

# METABOLISM AND NUTRITION

## Effects of genetic selection and threonine on meat quality in Pekin ducks

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**ABSTRACT** The present study was conducted to investigate the effects of genetic selection and threonine levels on meat quality in Pekin ducks. At 15 D of age, 192 lean ducks and 192 fatty ducks were selected and allotted to one of three treatments with 8 replicates with similar BW (8 ducks/cage), respectively. All ducks were fed the experimental diets (0.00, 0.15, and 0.30% added threonine) for 21 D from 15 to 35 D of age. The results showed that fatty ducks had higher ( $P < 0.001$ ) feed intake, feed/gain ratio, abdominal fat percentage, and sebum percentage and lower ( $P = 0.001$ ) breast muscle percentage compared with that of lean ducks. The fatty-type and lean-type ducks had similar weight gain and BW. Dietary threonine supplementation improved ( $P < 0.05$ ) growth performance and increased breast muscle percentage in lean-type ducks, but it did not affect ( $P > 0.05$ ) those indices in fatty-type ducks. Lean ducks had higher ( $P < 0.001$ ) hepatic contents of total lipids, triglyceride, cholesterol, and plasma low-density

lipoprotein cholesterol concentration, and dietary threonine supplementation decreased ( $P < 0.05$ ) hepatic total lipid, cholesterol, and triglyceride contents in lean ducks, but it had no influence on hepatic lipids in fatty ducks ( $P > 0.05$ ). Lean ducks had higher ( $P < 0.05$ ) concentrations of monounsaturated fatty acid (MUFA), and C18-polyunsaturated fatty acid (PUFA) in the liver, PUFA in the breast muscle, and C18:3n6 and C18:3n3 in plasma and lower C20-PUFA and C22-PUFA in the liver and MUFA in plasma, compared with fatty ducks. Threonine supplementation increased PUFA, N3-PUFA, and n6-PUFA in plasma and hepatic fatty acids profiles in lean ducks ( $P > 0.05$ ) but had no influence on total MUFA and total PUFA in the liver, breast muscle, and plasma in fatty ducks ( $P > 0.05$ ). In conclusion, genetic selection toward meat production and threonine supplementation increases meat production and PUFA contents, which would influence eating quality, but it is benefit for human health.

**Key words:** duck, threonine, fatty acids profile, meat quality

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## INTRODUCTION

Intramuscular fat (IMF) is an important determinant of meat quality, which positively affects meat sensory traits, such as flavor, tenderness, and juiciness (Wood et al., 2008). The IMF is affected by breeding, nutritional, hormonal, and metabolic factors. To meet the increasing need for lean meat, genetic selection has increased the carcass lean content of commercial swine

lines (Cameron and Enser, 1991), which simultaneously reduced the subcutaneous fat and IMF content (Jeremiah et al., 1999). When IMF content is reduced below 2~2.5%, the sensory traits of pig meat are negatively affected (DeVol, et al., 1988). In the last decades, continuous genetic selection forms fatty-type and lean-type Pekin duck in China, which results in tremendous differences in meat production, sebum, and abdominal fat contents (Zheng et al., 2014). However, whether the IMF content of breast muscle was different between the lean-type duck and fatty-type duck is still unknown.

In addition, some nutritional strategies have been proved to successfully increase IMF content in muscle, such as reducing dietary CP or amino acid (Doran et al., 2006; D'Souza et al., 2008; Madeira et al., 2013b; Tous et al., 2014; Pires et al., 2016; Madeira et al., 2017;

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**Table 1.** Composition and nutrient levels of basal diet for Pekin ducks from 15 to 35 D of age (as fed basis).

Ingredients	%	Nutrient levels	%
Corn	44.15	Metabolizable energy ( kcal/kg )	2,977
Peanut cake	11.80	Crude protein <sup>3</sup>	15.45
Wheat	40.07	Lysine <sup>3</sup>	0.69
Dicalcium phosphate	1.40	Methionine <sup>3</sup>	0.33
Limestone	1.10	Methionine + cysteine <sup>3</sup>	0.60
Salt	0.30	Tryptophan <sup>3</sup>	0.17
DL-Methionine	0.10	Arginine <sup>3</sup>	1.07
L-Tryptophan	0.03	Threonine <sup>3</sup>	0.41
L-Lysine HCl	0.25	Valine <sup>3</sup>	0.62
Premix <sup>1</sup>	0.50	Isoleucine <sup>3</sup>	0.51
Corn starch + threonine <sup>2</sup>	0.30	Calcium <sup>4</sup>	0.77
Total	100	Total phosphorus <sup>4</sup>	0.56
		Nonphytate phosphorus <sup>4</sup>	0.34

<sup>1</sup>Supplied per kilogram of total diet: Cu (CuSO<sub>4</sub> · 5H<sub>2</sub>O), 10 mg; Fe (FeSO<sub>4</sub> · 7H<sub>2</sub>O), 60 mg; Zn (ZnO), 60 mg; Mn (MnSO<sub>4</sub> · H<sub>2</sub>O), 80 mg; Se (NaSeO<sub>3</sub>), 0.3 mg; I (KI), 0.2 mg; choline chloride, 750 mg; vitamin A (retinyl acetate), 8,000 IU; vitamin D<sub>3</sub> (Cholcalciferol), 3,000 IU; vitamin E (DL- $\alpha$ -tocopheryl acetate), 20 IU; vitamin K<sub>3</sub> (menadione sodium bisulphate), 2 mg; thiamin (thiamin mononitrate), 1.5 mg; riboflavin, 4 mg; pyridoxine hydrochloride, 3 mg; cobalamin, 0.02 mg; calcium-D-pantothenate, 10 mg; nicotinic acid, 50 mg; folic acid, 1 mg; and biotin, 0.15 mg.

<sup>2</sup>Crystalline threonine supplements added in place of equivalent weights of cornstarch.

<sup>3</sup>These values determined by analysis based on triplicate determinations.

<sup>4</sup>These values are as formulated.

Palma-Granados et al., 2017). For example, restriction of dietary CP promoted the IMF content in pork (Doran et al., 2006; Madeira et al., 2013b; Tous et al., 2014), and feeding lysine deficiency diets could increase IMF content in fatty and lean-type pigs (Palma-Granados, et al., 2017). Whereas, Madeira et al. (2017) reported that dietary lysine restriction increased IMF content in lean pig but not in fatty pig (Madeira et al., 2013a). However, dietary threonine restriction did not change the fatty acid profile contents in porcine meat (Kobayashi et al., 2012). In ducks, dietary supplementation with *Clostridium butyricum* could increase IMF content in breast meat (Liu et al., 2018). Recently, in our laboratory, it was evaluated that threonine requirement is 0.61% for Pekin duck from 1 to 21 D of age. Meanwhile, we also observed that dietary threonine deficiency would increase hepatic total lipids of Pekin ducks (Jiang et al., 2017; Jiang et al., 2019a) and hepatic fatty acid profile contents (Jiang et al., 2019b). Whether dietary threonine supplementation affects fatty acid profile contents of breast muscle and responses of fatty-type duck to dietary threonine levels are similar to those of lean-type duck is still unknown.

Therefore, the present experiment was conducted to determine the effects of duck lines and dietary threonine levels on growth performance, carcass traits, hepatic lipids content, and fatty acids profiles in the liver, breast meat, and plasma of ducks and to evaluate the differences in IMF content between both duck lines and the role of dietary threonine supplementation on IMF content in fatty and lean-type Pekin ducks.

## MATERIALS AND METHOD

### Experimental Design and Treatments

A completely randomized design involving 2 duck lines × 3 dietary threonine levels was used in this

experiment. The 2 duck lines were either fatty-type Pekin ducks or lean-type Pekin ducks. The 3 dietary threonine treatments consisted of a corn–wheat–peanut meal basal diet with no threonine or supplemented with 0.15 and 0.30% crystalline threonine. Therefore, there were a total of 6 treatments in this experiment. The basal diets contained 15.45% CP and 2,977 kcal/kg metabolizable energy as-fed basis.

### Animals and Diets

The study was approved by the Animal Management Committee (in charge of animal welfare issue) of the Institute of Animal Science, Chinese Academy of Agricultural Sciences (IAS-CAAS, Beijing, China) and performed in accordance with the guidelines. Ethical approval on animal survival was given by the animal ethics committee of IAS-CAAS.

At 1 D of age, 500 Pekin ducks, 250 fatty-type ducks (high-sebum percentage) and 250 lean-type ducks (low-sebum percentage), were housed in raised wire floor pens (200 × 100 × 40 cm) from 1 to 14 D of age, respectively. The birds were raised in 50 cages of 10 birds per cage with nipple drinkers and tubular feeders and maintained under constant light. The temperature was maintained at 30°C from 1 to 3 D of age and then gradually reduced to 25°C until 14 D of age. Feed pellets and water were offered ad libitum. The birds were fed commercial diets.

At 15 D of age, a total of 384 ducks, 192 fatty-type and 192 lean-type ducks, were selected according to average BW and were randomly assigned to one of three diets with 8 replicate cages of 8 birds per cage for each treatment with similar BW, respectively. Birds were fed corn–wheat–peanut meal basal diet or the basal diet supplemented with 0.15 and 0.30% crystalline threonine for 21 D from 15 to 35 D of age. During the experimental

**Table 2.** Effects of threonine on growth performance of fatty-type and lean-type Pekin ducks from 14 to 35 D of age.

Lines	Threonine levels	BW at 14 D, g	BW at 35 D, g	ADFI, g	ADG, g	F/G, ( g/g )
Fatty <sup>1</sup>	0.41	607.7	2,198 <sup>a</sup>	191.7 <sup>a</sup>	75.76 <sup>a</sup>	2.53 <sup>b</sup>
	0.56	613.0	2,224 <sup>a</sup>	189.5 <sup>a</sup>	76.72 <sup>a</sup>	2.47 <sup>b</sup>
	0.71	605.0	2,183 <sup>a</sup>	184.6 <sup>a</sup>	75.15 <sup>a</sup>	2.46 <sup>b</sup>
	0.41	614.5	1,664 <sup>b</sup>	136.7 <sup>c</sup>	50.01 <sup>b</sup>	2.80 <sup>a</sup>
Lean <sup>1</sup>	0.56	617.6	2,169 <sup>a</sup>	171.4 <sup>b</sup>	73.90 <sup>a</sup>	2.28 <sup>c</sup>
	0.71	618.5	2,172 <sup>a</sup>	167.8 <sup>b</sup>	74.01 <sup>a</sup>	2.23 <sup>c</sup>
	Pooled SE line <sup>2</sup>	2.37	48.67	3.48	2.30	0.05
Pooled SE	Fatty	616.9 <sup>a</sup>	2,202	188.6	75.88	2.49
	Lean	608.6 <sup>b</sup>	2,002	158.6	65.97	2.45
Threonine <sup>3</sup>	0.41	1.37	28.10	2.01	1.33	0.03
	0.41	615.3	1,931	164.2	62.88	2.66
	0.56	611.8	2,196	180.4	75.31	2.38
Pooled SE	0.71	611.1	2,178	176.2	74.58	2.35
	1.68	34.41	2.46	1.63	0.04	
<i>P</i> -value	Lines	<0.001	<0.001	<0.001	<0.001	0.254
	Threonine	0.178	<0.001	<0.001	<0.001	<0.001
	Lines × Threonine	0.167	<0.001	<0.001	<0.001	<0.001

<sup>a-c</sup>Means within column with different superscripts differ ( $P < 0.05$ ).

Abbreviations: F/G, Feed/Gain; g, Gram.

<sup>1</sup>Data represent the means of 8 replicate cages ( $n = 8$ ).

<sup>2</sup>Data represent the means of 24 replicate cages ( $n = 24$ ).

<sup>3</sup>Data represent the means of 16 replicate cages ( $n = 16$ ).

period, ducks were reared in raised wire-floor pens (200 × 100 × 40 cm). Cages were equipped with nipple drinkers and tubular feeders. Feed (pellet form) and water were offered ad libitum. Experimental facility was maintained on a 24-h constant light, and the temperature was reduced gradually to 20°C from 15 to 21 D of age and then maintained at 20°C until the end of the experiment.

The basal diet (Table 1) was formulated to meet or exceed the nutrient needs of growth period as recommended by NRC (1994) except for threonine concentration. First, a single batch of basal diet was mixed and then divided into 3 sublots according to the experimental treatments. Each subplot was mixed with crystalline

threonine and corn starch, which was used to replace the crystalline threonine to achieve the experimental diets. The basal diets contained 15.45% CP and 0.41% threonine as-fed basis.

### Sample Collections and Analyses

At 35 D of age, BW and feed intake of birds per cage were recorded after a 12 h fast. Then, 3 birds per cage were selected according to the average BW of the cage. Blood samples were collected from each bird via wing vein puncture in 5-mL heparinized vacuum-tube syringes with stainless-steel needles, then centrifuged at 1,000 × *g* for 15 min at 4°C to separate plasma. The plasma samples

**Table 3.** Effects of threonine on carcass traits of fatty-type and lean-type Pekin ducks at 35 D of age (%).

Lines	Threonine levels	Breast muscle <sup>4</sup>	Thigh muscle <sup>4</sup>	Abdominal fat <sup>4</sup>	Sebum <sup>4</sup>
Fatty <sup>1</sup>	0.41	5.49 <sup>b</sup>	11.83 <sup>c</sup>	2.13	30.20 <sup>a</sup>
	0.56	6.04 <sup>b</sup>	13.27 <sup>a,b</sup>	2.40	29.50 <sup>a</sup>
	0.71	5.31 <sup>b</sup>	12.54 <sup>b,c</sup>	2.08	30.52 <sup>a</sup>
Lean <sup>1</sup>	0.41	5.46 <sup>b</sup>	13.95 <sup>a</sup>	1.68	23.99 <sup>c</sup>
	0.56	7.04 <sup>a</sup>	12.90 <sup>b</sup>	1.89	26.25 <sup>b</sup>
	0.71	7.83 <sup>a</sup>	13.00 <sup>b</sup>	1.63	24.16 <sup>c</sup>
Pooled SE line <sup>2</sup>	0.31	0.29	0.29	0.11	0.54
	Fatty	5.61	12.54	2.20 <sup>a</sup>	30.07
Pooled SE	Lean	6.78	13.28	1.73 <sup>b</sup>	24.80
	0.18	0.17	0.06	0.31	
Threonine <sup>3</sup>	0.41	5.48	12.89	1.90 <sup>b</sup>	27.09
	0.56	6.54	13.09	2.15 <sup>a</sup>	28.87
	0.71	6.56	13.77	1.85 <sup>b</sup>	27.34
Pooled SE	0.22	0.21	0.079	0.4	
	Lines	<0.001	0.004	<0.001	<0.001
<i>P</i> -value	Threonine	0.001	0.557	0.026	0.344
	Lines × Threonine	<0.001	<0.001	0.949	0.009

<sup>a-c</sup>Means within column with different superscripts differ ( $P < 0.05$ ).

<sup>1</sup>Data represent the means of 8 replicate cages ( $n = 8$ ).

<sup>2</sup>Data represent the means of 24 replicate cages ( $n = 24$ ).

<sup>3</sup>Data represent the means of 16 replicate cages ( $n = 16$ ).

<sup>4</sup>Organ index = Organ weight/Live body weight × 100%.

**Table 4.** Effects of threonine on serum lipid of fatty-type and lean-type Pekin ducks at 35 D of age.

Lines	Threonine levels	HDLC, mmol/L	LDLC, mmol/L	Total cholesterol, mg/dL	Triglyceride, mmol/L
Fatty <sup>1</sup>	0.41	2.15 <sup>b</sup>	0.93	3.49 <sup>b,c</sup>	0.58
	0.56	2.18 <sup>b</sup>	1.14	3.60 <sup>b,c</sup>	0.56
	0.71	2.34 <sup>b</sup>	1.19	3.99 <sup>a,b</sup>	0.61
Lean <sup>1</sup>	0.41	2.74 <sup>a</sup>	0.64	4.43 <sup>a</sup>	0.52
	0.56	2.13 <sup>b</sup>	0.67	3.47 <sup>b,c</sup>	0.61
	0.71	1.98 <sup>b</sup>	0.56	3.16 <sup>c</sup>	0.61
Pooled SE line <sup>2</sup>		0.13	0.07	0.21	0.05
	Fatty	2.28	1.09 <sup>a</sup>	3.69	0.58
Pooled SE	Lean	2.23	0.62 <sup>b</sup>	3.69	0.58
		0.07	0.04	0.12	0.03
Threonine <sup>3</sup>	0.41	2.44	0.79	3.96	0.55
	0.56	2.15	0.91	3.53	0.59
	0.71	2.16	0.87	3.57	0.61
Pooled SE		0.09	0.05	0.15	0.03
<i>P</i> -value	Lines	0.582	<0.001	0.099	0.475
	Threonine	0.047	0.260	0.974	0.949
	Lines × Threonine	0.002	0.079	0.001	0.518

<sup>a-c</sup>Means within column with different superscripts differ ( $P < 0.05$ ).  
 Abbreviations: HDLC, high density lipoprotein cholesterol; LDLC, low density lipoprotein cholesterol.  
<sup>1</sup>Data represent the means of 8 replicate cages (n = 8).  
<sup>2</sup>Data represent the means of 24 replicate cages (n = 24).  
<sup>3</sup>Data represent the means of 16 replicate cages (n = 16).

were stored at  $-20^{\circ}\text{C}$  until analysis for biochemical parameters and fatty acids profile. Then Pekin ducks were immediately killed by cervical dislocation. The liver samples were excised from the left lobe and were frozen ( $-20^{\circ}\text{C}$ ) for total lipid, triglyceride (TG), total cholesterol (TCHO), and fatty acid profile analysis. The breast muscle samples also were excised and were frozen ( $-20^{\circ}\text{C}$ ) for fatty acid profile analysis. In addition, another 2 birds from each cage were selected to isolate breast muscles, thigh muscle, abdominal fat, and liver and were weighed to calculate organ percentage.

**Crude Protein and Amino Acids**

Diet samples were ground to pass through a 0.50-mm screen and mixed thoroughly before analyses for CP

and amino acids. The CP concentration in the basal diet was determined by the Kjeldahl method (Thiex et al., 2002). The amino acids of basal diets were analyzed using ion-exchange chromatography by an amino acid analyzer (L-800, Hitachi, Tokyo, Japan) after diet samples were hydrolyzed with 6 mol/L HCl for 24 h at  $110^{\circ}\text{C}$ .

**Plasma Parameters**

Plasma concentrations of TCHO, TG, low-density lipoprotein cholesterol (LDLC), and high-density lipoprotein cholesterol (HDLC) were measured with an automatic analyzer (Hitachi 7080, Tokyo, Japan) by using commercial kits (Maccura, Sichuang, China).

**Table 5.** Effects of threonine on hepatic lipids of fatty-type and lean-type Pekin ducks at 35 D of age.

Lines	Threonine levels	Liver, %	Total lipid, %	Total cholesterol, mg/gProt	Triglyceride, mg/gProt
Fat <sup>1</sup>	0.41	2.20	5.12 <sup>c</sup>	0.0594 <sup>b,c</sup>	0.1522 <sup>c</sup>
	0.56	2.21	5.10 <sup>c</sup>	0.0525 <sup>c</sup>	0.1374 <sup>c</sup>
	0.71	2.19	5.03 <sup>c</sup>	0.0537 <sup>c</sup>	0.1519 <sup>c</sup>
Lean <sup>1</sup>	0.41	2.52	11.37 <sup>a</sup>	0.1015 <sup>a</sup>	0.5905 <sup>a</sup>
	0.56	2.23	6.57 <sup>b</sup>	0.0672 <sup>b</sup>	0.2406 <sup>b</sup>
	0.71	2.44	6.56 <sup>b</sup>	0.0674 <sup>b</sup>	0.2465 <sup>b</sup>
Pooled SE Line <sup>2</sup>		0.11	0.49	0.0045	0.0301
	Fatty	2.20	5.09	0.0552	0.1471
Pooled SE	Lean	2.40	8.17	0.0787	0.3592
		0.06	0.28	0.0026	0.0174
Threonine <sup>3</sup>	0.41	2.36	8.25	0.0804	0.3713
	0.56	2.22	5.84	0.0598	0.1890
	0.71	2.31	5.85	0.0606	0.1992
Pooled SE		0.074	0.35	0.0032	0.0213
<i>P</i> -value	Lines	0.062	<0.001	<0.001	<0.001
	Threonine	0.401	<0.001	<0.001	<0.001
	Lines × Threonine	0.348	<0.001	0.003	<0.001

<sup>a-c</sup>Means within column with different superscripts differ ( $P < 0.05$ ).  
<sup>1</sup>Data represent the means of 8 replicate cages (n = 8).  
<sup>2</sup>Data represent the means of 24 replicate cages (n = 24).  
<sup>3</sup>Data represent the means of 16 replicate cages (n = 16).

**Table 6.** Effects of threonine on hepatic fatty acid profiles of fatty-type and lean-type Pekin ducks at 35 D of age (mg/g).

Lines	Threonine levels	C14:0	C16:0	C20:0	C18:0	C22:0	C24:0	C16:1	C18:1n9c	C20:1	C24:1	C18:2n6c	C18:3n6
Fatty <sup>1</sup>	0.41	0.17 <sup>c</sup>	21.51 <sup>d</sup>	0.15 <sup>c</sup>	18.65 <sup>c</sup>	0.32	0.53	0.80 <sup>c</sup>	31.35 <sup>c</sup>	0.40 <sup>c,d</sup>	0.61	9.60	0.20 <sup>c</sup>
	0.56	0.17 <sup>c</sup>	21.71 <sup>d</sup>	0.14 <sup>c</sup>	18.32 <sup>c</sup>	0.30	0.54	0.76 <sup>c</sup>	29.22 <sup>c</sup>	0.40 <sup>c,d</sup>	0.64	9.42	0.21 <sup>c</sup>
	0.71	0.19 <sup>c</sup>	23.67 <sup>c,d</sup>	0.15 <sup>c</sup>	19.04 <sup>c</sup>	0.33	0.55	0.80 <sup>c</sup>	35.02 <sup>c</sup>	0.38 <sup>d</sup>	0.59	10.02	0.21 <sup>c</sup>
Lean <sup>1</sup>	0.41	1.09 <sup>a</sup>	54.74 <sup>a</sup>	0.29 <sup>a</sup>	24.28 <sup>a</sup>	0.37	0.57	2.76 <sup>a</sup>	97.98 <sup>a</sup>	0.65 <sup>a</sup>	0.37	14.45	0.37 <sup>a</sup>
	0.56	0.46 <sup>b</sup>	34.20 <sup>b</sup>	0.23 <sup>b</sup>	21.06 <sup>b</sup>	0.38	0.55	1.74 <sup>b</sup>	57.05 <sup>b</sup>	0.50 <sup>b</sup>	0.40	12.24	0.25 <sup>b,c</sup>
	0.71	0.39 <sup>b,c</sup>	32.08 <sup>b,c</sup>	0.22 <sup>b</sup>	20.86 <sup>b</sup>	0.39	0.58	1.23 <sup>b,c</sup>	48.71 <sup>b,c</sup>	0.46 <sup>b,c</sup>	0.42	12.33	0.31 <sup>b</sup>
Pooled SE line <sup>2</sup>		0.085	3.22	0.012	0.621	0.01	0.023	0.189	6.32	0.022	0.031	0.68	0.022
	Fatty	0.17	22.23	0.15 <sup>b</sup>	18.67	0.32 <sup>b</sup>	0.54	0.78	31.86	0.39	0.61 <sup>a</sup>	9.68 <sup>b</sup>	0.21
Pooled SE	Lean	0.64	40.34	0.25 <sup>a</sup>	22.06	0.38 <sup>a</sup>	0.57	1.91	67.91	0.54	0.40 <sup>b</sup>	13.01 <sup>a</sup>	0.31
		0.049	1.86	0.007	0.358	0.006	0.013	0.109	3.65	0.014	0.018	0.39	0.013
Threonine <sup>3</sup>	0.41	0.62	38.12	0.22	21.47	0.35	0.55	1.78	64.66	0.52	0.5	12.02	0.29
	0.56	0.31	27.96	0.19	19.95	0.34	0.54	1.25	43.13	0.45	0.52	10.83	0.23
	0.71	0.28	27.88	0.19	19.69	0.36	0.57	1.01 <sup>a</sup>	41.87	0.43	0.51	11.18	0.25
Pooled SE		0.060	2.27	0.008	0.439	0.007	0.016	0.133	4.67	0.017	0.022	0.48	0.016
P-value	Lines	<0.001	<0.001	<0.001	<0.001	<0.001	0.174	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
	Threonine	<0.001	0.003	0.004	0.013	0.246	0.563	0.022	0.001	<0.001	0.788	0.208	0.042
	Lines × Threonine	<0.001	0.001	0.005	0.010	0.362	0.831	0.008	<0.001	0.002	0.356	0.155	0.025

<sup>a-d</sup>Means within column with different superscripts differ ( $P < 0.05$ ).

Abbreviations: MUFA, monounsaturated fatty acid; PUFA, polyunsaturated fatty acid; SFA, saturated fatty acid; TFA, total fatty acid; UFA, unsaturated fatty acid.

1. Data represent the means of 8 replicate cages ( $n = 8$ ).

2. Data represent the means of 24 replicate cages ( $n = 24$ ).

3. Data represent the means of 16 replicate cages ( $n = 16$ ).

## Total Lipids, Triglycerides, and Cholesterol

Total lipids in the liver were measured according to the method described by Folch et al. (1957). Liver samples were homogenized in ice-cold 10% (wt/vol) physiological saline for 60 s and then sonicated with an ultrasonic wave cell grinder (JY92-11; Ningbo, Jiangsu, China) for 1 min (1 s on followed by 2 s interval for each sonication). The homogenates were centrifuged at  $1,000 \times g$  for 15 min at 4°C, and the supernatants were collected to determine the concentration of total protein, TCHO, and TG. Total protein concentration was determined using a BCA Protein Assay kit (Pierce, Rockford, IL). The concentrations of TG and TCHO

were determined with a microplate reader by using commercial kits (Nanjing Jiangcheng Bioengineering Institute, NanJing, China) and expressed as milligram per milligram protein.

## Fatty Acids Compositions

The liver, breast muscle, and 200  $\mu$ L plasma samples were freeze-dry prepared according to the method described previously (Yang, et al., 2010). Briefly, fatty acids were extracted from total lipids and methylated by adding 1 mL acetylchloride/methanol (1/10, v/v) and 20  $\mu$ L of 5 mg/mL nonadecanic acid (used as

**Table 7.** Effects of threonine on breast muscle fatty acid profiles of fatty-type and lean-type Pekin ducks at 35 D of age (mg/g).

Lines	Threonine levels	C14:0	C16:0	C18:0	C20:0	C22:0	C24:0	C16:1	C18:1n9c	C20:1	C24:1	C18:2n6c	C18:3n6
Fatty <sup>1</sup>	0.41	0.078	7.6	6.14	0.23 <sup>b</sup>	0.18 <sup>b</sup>	0.29 <sup>b,c</sup>	0.28	10.15	0.16 <sup>c</sup>	0.12 <sup>a,b</sup>	5.10 <sup>c,d</sup>	0.078
	0.56	0.1	8.52	6.22	0.23 <sup>b</sup>	0.17 <sup>b</sup>	0.29 <sup>b,c</sup>	0.43	12.56	0.17 <sup>b,c</sup>	0.12 <sup>a,b</sup>	5.63 <sup>b,c</sup>	0.087
	0.71	0.088	7.61	6.34	0.23 <sup>b</sup>	0.17 <sup>b</sup>	0.29 <sup>c</sup>	0.4	10.89	0.16 <sup>c</sup>	0.12 <sup>a,b,c</sup>	5.54 <sup>b,c</sup>	0.084
Lean <sup>1</sup>	0.41	0.081	7.3	6.68	0.28 <sup>a</sup>	0.21 <sup>a</sup>	0.36 <sup>a</sup>	0.23	9.79	0.14 <sup>c</sup>	0.15 <sup>a</sup>	4.73 <sup>d</sup>	0.098
	0.56	0.124	8.99	6.9	0.22 <sup>b</sup>	0.18 <sup>b</sup>	0.31 <sup>b</sup>	0.4	12.7	0.22 <sup>a</sup>	0.08 <sup>c</sup>	6.70 <sup>a</sup>	0.09
	0.71	0.089	8.37	6.49	0.22 <sup>b</sup>	0.17 <sup>b</sup>	0.30 <sup>b,c</sup>	0.33	11.48	0.19 <sup>b</sup>	0.09 <sup>b,c</sup>	6.14 <sup>a,b</sup>	0.088
Pooled SE line <sup>2</sup>		0.011	0.344	0.15	0.007	0.007	0.008	0.054	0.8	0.01	0.012	0.213	0.005
	Fatty	0.09	7.91	6.24 <sup>b</sup>	0.23	0.17	0.29	0.37	11.2	0.16	0.12	5.42	0.083 <sup>b</sup>
Pooled SE	Lean	0.1	8.22	6.69 <sup>a</sup>	0.24	0.19	0.32	0.32	11.32	0.18	0.11	5.86	0.092 <sup>a</sup>
		0.007	0.199	0.086	0.004	0.004	0.005	0.031	0.46	0.006	0.007	0.123	0.003
Threonine <sup>3</sup>	0.41	0.08 <sup>b</sup>	7.45 <sup>b</sup>	6.41	0.26	0.19	0.33	0.26 <sup>b</sup>	9.97 <sup>b</sup>	0.15	0.13	4.913	0.088
	0.56	0.11 <sup>a</sup>	8.76 <sup>a</sup>	6.56	0.23	0.18	0.3	0.42 <sup>a</sup>	12.63 <sup>a</sup>	0.19	0.1	6.168	0.09
	0.71	0.09 <sup>b</sup>	7.99 <sup>b</sup>	6.42	0.23	0.17	0.29	0.37 <sup>a</sup>	11.19 <sup>a,b</sup>	0.1	0.1	5.842	0.086
Pooled SE		0.008	0.244	0.106	0.005	0.005	0.006	0.038	0.567	0.007	0.009	0.15	0.004
P-value	Lines	0.317	0.279	0.001	0.008	0.003	<0.001	0.305	0.856	0.006	0.279	0.017	0.034
	Threonine	0.021	0.002	0.534	<0.001	0.005	<0.001	0.017	0.008	<0.001	0.013	<0.001	0.865
	Lines × Threonine	0.541	0.291	0.202	<0.001	0.042	0.005	0.935	0.839	0.008	0.019	0.005	0.209

<sup>a-d</sup>Means within column with different superscripts differ ( $P < 0.05$ ).

Abbreviations: MUFA, monounsaturated fatty acid; PUFA, polyunsaturated fatty acid; SFA, saturated fatty acid; TFA, total fatty acid; UFA, unsaturated fatty acid.

1. Data represent the means of 8 replicate cages ( $n = 8$ ).

2. Data represent the means of 24 replicate cages ( $n = 24$ ).

3. Data represent the means of 16 replicate cages ( $n = 16$ ).

**Table 6.** (Continued)

C18:3n3	C20:2	C22:2	C20:3n6	C20:4n6	C20:5n3	C22:6n3	SFA	MUFA	PUFA	UFA	TFA	N3	N6	N6:N3	PUFA/SFA
0.10	3.40	0.40	3.32	24.13	0.19	0.49 <sup>a</sup>	41.34 <sup>d</sup>	33.16 <sup>c</sup>	41.82	74.98 <sup>c</sup>	116.3 <sup>c,d</sup>	0.77	37.25	48.48 <sup>b</sup>	1.01 <sup>a</sup>
0.10	3.27	0.39	3.29	24.67	0.18	0.47 <sup>a,b</sup>	41.18 <sup>d</sup>	31.01 <sup>c</sup>	42.00	73.00 <sup>c</sup>	114.2 <sup>d</sup>	0.75	37.59	50.01 <sup>b</sup>	1.02 <sup>a</sup>
0.10	3.37	0.41	3.44	24.1	0.18	0.47 <sup>a,b</sup>	43.93 <sup>c,d</sup>	36.74 <sup>c</sup>	42.31	79.05 <sup>c</sup>	123.0 <sup>c,d</sup>	0.76	37.77	49.87 <sup>b</sup>	0.99 <sup>a</sup>
0.18	2.19	0.42	2.75	18.48	0.13	0.36 <sup>c</sup>	81.33 <sup>a</sup>	101.76 <sup>a</sup>	39.33	141.09 <sup>a</sup>	222.42 <sup>a</sup>	0.67	36.05	55.41 <sup>a</sup>	0.50 <sup>c</sup>
0.14	2.68	0.38	3.10	20.98	0.19	0.43 <sup>b</sup>	56.87 <sup>b</sup>	59.68 <sup>b</sup>	40.39	100.07 <sup>b</sup>	156.9 <sup>b</sup>	0.76	36.57	48.31 <sup>b</sup>	0.75 <sup>b</sup>
0.16	2.51	0.37	3.20	21.66	0.20	0.43 <sup>b</sup>	54.52 <sup>b,c</sup>	50.82 <sup>b,c</sup>	41.18	92.00 <sup>b,c</sup>	146.5 <sup>b,c</sup>	0.80	37.5	46.95 <sup>b</sup>	0.77 <sup>b</sup>
0.015	0.156	0.021	0.171	0.989	0.016	0.017	3.779	6.452	1.363	6.569	10.13	0.032	1.289	1.704	0.045
0.10 <sup>b</sup>	3.34 <sup>a</sup>	0.4	3.35 <sup>a</sup>	24.30 <sup>a</sup>	0.1825	0.48	42.15	33.64	42.04	75.68	117.8	0.76	37.54	49.45	1.01
0.16 <sup>a</sup>	2.46 <sup>b</sup>	0.39	3.02 <sup>b</sup>	20.37 <sup>b</sup>	0.1754	0.41	64.24	70.75	40.3	111.05	175.3	0.74	36.71	50.22	0.67
0.008	0.09	0.012	0.099	0.571	0.009	0.01	2.182	3.725	0.787	3.792	5.85	0.019	0.744	0.984	0.026
0.14	2.79	0.41	3.04	21.31	0.16	0.42	61.34	67.46	40.57	108.03	169.4	0.72	36.65	51.95	0.75
0.12	2.97	0.39	3.19	22.83	0.18	0.45	49.03	45.34	41.19	86.54	135.6	0.76	37.08	49.16	0.88
0.13	2.94	0.39	3.32	22.88	0.19	0.45	49.23	43.78	41.74	85.53	134.8	0.78	37.64	48.41	0.88
0.010	0.110	0.015	0.121	0.699	0.011	0.012	2.672	4.562	0.964	4.645	7.16	0.023	0.911	1.205	0.032
<0.001	<0.001	0.718	0.021	<0.001	0.582	<0.001	<0.001	<0.001	0.125	<0.001	<0.001	0.455	0.436	0.583	<0.001
0.45	0.483	0.483	0.253	0.208	0.104	0.099	0.003	0.001	0.695	0.002	0.002	0.197	0.748	0.104	0.008
0.465	0.152	0.352	0.485	0.274	0.070	0.010	0.001	<0.001	0.880	0.0006	0.001	0.071	0.93	0.012	0.005

internal standard) for 4 h at 80°C. Then, 1 mL of hexane and 1.5 mL of 6% (m/v) K<sub>2</sub>CO<sub>3</sub> were added into the tube. After shaking for 10 min, 400 µL of hexane layer was transferred to a new injection vial after centrifugation for 10 min at 3,000 × *g*. One microliter of the prepared sample was injected into the 7890A GC-FID system (Agilent Technologies, Palo Alto, CA) to determine the fatty acid profiles. The system was equipped with a DB-23 column (Agilent Technologies, 60 m × 0.25 mm × 0.25 µm) at a split ratio of 1:50. Injector and detector temperatures were 250°C and 250°C, respectively. The oven was programmed as follows: 50°C for 2 min, ramp to 175°C at 20°C/min, ramp to 220°C at 2°C/min, and finally ramp to 230°C at 4°C/min, holding 5 min with 1.1 mL/min nitrogen

as carrier gas. Peaks were identified by comparing retention times with those of the corresponding standards (Sigma Aldrich, Santa Clara, CA).

### Statistical Analysis

Data from the experiment were subjected to two-way ANOVA using general linear model procedure of SAS software (SAS Institute Inc., Cary, NC, 2003). Differences among means were tested by Duncan’s method. The level of statistical significance was set at *P* < 0.05. The cage served as the experimental unit for growth performance, and selected birds were served as the experimental unit for other measurements.

**Table 7.** (Continued)

C18:3n3	C20:2	C20:3n6	C20:4n6	C20:5n3	C22:2	C22:6n3	SFA	MUFA	PUFA	UFA	TFA	N3	N6	N6/n3	PUFA/SFA
0.044	1.95 <sup>a</sup>	1.01 <sup>a</sup>	5.58 <sup>b</sup>	0.085 <sup>a</sup>	0.19 <sup>a</sup>	0.28	16.47	11.97	14.31	26.28	42.74	0.41 <sup>b,c</sup>	11.77	28.99 <sup>b</sup>	0.87
0.069	1.87 <sup>a,b</sup>	1.03 <sup>a</sup>	5.44 <sup>b</sup>	0.093 <sup>a</sup>	0.18 <sup>a</sup>	0.26	17.4	13.28	14.66	27.84	45.34	0.42 <sup>b,c</sup>	12.19	29.03 <sup>b</sup>	0.84
0.056	1.98 <sup>a</sup>	1.01 <sup>a</sup>	5.18 <sup>b</sup>	0.091 <sup>a</sup>	0.18 <sup>a</sup>	0.27	16.71	11.57	14.38	25.95	42.66	0.41 <sup>b,c</sup>	11.81	28.61 <sup>b</sup>	0.86
0.053	1.22 <sup>c</sup>	0.83 <sup>b</sup>	6.81 <sup>a</sup>	0.049 <sup>b</sup>	0.15 <sup>b</sup>	0.27	16.13	10.32	14.21	24.53	40.66	0.37 <sup>c</sup>	12.46	34.02 <sup>a</sup>	0.88
0.101	1.77 <sup>a,b</sup>	1.00 <sup>a</sup>	5.66 <sup>b</sup>	0.095 <sup>a</sup>	2.00 <sup>a</sup>	0.27	18.5	13.39	15.89	29.28	47.78	0.47 <sup>a</sup>	13.45	28.79 <sup>b</sup>	0.86
0.071	1.70 <sup>b</sup>	0.98 <sup>a</sup>	5.53 <sup>b</sup>	0.086 <sup>a</sup>	0.19 <sup>a</sup>	0.28	17.34	12.1	15.06	27.16	44.51	0.43 <sup>a,b</sup>	12.73	29.54 <sup>b</sup>	0.87
0.009	0.069	0.032	0.176	0.006	0.007	0.004	0.466	0.986	0.294	1.17	1.591	0.015	0.264	1.00	0.015
0.056 <sup>b</sup>	1.93	1.02	5.40	0.089	0.18	0.27	16.86	12.27	14.45 <sup>b</sup>	26.72	43.58	0.41	11.92 <sup>b</sup>	28.88	0.86
0.075 <sup>a</sup>	1.56	0.93	6.00	0.077	0.18	0.28	17.32	11.94	15.05 <sup>a</sup>	26.99	44.32	0.43	12.88 <sup>a</sup>	30.78	0.87
0.005	0.04	0.019	0.102	0.004	0.004	0.005	0.269	0.569	0.17	0.676	0.919	0.008	0.152	0.578	0.009
0.048 <sup>b</sup>	1.58	0.92	6.92	0.067	0.17	0.28	16.30 <sup>b</sup>	11.14	14.26 <sup>b</sup>	25.40 <sup>b</sup>	41.70 <sup>b</sup>	0.39	12.12 <sup>b</sup>	31.5	0.88
0.085 <sup>a</sup>	1.82	1.02	5.55	0.094	0.19	0.27	17.95 <sup>a</sup>	13.34	15.27 <sup>a</sup>	28.61 <sup>a</sup>	46.56 <sup>a</sup>	0.45	12.82 <sup>a</sup>	29.08	0.85
0.063 <sup>b</sup>	1.84	0.99	5.35	0.089	0.19	0.27	17.03 <sup>a,b</sup>	11.84	14.72 <sup>a,b</sup>	26.56 <sup>a,b</sup>	43.58 <sup>a,b</sup>	0.42	12.27 <sup>b</sup>	28.91	0.87
0.006	0.049	0.023	0.124	0.005	0.005	0.007	0.33	0.697	0.208	0.828	1.125	0.01	0.187	0.707	0.011
0.013	<0.001	0.003	<0.001	0.016	0.889	0.219	0.227	0.681	0.017	0.782	0.576	0.284	<0.0001	0.025	0.342
0.001	0.001	0.014	<0.001	<0.001	0.026	0.465	0.004	0.087	0.005	0.029	0.014	0.002	0.027	0.021	0.347
0.418	<0.001	0.033	0.012	0.011	0.001	0.392	0.302	0.506	0.089	0.337	0.313	0.032	0.561	0.030	0.968

**Table 8.** Effects of threonine on serum fatty acid profiles of fatty-type and lean-type Pekin ducks at 35 D of age (ug/mL).

Lines	Threonine levels	C14:0	C16:0	C18:0	C20:0	C22:0	C24:0	C16:1	C18:1n9c	C18:2n6c	C20:1	C18:3n6	C18:3n3
Fatty <sup>1</sup>	0.41	9.5	823.81	414.17 <sup>b</sup>	2.21	5.09	15.00 <sup>c</sup>	43.01	1,100.58	342.89	9.39	16.77	3.71
	0.56	9.02	856.9	412.92 <sup>b</sup>	2.46	5.17	15.52 <sup>c</sup>	47.24	1,148.71	343.03	9.95	16.56	3.67
	0.71	8.66	851.7	413.83 <sup>b</sup>	2.27	5.32	15.73 <sup>b,c</sup>	41.24	1,110.4	363.11	9.31	18.64	3.63
Lean <sup>1</sup>	0.41	8.96	790.75	505.24 <sup>a</sup>	2.71	6.5	18.54 <sup>a,c</sup>	33.16	968.91	366.47	8.14	17.82	4.17
	0.56	8.62	791.84	417.41 <sup>b</sup>	2.55	5.71	15.94 <sup>b,c</sup>	39.41	1,031.59	389.83	8.72	19.43	4.57
	0.71	9.93	886.37	465.36 <sup>a,b</sup>	2.99	6.84	17.39 <sup>a,b</sup>	39.41	1,064.54	408.18	8.96	19.85	4.87
Pooled SE Line <sup>2</sup>		0.43	23.03	16.62	0.192	0.28	0.578	3.47	39.02	11.52	0.52	0.94	0.292
	Fatty	9.06	844.14	413.64 <sup>b</sup>	2.75 <sup>a</sup>	5.19 <sup>b</sup>	15.42 <sup>b</sup>	43.83 <sup>a</sup>	1,119.90 <sup>a</sup>	349.68 <sup>b</sup>	9.55 <sup>a</sup>	17.33 <sup>b</sup>	3.67 <sup>b</sup>
Pooled SE	Lean	9.17	822.99	462.67 <sup>a</sup>	2.31 <sup>b</sup>	6.35 <sup>a</sup>	17.29 <sup>a</sup>	37.33 <sup>b</sup>	1,021.68 <sup>b</sup>	388.16 <sup>a</sup>	8.61 <sup>b</sup>	19.04 <sup>a</sup>	4.54 <sup>a</sup>
		0.25	13.30	9.59	0.11	0.16	0.33	2.00	22.52	6.54	0.30	0.54	0.17
Threonine <sup>3</sup>	0.41	9.226	807.28 <sup>b</sup>	459.70 <sup>a</sup>	2.46	5.8	16.77	38.08	1,030.74	354.68 <sup>b</sup>	8.77	17.29	3.94
	0.56	8.821	824.37 <sup>a,b</sup>	439.60 <sup>a,b</sup>	2.5	5.44	15.73	43.33	1,090.15	366.43 <sup>a,b</sup>	9.33	18	4.12
	0.71	9.293	869.04 <sup>a</sup>	415.17 <sup>b</sup>	2.63	6.08	16.56	40.32	1,087.47	385.64 <sup>a</sup>	9.14	19.25	4.25
Pooled SE		0.301	16.28	11.749	0.135	0.196	0.404	2.454	27.59	8.149	0.366	0.664	0.208
P-value	Lines	0.746	0.267	0.001	0.008	<0.001	<0.001	0.027	0.004	<0.001	0.032	0.031	0.001
	Threonine	0.494	0.03	0.036	0.653	0.084	0.167	0.326	0.288	0.034	0.544	0.121	0.564
	Lines × Threonine	0.072	0.099	0.043	0.261	0.163	0.030	0.491	0.506	0.538	0.616	0.566	0.422

<sup>a-d</sup>Means within column with different superscripts differ ( $P < 0.05$ ).

Abbreviations: MUFA, monounsaturated fatty acid; PUFA, polyunsaturated fatty acid; SFA, saturated fatty acid; TFA, total fatty acid; UFA, unsaturated fatty acid.

1. Data represent the means of 8 replicate cages ( $n = 8$ ).

2. Data represent the means of 24 replicate cages ( $n = 24$ ).

3. Data represent the means of 16 replicate cages ( $n = 16$ ).

## RESULTS

### Growth Performance

Growth performance data were presented in Table 2. The fatty-type ducks had higher ( $P < 0.001$ ) initial BW than lean-type ducks at 14 D of age. The duck lines, threonine level, and its interaction affected ( $P < 0.001$ ) BW, ADFI, and ADG of the Pekin ducks from 14 to 35 D of age. The threonine levels and interaction between duck lines and threonine level affected ( $P < 0.001$ ) feed/gain (F/G) of the Pekin ducks from 14 to 35 D of age, while duck lines had no influence ( $P = 0.254$ ) on F/G. The lean duck fed diet with 0.56 and 0.71% threonine had higher ( $P < 0.05$ ) BW, ADFI, and ADG and lower ( $P < 0.05$ ) F/G than those fed diet with 0.41% threonine, but fatty-type ducks fed diet with all the 3 level threonine had no difference ( $P > 0.05$ ) in BW, ADFI, ADG, and F/G.

### Carcass Traits

Carcass traits data were presented in Table 3. Duck lines, threonine levels, and its interaction affected ( $P \leq 0.001$ ) breast muscle percentage. Duck lines and interaction between duck lines and threonine levels affected ( $P \leq 0.004$ ) the percentage of thigh muscle and sebum. Duck lines and threonine levels affected ( $P \leq 0.026$ ) abdominal fat percentage, and its interaction did not affect ( $P = 0.949$ ) abdominal fat percentage. Fatty-type ducks had higher ( $P < 0.05$ ) abdominal fat percentage than that in lean-type ducks, and the both ducks fed diet with 0.56% threonine had higher ( $P = 0.026$ ) abdominal fat percentage than those fed diets with 0.41 and 0.71% threonine. The lean-type ducks fed diets with 0.56 and 0.71% threonine had higher

( $P < 0.05$ ) breast muscle percentage than those fed diet with 0.41% threonine, but breast muscle percentage in fatty-type ducks was not affected ( $P > 0.05$ ) by the 3 diets. The lean-type ducks fed diets with 0.41 and 0.71% threonine had lower ( $P < 0.05$ ) sebum percentage than those fed diet with 0.56% threonine, but in fatty-type ducks, the 3 diets did not affect ( $P > 0.05$ ) sebum percentage. At 0.41% threonine diets, fatty-type ducks had lower ( $P < 0.05$ ) thigh muscle percentage than that in lean-type ducks. But at 0.56 and 0.71% threonine diets, the both line ducks had similar thigh muscle percentage.

### Plasma Lipid Concentrations

Plasma lipid data were presented in Table 4. The threonine level and interaction between duck line and threonine levels affected ( $P < 0.05$ ) plasma HDLC. The duck line ( $P < 0.001$ ) affected plasma LDLC, and the threonine levels and the interaction between duck line and threonine levels did not affect ( $P > 0.30$ ) plasma LDLC. The interaction between duck line and threonine affected ( $P = 0.001$ ) plasma TCHO. Duck line did not affect ( $P = 0.582$ ) plasma HDLC, and threonine level and interaction between duck line and threonine level had no influence ( $P > 0.08$ ) on plasma TCHO. Duck line, threonine level, and its interaction did not affect ( $P > 0.47$ ) the plasma TG. Fatty-type ducks had higher ( $P < 0.001$ ) plasma LDLC than that in lean-type ducks. Lean-type ducks fed with diet containing 0.41% threonine had higher ( $P < 0.05$ ) plasma HDLC and TCHO concentration than those fed with diet containing 0.56 and 0.71% threonine, whereas there was similar plasma concentration of HDLC and TCHO in fatty-type ducks fed diets with the 3 threonine levels.

**Table 8.** (Continued)

C20:2	C20:3n6	C20:4n6	C20:5n3	C22:2	C22:6n3	SFA	MUFA	PUFA	UFA	TFA	N3	N6	N6/n3	PUFA/SFA
177.92	78.94	765.81	8.66	10.57	19.75	1,270	1,160	1,414	2,574	3,844	32.12	1,204	37.85	1.12
174.55	81.12	772.1	9.03	9.95	20.28	1,302	1,213	1,420	2,633	3,835	32.99	1,212	36.79	1.09
185.46	82.63	756.51	10.04	9.85	20.3	1,298	1,168	1,440	2,608	3,906	33.97	1,221	36.59	1.11
98.79	65.28	832.74	8.57	9.39	19.2	1,333	1,018	1,413	2,431	3,763	31.94	1,282	40.46	1.06
120.71	71.05	704.7	9.63	8.62	20.23	1,242	1,087	1,340	2,427	3,670	34.43	1,185	34.53	1.08
123.13	72.48	762.34	8.97	9.81	20.47	1,389	1,119	1,420	2,540	3,929	34.31	1,263	36.85	1.02
8.30	1.70	31.20	0.708	0.498	0.87	32.80	40.10	37.8	64.9	92.5	1.379	35.8	1.273	0.02
179.31 <sup>a</sup>	80.90 <sup>a</sup>	764.81	9.24	10.12 <sup>a</sup>	20.117	1,289	1,180 <sup>a</sup>	1,425	2,605 <sup>a</sup>	3,895	33.02	1,212	37.08	1.11 <sup>a</sup>
114.21 <sup>b</sup>	69.61 <sup>b</sup>	766.59	9.06	9.27 <sup>b</sup>	19.97	1,321	1,074 <sup>b</sup>	1,391	2,466 <sup>b</sup>	3,787	33.56	1,243	37.28	1.05 <sup>b</sup>
4.79	1.70	18.01	0.41	0.28	0.50	18.9	23.1	21.8	37.4	53.4	0.796	20.7	0.735	0.012
138.35	72.11	799.27	8.61	0.98	19.48	1,301	1,089	1,414	2,503	3,804	32.03	1,243	39.16 <sup>a</sup>	1.09
147.63	76.09	738.4	9.33	9.28	20.26	1,272	1,150	1,380	2,530	3,803	33.71	1,199	35.66 <sup>b</sup>	1.09
154.3	77.56	759.42	9.5	9.83	20.38	1,343	1,143	1,430	2,574	3,917	34.14	1,242	36.72 <sup>a,b</sup>	1.07
5.87	2.09	22.06	0.50	0.348	0.614	23.2	28.4	26.7	45.9	65.4	0.975	25.30	0.90	0.014
<0.001	<0.001	0.944	0.746	0.040	0.841	0.247	0.002	0.279	0.012	0.16	0.636	0.299	0.845	0.003
0.168	0.174	0.153	0.419	0.339	0.530	0.105	0.254	0.41	0.544	0.372	0.280	0.378	0.026	0.513
0.310	0.786	0.110	0.503	0.368	0.913	0.061	0.467	0.557	0.575	0.298	0.835	0.333	0.173	0.177

**Hepatic Lipid Concentrations**

Hepatic lipid data were presented in Table 5. Duck lines, dietary threonine, and its interaction affected ( $P < 0.004$ ) the hepatic total lipids, TCHO, and TG of ducks, but it did not affect ( $P > 0.348$ ) the relative liver weight. The hepatic total lipids, TCHO, and TG concentrations were higher ( $P < 0.05$ ) in lean-type duck fed diet with 0.41% threonine than those fed diet with 0.56 and 0.71% threonine, while no difference ( $P > 0.05$ ) was observed in fatty-type ducks fed diet with 0.41, 0.56, and 0.71% threonine.

**Hepatic Fatty Acid Profiles**

Hepatic fatty acid data were presented in Table 6. The duck lines affected ( $P < 0.020$ ) the hepatic concentration of C22:0, C24:1, C18:2n6c, C18:3n3, C20:2, C20:3n6, and C20:4n6 of ducks, but threonine level and the interaction between duck lines and threonine level did not affect ( $P > 0.05$ ) these indices. Duck line, threonine level, and its interaction affected ( $P < 0.05$ ) the C14:0, C16:0, C18:0, C20:0, C16:1, C18:1n9c, C20:1, C18:3n6, saturated fatty acid (SFA), monounsaturated fatty acid (MUFA), unsaturated fatty acid (UFA), total fatty acid (TFA), and polyunsaturated fatty acid (PUFA)/SFA, but did not affect ( $P > 0.05$ ) C24:0, C22:2, C20:5n3, PUFA, and N3 and N6 PUFA. The interaction between duck lines and threonine level affected ( $P < 0.020$ ) C22:6n3 and N6/N3. The lean-type ducks had higher ( $P < 0.001$ ) concentration of C22:0, C18:2n6c, and C18:3n3 fatty acids, but lower ( $P < 0.020$ ) concentration of C24:1, C20:2, C20:3n6, and C20:4n6 fatty acids than that in fatty-type ducks had. Diets with 0.41% threonine increased ( $P < 0.05$ ) the concentrations of C14:0, C16:0, C18:0, C20:0, C16:1, C18:1n9c, C20:1, C18:3n6, SFA, MUFA, UFA, TFA, and N6/N3 and reduced C22:6n3 in lean-type

ducks, compared with those fed with diet containing 0.56 and 0.71%. However, no differences were observed ( $P > 0.05$ ) in the concentration of C14:0, C16:0, C18:0, C20:0, C16:1, C18:1n9c, C20:1, C18:3n6, C22:6n3, SFA, MUFA, UFA, TFA, and N6/N3 in fatty-type ducks fed diets with 0.41, 0.56, and 0.71% threonine.

**Breast Muscle Fatty Acid Profile**

Breast muscle fatty acid data were presented in Table 7. The duck lines affected ( $P = 0.001$ ) the concentration of C18:0, C18:3n3, and PUFA in breast muscle, whereas dietary threonine levels affected ( $P < 0.03$ ) C14:0, C16:0, C16:1, C18:3n3, SFA, PUFA, UFA, TFA, N3, and N6 in breast muscle. The duck line, threonine levels, and its interaction affected ( $P < 0.05$ ) C20:0, C22:0, C24:0, C20:1, C18:2n6c, C20:2, C20:3n6, C20:4n6, C20:5n3, N6, and N3/N6 in breast muscle. Lean-type ducks had higher ( $P < 0.02$ ) concentrations of C18:0, C18:3n3, PUFA, and N6 in breast muscle than that in fatty-type ducks. The threonine levels and interaction between threonine level and duck lines affected ( $P < 0.04$ ) the C24:1 and N3 in breast muscle of ducks. The both type ducks fed diets with 0.56% threonine had higher ( $P < 0.03$ ) concentrations of C14:0, C16:0, C18:3n3, SFA, PUFA, UFA, TFA, and N6 compared with 0.41% threonine. Lean ducks fed diet with 0.41% threonine had higher ( $P < 0.05$ ) C20:0, C22:0, C24:0, C24:1, C18:2n6c, C18:3n3, C20:2, C20:3n6, and C20:4n6 and lower ( $P < 0.02$ ) C20:2 and C20:5n3 in breast muscle compared with the ducks fed diet with 0.56 and 0.71% threonine.

**Plasma Fatty Acid Profile**

Plasma fatty acid data were presented in Table 8. Duck lines affected ( $P < 0.05$ ) C20:0, C22:0, C16:1, C18:1n9c, C18:2n6c, C20:1, C18:3n6, C18:3n3, C20:2,



C20:3n6, C22:2, MUFA, and the ratio of PUFA/SFA, but it did not affect ( $P > 0.05$ ) C14:0, C16:0, C20:4n6, C20:5n3, C20:6n3, SFA, MUFA, UFA, TFA, N3, N6, and the ratio of N6/N3 in plasma of duck. Threonine levels only affected C16:0 and the ratio of N6/N3, and it did not affect ( $P > 0.05$ ) the other fatty acids in plasma. The duck lines, threonine level, and its interaction affected ( $P = 0.043$ ) the C16:0, and duck lines and the interaction between duck lines and threonine affected ( $P = 0.03$ ) the C24:0 concentration in plasma. The fatty-type ducks had higher ( $P < 0.03$ ) C20:0, C16:1, C18:1n9c, C20:1, C20:2, C20:3n6, and MUFA and lower ( $P < 0.04$ ) C22:0, C18:2n6c, C18:3n6, and C18:3n3 than that in lean-type ducks. Lean-type ducks fed diet with 0.41% threonine had higher ( $P < 0.05$ ) C18:0 and C24:0 than those fed diet with 0.56% threonine, whereas dietary threonine levels did not affect ( $P > 0.05$ ) the C18:0, and C24:0 in fatty-type ducks.

## DISCUSSION

To investigate the differences of lipid deposition between fatty-type and lean-type ducks fed diet with different threonine levels, we determine the fatty acids contents in fatty-type and lean-type Pekin ducks after feeding diets with diverse levels of threonine for 21 D from 15 to 35 D of ages. In the present study, the reduced abdominal fat percentage was observed in Pekin ducks fed diets with 0.41 or 0.71% threonine, which is in agreement with previous studies (Jiang et al., 2017; Jiang et al., 2019a). In addition, it was also observed that the fatty-type ducks had higher sebum percentage compared with lean-type ducks, and the dietary threonine levels affected the sebum percentage in lean ducks, but not in fatty ducks. A previous study in our laboratory reported that fatty-type ducks had higher percentage in abdominal fat and sebum and lower percentage in breast muscle and thigh muscle (Zheng et al., 2014). Obviously, the mechanism of lipid deposition in sebum and abdominal fat is different between fatty-type and lean-type ducks, and the response to threonine levels also is different, which needs further studies to prove it.

Fatty-type ducks showed higher feed intake and feed to gain ratio than lean-type ducks, and the BW and weight gain were similar with lean-type duck throughout the experiment (15 to 35 D of age), but fatty-type ducks deposited more fat and less muscle mass than lean-type ducks, which indicated that the energy value is greater for BW gain in fatty-type ducks than lean-type ducks. Previous studies demonstrated that dietary threonine deficiency reduced growth performance of Pekin ducks (Zhang et al., 2014; Jiang et al., 2016; Zhang et al., 2016; Jiang et al., 2017; Jiang et al., 2018; Jiang et al., 2019a). In the present study, dietary threonine deficiency reduced the BW, feed intake, weight gain, and feed to gain ratio in lean-type ducks but not in fatty-type ducks. This might be because fatty-type ducks had higher feed intake, which meet the need for metabolism and growth in fatty-type ducks, or fatty-

type ducks had lower threonine requirement from 14 to 35 D of age.

Liver is a major site of *de novo* synthesis of fatty acid for poultry (Leveille et al., 1975; Patel et al., 1975; Pullen et al., 1990). In our laboratory, it was observed that hepatic contents of total lipid, TCHO, and TG were increased by dietary threonine deficiency (Jiang et al., 2017; Jiang et al., 2019a). In present study, the dietary threonine deficiency also increased the hepatic contents of total lipid, TCHO, and TG in lean-type ducks but not in fatty-type ducks. Meanwhile, dietary threonine deficiency increased plasma HDLC and TCHO concentrations. A previous study from our laboratory showed that dietary threonine deficiency increased the gene expression related to fatty acid and triglyceride synthesis, and reduced the gene expression involving in fatty acid oxidation and triglyceride degradation (Jiang et al., 2019b), although it did not detect the changes of genes related to lipid transportation to extrahepatic tissue, we speculated that threonine deficiency influences the transfer of lipid from liver to blood by decreasing the protein synthesis in lean-type ducks, hence the lipid was deposited in the liver. Whereas, threonine-deficient diet reduced hepatic total lipids in rat (Ross-Inta et al., 2009). Interestingly, the higher hepatic contents of total lipid, TCHO, and TG were observed in lean-type ducks compared with fatty-type ducks. These results were in line with plasma LDLC content, which was higher in fatty ducks than lean-type ducks. LDLC is converted from VLDL, which transport endogenous triglycerides, phospholipids, cholesterol, and cholesteryl esters to extrahepatic tissue, whereas these cholesterols unabsorbed by extrahepatic tissue is returned to the liver in the form of LDLC and excreted by bile metabolism. In the present study, it was observed that plasma concentrations of LDLC and TCHO in fatty-type ducks were higher than that in lean-type ducks, whereas hepatic TCHO content in fatty-type ducks is lower than that in lean-type ducks, which indicated that the liver of fatty-type ducks might have lower ability to absorb LDLC, but higher excretion of TCHO. These might be because of the high expression of fatty acid-binding protein 1 (FABP1) in lean ducks (Zheng et al., 2014). FABP1 facilitates the transfer of fatty acid between extracellular and intracellular membranes (Weisiger, 2002) and plays an important role in cholesterol uptake in hepatocytes (Huang et al., 2015). In addition, protein expressions of *de novo* fatty acid synthesis were higher in fatty ducks compared with lean ducks (Zheng et al., 2014). Therefore, it was speculated that fatty acid synthesized of liver in fatty-type ducks was transported to adipose tissue by blood circulation and more lipid was taken up by liver in lean-type ducks to supply energy for protein synthesis in lean-type ducks. But, this hypothesis need to be demonstrated by further studies.

Generally, eating quality traits could be improved by increase of IMF content and monosaturated fatty acid percentage and decrease of polysaturated fatty acid (Cameron and Enser, 1991). PUFA is susceptible to produce undesired volatile compounds during cooking

(Larick et al., 1992), which results in off flavors. In addition, pork eating quality is positively correlated with MUFA contents (Rhee et al., 1990). In present study, it was found that lean duck had higher PUFA content, and no difference in MUFA content in breast muscle, compared with fatty-type ducks. In addition, threonine supplementation increased breast muscle PUFA content in lean duck, but not MUFA content in fatty ducks. Obviously, genetic selection toward lean muscle production increased PUFA deposition, which might reduce the eating quality of lean ducks. In addition, dietary threonine deficiency reduced PUFA content in breast muscle of ducks, and dietary high levels threonine had a trend to reduce PUFA content in breast muscle. Interestingly, it was found hepatic MUFA content is higher in lean-type duck than fatty-type ducks, and dietary threonine deficiency increased MUFA content in lean-type ducks, but not in fatty-type ducks.

N3-and N6-PUFA are obtained through diet. Linoleic acid (18:2n-6) is converted into  $\gamma$ -linolenic acid (18:3n-6) and dihomogamma-linolenic acid (20:3n-6), which further form the key intermediate arachidonic acid (20:4n-6) catalyzed by various enzymes. The N3 fatty acid  $\alpha$ -linolenic acid (18:3n-3) is converted into stearidonic acid (18:4n-3) and eicosatetraenoic acid (20:4n-3), which further converted to DHA (22:6n-3). In the present study, lean-type ducks had higher concentrations of 18:2n-6 and 18:3n-6 fatty acid and lower 20:3n-6 fatty acid in the liver, breast muscle, and plasma compared with fatty-type ducks. In addition, genetic selection toward meat production reduced hepatic 20:4n-6 fatty acid content, whereas it increased C20:4n-6 fatty acids content in breast muscle. Previous studies had demonstrated that C20:4n-6 is positively associated with antithrombotic and antiarrhythmic cardiac effects (Conquer and Holub, 1998) and reduced cardiovascular diseases in rats (McLennan et al., 1996) and in neural and retinal function (Alessandri et al., 1998). In the present study, dietary threonine supplementation increased breast muscle N3-and N6-PUFA contents in fatty ducks. These results indicated that the genetic selection and threonine supplementation would affect eating quality traits of duck meat.

In conclusion, threonine supplementation increase growth performance and breast muscle PUFA of lean ducks, but not in fatty ducks. Genetic selection toward lean meat production reduce abdominal fat and sebum percentage, increase breast muscle percentage, the PUFA contents in breast muscle, which influence eating quality traits of duck meat, but it is benefit to human health. Further studies should be conducted to explore the molecular mechanism of threonine regulation on fatty acids synthesis in lean ducks and reveal the molecular changes of fatty acids synthesis in breast muscle caused by genetic selection for lean meat production.

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