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# Association of a missense mutation of the *MC4R* gene with growth traits in cattle (Brief report)

(Assoziation einer Mutation im MC41 Gen mit Wachstumsmerkmalen beim Rind)

**Background**: Melanocortin-4 receptor (*MC4R*) is one of five G-protein-coupled receptors binding melanocortins that is implicated in the control of ingestive behavior and energy homeostasis. Mutations have been described in the human and mouse *MC4R* genes which are associated with obesity (YEO et al., 1998; HUSZAR et al., 1997). Moreover, a mutation in porcine *MC4R* is associated with economically important traits in the pig (KIM et al., 2000). The SNPs reported in bovine *MC4R* coding region were specific to breeds (VALLE et al., 2004; THUE et al., 2001; HAEGEMAN et al., 2001). In the present experiment over 95% of the coding region of *MC4R* were screened to detect the SNPs in the predominant cattle breeds of China. Association of a missense mutation of *MC4R* gene with growth traits was analyzed.

**Procedures**:

*Primer sequences:*

The primer pairs 1-3 were designed based on bovine *MC4R* gene (GenBank accession No. AF265221).

1: 5'-ACGGAATGCCCACCAATG-3', 5'-AACAAGACGCCCGACACC-3';
2: 5'-TGTTGGTGAGCGTTTC-3', 5'-ATGGTCAGGGTAATCG-3';
3: 5'-CATCACCGTGTTCTTCA-3', 5'-GCAGGGCATAAATCAG-3'.

*PCR-SSCP conditions:*

The 15 µl polymerase chain reaction (PCR) contained 100 ng of genomic bovine DNA, 1 µM of each primer, dNTPs (0.2 mM), $MgCl_2$ (1.5 mM), and 0.50 U Taq DNA polymerase (TaKaRa, Dalian, China). The cycling protocol was 4 min at 95℃, 35 cycles of 94℃ for 45 s, annealing at 63℃, 60℃ or 55℃ corresponding to 3 different primer pairs for 45 s, 72 ℃ for 1 min, with a final extension at 72℃ for 4 min. The single stranded conformation polymorphisms (SSCP) were analyzed according to previous description (HAEGEMAN et al., 2001). The PCR fragments from different SSCP patterns were subcloned and sequenced (GenBank # DQ665825).

**Results**: Only two SNPs 927C>T and 1069C>G (published in GenBank #AF265221 correspond to position 653 and 795 in DQ665825, respectively) were detected in 698 unrelated cattle from seven breeds (Nanyang 240, Qinchuan 97, Jinnan 60, Angus 43, Chinese Holstein 61, Luxi 57, Jiaxian 140). The C-to-G (1069) mutation resulted in a valine to leucine substitution. The previously described 647T>A, 727G>A, 747G>A SNPs were not detected in present populations (VALLE et al., 2004; THUE et al., 2001; HAEGEMAN et al., 2001) confirming the breed-specificity of the polymorphisms. Interestingly, the frequency of the `C´ allele at nt1069 of the Chinese cattle breeds (0.910-1.000) was significantly higher than that of Limousin (0.375; VALLE, 2004) ($\chi^2$=183.905, $P$<0.001). Limousin has been bred and selected for meat production for a very long time that may decrease the frequency of `C´ allele at nt1069. If that is true, the 1069C>G SNP may have significantly negative association with meat production traits. To address the previous hypothesis, the association of genotypes `CC´, `CG´ or `GG´ with average daily gain and birth weight of Nanyang and Jiaxian were analyzed (Table). A total of 380 cows of the Nanyang and Jiaxian breeds with known pedigree originated from our Institute were used. Fixed effects of breed, genotype, year, season of birth were included as independent variables in the linear model. The 380 cows were daughters of 54 sires. There were only 4 GG animals in the tested population, so the effect was not analyzed. The result indicated that the *MC4R* genotype was significantly associated with birth weight ($P<0.01$) and average daily gain of 6 month of age ($P<0.05$). The CG genotype had higher average daily gain and lower birth weight than the CC genotype. The bovine *MC4R* gene was located at BTA24q27 (VALLE et al., 2004). Two QTLs associated with dressing percentage and hot carcass weight were detected on BTA24. Taken together, *MC4R* gene might contribute to meat production traits of beef cattle, but the association analysis with more animals is needed before this gene can be considered as a genetic marker.

Table
Least square mean (means ± standard error of means) of average daily gain and birth weight for the alternative *MC4R* genotypes

| Trait | CC (n= 336) | CG (n=40) | *P*-value |
|---|---|---|---|
| Average daily gain of 6 months (g) | 728±8 | 840±20 | 0.010 |
| Birth weight (kg) | 29.87±0.20 | 28.40±0.53 | 0.027 |

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