The Journal of Animal & Plant Sciences, 27(3): 2017, Page: 756-762
ISSN: 1018-7081

ASSOCIATION OF TWO *SUMO3* GENE SINGLE NUCLEOTIDE POLYMORPHISMS WITH CARCASS AND MEAT QUALITY TRAITS OF THE CHINESE SIMMENTAL CATTLE

A. Li[1,#], Q. Qin[2,#], H. Xiao[1], X. Fang[1], P. Jiang[1], Z. Zhao[1] and R. Yang[1,*]

[1]College of Animal Science, Jilin University, Xi An Road 5333, Changchun, Jilin 130062, P. R. China;
[2]Department of Pharmaceutical Science and Technology / Animal Science, Heze University, Heze, Shandong, 274015, P. R. China.
[#]Aonan Li and Qiaomei Qin contributed equally to this work, and share the first authorship.
[*]Corresponding author E-mail: yrj@jlu.edu.cn

ABSTRACT

Small ubiquitin-related modifier 3(SUMO3) is a member of a growing family of ubiquitin-like proteins which is related to post-translational protein modification. Our previous results from gene microarray analysis showed that *SUMO3* gene might be associated with carcass and meat quality traits of cattle. In the present study, the association of two novel SNPs, I1-2653A>G and 3'UTR-1120T>C in the intron and 3'UTR region of bovine SUMO3, with 36 traits related to carcass and meat quality was investigated among 375 Chinese Simmental cattle. Statistical analysis demonstrated that the two SNPs were correlated with the carcass and meat quality traits. Individuals with the genotype AA of SUMO3-I1-2653A>G had significantly heavier carcass weight, thicker loin, higher fat coverage and larger eye muscle area (p<0.05). The individuals with genotype TT of SUMO3-3'UTR-1120T>C also showed a significant association with carcass weight, dressing percentage, back fat thickness, fat cover of carcass and rib eye area (p<0.05). SNPs in SUMO3 might be a valuable marker of meat quality traits for marker-assisted selection program in beef cattle breeding and production.

Keywords: *SUMO3*gene,Mutation,ChineseSimmentalcattle,Meatqualitytrait.

INTRODUCTION

Small Ubiquitin-like Modifier(SUMO), a small protein resembling ubiquitin in structure, can reversibly conjugate covalently to a lysine residue in target proteins through a process called sumoylation (Kroetz, 2005; Vertegaal, 2010) (Feng *et al*., 2013). Thus far, four members of SUMO family are discovered in vertebrate, SUMO1, SUMO2, SUMO3, and recently SUMO4 (Tatham *et al*., 2001). In higher mammals, SUMO2 and SUMO3 share 97% amino acid identity, but only 48% and 46% are identified with SUMO1 (Sekiyama *et al*., 2008). *SUMO3* gene is conserved from yeast to human (Takahashi *et al*., 2001).

Preliminary microarray analysis on the differences of gene expression in longissimus dorsi muscle tissue between 1 and 24 months Chinese Red Steppes have been performed by our lab(Qin *et al*., 2011), which indicated that *SUMO3*gene maybe related to beef quality traits in microarray analysis. However, there is no study report on the effects of *SUMO3* gene polymorphism on economic traits of domestic animals. The present study aims to validate whether *SUMO3* gene is associated with beef quality traits.

Single nucleotide polymorphisms(SNP), known as the third generation of molecular markers, play an important role in the study of animal genetics and breeding(Thomson *et al*., 1998). In the present study, the correlation between I1-2653A>G and 3'UTR-1120T>C of *SUMO3* gene with 36 traits related to carcass and meat quality was investigated among Chinese Simmental cattle population.

MATERIALS AND METHODS

Ethics statement: Animal experiments were carried out strictly in accordance with the guideline for the care and use of laboratory animals by the Jilin university animal care and use committee (Permit number: SYXK (ji) 2012-0010/0011). All operations are carried out under sodium pentobarbital anesthesia and every effort was made to reduce the pain. All measurements of carcass and meat quality were performed in the Meat Laboratory of Chinese Academy of Agricultural Sciences.

Materials:375 Chinese Simmental-cross steers that were crossbred with Simmental bull frozen semen were randomly selected from 15 cattle farms in Wulagai animal husbandry management district of Xilin Gol league in Inner Mongolia. The cattle were fed in two fattening farms(135 head of cattle in Bao Longshan fattening farm and 240 head of cattle in JinWeifuren fattening farm) and fattened under the same feeding and management conditions until 28 months of age. Blood samples (10mL each) were collected from jugular vein with anticoagulant ACD (Acid citrate dextrose) and

stored in -70℃. DNA was extracted from 1mL whole blood using the DNA extraction kit (TianGen, Beijing, China) based on the manufacturer's protocol.

Traitsanalyzed: The carcasses were stored in the freezing chamber for 24 hrs before all the carcass traits and meat quality traits were measured based on the cutting standard of fresh and chilled beef GB/T17238-1998 of China (China Standard Publishing House).

Certain traits, such as final body weight, living QIB, percentage intramuscular fat, rib eye area, live back-fat thickness, were measured before slaughter by ultrasound. Most of traits were recorded at the slaughter plant, for example, muscle color score, the weights of carcass, eye muscle area, carcass length, hind leg circumference, marbling score, fat color score, mesenteric fat, kidney fat, hind leg length, and so on. Muscle color was measured by colorimeter flesh or flesh-colored panels within the appropriate temperature and time (Fang *et al*., 2014; Tian *et al*., 2013).

Primers and PCR amplification: Primers were designed according to the published sequence of the bovine *SUMO3* gene (Gene Bank accession number: NM_001076449) with Primer Premier 5. The primer pair was synthesized by Sangon biology company (Shanghai, China). The primer sequences were shown in table 1. Which targeted the first intron region P1 (from 2554 bp to 3068 bp of the first intron region for *SUMO3* gene) and 3'UTR region P2 (from 195 bp to 1352 bp of the 3'UTR region for *SUMO3* gene). PCR was performed in a 25 μL mixture: 100 ng bovine genomic DNA, 4 pmol of each primer, 12.5 μL 2×Taq PCR Green Mix (Dingo, Beijing, China).

PCR-RFLP analysis: The PCR products of *SUMO3* gene were subjected to PCR-RFLP analysis. PCR products P1 (I1-2653A>G) were digested with MseI (Thermo, USA) enzymes in a 20 μL reaction with 8 μL PCR products; 0.6 U of MseI, and 2 μL 10×buffer. PCR products P2 (3'UTR-1120T>C) were digested with Bc1I (New England BioLabs, USA) enzymes in a 20 μL reaction with 6 μL PCR products; 0.5 U of Bc1I, and 2 μL 10×NEBbuffer3.1.The enzyme digested products were detected by electrophoresis on 2% agarose gels. EB stained agarose gels were analyzed with a Multimage™ Light Cabinet Filter Positons gel imaging system (Alpha Innotech, US).

Statistical analysis: The Hardy–Weinberg equilibrium of the mutation was determined by Chi squared ($\times^2$) test. Values of the genotype frequencies were calculated for the examined Chinese Simmental Cattle and were analyzed by the significance test. Associations of *SUMO3* gene polymorphisms with the carcass traits were carried out using two-way ANOVA with SPSS 13.0. The fixed model was:

$Y_{ijk} = u + ff_i + m_j + e_{ijk}$

Where, Y_{ijk} is the observed value of kth individual from the Simmental breed, of genotype j, in the i fattening farm; u is the least square means of the observed values; ffi is the effective value of the i fattening farm; mj is the effective value of the genotype j; and eijk is the random residual effect corresponding to the observed value (Fang *et al*., 2014).

RESULTS

***SUMO3* PCR amplification:** P1 (524 bp) and P2 (1158 bp) of *SUMO3* gene were PCR amplified from the 375 Chinese Simmental cattle, as shown in figure.1-A and figure.2-A. The PCR amplified product was directly used for polymerase chain reaction-restriction fragment length polymorphism.

RFLP analysis and sequencing of different genotypes: PCR products from the DNA mixture of 30 cattle were sequenced. Polymorphisms were found in the PCR products. As shown in figure 1-B, an A/G peak at I1-2653 appeared, which suggested a SNP site of an A/G substitution at position 2653 bp of *SUMO3* gene intron1. The result of sequencing showed that there was a recognition site of MseI in GG genotype, not in AA genotype. The PCR products were further analyzed with PCR-RFLP resulting in three genotypes, named AA (figure.1-C, electrophoresis path 6 and 9), GA (figure.1-C, electrophoresis path 3,4 and10) and GG (figure.1-C, electrophoresis path 1,2,5 and 8). GG genotypes had two bands, 424 bp and 100 bp; GA genotypes had three bands, 524 bp, 424 bp and 100 bp; AA genotypes had only one band, 524 bp.

A C/T peak at 3'UTR-1120 of SUMO3 was found, which suggested a SNP site of a C/T substitution, as shown in figure.2-B. There was a recognition site of Bc1I in CC genotype from the result of sequencing. The PCR products were further analyzed with PCR-RFLP resulting in three genotypes, named CC(figure.2-C, electrophoresis path 1,2,4,6 and 8), TC(figure.2-C, electrophoresis path 3, 5 and 7) and TT(figure.2-C, electrophoresis path 9 and 10). CC genotypes had two bands, 924 bp and 235 bp; TC genotypes had three bands, 1158 bp, 924 bp and 235 bp; TT genotypes had only one band, 1158bp. As shown in table2, allele A had a frequency of 0.70 at I1-2653A>G polymorphism site in Chinese Simmental cattle population. At 3'UTR C>T polymorphism site, allele T had a frequency of 0.71.

Association analysis of *SUMO3* SNPs with the carcass and meat quality traits: In this study, the associations of *SUMO3* gene polymorphisms with the carcass and meat quality traits were analyzed by Two-Way ANOVA using SPSS 13.0. Our data suggested that two SNPs (Intron1 2653A>G and 3'UTR-1120C>T) were significantly associated with the carcass and meat quality traits.

Our data revealed that cattle with the A allele

homozygotes of Intron1 2653A>G had longer hind leg length (average hind leg length of each genotypes is 80.27+4.37 cm, $p<0.05$), hind leg circumference (49.33+3.45 cm, $p<0.05$) and loin thickness (6.94+0.96 cm, $p<0.05$), heavier carcass (259.96+38.17 kg, $p<0.05$), larger eye muscle area (81.34+ 13.29 cm^2, $p<0.05$) and fat coverage of carcass (49.32+20.47%, $p<0.01$) than cattle with the G allele homozygotes(78.00+4.54 cm, 47.29+2.85cm, 6.71+0.92 cm, 242.27+38.78 kg, 75.61+10.96 ^{cm2}, 42.65+21.45%). Among them, Individuals with the A allele homozygotes also had longer loin meet thickness and higher eye muscle area than those with the AG heterozygotes in population ($p<0.05$). And cattle with the AG heterozygotes of *SUMO3* gene had higher score of fat coverage than those with the G allele homozygotes ($p<0.05$).

For 3'UTR SNPs of *SUMO3* gene, individuals with the TT genotype was significantly associated with higher dressing percentage, hind leg circumference, eye muscle area ($p<0.05$) than those with the C allele homozygotes. Cattle with the TC heterozygotes also had lower hind hoof weight ($p<0.01$), higher back fat thickness ($p<0.05$) and higher fat coverage of carcass ($p<0.05$) than those with the C allele homozygotes. In addition, individuals with the TC heterozygotes had lower carcass weight than that of TT homozygotes in simmental cattle population($p<0.05$).

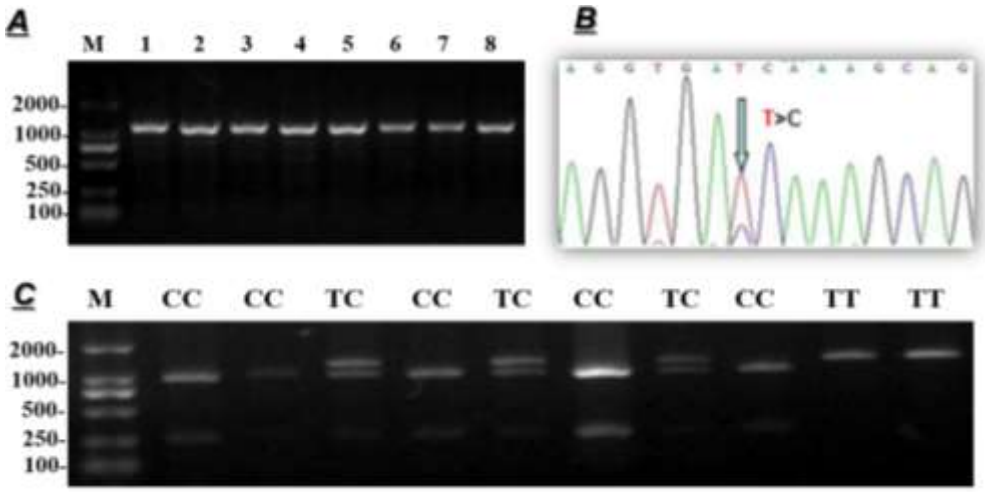

Figure1.Sequencing and PCR-RFLP analysis I1-2653A>G of bovine *SUMO3* gene.
A: PCR amplification result; B: Sequence resultof PCR products from 30 cattle DNA mixture; C: PCR-RFLP detection of *SUMO3* gene, a A/G substitution at position 2653bp of *SUMO3* gene intron1.

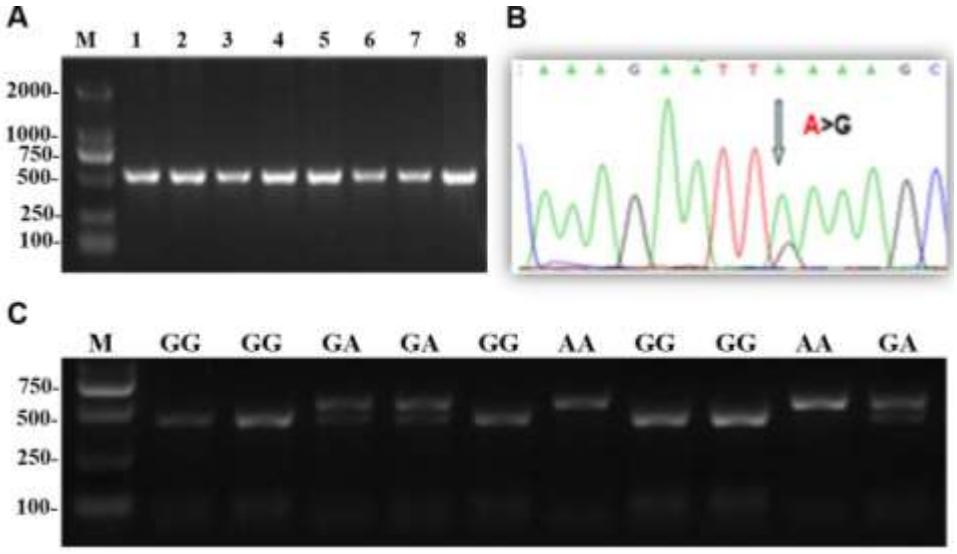

Figure 2. Sequencing and PCR-RFLP analysis 3'UTR-1120T>C of bovine *SUMO3* gene.
A: PCR amplification result; B: Sequence resultofPCR products from 30 cattle DNA mixture; C: PCR-RFLP detection of *SUMO3* gene, a T/C substitution at position 1119bp of *SUMO3* gene 3'UTR.

Table1. The conditions of *SUMO3* gene PCR amplification.

Name	Primer sequence	Conditions
MseI-f	5’GCAAGTGCTGGAGAGGATGTG3’	35cycles
MseI-r	5’GCCACTGACAACCACCATTCTTC3’	(95℃ 5min, 64℃ 30s, 72℃ 50s)
BcII-f	5'AGAAGGACTTCGCTCTGATGC 3'	35cycles
BcII-r	5'GATGTCCAATCTCCAAACGCAG 3'	(95℃ 5min, 60℃ 30s, 72℃ 1min)

Table 2. Genotypes and allele frequencies of *SUMO3* gene polymorphisms in Chinese Simmental cattle population.

SNPs	Allele Frequency		Genotype Frequency			P	χ^2
3’UTR-1120T>C	T	C	TT	TC	CC	0.36	0.84
	0.71	0.29	0.49	0.43	0.08		
I1-2653A>G	A	G	AA	AG	GG	0.00	15.96
	0.70	0.30	0.44	0.51	0.05		

Table 3. Association analyses of *SUMO3* gene polymorphisms with carcass and meat quality traits in Chinese Simmental cattle population.

Trait	Genotypes (I1-2653A>G)						Genotypes (3’UTR-1120C-T)					
	AA (n=159)		AG (n=184)		GG (n=17)		CC (n=28)		CT (n=161)		TT (n=183)	
	LSM	SE	LSM	SE	LSM	SE	LSM	SE	LSM	SE	LSM	SE
CW	259.96*	38.17	256.21	38.68	242.27	38.78	247.33	36.20	252.26	38.26	260.54*	38.95
DP	52.41	2.51	52.00	2.32	51.78	2.32	51.32	3.49	51.93	2.33	52.35*	2.18
FW	5.98	0.73	5.94	0.67	5.76	0.48	5.86	0.48	5.86	0.68	5.97	0.72
HHW	3.48	1.05	3.50	0.99	3.43	1.04	3.73	1.19	3.43**	1.01	3.49**	1.02
HLL	80.27*	4.37	80.50	3.88	78.99	4.54	80.73	4.09	80.05	4.26	80.39	4.06
HLC	49.33*	3.45	48.86	4.09	47.29	2.85	47.50*	2.66	48.70	3.87	49.38	3.75
TMT	18.02	1.71	17.83	1.70	18.04	1.62	18.00	1.34	17.84	1.64	17.89	1.80
MCS	5.70	1.06	5.57	1.06	5.29	1.05	5.64	1.03	5.66	1.05	5.64	1.03
FCS	2.70	0.94	2.71	0.99	2.94	1.20	2.86	1.15	2.69	0.96	2.72	0.94
LMT	6.94*	0.96	6.79	0.88	6.71	0.92	6.76	0.90	6.84	0.90	6.86	0.92
BFT	1.00	0.63	0.95	0.64	0.83	0.52	0.78	0.55	0.97*	0.64	0.99**	0.64
GW	495.15	62.23	491.19	59.25	466.82	62.73	481.00	52.96	484.57	61.98	496.39	61.68
FCR	49.32**	20.47	49.15*	22.45	42.65	21.45	43.54	22.57	48.75*	21.45	48.89**	21.53
Mb	5.44	0.70	5.34	0.70	5.41	0.71	5.54	0.64	5.40	0.71	5.36	0.73
RRA	7.56*	0.95	7.53*	0.93	7.13	0.97	7.67	0.93	7.51	1.01	7.49	0.87
Om	4.00*	0.96	3.95	0.72	3.67	0.72	4.17*	0.73	3.92	0.74	3.97	0.65
ADPH	5.59	0.34	5.56	0.33	5.64	0.47	5.58	0.35	5.56	0.34	5.58	0.34
SPH	6.25	0.48	6.24	0.54	6.28	0.58	6.29	0.56	6.27	0.54	6.29	0.56
EMA	81.34	13.29	78.39*	12.72	75.61*	10.96	74.79	9.82	78.52	12.69	80.29*	13.18
Testis	0.69	0.14	0.65*	0.15	0.61*	0.16	0.66	0.13	0.65	0.16	0.68	0.14
NBW	20.60	3.50	19.96	2.76	20.31	2.49	20.53	2.66	19.79	2.93	20.41	3.31
HW	23.65	2.42	23.63	2.46	22.92	2.58	23.44	2.71	23.47	2.58	23.59	2.31
Tare	42.46	6.27	42.20	6.35	40.55	5.14	43.13	6.97	41.87	6.32	42.35	6.13
Heart	1.83	0.36	1.87	0.34	1.72	0.31	1.72*	0.33	1.78*	0.33	1.85	0.37
Liver	5.95	1.09	5.88	1.12	5.81	1.28	5.79	1.07	5.81*	1.07	5.97	1.15
LT	3.13	0.49	3.17	0.47	3.01	0.39	3.09	0.42	3.12	0.47	3.16	0.50
Kidney	1.17	0.20	1.17	0.22	1.15	0.18	1.16	0.18	1.15	0.20	1.18	0.22
KF	4.71	2.85	4.69	2.71	4.62	3.07	4.24	2.87	4.79	2.90	4.64	2.74
BW	0.44	0.09	0.44	0.08	0.42	0.08	0.46	0.08	0.43	0.09	0.45	0.08
GF	0.89	0.36	0.90	0.35	0.93	0.41	0.97	0.40	0.89	0.34	0.89	0.36
Spleen	0.86	0.17	0.86	0.20	0.81	0.18	0.84	0.15	0.84	0.18	0.86	0.19
Oxtail	1.36	0.25	1.36	0.24	1.32	0.24	1.31	0.22	1.34	0.25	1.37	0.25
CL	139.67	8.74	140.14	7.70	138.18	8.85	140.57	9.20	139.74	8.85	139.81	7.38

CD	64.31	3.34	64.65	3.33	64.29	2.11	64.48	3.04	64.35	3.35	64.57	3.18
CCD	65.46	3.78	65.42	3.58	64.09	3.01	64.91	2.82	65.27	4.03	65.58	3.43
HLW	44.87	2.73	44.51	2.55	44.26	3.49	44.71	2.27	44.41	2.86	44.77	2.49

Significance of difference from each other: the symbol** indicate extremely significant difference ($p<0.01$); the symbol * indicate significant difference ($p<0.05$). LSM, least square mean; SE, standard error of mean. Traits: CW, carcass weight; DP, dressing percentage; FW, fore-hoof weight; HHW: hind-hoof weight; HLL: hind leg length; HLC, hind leg circumference; TMT, thigh meat thickness; LMT, loin meat thickness; BFT, back-fat thickness; GW, gross weight; FCR: fat coverage rate; Mb: marbling; RRA: rumen reticulum and abomasum; Om: omasum; ADPH: acid discharge PH; SPH: slaughter PH; EMA: eye muscle area; NBW: net bone weight; HW: head weight; LT: lung, trachea; KF: kidney fat; BW: bullwhip; GF: genital fat; CL: carcass length; CD: carcass depth; CCD: carcass chest depth; HLW: hind legs wide.

DISCUSSION

Traditionally, beef quality traits were mainly defined by indicators such as muscle pH, tenderness, back-fat thickness, rib eye area, marbling, meat color, muscle color score and etc(Maltin *et al*., 2003). However, regular measurements of these indicators for meat quality traits are not accurate, which lead to alternative strategies such as molecular marker assisted selection (Casas *et al*., 2005; Hou *et al*., 2011). In our study, two polymorphic loci, one in the first intron 27 and the other in the 3'UTR of the *SUMO3* gene were found to be significantly or extremely significantly associated with the carcass weight, hind leg circumference, hind leg length, loin thickness, fat coverage of carcass, rib eye area, dressing percentage, hind hoof weight and back fat thickness. All these traits have been reported to be associated with meat and carcass quality.

Carcass quality, one of the most important indicators for the economic value of beef cattle, had become the most important performance measurement of beef cattle. Carcass quality includes hot carcass weight, cold carcass weight, dressing percentage, meat percentage, carcass fat cover, loin thickness, back-fat thickness and other parts of slaughtered meat production traits. Ultrasonic measurement is generally used for in vivo intramuscular fat content, back and flesh, back-fat thickness, loin eye area, buttocks fat thickness traits; As a complex trait, meat quality is usually graded by various indicators, such as muscle color score, tenderness, intramuscular fat content, marbling, fat color score, line or dripping water loss, carcass level, pH, flavor and other indicators(Doran *et al*., 2014; San Vito *et al*., 2014).

SUMO is a newly discovered ubiquitin-like proteins participating in protein post-translational modification (Anckar&Sistonen, 2007; Benson *et al*., 2007). SUMO mediated reversible modification of target proteins involved in target protein positioning and functional regulatory process, but not proteasome mediated degradation of target protein (Eaton & Sealy, 2003; Yang *et al*., 2003). Kristen N *et al*, had been reported that SUMO was associated with high grade breast cancer risk (Stevens *et al*., 2011). SUMO was confirmed to be associated with tumorigenesis, inflammatory reaction, and other human diseases (Kroetz, 2005; Sarge& Park-Sarge, 2011). However, there have been very few reports about the association of SUMO genes with carcass and meat quality traits. The preliminary job of our team showed that SUMO3 perhaps associated with meat quality and carcass traits by gene microarray sequencing. The study that association of *SUMO3* gene SNPs with carcass and meat quality traits had been examined. The intron region contented some regulatory sequence, may be involved in gene expression. Introns plays a modified role for the efficiency of gene transcription. And the UTR (untranslated regions) were of transcriptional regulation function. Regulation of gene expression is an important part of the growth and development of the organism. Thus the non-coding region was essential for gene function. So the intron 1 and 3'UTR of *SUMO3* gene were chosen for further study.

Our results showed that intron1 2653A>G and 3'UTR-1120T>C of bovine *SUMO3* gene were significantly associated with the carcass and meat quality traits. Intron1 2653A>G of SUMO3 was significantly corresponding with carcass weight, hind leg circumference, hind leg length, loin thickness, fat coverage of carcass, and eye muscle area ($p<0.05$). Chi-square test result showed that the population in the 3'UTR-1120T>C sites do not conform to the Hardy-Weinberg equilibrium, which could be attributed to nonrandom mating or selections existed in this Chinese Simmental cattle population. It is demonstrated that A allelomorphic gene of intron1 2653A>G was the dominance gene in Chinese Simmental cattle. A allele of Intron1 2653A>G individuals were better than that of other individuals in carcass and meat quality traits.

For 3'UTR-1120T>C of *SUMO3* gene, CC genotype was correlated with carcass weight, dressing percentage, hind hoof weight, hind leg circumference, back fat thickness, fat of coverage, and eye muscle area ($p<0.05$). The chi-square test indicated that the locus was in accordance with Hardy-Weinberg equilibrium. Our data also revealed that CC genotype had better measurements for hind hoof weight, back fat thickness, fat coverage and eye muscle area, which suggested that C allelomorphic gene of 3'UTR-1120T>C was the dominance gene in Chinese Simmental cattle on these traits. In addition, T allele was associated with better

traits in carcass weight, dressing percentage and hind leg circumference.

In summary, *SUMO3* gene polymorphism in Chinese Simmental cattle is associated with carcass and meat quality traits, which could be further explored to be used in marker-assisted selection in beef cattle breeding.

Acknowledgments: This work was supported by the National High Technology Research and Development Program (863 Program, no. 2013AA102505), the National Natural Science Foundation of China (no. 31372278 and 31672389), the Jilin Scientific and Technological Development Program (No. 20170519014JH), and the National R & D Project of Transgenic Organisms of Ministry of Science and Technology of China (2016ZX08009003-006).

REFERENCES

Anckar, J. and L. Sistonen (2007). SUMO: getting it on. Biochem Soc Trans,35(Pt6),1409-13.

Benson, M. D., Q. J. Li, K. Kieckhafer, D. Dudek, M. R. Whorton, R. K. Sunahara, J. A. Iniguez-Lluhi, and J. R. Martens (2007). SUMO modification regulates inactivation of the voltage-gated potassium channel Kv1.5. Proc Natl Acad Sci U S A,104(6):1805-10.

Casas, E., S. N. White, D. G. Riley, T. P. Smith, R. A. Brenneman, T. A. Olson, D. D. Johnson, S. W. Coleman, G. L. Bennett and C. C. Jr. Chase (2005). Assessment of single nucleotide polymorphisms in genes residing on chromosomes 14 and 29 for association with carcass composition traits in Bos indicus cattle. J Anim Sci, 83(1): 13-9.

Doran, A. G., D. P. Berry C. J. and Creevey (2014). Whole genome association study identifies regions of the bovine genome and biological pathways involved in carcass trait performance in Holstein-Friesian cattle. BMC Genomics,15(1):837.

Eaton, E. M. and L. Sealy (2003). Modification of CCAAT/enhancer-binding protein-beta by the small ubiquitin-like modifier (SUMO) family members, SUMO-2 and SUMO-3. J Biol Chem,278(35):33416-21.

Fang, X. B., L. P. Zhang, X. Z. Yu, J. Y. Li, C. Y. Lu, Z. h. Zhao, and R. J. Yang (2014). Association of HSL gene E1-c.276C>T and E8-c.51C>T mutation with economical traits of Chinese Simmental cattle. Mol Biol Rep,41(1):105-12.

Feng, Z. J., B. Gurung, G. H. Jin, X. L. Yang and X. X. Hua (2013). SUMO modification of menin. Am J Cancer Res,3(1):96-106.

Hou, G. Y., Z. R. Yuan, H. L. Zhou, L. P. Zhang, J. Y. Li, X. Gao, D. J. Wang, H. J. Gao and S. Z. Xu (2011). Association of thyroglobulin gene variants with carcass and meat quality traits in beef cattle. Mol Biol Rep,38(7):4705-8.

Kroetz, M. B. (2005). SUMO: a ubiquitin-like protein modifier. Yale J Biol Med, 78(4):197-201.

Maltin, C., D. Balcerzak, R. Tilley and M. Delday (2003). Determinants of meat quality: tenderness. Proceedings of the Nutrition Society, 62(2):337-347.

Qin, L. H., G. L. Zhang, Y. Cao, J. B. Zhang, Y. M. Zhao and Z. H. Zhao (2011). Microarray Analysis on the Differences of Gene Expression in Longissimus Dorsi Muscle Tissue Between 1 and 24 Months Chinese Red Steppes. J. Anim. Vet. Advances, 10(4):428-436.

San Vito, E., J. F. Lage, A. F. Ribeiro, R. A. Silva and T. T. Berchielli (2014). Fatty acid profile, carcass and quality traits of meat from Nellore young bulls on pasture supplemented with crude glycerin. Meat Sci, 100C:17-23.

Sarge, K. D., and O. K. Park-Sarge (2011). SUMO and its role in human diseases. Int Rev Cell Mol Biol, 288: 167-83.

Sekiyama, N., T. Ikegami, T. Yamane, M. Ikeguchi, Y. Uchimura, D. Baba, M. Ariyoshi,H. Tochio, H. Saitoh, and M. Shirakawa (2008). Structure of the small ubiquitin-like modifier (SUMO)-interacting motif of MBD1-containing chromatin-associated factor 1 bound to SUMO-3. J Biol Chem, 283(51):35966-75.

Stevens, K. N., X. Wang, Z. Fredericksen, V. S. Pankratz, J. Cerhan, C. M. Vachon, J. E. Olson and F. J. Couch (2011). Evaluation of associations between common variation in mitotic regulatory pathways and risk of overall and high grade breast cancer. Breast Cancer Res Treat, 129(2):617-22.

Takahashi,Y., A. Toh-e, and Y. Kikuchi (2001). A novel factor required for the SUMO1/Smt3 conjugation of yeast septins.Gene, 275(2):223-31.

Tatham, M. H., E. Jaffray, O. A. Vaughan, J. M. Desterro, C. H. BottingJ. H. Naismith and R. T. Hay (2001). Polymeric chains of SUMO-2 and SUMO-3 are conjugated to protein substrates by SAE1/SAE2 and Ubc9. J Biol Chem, 276(38):35368-74.

Thomson, J. A., J. Itskovitz-Eldor, S. S. Shapiro, M. A. Waknitz, J. J. Swiergiel, V. S. Marshall and J. M. Jones (1998). Embryonic stem cell lines derived from human blastocysts. Science, 282 (5391): 1145-7.

Tian, J., Z. Zhao, L. Zhang, Q. Zhang, Z. Yu, J. Li and R. Yang (2013). Association of the leptin gene E2-169T>C and E3-299T>A mutations with carcass

and meat quality traits of the Chinese Simmental-cross steers. Gene,518(2):443-8.

Vertegaal, A. C. (2010). SUMO chains: polymeric signals. Biochem Soc Trans, 38(Pt1):46-9.

Yang, S. H., E. Jaffray, B. Senthinathan, R. T. Hay and A. D. Sharrocks (2003). SUMO and transcriptional repression: dynamic interactions between the MAP kinase and SUMO pathways. Cell Cycle, 2(6):528-30.