Effects of rapeseed meal on laying performance and egg quality in laying ducks

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ABSTRACT This study was conducted to investigate the effect of different varieties of rapeseed meal (**RSM**) with different concentrations of glucosinolates (**Gls**) and erucic acid (**EA**) on performance and egg quality of laying ducks. A total of 576 twenty eight-wk-old laying ducks were randomly allocated to 4 treatments. Each treatment had 8 replicates of 18 laying ducks raised in 6 adjacent cages with 3 laying ducks per cage. The control diet was corn soybean meal based without RSM. Three varieties of RSM varying in Gls concentrations were supplemented to the base diet at 10% by substituting soybean meal to formulate the three RSM diets. The experiment lasted 12 wk. Diets with 10% RSM decreased average egg weight ($P < 0.01$) and feed intake ($P = 0.07$) compared with the control diet, but there was no significant difference in laying performance among the 3 RSM diets. RSM increased color value ($P < 0.05$) and crude protein (**CP**) content ($P < 0.05$) of yolk compared with the control diet, but had no significant effects on the other egg quality indexes including eggshell strength, albumen height, Haugh unit, and the composition ratio of eggshell, albumen and yolk. RSM decreased total monounsaturated fatty acids (**MUFA**) ($P < 0.01$) and increased total polyunsaturated fatty acids (**PUFA**) ($P < 0.01$) of yolk, but total saturated fatty acids (**SFA**) proportions and UFA/SFA ratio of egg yolk were not significantly affected by RSM. RSM increased deposition of trimethylamine (**TMA**) and 5-vinyl-1,3-oxazolidine-2-thione (**5-VOT**) in yolk ($P < 0.01$); moreover, the high Gls RSM increased deposition of TMA ($P < 0.01$) and 5-VOT in yolk ($P < 0.01$) compared with the RSM varieties low in Gls. These results suggested that dietary inclusion of 10% RSM decreased egg weight of laying ducks, and affected yolk quality especially yolk color, fatty acid profile, CP, TMA, and 5-VOT content of yolk. Moreover, RSM with higher Gls concentration resulted in higher deposition of TMA and 5-VOT in egg yolk.

Key words: rapeseed meal, laying duck, laying performance, egg quality

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INTRODUCTION

Rapeseed meal (**RSM**) is used widely in feed industry as an economic plant protein raw material. However, RSM inclusion rate is usually limited in diets due to its high content of antinutritional factors including glucosinolates (**Gls**), erucic acid, sinapine, and tannin (Bell, 1993). Gls can be degraded to isothiocyanate (**ITC**) and oxazolidinethione (**OZT**) which may have toxic effects on thyroid, liver, kidney, and other organs of animals (Derycke et al., 1999; Mabon et al., 2000; Tripathi and Mishra, 2007), resulting in a decrease of performance, meat or egg quality in animals (Taraz et al., 2006; Zhu et al., 2018a). Two metabolites of Gls, 5-vinyl-1,3-oxazolidine-2-thione (**5-VOT**) and thiocyanate ion (**SCN^-**), were recognized as poisonous substances according to FDA Poisonous Plant Database. According to the latest national hygienical standard for feeds in China, ITC content in RSM and poultry feed cannot be higher than 4,000 mg/kg and 500 mg/kg respectively, and OZT content in RSM and egg-laying poultry feed cannot be higher than 2,500 mg/kg and 500 mg/kg respectively (State Administration for Quality Supervision and Inspection and Quarantine of the People's Republic of China and the Standardization Administration of China, 2017). The previous studies in our lab showed that layer diets with high levels of RSM led to residue of 5-VOT and SCN^- in eggs, which may threaten food safety and health for human (Zhu et al., 2018a), and high levels of RSM with higher Gls in layer diets were found to decrease egg weight, nutrient digestibility, intestinal absorptive area, and egg internal quality (Zhu et al., 2019).

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Sinapine is another antinutritional factor in RSM, which is hydrolyzed to choline by microbial fermentation in ceca and then catabolized to trimethylamine (**TMA**) (Qiao and Classen, 2003). Laying hen diets with high levels of RSM resulted in TMA metabolism load, thus inducing fishy odor eggs (Ward et al., 2009; Long et al., 2017). TMA levels in duck eggs were significantly higher than those in chicken eggs under normal dietary conditions, and the excessive TMA deposition in duck eggs was one of the main factors causing the fishy odor in duck eggs (Li et al., 2018). However, there was very little information about TMA deposition in duck eggs, and there was some doubt whether the inherent fishy smell of duck egg was induced by RSM.

China is the largest producer of duck eggs in the world. More than 4 million tons duck eggs are produced in China each year. There were some studies to investigate the effect of grain by-products on performance and egg quality of laying ducks (Ruan et al., 2015; Ruan et al., 2018a), but little is known about the effects of RSM derived from different varieties of rapeseeds on laying performance and egg quality of laying ducks. The mechanism of RSM influencing laying performance and egg quality of laying ducks has not been investigated so far. Therefore, the objective of this study was to investigate the effects of different varieties of RSM with different Gls contents on laying performance and egg quality of laying ducks.

MATERIALS AND METHODS

Rapeseed and Rapeseed Meal

Three varieties of rapeseed were collected from rapeseed planting bases: the high erucic acid rapeseed H405 (**H405**), and two double-low rapeseeds Deyou No.6 (**DY6**) and Deyou No.5 (**DY5**). RSM was produced from the rapeseed after oil extraction by the hot expeller method. The content of Gls and erucic acid in rapeseed, and total Gls, sinapine, ITC, and OZT in RSM (Table 1) were analyzed (Oil Crops Research Institute, Chinese Academy of Agricultural Sciences, Wuhan, China) by high-performance liquid chromatography (Agilent 1200, Santa Clara, CA) and gas chromatography (Agilent 7890A, Agilent Technologies Inc., Santa Clara, CA).

Animals, Diets and Management

This study was approved by the Institutional Animal Care and Use Committee of Sichuan Agricultural University (Chengdu, Sichuan, China). A total of 576 laying Shaoxing ducks (28-wk-old) with similar BW and laying performance were selected and randomly allocated to 4 treatments with 8 replicates per treatment. Each replicate had 18 laying ducks in 6 adjacent cages with 3 laying ducks per cage. Diets with or without 10% RSM (Table 2) were formulated to meet the requirements of laying ducks (Ruan et al., 2015). Before the trial, all ducks were fed the same diet without RSM for 8 wk. The diets were pelleted and sampled for nutrients and fatty acid profile analysis (Table 3). Diets and water were

Table 1. Chemical and antinutritional factor analysis of rapeseed and rapeseed meal[1].

Item	DY6	DY5	H405
Rapeseed			
Glucosinolates (μmol/g) [2]	40.07	51.08	104.89
Erucic acid (%) [3]	8.69	5.55	54.9
Rapeseed meal			
Sinapine (mg/kg)	9,390	9,360	8,480
Glucosinolates (μmol/g)	8.92	7.67	22.65
Isothiocyanate (mg/kg)	557	400	1756
Oxazolidine thione (mg/kg)	170	150	1410
Crude protein (%)	37.67	38.07	45.2
Crude fat (%)	6.54	7.7	7.62
Crude fiber (%)	16.72	18.38	13.52
Ash (%)	8.76	8.35	6.82

[1]DY6 = Deyou No.6, DY5 = Deyou No.5, H405 = High erucic acid H405.

[2]Glucosinolates content of rapeseed was calculated based on rapeseed meal.

[3]Erucic acid content of rapeseed was relative to total fatty acids content.

Table 2. Composition and nutrient levels of the four diets (as-fed basis)[1].

Item	Control	DY6	DY5	H405
Ingredients (%)				
Corn	55.59	55.44	55.44	55.44
Soybean meal, 43%	25.91	18.96	18.96	18.96
Rapeseed meal	0	10.00	10.00	10.00
Calcium carbonate	8.36	8.25	8.25	8.25
Wheat flour	5.00	5.00	5.00	5.00
Soybean oil	0	0.30	0.30	0.30
Wheat bran	3.19	0.00	0.00	0.00
Dicalcium phosphate	0.98	1.03	1.03	1.03
Sodium chloride	0.33	0.33	0.33	0.33
DL-Methionine	0.17	0.13	0.13	0.13
L-Lysine•HCl	0	0.08	0.08	0.08
Choline chloride	0.15	0.15	0.15	0.15
L-tryptophan	0.01	0.02	0.02	0.02
Phytase (10,000 IU/g)[2]	0.01	0.01	0.01	0.01
Premix[3]	0.30	0.30	0.30	0.30
Total	100.00	100.00	100.00	100.00
Nutrient levels[4]				
Metabolizable energy (kcal/kg)	2700	2700	2700	2700
Crude protein, %	17.78	17.75	17.56	18.42
Crude fat, %	2.19	2.98	2.81	2.75
Crude fiber, %	2.44	3.53	3.3	3.41
Ash, %	12.19	11.54	11.76	11.26
Ca, %	3.60	3.60	3.62	3.59
Total P, %	0.53	0.53	0.54	0.52
Available P, %	0.35	0.35	0.35	0.35
Total Lys, %	0.89	0.88	0.88	0.91
Total Met, %	0.40	0.40	0.40	0.41
Total Met+Cys, %	0.73	0.73	0.75	0.76
Total Trp, %	0.21	0.21	0.21	0.22
Total Thr, %	0.69	0.69	0.69	0.71

[1]DY5 = Deyou No.5, DY6 = Deyou No.6, H405 = High erucic acid H405.

[2]Supplemented with phytase 1,000 U/kg in diet to provide 0.07% available phosphorus.

[3]The premix provided per kilogram diet: vitamin A, 10,000 IU; vitamin D, 3,000 IU; vitamin E, 40 mg; vitamin K3, 2.5 mg; vitamin B1, 4.0 mg; vitamin B2, 6.0 mg; vitamin B6, 6.0 mg; vitamin B12, 0.02 mg; nicotinic acid, 15 mg; D-pantothenic acid, 20 mg; folic acid, 1.0 mg; biotin, 0.2 mg; Fe, 75 mg; Cu, 10 mg; Mn, 125 mg; Zn, 115 mg; I, 0.25 mg; Se, 0.25 mg; Co, 0.3 mg.

[4]The metabolizable energy and available phosphorus were calculated values, while the other nutrient levels were measured values.

Table 3. Fatty acid content and profile of the four diets (as-fed basis).

Item	Diets[1] Control	DY6	DY5	H405
Crude fat (% Diet)	2.19	2.98	2.81	2.75
Total fatty acids (% Diet)	1.70	2.39	2.50	2.40
Fatty acid profile (%)[2]				
Myristic acid (C14:0)	0.19	0.15	0.15	0.16
Pentadecanoic acid (C15:0)	0.06	0.05	0.05	0.05
Palmitic acid (C16:0)	17.29	13.66	13.31	13.06
Heptadecanoic acid (C17:0)	0.11	0.09	0.09	0.08
Stearic acid (C18:0)	2.50	2.32	2.32	2.04
Eicosanoic acid (C20:0)	0.35	0.37	0.39	0.41
Behenic acid (C22:0)	0.22	0.22	0.24	0.37
TricosanoiCacid (C23:0)	0.05	0.00	0.04	0.00
Eicosatetraenoic acid(C24:0)	0.24	0.19	0.22	0.21
Palmitoleic acid (C16:1)	0.13	0.23	0.23	0.20
Oleic acid (C18:1 c9)	22.39	28.21	29.43	20.69
Gadoleic acid (C20: c1)	0.37	0.54	0.65	0.93
Erucic Acid(C22:1n9)	0	0.86	0.84	9.51
Linoleic acid (C18:2n-6)	53.41	49.00	47.89	46.89
α-Linolenic acid (C18:3n-3)	2.69	4.07	4.08	4.66
Eicosadienoic acid (C20:2n-6)	0	0.05	0.06	0.12
Eicosatrienoic acid (C20:3n3)	0	0	0	0.43
Docosadienoic acid (C22:2)	0	0	0	0.20
Total fatty acid profile	100	100	100	100
Saturated fatty acids (SFA)	21.01	17.05	16.82	16.38
Monounsaturated fatty acids (MUFA)	22.90	29.83	31.15	31.32
Polyunsaturated fatty acids (PUFA)	56.10	53.12	52.03	52.30
Unsaturated fatty acids (UFA)	78.99	82.95	83.18	83.62
UFA/SFA	3.76	4.87	4.95	5.11

[1]DY5 = Deyou No.5, DY6 = Deyou No.6, H405 = High erucic acid H405.
[2]Expressed as the relative proportion of the total fatty acids content.

provided for ad libitum consumption, and 16 h of light was provided throughout the trial.

Laying Performance

Feed intake, egg production, and egg weight on a replicate basis were recorded daily to calculate laying performance indexes including average daily feed intake (**ADFI**), egg production, average egg weight, egg mass, and feed conversion ratio (**FCR**) each week.

Egg Quality

Four eggs from each replicate were collected at the 12th week of the trial period for egg quality analysis. Eggshell strength and thickness were evaluated by eggshell force gauge model II (Robotmation Co., Ltd., Tokyo, Japan) and eggshell thickness gauge (Robotmation Co., Ltd.) respectively. Egg weight, yolk color, and Haugh unit were evaluated by an egg multitester (EMT-7300; Robotmation, Co., Ltd.). Yolk, albumen, and eggshell from each egg were separated and weighted to calculate the composition ratio of eggs. Yolks of the eggs from each replicate were mixed and stored at −20°C for yolk quality analysis.

Yolk Composition and Fatty Acid Profile Analysis

Yolks were separated and dried in a freezer dryer, then ground by a pulverizer for yolk composition and fatty acid profile analysis. Dry matter, crude protein and crude fat content of yolks were analyzed as described by AOAC Int (2012). Fatty acid profile of the yolks collected in the 12th week of the trial period was analyzed by gas chromatography with a flame ionization detector according to the method as described (Ruan et al., 2018a). Fatty acid compositions were expressed as percentages of total fatty acids.

5-VOT and SCN^- Analysis

Yolk powder was used for 5-VOT and SCN− analysis. 5-VOT and SCN^- content of the yolk was quantified by the high-performance liquid chromatography (**HPLC**) and high-performance ion chromatography (**HPIC**) method, respectively (Zhu et al., 2018a).

TMA Analysis

TMA content in fresh egg yolk was analyzed by head-space gas chromatography as proposed with minor modification (Long et al., 2017). Fresh yolk (~1 g) and 5 mL 1.17 g/mL KOH were placed in a vial. The vial capped with a Teflon-lined septum was kept at 30°C for 20 min by ultrasonic treatment, then kept at 40°C for 30 min. The head-space gas (250 μL) was subjected to gas chromatography analysis with a gas chromatographer (Agilent Technologies, Palo Alto, CA) equipped with a flame ionization detector. The chromatographer was also fitted with a 30 m × 0.32 mm id fused silica capillary column coated with 0.5-μm films of DBWax (Agilent J&W Scientific, Folsom, CA). Operating conditions were set as 40°C for 3 min, then increased to 220°C (30 °C/min), and kept at 220°C for 1 min; injector 220°C, detector 220°C, split mode (split ratio 2:1); hydrogen (carrier gas) 40 mL/min.

Statistical Analysis

Replicate (n = 8) was taken as the experimental unit. Except where noted otherwise, 4 sampled eggs per replicate were used. The effect of RSM in the diets was analyzed by one-way ANOVA using GLM procedure of SAS 9.1 (SAS Institute Inc., Cary, NC, 2004). Differences between means were assessed by Duncan's multiple range tests at 5% probability level. Results were expressed as means and standard error of the mean (**SEM**).

RESULTS

Laying Performance

The effect of RSM on performance of laying ducks was presented in Table 4. The average egg weight of DY6, DY5, and H405 groups was decreased ($P < 0.01$), respectively by 2.24, 2.38, and 2.8% compared with that of the control diet, but only a numerical depression of ADFI ($P = 0.07$), and no effect on egg production, egg mass, or

Table 4. Effect of RSM on the laying performance of laying ducks[1].

Item	Treatment				SEM	P-value
	Control	DY6	DY5	H405		
Egg-laying rate (%)	84.5	81.1	79.5	82.0	2.08	0.61
ADFI (g/d)	152.3	146.1	148.3	148.8	1.52	0.07
Egg weight (g)	71.4[a]	69.8[b]	69.7[b]	69.4[b]	0.26	0.0003
Egg mass[2] (g/d)	60.4	56.6	55.4	56.9	1.46	0.29
FCR	2.53	2.60	2.68	2.62	0.06	0.65

DY5 = Deyou No.5, DY6 = Deyou No.6, H405 = High erucic acid H405.

Abbreviations: ADFI, average daily feed intake; FCR, feed conversion ratio.

[ab]Different superscript alphabets in the same row means significantly different ($P < 0.05$).

[1]Data are expressed as mean and SEM (n = 8).

[2]Egg mass = (egg production × egg weight)/100.

FCR was observed in the 3 RSM diets ($P > 0.05$). There was no significant difference in laying performance among the three RSM diets ($P > 0.05$).

Egg Quality

The effect of RSM on egg quality of laying ducks was presented in Table 5. Dietary 10% RSM of H405 variety decreased eggshell thickness ($P < 0.05$) compared with DY5 group, and RSM of H405 variety increased yolk color ($P < 0.05$) compared with control group. However, RSM had no significant effects on the other egg quality indexes including eggshell strength, albumen height, Haugh unit, and composition ratio of eggshell, albumen, and yolk compared with the control group.

Yolk Composition and Fatty Acid Profile

Yolk composition and fatty acid profile were presented in Table 6. Diet with 10% RSM increased crude protein content ($P < 0.01$) and total fatty acids content ($P < 0.05$) of yolk, and a numerical increase in yolk dry matter ($P = 0.077$) was observed in all three RSM diets. Although total polyunsaturated fatty acids (**PUFA**) proportions were increased ($P < 0.01$), total saturated fatty acids (**SFA**) proportions, total unsaturated fatty acids (**UFA**), and UFA/SFA ratio of egg yolk were not significantly affected by RSM. Dietary 10% RSM of DY6 or H405 variety significantly decreased total monounsaturated fatty acids (**MUFA**) proportions of yolk when compared with the control group ($P < 0.01$) and DY5 group ($P < 0.05$). RSM had no significant effects on proportions of most of the SFA except for stearic acid, which was increased by DY6 or H405 RSM significantly compared with control group ($P < 0.01$) and DY5 group ($P < 0.01$). RSM decreased proportions of myristoleic acid ($P < 0.01$), palmitoleic acid ($P < 0.01$), and oleic acid ($P < 0.05$) of yolk, but increased proportions of linoleic acid ($P < 0.01$), α-linolenic acid ($P < 0.01$), arachidonic acid ($P < 0.05$), and docosahexaenoic acid ($P < 0.01$) of yolk. In addition, RSM H405 increased α-linolenic acid proportion ($P < 0.01$) while decreased arachidonic acid proportion ($P < 0.01$) when compared with the DY6 and DY5 group, however, RSM DY5 decreased stearic acid proportion ($P < 0.01$) and γ-linolenic acid proportion ($P < 0.05$) when compared with the DY6 and H405 group, which means that yolk fatty acid profile was affected by RSM variety.

Table 5. Effect of RSM on the egg quality of 40-wk-old laying ducks[1].

Item	Treatment				SEM	P-value
	Control	DY6	DY5	H405		
Egg weight(g)	74.76	74.43	73.97	72.54	1.09	0.19
Yolk weight (g)	25.58	25.17	25.25	24.70	0.53	0.44
Albumen weight (g)	41.92	41.66	41.35	40.69	0.71	0.35
Eggshell weight (g)	7.27	7.60	7.36	7.16	0.17	0.06
Yolk rate (%)	34.20	33.79	34.15	34.05	0.50	0.84
Albumen rate (%)	56.07	56.00	55.88	56.09	0.49	0.97
Eggshell rate (%)	9.73	10.21	9.97	9.86	0.20	0.11
Eggshell strength (kg/cm^2)	4.89	4.90	4.82	4.84	0.19	0.98
Eggshell thickness (mm)	0.390[a]	0.403[ab]	0.414[b]	0.397[a]	0.008	0.02
Haugh unit	85.73	85.76	82.83	81.68	2.73	0.33
Yolk color	2.24[a]	2.35[ab]	2.68[ab]	2.88[b]	0.27	0.05

DY5 = Deyou No.5, DY6 = Deyou No.6, H405 = High erucic acid H405.

[ab]Different superscript alphabets in the same row means significantly different ($P < 0.05$).

[1]Data are expressed as mean and SEM (n = 32).

5-VOT, SCN^-, and TMA Deposition in Yolk

5-VOT, SCN^-, and TMA deposition in egg yolk are presented in Table 7. 5-VOT in yolk was not be detected in the control group, but 10% DY6, DY5, and H405 RSM inclusion resulted in deposition of 5-VOT at 283.4 μg/kg, 573.5 μg/kg, and 1,276.5 μg/kg respectively in egg yolk ($P < 0.01$). SCN^- in yolk could not be detected in any treatment. The content of TMA was 3.9 mg/kg in egg yolk of the control group, and all RSM varieties increased TMA content in egg yolk ($P < 0.01$), which was the highest for H405, the lowest for DY6, and intermediate for DY5.

DISCUSSION

Effect of RSM on Performance and Egg Quality of Laying Ducks

RSM inclusion in animal diets is limited because of presence of antinutritional factors which may have negative effects on feed intake, performance and product quality. It was recommended that inclusion of high Gls RSM (>30 μmol/g) and low Gls RSM should be restricted to a maximum of 5 and 10%, respectively in hen diets (Mawson et al., 1994a). Gls are precursors of compounds that exert goitrogenic activity in birds and induce metabolic disorders, and they might also cause abnormalities in internal organs such as liver, kidney, and thyroid gland (Mawson et al., 1994b). When substituting 10% canola meal for soybean meal on an iso-caloric basis in laying hen diets, egg production declined and eggs were smaller (Summers et al., 1985). Diets containing 20% RSM decreased egg production and feed

Table 6. Effect of RSM on the yolk composition and fatty acid profile of duck egg yolk[1].

Item	Treatment				SEM	*P*-value
	Control	DY6	DY5	H405		
Yolk composition						
Dry matter (% fresh yolk)	53.09	54.14	54.09	53.59	0.44	0.077
Crude fat (% freeze dried yolk)	57.09	57.24	58.17	58.03	0.59	0.185
Crude protein (%freeze dried yolk)	29.94[a]	32.09[b]	31.77[b]	31.92[b]	0.33	<0.001
Fatty acids (%freeze dried yolk)	54.76[a]	56.74[b]	56.56[b]	56.20[b]	0.70	0.035
Yolk fatty acid profile[2]						
Lauric acid (C12:0)	0.01	0.01	0.01	0.01	0.01	0.384
Myristic acid (C14:0)	0.44	0.43	0.43	0.45	0.02	0.366
Pentadecanoic acid (C15:0)	0.03	0.03	0.03	0.03	0.00	0.053
Palmitic acid (C16:0)	25.45	25.18	24.76	25.04	0.27	0.111
Heptadecanoic acid (C17:0)	0.09	0.10	0.10	0.10	0.01	0.136
Stearic acid (C18:0)	6.43[a]	6.76[b]	6.43[a]	6.98[b]	0.14	0.001
Eicosanoic acid (C22:0)	0.07	0.07	0.07	0.06	0.02	0.879
Myristoleic acid (C14:1)	0.04[b]	0.03[a]	0.03[a]	0.03[a]	0.00	0.005
Palmitoleic acid (C16:1)	2.40[b]	1.91[a]	2.04[a]	1.92[a]	0.08	0.000
Monoheptadecenoin (C17:1)	0.05	0.05	0.05	0.06	0.01	0.653
Oleic acid (C18:1 c9)	52.19[b]	50.68[a]	51.71[ab]	50.59[a]	0.53	0.012
Gadoleic acid (C20: c1)	0.18	0.19	0.19	0.27	0.10	0.711
Linoleic acid (C18:2n-6)	7.41[a]	8.57[b]	8.13[b]	8.42[b]	0.25	0.000
γ-Linolenic acid (C18:3n-6)	0.17[a]	0.19[b]	0.17[a]	0.19[b]	0.01	0.015
α-Linolenic acid (C18:3n-3)	0.53[a]	0.78[b]	0.79[b]	1.09[c]	0.12	0.001
Eicosadienoic acid (C20:2n-6)	0.20[a]	0.23[b]	0.21[ab]	0.22[b]	0.01	0.010
Dihomo-γ-linolenic acid (C20:3n-6)	0.39	0.42	0.42	0.42	0.02	0.323
Arachidonic acid (C20:4n-6)	3.69[ab]	3.93[c]	3.89[bc]	3.66[a]	0.10	0.020
Docosahexaenoic acid (C22:6n-3)	0.29[a]	0.46[b]	0.45[b]	0.46[b]	0.01	0.000
Saturated fatty acids (SFA)	32.50	32.57	31.82	32.66	0.33	0.063
Monounsaturated fatty acids (MUFA)	54.84[c]	52.85[a]	54.03[bc]	52.88[a]	0.50	0.001
Polyunsaturated fatty acids (PUFA)	12.67[a]	14.58[b]	14.06[b]	14.46[b]	0.33	0.000
Unsaturated fatty acids (UFA)	67.51	67.43	68.09	67.34	0.36	0.175
UFA/SFA	2.08	2.07	2.14	2.06	0.03	0.068

[a-c]Different superscript alphabets in the same row means significantly different ($P < 0.05$).
[1]Data are expressed as mean and SEM (n = 8). DY5 = Deyou No.5, DY6 = Deyou No.6, H405 = High erucic acid H405.
[2]Fatty acid profile was expressed as percentages of total fatty acids in freeze dried yolks.

consumption compared with corn soybean meal control diet (Leslie et al., 1973). However, egg production performance and egg quality including shell thickness, albumen height, and Haugh unit were not significantly affected by the presence of RSM in the diet, but yolk color was increased (Blair et al., 1975). Previous studies by our team found that egg production and egg mass linearly decreased and FCR increased linearly with the increased content of RSM (Zhu et al., 2018b). Zhu et al. (2019) reported that exposure of laying hens to RSM with higher Gls led to lower egg weight, poor digestibility, and poor egg quality compared to RSM with lower Gls. However, there was little research about the effect of RSM on performance and egg quality of laying ducks. Our study demonstrated that laying duck diets with 10% RSM decreased average egg weight and increased yolk color value compared with the corn-soybean control diet, but had no significant effects on other performance measurements (FI tended to be decreased) and egg quality indexes including eggshell strength, albumen height, Haugh unit, and ratio of eggshell, albumen, and yolk. There were some differences in nutritional and antinutritional composition among the 3 varieties RSM, that resulted in different crude protein, Gls, ITC and OZT content in the diets with 10% RSM. In this study, laying performance of duck was not affected by RSM variety, but eggshell thickness was decreased by the high Gls RSM H405 compared with the low Gls RSM DY5.

Effect of RSM on Yolk Quality of Laying Ducks

Duck eggs were consumed mainly as further processed products such as preserved eggs, salted eggs,

Table 7. Effect of rapeseed cake on egg yolk quality in laying ducks[1].

Item[2]	Treatment				SEM	*P*-value
	Control	DY6	DY5	H405		
5-VOT (μg/kg)	ND	283.4[a]	573.5[b]	1276.5[c]	37.45	<0.001
SCN^- (mg/kg)	ND	ND	ND	ND	-	-
TMA (μg/g)	3.90[a]	6.57[b]	8.45[c]	9.91[d]	0.58	<0.001

[a-d]Different superscript alphabets in the same row means significantly different ($P < 0.05$).
[1]Data are expressed as mean and SEM (n = 8). DY5 = Deyou No.5, DY6 = Deyou No.6, H405 = High erucic acid H405, and ND = not detected.
[2]5-VOT = 5-vinyl-1,3-oxazolidine-2-thione, SCN^- = thiocyanate ion, TMA = trimethylamine. 5-VOT, SCN^- content was analyzed with freeze dried yolk samples, and TMA content was analyzed with fresh egg yolk samples.

mayonnaise, and mooncake filling, thus yolk quality of duck egg is very important because it may affect nutrition value, flavor, texture, and sale price of yolks. Egg yolks from ducks fed diets supplemented with sorghum, rapeseed meal or DDGS were more sensitive to lipid and protein oxidation during salting, reducing quality of salted yolks (Ruan et al., 2018b). Our study indicated that diets supplemented with 10% RSM increased protein and total fatty acids content of yolk but had no significant effects on dry matter and crude fat content of yolk. Lipids are important functional and nutritional components of yolk. Adding canola oil in laying hen diets restored the level of egg yolk SFA to UFA, and led to a higher level of C18:3 n-3 and CLA in the egg (Aydin, 2007). Increasing amounts of rice bran in laying duck diet linearly increased egg yolk concentrations of key fatty acid like C18:2 n-6 and C18:3 n-3 (Ruan et al., 2015). Laying duck diets with DDGS linearly increased egg yolk concentrations of key fatty acid like C18:2n-6 and C18:3n-3 (Ruan et al., 2018a). Laying ducks diets containing 8% RSM reduced antioxidant capacity and increased egg yolk concentrations of PUFA (Ruan et al., 2018b). In this study, laying duck diets containing 10% RSM decreased yolk MUFA and increased PUFA, but SFA proportions and UFA/SFA ratio of egg yolks were not significantly affected by RSM. Yolk fatty acid profile may be affected by dietary fatty acid composition, digestion, absorption, and nutritional metabolism. Thus, lipids content and fatty acid composition of RSM may affect yolk fatty acid profile. In this study, crude fat and total fatty acids content in the three RSM diets were higher than those in the control diet. Moreover, SFA proportions in the three RSM diets were lower than those in the control diet, and UFA proportions were higher than those in the control diet, thus UFA/SFA ratio of the 3 RSM diets was higher than that of the control diet. Although the crude fat and total fatty acid content were almost the same among the 3 RSM diets, the fatty acid profile especially oleic acid, linoleic acid, and erucic acid proportions was quite different. In addition, the ANFs of RSM such as Gls, erucic acid might affect the function of internal organs such as liver, kidney, and thyroid gland, thus might change the metabolism and deposition of fatty acids. The erucic acid proportions of RSM H405 was higher than those of RSM DY6 and DY5, leading to a diet with a higher erucic acid proportion. However, erucic acid was not detected in yolks from all treatments because the erucic acid content in all diets was still low.

Gls is the main antinutritional factor in RSM, which can be hydrolyzed to some degradation products with detrimental effects (Mawson et al., 1994a). The most important of these by-products are 5-VOT and SCN^- because of their toxic effects on liver, kidney, and thyroid gland, which could cause metabolic disturbances (Fenwick et al., 1983). As an economic raw material, RSM is used in animal diets, thus 5-VOT and SCN^- could be deposited in meat, eggs or milk, which might threaten food safety and human health. Laying hen diets containing RSM resulted in deposition of 5-VOT and SCN^- in hen eggs, moreover, both 5-VOT and SCN^- levels of eggs were correlated with RSM intake and RSM feeding time (Zhu et al., 2018a). Although there were some studies about 5-VOT and SCN^- deposition in organs and chick eggs (Mabon et al., 2000; Zhu et al., 2018a), there was no information about 5-VOT and SCN^- deposition in duck eggs. Our study found that diets with 10% RSM resulted in deposition of 5-VOT in egg yolk, and high Gls RSM H405 increased deposition of 5-VOT in egg yolk compared with low Gls RSM DY6 and DY5. However, deposition of SCN^- in yolk was not be detected in any treatment. Chinese government has issued the standard about the ITC and OZT content in raw material and feed products (GB 13078-2017), and some researchers reported the health threat of 5-VOT and SCN^- to human or animals (Langer et al., 1971; Tripathi and Mishra, 2007), but the related standard about 5-VOT and SCN^- deposition in food was scarce.

As a common feed ingredient, RSM is rich in sinapine, which is an ester of choline and sinapic acid (Pearson et al., 1979). As a TMA precursor, sinapine was hydrolyzed to choline by microbial fermentation in the cecum and then catabolized to TMA (Qiao and Classen, 2003), which was deposited in yolk and induced fish odor syndrome in laying hens (Ward et al., 2009). Laying hen diet containing 14% RSM increased TMA synthesis in the cecum and increased TMA deposition in yolk (Long et al., 2017). It was reported that excessive TMA deposition in duck eggs was one of the main factors causing the fishy odor in duck eggs (Li et al., 2018). It was speculated that the tolerance threshold of TMA levels in chick egg yolks for the human-perceived fishy odor is approximately 4 μg/g (Ward et al., 2009). The results of this study showed that diets with 10% RSM DY6, DY5 or H405 significantly increased TMA content in yolk compared with the control diet. The TMA content of duck egg yolks from control group was 3.90 μg/g, which was lower than the human-perceived fishy odor level. However, the TMA content of duck egg yolks from DY6, DY5, and H405 groups was 6.57 μg/g, 8.45 μg/g, and 9.91 μg/g, respectively, which were much higher than the human-perceived fishy odor level (Ward et al., 2009). Although there was little difference in sinapine content among the 3 RSM diets, the diet supplemented with high Gls RSM (H405) increased deposition of TMA in egg yolk compared with the diets supplemented with low Gls RSM (DY6 &DY5). This might be caused by the microbial fermentation in the cecum and the antinutritional factor Gls. Laying hen diet with RSM inclusion increased bacterial diversity and modified the structure of the microbial community (Long et al., 2017). The chemical composition and antinutritional factors content were different among different RSM and diets, which might change microbial composition and TMA content of the cecum. After being absorbed and transported to liver, TMA was metabolized to the odorless trimethylamine N-oxide (**TMAO**) (Yeung et al., 2007). It was reported that goitrin formed by the action of myrosinase on Gls, inhibited the oxidation of

TMA to TMAO by competing for the active site of flavin-containing monooxygenase 3 (**FMO3**) (Goh et al., 1985), thus the Gls content of the RSM might be another factor affecting the TMA deposition in yolks.

In conclusion, the current study indicated that diets with 10% RSM decreased egg weight of laying ducks, and affected yolk quality especially yolk color, fatty acid profile, CP, TMA, and 5-VOT content of yolk. Moreover, the deposition of TMA and 5-VOT in egg yolks was affected by the Gls concentration of RSM.

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DISCLOSURES

The authors declare that there is no conflict of interest, financial or otherwise.

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