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Effects of daidzein on growth performance, blood metabolites and meat quality of finishing Xianan beef cattle

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Abstract

An experiment was conducted to determine the effects of supplementing different amounts of daidzein in a diet on the growth performance, blood biochemical parameters and meat quality of finishing beef cattle. Thirty finishing Xianan steers were distributed in three groups equilibrated by weight and fed three different dietary treatments (concentrate ratio = 80%): (1) control; (2) 500 mg/kg daidzein and (3) 1000 mg/kg daidzein, respectively. Steers were slaughtered after an 80-day feeding trial. Results showed that daidzein supplementation had no effect on the final body weight, average daily gain and feed conversion rate of steers. Steers fed with 1000 mg/kg daidzein had greater dry matter intake than those fed with control diets. Compared with the control group, the 1000 mg/kg daidzein group had a higher fat thickness, lower shear force and lightness. The pH, drip loss, cooking loss, redness (a^*), yellowness (b^*), moisture, ash, crude protein and intramuscular fat of the *Longissimus dorsi* muscle were unaffected by daidzein supplementation. Compared with the control group, the 1000 mg/kg daidzein group significantly increased the serum concentrations of insulin, free fatty acid and Glutamic-pyruvic transaminase. The 500 mg/kg daidzein group significantly increased the serum concentration of tetraiodothyronine compared with the control group. Supplemental daidzein did not affect the blood antioxidant ability and blood immune parameters in serum. In conclusion, daidzein supplementation above 500 mg/day modifies feed intake and metabolic and hormonal profile, with positive and negative effects on meat quality.

Introduction

Phytoestrogens are widely found in soybeans, alfalfa and other plant feedstuffs. Daidzein is the main component of soybean isoflavones (Franke *et al.*, 1994; Liggins *et al.*, 2002). Due to its chemical structure, similar to mammalian oestrogens, daidzein displays weak oestrogenic activity in animals and has been studied extensively for possible biological activities such as antioxidant, anti-stress, modulating glucolipid metabolism and promoting animal growth (Wong *et al.*, 2008; Liu *et al.*, 2012). Researchers have shown that daidzein improves the feed efficiency of chicken, castrated piglets and weaned piglets (Wang *et al.*, 1994; Guo *et al.*, 2002; Li *et al.*, 2015), increases the daily weight gain of chicken, castrated piglets and Northeast fine wool sheep (Guo *et al.*, 2002; Ren *et al.*, 2009; Li *et al.*, 2015) and elevates milk production, milk protein content, milk fat content and serum antioxidant capacity of dairy cows (Liu *et al.*, 2013). In beef cattle, research has focused mainly on the effect of daidzein on rumen fermentation characteristics (Zhao *et al.*, 2018). In a previous study, supplementation with daidzein changed the bacterial community composition and enhanced the rumen fermentation function of beef cattle (Liang *et al.*, 2018).

As a result of rapid economic development, the demand for high-quality beef has grown in popularity in China. Improving the production and quality of beef has become an urgent issue for the Chinese beef industry. Daidzein has been reported to be effective at affecting the meat quality of animals, but a few animal studies published have given conflicting results. Zhao *et al.* (2015) found that daidzein supplementation improved the tenderness of beef by increasing the intramuscular fat (IMF) content and marbling score. On the other hand, Rehfeldt *et al.* (2007) reported that supplemental daidzein changed the muscle fibre type in piglets. By measuring the IMF content and tenderness of beef, the current research was conducted to clarify the controversial results.

Xianan cattle (*Bos taurus*) is the first cattle breed developed especially for beef in China (Ru *et al.*, 2006). Xianan cattle are crossbreeds of Nanyang cattle (female parent) and Charolais cattle (male parent) which have the characteristics of fast growth, high-meat quality, coarse feedstuff tolerance and strong adaptability (Wang *et al.*, 2007, 2008). However, few

investigations have focused on the meat quality of Xianan cattle. Moreover, there are no reports about the effect of daidzein on growth performance, blood biochemical parameters and meat quality of finishing Xianan cattle. Besides, the supplemental dosages of daidzein in previous studies were below 500 mg/kg. A higher dosage (1000 mg/kg) of daidzein has not been tested. Therefore, the purpose of the current study was to evaluate the effects of different dosages of daidzein on growth performance, blood biochemical parameters and meat quality of finishing Xianan cattle, and explore the mechanisms of daidzein on improving meat quality.

Materials and methods

Animals, diets and experimental design

Thirty 2-year-old finishing Xianan steers (body weight (BW) = 686 ± 50.9 kg) were used in an 80-day feeding study during the finishing stage. These steers were segregated by live body weight (LBW) and then divided into three experimental groups using a randomization procedure to ensure that groups were equilibrated by LBW (similar number of animals from each weight group in experimental groups). Each group randomly received one of three dietary treatments (concentrate ratio = 80%): (1) control; (2) 500 mg/kg daidzein (500 mg daidzein/kg concentrate) and (3) 1000 mg/kg daidzein (1000 mg daidzein/kg concentrate). The cattle were untethered in individual stalls whose floors were paved with brick and fed with an 800 g/kg concentrate diet in quantities sufficient to provide *ad libitum* consumption (Table 1). For daidzein treatments, daidzein was mixed into the mineral premix and added to the concentrate. Diets were offered to the cattle twice daily at 07.00 and 14.00 h. Fresh water was available for *ad libitum* consumption throughout the study.

Growth performance

Amount of feed offered and refused were recorded daily for each steer. The feed intake was calculated daily for each steer as the dry matter (DM) offered minus DM refused. Steers were weighed for two consecutive days at the start and end of the experiment. The average daily gain (ADG) was determined for the overall experiment by subtracting initial BW from BW at the end of the experiment and dividing by the number of days. Feed conversion rate (FCR) was determined as the ratio of dry matter intake (DMI) to ADG.

Carcass characteristics

Steers were slaughtered at a commercial abattoir following the standard procedures. The carcasses were then put into a chiller at 0 °C and aged for 14 days. The *Longissimus dorsi* muscle (LM) area and 12th-rib fat thickness were obtained for each carcass after ageing. The ribeye area was obtained by tracing the transverse section of each LM on sulphate paper, and then calculated by the volume-curve method using Leica QWin software. Fat thickness over the LM was determined by means of vernier callipers (triplicate in each carcass).

Physicochemical analysis

The meat quality was measured from an LM sample removed from the 12th and 13th ribs on the right side of the carcass,

Table 1. Basic diet composition (daidzein was included according to treatment)

Item	Inclusion levels (g/kg as fed)
Diet ingredient	
Peanut vine	200
Maize	442
Barley	282
Soybean meal	20
Soybean oil	14
Baking soda	7
Salt	7
Mineral-vitamin pre-mix ^a	28
Chemical composition	
DM	851.0
Crude protein	111.0
NDF	175.2
ADF	104.2
Ether extract	35.0
Daidzein	0.0066

^aMineral-vitamin pre-mix (per kg): vitamin A, 250 000 IU; vitamin D3, 30 000 IU; vitamin E, 800 IU; Cu, 1 g; Fe, 5 g; Mn, 4 g; Zn, 3 g; Se, 10 mg; I, 50 mg; Co, 10 mg.

with no connective tissue or subcutaneous fat. The pH value was determined by using a Delta 320pH Meter (Mettler Toledo, Greifensee, Switzerland) with the probe inserted into the middle of the LM. Meat colour values of L^* , a^* and b^* were measured by using a colorimeter (WSC-S, Shanghai, China) with an illuminant D65, 10° observer and CIELAB system.

To determine the cooking loss and shear force values of LM, steaks ($3 \times 4 \times 5$ cm) were dissected from the LM sample, placed in package bags and subsequently cooked in a 75 °C water bath with automatic temperature control until the internal temperature reached 70 °C. Then, the cooked beef was cooled naturally to room temperature. Cooking losses were calculated as the difference between initial and cooked weight as a percentage of initial weight. Six beef pieces of $3 \times 1 \times 1$ cm (length \times width \times height) were removed parallel to the muscle fibre direction and cut with a Warner-Bratzler shear device (C-LM4, Haerbin, China) by running it perpendicularly in the muscle fibre direction. The shear force was expressed as the mean of six shear force values (kg f). To determine the drip losses of LM, the samples were covered with valve bags to avoid evaporation and placed in a container at 4 °C for 48 h. The samples were kept away from contacting the container and with the muscle fibre direction parallel to gravity, and drip losses were calculated as the difference between initial and refrigerated weight as a percentage of initial weight.

Determination of chemical composition of the basal diet and LM was carried out following the procedures of AOAC methods (AOAC, 2000). Briefly, the daidzein content of the basal diet was determined by high performance liquid chromatography (HPLC; LC-10A; Shimadzu, Tokyo, Japan) as described by Vyn *et al.* (2002). DM content was determined by drying feed and meat samples at 105 °C to constant weight. Neutral detergent fibre (NDF) and acid detergent fibre (ADF) contents were determined by using an automatic fibre analyser (Ankom 2000i full;

Ankom Technology Corporation, Macedon, NY, USA) (Van Soest *et al.*, 1991). Ether extract content was determined by Soxhlet extraction with diethyl ether, preceded by an acid hydrolysis treatment when analysing meat samples. Ash content was determined by combustion at 550 °C for 5 h. Crude protein content of meat was measured by the Kjeldahl nitrogen method.

Determination of blood biochemical parameters

At the end of the experiment, the feed was removed from the stalls and blood samples were collected from each steer at 07.00 h, 24 h before slaughter, via jugular venepuncture using 5 ml capacity evacuated blood-collecting tubes (YL003, Nanjing, China). After coagulation at room temperature for 30 min, the blood samples were centrifuged at 3000 g for 10 min at 4 °C. The serum samples were divided into tubes and stored at –20 °C for analysis.

Blood biochemical parameters including total cholesterol (TC), triglyceride (TG), free fatty acid (FFA), glucose, total protein (TP), albumin, high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), urea, glutamic-pyruvic transaminase (ALT), glutamic-oxaloacetic transaminase (AST), total superoxide dismutase (T-SOD), malondialdehyde (MDA), glutathione peroxidase (GSH-PX), total antioxidant capacity (T-AOC), immunoglobulin (Ig)A, IgM, IgG, triiodothyronine (T3), tetraiodothyronine (T4), growth hormone (GH), insulin, leptin and adiponectin were determined by assay kits (Table 2). All the assay kits were purchased from the same biotech company (Jiancheng, Nanjing, China). All measurements were operated in an AU5421 Automatic Biochemistry Analyzer (Beckman-Kelt, USA) at the First Affiliated Hospital of Nanchang University.

Statistical analysis

Differences between means were tested for statistical significance with one-way analysis of variance. The least significant difference test was conducted to determine the significance level of the particular comparison between treatment means. All statistical procedures were performed with SPSS 17.0 for Windows (SPSS Inc., Chicago, IL, USA). Significance was declared at $P \leq 0.05$. The model including random and fixed effects was as follows:

$$Y_{ij} = \mu + T_j + E_{ij} \quad (1)$$

where Y_{ij} is the dependent variable, μ is the overall mean, T_j is the fixed effect of daidzein ($j = 1-3$) and E_{ij} is the error term.

Results

Intake and growth performance

Steers fed with the 1000 mg/kg daidzein diet had greater DMI than those fed with the control diets ($P = 0.025$; Table 3). However, the final BW, ADG and FCR did not differ among three groups.

Carcass characteristics and meat quality

Compared with the control steers, fat thickness was greater for steers fed with 1000 mg/kg daidzein diet ($P = 0.042$; Table 4). The shear force ($P = 0.038$) and L^* ($P = 0.026$) of LM in steers fed with 1000 mg/kg daidzein were significantly lower than those fed with control diets. No difference in LM area, pH, drip loss, cooking loss, a^* or b^* of LM were observed among the three groups.

Table 2. The assay kits for blood biochemical parameters

Blood biochemical parameter	Assay kit ^a
TC	TC assay kit
TG	TG assay kit
FFAs	FFA assay kit
Glucose	Glucose assay kit
TP	TP assay kit
Albumin	Albumin assay kit
HDL-C	HDL-C assay kit
LDL-C	LDL-C assay kit
Urea	Urea assay kit
ALT	ALT assay kit
AST	AST assay kit
T-SOD	T-SOD assay kit
MDA	MDA assay kit
GSH-PX	GSH-PX assay kit
T-AOC	T-AOC assay kit
IgA	IgA assay kit
IgM	IgM assay kit
IgG	IgG assay kit
T3	T3 assay kit
Thyroxine	Thyroxine assay kit
GH	GH assay kit
Insulin	Insulin assay kit
Leptin	Leptin assay kit
Adiponectin	Adiponectin assay kit

^aAll assay kits were purchased from the same biotech company (Jiancheng, Nanjing, China).

Supplementation with daidzein did not affect the moisture, ash, crude protein or IMF of LM (Table 4).

Blood biochemical parameters

Daidzein supplementation at 1000 mg/kg significantly increased the concentrations of FFA ($P = 0.002$) and ALT ($P = 0.024$), by 164.8 $\mu\text{mol/l}$ and 11.5 U/l, respectively, compared with the control group (Table 5). Supplemental daidzein did not affect the concentrations of TC, TG, glucose, TP, albumin, HDL-C, LDL-C, urea or AST.

Blood antioxidant ability and immune parameters

Daidzein supplementation did not affect the blood antioxidant ability including T-SOD, MDA, GSH-PX and T-AOC (Table 6), or blood immune parameters including IgA, IgM and IgG.

Blood hormone parameters

The concentration of T4 was 36% higher with 500 mg/kg daidzein than the control ($P = 0.016$; Table 7). The concentration of insulin was higher for steers fed with 1000 mg/kg daidzein in comparison

Table 3. Effects of supplemental daidzein on growth performance in finishing steers

Items	Daidzein supplementation levels (mg/kg)			S.E.M. ^a	P-Value
	0	500	1000		
Initial BW (kg)	686	688	686	9.6	0.796
Final BW (kg)	737	743	744	10.6	0.676
ADG (kg/day)	0.64	0.69	0.73	0.041	0.554
DMI (kg/day)	8.3	8.7	8.9	0.11	0.025
Forage intake (kg/day)	1.63	1.69	1.75	0.021	0.022
Concentrate intake (kg/day)	6.71	6.97	7.19	0.086	0.026
FCR	15	15	16	1.4	0.611

BW, body weight; ADG, average daily gain; DMI, dry matter intake; FCR, feed conversion rate.

^aS.E.M. = standard error of the mean ($n = 10$ steers per treatment).

Table 4. Effects of supplemental daidzein on meat quality of the *Longissimus dorsi* muscle

Items	Daidzein supplementation level (mg/kg)			S.E.M. ^a	P-Value
	0	500	1000		
Carcase characteristics					
<i>Longissimus</i> area (cm ²)	92	92	92	1.6	0.996
Fat thickness (cm)	1.37	1.54	1.83	0.086	0.042
pH	5.17	5.12	5.23	0.034	0.408
Shear force (kg f)	3.5	3.1	3.0	0.11	0.038
Drip loss (g/kg)	17.8	14.7	17.2	0.71	0.159
Cooking loss (g/kg)	312	325	348	7.9	0.198
<i>L</i> *	46.5	45.5	42.6	0.63	0.026
<i>a</i> *	22.6	22.6	22.1	0.30	0.803
<i>b</i> *	12.9	12.6	12.4	0.26	0.718
Chemical composition					
Moisture (g/kg)	686	682	690	3.7	0.679
Ash (g/kg)	18.9	19.0	19.4	0.31	0.746
Crude protein (g/kg)	210	210	215	4.8	0.887
IMF (g/kg)	84	88	85	4.1	0.947

*L**, lightness; *a**, redness; *b**, yellowness.

^aS.E.M. = standard error of the mean ($n = 10$ steers per treatment).

with the steers in the control group ($P = 0.043$). Concentrations of T3, GH, leptin and adiponectin did not differ among the three groups.

Discussion

To date, there are very few studies on the effects of daidzein supplementation in beef cattle diet and, in particular, limited information is available regarding the effects of daidzein on meat quality of beef cattle. Furthermore, in a few animal studies which have been published, the results of daidzein affecting meat quality have been contradictory. Since there are very few studies on the relationship between beef cattle and daidzein, this paper only compared the current results with those obtained from other animal species.

During high-grade beef production, feeding a high-concentrate diet continuously would undermine the health of finishing cattle, which might cause a reduction in feed intake and low efficiency in fat deposition (Brown *et al.*, 2000; Khafipour *et al.*, 2009). The results from the current study showed that daidzein supplementation at 1000 mg/kg increased the feed intake of steers, which was significant for high-grade beef production. The increase of feed intake did not affect ADG of steers but fat thickness increased significantly. These results could be related to changes in rumen fermentation parameters. In earlier studies, adding 1000 mg/kg daidzein significantly increased acetate content in the rumen (Liang *et al.*, 2018). As a precursor of adipose, acetate is conducive to lipogenesis in ruminants (Kristensen and Harmon, 2004).

Contrary to previous results, in which supplemental daidzein improved the capacities of antioxidant (Foti *et al.*, 2005; Mishra

Table 5. Effects of supplemental daidzein on blood biochemical parameters of finishing steers

Items	Daidzein supplementation levels (mg/kg)			S.E.M. ^a	P-Value
	0	500	1000		
TC (mmol/l)	2.8	3.2	3.4	0.18	0.294
TG (mmol/l)	0.54	0.57	0.39	0.069	0.637
FFA (μ mol/l)	421	439	586	23.0	0.002
Glucose (mmol/l)	11	9	8	1.5	0.770
TP (g/l)	73	80	79	4.0	0.785
Albumin (g/l)	26	27	27	1.1	0.965
HDL-C (mmol/l)	1.9	2.0	2.5	0.19	0.413
LDL-C (mmol/l)	0.78	1.01	0.95	0.081	0.469
Urea (mmol/l)	4.9	5.8	5.8	0.30	0.384
ALT (U/l)	25	31	36	1.9	0.024
AST (U/l)	79	90	86	4.2	0.628

TC, total cholesterol; TG, triglyceride; FFA, free fatty acid; TP, total protein; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; ALT, glutamic-pyruvic transaminase; AST, glutamic-oxaloacetic transaminase.

^aS.E.M. = standard error of the mean ($n = 10$ steers per treatment).

Table 6. Effects of supplemental daidzein on blood antioxidant ability and immune parameters of finishing steers

Items	Daidzein supplementation levels (mg/kg)			S.E.M. ^a	P-Value
	0	500	1000		
T-SOD (U/ml)	148.4	136.3	144.7	4.4	0.158
MDA (nmol/ml)	4.7	6.3	5.0	0.42	0.273
GSH-PX (μ mol/l)	913	1003	911	23.1	0.353
T-AOC (U/ml)	16	16	16	1.3	0.381
IgA (g/l)	0.94	0.93	1.13	0.078	0.585
IgM (g/l)	0.85	0.94	0.87	0.044	0.611
IgG (g/l)	8.4	9.3	9.2	0.27	0.320

T-SOD, total superoxide dismutase; MDA, malondialdehyde; GSH-PX, glutathione peroxidase; T-AOC, total antioxidant capacity.

^aS.E.M. = standard error of the mean ($n = 10$ steers per treatment).

et al., 2009; Liu *et al.*, 2013) and immunity (Morimoto *et al.*, 2009; Mohammad-Shahi *et al.*, 2011) in serum, these were not affected for finishing Xianan cattle in the current study. It should be noted that the regulative effect of daidzein in animals was influenced by several factors, such as supplementing amount of daidzein, animal breed, age, sex and methods of feed management (Lozupone *et al.*, 2012). In the current study, soybean meal in the basal diet could supply daidzein in significant amounts (Kašparovská *et al.*, 2017). Besides, soybean oil in the basal diet could supply di-*tert*-butylhydroxytoluene which is an antioxidant (Blanco *et al.*, 2017). These reasons might lead to a high antioxidant activity for the basal diet.

There are no consistent effects of daidzein on lipid metabolism in animals. Some researchers have shown that daidzein has a positive effect on fat deposition. Zhao *et al.* (2015) found that daidzein supplementation significantly increased the IMF content of cross-breed steers. Li *et al.* (2011) and Rehfeldt *et al.* (2007) reported that adding a high dose of daidzein significantly increased the fat proportion in carcasses of Guangxi minipig and piglets, respectively.

But there was also a viewpoint that supplemental daidzein inhibited the synthesis of lipids in animals. Szkudelska *et al.* (2002) and Choi *et al.* (2005) found that feeding daidzein reduced glucose uptake and inhibited the synthesis of TG in mouse cells. Liu *et al.* (2003) reported that supplementation with daidzein decreased the fat thickness and abdominal fat percentage of broilers. The results of the current study showed that daidzein supplementation significantly increased fat thickness and enhanced the synthesis of lipids in finishing Xianan cattle. This result may be caused by the weak oestrogenic activity of daidzein. Oestrogen could inhibit the β oxidation of fatty acids in the liver and lead to elevated serum concentrations of FFAs, which are precursors for the synthesis of lipids (Bauman *et al.*, 2011). Kadegowda *et al.* (2008) reported that adipose tissue increased the intake of FFAs and decreased the intake of β -hydroxybutyrate and acetic acid for synthesis of lipids with the increase of FFA concentrations in blood, which was consistent with the current study. Besides, there was an interaction between plasma FFA and insulin. It is known that plasma FFA could

Table 7. Effects of supplemental daidzein on blood hormone parameters of finishing steers

Items	Daidzein supplementation levels (mg/kg)			S.E.M. ^a	P-Value
	0	500	1000		
T3 (ng/ml)	2.6	3.1	2.6	0.11	0.128
T4 (ng/ml)	73	99	77	3.4	0.016
GH (ng/ml)	4.9	5.7	5.1	0.26	0.462
Insulin (ng/ml)	14	19	25	2.1	0.043
Leptin (ng/ml)	6.6	7.3	5.5	0.37	0.046
Adiponectin (ng/ml)	1.5	1.5	1.3	0.76	0.339

T3, triiodothyronine; T4, tetraiodothyronine; GH, growth hormone.

^aS.E.M. = Standard error of the mean ($n = 10$ steers per treatment).

inhibit insulin signalling in different physiological and metabolic processes, inducing insulin resistance. The increase of FFA content inhibited insulin's anti-lipolytic action and promoted the synthesis of lipids (Ovadia *et al.*, 2011).


As an important indicator to measure the tenderness of beef, shear force was affected by the IMF content and muscle fibre type of beef meat (Belew *et al.*, 2003; Koochmaraie and Geesink, 2006). Daidzein supplementation significantly decreased the shear force of LM in Xianan cattle. Considering that supplemental daidzein did not affect the IMF content of LM, the current study hypothesized that daidzein may change the muscle fibre type of LM to reduce the shear force. Due to the characteristic of weak oestrogenic activity, daidzein could regulate expression of the oestrogen receptor gene, which could affect the differentiation of marrow stromal cells (Zhou *et al.*, 2001) and change the muscle fibre type.

Meat colour plays an important role in purchasing decision and price (Liu *et al.*, 1995). Different meat colours are largely related to the relative proportions of metmyoglobin (grey-brown), myoglobin (dark red) and oxymyoglobin (bright red, the ferrous oxygenated form of myoglobin) (Kim *et al.*, 2009). There was a strong correlation between the content of myoglobin and L^* value in meat, and the L^* value decreased with the increase of myoglobin content (O'Keefe and Hood, 1982). In the current results, daidzein supplementation significantly decreased the L^* value of LM. The lowest values of L^* could be related to a lower proportion of white glycolytic fibres, which are known to be harder than oxidative ones (Chen *et al.*, 2012).

With respect to the dosage effect of daidzein, supplementing with high doses (1000 mg/kg) of daidzein in diet had a better effect than low dose (500 mg/kg). This result was consistent with studies by Li *et al.* (2011), Šošić-Jurjević *et al.* (2007) and Dang and Löwik (2004), in which daidzein supplementation at high doses had a better effect on enhancing the synthesis of lipids in swine, rat and mouse, respectively. However, the dosage of daidzein (1000 mg/kg) used in the current study was much higher than the supplemental dosage in Li *et al.* (2011) (500 mg/kg), Šošić-Jurjević *et al.* (2007) (3 mg/kg) and Dang and Löwik (2004) (30 mg/kg). The reason for those results might be that daidzein can be partly metabolized to equol by ruminal microorganisms (Lundh, 1995).

In conclusion, daidzein supplementation above 500 mg/day modifies feed intake and metabolic and hormonal profile, with

positive and negative effects on meat quality. Further studies should be conducted to optimize daidzein dose and to identify the modulating factors influencing the potential antioxidant activity of isoflavones.

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Ethical standards. All animal procedures such as ethical and animal welfare issues were approved by the ethics committee of Jiangxi Agricultural University. Daidzein was purchased from Ci Yuan Biotechnology Co., Ltd. (purity >98%, Shanxi, China).

References

- AOAC (Association of Official Analytical Chemists) (2000) *Official Methods of Analysis*, 17th Edn. Arlington, VA, USA: Association of Official Analytical Chemists.
- Bauman DE, Harvatine KJ and Lock AL (2011) Nutrigenomics, rumen-derived bioactive fatty acids, and the regulation of milk fat synthesis. *Annual Review of Nutrition* **31**, 299–319.
- Belew JB, Brooks JC, McKenna DR and Savell JW (2003) Warner-Bratzler shear evaluations of 40 bovine muscles. *Meat Science* **64**, 507–512.
- Blanco C, Giraldez JF, Morán L, Mateo J, Villalobos-Delgado LH, Andrés S and Bodas R (2017) Effects of sunflower soap stocks on light lamb meat quality. *Journal of Animal Science* **95**, 3455–3466.
- Brown MS, Krehbiel CR, Galyean ML, Rummenga MD, Peters JP, Hibbard B, Robinson J and Moseley WM (2000) Evaluation of models of acute and subacute acidosis on dry matter intake, ruminal fermentation, blood chemistry, and endocrine profiles of beef steers. *Journal of Animal Science* **78**, 3155–3168.
- Chen JY, Niu L, Huang M and Zhou GH (2012) Major bio-factors affecting beef color stability. *Scientia Agricultura Sinica* **45**, 3363–3372.
- Choi MS, Jung UJ, Kim MJ, Kim JY, Park SY, Jang JY and Lee MK (2005) Effect of genistein and daidzein on glucose uptake in isolated rat adipocytes; comparison with respective glycones. *Journal of Food Science and Nutrition* **10**, 52–57.
- Dang Z and Löwik CW (2004) The balance between concurrent activation of ERs and PPARs determines daidzein-induced osteogenesis and adipogenesis. *Journal of Bone and Mineral Research* **19**, 853–861.
- Foti P, Erba D, Riso P, Spadafranca A, Criscuoli F and Testolin G (2005) Comparison between daidzein and genistein antioxidant activity in primary and cancer lymphocytes. *Archives of Biochemistry and Biophysics* **433**, 421–427.
- Franke AA, Custer LJ, Cerna CM and Narala KK (1994) Quantitation of phytoestrogens in legumes by HPLC. *Journal of Agricultural and Food Chemistry* **42**, 1905–1913.
- Guo H, Han Z and Wang GJ (2002) Effect of supplementation of daidzein in diet on the performance and related endocrine secretion in castrated piglets. *Chinese Journal of Animal Science* **38**, 17–18.
- Kadegowda AKG, Piperova LS, Delmonte P and Erdman RA (2008) Abomasal infusion of butterfat increases milk fat in lactating dairy cows. *Journal of Dairy Science* **91**, 2370–2379.
- Kašparovská J, Dadáková K, Lochman J, Hadrová S, Křížová L and Kašparovský T (2017) Changes in equol and major soybean isoflavone contents during processing and storage of yogurts made from control or isoflavone-enriched bovine milk determined using LC-MS (TOF) analysis. *Food Chemistry* **222**, 67–73.
- Khafipour E, Krause DO and Plaizier JC (2009) A grain-based subacute ruminal acidosis challenge causes translocation of lipopolysaccharide and triggers inflammation. *Journal of Dairy Science* **92**, 1060–1070.
- Kim YH, Keeton JT, Smith SB, Berghman LR and Savell JW (2009) Role of lactate dehydrogenase in metmyoglobin reduction and color stability of different bovine muscles. *Meat Science* **83**, 376–382.

- Koohmarie M and Geesink GH (2006) Contribution of postmortem muscle biochemistry to the delivery of consistent meat quality with particular focus on the calpain system. *Meat Science* **74**, 34–43.
- Kristensen NB and Harmon DL (2004) Effect of increasing ruminal butyrate absorption on splanchnic metabolism of volatile fatty acids absorbed from the washed reticulorumen of steers. *Journal of Animal Science* **82**, 3549–3559.
- Li FN, Li LL, Yang HS, Yuan XX, Zhang B, Geng MM, Xiao CW and Yin YL (2011) Regulation of soy isoflavones on weight gain and fat percentage: evaluation in a Chinese Guangxi minipig model. *Animal: An International Journal of Animal Bioscience* **5**, 1903–1908.
- Li FF, Zhu T, Zhu Y, Gao Y, Meng L, Guo FL and Zhang Y (2015) Soybean isoflavone effects on growth performance, immune function, nutrient apparent digestibility and fecal microorganism in weaning piglets. *Swine Production* **5**, 25–29.
- Liang H, Xu LJ, Zhao XH, Bai J, Chen ZD, Zhou S, Song XZ, Ouyang KH, Pan K, Liu CJ and Qu MR (2018) Effect of daidzein on fermentation parameters and bacterial community of finishing Xianan cattle. *Italian Journal of Animal Science* **17**, 950–958.
- Liggins J, Mulligan A, Runswick S and Bingham SA (2002) Daidzein and genistein content of cereals. *European Journal of Clinical Nutrition* **56**, 961–966.
- Liu Q, Lanari MC and Schaefer DM (1995) A review of dietary vitamin E supplementation for improvement of beef quality. *Journal of Animal Science* **73**, 3131–3140.
- Liu X, Zhang G and Zhang W (2003) Effect of dietary addition cysteamine and daidzein on adipose metabolism in broilers. *Journal of Northeast Agricultural University* **34**, 171–175.
- Liu X, Suzuki N, Laxmi YRS, Okamoto Y and Shibutani S (2012) Anti-breast cancer potential of daidzein in rodents. *Life Sciences* **91**, 415–419.
- Liu DY, He SJ, Jin EH, Liu SQ, Tang YG, Li SH and Zhong LT (2013) Effect of daidzein on production performance and serum antioxidative function in late lactation cows under heat stress. *Livestock Science* **152**, 16–20.
- Lozupone CA, Stombaugh JI, Gordon JI, Jansson JK and Knight R (2012) Diversity, stability and resilience of the human gut microbiota. *Nature* **489**, 220–230.
- Lundh T (1995) Metabolism of estrogenic isoflavones in domestic animals. *Proceedings of the Society for Experimental Biology and Medicine* **208**, 33–39.
- Mishra P, Kar A and Kale RK (2009) Prevention of chemically induced mammary tumorigenesis by daidzein in pre-pubertal rats: the role of peroxidative damage and antioxidative enzymes. *Molecular and Cellular Biochemistry* **325**, 149–157.
- Mohammad-Shahi M, Haidari F, Rashidi B, Saei AA, Mahboob S and Rashidi MR (2011) Comparison of the effects of genistein and daidzein with dexamethasone and soy protein on rheumatoid arthritis in rats. *BioImpacts: BI* **1**, 161–170.
- Morimoto M, Watanabe T, Yamori M, Takebe M and Wakatsuki Y (2009) Isoflavones regulate innate immunity and inhibit experimental colitis. *Journal of Gastroenterology and Hepatology* **24**, 1123–1129.
- O'Keefe M and Hood DE (1982) Biochemical factors influencing metmyoglobin formation on beef from muscles of differing colour stability. *Meat Science* **7**, 209–228.
- Ovadia H, Haim Y, Nov O, Almog O, Kovsan J, Bashan N, Benhar M and Rudich A (2011) Increased adipocyte s-nitrosylation targets anti-lipolytic action of insulin: relevance to adipose tissue dysfunction in obesity. *Journal of Biological Chemistry* **286**, 30433–30443.
- Rehfeldt C, Adamovic I and Kuhn G (2007) Effects of dietary daidzein supplementation of pregnant sows on carcass and meat quality and skeletal muscle cellularity of the progeny. *Meat Science* **75**, 103–111.
- Ren DP, Geng ZC, Liu SJ and Guo J (2009) Effects of cysteamine and daidzein on gene expression of IGF-I mRNA in tissue of northeast fine-wool sheep. *Chinese Journal of Animal Nutrition* **21**, 967–973.
- Ru BR, Lou CH, Qi XL, Li PF, Wang ZB and Yang QC (2006) Project progress report on new beef line breeding by introducing the Charolais blood into Nanyang cattle. *China Cattle Science* **75**, 40–43.
- Šošić-Jurjević B, Filipović B, Ajdžanović V, Brkić D, Ristić N, Stojanoski MM, Nestorović N, Trifunović S and Sekulić M (2007) A brief communication: subcutaneously administered genistein and daidzein decrease serum cholesterol and increase triglyceride levels in male middle-aged rats. *Experimental Biology and Medicine* **232**, 1222–1227.
- Szkudelska K, Szkudelski T and Nogowski L (2002) Daidzein, coumestrol and zearalenone affect lipogenesis and lipolysis in rat adipocytes. *Phytomedicine* **9**, 338–345.
- Van Soest PJ, Robertson JB and Lewis BA (1991) Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *Journal of Dairy Science* **74**, 3583–3597.
- Vyn TJ, Yin X, Bruulsema TW, Jackson CC, Rajcan I and Brouder SM (2002) Potassium fertilization effects on isoflavone concentrations in soybean. *Journal of Agricultural and Food Chemistry* **50**, 3501–3506.
- Wang G, Hang T and Chen J (1994) Effects of daidzein on muscle growth in broilers and mechanism involved. *Guangdong Journal of Animal and Veterinary Science* **3**, 4–6.
- Wang Q, Garrity GM, Tiedje JM and Cole JR (2007) Naïve Bayesian classifier for rapid assignment of rRNA sequences into the new bacterial taxonomy. *Applied and Environmental Microbiology* **73**, 5261–5267.
- Wang XZ, Han ZK and Wang GJ (2008) Effects of ipriflavone on ruminal metabolism and concentration of sex hormones in rumen fluid of goat. *Chinese Journal of Animal Nutrition* **3**, 1–6.
- Wong MC, Emery PW, Preedy VR and Wiseman H (2008) Health benefits of isoflavones in functional foods? Proteomic and metabolomic advances. *Inflammopharmacology* **16**, 235–239.
- Zhao XH, Yang ZQ, Bao LB, Wang CY, Zhou S, Gong JM, Fu CB, Xu LJ, Liu CJ and Qu MR (2015) Daidzein enhances intramuscular fat deposition and improves meat quality in finishing steers. *Experimental Biology and Medicine* **240**, 1152–1157.
- Zhao XH, Zhou S, Bao LB, Song XZ, Ouyang KH, Xu LJ, Pan K, Liu CJ and Qu MR (2018) Response of rumen bacterial diversity and fermentation parameters in beef cattle to diets containing supplemental daidzein. *Italian Journal of Animal Science* **17**, 643–649.
- Zhou S, Zilberman Y, Wassermann K, Bain SD, Sadovsky Y and Gazit D (2001) Estrogen modulates estrogen receptor alpha and beta expression, osteogenic activity, and apoptosis in mesenchymal stem cells (MSCs) of osteoporotic mice. *Journal of Cellular Biochemistry* **36**(Suppl), 144–155.