111.55	120.65	122.13	123.21	119.26	116.09	119.89	121.94	2.762	0.076	0.964	0.157
Se (μg/L)	♀	0.30	0.29	0.27	0.27	0.27	0.28	0.30	0.30	0.025	0.998	0.597	0.627
	♂	0.26	0.28	0.25	0.36	0.25	0.26	0.30	0.35	0.031	0.026	0.909	0.698
VE (mg/L)	♀	0.54^{d}	5.35^{c}	5.48^{c}	6.84^{bc}	3.51^{cd}	5.93^{c}	9.99^{ab}	11.05^{a}	1.069	0.001	< 0.0001	0.280
	♂	0.64^{d}	4.33^{c}	6.10^{bc}	7.59^{bc}	5.81^{bc}	6.34^{bc}	8.97^{b}	13.41^{a}	1.219	0.0003	0.0001	0.373

TP, total protein; *ALB*, albumin; *GLB*, globulin; *GLU*, glucose; *TG*, triglycerides; *CHO*, cholesterol; *HDL*, highly density lipoprotein cholesterol; *LDL*, low density lipoprotein cholesterol; *BUN*, blood urea nitrogen; *CRE*, creatinine; *Se*, selenium; *VE*, vitamin E; *SEM*, standard mean error

VE × Se, the interaction between vitamin E and selenium supplementation

Means with different letters within the same line and the same item differ significantly ($P < 0.05$), data are expressed as least squares means with pooled SEM; $n = 6$ per treatment

is in line with other studies [41–43]. A relative increase in Se urinary excretion occurs with increasing dietary Se to decrease Se retention and maintain Se homeostasis [43]. Se supplementation in blue fox diets increased the urinary excretion of Se. In 1957, Schwartz and Foltz established Se as an essential trace element in nutrition for prevention of disease [44]. In addition, dietary levels of VE greatly influenced total Se excretion and deposition.

The present study demonstrated that supplementation of blue fox diets with VE and Se may affect the antioxidant capacity of the body and alter the concentration of antioxidant enzymes in serum. It is well known that supplementing VE and Se in feed causes multiple alterations in the antioxidant system [45, 46]. VE is a chain-breaking antioxidant that prevents the propagation of free radical reactions [45]. The nutritive effect of Se in animals is manifested in the composition of selenocysteine that is used as building block in selenoproteins [47]. Increasing dietary levels of antioxidants may decrease in vivo measure of oxidative damage [46].

In the present study, dietary VE and Se levels affected T-AOC and the level of MDA in the serum of foxes. Lan et al. [48] found that VE affects the activity of T-AOC and the concentrations of MDA in rat serum. Se supplementation to blue fox diets enhanced the serum GSH-PX activity in the present study which was similar to results obtained in poultry diets [49]. The GSH-Px enzyme contains selenocysteine residues as its active site [50]. Ebeid et al. [7] reported that VE and organic Se supplementation in rabbit diets increased serum GSH-PX concentrations. In the present study, the activity of SOD was influenced by diet. Interestingly, SOD activities were highest in groups fed with 200 mg VE/kg and approximately 0.1 mg Se/kg diet. The SOD enzyme also participates in the antioxidant defense system [51]. Akiyama et al. [52] suggested that SOD activation is a characteristic feature of ROM production. Interestingly, the concentrations of TG in serum were also higher in the abovementioned group. Kurahashi found that the concentrations of TG in wild-type mice serum were higher than in SOD1-knockout mice serum [53], which indicated that SOD could stimulate lipid secretion into the blood. Ziaie et al. [54] indicated that VE is a potent ROM scavenger that decreases the total SOD activity. In the present study, the activity of GSH-ST was significantly changed with the levels of VE and Se in diets. GSH-ST is part of a superfamily that is composed of antioxidant enzymes [55]. In female fox serum samples, the activity of TrxR was markedly affected by the Se levels in diets. TrxR is an enzyme that contains selenocysteine residues, which are essential for its catalytic activity [56]. Hence, the results indicated that the dietary Se levels may influence the synthesis of selenocysteine residues and further affect the activities of related enzymes. The current study found that the activity of TrxR in serum was not influenced by dietary levels of VE and Se in male blue fox diets. Dietary Se levels did not influence the expression of TrxR 3 in livers and kidneys tissue of mice [57] or of TrxR 1 in pig either [58]. The activity of TPX was significantly influenced by the dietary VE and Se contents in the present study. TPX is a major cellular protein disulfide reductase, which serves as electron donor for reducing enzymes [59]. Suitable concentrations of VE and Se in fox diets can improve the activities of TPX. The serum of T-GSH was also affected by the dietary VE and Se concentrations in this study. It is well known that VE together with glutathione and a membrane-bound enzyme can prevent against damaging effects of reactive oxygen species on PUFA in biomembranes (lipid peroxidation) [60]. Exposure to reactive oxygen and nitrogen species can increase the GSH concentrations by increasing the rate of GSH synthesis [60]. Hence, the serum of T-GSH was lower in the groups with the highest VE supplementation in diets.

In the present study, interactions between VE and Se affected serum TP and ALB in male blue foxes, but not in female. There were significant effects of dietary Se supplementation on the concentrations of GLB in females, but there was no effect on the GLB in male serum. Shinde et al. [61] revealed no significant difference in the concentrations of TP, ALB, and GLB in the serum of male buffalo calves fed different VE or Se supplemented diets. Some studies in animals indicated that high doses of Se might have gender-dependent effects on the immune system [33]. Marsh et al. [62] showed that high levels of Se impaired the antibody responses of male chickens but not of female chickens, and similar results have been found for male and female rats [63]. Adding VE to blue fox diets decreased the concentrations of serum GLU in the present study. The research in type-2 diabetes mellitus patients found that a significant decrease in fasting blood GLU was seen in groups supplemented with VE [64]. In the present study, the concentrations of TG in serum of foxes with Se supplementation were lower than in those fed diets without Se addition. Crespo et al. [65] indicated that Se is involved with TG in the metabolism. In the present study, there was no significant effect of VE and Se supplementation in blue fox diets on serum LDL, which agreed with the findings of Corino et al. [66] and Ebeid et al. [7] in growing rabbits. The concentrations of HDL were influenced by the concentrations of VE in diets, but they were not changed by the dietary Se addition. Shinde et al. [61] found that HDL concentrations in serum of calves were affected supplementation with VE and Se. In some studies, Se supplementation has been associated with modest reductions of CHO and increased HDL levels in serum of children [28, 67]. González-Estecha et al. [68] found significant correlations between serum Se and TG, CHO, and LDL in humans. Dietary treatments with VE significantly affected the CHO concentrations in male serum, which contradicted the findings of Shinde et al. [61] in male buffalo calves fed different dietary concentrations of VE and Se. Moreover, the effects of different levels of VE and

selenomethionine supplementation on serum biochemistry in laying hens have suggested that a combination of VE and Se added to the diet decreases serum CHO, TG, and LDL concentrations but increases HDL concentrations [69]. In the present study, interaction effects of different VE and Se supplementation levels were found on BUN. Sugden et al. [70] found that Se supplementation in ewe diets significantly increased the levels of BUN. Similar effects of VE and Se supplementation on blood chemistry have been reported in male calves [62]. The different results may be due to differences in species of animals, and further studies are needed. Regardless of gender, serum VE concentrations were significantly affected by dietary supplementation of VE and Se. Jewell et al. [46] found that increasing levels of dietary VE in dog feed caused significant increases in serum VE levels. These results further confirmed a synergistic effect between VE and Se [33].

Conclusion

The present study demonstrated that appropriate dietary levels of VE and Se in growing furring foxes were beneficial for feed conversion ratio, the nutrient digestibility, and the antioxidant abilities. In addition, some metabolic parameters were influenced by diet. These findings suggested that the suitable dietary concentration of VE is 200 mg/kg in combination with approximately 0.1 mg Se/kg from raw material.

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References

1. Van Metre DC, Callan RJ (2001) Selenium and vitamin E. Vet Clin N Am-Food A 17(2):373–402. https://doi.org/10.1016/S0749-0720(15)30034-7
2. Horwitt MK (1986) Interpretations of requirements for thiamin, riboflavin, niacin-tryptophan, and vitamin E plus comments on balance studies and vitamin B-6. Am J Clin Nutr 44(6):973–985. https://doi.org/10.1093/ajcn/44.6.973
3. Gossum AV, Kurian R, Whitwell J, Jeejeebhoy KN (1988) Decrease in lipid peroxidation measured by breath pentane output in normals after oral supplementation with vitamin E. Clin Nutr 7(1):53–57. https://doi.org/10.1016/0261-5614(88)90012-X
4. Surai PF, Fisinin VI (2016) Selenium in sow nutrition. Anim Feed Sci Technol 211:18–30. https://doi.org/10.1016/j.anifeedsci.2015.11.006
5. Surai PF (2002) Natural antioxidants in avian nutrition and reproduction. Dissertation, Nottingham University
6. Surai PF (2006) Selenium in nutrition and health. Dissertation, Nottingham University
7. Ebeid TA, Zeweil HS, Basyony MM, Dosoky WM, Badry H (2013) Fortification of rabbit diets with vitamin E or selenium affects growth performance, lipid peroxidation, oxidative status and immune response in growing rabbits. Livest Sci 155(2–3):323–331. https://doi.org/10.1016/j.livsci.2013.05.011
8. Avanzo JL, de Mendonça CX Jr, Pugine SM, de Cerqueira Cesar M (2001) Effect of vitamin E and selenium on resistance to oxidative stress in chicken superficial pectoralis muscle. Comp Biochem Physiol C 129 (2):163–173 https://doi.org/10.1016/S1532-0456(01)00197-1
9. Teixeira AGV, Lima FS, Bicalho MLS, Kussler A, Lima SF, Felippe MJ, Bicalho RC (2013) Effect of an injectable trace mineral supplement containing selenium, copper, zinc, and manganese on immunity, health, and growth of dairy calves. J Dairy Sci 97:4216–4226. https://doi.org/10.1016/j.tvjl.2013.02.022
10. Falk M, Bernhoft A, Framstad T, Salbu B, Wisløff H, Kortner TM, Kristoffersen AB, Oropeza-Moe M (2018) Effects of dietary sodium selenite and organic selenium sources on immune and inflammatory responses and selenium deposition in growing pigs. J Trace Elem Med Biol 50:527–536. https://doi.org/10.1016/j.jtemb.2018.03.003
11. NRC (1982) Nutrient requirements of mink and foxes. 2nd rev. National Academy of Sciences, Washington, DC
12. Ender F, Helgebostad A (1975) Unsaturated dietary fat and lipoperoxides as etiological factors in vitamin E deficiency in mink. Acta Vet Scand Suppl 16(55):1–25
13. Engberg RM, Jakobsen K, Børsting CF, Gjern H (1993) On the utilization, retention and status of vitamin E in mink (*Mustela vison*) under dietary oxidative stress. J Anim Physiol Anim Nutr 69(1–5):66–78. https://doi.org/10.1111/j.1439-0396.1993.tb00791.x
14. Ender F, Helgebostad A (1953) Yellow fat disease in fur bearing animals. Proc XV Int Vet Cong Stockholm 1 Pt 1:636
15. Stowe HD, Whitehair CK (1963) Gross and microscopic pathology of tocopherol-deficient mink. J Nutr 81(81):287–300. https://doi.org/10.1093/jn/81.4.287
16. Van Vleet JF (1975) Experimentally induced vitamin E-selenium deficiency in the growing dog. J Am Vet Med Assoc 166(8):769–774
17. Müller AS, Pallauf J, Most E (2002) Parameters of dietary selenium and vitamin E deficiency in growing rabbits. J Trace Elem Med Biol 16(1):47–55. https://doi.org/10.1016/S0946-672X(02)80008-6
18. Rd Miller E, Pastorbarriuso R, Dalal D, Riemersma RA, Appel LJ, Guallar E (2005) Meta-analysis: high-dosage vitamin E supplementation may increase all-cause mortality. Ann Intern Med 142(2):37–47. https://doi.org/10.7326/0003-4819-142-1-200501040-00110
19. Aburto A, Britton W (1998) Effects of different levels of vitamins A and E on the utilization of cholecalciferol by broiler chickens. Poult Sci 77(4):570–577. https://doi.org/10.1093/ps/77.4.570
20. Kim YY, Mahan DC (2001) Comparative effects of high dietary levels of organic and inorganic selenium on selenium toxicity of growing-finishing pigs. J Anim Sci 79(4):942–948. https://doi.org/10.2527/2001.794942x
21. Nolan B, Hess M, Howell A, Martin JV, Wagner GC, Fisher H (2005) High vitamin E and selenium elevate, whereas diphenyl-para-phenylenediamine plus caffeine lowers liver fat in alcohol-fed rats. Nutr Res 25(7):701–709. https://doi.org/10.1016/j.nutres.2005.07.004
22. Finch JM, Turner RJ (1989) Enhancement of ovine lymphocyte responses: a comparison of selenium and vitamin E supplementation. Vet Immunol Immunopathol 23(3–4):245–256. https://doi.org/10.1016/0165-2427(89)90138-4
23. Stabel JR, Nonnecke BJ, Reinhardt TA (1990) Effect of in vitro selenium repletion on bovine lymphocyte proliferation. Nutr Res 10(9):1053–1059. https://doi.org/10.1016/S0271-5317(05)80047-8
24. Yasunaga T, Kato H, Ohgaki K, Inamoto T, Hikasa Y (1982) Effect of vitamin E as an immunopotentiation agent for mice at optimal

dosage and its toxicity at high dosage. J Nutr 112(6):1075–1084. https://doi.org/10.1093/jn/112.6.1075
25. Canadian Council on Animal Care (1993) Guide to the care and use of expermental animals, vol 2nd. CCAC, Ottawa
26. AOAC (2003) Official methods of analysis. Association of Official Analytical Chemists, Inc, Arlington, p 1298pp
27. AOAC (2000) Official methods of analysis: selenium in feeds and premixes. Association of Official Analytical Chemists, Inc, Arlington
28. Błażewicz A, Klatka M, Astel A, Koronaglowniak I, Dolliver W, Szwerc W, Kocjan R (2015) Serum and urinary selenium levels in obese children: a cross-sectional study. J Trace Elem Med Biol 29: 116–122. https://doi.org/10.1016/j.jtemb.2014.07.016
29. Alvarez JC, De Mazancourt P (2001) Rapid and sensitive high-performance liquid chromatographic method for simultaneous determination of retinol, α-tocopherol, 25-hydroxyvitamin D-3 and 25-hydroxyvitamin D-2 in human plasma with photodiode-array ultraviolet detection. J Chromatogr B Biomed Sci Appl 755(1): 129–135. https://doi.org/10.1016/S0378-4347(01)00047-0
30. Eiben C, Végi B, Virág G, Gódor-Surmann K, Kustos K, Maró A, Odermatt M, Zsédely E, Tóth T, Schmidt J (2011) Effect of level and source of vitamin E addition of a diet enriched with sunflower and linseed oils on growth and slaughter traits of rabbits. Livest Sci 139(3):196–205. https://doi.org/10.1016/j.livsci.2011.01.010
31. Työppönen J, Hakkarainen J, Juokslahti T, Lindberg P (1984) Vitamin E requirement of mink with special reference to tocopherol composition in plasma, liver, and adipose tissue. Am J Vet Res 45(9):1790–1794
32. Chauhan SS, Ponnampalam EN, Celi P, Hopkins DL, Leury BJ, Dunshea FR (2016) High dietary vitamin E and selenium improves feed intake and weight gain of finisher lambs and maintains redox homeostasis under hot conditions. Small Rumin Res 137:17–23. https://doi.org/10.1016/j.smallrumres.2016.02.011
33. Finch JM, Turner RJ (1996) Effects of selenium and vitamin E on the immune responses of domestic animals. Res Vet Sci 60(2):97–106. https://doi.org/10.1016/S0034-5288(96)90001-6
34. Kiiskinen T, Maekelae J (1977) The results of mineral supplementation in the feeding of mink. Scientifur 1:41–42
35. Adkins RS, Ewan RC (1984) Effect of supplemental selenium on pancreatic function and nutrient digestibility in the pig. J Anim Sci 58(2):351–355. https://doi.org/10.2527/jas1984.582351x
36. Nuijten WGM, Morel PCH, Purchas RW (2010) Effect of lipid type, selenium and vitamin E on total tract nutrient digestibility in growing pigs. Proc N Z Soc Anim Prod 70:266–268
37. Malhi M (2015) Effects of dietary selenium supplementation on nutrient digestibility and gastrointestinal development of goats. International Conference on Agriculture Food and Animal Science
38. Tian JZ, Yun MS, Ju WS, Long HF, Kim JH, Kil DY, Chang JS, Cho SB, Kim YY, Han IK (2006) Effects of dietary selenium supplementation on growth performance, selenium retention in tissues and nutrient digestibility in growing-finishing pigs. Asian Austral J Anim 19(19):55–60. https://doi.org/10.5713/ajas.2006.55
39. Herdt TH, Rumbeiha W, Braselton WE (2000) The use of blood analyses to evaluate mineral status in livestock. Vet Clin North Am Food Anim Pract 16(3):423–444. https://doi.org/10.1016/S0749-0720(15)30078-5
40. Ullrey DE (1992) Basis for regulation of selenium supplements in animal diets. J Anim Sci 70(12):3922–3927. https://doi.org/10.2527/1992.70123922x
41. NRC (1983) Selenium in nutrition. National Academy Press, Washington, DC
42. Alhidary IA, Shini S, Al Jassim RA, Abudabos AM, Gaughan JB (2015) Effects of selenium and vitamin E on performance, physiological response, and selenium balance in heat-stressed sheep. J Anim Sci 93(2):576–588. https://doi.org/10.2527/jas2014-8419
43. Weiss WP, Eastridge ML (2005) Selenium sources for dairy cattle. tri-state dairy nutrition conference, Fort Wayne, Indiana USA
44. Schwartz K, Foltz GM (1957) Selenium as an integral part of factor 3 against diety necrotic liver degeneration. J Am Chem Soc 79: 3292–3293
45. Herrera E, Barbas C (2001) Vitamin E: action, metabolism and perspectives. J Physiol Biochem 57(1):43–56. https://doi.org/10.1007/BF03179812
46. Jewell DE, Toll PW, Wedekind KJ, Zicker SC (2000) Effect of increasing dietary antioxidants on concentrations of vitamin E and total alkenals in serum of dogs and cats. Vet Ther 1(4):264–272
47. Ferguson LR, Karunasinghe N, Zhu S, Wang AH (2012) Selenium and its' role in the maintenance of genomic stability. Mutat Res-Fund Mol M 733(1–2):100–110. https://doi.org/10.1016/j.mrfmmm.2011.12.011
48. Lan T, Yan LJ, Pei JR, Gao L, Li XZ, Liu MF, Li SC, Zhou LW, Liu ZC, Zeng XC (2012) Protective effect of selenium and vitamin E on rat myocardial oxidative injury. Chin J Catal 31(4):381–384. https://doi.org/10.3760/cma.j.issn.l000-4955.2012.04.009
49. De Vore VR, Colnago GL, Jensen LS, Greene BE (1983) Thiobarbituric acid values and glutathione perioxidase activity in meat from chickens fed a selenium-supplemented diet. J Food Sci 48(1):300–306
50. Patching SG, Gardiner R (1999) Recent developments in selenium metabolism and chemical speciation: a review. J Trace Elem Med Biol 13(4):193–214. https://doi.org/10.1016/S0946-672X(99)80037-6
51. Virgili F, Canali R, Figus E, Vignolini F, Nobili F, Mengheri E (1999) Intestinal damage induced by zinc deficiency is associated with enhanced CuZn superoxide dismutase activity in rats: effect of dexamethasone or thyroxine treatment. Free Radic Biol Med 26(9–10):1194–1201. https://doi.org/10.1016/S0891-5849(98)00307-4
52. Akiyama S, Inagaki M, Tsuji M, Gotoh H, Gotoh T, Washio K, Gotoh Y, Oguchi K (2005) Comparison of effect of vitamin E-coated dialyzer and oral vitamin E on hemodialysis-induced Cu/Zn-superoxide dismutase. Am J Nephrol 25(5):500–506. https://doi.org/10.1159/000088172
53. Kurahashi T, Konno T, Otsuki N, Kwon M, Tsunoda S, Ito J, Fujii J (2012) A malfunction in triglyceride transfer from the intracellular lipid pool to apoB in enterocytes of SOD1-deficient mice. FEBS Lett 586(24):4289–4295. https://doi.org/10.1016/j.febslet.2012.09.047
54. Ziaie S, Jamaati H, Hajimahmoodi M, Hashemian SM, Fahimi F, Farzanegan B, Moghaddam G, Radmand G, Vahdani B, Nadji SA (2011) The relationship between vitamin e plasma and BAL concentrations, SOD activity and ventilatory support measures in critically ill patients. Iran J Pharm Res 10(4):953–960
55. Hemachand T, Shaha C (2003) Functional role of sperm surface glutathione S -transferases and extracellular glutathione in the haploid spermatozoa under oxidative stress. FEBS Lett 538(1–3):14–18. https://doi.org/10.1016/S0014-5793(03)00103-0
56. Zhong L, Holmgren A (2000) Essential role of selenium in the catalytic activities of mammalian thioredoxin reductase revealed by characterization of recombinant enzymes with selenocysteine mutations. J Biol Chem 275:18121–18128. https://doi.org/10.1074/jbc.M000690200
57. Labunskyy VM, Lee BC, Handy DE, Loscalzo J, Hatfield DL, Gladyshev VN (2011) Both maximal expression of selenoproteins and selenoprotein deficiency can promote development of type 2 diabetes-like phenotype in mice. Antioxid Redox Signal 14(12): 2327–2336. https://doi.org/10.1089/ars.2010.3526
58. Pinto A, Juniper DT, Sanil M, Morgan L, Clark L, Sies H, Rayman MP, Steinbrenner H (2012) Supranutritional selenium induces alterations in molecular targets related to energy metabolism in skeletal muscle and visceral adipose tissue of pigs. J Inorg Biochem 114(9): 47–54. https://doi.org/10.1016/j.jinorgbio.2012.04.011

59. Arnér ES, Holmgren A (2010) Physiological functions of thioredoxin and thioredoxin reductase. FEBS J 267(20):6102–6109. https://doi.org/10.1046/j.1432-1327.2000.01701.x
60. Van Haaften R, Haenen GR, Evelo CT, Bast A (2003) Effect of vitamin E on glutathione-dependent enzymes. Drug Metab Rev 35(2–3):215–253. https://doi.org/10.1081/DMR-120024086
61. Shinde PL, Dass RS, Garg AK (2009) Effect of vitamin E and selenium supplementation on haematology, blood chemistry and thyroid hormones in male buffalo (Bubalus bubalis) calves. J Anim Feed Sci 18(2):241–256. https://doi.org/10.22358/jafs/66388/2009
62. Marsh JA, Dietert RR, Combs GF Jr (1981) Influence of dietary selenium and vitamin E on the humoral immune response of the chick. P Soc Exp Biol Med 166(2):228–236. https://doi.org/10.3181/00379727-166-41051
63. Siami G, Schulert AR, Neal RA (1972) A possible role for the mixed function oxidase enzyme system in the requirement for selenium in the rat. J Nutr 102(7):857–861. https://doi.org/10.1093/jn/102.7.857
64. Panda P, Behera AA, Panda SK (2016) Evaluation of effect of vitamin-E on blood glucose and serum lipids in type-2 diabetes mellitus treatment. Glob J Res Anal 5(8):9–10
65. Crespo AMV, Lança MJ, Vasconcelos S, Andrade V, Rodrigues H, Santos MC (1995) Effect of selenium supplementation on some blood biochemical parameters in male rats. Biol Trace Elem Res 47(1–3):343–347. https://doi.org/10.1007/BF02790136
66. Corino C, Lo Fiego DP, Macchioni P, Pastorelli G, Di GA, Domeneghini C, Rossi R (2007) Influence of dietary conjugated linoleic acids and vitamin E on meat quality, and adipose tissue in rabbits. Meat Sci 76(1):19–28. https://doi.org/10.1016/j.meatsci.2006.10.007
67. Rayman MP, Stranges S, Griffin BA, Pastorbarriuso R, Guallar E (2011) Effect of supplementation with high-selenium yeast on plasma lipids: a randomized trial. Ann Intern Med 154(10):656–665. https://doi.org/10.7326/0003-4819-154-10-201105170-00005
68. González-Estecha M, Palazón-Bru I, Bodas-Pinedo A, Trasobares E, Palazón-Bru A, Fuentes M, Cuadrado-Cenzual MÁ, Calvo-Manuel E (2017) Relationship between serum selenium, sociodemographic variables, other trace elements and lipid profile in an adult spanish population. J Trace Elem Med Biol 43:93–105. https://doi.org/10.1016/j.jtemb.2016.12.002
69. Kor NM, Ziaei N, Moradi K (2013) Effect of different levels of vitamin-E and selenomethionine supplementation on serum biochemistry in Lohman LS laying hens. Online Journal of Veterinary Research 17(7):383–390
70. Sugden EA, Hidiroglou M, Mitchell D (1978) Lack of an effect of dietary selenium on serum albumin, glucose and urea nitrogen in ewes. Can J Comp Med 42(3):376–378