

The effect of dietary vitamin A supplementation in maternal and its offspring on the early growth performance, liver vitamin A content, and antioxidant index of goslings

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ABSTRACT This study investigated the effect of dietary VA supplementation on maternal and its offspring in terms of the early growth performance, antioxidant index, and tissue VA content of the goslings. Yangzhou geese aged 180 D were selected and randomly distributed into 5 experimental groups with 15 female geese and 3 male geese in each group. The geese were fed a basal diet supplemented with 0, 4,000, 8,000, 12,000, or 16,000 IU/kg VA. Eggs were collected from each group starting at 300 D. After hatching, 96 goslings were selected from each maternal group and randomly distributed into 2 experimental groups with factorial arrangement (6 replicates × 8 geese), including 2 levels of VA supplementations, 0 and 9,000 IU/kg. The results are as follows: (1) Different levels of maternal VA supplementation significantly affected the BW and weight gain of 7-day-old offspring ($P < 0.05$). The

weight gain of offspring administered 9,000 IU/kg VA was significantly higher than that of offspring administered the basal diet ($P < 0.05$). (2) Maternal VA levels significantly affected the T3, T4, and insulin levels of the offspring ($P < 0.05$). (3) The GSH-PX, SOD, T-AOC, CAT, and tissue VA content of the offspring were significantly higher and MDA was significantly lower in the 9,000 IU/kg VA group than in the no VA group ($P < 0.05$). (4) Maternal VA levels had a significant effect on offspring GSH, GSH-PX, SOD, MDA, T-AOC, and CAT ($P < 0.05$). Maternal and offspring VA supplementation interact with the weight gain, tissue VA content, GSH, GSH-PX, SOD, MDA, and CAT of goslings ($P < 0.05$). Maternal supplementation with 12,000 IU/kg VA and offspring supplementation with 9,000 IU/kg VA was conducive to gosling growth.

Key words: vitamin A, maternal and offspring, growth performance, liver vitamin A content, antioxidant index

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INTRODUCTION

Vitamin A (VA) is an important fat-soluble vitamin (Clagett and Knutson, 2011). VA plays an important role in maintaining the normal visual function of animals and the structural integrity of epithelial tissue. Vitamin A improves bone growth and ensures normal immune function (Kheirouri and Alizadeh, 2014), and it maintains a normal intestinal environment and enhances antioxidant function (Pedro et al., 2018). Under the condition of VA deficiency, mucosal immune responses are reduced in animals (Sommer and Alfred, 2008; West and Mehra, 2010; Sihag et al., 2019). Wiseman et al. (2017) found that a lack of VA affects cell development, growth, and normal metabolism,

thereby reducing animal resistance. However, excessive VA in animals cause delayed bone growth (Li et al., 2019).

The US NRC (1994) recommended 4,000 IU/kg VA as the optimal supplementation level for breeding geese. However, Han (2018) believed that this amount was only the minimum requirement and failed to promote the optimal physiological state of the body (2018). While research on VA has been carried out on chickens and ducks, few studies have been performed with geese. In this experiment, 0, 4,000, 9,000, 12,000, and 16,000 IU/kg VA was added to the diet of maternal geese, 0 and 9,000 IU/kg VA was added to the diet of the offspring. This study investigated the effect of dietary VA supplementation in maternal and its offspring in terms of early growth performance, antioxidant performance, and tissue VA content of goslings. The results of this study will be beneficial to find out the appropriate amount of VA supplementation in geese diet, and provide reference for the feed preparation of the geese.

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MATERIALS AND METHODS

Experimental Design and Diets

The Yangzhou University (Yangzhou, China) Animal Care and Use Committee approved all of the procedures conducted in this study. All test geese were healthy and were obtained from the Gaoyou Yangzhou Geese Breeding Farm (China).

The experimental setup consisted of 180-day-old Yangzhou geese randomly distributed into 5 experimental groups (I, II, III, IV, and V) with 15 female geese and 3 male geese in each group. The test period lasted 120 D. The basal diet was supplemented with 5 concentrations of VA (0, 4,000, 8,000, 12,000, and 16,000 IU/kg) in groups I, II, III, IV, and V, respectively. VA (produced by Diesman Vitamin Co., Ltd, Shanghai, China) was added in the form of acetate with the concentration of the active ingredient at 5×10^5 IU/g. Vitamin A purchased from Diesman Vitamin Co., Ltd. The geese were raised in concrete pens with straw litter (2 to 3 cm thickness). Natural mating of geese, for the entire experimental period, the geese were housed under conditions with 12 h of light per day, and the room temperature was $24 \pm 3^\circ\text{C}$. Eggs were collected when the geese reached the age of 300 D (the peak time of egg laying). Subsequently the eggs were hatched. The BW of the offspring after hatching was measured. The offspring of each treatment group were randomly divided into 2 groups (6 replicates \times 8 geese). The offspring control group received only the basal diet. The VA-treated group received the basal diet supplemented with an additional 9,000 IU/kg VA (this supplementation level is based on the level of VA added to the feed for domestic chicks). A basal maize-soybean meal diet was formulated to provide adequate concentrations of all the nutrients required by geese (NRC, 1994) except for VA (Table 1). Diets are formulated on the basis of NRC and our laboratory's achievements over the years. The goslings were raised in concrete pens with straw litter (2 to 3 cm thickness). The offspring are fed from birth. Water and feed were provided ad libitum. The housing was kept clean and well ventilated. The geese were exposed to light for 23 h per day, and the temperature was maintained at approximately 29°C . The gosling test period was 7 D. No VA was added to the basic diet premix of geese and their offspring.

Sample Collection and Index Determination

BW The primary weight of the offspring after hatching was measured, and the BW and weekly weight gain were measured after 1 wk of feeding. BW was recorded by electronic platform scale (acs-30 Shanghai Yousheng Co., Ltd, Shanghai, China).

Hormone Triiodothyronine (**T3**), tetraiodothyronine (**T4**), insulin, and growth hormone (**GH**) were measured using a kit (purchased from Beijing North Biotechnology Research Institute Co., Ltd, Beijing,

Table 1. Composition and nutrient levels of the basal diets of geese and offspring (dry basis).

Item	Geese	Offspring
Ingredients (%)		
Corn	63.00	63.00
Soybean meal	23.00	30.20
Rice husk	8.70	3.20
Methionine	0.26	0.10
Salt	0.30	0.30
Stone powder	2.00	1.10
Calcium hydrogen phosphate	1.50	1.10
Choline chloride	0.24	–
Premix ¹	1.00	1.00
Total	100.00	100.00
Nutritional level ²		
ME (MJ/kg)	11.12	11.34
Crude protein (%)	15.54	18.98
Crude fiber (%)	5.86	4.07
Ca (%)	1.29	0.83
Total phosphorus (%)	0.59	0.56
Effective phosphorus (%)	0.37	0.32
Lysine (%)	0.76	0.99
Methionine (%)	0.49	0.42
Vitamin A (IU/kg)	1,210.00	1,277.00

¹Geese received the following per kilogram of premix: VD₃, 2,250,000 IU; VE, 3,600 IU; VK, 225 mg; VB₁, 220 mg; VB₂, 975 mg; VB₆, 375 mg; VB₁₂, 2.75 mg; nicotinic acid, 3,750 mg; pantothenic acid, 1,500 mg; folic acid, 140 mg; biotin, 9.5 mg; Choline chloride, 55 g; Fe, 8 g; Cu, 0.5 g; Mn, 10 g; Zn, 10 g; Se, 30 mg; and I, 125 mg.

²Offspring received the following per kilogram of premix: VD, 400,000 IU; VE, 1,800 IU; VK, 150 mg; VB₁, 60 mg; VB₂, 600 mg; VB₆, 200 mg; VB₁₂, 1 mg; niacin, 3,000 mg; D-pantothenic acid, 900 mg; folic acid, 50 mg; biotin, 4 mg; choline, 35 g; Fe, 6 g; Cu, 1 g; Mn, 9.5 g; Zn, 9 g; Se, 30 mg; and I, 50 mg.

³Vitamin A is the measured value, and the rest are calculated values.

China). The GC-911 γ radioimmunoassay was produced by Anhui Zhongke Zhongjia Scientific Instrument Co., Ltd. (Anhui, China).

Liver VA Content After 1 wk of feeding, the offspring were slaughtered and bled, and an appropriate amount of liver was collected and placed in a centrifuge tube for measurements of the VA content. The measurement method is described in GB/T 5009.82-2016 "Determination of VA and Vitamin E in Foods—Reversed Phase High Performance Liquid Chromatography".

Preparation of potassium hydroxide solution: 50 g of potassium hydroxide was weighed and dissolved in 50 mL of water, cooled, and stored in a polyethylene bottle.

Preparation of VC-ethanol solution: 2 g of ascorbic acid was weighed and dissolved in 10 mL of water, and then added with 90 mL of absolute ethanol.

Sample pretreatment method: a sample of about 5 g was accurately weighed, and saponification was carried out by adding 100 mL of a VC-ethanol solution and 25 mL of a potassium hydroxide solution. After saponification for 30 min, the saponification solution was extracted twice with 50 mL of petroleum ether, and the ether layer was combined. The ether layer was washed with 100 mL of water to neutral. The ether layer was dried over anhydrous sodium sulfate and then evaporated to dryness. The solution was made up to 10 mL of methanol, and the solution was passed through a 0.22 μm organic filter, and then measured by an

Agilent Model 1260 high performance liquid chromatograph manufactured by Agilent Technologies.

High performance liquid chromatography conditions:

Column: Agilent C₁₈ column (column length 150 mm, inner diameter 4.6 mm, particle size 5 μ m), DAD-FLD detector.

Mobile phase: gradient mobile phase

Time (min)	Methanol (%)	Water (%)
0	98	98
30.0	2	2

Flow rate: 1.0 mL/min, injection volume: 10 μ L

Column temperature: 20°C

Wavelength: VA: 325 nm

Quantification by external standard method, the standard working solution of VA was injected into high performance liquid chromatograph, and the corresponding peak area was determined. The peak area was plotted on the ordinate and the standard test solution concentration was plotted on the abscissa to calculate the standard curve equation. The sample solution was analyzed by high performance liquid chromatography, and the peak area was measured, and the concentration was calculated by the above standard curve by an external standard method.

Antioxidant Index The offspring were slaughtered and bled after 1 wk of feeding. An appropriate amount of liver sample was collected, placed in a centrifuge tube and divided into 2 portions. One portion was quickly stored in liquid nitrogen and another portion was stored in a refrigerator at 4°C, the liver sample was accurately weighed 0.5 g, 9 times volume of physiological saline by weight was added (g): volume (mL) = 1:9, ice bath conditions were homogenized by a variety of fast homogenizers. Various fast homogenizers are purchased from Shanghai Jingxin Industrial Development Co., Ltd, (Shanghai, China). By using a DL-5 M low-speed refrigerated centrifuge 2,500 r/min, the liver samples were centrifuged for 10 min, and 6 supernatants were separated into 1.5 mL enzymatic-free tubes, which were stored at -70°C for testing. The DL-5 M low-speed refrigerated centrifuge was purchased from Hunan Xiangyi Centrifuge Instrument Co., Ltd, (Hunan, China). The assay kits of Glutathione (**GSH**), glutathione peroxidase (**GSH-PX**), superoxide dismutase (**SOD**), malondialdehyde (**MDA**), total antioxidant capacity (**T-AOC**), and catalase (**CAT**) were purchased from Nanjing Jiancheng Bioengineering Research Institute (Nanjing, China). The operation method is the same as the manual. Multiskan FC type enzyme label instrument and HH-8 digital thermostat water bath pot were used in the determination process. Among them, Multiskan FC type enzyme label instrument was purchased from Semerfer Instrument Co., Ltd., (Chongqing, China). HH-8 digital thermostat water bath pot was purchased from Changzhou Guohua Electrical Appliances Co., Ltd, (Changzhou, China).

Statistical Analysis Data were analyzed in a 5 \times 2 factorial arrangement with 5 levels of geese VA and 2 levels of gosling VA using the following statistical model. Data were subjected to ANOVA using the general linear models procedure in SPSS 17.0 (SPSS, 2009), and single degree of freedom orthogonal contrasts were used to partition the treatment sums of squares into their linear effects. Deviations from linearity means were determined at $P < 0.05$.

RESULTS

BW

The effect of different VA levels in the maternal and offspring diet on the early weight and weekly weight gain of the goslings are shown in Table 2. At 7 D of age, the maternal 12,000 IU/kg VA level had increased on the offspring weight ($P < 0.05$). The maternal 12,000 IU/kg VA level had improved on the weight gain of the goslings ($P < 0.05$). The weight gain of the offspring treated with 9,000 IU/kg VA was significantly higher than that of the group without VA addition ($P < 0.05$). Different VA levels in the maternal and offspring diets had significant interaction effects on the weight gain of the goslings ($P < 0.05$). When breed geese fed diet with 12,000 IU/kg VA and its offspring fed diet with 9,000 IU/kg VA, the BW and weight gain of goslings were highest compared to the other groups.

Hormone

The effect of different VA levels in the maternal and its offspring diets on the hormone levels of the goslings are summarized in Table 3. Maternal 8,000 and 12,000 IU/kg VA levels had increased on the T3 and GH levels of the offspring ($P < 0.05$). The maternal VA 12,000 IU/kg level had increased on the T4 content of the offspring ($P < 0.05$). The maternal VA 4,000, 8,000, 12,000, and 16,000 IU/kg levels had a significant effect on the insulin content of the offspring ($P < 0.05$). The contents of VA T3, T4, and GH in offspring were significantly higher than that in the addition of 9,000 IU/kg ($P < 0.05$). There was a quadratic linear relationship between the levels of VA in maternal diets and the levels of T3, T4, and insulin in the offspring ($P < 0.05$), and a linear relationship with the content of GH ($P < 0.05$). There was no significant interaction between the maternal and offspring diets with different VA levels on the T3, T4, insulin, and GH levels of the offspring ($P > 0.05$).

Liver VA Content

The results of different VA levels in the maternal and offspring diets on the liver VA content of the goslings are presented in Table 4. The liver VA content of the offspring with 9,000 IU/kg VA added

Table 2. Effect of different VA levels in the maternal and its offspring diets on the early weight and weight gain of goslings.

Item	Maternal VA level/(IU/kg)	Offspring VA level/(IU/kg)	0 D weight/g	7 D weight/g	7 D weight gain/g
T1	0	0	92.95	212.00	119.05
T2		9,000	88.12	229.00	140.88
T3	4,000	0	91.20	250.58	159.37
T4		9,000	92.38	259.44	167.06
T5	8,000	0	90.16	244.09	153.92
T6		9,000	90.50	247.90	157.40
T7	12,000	0	91.75	255.87	164.12
T8		9,000	91.29	265.03	173.74
T9	16,000	0	88.71	249.43	160.72
T10		9,000	92.61	240.50	147.89
	SEM ¹		0.67	2.54	2.45
	0		90.53	220.50 ^c	129.97 ^c
	4,000		91.79	255.01 ^{a,b}	163.22 ^{a,b}
	8,000		90.33	245.99 ^b	155.66 ^b
	12,000		91.52	260.45 ^a	168.93 ^a
	16,000		90.66	244.96 ^b	154.31 ^b
	SEM ¹		0.67	2.54	2.45
		0		242.39	139.21 ^b
		9,000		248.37	157.34 ^a
		SEM ¹		5.07	6.57
P-value	Maternal VA level		0.950	<0.001	<0.001
	Linear		0.996	<0.001	<0.001
	Quadratic		0.873	<0.001	<0.001
	Offspring VA level			0.243	0.009
	Maternal VA level and offspring VA level interaction			0.256	0.033

¹SEM is the standard error of the mean.

The results are average values, n = 6 in T1 to T10, n = 12 when the maternal VA level is the main factor, and n = 30 when the VA level of the offspring is the main factor.

The same letter or no letter in the same column indicates that the difference is not significant ($P > 0.05$), and different lowercase letters indicate significant difference ($P < 0.05$).

Table 3. Effect of different levels of VA in the maternal and its offspring diets on early related hormones in goslings.

Item	Maternal VA level/(IU/kg)	Offspring VA level/(IU/kg)	T3 (ng/mL)	T4 (ng/mL)	Insulin (μIU/mL)	Growth hormone(ng/mL)
T1	0	0	0.98	19.21	5.67	0.64
T2		9,000	0.88	16.30	5.70	0.56
T3	4,000	0	1.29	19.82	6.69	0.71
T4		9,000	0.94	18.97	6.61	0.65
T5	8,000	0	1.40	19.92	6.69	0.73
T6		9,000	1.10	19.55	7.38	0.67
T7	12,000	0	1.46	21.05	6.82	0.73
T8		9,000	1.12	20.88	7.43	0.68
T9	16,000	0	0.99	20.86	6.70	0.71
T10		9,000	1.00	17.44	7.27	0.65
	SEM ¹		0.03	0.32	0.13	0.01
	0		0.93 ^b	17.76 ^b	5.69 ^b	0.60 ^b
	4,000		1.12 ^{a,b}	19.40 ^{a,b}	6.65 ^a	0.68 ^{a,b}
	8,000		1.25 ^a	19.73 ^{a,b}	7.04 ^a	0.70 ^a
	12,000		1.29 ^a	20.97 ^a	7.12 ^a	0.71 ^a
	16,000		0.99 ^b	19.15 ^{a,b}	6.99 ^a	0.68 ^{a,b}
	SEM ¹		0.03	0.32	0.13	0.01
		0	1.22 ^a	20.17 ^a	6.52	0.70 ^a
		9,000	1.01 ^b	18.63 ^b	6.88	0.64 ^b
		SEM ¹	0.06	0.61	0.23	0.03
P-value	Maternal VA level		0.001	0.028	0.001	0.091
	Linear		0.175	0.045	<0.001	0.047
	Quadratic		<0.001	0.020	0.012	0.051
	Offspring VA level		0.001	0.014	0.167	0.023
	Maternal VA level and offspring VA level interaction		0.120	0.227	0.746	0.998

¹SEM is the standard error of the mean.

The results are average values, n = 6 in T1 to T10, n = 12 when the maternal VA level is the main factor, and n = 30 when the VA level of the offspring is the main factor.

The same letter or no letter in the same column indicates that the difference is not significant ($P > 0.05$), and different lowercase letters indicate significant difference ($P < 0.05$).

Table 4. Effect of different VA levels in the maternal and its offspring diets on the VA content in early liver of goslings.

Item	Maternal VA level/(IU/kg)	Offspring VA level/(IU/kg)	Liver VA content (mg/kg)
T1	0	0	5.78
T2		9,000	115.68
T3	4,000	0	19.68
T4		9,000	140.78
T5	8,000	0	22.45
T6		9,000	134.77
T7	12,000	0	41.71
T8		9,000	193.97
T9	16,000	0	43.83
T10		9,000	130.77
	SEM ¹		8.24
	0		60.70
	4,000		80.23
	8,000		78.61
	12,000		117.84
	16,000		87.30
	SEM ¹		8.24
		0	26.69 ^b
		9,000	143.18 ^a
		SEM ¹	6.48
<i>P</i> -value	Maternal VA level		0.278
	Linear		0.121
	Quadratic		0.388
	Offspring VA level		<0.001
	Maternal VA level and offspring VA level interaction		<0.001

¹SEM is the standard error of the mean.

The results are average values, n = 6 in T1 to T10, n = 12 when the maternal VA level is the main factor, and n = 30 when the VA level of the offspring is the main factor.

The same letter or no letter in the same column indicates that the difference is not significant ($P > 0.05$), and different lowercase letters indicate significant difference ($P < 0.05$).

was significantly higher than that of the group with no VA added ($P < 0.05$). Different levels of VA in the diets of the offspring and the maternal had significant interaction effects on the early liver VA content of the offspring ($P < 0.05$). The VA content of the liver was highest in offspring who received 9,000 IU/kg VA who hatched from maternal geese in the group supplemented with 12,000 IU/kg VA.

Antioxidant Index

Table 5 showed the effect of different VA levels in the maternal and offspring diets on the liver antioxidant performance of the goslings at sampling day. Maternal VA supplementation at 8,000 IU/kg increased on gosling GSH and GSH-PX activity ($P < 0.05$), and the effect of 8,000 IU/kg was not significantly different from that of 12,000 IU/kg ($P > 0.05$). Maternal 4,000, 8,000, 12,000, and 16,000 IU/kg VA levels had significantly affected the offspring SOD and MDA activity ($P < 0.05$). Maternal 8,000, 12,000, and 16,000 IU/kg VA levels had a significant increase on offspring T-AOC activity ($P < 0.05$). The maternal 8,000 and 12,000 IU/kg VA levels had a significant increase on offspring CAT activity ($P < 0.05$). The GSH-PX, SOD, MDA, T-AOC, and CAT of the offspring 9,000 IU/kg VA addition group were significantly higher than those in the group without VA addition ($P < 0.05$). Different VA levels in

maternal diets had a quadratic linear relationship with GSH, GSH-PX, SOD, MDA, T-AOC, and CAT in the offspring ($P < 0.05$). Different VA levels in the maternal and its offspring diets had significant interaction effects on GSH, SOD, MDA, and CAT activity in the livers of the offspring ($P < 0.05$), and had no significant interaction effect on GSH-PX and T-AOC activity ($P > 0.05$). The goslings exhibited the best antioxidant performance when the maternal 12,000 IU/kg VA level and the offspring received 9,000 IU/kg VA.

DISCUSSION

BW

Vitamin A has an important impact on animal growth and development. Vahid et al. (2014) and Xiao et al. (2019) determined that the addition of VA to the diet can improve intestinal epithelial cells and improve the digestion and absorption of nutrients. Barbalho et al. (2018) and Tian et al. (2018) reported that VA regulates the microenvironment of the intestines and that the amount of VA supplemental can alter the amount of protein in the body. Li et al. (2008) reported that the addition of 35,512 IU/kg and 65,512 IU/kg of VA to the broiler diet resulted in the best growth performance. Jensen et al. (1983) observed that excess dietary VA supplementation slows poultry growth. The results of our experiment showed that the amount of VA in the diet of geese affects the growth of goslings. This finding may be because the addition of VA benefits the intestinal environment and accelerates the absorption of nutrients, thereby promoting the growth rate of goslings. Notably, excessive amount of VA supplementation resulted in weight loss in the goslings. This result is consistent with the findings of Jensen et al. (1983). The best growth performance in goslings was observed when the maternal diet was supplemented with 12,000 IU/kg VA, and the offspring were supplemented with 9,000 IU/kg VA.

Hormone

The physiological role of GH is to promote the metabolism and growth of substances. GH affects various organs and tissues in the body and can also enhance the immune performance (Farhat et al., 2018). Thyroid hormones include T3 and T4. Thyroid hormones exert wide ranging and long-lasting effects in animals, primarily by regulating physiological processes such as metabolism, growth and development. Normal growth and development of the body is generally coordinately regulated by the thyroid hormones and GH. Growth hormone primarily promotes tissue growth, while thyroid hormones promote organ and tissue cell differentiation. Insulin is an important hormone that regulates the metabolism of the 3 major nutrients, and GH can promote the secretion of insulin. Burnside et al. (1991) and Harvey et al. (2001) showed that GH promotes

Table 5. Effect of different vitamin A levels in maternal and its offspring diets on liver antioxidant index of goslings at day old.

Item	Maternal VA level/(IU/kg)	Offspring VA level/(IU/kg)	GSH ($\mu\text{mol/gprot}$)	GSH-PX (U/mgprot)	SOD (U/mgprot)	MDA (nmol/mgprot)	T-AOC ($\mu\text{mol/gprot}$)	CAT (U/mgprot)
T1	0	0	29.82	142.49	179.63	1.83	69.35	114.63
T2		9,000	35.05	165.73	152.15	0.41	74.43	117.91
T3	4,000	0	47.25	158.50	241.64	0.89	70.24	109.90
T4		9,000	43.84	190.10	292.20	0.33	86.23	154.40
T5	8,000	0	52.47	197.83	283.24	0.78	76.71	126.01
T6		9,000	50.29	225.67	309.22	0.33	93.47	161.37
T7	12,000	0	43.57	175.03	243.92	0.79	72.51	116.46
T8		9,000	51.49	220.76	340.63	0.29	96.84	170.95
T9	16,000	0	39.43	169.59	238.69	0.78	76.08	110.46
T10		9,000	44.35	205.20	303.05	0.40	87.11	145.30
	SEM ¹		1.06	4.10	8.03	0.06	1.53	3.27
	0		32.43 ^d	154.11 ^d	165.89 ^b	1.12 ^a	71.89 ^b	116.27 ^b
	4,000		45.54 ^{b,c}	174.30 ^{c,d}	266.92 ^a	0.61 ^b	78.24 ^{a,b}	132.15 ^{a,b}
	8,000		51.38 ^a	211.75 ^a	296.23 ^a	0.55 ^b	85.09 ^a	143.69 ^a
	12,000		47.53 ^{a,b}	197.89 ^{a,b}	292.28 ^a	0.54 ^b	84.67 ^a	143.71 ^a
	16,000		41.89 ^c	187.40 ^{b,c}	270.87 ^a	0.59 ^b	81.59 ^a	127.88 ^{a,b}
	SEM ¹		1.06	4.10	8.03	0.06	1.53	3.27
		0	42.51	170.93 ^b	237.42 ^b	1.01 ^a	71.01 ^b	115.49 ^b
		9,000	45.00	199.25 ^a	279.45 ^a	0.35 ^b	78.67 ^a	149.99 ^a
		SEM ¹	2.12	7.40	15.23	0.08	2.55	4.80
<i>P</i> -value	Maternal VA level		<0.001	<0.001	<0.001	0.004	0.032	0.034
	Linear		<0.001	<0.001	<0.001	0.004	0.014	0.117
	Quadratic		<0.001	<0.001	<0.001	0.011	0.035	0.005
	Offspring VA level		0.243	<0.001	0.008	<0.001	0.004	<0.001
	Maternal VA level and offspring VA level interaction		0.017	0.684	<0.001	<0.001	0.053	<0.001

¹SEM is the standard error of the mean.

The results are average values, $n = 6$ in T1 to T10, $n = 12$ when the maternal VA level is the main factor, and $n = 30$ when the VA level of the offspring is the main factor.

The same letter or no letter in the same column indicates that the difference is not significant ($P > 0.05$), and different lowercase letters indicate significant difference ($P < 0.05$).

the growth of chicks and chicken embryos. To date, few studies have investigated the effects of VA on poultry hormones. The results of our experiment showed that adding VA to the diet of geese can increase the levels of T3, T4, insulin, and GH in the offspring. The levels of VA in the diet were linearly related to T3, T4 and insulin levels and showed a linear relationship with GH. Perhaps because high doses of VA in the diet inhibit the activation of immune cells, there by suppressing the release of T3 and T4. An appropriate amount of GH in the serums stimulate islet B cells and causes insulin secretion. However, when GH is excessive, insulin secretion is inhibited. Therefore, the levels of T3, T4, and insulin in the offspring initially increased, but then decreased with increasing VA addition, whereas the GH level continued to rise.

Liver VA Content

Goslings can obtain a certain amount of VA from the egg after hatching to meet the maintenance needs of the early growth stage. Lima and Souza (2018) and Noble (1990) observed VA retention in chicks, and the amount of VA retained was significantly positively correlated with the VA content of the eggs approximately 40% of the VA in the eggs is transported to the newborn. Surai et al. (2000) added different levels of VA to the diets of laying hens and showed that the VA content of eggs was significantly increased with increasing dietary VA

supplementation. The present study showed that when the geese lack VA, the VA content in the liver of the offspring is significantly reduced. Moreover when VA is added to the gosling diet, the VA content in the liver is significantly increased. The offspring cannot receive VA in their diet by supplementing VA from exterior sources. They consume only the VA obtained from the egg to maintain growth. When the offspring diet is lacking in VA, the VA content of the liver decreases, and normal growth and development are adversely affected. High dose VA supplementation in the diets of maternal and offspring reduced the VA content in the liver of goslings, indicating that high levels of VA exceed the regulatory ability of the livers of the offspring, impacting VA absorption and resulting in a decrease in the VA content of the liver. The highest VA content in the gosling liver was observed when the maternal diet was supplemented with 12,000 IU/kg VA, the offspring were supplemented with 9,000 IU/kg VA.

Antioxidant Index

GSH is the most important non-enzymatic antioxidant in the body and it has many important physiological functions, such as scavenging free radicals, detoxifying, promoting iron absorption, maintaining

erythrocyte membrane integrity, maintaining DNA biosynthesis, normal cell growth and development, and promoting cellular immunity. GSH is a low molecular scavenger that removes O₂, H₂O₂, and LOOH, therefore, the amount of GSH in animals is an important factor for measuring the antioxidant capacity of the body. GSH-PX is an important enzyme that catalyzes the decomposition of hydrogen peroxide, which is widely present in the body. GSH-PX specifically catalyzes the reduction of GSH to hydrogen peroxide, which protects the structure and function of the cell membrane. SOD is an antioxidant enzyme that can effectively scavenge superoxide anion free radicals. This enzyme catalyzes the disproportionation reaction of superoxide anion radicals and alleviates the damage of the free radical chain reaction. The activity of MDA often matches the activity of SOD. The level of SOD activity indirectly reflects the body's ability to scavenge oxygen free radicals, and the level of MDA indirectly reflects the severity of free radical attack in the body. T-AOC is a comprehensive indicator of the functional status of antioxidant systems and is a result of the interaction of antioxidant enzymes in the body. T-AOC can reflect the compensatory ability of the body in response to external stimuli and indicates the state of free radical metabolism in the body. CAT is an enzyme that catalyzes the decomposition of hydrogen peroxide into oxygen and water and is located in the peroxisomes of cells. CAT is also a peroxidase marker. Mahmoud and Hijazi (2007) showed that the addition of VA to the diet reduces the degree of damage to the liver and cells of the animal body. Vollmar et al. (2002) reported that excessive VA in the rat diet damages the liver. Mccuaig and Motzok (1970) found that adding excess VA to broiler diets causes porcine liver toxicity. The results of our experiment showed that when the diets of geese lack VA, the offspring exhibit poor antioxidant properties. However, the addition of a proper amount of VA to the diets of maternal and the resulting offspring improves the antioxidant capacity in the liver of the offspring. Based on the comprehensive GSH, GSH-PX, SOD, MDA, T-AOC, and CAT analyzes the best antioxidant performance was observed when 12,000 IU/kg VA was added to the maternal diet and 9,000 IU/kg of VA was added to the offspring diet. When the amount of VA added to the diet exceeded the optimal amount, the antioxidant performance tended to decrease, which is consistent with the results of previous studies. This finding may be because adding excessive VA to the diet causes liver damage in geese, the enzymes and antioxidants in the liver that scavenge oxygen free radicals. After the livers of the geese were damaged, the levels of oxygen free radicals increased, which reduced the antioxidant capacity of the geese.

In summary, for goslings, adding 12,000 IU/kg VA to maternal diet and 9,000 IU/kg VA to offspring diet is beneficial to growth and development of goslings. Excessive VA has adverse effects on the growth of goslings.

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