Animal Nutrition 3 (2017) 33-38

Contents lists available at ScienceDirect

Animal Nutrition

journal homepage: http://www.keaipublishing.com/en/journals/aninu/

Original Research Article

Effects of flavonoids from *Allium mongolicum Regel* on growth performance and growth-related hormones in meat sheep

Muqier, Sarula Qi, Terigele Wang, Renwei Chen, Cuifang Wang, Changjin Ao*

College of Animal Science, Inner Mongolia Agricultural University, Hohhot 010018, China

ARTICLE INFO

Article history: Received 29 August 2016 Accepted 10 January 2017 Available online 17 January 2017

Keywords: Allium mongolicum Regel Flavonoids Growth performance Hormones Meat sheep

ABSTRACT

This study was conducted to investigate the effects of different doses of flavonoids from Allium mongolicum Regel on the production performance and neuroendocrine hormones in meat sheep and to determine the optimum dosage of Allium mongolicum Regel flavonoids to add to the basal diet of dry lotfeeding meat sheep. Sixty meat sheep (initial body weight = 39.9 ± 3.2 kg; 6-month-old) were randomly assigned to 4 groups (15 sheep per group). The sheep in the control group were fed a basal diet, and the 3 experimental groups were fed the basal diet supplemented with flavonoids at 11, 22 and 33 mg/kg. Blood samples were collected via the jugular vein at d 0, 15, 30, 45, and 60 to determine the neuroendocrine hormone levels. The fasting weight of the sheep was measured during the experimental period, and feed offered and refusals were recorded daily. The basal diet supplemented with flavonoids from 11 to 33 mg/ kg significantly increased the daily weight gain and average daily feed intake (P < 0.05) and significantly decreased the feed conversion ratio (P < 0.05), but there were no differences among the supplementation groups (P > 0.05). Starting on d 30, the growth hormone (GH) and insulin-like growth factor-1 (IGF-1) levels in the sera of the sheep in the supplementation groups increased significantly (P < 0.05), and the increases occurred in a time-dependent manner. Compared with control group, after d 30, the serum corticosterone (CORT) levels were reduced in the sheep that consumed the basal diet supplemented with 22 mg/kg flavonoids (P < 0.05), but among the other experimental groups, there was a non-significant effect (P > 0.05). The serum adrenocorticotropic hormone (ACTH) levels were increased by the supplementation of flavonoids, but compared with the control group, the effect was not significant. The basal diet supplemented with flavonoids at levels from 11 to 33 mg/kg had a significant effect on the production performance and neuroendocrine hormone levels of meat sheep, and the effect occurred in a time-dependent manner. The effect was especially obvious after 30 d of feeding.

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1. Introduction

Allium mongolicum Regel, also known as the Mongolia leek, is a type of Liliaceous allium plant that grows in high altitude desert steppe and desert areas; it is perennial and xerophytic (Wang et al.,

* Corresponding author.

E-mail address: changjinao@aliyun.com (C. Ao).

Peer review under responsibility of Chinese Association of Animal Science and Veterinary Medicine.

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2013). It is mainly found in desert land in Qinghai, Gansu, Xinjiang and Inner Mongolia, and is especially abundant in the western areas of Xilingol, Ordos, and Alxain Inner Mongolia. *Allium mongolicum Regel*, as a characteristic wild vegetable, is a natural and healthy food with a high nutritional value, unique flavour and supreme palatability (Chen et al., 2000). *Allium mongolicum Regel* and its extracts have been shown to increase the average daily gain (ADG), feed intake and feed remuneration of broiler chickens (Ha, 2008) and to significantly affect growth-related hormones (Zhang, 2005).

Flavonoids are active ingredients in natural plants that can promote growth, neuroendocrine and immune function of animals. Related research found that the isoflavone daidzein promoted animal growth (Han, 1999), improved immune function (Guo and Zhao, 2004), and enhanced lactation and laying (Hu and Zhang, 2009; Yang et al., 2006). Flavonoids are an important active

http://dx.doi.org/10.1016/j.aninu.2017.01.003







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substance in *Allium mongolicum Regel* (Sa et al., 2014). According to research, *Allium mongolicum Regel* flavonoids show biological efficacy as anti-oxidants and have anti-bacterial and immune regulation abilities. *Allium mongolicum Regel* provides small doses, low toxicity, and high efficacy in practical application; consequently, it offers good latent capacity for new types of feed additives. Animal growth is a complicated metabolic process influenced by genes, nutrients, hormones and the environment and regulated by neuroendocrine function (Tao, 2006), of which the hypothalamic-pituitary-growth axis is the key regulator (Sarah, 2003).

Currently, research on the flavonoids from *Allium mongolicum Regel* has mainly focused on its *in vitro* anti-oxidant, anti-bacterial and anti-viral activities; studies of growth performance and growth-related hormones in meat sheep are rare. On the basis of a preliminary *in vitro* study (Bao, 2015; Sa, 2014), this study selected the flavonoids from *Allium mongolicum Regel* as feed additives for meat sheep. Different doses of flavonoids from *Allium mongolicum Regel* were added to the basal diet, and the effect on growth performance and growth-related hormones in meat sheep was explored. A suitable amount of flavonoids from *Allium mongolicum Regel* was determined for use as a natural feed additive.

2. Materials and methods

2.1. Experimental flavonoids from Allium mongolicum Regel

The flavonoids from Allium mongolicum Regel were extracted using the extraction processing method described by Saruli (Sa, 2014). An ultrasonic extraction method was adopted using the following parameters: extraction time = 15 min, extraction temperature = 40 °C, alcohol density = 75%, and material-to-liquid ratio = 1:30. The projected total flavonoid yield was 12.85 mg/g. The flavonoids from Allium mongolicum Regel are yellow in colour and insoluble in water. According to early research regarding structure identification (Sa, 2014), the structure contains saccharides, naphthenic hydrocarbon, 3',4'-epoxygroup-7-0-5-methoxy flavonols, 7-0-5,4'-dimethoxy-3'-hydroxide radical flavones, rutin, quercitrin, saccharides, mignonette element - 5 0-glucose-4-hydroxybenzoic acid, and acacia.

2.2. Reagents and apparatus

Testing kits were used to determine the levels of serum growth hormone (GH; HY-C0018), serum insulin-like growth factor-1 (IGF-1; HY-H0024), serum corticosterone (CORT; HY-D0032), and serum adrenocorticotropic hormone (ACTH; HY-D0023). The testing method was radioimmunoassay, and the test apparatus was an r-911 auto radio free counter (provided by the Science and Technology Industrial Company of the University of Science and Technology of China). All reagents and apparatuses were provided by the Beijing Sino-UK Institute of Biological Technology.

2.3. Experimental method

2.3.1. Selection of experimental animals

A total of 60 six-month-old healthy meat sheep with initial body weight of 39.9 ± 3.2 kg were divided into 4 groups of 15 sheep, which included the control group, test group 1, test group 2 and test group 3.

2.3.2. Experimental diet

The control group was fed a basal diet, and the 3 test groups were supplemented with 11, 22, and 33 mg/kg flavonoids from *Allium mongolicum Regel*. According to the stability test of flavonoids from *Allium mongolicum Regel* in the rumen, adding 22 mg/kg

of total flavonoids from Allium mongolicum Regel to the rumen could improve parameters related to rumen microbial fermentation gas production, pH, ammonia nitrogen, microbial protein, and volatile fatty acids and could optimize the effects of the flavonoids (Bao, 2015). This study referred to these previous results when choosing the supplementation doses (11, 22, and 33 mg/kg) of flavonoids from Allium mongolicum Regel. The components of the experimental diet testing method (Zhang, 2003) were as follows: 1) The crude protein of the feedstuff was tested using the Kjeldahl Nitrogen Determination method; 2) The Van Soest detergent fibre analysis method was used for the determination of neutral detergent fibre (NDF) and acid detergent fibre (ADF) contents; 3) The calcium content of feedstuff was tested using potassium permanganate titration; 4) The phosphorus content was determined using colorimetry with molybdenum yellow. The basic dietary composition and nutritional levels are shown in Table 1.

2.3.3. Feeding management

The entire experimental period lasted 75 d, and the adaptation period lasted 15 d. Parasites were expelled, and disinfection and epidemic prevention efforts were applied to the experimental sheep. The normal experimental period was 60 d, during which feed was given at 06:00 and 18:00, and water was given *ad libitum*. The ratio of the roughage to concentrate was 7:3. The control group was given the basal diet, and the test groups were given the appropriate experimental diet.

2.3.4. Sample collection and treatments

During the experimental period, feed offered and refusals were recorded, and sheep was weighted before feeding at 0, 15, 30, 45, and 60 d, which were prepared to calculate the performance index. Meanwhile, sterile blood samples from the jugular veins of the sheep were bottled into 5-mL tubes containing no anti-coagulant. Blood samples were stood for 40 min, and centrifuged by an $800 \times g$ vertical centrifuge for 10 min. After sera were separated out, samples were stored at -20 °C and prepared for hormones testing.

Table	1

Composition and nutrient levels of the basal diets (air-dry basis).

Item	Content
Ingredient, %	
Chinese wild rye grass hay	32.00
Alfalfa	17.80
Corn	23.00
Wheatbran	2.87
Sunflower meal (expeller)	16.92
Pea plant	2.45
Pomace	2.45
CaHPO ₄	0.72
NaCl	0.79
Premix ¹	1.00
Total	100.00
Nutrient levels, %	
DE, MJ/kg ²	16.88
CP	17.21
NDF	65.01
ADF	32.59
Ca	1.39
Р	0.51

DE = digestible energy; CP = crude protein; NDF = neutral detergent fibre; ADF = acid detergent fibre.

¹ The premix provides the following nutrients per kilogramme: 25 mg Fe (as ferrous sulphate), 29 mg Zn (as zinc sulphate), 8 mg Cu (as copper sulphate), 30 mg Mn (as manganese sulphate), 0.04 mg I (as potassium iodide), 0.1 mg Co (as cobalt sulphate), 3,200 IU vitamin A, 1,200 IU vitamin D₃, 20 IU vitamin E.

² Digestible energy is a calculated (Zhang, 2003), while the others are measured values.

2.3.5. Determination index

The following variables were measured: ADG, average daily intake, average feed conversion ratio, growth hormone (GH), insulin-like growth factor-1 (IGF-1), corticosterone (CORT), and adrenocorticotropic (ACTH).

2.4. Statistical analysis

Statistical analyses were performed using the method for double-factor variance analysis GLM in SAS9.0. The values are given as means \pm standard deviations. The significance level of 0.05 was adopted.

3. Results

3.1. Effect of flavonoids from Allium mongolicum Regel on growth performance

The growth performance included ADG, average daily feed intake and feed conversion ratio.

3.1.1. Average daily gain

The results indicated the *F* value of experimental time was 2.97, which had an obvious effect on ADG (P = 0.03). The results show after 30 d of feeding, ADG changed significantly (P < 0.05). However the F value of the interaction effect between experimental time and treatment was 0.95, so there were no significant interaction effects between them (P = 0.48). Table 2 shows the flavonoids from Allium mongolicum Regel had a clear effect on ADG. The ADG of the sheep that consumed the diet supplemented with flavonoids for 15 d showed no significant change (P > 0.05) compared with the control group. However, after 30 d, the ADG of the 3 test groups showed an increasing trend, and far surpassed that of the control group (P < 0.05). After 45 d, the ADG in test group 3 was significantly higher than that of the control group (P < 0.05), but the ADG did not vary significantly among the experimental groups (P > 0.05). Between d 45 and 60, the flavonoids from Allium mongolicum Regel continued to increase the ADG, but increased rate was lower in test group 3. At d 60, ADG of the 3 test groups was much higher than that of the control group (P < 0.05); the ADG of test group 2 was the highest, but it did not differ significantly from the ADG of the other 2 test groups (P > 0.05).

3.1.2. Average daily feed intake

The analysis showed the experimental time and treatment had an interaction effect, and the *F* value was 12.34. Table 2 shows flavonoids from *Allium mongolicum Regel* could improve the daily feed intake (DFI) of sheep. After 15 d of feeding, the DFI did not differ among groups, nor did the test groups exhibit any significant alteration (P > 0.05) compared with the control group. However, starting at d 30, the DFI of all groups gradually changed; the DFI of test group 3 was the highest and significantly higher than that of the control group (P < 0.05). After d 45, the DFI of the 3 test groups showed an increasing trend but no big difference (P > 0.05), and their DFI were higher than that of the control group. At d 60, the DFI of all groups showed increases, but those of 3 test groups were significantly higher than that of the control group (P < 0.05). At d 60, the DFI of test group 3 was the highest, but it did not differ significantly from that of other 2 test groups (P > 0.05). Thus, when the experimental time was 60 d, the basal diet supplemented with 33 mg/kg flavonoids groups plays an important role to the DFI.

3.1.3. Feed conversion ratio

The results showed the experimental time and treatment had an interaction effect, and the *F* value was 3.1. Table 2 indicates after d 15. basal diet supplement with flavonoids from Allium mongolicum Regel had no obvious effect on the feed conversion ratio (P > 0.05). After d 30, the feed conversion ratios of the 3 test groups decreased and were significantly lower than that of the control group (P < 0.05). At d 45, the feed conversion ratios of all groups continued to decrease, with the same difference among groups, and the feed conversion ratio of test group 2 was the lowest (P < 0.05). From d 45 to 60, the feed conversion ratios of the 3 test groups were significantly lower than that of the control group (P < 0.05). The feed conversion ratio of test group 3 was lower than that of test group 2, by an insignificant amount (P > 0.05), and was significantly lower than that of test group 1 (P < 0.05). The flavonoids from Allium mongolicum Regel affected the feed conversation ratio from d 30, and the effect of test group 3 was the best, until d 60, the feed conversion ratio in test group 3 was the lowest.

3.2. Serum GH, IGF-1, ACTH and CORT contents in sheep

3.2.1. Serum GH levels

At d 45, the content of serum GH was significantly changed (P < 0.05), and increased until d 60 when the content was the highest. But the results indicated the experimental time and treatment did not have interaction effect (F = 1.48). Thus, Table 3 is the analysis of treatment; it shows at d 0, the GH levels of the control group and test groups showed no obvious change (P > 0.05). At d 15, the GH content of test group 1 was higher than that of the control group (P < 0.05), but no significant difference among test groups (P > 0.05). At d 30, the GH content of the control group increased but did not differ significantly from those of other groups (P > 0.05). After d 45, the GH contents of all 3 test groups were higher than that of the control group (P < 0.05), but values of test group 2 and 3 did not differ significantly (P > 0.05). After d 60, the GH contents of test groups 1 and 2 were higher than that of the

Table 2

Effect of flavonoids from Allium mongolicum Regel on the average daily gain, the daily feed intake and the feed-to-gain ratio in sheep.

Item	Groups	15 d	30 d	45 d	60 d
Average daily gain, g/d	Control	150.67 ± 56.77	194.22 ± 82.37 ^b	218.67 ± 169.43 ^b	226.22 ± 67.48^{b}
	Test group 1	156.89 ± 50.35	226.67 ± 65.37^{a}	259.56 ± 50.92^{ab}	311.56 ± 60.56^{a}
	Test group 2	126.22 ± 52.69	250.22 ± 75.60^{a}	279.56 ± 74.34^{ab}	318.22 ± 61.61^{a}
	Test group 3	117.33 ± 53.65	239.56 ± 63.73^{a}	307.56 ± 119.45^{a}	289.33 ± 47.44^{a}
Daily feed intake, g/d	Control	1,148.59 ± 50.36	$1,148.15 \pm 42.64^{b}$	1,243.45 ± 87.79	$1,408.63 \pm 103.07^{b}$
	Test group 1	1,151.42 ± 43.50	1,107.26 ± 45.79 ^c	1,308.34 ± 146.97	1,563.26 ± 53.91 ^a
	Test group 2	$1,156.84 \pm 40.48$	$1,139.32 \pm 42.89^{b}$	1,305.51 ± 122.38	$1,551.72 \pm 68.14^{a}$
	Test group 3	1,183.79 ± 57.95	$1,191.69 \pm 40.99^{a}$	1,337.91 ± 157.64	$1,593.47 \pm 64.06^{a}$
Feed-to-gain ratio	Control	8.058 ± 0.701	6.227 ± 0.472^{a}	5.912 ± 0.227^{a}	5.686 ± 0.416^{a}
	Test group 1	8.392 ± 0.333	5.018 ± 0.179^{b}	4.885 ± 0.209^{b}	5.041 ± 0.586^{b}
	Test group 2	8.379 ± 0.788	4.876 ± 0.222^{b}	4.553 ± 0.177 ^c	4.669 ± 0.453^{bc}
	Test group 3	8.307 ± 0.688	5.507 ± 0.229^{b}	4.974 ± 0.177^{b}	$4.348 \pm 0.531^{\circ}$

^{a-c} Within a column, means without a common superscript differ at P < 0.05, n = 15.

Table 3

Effect of flavonoids from Allium mongolicum Regel on the serum growth hormone (GH) content, insulin-like growth factor-1 (IGF-1) content, corticosterone (CORT) content and adrenocorticotropic hormone (ACTH) content in sheep.

Item	Groups	0 d	15 d	30 d	45 d	60 d
GH, ng/mL	Control	4.29 ± 0.59	4.26 ± 0.54^{b}	4.50 ± 0.48	$4.44 \pm 0.68^{\circ}$	4.38 ± 0.87^{c}
	Test group 1	4.31 ± 0.73	4.75 ± 0.54^{a}	4.78 ± 0.62	5.05 ± 0.64^{ab}	5.49 ± 0.86^{ab}
	Test group 2	4.41 ± 0.67	4.54 ± 0.58	4.75 ± 0.77	4.87 ± 0.61	5.60 ± 0.95^{ab}
	Test group 3	4.43 ± 0.52	4.34 ± 0.48	4.36 ± 0.45	4.80 ± 0.29	5.05 ± 0.91
IGF-1, ng/mL	Control	190.18 ± 15.16	191.83 ± 6.50	191.65 ± 9.87	191.14 ± 8.15^{b}	191.36 ± 11.91 ^b
	Test group 1	190.21 ± 12.76	192.48 ± 9.96	199.73 ± 11.77	202.55 ± 12.77^{a}	202.18 ± 10.76^{a}
	Test group 2	188.96 ± 12.78	190.93 ± 11.32	199.30 ± 8.79	196.10 ± 9.03	205.38 ± 9.44^{a}
	Test group 3	190.75 ± 13.18	191.07 ± 10.74	199.89 ± 12.68	203.79 ± 13.55^{a}	206.78 ± 7.95^{a}
CORT, ng/mL	Control	59.67 ± 6.32	58.59 ± 8.08	60.41 ± 5.99^{a}	54.12 ± 5.82	54.02 ± 4.02
	Test group 1	61.43 ± 6.71	61.98 ± 7.28	56.89 ± 7.71^{ab}	51.47 ± 5.89	51.69 ± 7.08
	Test group 2	62.74 ± 6.99	60.75 ± 5.54	53.50 ± 6.81^{b}	51.59 ± 5.39	51.13 ± 8.01
	Test group 3	62.13 ± 4.72	60.78 ± 7.12	58.75 ± 5.23^{ab}	51.77 ± 8.09	51.08 ± 8.21
ACTH, pg/mL	Control	23.80 ± 2.70	24.04 ± 1.96	24.67 ± 1.65	25.82 ± 2.36	24.95 ± 4.27
	Test group 1	23.64 ± 2.15	24.76 ± 2.6	25.86 ± 2.09	26.00 ± 2.86	27.82 ± 2.66
	Test group 2	24.16 ± 1.28	25.75 ± 2.59	25.71 ± 2.45	26.85 ± 4.45	27.58 ± 3.30
	Test group 3	23.86 ± 3.35	23.92 ± 3.60	24.40 ± 3.59	26.52 ± 3.77	26.87 ± 3.78

^{a-c} Within a column, means without a common superscript differ at P < 0.05, n = 15.

control group (P < 0.05), but there was no significant difference between groups in the amount of GH change (P > 0.05). The GH content showed an increasing trend with feeding time, and after d 45, the GH content showed significant changes (P < 0.05). Although there were no interaction effect on the experimental time and treatment, but at d 60, the test group 2 exerted its' best effect on the content of GH levels.

3.2.2. Serum IGF-1 content

The content of serum IGF-1 was affected by experimental time. Result shows from d 30 the serum content was changed (P < 0.05). And the results indicated the experimental time and treatment did not have interaction effect (F = 0.72). Table 3 clearly shows at d 0, the addition of flavonoids from Allium mongolicum Regel to the basal diet had no obvious effect on the serum IGF-1 content, and the control group and all test groups did not show significant differences (P > 0.05). At d 15, the IGF-1 content increased in all test groups with no significant difference (P > 0.05); furthermore, the increase since d 0 was not significant (P > 0.05). After d 30, the IGF-1 content of the 3 test groups increased with no significant changes compared with that of the control group (P > 0.05). Forty-five days later, the IGF-1 contents of test groups 1 and 3 became significantly higher than that of the control group (P < 0.05) and nonsignificantly higher than that of test group 2 (P > 0.05). The IGF-1 contents of 3 groups were significantly higher than that of the control group at d 60 (P < 0.05); test group 3 had the highest IGF-1 levels at this time point, but with no significant difference (P > 0.05). The IGF-1 content was the highest after 60 d of feeding and showed significant changes (P < 0.05) compared with the contents at d 0 and 15. But there were no interaction effect between the experimental time and treatment, thus the basal diet supplemented with 33 mg/ kg flavonoids from Allium mongolicum Regel and fed 60 d would play the best role in the secretion of serum IGF-1.

3.2.3. Serum CORT content

Result shows at d 30 of feeding, the content of serum CORT level was changed obviously (P < 0.05), and at d 45 and 60, the content was higher than values at d 0, 15 and 30 (P < 0.05). Table 3 shows the basal diet with flavonoids from *Allium mongolicum Regel* did not lead to variations in the serum CORT content (P > 0.05) at d 0. After 15 d of feeding, the CORT contents of all test groups decreased, and test groups 2 and 3 showed the lowest levels; however, there was no significant difference between any of the test groups and the control group (P > 0.05). At d 30, all test groups still showed a decreasing trend, and the CORT content of test group 2 was

significantly lower than that of the control group (P < 0.05) and non-significantly lower than those of the other 2 test groups (P > 0.05). At d 45, all of the CORT contents of the 3 test groups and the control group decreased; the CORT contents of the 3 test groups were lower than that of the control group with no significant difference (P > 0.05). At d 60, the CORT contents of the 3 test groups were lower than that of the control group, still not to a significant degree (P > 0.05). These findings indicate the flavonoids from *Allium mongolicum Regel* could decrease the serum CORT contents in sheep. At d 60, the content of serum CORT in 3 test groups was the highest.

3.2.4. Serum ACTH content

Results indicated feeding time affected the serum ACTH levels from d 30 (P < 0.05), and the contents at d 45 and 60 were higher than the level at d 30 (P < 0.05). And the content at d 60 was the highest (P < 0.05). However, experimental time and treatment had no interaction effect (F = 0.48). Table 3 shows at d 0, consumption of basal diet supplemented with flavonoids from Allium mongolicum Regel did not have significant effect on the ACTH content (P > 0.05). At d 15, the serum ACTH content was higher than that at d 0, but there was no significant difference between the control and test groups (P > 0.05); furthermore, although test group 2 had the highest ACTH content at this time point, the differences among the test groups were not significant (P > 0.05). At d 30, the ACTH contents of all groups were higher than those at d 0 and 15; despite the obvious increasing trend, the change was still not significant (P > 0.05). The ACHT contents of test groups 1 and 2 increased with no significant difference (P > 0.05). At d 45, the ACHT contents of 3 test groups were higher than that of the control group; although test group 2 had the highest ACHT levels, it did not differ significantly from other 3 groups (P > 0.05). At d 60, the ACTH contents of test groups 1 and 2 were the highest, but the difference was not significant (P > 0.05). These results showed the flavonoids from Allium mongolicum Regel could increase the serum ACTH content of sheep, but there was no significant difference among the groups (P > 0.05).

4. Discussions

4.1. Effect of flavonoids from Allium mongolicum Regel on growth performance in meat sheep

Flavonoids are a type of plant oestrogen that improves growth in animals. Many studies have examined animal growth performance. Xue (2000) found that a basal diet with the appropriate amount of daidzein could increase the daily gain of pigs and decrease the feed conversion ratio. Zhou et al. (2004) confirmed that a basal diet supplemented with 5 mg/kg daidzein could significantly increase the laying rate of *Shaoxing* ducks. Cheng et al. (2005) found that basal diets supplemented with daidzein could improve the ADG and decrease the feed conversion ratio of fatting pigs. The present study found that a basal diet supplemented with flavonoids at levels from 11 to 33 mg/kg from *Allium mongolicum Regel* could increase the ADG and average feed intake and decrease the feed conversion ratio of meat sheep. This result was similar to previous findings and proved that flavonoids from *Allium mongolicum Regel* could improve sheep's growth performance and increase meat production.

4.2. Effect of flavonoids from Allium mongolicum Regel on the serum GH, IGF-1, ACTH and CORT content in sheep

Body hormones regulate the immune function through neuroendocrine pathways to improve the disease resistance of animals, thus playing an important role in animal production and immune function. This knowledge suggests a promising study direction (Draper et al., 1997). Growth hormone is a type of peptide hormone, a protein secreted by pituitary cells. The function of animal GH is mediated by IGF-1 in animal bodies. Serum IGF-1, as an animal growth regulator, is closely related to animal growth (Hossner et al., 1997). Growth hormone is a core aspect of animal growth that can promote the growth of body tissue and increase the decomposition of protein and fat. Numerous studies have shown that flavonoids can increase GH levels in the body. Liu et al. (1999) found that daidzein could significantly increase the GH content in the serum and colostrum of sows. Jiang et al. (2012) proved that a broiler chicken basal diet supplemented with 20 mg/kg of isoflavones from Trifolium pratense could increase the serum GH content resulting in a significant change, and a basal diet supplemented with isoflavones at 10 and 30 mg/kg could also increase the serum GH content, thus promoting the growth of broiler chickens. Animals' cellular amino acid intake and utilization is increased by IGF-1. Li et al. (2008a) found that sea buckthorn flavones could increase the serum IGF-1 content of broiler chickens.

The animal growth process is greatly affected by environmental factors, nutrition, age and physiological function in addition to genes and neuroendocrine hormones. Growth hormone secreted from the hypothalamus can act directly on animal target organs and secretes IGF-1 and receptors. Insulin-like growth factor-1 promotes body cell proliferation, regulates protein synthesis, and contributes to animal tissue and organ development and bone growth (Liu et al., 2003; Wang et al., 1999). The stress of the feeding process can affect animals' growth performance. Under stress, animals sense the threat of homeostasis imbalance through the hypothalamic-pituitary-adrenal (HPA) axis; consequently, HPA axis excitability increases, and the stress results in the increase of small corticotrophin-releasing factor (CRF), which is distributed inside the par ventricular nucleus of the small nerve cells of the hypothalamus (Li et al., 2008b). Corticotrophin-releasing facto is then released into the anterior lobe of the pituitary gland through a portal, and the anterior lobe of the pituitary gland then releases ACTH, which activates adrenal cortical cells to synthesize and release CORT. The serum ACTH and CORT levels are important indicators of the impact of stress on an animal's body. Under normal conditions, CORT provides negative feedback inhibition along the HPA axis, thus maintaining body immunity. Jiang et al. (2004) showed that the total flavones of epimedium adjust the HPA axis in rats with kidney deficiency and improve it by controlling the expression of the calmodulin-like protein gene. In addition, Shen and Chen (2002) found that epimedium flavones and polysaccharides also promote the endocrine immune capacity. Animals under stress transfer their immunity and growth energy, which affects their disease resistance. Optimal nutrition or feed control could improve the animal's anti-stress defences and contribute to growth. The results of this study show that flavonoids from Allium mongolicum Regel could significantly increase serum GH and IGF-1 levels. Three different doses of flavonoids from Allium mongolicum Regel could have an increasing effect on GH. After d 60. feeding with flavonoids from Allium mongolicum Regel significantly increased the IGF-1 contents of all groups. Additionally, it increased the serum ACTH contents of all groups, but not to a significant degree. Adrenocorticotropic hormone is a key signal delivery factor in the HPA axis and could be involved in stress-induced intake reductions; however, our results indicated that increased ACTH did not inhibit feed intake, possibly because of the regulatory effect of the Allium mongolicum Regel flavonoid's regulation of HPA through the nervous system. The Allium mongolicum Regel flavonoids had an inhibitory effect on the serum CORT content, which, according to this research, might be related to the regulation of the inflammatory cytokines IL-1, IL-6, TNF-α (Cai et al., 1995).

The Allium mongolicum Regel flavonoids might improve the levels of body hormones via those cytokines' effects on the neuroendocrine system. The flavonoids from Allium mongolicum Regel have a promoting effect on the neuroendocrine hormones GH, IGF-1 and ACTH and an inhibitory effect on CORT. A basal diet with Allium mongolicum Regel flavonoids could regulate the HPA axis via the nervous system, thereby assuring the balance of the neuroendocrine system. Consequently, a basal diet supplemented with flavonoids from Allium mongolicum Regel could facilitate the animals' adjustment to their environment, increase their anti-stress ability, and promote the growth performance and immune function of meat sheep.

5. Conclusions

The optimum supplementation of flavonoids from *Allium mongolicum Regel*, which is from 11 to 33 mg/kg, could increase the ADG and average DFI, and decrease the feed conversion ratio in meat sheep; furthermore, it could promote increases in GH, IGF-1 and ACTH levels while inhibiting CORT levels. The results confirmed that the flavonoids from *Allium mongolicum Regel* regulate the growth performance of sheep via their effects on GH, IGF-1 and the HPA axis. This effect was especially evident after 30 d of feeding, indicating that it worked in a time-dependent manner. In future, flavonoids from *Allium mongolicum Regel* must be well-mixed in basal diet, and it needs to solve the problem of the production costs.

Acknowledgements

This work was funded by the National Natural Science Foundation of China (31260558; 31160474); the National Science & Technology Pillar Program during the Twelfth Five-year Plan Period (2013BDA10B04). We express our appreciation to the post graduate students of Animal Science College of Inner Mongolia Agricultural University for their help with the collection of the experimental samples.

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