Full Length Research Paper

# Effects of different diets on growth performance, physiological parameters of digestive tract and apparent digestibility in geese

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This experiment was conducted to compare the effects of different diets, containing different fibre sources, on growth performance, hormones level, digestive enzyme activity, intestinal canal morphology, and apparent digestibility. In the first experiment, a total of 216 three-weeks-old Yangzhou goslings were distributed into six pens of 36 birds/pen (18 males and 18 females). Each treatment was represented by three replicates. The experimental diets for the six different treatments with corresponding Groups 1 to 6 were supplemented with corn-soybean-based, alfalfa meal, wheat bran and ryegrass, respectively as the major source of fibre, and the contents of these three fodders were 14, 16, 18, 20, 22 and 24% respectively, severally corresponding to diets 1 to 6. In the second experiment, eighteen 10-weeks-old male geese were placed in metabolism cages, and the birds of treatments 1 to 6 received diets 1 to 6, respectively. The results show that the feed intakes of Group 3 were the most, but there were no significant differences among these six groups (P > 0.05). The body weight (BW) of Group 3 geese was the biggest; moreover they appeared different to a certain extent. The contents of triiodothyronine  $(T_3)$ and growth hormone (GH) in Group 3 were the highest, thereinto the content of  $T_3$  in Group 3 was more than other groups except Group 2, and the content of GH in Group 3 was more than Groups 5 and 6 (P <0.05). The digestive enzyme activity in intestinal tract in Group 3 was the most, but there was no significant difference among these six groups (P > 0.05). The intestinal wall thickness of duodenum, jejunum and cecum in Group 3 was the most, in the same way, the villus height of duodenum in Group 3 was also the most. However, the intestinal wall thickness and villus height in Groups 5 and 6 were generally less. When the proportion of alfalfa, ryegrass and wheat bran in dietary was more than 18%, the apparent nutrient digestive ratios decreased. The current study indicates that the feasible proportion of alfalfa, ryegrass and wheat bran in diet in geese was 18%.

Key words: Fibre, goose, growth performance, digestive enzyme, digestibility.

## INTRODUCTION

Geese are herbivorous waterfowl and they consume and digest large amounts of green grass, clover, and some plants (Kropp, 1975), and have stronger ability of digesting plant structural materials than other birds (Jamroz et al., 1992). Lu et al. (2011) reported that rice

husk (RH) diluted diet in which the basal diet was diluted by 400 g/kg RH could not affect BW, carcass yield or digestive tract, except for thigh yield and cecum weight, and RH could use as a nutrient diluent feedstuff in diets of adult geese. The geese are able to utilize a high-fibre diet because they have an efficient and powerful gizzard and because of the effective microbial breakdown of fibre in the cecum and large intestine (Yang et al., 2009). The fibre may be partially digested by acid hydrolysis early in the digestive tract, and the small particles of the fluid

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digesta obtained may undergo a limited fermentation in the caecae (Durant, 2003). The digestibility of neutral detergent fibre (NDF), acid detergent fibre (ADF) and hemicellulose (HC) was the highest in duodenum, gizzard and duodenum and cecum, respectively.

Comparing the activity of amylase, protease, lipase and cellulose between chicken and duck, it is easy to find that there are significant differences of utilizing fibre of plant feedstuffs between fowls (Fan, 2003). Yu et al. (1998) have studied the influence of different dietary fibres on growth performance, small intestinal morphology and cecum carbohydrase in domestic geese. Furthermore, the apparent nutrient digestibility of different roughage sources are different. Apparent digestibility of organic matter, crude fibre (CF), NDF and ADF in alfalfa meal, grass meal and dried sugar beet pulp were determined as 47.9, 41.2 and 47.6; 43.8, 27.9 and 24.6; 22.8, 29.3 and 45.7; and 6.9, 16.2 and 19.8%, respectively (Arslan and Inal, 2003). In addition, Dawson et al. (2000) reported that fibre digestion on both diets was small, 19 and 27% of NDF for rice and grass, respectively.

The utilization of fibre by geese has attracted more and more attentions. Comparing the effects of different dietary, different fibres sources. containing on growth performance, hormones level, digestive enzyme, alimentary canal morphologic, and apparent nutrient digestive ratio can ascertain the content of fodders so as to provide reference material for the raisers.

#### MATERIALS AND METHODS

#### **Experimental design**

#### Experiment 1

A total of 216 Yangzhou goslings, three-weeks-old and with similar BW, produced by the same flock of geese, were allocated to six groups of thirty-six each (18 males, 18 females) according to similar pen weight. Each treatment was represented by 3 replicates (pens). All birds, under natural daylight, were reared on the floor with padding and received the experimental diets at fixed time from 3 to 10 weeks of age. Body weight was recorded at 21, 28, 35, 42, 49, 56, 63 and 70 days of age, and feed intake by pen was measured on a daily basis. Mortality was recorded as it occurred, and the feed intake was corrected for any bird that died during the course of the experiment.

At 10th week of age, from the six pens per experimental group, six geese (three males and three females) approaching average weight were taken, and the blood samples of them were collected using anticoaglulant from a wing vein. Afterwards, the blood samples were immediately centrifuged (3,000 rpm) for a 10 min and the plasma stored frozen at -20°C for hormone analyses. Then, all the birds were euthanized, and the chyme in digestive tract was collected. Finally, the activity of amylase, protease and lipase in pancreatic gland, duodenum, jejunum, and ileum was determined, respectively.

#### Experiment 2

At 70th day of age, three male geese from each group (one

bird/pen) with BW closest to the mean weight of the group were selected and housed separately in metabolism cages (75 x 65 x 35 cm) with wire floors, equipped with individual feeders and self-drinking systems, and the data were recorded individually. The daily dry matter amount of feed per goose was adjusted to 4% of body weight through force feeding (Yang, 1999). The whole experiment involved a four-day adaptation and four-day collection period. During the collection period, excreta was collected into a bottle, dried to a constant weight at 65°C, allowed to reach equilibrium with the atmospheric moisture for a 24 h, weighed and ground to pass through a 0.4 mm mesh sieve. Total amount of collected excreta was immediately frozen and stored at -20°C for further analysis. At last, all the experiment geese were euthanized and then the gizzard, duodenum, jejunum, ileum and cecum were extracted to measure empty gizzard weight, intestinal wall thickness and villus height.

The experimental procedures were approved by the Yangzhou University Animal Care and Use Committee.

#### **Experimental diets**

The experimental diets for the six different treatments, formulated into isoenergetic and isonitrogenous diets, were supplemented with corn-soybean-based, alfalfa meal, wheat bran and ryegrass, respectively as the major source of fibre. The contents of these three fodders in different treatments were 14, 16, 18, 20, 22 and 24%, respectively.

During the experimental period, goslings in treatments 1 to 6 received diets 1 to 6, respectively (Table 1). The diets were pulverous and offered *ad libitum* to each replicate. Water was freely available throughout the trial.

#### **Digestive enzyme studies**

#### Enzyme extraction

Assay for amylase activity was determined using the method of Somogyi (1960). One unit of amylase activity was defined as the amount of amylase that caused formation of reducing power equivalent to 1 mg of glucose in 30 min at 38°C per mg of intestinal digesta protein or pancreas. The substrate used in the analysis was cornstarch.

#### Digestive enzyme assays

Protease activity was analyzed using the method of Lynn and Clevette-Radford (1984). The protease activity unit was defined as milligrams of azocasein degraded during a 2 h incubation at 38°C per mg of intestinal digesta protein or pancreas. Azocasein was used as the substrate.

Lipase activity was assayed using the method described by Tietz and Fiereck (1966). Lipase activity unit was equal to the volume (in mL) of 0.05 M NaOH required to neutralize the fatty acid liberated during a 6 h incubation with 3 mL of lipase substrate at 38°C per mg of intestinal digesta protein or pancreas. Olive oil was used as the substrate in this assay.

#### Hormone studies

Plasma triiodothyronine  $(T_3)$  and thyroxine  $(T_4)$  content were measured by radioimmunoassay as described by Darras et al. (1992). Intraassay coefficients of variation were 4.5 and 5.4% for  $T_3$ and  $T_4$ , respectively. Growth hormone (GH) content was measured by homologous radioimmunoassay (RIA) as developed and Table 1. Composition and calculated analysis (g/kg, as fed) of the experimental diets<sup>1</sup>

	Diet								
Item -	1	2	3	4	5	6			
Ingredient (%)									
Corn	57.67	56.28	55.92	53.68	51.59	49.34			
Soybean meal	17.54	16.89	15.28	15.28	14.43	13.83			
Vitamin and trace mineral premix <sup>2</sup>	4.72	4.72	4.72	4.72	4.72	4.72			
Alfalfa	7.05	0	2.83	0	19.53	23.86			
Wheat bran	0	7.55	4.84	9.43	0	0			
Ryegrass	6.95	8.49	10.34	10.82	2.47	0			
Limestone	0.2	0.2	0.2	0.2	0	0			
Lysine	0.16	0.16	0.16	0.16	0.16	0.16			
Methionine	0.04	0.04	0.04	0.04	0.04	0.04			
Vegetable oil	0	0	0	0	1.39	2.38			
Fish meal	5.67	5.67	5.67	5.67	5.67	5.67			
Total	100	100	100	100	100	100			
Composition <sup>3</sup>									
ME (Mcal/Kg)	2.53	2.53	2.54	2.51	2.51	2.52			
Crude protein (%)	17.55	17.73	17.54	17.74	17.59	17.45			
Crude fibre (%)	6.92	6.90	6.92	6.90	6.90	6.96			
Calcium (%)	0.99	1.08	1.02	0.99	1.13	1.18			
Available phosphorus (%)	0.71	0.73	0.75	0.81	0.67	0.64			
Lysine (%)	0.79	0.82	0.79	0.80	0.81	0.80			
Methionine (%)	0.40	0.42	0.41	0.41	0.41	0.41			

<sup>1</sup>Values are expressed on a DM basis. <sup>2</sup>Supplied, per kilogram of diet: vitamin A (retinyl acetate), 30 mg; vitamin D<sub>3</sub> (cholecalciferol), 0.5625 mg; vitamin E (di-α-tocopheryl acetate), 150 mg; vitamin K (2-methyl-1,4-naphthoquinone), 100 mg; thiamin, 50 mg; riboflavin, 600 mg; pyridoxine, 100 mg; vitamin B<sub>12</sub> (cobalamin), 1 mg; nicotinic acid, 3 g; pantothenic acid, 900 mg; folic acid, 50 mg; biotin, 4 mg; choline, 35 g; Fe, 6 g; Cu, 1 g; Mn, 9.5 g; Zn, 9 g; I, 50 mg; Se, 30 mg. <sup>3</sup>Analyzed values given for CP, crude fibre, calcium, and available phosphorus. Calculated values given for lysine and methionine.

validated by Berghman et al. (1988). The intraassay coefficient of variation was 4.0%. All measurements of a given hormone were performed in a single assay.

#### Intestinal morphology

Intestinal sections were collected and placed in phosphate-buffered formalin for preservation. All samples were collected within 15 min of the time of death. Tissues were embedded in paraffin and sliced into 3  $\mu$ m sections using a microtome. Samples then were placed on glass slides followed by staining with hematoxylin and eosin. Digital images of tissues were taken using a Nikon Optiphot microscope. Height and width measurements of small intestinal villi were taken using Image Pro Plus® software.

#### Determination of apparent digestibility

Samples were analyzed for total N using the micro-Kjeldahl method (990.03, AOAC, 2000). Crude protein was calculated as N  $\times$  6.25. Calcium (Ca) was determined by atomic absorption spectrophotometry according to the methods of the Association of

Official Analytical Chemists. Phosphorus (P) was determined photometrically as orthophosphate from filtered ash solutions, using the vanado-molybdate method.

Ashing of samples was performed at 550°C for a 12 h. CF was measured by sequential extraction with diluted acid and alkali (962.09; AOAC, 2000). NDF and ADF were determined sequentially as described by Van Soest et al. (1991) and expressed on an ash-free basis.

The apparent digestibility was calculated using the following formula:

Apparent digestibility = (feed intake  $\times$  nutrient diet – excreta output  $\times$  nutrient excreta)/ (feed intake  $\times$  nutrient diet)  $\times$  100%.

#### Statistical analysis

All data from the experiments were analysed using a one-way analysis of variance (ANOVA) (SPSS Inc., 1993). Differences among treatment means were compared by Duncan's multiple-range test. Statements of significance were based on P < 0.05.

Age	Group (g/bird)								
(week)	1	2	3	4	5	6			
Number	36	36	36	36	36	36			
3-4	627 ± 93	608 ± 89	633 ± 97	613 ± 91	602 ± 79	582 ± 81			
4-5	842 ± 126	818 ± 121	915 ± 117	864 ± 130	805 ± 115	760 ± 131			
5-6	1076 ± 161	1164 ± 149	1107 ± 155	1020 ± 145	1033 ± 165	986 ± 129			
6-7	1110 ± 163	1180 ± 179	1278 ± 116	1164 ± 166	1030 ± 123	1093 ± 169			
7-8	1245 ± 151	1248 ± 178	1273 ± 162	1176 ± 184	1078 ± 173	1057 ± 187			
8-9	1190 ± 177	1255 ± 135	1318 ± 154	1140 ± 149	1055 ± 139	1022 ± 174			
9-10	1280 ± 213	1320 ± 124	1340 ± 173	1245 ± 171	1122 ± 159	1073 ± 181			

**Table 2.** Influence of different diets on weekly feed intakes of geese.

Table 3. Influence of different diets on weekly weight.

Age	Group (g/bird)								
(week)	1	2	3	4	5	6			
Number	36	36	36	36	36	36			
3	406.35 ± 81.66	405.41 ± 62.72	397.2 ± 67.23	405.15 ± 55.85	399.35 ± 60.38	409.48 ± 72.49			
4	779.17 ± 137.81	774.41 ± 135.45	778.09 ± 153.08	780.81 ± 167.87	771.91 ± 131.67	770.34 ± 175.25			
5	1239.89 ± 213.75 <sup>bd</sup>	1227.35 ± 190.74 <sup>bd</sup>	1304.53 ± 190.33 <sup>d</sup>	1212.89 ± 197.93 <sup>bd</sup>	1131.07 ± 214.47 <sup>ab</sup>	1093.17 ± 145.54 <sup>a</sup>			
6	1396.31 ± 187.10 <sup>abc</sup>	1469.59 ± 252.77 <sup>bc</sup>	1562.48 ± 147.02 <sup>c</sup>	1475.38 ± 190.74 <sup>bc</sup>	1361.30 ± 125.74 <sup>ab</sup>	1286.19 ± 169.01 <sup>a</sup>			
7	1787.50 ± 106.35 <sup>b</sup>	1779.33 ± 126.34 <sup>b</sup>	1848.44 ± 115.45 <sup>c</sup>	1783.04 ± 137.79 <sup>b</sup>	1694.68 ± 127.01 <sup>ab</sup>	1574.18 ± 142.57 <sup>a</sup>			
8	1948.60 ± 146.30 <sup>ab</sup>	2044.94 ± 130.04 <sup>ab</sup>	2204.91 ± 174.01 <sup>b</sup>	2166.83 ± 116.22 <sup>b</sup>	1898.92 ± 153.09 <sup>ab</sup>	1820.30 ± 188.54 <sup>a</sup>			
9	2184.20 ± 211.95	2467.83 ± 137.93	2574.17 ± 134.93	2452.00 ± 175.71	2221.17 ± 114.33	2118.50 ± 159.71			
10	2411.50 ± 248.71	2584.50 ± 107.44	2717.50 ± 174.27	2632.20 ± 162.90	2336.13 ± 181.36	2333.50 ± 200.54			

<sup>a,b,c,d</sup>Means within a row lacking a common superscript were differ significantly (P < 0.05).

## RESULTS

#### **Growth performance**

During the experiment periods, the sequence of weekly feed intake was Groups 3, 2, 1, 4, 5 and 6 in turns (Table 2), but there were no significant

differences among them (P > 0.05). Weekly weight of the six groups geese improved gradually with the time being (Table 3). During the experimental period, the weight of Group 3 geese was the biggest, and that of Group 6 geese was the least. The change trend of weekly weight was similar to that of weekly feed intake. The birds of Group 3 were weightier than those of Groups 5 and 6 from 5 to 7-weeks-old (P < 0.05). At the same time, the birds of Groups 1, 2 and 4 were heavier than those of Group 6 at 5 and 7-weeks-old (P < 0.05), and the weight of Groups 4 and 2 birds were more than those of Group 6 at 6-weeks-old too (P < 0.05). At eight-weeks age, the birds of Groups 3 and 4 were

ltere	Group							
Item	1	2	3	4	5	6		
Number	6	6	6	6	6	6		
Triiodothyronine (ng/ml)	$0.62 \pm 0.22^{abc}$	$0.67 \pm 0.18^{acd}$	$0.84 \pm 0.29^{d}$	$0.75 \pm 0.25^{\circ}$	$0.57 \pm 0.23^{a}$	$0.46 \pm 0.10^{a}$		
Tyroxine (ng/ml)	12.48 ± 1.98	11.98 ± 1.16	10.59 ± 1.44	10.61 ± 1.65	10.51 ± 1.52	10.71 ± 1.71		
Growth hormone (ng/ml)	$0.27 \pm 0.14^{ac}$	$0.24 \pm 0.11^{a}$	$0.40 \pm 0.12^{\circ}$	$0.31 \pm 0.16^{\circ}$	$0.21 \pm 0.09^{a}$	$0.23 \pm 0.09^{a}$		

Table 4. Influence of different diets on plasma concentrations of triiodothyronine, tyroxine and growth hormone.

<sup>a,b,c,d</sup>Means within a row lacking a common superscript were differ significantly (P < 0.05).

weightier than those of Group 6 (P < 0.05).

## Plasma hormone level

The level of  $T_3$  in Group 3 which was the highest was more than those of Groups 1, 4, 5, and 6 (*P* < 0.05), and the level of  $T_3$  in Group 4 was also evidently higher than those of Group 5 and Group 6 (*P* < 0.05) (Table 4). The levels of  $T_4$  were different, but there were no differences among them (*P* > 0.05). The level of GH in Group 3 which was the highest was distinctly higher than those in Groups 2, 5 and 6 (*P* < 0.05). The content of GH in Group 4 was also significantly higher than those in Groups 2, 5 and 6 (*P* < 0.05).

## **Digestive enzyme**

Activity of the same digestive enzyme in different groups was dissimilar, but there were no significant differences among them (Table 5). In pancreas, the activities of amylase, protease and lipase were higher in Groups 3 and 4, however those were lower in Groups 5 or 6. In duodenum, the activities of amylase and lipase in Group 3 were higher, but those in Groups 5 and 6 were lower. In the same way, the activity of protease in Group 3 was also higher, whereas that in Group 5 was lower. In jejunum, the activity of amylase in Group 3 was higher, nevertheless those in Groups 5 and 6 were lower. The activities of protease and lipase in Group 3 were higher, but those in Group 2 were lower. In ileum, the activity of amylase in Group 3 was higher, howbeit those in Groups 5 and 6 were lower. The activity of protease in Group 3 was higher, but that in Group 2 was lower. The activity of lipase in Group 6 was higher, and that in Group 4 was lower.

Relatively, digestive enzyme activity in pancreas was the highest. The amylase activity in pancreas was nearly one thousand times more than those in duodenum, jejunum or ileum. The protease activity in pancreas was almost thirty times higher those that in duodenum, jejunum or ileum. In like manner, the lipase activity in pancreas was feckly 200 times greater than those in duodenum, jejunum or ileum.

In addition, the activity of amylase was the highest, but that of lipase was the lowest.

## Intestinal morphology

Histological measurements of the intestinal wall

and villus were performed at 70 day of age (Table 6). Gizzard weight of Group 3 was the most, and significantly more than those of Groups 1 and 5 (P > 0.05). In duodenum, the intestinal wall thickness and villus height in Group 3 were the greatest, however the intestinal wall thickness in Group 1 and the villus height in Group 5 were the least, but the differences were not significant (P >0.05). In jejunum, the intestinal wall thickness in Group 3 was thicker than that in Group 5 (P <0.05), and the villus heights in Groups 1 to 4 were longer than those in Groups 5 and 6 (P < 0.05). In ileum, the intestinal wall thickness in Group 4 was thicker than those in Groups 1, 5 and 6 (P < 0.05), and the villus heights in Groups 3 and 4 were longer than that in other Groups (P < 0.05), thereinto the villus height in Group 5 was the shortest. In cecum, though the intestinal wall thickness in Group 3 was the thickest, and that in Group 5 was the least, but there were no differences among them (P > 0.05).

### Apparent digestibility

The apparent retention ratio of crude protein, calcium phosphorus, and dietary fibre components are presented in Table 7. No significant differences (P > 0.05) were found in the retention

-		Group (u/g)									
Enzyme		1	2	3	4	5	6				
Number		6	6	6	6	6	6				
	Amylase	93 272.68 ± 13 284.94	85 945.86 ± 11 353.42	96 303.65 ± 16 101.82	108 644.10 ± 9 614.06	77 507.61 ± 12 072.61	80 919.83 ± 6 242.75				
Pancreas	Protease	874.63 ± 145.57	943.83 ± 146.71	1252.61 ± 265.86	1081.56 ± 191.50	856.46 ± 114.42	928.43 ± 200.43				
	Lipase	$9.54 \pm 7.50$	$10.54 \pm 9.86$	14.07 ± 7.75	13.62 ± 3.47	9.51 ± 4.55	7.17 ± 5.45				
	Amylase	86.05 ± 9.16	95.23 ± 1.79	110.83 ± 8.81	97.68 ± 2.79	63.46 ± 8.76	56.67 ± 5.17				
Duodenum	Protease	30.42 ± 8.25	28.61 ± 10.45	44.17 ± 8.20	34.93 ± 13.35	27.06 ± 18.81	31.28 ± 18.70				
	Lipase	4.48 E-02 ± 0.01	$4.85E-02 \pm 0.03$	6.40 E-02 ± 0.04	3.03 E-02 ± 0.02	4.23 E-02 ± 0.01	2.94 E-02 ± 0.01				
	Amylase	129.63 ± 35.02	114.01 ± 18.93	152.47 ± 30.41	138.27 ± 32.42	91.33 ± 15.97	98.25 ± 26.77				
Jejunum	Protease	24.81 ± 21.68	19.84 ± 17.34	30.48 ± 26.83	27.22 ± 22.98	24.21 ± 25.41	28.96 ± 10.98				
	Lipase	4.60 E-02 ± 0.03	$4.39E-02 \pm 0.04$	6.81 E-02 ± 0.02	5.20 E-02 ± 0.02	4.40 E-02 ± 0.03	4.41 E-02 ± 0.02				
	Amylase	105.05 ± 38.18	107.81 ± 34.07	143.17 ± 35.91	127.99 ± 53.34	84.02 ± 57.44	95.71 ± 30.64				
lleum	Protease	26.23 ± 10.67	24.68 ± 8.13	39.44 ± 11.16	28.82 ± 15.31	25.56 ± 8.03	30.21 ± 13.59				
	Lipase	2.95E-02 ± 0.03	3.03E-02 ± 0.01	2.97 E-02 ± 0.01	1.97 E-02 ± 0.02	2.08 E-02 ± 0.02	3.20 E-02 ± 0.02				

**Table 5.** Effects of different diets on digestive enzyme activity in intestinal tract of geese.

**Table 6.** Influence of different diets feeding on villus height, crypt depth, and intestinal wall thickness of geese at 70 d of age.

		Cimeral	Duodenum		Jejunum		lle	Cecum	
Group	Number	Gizzard weight	Intestinal wall thickness (µm)	Villus height (µm)	Intestinal wall thickness (µm)	Villus height (µm)	Intestinal wall thickness(µm)	Villus height (µm)	Intestinal wall thickness (µm)
1	3	99.67 ± 17.29 <sup>a</sup>	65.00 ± 8.94	89.67 ± 9.07	62.83 ± 8.72 <sup>ab</sup>	112.00 ± 7.45 <sup>c</sup>	59.52 ± 6.47 <sup>abc</sup>	90.53 ± 9.10 <sup>d</sup>	66.17 ± 9.65
2	3	114.62 ± 31.83 <sup>ab</sup>	67.83 ± 6.11	90.33 ± 7.28	$70.33 \pm 6.65^{ab}$	104.83 ± 3.23 <sup>bc</sup>	80.67 ± 20.58 <sup>cd</sup>	69.17 ± 11.58 <sup>b</sup>	72.00 ± 8.21
3	3	128.73 ± 8.22 <sup>b</sup>	75.33 ± 9.93	95.33 ± 6.31	73.33 ± 8.07 <sup>b</sup>	105.33 ± 9.61 <sup>bc</sup>	$74.0 \pm 33.27^{bcd}$	104.17 ± 12.26 <sup>e</sup>	72.17 ± 4.70
4	3	121.24 ± 14.45 <sup>ab</sup>	71.00 ± 4.00	88.83 ± 6.14	65.83 ± 8.54 <sup>ab</sup>	110.17 ± 10.92 <sup>c</sup>	88.33 ± 20.65 <sup>d</sup>	107.17 ± 11.79 <sup>e</sup>	65.33 ± 11.69
5	3	103.40 ± 20.81 <sup>a</sup>	73.17 ± 8.72	81.00 ± 8.53	55.17 ± 3.31 <sup>a</sup>	$86.00 \pm 8.46^{a}$	$48.83 \pm 6.40^{a}$	55.83 ± 10.32 <sup>a</sup>	65.00 ± 4.98
6	3	119.85 ± 11.44 <sup>ab</sup>	72.83 ± 7.57	90.17 ± 11.35	$60.00 \pm 2.75^{ab}$	$85.00 \pm 8.94^{a}$	54.83 ± 5.71 <sup>ab</sup>	$70.67 \pm 9.90^{b}$	65.17 ± 6.91

 $^{a,b,c,d,e}$ Means within a row lacking a common superscript were differ significantly (P < 0.05).

Item	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6
Number	3	3	3	3	3	3
Crude protein (%)	66.26 ± 3.18	67.22 ± 3.33	66.18 ± 3.61	65.88 ± 2.01	61.59 ± 3.95	60.97 ± 4.12
Calcium (%)	34.16 ± 2.78	35.24 ± 4.08	35.64 ± 3.21	35.47 ± 3.06	33.71 ± 2.96	31.39 ± 4.34
Phosphorus (%)	35.40 ± 4.69	38.29 ± 5.67	43.29 ± 5.14	37.10 ± 4.89	28.78 ± 6.99	29.95 ± 7.33
Crude fibre (%)	16.43 ± 3.35 <sup>bce</sup>	18.33 ± 3.81 <sup>c</sup>	19.65 ± 3.54 <sup>c</sup>	11.67 ± 5.24 <sup>ae</sup>	12.32 ± 2.01 <sup>ab</sup>	$10.03 \pm 2.81^{a}$
Neutral detergent fibre (%)	41.67 ± 3.74	46.83 ± 4.95	43.34 ± 2.59	38.40 ± 3.61	38.07 ± 8.19	36.96 ± 6.68
Acid detergent fibre (%)	13.94 ± 4.00 <sup>abc</sup>	20.29 ± 5.13 <sup>c</sup>	18.39 ± 10.88 <sup>bc</sup>	17.28 ± 2.83 <sup>bc</sup>	10.32 ± 2.15 <sup>a</sup>	$9.79 \pm 6.38^{a}$

Table 7. Apparent retention ratio of crude protein, calcium, phosphorus, and dietary fibre components.

<sup>a,b,c,d,e</sup>Means within a row lacking a common superscript were differ significantly (*P* < 0.05).

ratios of crude protein, calcium, phosphorus and NDF. The retention ratio of CF in Group 3 was the highest, and it was obviously more than those in Groups 4, 5 and 6, especially, it was two times as much as that in Group 6. The retention ratios of ADF in Groups 5 and 6 were lower than those in Groups 2, 3 and 4 (P > 0.05), and that in Group 5 was also about half of Group 3.

## DISCUSSION

It has come to light that the feed intakes of poultries are for their energy demands. Therefore, no significant differences appeared among the six groups in weekly feed intakes (Table 2). Geese can digest large amounts of fibre feedstuff, and the structure and function of their digestive system allow them to utilise and thrive on a high-fibre diet (Buckland and Guy, 2002). In this experiment, the weekly feed intake of Group 3 was the most, that is, when the proportion of alfalfa, ryegrass and wheat bran in diets was more than or less than 18%, the weekly feed intake of testing birds decreased. On the one hand, the appetite of the birds became week when the proportion of these three fodders was less. On the other hand, the more the contents of these three fodders were, the bigger the feedstuff cubage was. These maybe, made birds reduce their feed intakes. At the same time, the trend of weekly weight was unanimous with the weekly feed intake on the whole. The experimental diets for the six different treatments, supplemented with corn-soybean-based, were formulated into isoenergetic and isonitrogenous diets, and only the contents of alfalfa meal, wheat bran and ryegrass were different, so the weekly weight of different groups birds were connected with feed intake.

The level of  $T_3$  was in the sequence: Group 3 > Group 4 > Group 2 > Group 1 > Group 5 > Group 6. At the same time, the levels of GH in different groups were similar with that of  $T_3$ .  $T_3$  and GH stimulates growth, cell reproduction and regeneration. As a result, these trends were consistent with the weekly weight. According to the level of  $T_3$  and GH at 10-weeks of age, it can be speculated that the next weekly weight of different groups would possibly appear significant differences.

In this experiment, the activities of digestive enzyme in different groups did not appear obvious differences. However, those in Group 3 were higher than other groups, and those in Groups 5 and 6 were lower than other groups. As a matter of fact, these trends were similar to the weekly feed intake. In other words, the activities of digestive enzyme in intestinal tract were affected by the proportion of alfalfa, ryegrass and wheat bran. When the proportion was 18%, the activities of digestive enzyme were the highest. When the proportion was more than or less than 18%, the activities of digestive enzyme would present decline trends, perhaps the content and composing of this diet can utmostly stimulate digestive enzyme secrete.

Comparing with other tracts of goslings, the activity of digestive enzyme was the highest in pancreas. This could be because, pancreas was the most important digestive gland, and most amylase, protease and lipase in the small intestine tract came from here, so the activity of digestive enzyme in pancreas was higher than other parts of intestinal tract.

The amount and type of dietary fibre might influence its digestibility and the utilisation of other nutrients, and might even affect the development of the gastrointestinal tract (Jamroz et al., 1992).

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In the present experiments, different amounts of fodders significantly influenced the intestinal wall thickness and villi height in jejunum and ileum, thereinto the intestinal wall thickness and villi height in Groups 5 and 6 which fodder contents were more than 20% were distinctly less. In addition, the intestinal wall thickness in duodenum and cecum and the villi height in duodenum in Group 3 were more. The results of the present study show that the fodder contents of 18% might accelerate the small intestine growth.

Digestive constraints may, in part, influence traits such as growth, reproduction and survival (Naya et al., 2005). For example, a more developed gizzard may enhance the grinding of feed that will increase the exposure of nutrients to digestive enzymes and improve nutrient digestion (Hetland et al., 2002; Gabriel et al., 2003), energy utilization (Preston et al., 2000) and performance (Ravindran et al., 2006). Lou et al. (2010) reported that the gizzard played important roles in the digestion of crude fibre in geese. In this experiment, the gizzard weight of Group 3 was the most, and the apparent retention ratio of several nutrition almost was the most too. Though there were no distinct differences in gizzard weight between Groups 6 and 3, the apparent digestibility of Group 6 decreased. The reason perhaps was that the fodder contents were too many. In fact, when the fodder contents were more than 18%, the apparent digestibility, especially crude fibre and ADF, evidently decreased.

Cellulose, hemicelluloses. pectins and some oligosaccharides are partly digested by the microflora of the large intestine (Champ, 1985). Total digestibility of fibre varies considerably and depends on the nature of the fibre and the animal species. In the experiments, the ryegrass was got from farmland, and it was very tender. The alfalfa had entered the autumn, and it had lignified. The alfalfa content in Groups 5 and 6 was high, so the apparent retention ratio of crude fibre was lower. However, the ryegrass content in Group 3 was high, so the apparent retention ratio of crude fibre was also higher. The ryegrass content in Group 4 was high, but the apparent retention ratio of crude fibre was lower than that in Group 3, perhaps the crude fibre retention ratio of wheat bran was distinctly lower than that of alfalfa and corn.

## Conclusions

The proportion of fodders which were comprised of alfalfa, ryegrass and wheat bran in dietary influenced feed intake, plasma hormone level, digestive enzyme activity, gizzard weight, intestinal wall thickness and villus height and apparent retention ratio of partial nutrition. The proportion of fodders cannot exceed 20%, and the feasible proportion should be 18%.

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## REFERENCES

- AOAC (Association of Official Analytical Chemists) (2000). Official Methods of Analysis. 17th ed. AOAC, Gaithersburg, MD.
- Arslan C, Inal F (2003). Determination of apparent nutrient digestibility of different roughage sources in geese. Arch. Geflugelkd. 67:116-119.
- Berghman LR, Van Beemen J, Decuypere E, Kuhn ER, Vandesande F (1988). One step purification of chicken growth hormone from a crude pituitary extract by use of a monoclonal immune-adsorbent. J. Endocrinol. 118:381-387.
- Buckland R, Guy G (2002). Goose Production. FAO Animal Production and Health Paper 154. FAO, Rome.
- Champ M (1985). Carbohydrate digestion in monogastric animals. Reprod. Nutr. Dev. 25:819-842.
- Darras VM, Visser TJ, Berghman LR, Kuhn ER (1992). Ontogeny of type I and III deiodinase activities in embryonic and posthatch chicks: Relationship with changes in plasma triiodothyronine and growth hormone levels. Comp. Biochem. Physiol. A-mol Int. Physiol. 103:131-136.
- Dawson TJ, Whitehead PJ, McLean AF, Fanning D, Dawson WR (2000). Digestive function in *Australian magpie geese (Anseranas semipalmata*). Aust. J. Zool. 48:265-279.
- Durant D (2003). The digestion of fibre in herbivorous Anatidae. Wildfowl 54:7-24.
- Fan HP (2003). Comparative study of the digestion of feed nutrients between cockerel and drake. PhD Dissertation, Chinese Academy of Agricultural Sciences, Beijing.
- Gabriel I, Mallet S, Leconte M (2003). Differences in the digestive tract characteristics of broiler chickens fed on complete pelleted diet or whole wheat added to pelleted protein concentrate. Br. Poult. Sci. 44:283-290.
- Hetland H, Svihus B, Olaisen V (2002). Effect of feeding whole cereals on performance, starch digestibility and duodenal particle size distribution in broiler chickens. Br. Poult. Sci. 43:416-423.
- Jamroz D, Wiliczkiewicz A, Skorupinska J (1992). The effect of diets containing different levels of structural substances on morphological changes in the intestinal walls and the digestibility of the crude fibre fractions in geese (Part 3). J. Anim. Feed. Sci. 1:37-50.
- Kropp LB (1975). Geese: America's fourth fowl. Feedstuffs 16:32-33.
- Lou YJ, Liu HL, Wang J, Sun ZJ (2010). Determination and comparison of digestion kinetics of two fibre sources in geese (Anseris). S. Afr. J. Anim. Sci. 40:70-77.
- Lu J, Wang ZY, Yang HM, Shi SR, Zou JM (2011). Effect of rice husk diluted dietary switching on body weight, carcass yield and digestive tract of adult ganders. Arch. Geflugelkd 75:120-124.
- Lynn KR, Clevette-Radford NA (1984). Purification and characterization of hevain, a serine protease from *Hevea brasiliensis*. Phytochemistry 23:963-964.
- Naya DE, Farfan G, Sabat P, Mendez MA, Bozinovic F (2005). Digestive morphology and enzyme activity in the Andean toad *Bufo spinulosus*: Hard-wired or flexible physiology? Comp. Biochem. Physiol. A-mol Int. Physiol. 140:165-170.
- Preston CM, McCracken KJ, McAllister A (2000). Effect of diet form and enzyme supplementation on growth, efficiency and energy utilisation of wheat-based diets for broilers. Br. Poult. Sci. 41:324-331.
- Ravindran V, Wu YB, Thomas DG, Morel PCH (2006). Influence of whole wheat feeding on the development of gastrointestinal tract and performance of broiler chickens. Aust. J. Agric. Res. 57:21-26.
- Somogyi M (1960). Modification of two methods for the assay of amylase. Clin. Chem. 6:23-27.
- Tietz NW, Fiereck EA (1966). A specific method for serum lipase determination. Clin. Chim. Acta. 13:352-355.
- Van Soest PJ, Robertson JB, Lewis A (1991). Methods for dietary fibre, neutral detergent fibre, and nonstarch polysaccharides in relation to

animal nutrition. J. Dairy Sci. 74:3583-3597.

Yang F (1999). Animal nutrition. 2nd ed. China Agric Press, Beijing. Yang HM, Wang ZY, Wang J, Shi SR, Zhu XH (2009). Effects of caecectomy on digestibility of crude protein, calcium, phosphorus, neutral detergent fibre and acid detergent fibre in geese. Arch. Geflugelkd 73:189-192. Yu B, Tsai CC, Hsu JC, Chiou PWS (1998). Effect of sources of dietary fibre on growth performance, intestinal morphology and caecal carbohydrases of domestic geese. Br. Poult. Sci. 39:560-567.