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Original Research Article

# Effects of dietary alfalfa flavonoids extraction on growth performance, organ development and blood biochemical indexes of Yangzhou geese aged from 28 to 70 days



Yinyin Chen<sup>a</sup>, Xiaoxiao Gong<sup>a</sup>, Guodong Li<sup>a</sup>, Miao Lin<sup>a</sup>, Yongjiu Huo<sup>a</sup>, Shengli Li<sup>b</sup>, Guoqi Zhao<sup>a,\*</sup>

<sup>a</sup> College of Animal Science and Technology, Yangzhou University, Yangzhou 225009, China
 <sup>b</sup> College of Animal Science and Technology, China Agricultural University, Beijing 100193, China

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#### ABSTRACT

This experiment was conducted to study the effects of dietary alfalfa flavonoids extraction supplemental level on growth performance, organ development and blood biochemical indexes of Yangzhou geese at the age of 28 to 70 days. Two hundred and forty 21-day-old healthy male geese with similar body weight were randomly distributed into 4 groups with 6 replicates per group and 10 geese per replicate. Geese in the control group were fed a basal diet and the others in the experimental groups (groups 1, 2, and 3) were fed experimental diets supplemented with 150, 300 and 450 mg/kg alfalfa flavonoids extraction (the concentration of it was 81%), respectively. The experiment had 7 days for pre-test and 42 days for formal test. The results showed that the final body weight and average daily intake of group 2 were significantly higher than those of other groups (P < 0.05). The average daily gain of group 2 was significantly higher than that in the control group and group 1 (P < 0.05). There was no significant difference in feed-to-gain ratio between each group (P > 0.05). Pre-slaughter live weight, carcass weight, slaughter rate, semieviscerated weight, semi-eviscerated rate, eviscerated weight, eviscerated rate, leg muscle weight and leg muscle rate had no significant difference between each group (P > 0.05). The breast muscle weight and ratio of each test group were significantly higher than those in the control group (P < 0.05) and the group 2 was the best. The abdominal fat weight and ratio in the group 1 were significantly higher than those in the control group and group 3 (P < 0.05) and the tibia weight in the group 2 was significantly higher than that in the control group and group 1 (P < 0.05); There were no significant differences in heart weight, liver weight and the gland stomach weight among all groups (P > 0.05). Spleen weight in test groups was significantly higher than that in the control group (P < 0.05). The bursa weight and muscular stomach weight in the group 2 were significantly higher than those in the control group and group 1 (P < 0.05). In serum, total cholesterol, triglycerides, low-density lipoprotein and urea nitrogen in the group 2 were significantly lower comparing with those in the control group (P < 0.05). High-density lipoprotein in the group 2 was significantly higher than that in other groups (P < 0.05). There were no significant differences in total serum protein, albumin, globulin and albumin/globulin among all groups (P > 0.05). Alanine aminotransferase and aspartate transaminase (AST) in groups 2 and 3 were higher than those in the group 1 and control group but not obvious (P > 0.05) and alkaline phosphatase (ALP) in groups 1 and 2 was higher than that in the control group and group 3 (P > 0.05). It is concluded that alfalfa flavonoids extraction added in dietary feed improve the growth performance, organ development and blood

\* Corresponding author.

E-mail address: gqizhao@yzu.edu.cn (G. Zhao).

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biochemical indexes of Yangzhou geese. It is concluded that 300 mg/kg supplemental level of the dietary alfalfa flavonoids extraction is optimal in this experiment.

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#### 1. Introduction

Alfalfa (Medicago sativa L.) is a high-yield and perennial legume which is regarded as the "King of Grass" in the world (Yu et al., 2014). It was introduced and cultivated all over the world. With rich nutritional value and rich in active substances, utilization of alfalfa resources has become a good prospective projects in the development of forage resources in China which also aroused a growing concern and attention domestic and abroad (Yu et al., 2014). Flavonoids as one of the biologically active ingredient of alfalfa was shown to significantly promote growth, improve carcass quality and enhance immunity in a certain range of additive on livestock (Xiong et al., 2012; Zhu et al., 2009). Alfalfa meal in the diet of laying hens can improve egg quality and yolk color (Xia et al., 2011). With alfalfa flavonoids adding in the diet of mice, the researchers showed that it can significantly improve the growth performance of male mice and the specific and nonspecific immune function in mice has improved to some extent (Zhu et al., 2008). Research of flavonoids adding in pigs, mice, chickens and other ruminant animals has been illustrated in some related studies (Zhang et al., 2006; Gao et al., 2011; Liu et al., 2004), whereas the impact on the study of Yangzhou geese has not been reported. Yangzhou geese origin from local geese resources using modern genetic breeding method of new breed with fast early growth, excellent adaptability, good tolerance with crude feed and have fresh meat as well as other advantages. This experiment intended to determine how the supplementation of alfalfa flavonoids extraction influences growth performance, organ development and blood biochemical indexes in Yangzhou geese. This may provide a scientific basis for the rational use of alfalfa flavonoids in the application and efficient development of alfalfa resources.

#### 2. Materials and methods

#### 2.1. Materials

Choosing 1-day-old Yangzhou male geese (available from Yangzhou Goosing Agricultural Science and Technology Co. Ltd in Gaoyou, China), breeding until 21-day-old and then all geese were converted to the experiment. Alfalfa flavonoids: using supercritical CO<sub>2</sub> extraction method by high-speed countercurrent chromatography separation and purification and then measure it by UV spectrophotometry, the concentration of alfalfa flavonoids was 81%. Methods of measurement relating to alfalfa flavonoids can be consulted according to the reference (Cong et al., 2006).

#### 2.2. Experimental design and feeding management

The experiment was conducted at geese field of Yangzhou Goosing Agricultural Science and Technology Co., Ltd which belongs to the teaching practice base of Yangzhou University from April to June in 2015. Two hundred and forty 21-day-old healthy male geese with similar body weight were randomly distributed into 4 groups with 6 replicates per group and 10 geese per replicate. Geese in the control group were fed a basal diet and the others in the experimental groups (groups 1, 2, and 3) were fed experimental

diets supplemented with 150, 300 and 450 mg/kg alfalfa flavonoids extraction (81%), respectively. In each experimental group, alfalfa flavonoids were mixed in proportion to the premix and then added in diet with remixing. All geese using methods of indoor online rearing, free feeding with mash diets, enough water and natural light, were treated according to routine immunization program. During 7 days for pre-test, all experimental geese were fed the basal diet at 1 to 2 days, and 1/3 experimental diets with 2/3 basal diets were fed at 3 to 5 days, 2/3 experimental diets with 1/3 basal diets were fed at 6 to 7 days according to suitable proportion. Feeding trial period starts from 28 to 70 days.

Experimental diets were based on corn-soybean raw materials, referring to the US NRC (1994) standards, designing same or similar level of the nutrient on diets, its component and nutrient levels are shown in Table 1.

#### 2.3. Determination of indexes

#### 2.3.1. Production performance indexes

Feed intake and weight gain were recorded weekly from 28 to 70 days post-hatch, and feed was stopped 6 h before weighting; death and cull were recorded at various ages, and the average daily feed intake (ADFI), average daily gain (ADG) and feed-to-gain ratio (F:G) were calculated.

#### 2.3.2. Slaughter performance indexes

Table 1

At the end of experiment (70 days), 2 Yangzhou geese of average BW from each replicate (selecting 12 geese from each group) were chosen, weighted and slaughtered, measuring carcass weight, eviscerated weight, semi-eviscerated weight, breast muscle weight, leg muscle weight, abdominal weight, tibia weight while calculating the indexes of carcass yield, eviscerated rate, semi-eviscerated rate, breast muscle yield, leg muscle yield and abdominal rate; Carcass yield (%) = 100 × carcass weight/live weight; eviscerated rate (%) = 100 × eviscerated weight/live weight; semi-eviscerated rate (%) = 100 × ambilateral breast muscle weight/live weight; breast muscle yield (%) = 100 × ambilateral breast muscle weight/eviscerated weight/live weight; breast muscle yield (%) = 100 × ambilateral breast muscle weight/eviscerated

Composition and nutrient levels of	experimental	diets at the	age of 28	to 70	) days
(air-dry basis).					

Ingredients	Content, %	Nutrient levels <sup>2</sup>	Content, %
Corn	51.50	Metabolic energy, MJ/kg	10.95
Soybean meal	23.00	CP	16.42
Wheat bran	10.00	CF	5.58
Oat grass	8.00	EE	3.07
Limestone	1.50	Ca	0.97
CaHPO <sub>4</sub>	1.00	Р	0.61
Premix <sup>1</sup>	5.00	Met	0.40
Total	100.00	Lys	1.03

 $^1$  The premix provided the following per kg of diets: VA 1, 500 IU, VD<sub>3</sub> 200 IU, VE 12.5 IU, VB<sub>1</sub> 2.4 mg, VB<sub>2</sub> 5.0 mg, VB<sub>6</sub> 2 mg, VB<sub>12</sub> 0.01 mg, nicotinic acid 65 mg, <code>p-pantothenic</code> acid 15 mg, folic acid 0.5 mg, biotin 0.2 mg, choline 15 mg, Fe (as ferrous sulfate) 90 mg, Cu (as copper sulfate) 5 mg, Mn (as manganese sulfate) 95 mg, Zn (as zinc sulfate) 90 mg, I (as potassium iodide) 0.5 mg, Se (as sodium selenite) 0.3 mg.  $^2$  Metabolic energy was a calculated value, while the others were measured values.

weight; leg muscle yield (%) =  $100 \times$  ambilateral leg muscle weight/ eviscerated weight; abdominal fat rate (%) =  $100 \times$  abdominal fat weight/(eviscerated weight + abdominal fat weight).

#### 2.3.3. Organ indexes

Fat around the glandular stomach and gizzard (removing contents) was eradicated; And then heart, liver, spleen and bursa were quickly weighed and calculated viscera indexes; Formula: organ index (%) =  $100 \times \text{fresh organ weight (g)/live weight (g)}$ . References for methods refer to (NY/T823-2004).

#### 2.3.4. Blood biochemical indexes

At the end of experiment (70 days), 2 Yangzhou geese of average BW from each replicate (selecting 12 geese from each group) were chosen to take blood sample 10 mL from wing's vein;  $1,300 \times g$  centrifugal with 15 min prepared serum (stored at -20 °C). Serum samples were tested in Yangzhou Municipal Center for Disease Control (China), measuring indexes including: total serum protein (TP), albumin (ALB), globulin (GLB), albumin:globulin (A:G), alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), urea nitrogen (UN), total cholesterol (TC), triglycerides (TG), high-density lipoprotein (HDL) and low-density lipoprotein (LDL).

#### 2.4. Statistical analysis

The data of the experiment was treated by using Excel for preliminary processing including statistical processing etc and then analyzed by SPSS 21.0 statistical software (one-way ANOVA) through analysis of variance, multiple comparisons with *t*-test, data was shown by mean  $\pm$  standard deviation, *P* < 0.05 was considered as statistically significant criteria.

#### 3. Results and analysis

## 3.1. Effects of alfalfa flavonoids on growth performance of Yangzhou geese aged from 28 to 70 days

Table 2 shows that the final body weight and average daily intake of group 2 were significantly higher than those of the other three groups (P < 0.05). The average daily gain of group 2 was significantly higher than that in the control group and group 1 (P < 0.05). There was no significant difference in F:G between each group (P > 0.05). From the results of this experiment, supplementation of alfalfa flavonoids in diet could improve growth performance of Yangzhou geese, group 2 was the best.

#### Table 2

Effects of dietary alfalfa flavonoids supplement on growth performance of Yangzhou
geese at the age of 28 to 70 days.

	Item	Groups			
_		Control	1	2	3
	IBW, kg	$1.21 \pm 0.04$	$1.22 \pm 0.05$	$1.22 \pm 0.06$	$1.23 \pm 0.06$
	FBW, kg	$3.18 \pm 0.14^{b}$	$3.20 \pm 0.26^{b}$	$3.42 \pm 0.13^{a}$	$3.25 \pm 0.19^{b}$
	ADFI, g/d	245.17 ± 32.71 <sup>b</sup>	$247.25 \pm 23.63^{b}$	$258.80 \pm 33.31^{a}$	$248.94 \pm 26.57^{b}$
	ADG, g/d	$46.90 \pm 4.01^{b}$	$47.14 \pm 3.97^{b}$	$52.42 \pm 3.88^{a}$	$48.10 \pm 3.73^{ab}$
	F:G	$5.22 \pm 0.98$	$5.24 \pm 1.65$	$4.94 \pm 0.62$	$5.18 \pm 0.58$

IBW = initial body weight; FBW = final body weight; ADFI = average daily feed intake; ADG = average daily gain; F:G = feed-to-gain ratio.

<sup>a,b</sup> In the same row, values with different small letter superscripts mean significant difference (P < 0.05), while with the same or no letter superscripts mean no significant difference (P > 0.05).

## 3.2. Effects of alfalfa flavonoids on slaughter performance of Yangzhou geese aged from 28 to 70 days

Table 3 shows that pre-slaughter live weight, carcass weight, slaughter rate, semi-eviscerated weight, semi-eviscerated rate, eviscerated weight, eviscerated rate, leg muscle weight and leg muscle rate had no significant difference between each group (P > 0.05). The breast muscle weight and ratio of each test group were significantly higher than those in the control group (P < 0.05) and the group 2 was the best. The abdominal fat weight and ratio in group 1 were significantly higher than those in the control group and group 3 (P < 0.05) and the tibia weight in the group 2 was significantly higher than that in the control group and group 1 (P < 0.05). From the results of this experiment, supplementation of alfalfa flavonoids in diet could promote most part of the indexes on slaughter performance of Yangzhou geese, group 2 was the best.

## 3.3. Effects of alfalfa flavonoids on organ development of Yangzhou geese aged from 28 to 70 days

Table 4 shows that there were no significant differences in heart weight, liver weight and the gland stomach weight among all groups (P > 0.05). Spleen weight in test group was significantly higher than that in the control group (P < 0.05). The bursa weight and muscular stomach weight in the group 2 were significantly higher than those in the control group and group 1 (P < 0.05). From the results of this experiment, supplementation of alfalfa flavonoids in diet increased the internal organs weight of Yangzhou geese, which indicated that alfalfa flavonoids had a certain role in promoting the growth performance and helping the development of internal organs of Yangzhou geese, group 2 was the best.

## 3.4. Effects of alfalfa flavonoids on blood biochemical indexes of Yangzhou geese aged from 28 to 70 days

Table 5 shows that TC, TG, LDL and UN in the group 2 were significantly lower comparing with those in the control group (P < 0.05). The HDL in the group 2 was significantly higher than that in other groups (P < 0.05). There were no significant differences in TP, ALB, GLB and A:G among all groups (P > 0.05). The ALT and AST in groups 2 and 3 were higher than that in the group 1 and control group but not obvious (P > 0.05) and ALP in groups 1 and 2 was higher than that in the control group and group 3 (P > 0.05). From the results of this experiment, supplementation of alfalfa flavonoids in diet improved the blood biochemical indexes of Yangzhou geese, group 2 was the best.

#### 4. Discussion

Flavonoids belong to the secondary metabolites of plant, which are derived from a class of yellow pigment regarding as the mother of flavonoids (2-phenyl chromone); A number of studies have shown that flavonoids extracted from Alfalfa has mild estrogenic effect, which could improve production performance of livestock and poultry, remove free radicals, increase body's antioxidant and anti-stress ability (Dombos et al., 2006; Xie, 2001). Flavonoids can promote animal growth through acting on the animal's hypothalamic—pituitary—target organ growth axis (Zhou, 2011). Flavonoids extracted from alfalfa were added in the hens' diets, after feeding a certain period, the experimental group compared with the control group showed that alfalfa flavonoids extraction stimulated the hens' intake, improved hens' body weight and daily gain, and on the late stage effects of weight gain was more significant than the pre-stage. Li et al. (2008) study showed that Effects of dietary alfalfa flavonoids supplement on slaughter performance of Yangzhou geese at the age of 28 to 70 days.

Item	Groups			
	Control	1	2	3
Slaughter live weight, kg	3.21 ± 0.36	$3.22 \pm 0.51$	$3.29 \pm 0.40$	$3.23 \pm 0.48$
Slaughtering weight, kg	2.78 ± 0.31	$2.85 \pm 0.47$	$2.78 \pm 0.34$	$2.68 \pm 0.40$
Carcass ratio, %	$0.83 \pm 0.16$	$0.85 \pm 0.17$	0.87 ± 0.15	$0.83 \pm 0.17$
Semi-eviscerated weight, kg	$2.23 \pm 0.48$	$2.24 \pm 0.29$	$2.28 \pm 0.34$	$2.18 \pm 0.40$
Semi-eviscerated ratio, %	$0.70 \pm 0.03$	$0.69 \pm 0.03$	$0.69 \pm 0.03$	$0.68 \pm 0.04$
Eviscerated weight, kg	$2.01 \pm 0.27$	$2.06 \pm 0.43$	$2.02 \pm 0.31$	$2.01 \pm 0.38$
Eviscerated ratio, %	$0.60 \pm 0.03$	$0.61 \pm 0.04$	$0.61 \pm 0.03$	$0.60 \pm 0.04$
Leg muscle weight, g	250.58 ± 9.52	$254.86 \pm 10.54$	$260.11 \pm 11.67$	$259.00 \pm 9.34$
Leg muscle ratio, %	$12.44 \pm 1.08$	$12.50 \pm 1.36$	$13.26 \pm 1.95$	$12.98 \pm 1.32$
Breast muscle weight, g	$183.77 \pm 12.54^{b}$	$222.16 \pm 15.83^{a}$	$235.16 \pm 19.25^{a}$	$223.51 \pm 12.71^{a}$
Breast muscle ratio, %	$9.38 \pm 0.14^{b}$	$11.21 \pm 0.45^{a}$	$11.75 \pm 0.74^{a}$	$11.20 \pm 0.64^{a}$
Abdominal fat weight, g	$48.04 \pm 25.31^{b}$	$55.53 \pm 20.96^{a}$	$51.57 \pm 18.49^{ab}$	$47.73 \pm 13.66^{b}$
Abdominal fat ratio, %	$2.19 \pm 0.92^{b}$	$2.65 \pm 0.72^{a}$	$2.51 \pm 0.74^{ab}$	$2.08 \pm 0.63^{b}$
Tibia weight, g	$48.57 \pm 8.43^{b}$	$49.93 \pm 10.55^{b}$	$62.14 \pm 12.93^{a}$	$54.60 \pm 12.58^{ab}$

<sup>a,b</sup> In the same row, values with different small letter superscripts mean significant difference (P < 0.05), while with the same or no letter superscripts mean no significant difference (P > 0.05).

#### Table 4

Effects of dietary alfalfa flavonoids supplement on organ weight (g) of Yangzhou geese at the age of 28 to 70 days.

Item	Groups			
	Control	1	2	3
Hearth weight	21.89 ± 3.21	22.21 ± 3.86	$22.46 \pm 2.49$	22.14 ± 3.48
Liver weight	$74.25 \pm 8.57$	$76.06 \pm 12.28$	77.13 ± 6.98	75.33 ± 11.68
Spleen weight	$3.01 \pm 0.73^{b}$	$3.90 \pm 1.20^{a}$	$4.06 \pm 1.08^{a}$	$3.98 \pm 0.78^{a}$
Bursa weight	$1.40 \pm 0.49^{\rm b}$	$1.46 \pm 0.46^{\rm b}$	$1.80 \pm 0.52^{a}$	$1.61 \pm 0.69^{ab}$
Muscular stomach weight	$108.13 \pm 16.65^{b}$	$111.68 \pm 21.85^{b}$	$130.05 \pm 22.18^{a}$	$129.89 \pm 21.30^{a}$
Gland stomach weight	$12.99 \pm 1.36$	$13.08 \pm 2.09$	$13.50 \pm 2.14$	$13.32 \pm 2.47$

<sup>a,b</sup> In the same row, values with different small letter superscripts mean significant difference (P < 0.05), while with the same or no letter superscripts mean no significant difference (P > 0.05).

adding 0.2% Sea buckthorn in the basal diets could significantly improve the AA broilers' slaughter rates. Sea buckthorn leaves flavonoids can promote the utilization of calcium and phosphorus in feed, which will help the deposition of calcium and phosphorus in tibia and promote development of tibia as well as bone strength (Chen et al., 2010). The results of this experiment and the above

 Table 5

 Effects of dietary alfalfa flavonoids supplement on serum biochemical indexes of Yangzhou geese at the age of 28 to 70 days.

Item	Groups			
	Control	1	2	3
TC, mmol/L	$3.92 \pm 0.57^{a}$	$3.63 \pm 0.43^{ab}$	$3.20\pm0.62^b$	$3.18\pm0.47^{b}$
TG, mmol/L	$1.07 \pm 0.27^{a}$	$0.89 \pm 0.35^{ab}$	$0.73 \pm 0.36^{b}$	$0.90 \pm 0.27^{ab}$
HDL, mmol/L	$1.50 \pm 0.26^{b}$	$1.73 \pm 0.25^{a}$	$1.76 \pm 0.40^{a}$	$1.47 \pm 0.23^{b}$
LDL, mmol/L	$1.12 \pm 0.30^{a}$	$1.01 \pm 0.34^{b}$	$1.04 \pm 0.27^{b}$	$1.02 \pm 0.33^{b}$
UN, mmol/L	$8.13 \pm 0.19^{a}$	$8.01 \pm 0.12^{a}$	$7.62 \pm 0.13^{b}$	$7.70 \pm 0.15^{b}$
TP, g/L	$45.08 \pm 4.39$	$45.24 \pm 2.04$	$46.89 \pm 5.52$	44.70 ± 6.15
ALB, g/L	$14.37 \pm 0.99$	$15.71 \pm 0.67$	15.28 ± 1.25	$14.40 \pm 1.02$
GLB, g/L	30.11 ± 3.65	30.52 ± 1.84	$31.62 \pm 4.74$	30.30 ± 5.82
A:G	$0.47 \pm 0.04$	$0.52 \pm 0.03$	$0.49\pm0.06$	$0.49 \pm 0.07$
ALT, IU/L	$14.10 \pm 1.67$	$14.55 \pm 1.99$	16.27 ± 1.33	19.70 ± 1.32
AST, IU/L	$54.00 \pm 8.58$	55.45 ± 10.56	64.75 ± 10.77	61.20 ± 13.36
ALP, IU/L	$743.50 \pm 29.86$	$748.30 \pm 58.11$	$748.33 \pm 27.25$	$742.50 \pm 58.89$

TC = total cholesterol; TG = triglycerides; HDL = high-density lipoprotein; LDL = low-density lipoprotein; UN = urea nitrogen; TP = total serum protein; ALB = albumin; CLB = globulin; A:G = albumin:globulin; ALT = alanine aminotransferase; AST = aspartate aminotransferase; ALP = alkaline phosphatase. <sup>a,b</sup> In the same row, values with different small letter superscripts mean significant difference (P < 0.05), while with the same or no letter superscripts mean no sig-

nificant difference (P > 0.05).

conclusion have some similarities, alfalfa flavonoids adding in the diet with 300 mg/kg significantly increased feed intake, average daily gain and final weight, and reduced abdominal fat; Breast muscle and tibia indexes of each experimental group were significantly higher than those in the control group, this may be on account of the amount of alfalfa flavonoids acting as a role of estrogen-like which promotes the growth performance, slaughter rate and feed rate of the Yangzhou geese to some extent without increasing deposition of abdominal fat; This is similar to the conclusion of Zhao et al. (2012); These results suggest that an appropriate amount of alfalfa flavonoids is feasible in the diets of Yangzhou geese.

Growth and development of the various organs can indirectly reflect the digestion and absorption of nutrients on geese, whereas the heart and the liver are the vital organs for the body's metabolism; Spleen and bursa are the important immune organs of the geese; As early as 1985, Rivas and Abricant (1985) deemed that the bursa and spleen weight could be used to evaluate the immune status of chicks; Bursa was regarded as mature place of B lymphocytes; When pluripotent stem cells derived from bone went into the bursa, which would differentiate into mature B lymphocytes under the function of bursa's pigment, and also played an important role in humoral immunity; Length of time of the bursa in chicken's body had an important influence on immune functions especially the humoral immunity (Liao et al., 1996); The increase of its mass might promote the differentiation and maturation of B cells as well as humoral immunity (Zhang and Han, 1993). Stomach is the main digestive organs of geese which has developed muscular wall covering with cuticle inside and is capable of grinding food (Liu et al., 2009). In this experiment, the effect of dietary alfalfa flavonoids promoted the organ index, where liver, spleen and bursa weight increased with the adding levels; and the overall trend increased in the first place and then declined, which might due to the fact that alfalfa flavonoids increased the livers' sugar storage, original protein synthesis and secretion as well as fully promoting the immune mechanism of the spleen and bursa. The weight of the gizzard and proventriculus within a certain range was on an increasing trend and would also lead to decline if over-feeding, this might due to the factor that excessive alfalfa flavonoids affected the palatability of the diet.

The TG and TC are an integral components of blood fat, content level of which reflects the absorption, metabolism and utilization of lipids; the lower the content, the higher utilization of fat will be indicated; And the main physiological functions of HDL is to transport cholesterol to cells throughout the body, and finally to the liver for bile acid synthesis (Hao et al., 2010; Cong et al., 2006); LDL enrich cholesterol, and high levels will lead to atherosclerosis (Dombos et al., 1990). The results of this experiment showed that alfalfa flavonoids added in diet decreased the serum levels of TG, TC and LDL on Yangzhou geese, and improved the HDL content, which indicated that a certain level of alfalfa flavonoids could promote cholesterol metabolism, reduce cholesterol deposition in the body thereby improving the quality of meat (Zhou, 2011). Studies have shown that diets supplemented with 150 to 450 mg/ kg alfalfa flavonoids can increase the serum levels of TP, ALB and GLB in chicken, which is consistent with the results of this study indicating that alfalfa flavonoids in a certain level will have no adverse effect on protein metabolism and liver function: In contrast, it is conducive to protein synthesis on Yangzhou geese: Serum UN in poultry is the major end metabolic product of amino acid, and protein metabolism can be reflected by serum UN level of concentration; Elevated serum UN levels reflect that it will strengthen reaction of protein decomposition, but if it has declining trend which means that it will increase deposition of protein synthesis and improve the utilization of nitrogen (Qiu et al., 2009). In this experiment, compared with the control group, experimental group showed a declining trend of UN but maintained in the normal range; the results of this study and Zhou's experiment (Zhou, 2011) showed a consistent explanation of alfalfa flavonoids added in diets which increased the utilization of nitrogen and deposition of protein on Yangzhou geese. Serum ALT and AST are animals' important aminotransferase, which play an important intermediary role in the catabolism of protein and non-essential amino acids synthesis, the level of which reflect the status of protein synthesis and catabolism (Wang et al., 2015a, b). In this experiment, the levels of ALT and AST in each group has increased compared with the control group, but the trend was not significant; Probably because adding alfalfa flavonoids increased the utilization of protein and amino acid while accelerating the use of these two kinds of transaminases, but they had no adverse effect on Yangzhou geese. This is consistent with Wang et al. (2015a, b) and other findings. Usually under normal growth conditions of livestock, activity of ALP will rise, and it will decline only under the intestinal malabsorption. ALP is pyrophosphatase, which is capable of catalytic decomposition on pyrophosphate, and can promote deposition of bone salt (Liu et al., 2006). This experiment results showed that the level of ALP in experimental groups 1 and 2 were higher than those in the control group, which indicated alfalfa flavonoids could increase the activity of ALP while intestinal malabsorption did not occur; and it could also promote the growth of bone on Yangzhou geese to some extent, which were basically consistent with tibia measured results. But alfalfa flavonoids might contain some other substances that might affect serum biochemical indexes, and the specific mechanism would be demanded to do further research.

#### 5. Conclusion

Alfalfa flavonoids extraction adding in the diet of Yangzhou geese aged from 28 to 70 days can improve dietary intake, but shows no adverse effects on slaughter performance, organs and blood biochemical indexes. However, some improvement show in this experiment when the amount of supplementation reaches 300 mg/kg, which will increase economic benefits of Yangzhou geese.

In order to explore the best supplemental value of the amount of alfalfa flavonoids in Yangzhou geese s' diet, we can design more density gradient around 300 mg/kg to do further research.

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