
ASSOCIATION OF SINGLE NUCLEOTIDE POLYMORPHISM IN *TG*, *LEP* AND *SCD1* GENES WITH CARCASS TRAITS IN HIGHLAND AND GALLOWAY CATTLE

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ABSTRACT

This work deals with the influence of selected polymorphisms in *TG*, *LEP* and *SCD1* genes on quality of beef (composition and fatty acids content).

Testing of selected polymorphisms for meat quality was carried out on 19 bulls and 58 mothers of extensive breeds of cattle (Galloway and Highland). For detection of SNPs PCR-RFLP method was used. In a group of 19 bulls and 58 mothers the frequency of alleles and genotypes was detected.

Based on the association analysis no significant influence was found on meat composition with emphasis on IMF content. The polymorphisms in *TG*, *LEP* and *SCD1* genes differs significantly in fatty acid composition. C422T polymorphism of the *TG* gene has a significant effect ($p < 0.05$) on myristic acid content, C73T polymorphism of the *LEP* gene has a highly significant effect ($p < 0.01$) in palmitic acid content and a significant effect ($p < 0.05$) on myristoleic acid content, C878T polymorphism of the *SCD1* gene has a significant effect ($p < 0.05$) on myristic acid, linoleic acid, γ -linolenic acid, α -linolenic acid content, CLA and total omega 6 fatty acids content.

Key words: cattle, meat quality, *TG*, *LEP*, *SCD1*

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INTRODUCTION

Meat quality is defined by those traits the consumer perceives as desirable which includes both visual and sensory traits and credence traits of safety, health and more intangible traits such as 'clean' and 'green' or welfare status of the production system (Warner *et al.*, 2010). Meat quality depends on organoleptic properties, such as colour, texture, flavour and juiciness, which are related to zootechnical characteristics such as breed, age and sex, anatomical characteristics such as type of muscle, characteristics of handling and feeding or technological characteristics (de Huidobro *et al.*, 2003).

Fat, especially animal fat, has been the subject of much interest and debate because of risks of some diseases when consumed in excess. Fat however is not only a concentrated source of energy for the body, the fat in meat provides flavour, aroma and texture (Nuernberg *et al.*, 2005). Marbling is a quantitative trait that is controlled by interactions among several quantitative trait loci (QTLs) combined with environmental influences. Beef industry is looking for gene markers that would identify animals that have a high propensity to accumulate intramuscular fat in order to produce tasty and tender meat (Lim *et al.*, 2011).

Consumers are becoming more aware of the relationships between diet and health and this has increased consumer interest in the nutritional value of food. This is impacting on the demand for food which contain functional components that play important roles in health maintenance and disease prevention (Scollan *et al.*, 2006). The fatty acid composition in beef has emerged as an economically relevant trait to the beef industry due to increasing consumer awareness of the health implications of fat intake associated with red meat consumption. Accumulated evidence suggests the type of dietary fat (or the fatty acid composition) has a more profound impact on human health than the amount of fat in the diet (Han *et al.*, 2013).

MATERIAL AND METHODS

In total 20 head of Highland breed (3 bulls and 17 mothers) and 57 head of Galloway breed (16 bulls and 41 mothers) has been tested.

DNA samples were extracted from meat or hair bulbs samples by GenElute™ Mammalian Genomic DNA Miniprep Kit (Sigma-Aldrich Corp., St Louis, USA) and stored at -20°C until genotyping.

The PCR-RFLP method was used to detect the genotypes in *TG*, *LEP* and *SCD1* genes. Methodologies for marker use were adopted from the work of Barendse *et al.* (2001) for *TG* gene, Buchanan *et al.* (2002) for *LEP* gene and Tsuji *et al.* (2004) for *SCD1* gene. After digestion of PCR products the DNA fragments were separated on 2-3% agarose gels in electrophoresis visualised by ethidium bromide.

The statistical analysis was performed by the general linear model (GLM) by SAS v8.2 for Windows 9.1.4. (SAS Institute Inc., Cary, USA). The genotypes of relevant genes (Gen_i), SEURO classification (tz_j) and fat classification (tp_k) were used as fixed effect, age ($b * vek_l$) and total weight ($b * phm_m$) were used as regression.

$$y_{ijklmn} = \mu + Gen_i + tz_j + tp_k + b * vek_l + b * phm_m + e_{ijklmn}$$

RESULT AND DISCUSSION

The first objective of this study was to discover the frequency of alleles and genotypes in selected populations. All three markers were polymorphic in selected population. The observed frequencies of *TG*, *LEP* and *SCD1* markers were in accordance with various authors (Casas *et al.*, 2005; Barendse *et al.*, 2005; Milanese *et al.*, 2008).

Tab. 1 Number of animals with genotype of genes and allelic frequency of markers

Gene	Genotype count			Allelic frequency	
	CC	CT	TT	C	T
<i>TG</i>	CC	CT	TT	C	T
	68	9	0	0,942	0,058
<i>LEP</i>	CC	CT	TT	C	T
	5	35	37	0,292	0,708
<i>SCD1</i>	CC	CT	TT	C	T
	1	32	44	0,221	0,779

For *TG5* marker of *TG* gene was reported, that animals with *TT* genotype had significantly higher marbling score than the animals with the genotype *CC* and *CT* (Burrell *et al.*, 2004). On the contrary, Shin & Chung (2007) in their study assigned higher marbling in Korean cattle to *CC* and *CT* genotypes ($p < 0.05$). In this study there was no significant difference in IMF. Significant difference was only in the content of myristic acid ($p < 0.05$).

Tab. 2 Association analysis for selected indicators of *TG5* marker

Trait	Polymorphism <i>TG</i> (C422T)		p-value
	CC (n = 17)	CT (n = 2)	
	LSM ± SE	LSM ± SE	
IMF (%)	1,670 ± 0,219	2,500 ± 0,577	0,0735
Protein (%)	23,57 ± 0,206	23,80 ± 0,541	0,5697
C14:0	2,639 ± 0,082	3,001 ± 0,218	0,0430
C14:1	0,166 ± 0,029	0,084 ± 0,077	0,1698
C16:0	27,67 ± 0,600	27,32 ± 1,578	0,7716
C16:1	2,074 ± 0,198	1,650 ± 0,521	0,2889
C18:0	24,94 ± 1,692	28,09 ± 4,447	0,3522
C18:1, n9c	36,12 ± 1,383	32,81 ± 3,636	0,2387

C73T polymorphism in exon 2 of the *LEP* gene with impact on IMF and marbling score (Buchanan *et al.*, 2002) has been studied in many breeds and populations in independent studies by many authors around the world. Buchanan *et al.* (2002) demonstrated a significant difference between this polymorphism and carcass fat content. Allele *C* was associated with a lower fat content, and conversely the *T* allele with a higher fat content (Kononoff *et al.*, 2005). In this study significant differences in the content of palmitic acid ($P < 0.01$) and myristoleic acid were found ($p < 0.05$) but no significant influence on IMF content.

Tab. 3 Association analysis for selected indicators of *LEP* marker

Trait	Polymorphism <i>LEP</i> (C73T)			p-value		
	CC (n = 4)	CT (n = 11)	TT (n = 4)	CC-	CC-	CT-
	LSM ± SE	LSM ± SE	LSM ± SE	CT	TT	TT
IMF (%)	1,584 ± 0,236	1,392 ± 0,281	1,117 ± 0,292	0,538	0,197	0,254
Protein (%)	23,44 ± 0,212	23,57 ± 0,252	23,60 ± 0,262	0,641	0,621	0,902
C14:0	2,609 ± 0,092	2,438 ± 0,110	2,420 ± 0,114	0,174	0,182	0,838
C14:1	0,152 ± 0,022	0,256 ± 0,026	0,239 ± 0,027	0,003	0,018	0,447
C16:0	28,65 ± 0,376	26,73 ± 0,448	26,43 ± 0,467	0,002	0,002	0,434
C16:1	2,078 ± 0,180	2,520 ± 0,214	2,301 ± 0,223	0,08	0,408	0,236
C18:0	24,47 ± 1,697	22,38 ± 2,022	23,80 ± 2,106	0,355	0,790	0,408

The substitution C878T of *SCD1* gene has been proved to associate alanine with increasing percentage of MUFA ($p < 0.001$) contrary to valine (Taniguchi *et al.*, 2004). Ohsaki *et al.* (2009) found an association of this mutation with the composition of fatty acids. In this study, C878T polymorphism of *SCD1* gene had significant effect ($p < 0.05$) on the content of the myristic, linoleic, γ -linolenic, α -linolenic acid, CLA and total omega 6 fatty acids content.

Tab. 4 Association analysis for selected indicators of *SCD1* marker

Trait	Polymorphism <i>SCD1</i> (C878T)		p-value
	CT (n = 8)	TT (n = 10)	
	LSM \pm SE	LSM \pm SE	
IMF (%)	0,873 \pm 0,369	1,262 \pm 0,223	0,130
Protein (%)	23,48 \pm 0,349	23,50 \pm 0,211	0,938
C14:0	2,232 \pm 0,115	2,442 \pm 0,069	0,016
C18:2, n6c	3,463 \pm 0,384	2,813 \pm 0,232	0,023
C18:3, n6	0,260 \pm 0,026	0,222 \pm 0,015	0,042
C18:3, n3	2,290 \pm 0,242	1,883 \pm 0,146	0,024
C18:2, n9	0,188 \pm 0,132	0,390 \pm 0,080	0,037
n6	4,216 \pm 0,557	3,427 \pm 0,336	0,050

CONCLUSIONS

Recommendation based on these results would be to keep those animals, which has at least one *T* allele in *TG* gene, at least one allele *C* in *LEP* gene and at least one allele *T* in *SCD1* gene, in population to produce better meat in terms of fatty acid composition and impact on human health.

REFERENCES

- BARENDSE, W., BUNCH, R., THOMAS, R., ARMITAGE, S., BAUD, S., DONALDSON, N., 2001: The TG5 DNA marker test for marbling capacity in Australian feedlot cattle. Marbling Symposium.
- BARENDSE, W., BUNCH, R.J., HARRISON, B.E., 2005: The LEP C73T missense mutation is not associated with marbling and fatness traits in a large gene mapping experiment in Australian cattle. *Animal Genetics* 36: 86 – 88.
- BUCHANAN, F.C., FITZSIMMONS, C.J., VAN KESSEL, A.G., THUE, T.D., WINKELMAN-SIM, D.C., SCHMUTZ, S.M., 2002: Association of a missense mutation in the bovine leptin gene with carcass fat content and leptin mRNA levels. *Genetics Selection Evolution* 34: 105 – 116.
- BURRELL, D.N., MOSER, G.H.D., HETZEL, J., MIZOGUCHI, Y.S.S., HIRANO, T.K.S., SUGIMOTO, Y.S.K.Z., Mengersen, K.R., 2004: Meta analysis confirms associations of the TG5 thyroglobulin polymorphism with marbling in beef cattle. 29th International Conference on Animal Genetics ISAG 2004/TOKYO, s.135.
- CASAS, E., WHITE, S.N., RILEY, D.G., SMITH, T.P.L., BRENNEMAN, R.A., OLSON, T.A., JOHNSON, D.D., COLEMAN, S.W., BENNETT, G.L., CHASE JR., C.C., 2005: Assessment of single nucleotide polymorphisms in genes residing on chromosomes 14 and 29 for association with carcass composition traits in *Bos indicus* cattle. *Journal of Animal Science* 83: 13 – 19.
- DE HUIDOBRO, R.F., MIGUEL, E., ONEGA, E., BLÁZQUEZ, B., 2003: Changes in meat quality characteristics of bovine meat during the first 6 days post mortem. *Meat Science* 65 (4): 1439 – 1446.

- HAN, C., VINSKY, M., ALDAI, N., DUGAN, M.E.R., MCALLISTER, T.A., LI, C., 2013: Association analyses of DNA polymorphism in bovine SREBP-1, LXR α , FADS1 genes with fatty acid composition in Canadian commercial crossbred beef steers. *Meat Science* 93: 429 – 436.
- KONONOFF, P.J., DEOBALD, H.M., STEWART, E.L., LAYCOCK, A.D., MARQUESS, F.L.S., 2005: The effect of a leptin single nucleotide polymorphism on quality grade, yield grade, and carcass weight of beef cattle. *Journal of Animal Science* 83: 927 – 932.
- LIM, D., KIM, N.K., PARK, H.S., LEE, S.H., CHO, Y.M., OH, S.J., KIM, T.H., KIM, H., 2011: Identification of Candidate Genes related to Bovine Marbling using Protein-Protein Interaction Networks. *International Journal of Biological Sciences* 7 (7): 992 – 1002.
- MILANESI, E., NICOLOSO, L., CREPALDI, P., 2008: Stearoyl CoA desaturase (SCD) gene polymorphisms in Italian cattle breeds. *Journal of Animal Breeding and Genetics* 125: 63 – 67.
- NUERNBERG, K., DANNENBERGER, D., NUERNBERG, G., ENDER, K., VOIGT, J., SCOLLAN, N.D., WOOD, J.D., NUTE, G.R., RICHARDSON, R.I., 2005: Effect of a grass-based and a concentrate feeding system on meat quality characteristics and fatty acid composition of longissimus muscle in different cattle breeds. *Livestock Production Science* 94: 137 – 147.
- OHSAKI, H., TANAKA, A., HOASHI, S., SASAZAKI, S., OYAMA, K., TANIGUCHI, M., MUKAI, F., MANNEN, H., 2009: Effect of SCD and SREBP genotypes on fatty acid composition in adipose tissue of Japanese Black cattle herd. *Animal Science Journal* 80: 225 – 232.
- SCOLLAN, N., HOCQUETTE, J.F., NUERNBERG, K., DANNENBERGER, D., RICHARDSON, I., MOLONEY, A., 2006: Innovations in beef production systems that enhance the nutritional and health value of beef lipids and their relationship with meat quality. *Meat Science* 74: 17 – 33.
- SHIN, S.C., CHUNG, E.R., 2007: Association of SNP marker in the thyroglobulin gene with carcass and meat quality in Korean cattle. *Asian – Australian Journal of Animal Sciences* 20 (2): 172 – 177.
- TANIGUCHI, M., MANNEN, H., OYAMA, K., SHIMAKURA, Y., OKA, A., WATANABE, H., KOJIMA, T., KOMATSU, M., HARPER, G.S., TSUJI, S., 2004: Differences in stearoyl-CoA desaturase mRNA levels between Japanese Black and Holstein cattle. *Livestock Production Science* 87: 215 – 220.
- TSUJI, S., MANNEN, H., TANIGUCHI, M., 2004: Method of evaluating beef taste and texture based on the stearoyl-CoA desaturase haplotype. European Patent Specification (EP 1 598 430 B1).
- WARNER, R.D., GREENWOOD, P.L., PETHICK, D.W., FERGUSON, D.M., 2010: Genetic and environmental effects on meat quality. *Meat Science* 86 (1): 171 – 183.