Arch. Tierz., Dummerstorf **51** (2008) 1, 33-41

[1] Laboratory of Molecular Biology and Bovine Breeding, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China
[2] Department of Animal Science, Tianjin Agricultural College, Tianjin 300384, P. R. China
[3] Department of Dairy and Animal Science, Pennsylvania State University, University Park, PA, 16802, USA
[4] Shirakawa Institute of Animal Genetics, Odakura, Nishigo, Nishi-shirakawa, Fukushima 961-8061, Japan
[5] Department of Animal Biotechnology, College of Agriculture, Biotechnology and Natural Resources, University of Nevada, Reno, Nevada 89557, USA

HONG GUO[1,2], WAN-SHANG LIU[3], AKIKO TAKASUGA[4], KATIE EYER[5], EARL LANDRITO[5], SHANG-ZHONG XU[1], XUE GAO[1] and HONG-YAN REN[1]

Mapping and association of *GAD2* and *GIP* gene variants with feed intake and carcass traits in beef cattle (short communication)

Abstract

Glutamate decarboxylase 2 (GAD2) is a major autoantigen in insulin-dependent diabetes, while glucose-dependent insulinotropic polypeptide (GIP) is a gastrointestinal hormone. Both genes are involved in insulin secretion and play a role in the regulation of feed intake and metabolic rate in animals. In the present study, we assigned the bovine *GAD2* and *GIP* by RH mapping to BTA 13 and BTA 19, respectively. We discovered SNPs from these genes by a PCR-sequencing approach. We identified one SNP (T/C transition) from the 14th intron of the bovine *GAD2* gene and another SNP (A/C transversion) from the 4th intron of the bovine *GIP* gene. Genotyping over 300 animals from five different beef populations revealed that the average allelic frequency for the *GAD2* allele A was 0.48 and allele B 0.52, while for the GIP allele A 0.80 and allele B 0.20, respectively. There were significant associations observed between the *GAD2* and *GIP* gene variants and Average Daily Feed Intake (ADFI) ($p<0.05$) in beef cattle. In addition, the *GAD2* SNP was also associated with Meat Percent (MP) ($p<0.05$), whereas the *GIP* SNP was also significantly associated with Backfat Thickness (BF) ($p<0.05$) and Ratio of Feed-to-Meat (RFM) ($p<0.05$). There were no significant effects found for other traits. Although both SNPs could be used as candidate markers for MAS to improve feed intake and carcass traits, further investigations in large populations and in other breeds are recommended in order to understand the effect of the *GAD2* and *GIP* polymorphisms on these quantitative traits in beef cattle.

Key Word: beef cattle, *GAD2*, *GIP*, RH mapping, single nucleotide polymorphism, feed intake, carcass traits

Zusammenfassung

Titel der Arbeit: **Kartierung von *GAD2* und *GIP* Genvarianten sowie ihre Beziehungen zu Futteraufnahme und Schlachtmerkmalen bei Fleischrindern (Kurzmitteilung)**

Glutamat Decarboxylase 2 (*GAD2*) ist ein bestimmender Faktor insulinabhängiger Diabetes, während es sich bei Glukose abhängigen Insulinpolypeptiden (*GIP*) um Magen-Darm Hormone handelt. Beide Gene sind an der Insulinsekretion beteiligt. Sie spielen bei Rindern für die Regulation der Futteraufnahme und der Stoffwechselrate eine das Wachstum beeinflussende Rolle. Die vorliegende Studie beschäftigt sich mit der Kartierung boviner *GAD2* und *GIP* Genvarianten und deren Effekte bei Leistungsmerkmalen von Fleischrindern. Es wurden SPN's dieser Gene mittels der PCR sequenziert. Dabei wurden ein SNP (T/C Transversion) des 14. Intron des bovinen *GAD2* Genes und ein weiteres (A/C Transversion) des 4. Introns des bovinen *GIP* Genes identifiziert. Die Genotypisierung von 300 Tieren aus fünf unterschiedlichen Rinderpopulationen zeigte, dass die durchschnittliche Allelfrequenz für das *GAD2* Allel A 0,48, das Allel B 0,52 und für das *GIP* Allel A 0,80 beziehungsweise 0,20 für das Allel B betrug. Nachgewiesen wurden signifikante ($p<0,05$) Beziehungen zwischen den *GAD2* und *GIP* Genvarianten und der täglichen Futteraufnahme bei Fleischrindern. Ebenfalls signifikante Beziehungen ($p<0.05$) fanden sich zwischen *GAD2* SNP und dem Fleischanteil sowie zwischen *GIP* SPN und der Rückenfettdicke sowie dem Futter : Fleischverhältnis. Bei anderen untersuchten Merkmalen ergaben sich keine signifikanten Beziehungen. Obwohl beide SPN's als Marker zur Verbesserung von Futteraufnahme sowie Schlachtmerkmalen genutzt werden können, werden weitere Untersuchungen mit größeren Tierzahlen anderer Rassen zum besseren Verständnis des *GAD2* und *GIP* Polymorphismus, im Zusammenhang mit den hier untersuchten quantitativen Merkmalen bei Fleischrindern, empfohlen.

Schlüsselwörter: Fleischrinder, *GAD2*, *GIP*, RH Kartierung, Nucleotid Polymorphismen, Futteraufnahme, Schlachtmerkmale

Introduction

The GAD2 gene encodes the glutamic acid decarboxylase enzyme (GAD65), catalyzes the formation of Gamma-aminobutyric acid (GABA) from L-glutamic acid and is expressed in both pancreatic islets and brain (ERDO et al., 1990). GABA is co-localized in neuropeptide Y (NPY) neurons, and is involved in the leptin pathway through the arcuate nucleus in the hypothalamus, suggesting that GABA and NPY interact in the paraventricular nucleus to stimulate food intake (OVESJO et al., 2001). Several lines of investigation support a role for GABA in regulation of feed intake and growth. For example, in turkey, it has been demonstrated that injection of varying doses of muscimol, a potent GABA agonist, caused a dose-dependant increase of food intake (DENBOW, 1991); Transgenic mice over-expressing the GABA transporter exhibited heritable obesity, with increased body weight and fat deposition (MA et al., 2000). A genome-wide scan performed in 158 multiplex French obese Caucasian families (514 individuals) revealed a microsatellite marker (D10S197) on human chromosome (HSA) 10p that is significantly linked to obesity (HAGER et al., 1998). This marker is located within the 7^{th} intron of the human GAD2 gene (http://www.ncbi.nlm.nih.gov/genome/sts/sts.cgi?uid=8782). BOUTIN et al. (2003) investigated SNPs of the GAD2 gene and performed association studies with obesity, obtaining evidence to implicate GAD2 as a candidate gene for human obesity. The study in cases with extreme forms of obesity have shown mutations in the gene involved in the leptin–melanocortin pathway (CLEMENT et al., 2002), which primarily impairs food intake regulation. Recently, MEYRE et al. found that the GAD2 promoter variation was significantly associated with childhood obesity in the French population and also influenced fetal growth and feeding behavior (MEYRE et al., 2005). Therefore, GAD2 could play a role in regulating growth and development in animals (ERLANDRE et al., 1992).

Glucose-dependent insulinotropic polypeptide (GIP, formerly known as gastric inhibitory polypeptide) is a gastrointestinal hormone (BUCHAN et al., 1978). The role of GIP generally has been thought to stimulate insulin secretion from pancreatic beta-cells. In humans, GIP appears to play a role in lipid physiology and is associated with obesity (MEIER et al., 2003). However, obesity is a result of the interaction of environmental and genetic factors that mediate energy intake and expenditure (SPIEGELMAN et al., 2001). There is experimental evidence indicating that GIP regulates fat metabolism in adipocytes, including enhanced insulin-stimulated incorporation of fatty acids into triglycerides, stimulation of lipoprotein lipase activity, and stimulation of fatty acid synthesis (YIP et al., 2000). In 1999, by investigating the effect of gastric inhibitory polypeptide on bovine fat metabolism, DAWSON et al. found that cattle fed on grass silage diets tend to deposit more fat than animals fed on dried forages. They concluded that, although GIP may regulate nutrient utilization, it is unlikely that the gene plays a major role in promoting fat accretion in cattle (DAWSON et al., 1999). GIP also stimulates the growth, differentiation, proliferation and survival of pancreatic b-cells (POSPISILIK et al., 2003; TRUMPER et al., 2001).

In this study, our main goal is to identify polymorphisms from the bovine *GAD2* and *GIP* genes and to determine whether the polymorphisms are associated with

quantitative traits, such as feed intake, growth and meat quality. This is the first step towards understanding whether bovine *GAD2* and *GIP* gene variants can serve as genetic markers for marker assisted selection (MAS) to improve feed intake, growth and carcass traits in cattle.

Materials and Methods

Animals

A total of 320 animals including Simmental (n = 25), Angus (n = 22), Hereford (n = 21), Sanhe (n = 201) and Simmental crossbred steers (Simmental crossed with indigenous female yellow cattle in China) (n = 51) were randomly selected from commercial populations and used to analyze the GAD2 allelic frequencies. A total of 331 animals including Simmental (n = 28), Angus (n = 23), Hereford (n = 28), Sanhe (n = 201) and Simmental crossbred (n = 51) were used to analyze GIP allelic frequencies. Only the 51 Simmental crossbreed steers were used for the association study. The crossbreed steers (405 ± 50.5 kg; 30 ± 2 months of age) were housed in a concrete-floored cowshed (in a single pan for each animal). The pre-trial period (for adaptation to treatment) was 15 days, and the test period was 180 days. Steers were fed according to the NY/T 815-2004 feeding standard of beef cattle (Agricultural Department of People's Republic of China 2004). The composition of the diet fed is 60% maize, 20% artifactitious soybean, 5% artifactitious cotton seed, 10% bran and 5% specific mixture feed (including vitamins, microelements and mineral elements), which contained a total of energy 45.98 MJ, 18% CP, 69% TDN, 2.3% Ca, and 1.0% P. Animals had free access to water and feed during the entire 195 days of the experimental period. The average dry matter intake (DMI) is 7.46 kg/d for each cow during the test period. All animals were weighed on days 1, 75 and 195 at 8:00 AM, with prior removal of feed and water (12 hours). Feed not consumed was weighed and recorded at 10:00 PM each day. Immediately after the trial, steers were harvested at a processing facility (KeErQing Beef cattle Co., Ltd. P. R. China). Carcass traits were measured according to the criterion GB/T 17238-1998 Cutting Standard of Fresh and Chilled Beef in China (China Standard Publishing House). The following traits, Dressing Percent (DP), Backfat Thickness (BF), Carcass Weight (CW), Meat Percent (MP), Average Daily Gain (ADG), Average Daily Feed Intake (ADFI), Ratio of Feed-to-Meat (RFM), Beginning Average Daily Gain (BADG; an average daily gain in the test period from day 1 to day 75), Finishing Average Daily Gain (FADG; an average daily gain in the test period from day 75 to day 180), Stomach Weight (SW) and Intestine Weight (IW) were measured or calculated.

DNA preparation, SNP detection and genotyping

Blood samples were collected from all 331 animals studied. The routine phenol chloroform extraction method was used to isolate the genomic DNA, which was diluted to 50 ng/µl for PCR.

The mRNA sequence for the human *GAD2* (NM_000818) and *GIP* (NM_004123) genes were obtained from NCBI and used to search for bovine expressed sequence tags (ESTs) from EST databases using a standard BLAST (http://www.ncbi.nlm.nih.gov/blast/) search (WANG et al., 2003; YANG et al., 2003; ZHU et al., 2005). The bovine ESTs that are ~ 500 bp in length and have more than 80% similarity with the corresponding human mRNA were identified as candidates.

These candidate ESTs were then assembled into contigs. Finally, a contig of bovine ESTs (GenBank acc. nos. CK846169, DY453982, BE899934) was used for *GAD2* PCR primer design, and an EST (GenBank acc. no. CK833953) was used for *GIP* primer design. Primer pairs (Table 1) were designed using Primer 5.0 (PREMIER Biosoft International, Palo Alto, CA, USA) and synthesized by Shanghai Bioasia Biotechnology Co. Ltd. in China. PCR was performed in a PTC-200 Peltier Thermal Cycler (MJ Research Inc, Hercules, CA, USA).

Amplified products were obtained for *GAD2* (primers: F5-R5) and for *GIP* (primers: F1-R1) in a 20 µl reaction volume consisting of 50 ng of bovine genomic DNA, 1× PCR buffer, 0.3 µM of each primer, 75 µM of each dNTP, 1.5 mM $MgCl_2$ (2 mM $MgCl_2$ for *GIP*) and 1U Taq DNA polymerase (Promega, Madison, WI, USA). PCR conditions were: 5 min at 95 ˚C followed by 30 s at 94 ˚C, 30 s at the annealing temperature (Table 1) and 30 s at 72 ˚C for 34 cycles and a final extension at 72 ˚C for 10 min. PCR products were analyzed on a 1.5% agarose gel stained with 0.5 µg/mL ethidium bromide. To verify their identity, the products were purified using a Wizard Prep PCR Purification Kit (Shanghai Bioasia Biotechnology Co., Ltd. P. R. China), cloned into the pGEM-T Easy Vector Kit (Promega, Madison, WI, USA), and sequenced (Applied Biosystems 3730xl DNA Analyzer, Foster City, CA, USA). Sequences of the fragments were used in BLASTn searches at NCBI to ensure that they were the expected bovine sequences. After alignment of the amplified sequences from different individuals, several putative single polymorphism sites were identified.

PCR-RFLP was used to genotype the SNPs in the bovine *GAD2* and *GIP* genes. The PCR products for *GAD2* and *GIP* were digested with *Hae* III and *Msp* I at 37 ˚C for 4 hrs. respectively. Restriction fragments were scored on 1.5% agarose gels. A total of 320 cattles from five breeds (Simmental, Angus, Hereford, Sanhe, crossbreed) were genotyped and allele frequencies were determined (Table 2).

Statistical analyses

The collected data were analyzed using SAS (SAS Institute Inc. Cary, NC, USA) according to the following model:

$$Y_{ij} = \mu + G_i + b_{ij}w_{ij} + \varepsilon_{ij}$$

Where Y_{ij} stands for observed value; µ: population mean; G_i: ith genotype; b_{ij}: regression coefficient; w_{ij}: weight; ε_{ij}: random error.

Table 1
Bovine *GAD2* and *GIP* gene primers used for SNP analysis and RH mapping (Nutzung boviner *GAD2* und *GIP* Genprimer für die SNP Analyse sowie RH Kartierung)

Gene	Primer	Sequences	Tm (˚C)	Size (bp)	Region
GAD2[a]	(F5)	5'-AGTGTTTGGAGCTGGCGGA-3'	60	630	exon 14
	(R5)	5'-AGACATTGGTGTGCTGAGG-3'			exon 15
	(F6)	5'-GTCCCCATTCCAGTTCTTCT-3'	63	346	intron 14
GIP[b]	(F1)	5'-TACACAACATCACCCAGAGG-3'	62	902	exon 4
	(R1)	5'-CAGCAGCTCTCGAATCAGTA-3'			exon 5
	(F2)	5'-GGAAGAGCCTATATTCCCTG-3'	65.5	262	intron 4

[a] Primer pair F5-R5 was used to isolate the 630 bp genomic DNA fragment of *GAD2* and primer pair F6-R5 was used for RH mapping.
[b] Primer pair F1-R1 was used to isolate the 902 bp genomic DNA fragment of *GIP* and primer pair F2-R1 was used for RH mapping.

RH mapping

A 7000-rad whole-genome cattle-hamster RH panel (SUNbRH), consisting of 92 hybrids, was used to map the *GAD2* gene with the primer pair F6-R5 and the *GIP* gene with the primer pair F2-R1 (Table 1). PCR typing, vector scoring and RH mapping were conducted following the methods described by GUO et al. (2005) and LIU et al. (2005).

Results and Discussion

SNP identification

Using the primer pair F5-R5 that flanks the 14th intron of the *GAD2* gene (Table 1), we amplified a fragment of 630 bp (GenBank acc. no. DQ139317) by PCR from 10 individuals, which represent four breeds (Simmental, Angus, Hereford, and Sanhe). Sequencing of the fragment confirmed the origin of the bovine *GAD2* gene. By careful comparison of the sequences from the 10 individuals, we found a SNP (T/C transition) at position nt 185 in the fragment located within the 14th intron of the gene. Digestion of the PCR fragments with *Hae* III revealed two alleles: allele A (T-base) – one fragment of 630 bp, and allele B (C-base) – two fragments of 445 bp and 185 bp (Table 2). Similarly, using the primer pair F1-R1 (Table 1) that flanks the 4th intron of *GIP*, a fragment of 902 bp was amplified (GenBank acc. no. DQ160296). Sequence comparison among the 10 individuals revealed a SNP (A/C transversion) at position nt 758 of the PCR fragment. Digestion of the 902 bp PCR product with *Msp* I revealed two alleles: allele A (A-base) – two bands with sizes of 740 bp and 162 bp, and allele B (C-base) – three bands with sizes of 596 bp, 162 bp and 144 bp, which were observed on agarose gels. Allele frequency for both the *GAD2* and *GIP* genes was investigated in five different beef populations (Table 2). The frequency of the *GAD2* allele A ranged from 0.43 to 0.55. The frequency of the *GIP* allele A ranged from 0.58 to 0.98) (Table 2). We found that the allelic frequency varied among the five cattle populations studied. For the *GAD2* gene, the Chinese breed Sanhe cattle and the crossbreed that contains 50% of Chinese Yellow Cattle blood had frequency of allele A ($\geq$ 0.51), in contrast to the Western beef breeds (Simmental, Angus and Hereford) ($\leq$ 0.48); whereas for the *GIP* gene, the Chinese breed and the crossbreed had a frequency of $\leq$ 0.60 for allele A, which was significantly lower than that of $\geq$ 0.89 in the Western beef breeds (Table 2).

Table 2
Allelic frequencies of the bovine *GAD2* and *GIP* genes in different cattle breeds (Allelfrequenzen boviner *GAD2* und *GIP* Gene verschiedener Rassen)

Breeds	*GAD2* allelic frequency			*GIP* allelic frequency		
	No. of animals	A	B	No. of animals	A	B
Simmental	25	0.48	0.52	28	0.89[a]	0.11[a]
Angus	22	0.43	0.57	23	0.98[a]	0.02[a]
Hereford	21	0.45	0.55	28	0.95[a]	0.05[a]
Sanhe	201	0.51	0.49	201	0.58[b]	0.42[b]
Crossbreed	51	0.55	0.45	51	0.60[b]	0.40[b]
Total or Average	320	0.48	0.52	331	0.80[a]	0.20[a]

Within columns means with different superscripts indicate significant differences at $P < 0.001$

The X^2 test of allelic frequency of both the *GAD2* and *GIP* genes was performed among 5 breeds of cattle. For the *GAD2* gene, there was not a significant difference in the allelic frequency among breeds (P>0.05). For the *GIP* gene, there was a significant difference in allelic frequency between the Chinese native breed (and the crossbreed) and the Western beef breeds (Simmental, Hereford and Angus) ($p \leq 0.001$).

RH mapping of the bovine GAD2 and GIP genes

By typing on the 7000-rad SUNbRH panel and analyzing the vector together with 3,216 framework markers (http://www.animalgenome.org/cattle/maps) (ITOH et al. 2005), the *GAD2* gene was assigned, at a 2-point (*DIK5201*) LOD score of 32, to the bovine chromosome (BTA) 13 (Table 3), mapping between *DIK2709* and *DIK5201* with a distance of ~ 8.08 cR to *DIK2709* and ~ 4.48 cR to *DIK5201*. Comparative mapping has demonstrated that human chromosome (HSA) 10, where the human *GAD2* gene is located, is conserved with homologous segments to BTA13, BTA26 and BTA28 (EVERTS- VAN DER WIND et al., 2004; ITOH et al., 2005), which supports the mapping of the bovine *GAD2* gene to BTA13 in this work. *GIP* was assigned, at a 2-point (*BM17132*) LOD score of 38, to BTA19 between *BM17132* and *DIK5224* with a small interval of ~ 0.6 cR to *BM17132* and ~ 0.61 cR to *DIK5224*. The human *GIP* gene maps to HSA17, which is completely conserved with BTA19 (EVERTS- VAN DER WIND et al., 2004), supporting the present assignment of *GIP* to BTA19.

Table 3
Chromosomal assignment of bovine *GAD2* and *GIP* genes (Chromosomale Zuordnung boviner *GAD2* und *GIP* Gene)

Gene	GenBank acc. no.	Lod score	Location in Human	Assignment in cattle				
				Marker 1	Interval marker 1 (cR)	Marker 2	Interval marker 2 (cR)	Chr
GAD2	DQ118401	31.65	10p11.23	*DIK2709*	8.08	*DIK5201*	4.48	13
GIP	DQ120512	38.09	17q21.3-22	*BM17132*	0.6	*DIK5224*	0.61	19

the locations of genes on the human map were collected from http://www.ncbi.nlm.nih.gov/locuslink/

Association analysis of the GAD2 185-SNP and GIP 758-SNP

It is interesting the bovine GAD2 and GIP genes are located into several QTL regions in bovine chromosome 13 and 19, which have been indicated to affect body growth and carcass traits (http://bovineqtlv2.tamu.edu/home.php). A preliminary association study was performed to test the candidacy of GAD2 185-SNP and GIP 758-SNP as QTL contributors. Three genotypes, namely AA, AB and BB, were observed for both the *GAD2* and *GIP* genes among the animals investigated. The genotypes of 51 crossbreed steers were compared with their phenotypic data for 11 traits. Effects of different genotypes on these traits are given in Table 4. It is interesting to see that both the *GAD2* and *GIP* genes were significantly associated with ADFI ($p<0.05$). Animals with the AA and AB genotype of *GAD2* had a higher ADFI and BB had a lower ADFI (Table 4). For the *GIP* gene, it was the BB genotype that had a higher ADFI, while AA and AB had a lower ADFI. Besides feed intake, the *GAD2* 185-SNP was also significantly associated with Meat Percent (MP) ($p<0.05$), whereas the *GIP* SNP was also significantly associated with Backfat Thickness (BF) ($p<0.05$) and Ratio of Feed-

to-Meat (RFM) ($p<0.05$) (Table 4). Although we noticed differences among different genotypes of both the *GAD2* and *GIP* genes for the rest of the traits (DP, CW, ADG, BADG, FADG, SW, and IW), these differences were not statistically significant ($p\geq0.05$). Based on the data in Table 4, selection of animals with the BB genotype of *GAD2* would have ~ 7.8 % improvement in ADFI over the AA and AB genotypes. It also leads to ~ 4.6 % increase in MP. Selection of animals with the AB genotype of *GIP* would have ~ 18.8 % improvement in ADFI over the BB genotype. At the same time, it would also result in a ~ 35.5 % increase in RFM and ~ 15.8 % decrease in BF. Therefore, we propose that both the *GAD2* and *GIP* SNPs are markers for feed intake and could also be used as markers for carcass traits such as MP, BF and RFM.

Table 4
Effects of *GAD2* and *GIP* genotypes on phenotypic traits in beef cattle (Zusammenhänge zwischen *GAD2* und *GIP* Genotypen und phänotypischen Merkmale der Fleischrinder)

Genes	Genotypes	Traits[1] (mean ± SE)[2]					
		ADG (kg)	CW (kg)	DP (%)	MP (%)	BF (cm)	BADG (kg)
GAD2	AA	0.70±0.05	49.86±0.55	53.04±0.32	49.86^{a}±0.55	1.19±0.11	0.79±0.10
	AB	0.77±0.03	49.73±0.65	53.24±0.27	49.73^{a}±0.65	1.11±0.06	0.66±0.10
	BB	0.66±0.07	52.2±1.07	52.78±0.48	52.16^{b}±1.07	1.21±0.08	0.62±0.11
GIP	AA	0.74±0.03	50.36±0.54	53.20±0.32	62.68±2.52	1.23^{a}±0.07	0.69±0.06
	AB	0.81±0.05	49.73±1.71	52.9±0.46	66.5±3.03	0.95^{b}±0.10	0.56±0.26
	BB	0.74±0.06	50.23±0.55	53.15±0.24	65.6±2.12	1.10^{a}±0.07	0.72±0.08

Genes	Genotypes	Traits[1] (mean ± SE)[2]				
		FADG kg	RFM	ADFI kg	SW kg	IW kg
GAD2	AA	0.63±0.04	8.82±1.35	6.19^{a}±0.66	14.70±0.54	17.65±0.88
	AB	0.65±0.03	8.47±0.76	6.18^{a}±0.45	15.59±0.39	18.18±0.60
	BB	0.55±0.06	10.18±2.10	5.74^{b}±0.72	14.12±0.63	16.78±0.95
GIP	AA	0.62±0.03	8.64^{a}±0.86	5.90^{a}±0.44	15.07±0.45	17.43±0.65
	AB	0.68±0.06	7.53^{b}±1.29	5.68^{a}±0.81	15.72±0.49	19.35±0.91
	BB	0.61±0.05	10.2^{a}±1.48	6.75^{b}±0.64	14.82±0.56	17.14±0.74

[1] Average Daily Gain (ADG), Carcass Weight (CW), Dressing Percent (DP), Meat Percent (MP), Backfat Thickness (BF), Beginning Average Daily Gain (BADG; average daily gain in the test period from day 1 to day 75), Finishing Average Daily Gain (FADG; average daily gain in the test period from day 75 to day 180), Ratio of Feed-to-Meat (RFM), Average Daily Feed Intake (ADFI), Stomach Weight (SW), and Intestine Weight (IW).
[2] Within columns means with different superscripts indicate significant differences at $P < 0.05$.

Usually animals with higher feed intake have either a higher growth rate or a higher requirement for maintenance. The latter may be true for the Chinese cattle, as they, in general, have a lower growth rate in comparison to the Western beef breeds.
Although both SNPs could be used as candidate markers for MAS to improve feed intake and carcass traits, precautions must be taken when applying the markers, as the animal population used in this study is relatively small. In order to validate the findings present in this paper and to understand the effects of the *GAD2* and *GIP* polymorphisms on these quantitative traits and the efficiency of using these markers for MAS, further investigations in large populations and in other beef breeds are recommended.

Acknowledgements
This work was supported by the National High Technology Research and Development Program (2002AA242011) of China (863 Program).

References

AGRICULTURAL DEPARTMENT OF PEOPLE'S REPUBLIC OF CHINA.
Feeding standard of beef cattle NY/T 815-2004. Beijing, China, 8, 1-29, 2004

BOUTIN, P.; DINA, C.; VASSEUR, F.; DUBOIS, S., CORSET, L.; SERON, K.; BEKRIS, L.; CABELLON, J.; NEVE, B.; VASSEUR-DELANNOY, V.; CHIKRI, M.; CHARLES, M. A.; CLEMENT, K.; LERNMARK, A.; FROGUEL, P.:
GAD2 on chromosome 10p12 is a candidate gene for human obesity. PLoS Biol. **1** (2003), E68

BUCHAN, A. M.; POLAK, J. M.; CAPELLA, C.; SOLCIA, E.; PEARSE, A. G..:
Electronimmunocytochemical evidence for the K cell localization of gastric inhibitory polypeptide (GIP) in man. Histochemistry. **56** (1978), 37-44

CLEMENT, K.; BOUTIN, P.; FROGUEL, P.:
Genetics of obesity. Am J Pharmacogenomics. **2** (2002), 177-187

DAWSON, J. M.; GREATHEAD, H. M.; SESSIONS, V. A.; TYE, F. M.; BUTTERY, P. J.:
Effect of gastric inhibitory polypeptide on bovine fat metabolism. Comp Biochem Physiol B Biochem Mol Biol. **123** (1999), 79-88

DENBOW, D. M.:
Induction of food intake by a GABAergic mechanism in the turkey. Physiol Behav. **49** (1991), 485-488

ERDO, S. L.; WOLFF, J. R.:
gamma-Aminobutyric acid outside the mammalian brain. J Neurochem. **54** (1990), 363-372

ERLANDER, M. G.; TOBIN, A. J.:
A transcriptional regulatory element of the gene encoding the 67,000-M(r) form of human glutamate decarboxylase is similar to a Drosophila regulatory element. J Neurochem. **58** (1992), 2182-2190

EVERTS-VAN DER WIND, A.; KATA, S. R.; BAND, M. R.; REBEIZ, M.; LARKIN, D. M.; EVERTS, R. E.; GREEN, C. A.; LIU, L.; NATARAJAN, S.; GOLDAMMER, T.; LEE, J. H.; MCKAY, S.; WOMACK, J. E. &; LEWIN, H. A.:
A 1463 gene cattle-human comparative map with anchor points defined by human genome sequence coordinates. Genome Res. **14** (2004), 1424-1437

GUO, H.; LIU, W.S.; TAKASUGA, A.; EYER, K.; LANDRITO, E.; XU, S.Z.; GAO, X.; REN, H.Y.:
Mapping of the CCK, PSMC2, PSMC4, PSMD1, CPB1and PSPH genes in cattle. Anim. Genet. **37** (2005), 73-75

HAGER, J.; DINA, C.; FRANCKE, S.; DUBOIS, S.; HOUARI, M.; VATIN, V.; VAILLANT, E.; LORENTZ, N.; BASDEVANT, A.; CLEMENT, K.; GUY-GRAND, B.; FROGUEL, P.:
A genome-wide scan for human obesity genes reveals a major susceptibility locus on chromosome 10. Nat Genet. **20** (1998), 304-308

ITOH, T.; WATANABE, T.; IHARA, N.; MARIANI, P.; BEATTIE, C. W.; SUGIMOTO, Y.; TAKASUGA, A.:
A comprehensive radiation hybrid map of the bovine genome comprising 5593 loci. Genomics. **85** (2005), 413-424

LIU, W. S.; EYER, K.; YASUE, H.; ROELOFS, B.; HIRAIWA, H.; SHIMOGIRI, T.; LANDRITO, E.; EKSTRAND, J.; TREAT, M.; RINK, A.; YERLE, M.; MILAN, D.; BEATTIE, C. W.:
A 12,000-rad porcine radiation hybrid (IMNpRH2) panel refines the conserved synteny between SSC12 and HSA17. *G*enomics. **86** (2005), 731-738

MA, Y. H.; HU, J. H.; ZHOU, X. G.; ZENG, R. W.; MEI, Z. T.; FEI, J.; GUO, L. H.:
Transgenic mice overexpressing gamma-aminobutyric acid transporter subtype I develop obesity. Cell Res. **10** (2000), 303-310

MEIER, J. J.; GALLWITZ, B.; SIEPMANN, N.; HOLST, J. J.; DEACON, C. F. SCHMIDT, W. E.; NAUCK, M. A.:
Gastric inhibitory polypeptide (GIP) dose-dependently stimulates glucagon secretion in healthy human subjects at euglycaemia. Diabetologia. **46** (2003), 798-801

MEYRE, D.; BOUTIN, P.; TOUNIAN, A.; DEWEIRDER, M.; AOUT, M.; JOURET, B.; HEUDE, B.; WEILL, J.; TAUBER, M.; TOUNIAN, P.; FROGUEL, P.:
Is glutamate decarboxylase 2 (GAD2) a genetic link between low birth weight and subsequent development of obesity in children? J Clin Endocrinol Metab. **90** (2005), 2384-2390

OVESJO, M. L.; GAMSTEDT, M.; COLLIN, M.; MEISTER, B.:
GABAergic nature of hypothalamic leptin target neurones in the ventromedial arcuate nucleus. J Neuroendocrinol. **13** (2001), 505-516

POSPISILIK, J. A.; MARTIN, J.; DOTY, T.; EHSES, J. A.; PAMIR, N.; LYNN, F. C.; PITEAU, S.; DEMUTH, H. U.; MCINTOSH, C. H.; PEDERSON, R. A.:
Dipeptidyl peptidase IV inhibitor treatment stimulates beta-cell survival and islet neogenesis in streptozotocin-induced diabetic rats. Diabetes. **52** (2003), 741-750

SPIEGELMAN, B. M.; FLIER, J. S.:

Obesity and the regulation of energy balance. Cell. **104** (2001), 531-543

TRUMPER, A.; TRUMPER, K.; TRUSHEIM, H.; ARNOLD, R.; GOKE, B.; HORSCH, D.:
Glucose-dependent insulinotropic polypeptide is a growth factor for beta (INS-1) cells by pleiotropic signaling. Mol Endocrinol. **15** (2001), 1559-1570

WANG, Y. F.; YU, M.; YERLE, M.; LIU, B.; ZHAO, S. H.; XIONG, T. A.; FAN, B.; LI, K.:
Mapping of genes encoding four ATPase and three non-ATPase components of the pig 26S proteasome. Anim Genet. **34** (2003), 393-395

YANG, J.; LI, K.; LIU, B.; YERLE, M.; FAN, B.; XIONG, T.; YU, M.:
Assignments of GRB7, GCN5L2, COL1A1 and TBCD to pig chromosome 12 by radiation hybrid mapping. Cytogenet Genome Res. **103** (2003), 204L

YIP, R. G.; WOLFE, M. M.:
GIP biology and fat metabolism. Life Sci. **66** (2000), 91-103

ZHU, Z. M.; ZHANG, J. B.; LI, K.; ZHAO, S. H.:
Cloning, mapping and association study with carcass traits of the porcine SDHD gene. Anim Genet., **36** (2005), 191-195

Received: 2007-03-19

Accepted: 2007-09-14

Corresponding author
SHANG-ZHONG XU, Laboratory of Molecular Biology and Bovine Breeding, Institute of Animal Science, Chinese Academy of Agricultural Sciences, BEIJING 100094, P.R. CHINA

E-Mail: guohong882003@yahoo.com.cn or simmenta@vip.sina.com