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## Effects of Nutritional Level of Concentrate-Based Diets on Meat Quality and Expression Levels of Genes Related to Meat Quality in Hainan Black Goats

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## ORIGINAL ARTICLE

## Effects of nutritional level of concentrate-based diets on meat quality and expression levels of genes related to meat quality in Hainan black goats

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## ABSTRACT

The present study investigated the effects of the nutritional levels of diets on meat quality and related gene expression in Hainan black goat. Twenty-four goats were divided into six dietary treatments and were fed a concentrate-based diet with two levels of crude protein (CP) (15% or 17%) and three levels of digestive energy (DE) (11.72, 12.55 or 13.39 MJ/kg DM) for 90 days. Goats fed the concentrate-based diet with 17% CP had significantly ( $P < 0.05$ ) higher average daily gains (ADG) and better feed conversion rates (FCR). The pH<sub>24h</sub> value tended to decrease ( $P < 0.05$ ) with increasing DE levels. The tenderness of Longissimus dorsi muscle (LD) and Semimembranosus muscle (SM) reduced with increasing CP levels ( $P < 0.05$ ). With increasing DE levels, tenderness was increased ( $P < 0.05$ ). The heart fatty acid-binding protein (H-FABP) mRNA expression levels in LD and SM increased with increasing DE levels ( $P < 0.05$ ), but decreased with increasing CP levels ( $P < 0.05$ ). The calpastatin (CAST) and  $\mu$ -calpain mRNA expressions levels in LD and SM were affected significantly ( $P < 0.05$ ) by CP and DE levels in the diet. Therefore, the nutritional levels of diets affect meat quality and expression levels of genes associated with meat quality in Hainan black goats.

**Key words:** calpastatin, Hainan black goat, heart fatty acid-binding protein, meat quality,  $\mu$ -calpain.

## INTRODUCTION

Goats are good sources of lean meat. Goats store higher proportions of polyunsaturated fatty acids in their tissues (Mushi *et al.* 2008). The demand for goat production will continue to increase in the future (Kannan *et al.* 2006). Goats fed concentrate-based diets generally have higher growth rates, dressing percentage and carcass quality than goats that are grass-fed (Kosum *et al.* 2003). Furthermore, goats fed on suitable nutrition levels of concentrate-based diets have coordinated meat quality with those goats fed mainly on pasture (Kannan *et al.* 2006). Several studies have shown that the nutritional level of diets can affect meat quality and the expression levels of genes associated with meat quality (Patterson *et al.* 2009; Hocquette *et al.* 2010; Guo *et al.* 2011).

Hainan black goat, a meat goat breed, is raised in the south of China. This breed is not only tolerant to the warm and rainy climates of the region, it is also a source of delicious meat. In the past, Hainan black goats grazed freely. Recently, in an attempt to protect the environment and develop large-scale farming, goats have been confined. This breed of goat has great

flaws with slow growth rates and small body sizes; however, goat meat remains very popular in the south of China due to its delicious flavors.

Heart fatty acid-binding protein (H-FABP) has been associated with intramuscular fat (IMF) content (Gerbens *et al.* 1999). Even though H-FABP is widely distributed, the heart and skeletal muscle tissue have the greatest expression levels (Li *et al.* 2007). H-FABP may play an important role in the development of intramuscular adipocytes. Certain enzymes are associated with meat tenderness; for example,  $\mu$ -calpain is an enzyme largely responsible for postmortem muscle proteolysis (Geesink *et al.* 2006) and calpastatin (CAST) is an enzyme that decreases muscle proteolysis by inhibiting the action of  $\mu$ -calpain (Goll *et al.* 2003).

Few studies have been conducted to assess the effect of nutritional level of diets on growth performance

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**Table 1** Composition and nutrient levels of concentrate diets, as-fed basis

CP (%)	15			17		
	11.72	12.55	13.39	11.72	12.55	13.39
DE (MJ/kg)	11.72	12.55	13.39	11.72	12.55	13.39
Ingredients (%)						
Maize	53.00	61.00	74.00	48.00	59.00	68.50
Soy bean meal	19.50	19.00	19.50	26.00	25.00	25.50
Wheat bran	12.50	10.00	0.50	10.00	6.50	–
Chaff	9.00	4.00	–	10.00	3.50	–
Stone dust	0.50	0.50	0.50	0.50	0.50	0.50
Salt	1.00	1.00	1.00	1.00	1.00	1.00
Sodium bicarbonate	0.50	0.50	0.50	0.50	0.50	0.50
Premix†	4.00	4.00	4.00	4.00	4.00	4.00
Nutrition level‡						
Dry matter (%)	87.67	87.53	87.37	87.86	87.67	87.55
DE (MJ/kg)	11.80	12.60	13.39	11.78	12.58	13.40
Crude protein (%)	15.08	15.05	14.99	17.14	17.08	17.08
Ether extract (%)	2.35	2.85	3.14	2.77	3.75	3.95
NDF (%)	12.26	12.01	9.80	12.15	12.35	9.91
ADF (%)	5.35	5.26	4.53	5.48	5.31	4.85
Calcium (%)	0.78	0.79	0.80	0.79	0.80	0.81
Phosphorous (%)	0.37	0.36	0.31	0.36	0.36	0.33

†The premix provides the following per kg of diet: vitamin A 15000 IU, vitamin D 5000 IU, vitamin E 50 mg, Fe 9 mg, Cu 12.5 mg, Zn 100 mg, Mn 130 mg, Se 0.3 mg, I 1.5 mg, Co 0.5 mg. ‡The nutrient levels are measured values not including DE. CP, crude protein; DE, digestible energy; NDF, neutral detergent fiber; ADF, acid detergent fiber.

and meat quality of Hainan black goats, a local goat breed in South China. Therefore, our objective was to determine the effects of the nutritional level of diets on meat quality and expression of genes associated with meat quality in Hainan black goats.

## MATERIALS AND METHODS

### Animals and treatments

The animal experiment was approved by the Institutional Animal Care and Use Committee at the Chinese Academy of Tropical Agricultural Sciences in Haikou, China. The experiment was conducted in accordance with the National Institute of Health guidelines for the care and use of experimental animals.

The experiment had a 2 × 3 factorial design using a concentrate-based diet with a crude protein (CP) level of 15% or 17%, and a digestible energy (DE) level of 11.72, 12.55 or 13.39 MJ/kg. The ingredients and chemical composition of the concentrate-based diet are listed in Table 1. Twenty-four castrated Hainan black goats (3 months old and 10.56 ± 1.28 kg body weight (BW)) were randomly allocated to six dietary treatments, and each treatment was replicated four times with one goat per replicate.

### Physical and chemical composition of the concentrate-based diet and of King grass

The physical and chemical composition of concentrate-based diet and of King grass was determined according to the method of the AOAC (2000). The composition and nutritional levels of the diets are shown in Tables 1 and 2.

### Feeding and management

Animals were given a 15-day adaptation period during which they were treated with ivermectin against internal

**Table 2** Chemical composition of King grass (air-dry basis)

Items	Content (%)
Dry matter	22.50
Crude protein	8.27
Ether extract	1.04
Crude ash	3.26
Neutral detergent fiber	66.22
Acid detergent fiber	42.97
Calcium	0.10
Phosphorous	0.15

and external parasites. Fresh King grass (*Pennisetum purpureum* × *P.americanum* cv. Reyan No.4; its chemical composition is shown in Table 2) and the concentrate-based diet were fed twice daily (08.30 and 15.00 hours). The goats were fed the concentrate-based diet followed by the grass. Water was freely available to the goats. During the experimental period (90 days) animals were stall-fed in individual pens. Feeding allocations and refusals to eat were recorded daily for each goat. Each month, the goats were weighed before the morning feeding.

### Slaughter and samples collection

Goats were weighed on two consecutive days before slaughter to obtain the final live weight (FLW). Then they were fasted for 16 h and weighed again to obtain the slaughter live weight (SLW). The slaughter was performed over three consecutive days.

Approximately 100 g of Longissimus dorsi muscle tissue (LD, fifth lumbar vertebra region) and 100 g of Semimembranosus muscle tissue (SM) were removed from the left sides of carcasses and stored at –20°C for subsequent IMF determination.

For total RNA extraction, the LD and SM samples, obtained from the left side of the carcass within 15 min after

slaughter, were immediately frozen in liquid nitrogen and stored at  $-80^{\circ}\text{C}$  until analysis.

Two  $5 \times 2.5 \times 2.5$  cm chops of LD and SM, respectively, were obtained from the right side of the carcass along the direction of the muscle fibers within 45 min after slaughter, and stored at  $-20^{\circ}\text{C}$  for 24 h. These chops were used in the determination of Warner-Bratzler shear force (WBSF).

### Muscle property

Two 2.5 cm thick chops of LD and SM were obtained. The first chop was used for pH measurement approximately 45 min and 24 h postmortem using a penetrating electrode (Mettler Toledo, Changzhou, China) attached to a portable pH-meter (FG2, Shanghai, China). The pH probe was calibrated with pH 4 and pH 7 standard buffer solutions. The chop was then stored at  $4^{\circ}\text{C}$  for 24 h before the final pH measurement. The second chop was weighed, placed in a Whirlpak bag, suspended in a  $4^{\circ}\text{C}$  cooler for 24 h, and then reweighed. Chop drip loss was calculated based on the weight loss; drip loss was expressed as percentage.

LD and SM chops (aged 24 h at  $-20^{\circ}\text{C}$ ) were thawed for 16 h at  $4^{\circ}\text{C}$  and then cooked to an internal temperature of  $70^{\circ}\text{C}$  in a thermostatic water-bath set at  $80^{\circ}\text{C}$ . After removal from the water bath, LD and SM chops (two chops in every muscle, respectively) were allowed to cool to  $4^{\circ}\text{C}$ , and then two  $5 \times 1 \times 1$  cm chops from each chop were cut parallel to the orientation of the muscle fiber. Each chop was then sheared four times at a crosshead speed of 1 mm/s using a Texture Measurement System (Food Technology Corporation, Stirling, VA, USA). WBSF for individual chops were averaged for each sample, and then were averaged over four replicates for each dietary treatment (Guo *et al.* 2011).

Approximately 50 g of LD and SM samples were thawed for 16 h at  $4^{\circ}\text{C}$ . External fat and connective tissue were removed prior to homogenization. Samples were placed in a drying oven at  $65^{\circ}\text{C}$  for at least 48 h and then IMF was extracted in petroleum ether using an automated extraction system (Gerhardt, Bonn, Germany).

### RNA extraction and mRNA expressions

Total RNA was extracted using a commercial kit (Biotek Corporation, RP1201, Beijing, China). Purity and concentration of RNA was checked with the RNA 6000 Nano LabChip kit using a bioanalyzer (Agilent Technologies, Shanghai, China). Total RNA (2  $\mu\text{g}$ ) was used in a 20  $\mu\text{L}$  reverse tran-

scription reaction volume (Biotek Corporation). The resulting cDNA was diluted to 200  $\mu\text{L}$  with diethylpyrocarbonate (DEPC)-treated water and stored at  $-20^{\circ}\text{C}$ .

The cDNA was amplified by real-time PCR using SYBR green real-time PCR master mix (Fermentas, Burlington, Canada) according to the manufacturer's instructions. All the primer sequences are listed as follows: glyceraldehyde-3-phosphate dehydrogenase (GAPDH), 3'- ACCTTCACTACATGGTCTAC, 5'- CTTTCCATTGATGACGAGCTTC, amplification length 101 bp, accession no. AJ431207.1; H-FABP, 3'- TGGAAGTTAGTGACAGC, 5'- GATTGTGGTAGGTTGGTC, amplification length 102 bp, accession no. AY466498.1; CAST, 3'- GCACCCCTCAGATACAAGAAGC, 5'- TTTGGTTTGTGATTCTCTG, amplification length 89 bp, accession no. GU944861.1;  $\mu$ -calpain, 3'- TCCTGCACCGAGTAGTTC, 5'- TCCACCCACTCACAAACTGC, amplification length 94 bp, accession no. HQ718593.1. GAPDH gene was used as an internal control. The melting peaks of the amplification products were determined by melting curve analyses in order to ascertain that only the expected amplification products had been generated. Relative gene expression levels were presented as  $2^{-\Delta\Delta\text{C}_T}$ ; the  $\Delta\Delta\text{C}_T$  method was used as described by Livak and Schmittgen (2001).

### Statistical analyses

Analysis of variance (ANOVA) was performed with the assumption of homogeneity of variance using SAS software (SAS 1990). The results were analyzed in a  $2 \times 3$  factorial design with DE and CP and their interaction included as the main effects. Statistically significant differences were identified using Tukey's multiple-range test at  $P < 0.05$ .

## RESULTS

### Diet intake and kid growth

The DE level of the diet had no significant effect on final weight, slaughter weight, average daily gains (ADG), total dry matter feed intake, or feed conversion ratio (FCR) (Table 3). On the other hand, the CP level of the diet significantly affected ADG and FCR ( $P < 0.05$ ). Goats fed the concentrate-based diet with 17% CP had a significantly ( $P < 0.05$ ) higher ADG ( $77.24 \pm 3.66$  g) and better FCR ( $6.76 \pm 0.40$ ) than

**Table 3** Effects of nutritional levels of diets on growth performance in Hainan black goats

CP (%)	15			17			SEM	P-values†		
	DE (MJ/kg)	11.72	12.55	13.39	11.72	12.55		13.39	P	E
Live weight (kg)										
Initial		10.56	10.53	10.67	10.50	10.56	10.56	0.02	0.94	0.99
Final		16.88	16.69	16.42	16.63	17.44	17.88	0.23	0.27	0.84
Slaughter‡		15.42	15.75	15.92	15.83	16.67	16.25	0.24	0.83	0.64
ADG (g/day)		70	68	66	68	76	81	1	0.02	0.48
Feed intake (g/day)										
Concentrate, as fed		281	289	274	284	288	285	2	0.16	0.08
King grass (dry matter)		240	231	239	232	246	235	2	0.79	0.89
Total dry matter		521	520	513	516	535	520	3	0.38	0.37
FCR		7.48	7.65	7.30	6.97	7.04	6.47	0.17	0.03	0.39

ADG, average daily gains; FCR, feed conversion ratio. †P and E means influence factors of crude protein levels and digestible energy levels in the diet, respectively. ‡After 16 h of fasting.

**Table 4** Effects of nutritional level of diets on meat quality in Hainan black goats

CP (%)	15			17			SEM	<i>P</i> -values†		
	DE (MJ/kg)	11.72	12.55	13.39	11.72	12.55		13.39	<i>P</i>	<i>E</i>
Longissimus dorsi muscle										
Drip loss (%)	1.63	1.79	1.47	1.76	1.61	1.68	0.03	0.73	0.74	0.55
pH <sub>45min</sub>	6.54 <sup>a</sup>	6.44 <sup>ab</sup>	6.51 <sup>ab</sup>	6.52 <sup>ab</sup>	6.15 <sup>b</sup>	6.57 <sup>a</sup>	0.02	0.22	0.02	0.11
pH <sub>24h</sub>	5.47 <sup>ab</sup>	5.33 <sup>ab</sup>	5.24 <sup>b</sup>	5.52 <sup>a</sup>	5.47 <sup>a</sup>	5.37 <sup>ab</sup>	0.01	0.05	0.04	0.94
IMF (%)	6.83	7.21	7.30	6.46	6.55	6.97	0.07	0.05	0.22	0.79
WBSF (N)	73.83 <sup>a</sup>	68.16 <sup>c</sup>	62.58 <sup>c</sup>	75.71 <sup>a</sup>	70.92 <sup>b</sup>	66.07 <sup>d</sup>	0.09	<0.0001	<0.0001	0.17
Semimembranosus muscle										
Drip loss (%)	1.78	2.14	1.78	1.58	1.67	1.59	0.10	0.20	0.62	0.82
pH <sub>45min</sub>	6.64	6.64	6.68	6.49	6.28	6.57	0.01	0.02	0.20	0.38
pH <sub>24h</sub>	5.70 <sup>a</sup>	5.60 <sup>a</sup>	5.16 <sup>b</sup>	5.54 <sup>a</sup>	5.53 <sup>a</sup>	5.44 <sup>ab</sup>	0.02	0.78	0.001	0.02
IMF (%)	6.58	6.79	7.33	6.33	6.47	6.66	0.06	0.07	0.15	0.69
WBSF (N)	69.02 <sup>d</sup>	66.06 <sup>e</sup>	61.69 <sup>f</sup>	75.05 <sup>a</sup>	73.31 <sup>b</sup>	70.49 <sup>c</sup>	0.10	<0.0001	<0.0001	0.002

IMF, intramuscular fat; WBSF, Warner-Bratzler shear force. <sup>a,b</sup>Values within a row with different superscripts differ significantly at  $P < 0.05$ . †*P* and *E* mean influence factors of crude protein levels and digestible energy levels in diets, respectively.

goats fed the concentrate-based diet with 15% CP (ADG  $69.81 \pm 1.21$  g, FCR  $7.48 \pm 0.17$ ).

### Muscle properties

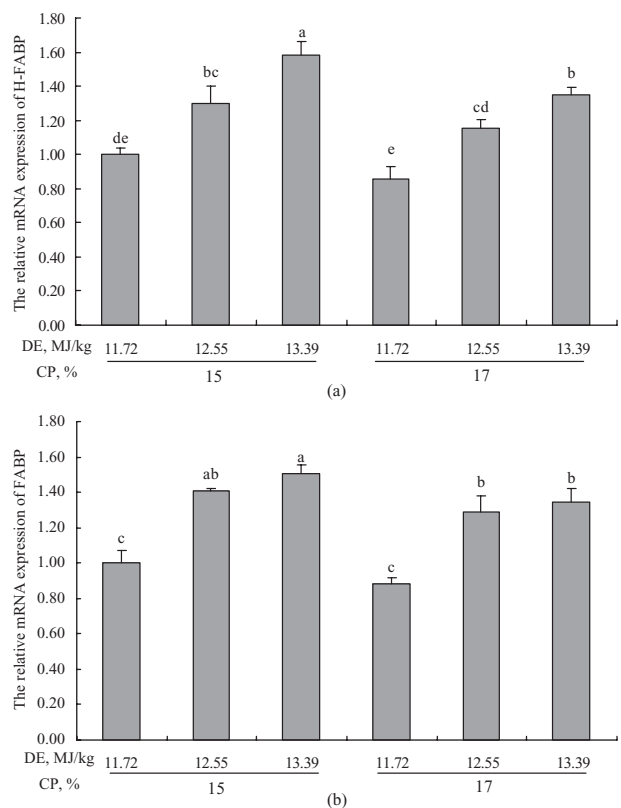
Drip loss and IMF of LD and SM samples were not affected by CP or DE levels (Table 4). The pH<sub>24h</sub> value tended to decrease ( $P < 0.05$ ) with increasing DE levels. On the other hand, CP levels did not affect pH<sub>24h</sub> (Table 4). The WBSF of goat muscle tissue was significantly ( $P < 0.05$ ) affected by CP and DE levels (Table 4). The shear force of LD and SM increased with increasing CP levels; the goat fed the concentrate-based diet with CP of 17% and DE of 11.72 MJ/kg had the highest WBSF ( $P < 0.05$ ). With increasing DE levels, WBSF was reduced ( $P < 0.05$ ).

### H-FABP mRNA expression levels

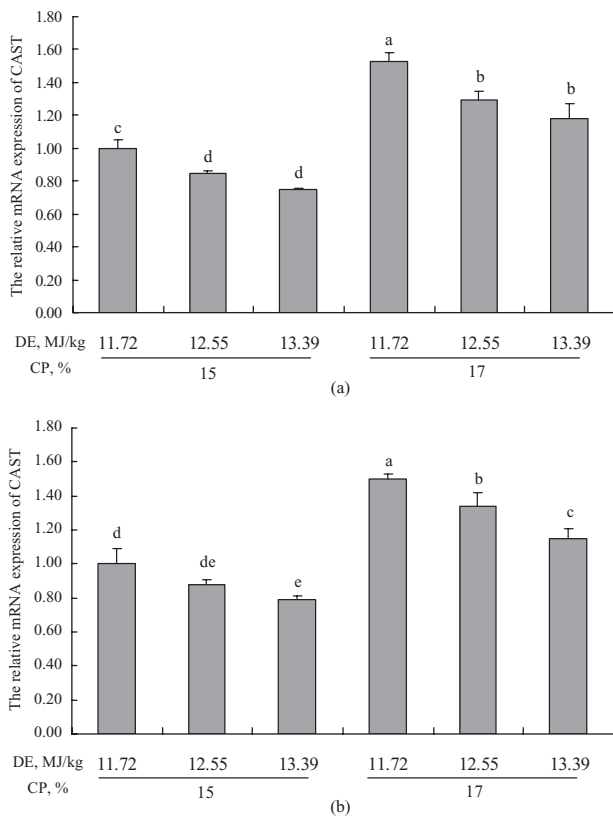
The CP and DE levels significantly affected the mRNA expression levels of H-FABP in LD and SM ( $P < 0.05$ ) (Fig. 1). The H-FABP mRNA expression levels in LD and SM increased with increasing DE levels ( $P < 0.05$ ), but decreased with increasing CP levels ( $P < 0.05$ ). Goats fed the concentrate-based diet with CP of 15% and DE of 13.39 MJ/kg had the highest H-FABP mRNA expression levels ( $P < 0.05$ ). Goats fed the concentrate-based diet with CP of 17% and DE of 11.72 MJ/kg had the lowest H-FABP mRNA expression levels ( $P < 0.05$ ).

### CAST and $\mu$ -calpain mRNA expressions levels

The mRNA expression levels of CAST in LD and SM increased with increasing CP levels ( $P < 0.05$ ) and decreased with increasing DE levels ( $P < 0.05$ ) (Fig. 2). Goats fed the concentrate-based diet with CP of 17% and DE of 11.72 MJ/kg had the highest CAST mRNA expression levels ( $P < 0.05$ ).



**Figure 1** Relative heart fatty acid binding protein (H-FABP) mRNA expression levels. Data are expressed as means  $\pm$  SD ( $n = 4$ ). Different superscripts indicate significant difference at  $P < 0.05$ . (a) Relative H-FABP mRNA expression levels in goat Semimembranosus muscle (SM). *P*-values of the main effects are: crude protein (CP),  $P = 0.0002$ ; digestible energy (DE),  $P < 0.0001$ ; and CP  $\times$  DE,  $P = 0.51$ . (b) Relative H-FABP mRNA expression levels in goat Longissimus dorsi (LD). *P*-values of the main effects are CP,  $P = 0.0003$ ; DE,  $P < 0.0001$  and CP  $\times$  DE,  $P = 0.78$ .



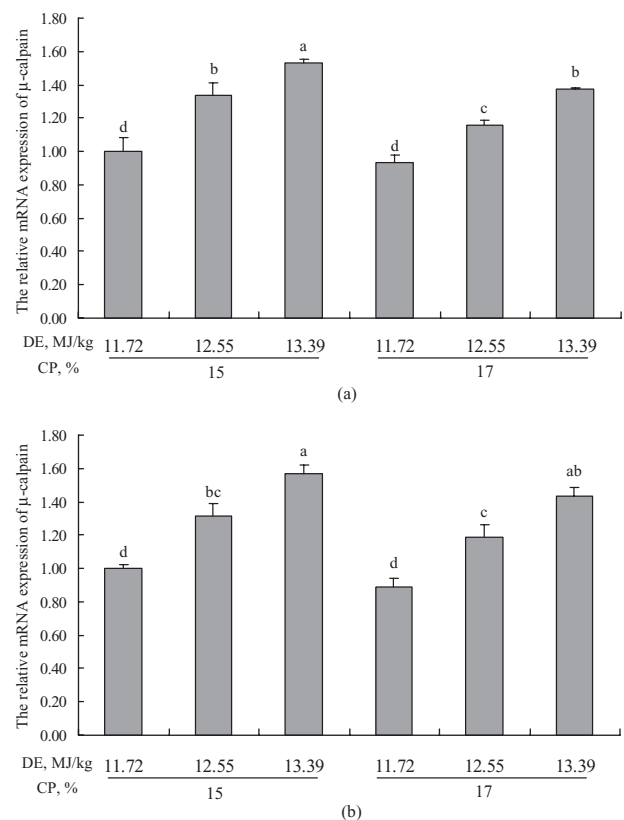
**Figure 2** Relative calpastatin (CAST) mRNA expression levels. Data are expressed as mean  $\pm$  SD ( $n = 4$ ). Different superscripts indicate significant difference at  $P < 0.05$ . (a) Relative CAST mRNA expression levels in goat Semimembranosus muscle (SM).  $P$ -values of the main effects are: crude protein (CP),  $P < 0.0001$ ; DE,  $P < 0.0001$ ; and CP  $\times$  DE,  $P = 0.18$ . (b) Relative CAST mRNA expression levels in goat Longissimus dorsi (LD).  $P$ -values of the main effects are CP,  $P < 0.0001$ ; DE,  $P < 0.0001$ ; and CP  $\times$  DE,  $P = 0.04$ .

In contrast, goats fed the concentrate-based diet with CP of 17% and DE of 11.72 MJ/kg had the lowest  $\mu$ -calpain mRNA expression levels ( $P < 0.05$ ) (Fig. 3) and goats fed the concentrate-based diet with CP of 15% and DE of 13.39 MJ/kg had the highest  $\mu$ -calpain mRNA expression levels ( $P < 0.05$ ). The CAST and  $\mu$ -calpain mRNA expressions levels in LD and SM were affected significantly ( $P < 0.05$ ) by CP and DE levels in the diet (Figs 2,3).

## DISCUSSION

### Effect of the nutritional level of diets on kid growth performance

The nutritional levels used in this study were based on the nutritional requirements of goats living in developing countries (BW 10 kg, ADG 75 g) as reported by Keral (1982) and obtained similar growth performances. In the present study, the CP level of the diet influenced ADG and FCR significantly, and DE level in



**Figure 3** Relative  $\mu$ -calpain mRNA expression levels. Data are expressed as mean  $\pm$  SD ( $n = 4$ ). Different superscripts indicate significant difference at  $P < 0.05$ . (a) Relative  $\mu$ -calpain mRNA expression levels in goat Semimembranosus muscle (SM).  $P$ -values of the main effects are: crude protein (CP),  $P < 0.0001$ ; digestible energy (DE),  $P < 0.0001$ ; and CP  $\times$  DE,  $P = 0.05$ . (b) Relative  $\mu$ -calpain mRNA expression levels of goat Longissimus dorsi (LD).  $P$ -values of the main effects are CP,  $P = 0.0006$ ; DE,  $P < 0.0001$ ; and CP  $\times$  DE,  $P = 0.94$ .

the diet had no significant effect on the growth performance of goats. This may be due to the fact that post-natal growth performance of Hainan black goat is slow and does not response to high energy. The kids may require high protein between 3 and 6 months of age to develop muscle mass, and during the remainder of their growth period, muscle development decreases in favor of fat synthesis. Studies have reported that concentrate-based diets with high nutritional levels result in better goat growth performance. Majdoub-Mathlouthi *et al.* (2013) reported that lambs receiving large concentrate diets (600 g) had high ADG and improved feed conversion rates (FCR).

### Effects of the nutritional level of diets on kid meat quality

The present study indicated that the nutritional levels of the diets had no significant effect on the drip loss of goat muscle. This was similar to the results by Guo

*et al.* (2011), who reported that different protein-carbohydrate ratios in the diet had no significant effect on the drip loss of pig muscle. Abdullah and Musallam (2007) also reported that drip loss was not affected by the energy contents of diets fed to male black goats.

Generally, muscle pH values are reduced during the immediate postmortem period; the rate of pH decline usually has remarkable effects on meat quality (Diaz *et al.* 2002). In the present study, the muscle pH<sub>24h</sub> values were within or near the acceptable range for goats (pH range: 5.6-5.8) (Pratiwi *et al.* 2007). Furthermore, the results revealed that the pH<sub>24h</sub> values declined ( $P < 0.05$ ) with increasing DE levels in the diet. Kadim *et al.* (2006) reported that increased energy intake leads to increased glycogen storage. Stored glycogen can be used in glycolysis, a pathway that is responsible for low ultimate pH values.

IMF is important for evaluating meat quality. IMF influences the juiciness, flavor and tenderness of meat. Sanudo *et al.* (2000) reported that meat from concentrate-supplemented goats had higher IMF content than unsupplemented goats. Young goats (3-6 months of age) mainly develop muscle mass, so the nutritional level of CP and DE had no significant effects on IMF content in goat muscle tissue in the present study. Meanwhile, the IMF content tended to increase with increasing DE levels and declined with increasing CP levels. These results are similar to those obtained in pigs. The IMF content in pig muscle tissue increased with low protein diets (Guo *et al.* 2011). The energetic efficiency of dietary protein is lower than that of starch, a carbohydrate (Van Milgen *et al.* 2001); therefore, reducing the protein/carbohydrate ratio increased the amount of energy available for fat deposition.

Tenderness is one of the most important factors influencing meat acceptability (Boleman *et al.* 1997). Apple *et al.* (2004) reported that low-protein diets improved tenderness. This could be attributed to an increase in IMF content and to enhanced myofibrillar protein degradation. In the present study, WBSF of goats fed the concentrate-based diet with 15% CP was lower than that of the goats fed the concentrate-based diet with 17% CP. Furthermore, WBSF declined with increasing DE levels, which is consistent with our research findings that IMF tended to increase with increasing DE levels. De Vol *et al.* (1988) and Hodgson *et al.* (1991) both reported that high IMF levels contribute to a significant reduction of WBSF in pigs.

### **Effect of the nutritional levels of diets on the mRNA expression of genes associated with meat quality**

The FABP family comprises a group of small cytosolic proteins. FABP is involved in the intracellular transport of fatty acids to fat storage sites or energy production sites (Krag *et al.* 2007). H-FABP has been

associated with IMF content in pigs (Gerbens *et al.* 1999). H-FABP is widely distributed, and the heart and skeletal muscle have the greatest expression levels (Li *et al.* 2007). In this study, H-FABP mRNA expression levels tended to increase with increasing IMF and DE levels, which is in agreement with the results obtained by Gerbens *et al.* (2001) who reported that H-FABP mRNA levels were positively correlated with IMF content. On the other hand, H-FABP mRNA expression levels declined with increasing CP levels which is in agreement with the results obtained by Guo *et al.* (2011) who reported that low-protein diets increase IMF content and H-FABP mRNA expression levels in pig muscle tissue.

Tenderness is the most important meat quality. The main factor that determines tenderness is the extent of protein proteolysis in muscle fibers (Koochmaraie & Geesink 2006). Calpains, which belong to the protease family, play a role in meat quality. As calpain activity increases, meat tenderness increases (Sentandreu *et al.* 2002). Geesink *et al.* (2006) reported that  $\mu$ -calpain, as opposed to m-calpain, is largely responsible for the myofibrillar protein degradation in skeletal muscle. CAST is the calpain-specific endogenous inhibitor (Wendt *et al.* 2004). Calpain systems are likely to be affected by the nutritional level of the diet (Thomson *et al.* 1997); research has indicated that there was a weak positive association between calpastatin activity and protein gain in weaned wether lambs. The results were similar to our study; in the present study, CAST mRNA expression levels significantly increased with increasing CP levels and significantly decreased with increasing DE levels. Interestingly, the  $\mu$ -calpain mRNA expression levels were in contrast to the CAST mRNA expression levels:  $\mu$ -calpain mRNA expression significantly increased with increasing DE levels and significantly decreased with increasing CP levels. These results may be attributed to the fact that CAST is a calpain-specific endogenous inhibitor (Wendt *et al.* 2004). Studies have reported that diets of low nutritional level had a trend to increase IMF content, and decreased WBSF of pork and improved mRNA level of  $\mu$ -calpain in skeletal muscle (Tang *et al.* 2010). These results suggested that a moderately reduced energy and protein diet could increase meat tenderness and intramuscular fat.

The nutritional levels of diets can influence the metabolism, synthesis and functional properties of collagen (McCormick 1989), which may be partly due to the up- or down-regulation of genes associated with lipid and protein metabolism in the muscle. These genes may have an effect on IMF content and meat tenderness.

### **Conclusions**

The CP level of concentrate-based diet did influence the ADG and FCR. Concentrate-based diet with low



nutritional levels increased muscle meat tenderness in Hainan black goats. The changes in muscle meat tenderness as a result of the nutritional levels of the diet were similar to those reported in other studies. These results suggested that the nutritional level of the diet affects meat quality of Hainan black goats; the results may be explained at least in part by the changes in mRNA expression levels of H-FABP, CAST and  $\mu$ -calpain.

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