Tropomodulin 1 (TMOD1) is associated with lean meat growth and meat quality in the pig (Brief Report)

Tropomodulin 1 (TMOD1) beeinflusst Fleischansatz und -qualität beim Schwein (Brief report)

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Background

Tropomodulin 1 (TMOD1) is a member of the tropomodulin family, which are highly conserved capping proteins of the pointed ends of the erythrocyte membrane and sarcomeric actin filaments. Tropomodulins are involved in the architecture of the sarcomere in muscle cells and the membrane skeleton in nonmuscle cells (COLUCCIO et al. 1994, GREGORIO et al. 1995). TMOD1 is predominantly expressed in vertebrate cardiac muscle and slow twitch muscle fibers, and it binds to one end of tropomyosin (TM) that plays important roles in regulating the function of actin filament (FISCHER et al. 2003, GUNNING et al. 2008). Here we identified a single nucleotide polymorphism (SNP) in the porcine TMOD1 gene and further analyzed the effects of this gene on the lean meat growth, meat quality and other related traits in pigs.

Procedures

The human TMOD1 mRNA (acc. no. NM_003275) was used to search for porcine ESTs in the EST-others database through BLAST (http://www.ncbi.nlm.nih.gov/blast). Porcine ESTs sharing more than 80% identity were aligned using SeqMan program of DNASTar (DNASTAR, Inc., Madison, WI, USA). The aligned sequence was corresponding to human exon7 and exon8 (acc. no. FJ428533). Two pairs of primers (F1: AGTTCAGCATCGTGGGGACA/R1: AATGCACCTGAAACACCACACAG and F2: AGATGCTCAAAGTGAACAAGGTG/ R2: CCAGAGAAGTGTTGTATGGAAGA) were designed for PCR amplification from pig genomic DNA with Primer Premier 5.0 (PREMIER Biosoft Inc, Palo Alto, CA, USA). SNP discovery was implemented by sequencing the pooled PCR products amplified from six DNA samples and each two were from Yorkshire, Landrace and Tongcheng pigs. One SNP c.816A>G (dbSNP acc. no. 107795099) in exon 7 causing a synonymous mutation Leu239Leu was identified with primers F2/R2, and it can be distinguished by BstNI with allele A revealing a 111bp and allele G revealing a 78bp and a 33bp fragments. A PCR-RFLP for SNP genotyping was developed as the followings, PCR mixture (10 μL) included 1×PCR buffer, 0.2 μM each primer, 150 μM each dNTP, 1.5 mM MgCl₂, 2U Taq DNA polymerase (Takara Company, Dalian, China) and 12.5ng genomic DNA. PCR reaction comprised of the initial denaturation at 95°C for 5 min, 30 cycles with 94°C for 30s, 62°C for 30s, 72°C for 30s,
followed by a final extension at 72 °C for 5 min; RFLP reaction mixture (10 μL) consisted of 1 μL 10× buffer, 2 U restricted enzyme BstNI and 5 μL PCR products. Samples were kept in 37 °C incubator overnight.

SNP genotyping was performed in two different pig populations. Population A (\(n=205\)) included Yorkshire (Y, \(n=26\)), Landrace (L, \(n=26\)), Tongcheng (T, \(n=49\)), L \(\times\) (Y \(\times\) T, \(n=54\)) and Y \(\times\) (L \(\times\) T, \(n=50\)). The association analysis was implemented using mixed procedure (SAS 9.0; SAS Institute, Cary, NC, USA) and this model treated population/population combination, sex, slaughter date and marker genotyping as fixed effects, dam as random effect and body weight as covariate (TANG et al. 2008). The genotyping was also performed in the ISU Berkshire × Yorkshire (B × Y) pig resource family comprised of 515 F₂ animals (MALEK et al. 2001). The association analyses was implemented using mixed model procedure, including sex, slaughter date and marker genotypes as fixed effects, dam (litter) as random effect and body weight as covariate.

**Results**

In population A, SNP c.816A>G was significantly associated (\(p<0.05\)) with loin pH. In B × Y family, c.816A>G was significantly associated with (\(p<0.05\)) loin eye muscle area, average back fat, back fat at the lumbar, at 10th rib and at last rib, ham pH and lab loin pH, and was suggestively associated (\(p<0.10\)) with hormel lion pH and average drip loss (Table 1). TM is one of the important regulatory proteins during the process of muscle contraction, and it is dominantly expressed in the thin filament with actin double helix, influencing interaction in actin and myosin (GREENFIELD et al. 2005, GUNNING et al. 2008). It is possible that TMOD1 functions in lean meat growth and affects water holding capacity of muscle cells by changing TMOD-TM binding ability (KOSTYUKOVA et al. 2004). TMOD1 was mapped between S0331 and Sw974 on SSC1, where numbers of QTL related to lean meat weight, LEA and pH have been located (http://www.animalgenome.org/cgi-bin/QTLdb; MALEK et al. 2001, BEECKMANN et al. 2003, GELDERMANN et al. 2003). Combining the association analyses and linkage mapping promote the porcine TMOD1 gene as a candidate gene for muscle production. Further work on additional causative mutation discovery and function analysis of TMOD1 gene in pigs are warranted.

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Table 1
Association of Leu239Leu of Tmod-1 gene with the analyzed traits in two pig populations respectively

<table>
<thead>
<tr>
<th>Population</th>
<th>Trait</th>
<th>LSM (SE)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>loin pH</td>
<td>6.36 (0.04)(^a)</td>
<td>6.48 (0.03)(^b)</td>
</tr>
<tr>
<td>B</td>
<td>LEA</td>
<td>33.22 (0.87)(^a)</td>
<td>35.29 (0.62)(^b)</td>
</tr>
<tr>
<td></td>
<td>AVBF</td>
<td>3.54 (0.09)(^a)</td>
<td>3.33 (0.05)(^b)</td>
</tr>
<tr>
<td></td>
<td>last rib BF</td>
<td>3.40 (0.09)(^a)</td>
<td>3.23 (0.05)(^b)</td>
</tr>
<tr>
<td></td>
<td>lumbar BF</td>
<td>3.83 (0.11)(^a)</td>
<td>3.63 (0.06)(^a)</td>
</tr>
<tr>
<td></td>
<td>10th rib BF</td>
<td>3.39 (0.11)(^a)</td>
<td>3.13 (0.06)(^b)</td>
</tr>
<tr>
<td></td>
<td>ham pH</td>
<td>5.95 (0.03)(^a)</td>
<td>5.86 (0.01)(^b)</td>
</tr>
<tr>
<td></td>
<td>hormel pH</td>
<td>5.77 (0.02)(^ab)</td>
<td>5.74 (0.01)(^a)</td>
</tr>
<tr>
<td></td>
<td>lab loin PH</td>
<td>5.81 (0.02)(^ab)</td>
<td>5.79 (0.01)(^a)</td>
</tr>
<tr>
<td></td>
<td>AVDRIPPR</td>
<td>5.92 (0.30)(^ab)</td>
<td>5.97 (0.15)(^a)</td>
</tr>
</tbody>
</table>

P-values are adjusted by Bonferroni. The traits are: loin pH: pH at 45 min post mortem, LEA: loin eye muscle area (cm²), AVBF: average back fat (cm), last rib BF: last rib back fat (cm), lumbar BF: lumbar back fat (cm), 10th rib BF: 10th rib back fat (cm), ham pH: pH at 24 h post mortem, hormel loin pH: pH at 24 h post mortem measured at the Hormel slaughter plant, lab loin pH: pH at 48 h post mortem measured at the Iowa State University Meat Laboratory, AVDRIPPR: average drip loss (%). LSM (SE) represents least squares means and their standard errors, Superscripts a, b, and/or c differ significantly (P<0.05) from each other.

References


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