Characterization of an X-chromosome PCR–RFLP marker associated with fat deposition and growth in the pig

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Summary

The X-chromosome, highly conserved within mammals, has been shown to contain major quantitative trait loci (QTL) for growth and fat deposition in the pig. We have discovered a BamHI polymerase chain reaction restriction fragment length polymorphism (PCR–RFLP) marker that was assigned to the porcine X-chromosome by two-point and multi-point linkage analysis following genotyping of a three-generation Berkshire by Yorkshire reference family. The marker was positioned 9 cM telomeric to SW2126 and 15.6 cM centromeric to SW1943. Sequence flanking the marker was found to have high similarity to existing database porcine DNA repeat elements. Association analyses of the BamHI marker for growth and meat quality traits in the reference family revealed significant association with marbling (P < 0.03), 10th rib back fat (P < 0.09) and total lipid percentage (P < 0.05), as well as with loin eye area (P < 0.04), average glycolytic potential (P < 0.03) and average lactate content (P < 0.04). Further studies are required to determine the X-chromosome functional gene affecting fat deposition and growth in the pig.

Keywords association analysis, deoxyribonucleic acid repeat element, fat deposition, growth, polymerase chain reaction restriction fragment length polymorphism, pig, X-chromosome.
Restriction enzyme digestion produced allelic fragments of 186 bp and 19 bp (allele 1) and 90 bp, 49 bp, 47 bp, and 19 bp (allele 2), respectively. The allele-specific PCR–RFLP products were separated by electrophoresis on a 3.5% agarose gel.

The X-linked inheritance of the marker was discovered following genotyping of a three-generation Berkshire by Yorkshire (BY) reference family (Malek et al. 2001). Within the pedigree, males always appeared homozygous for the marker, while the females displayed all three genotypes. Also, male offspring appeared to be homozygous for an allele inherited from the dam. Based on these facts, the X-linked inheritance of the marker was suggested and later confirmed by linkage mapping results. Genotype and allele frequencies were obtained from 284 unrelated female animals from five different pure breeds, one crossbred and three synthetic lines (Table 1).

The BamHI marker was assigned to SSCX by two-point and multi-point linkage analysis using genotypes from the reference family mentioned previously and CRI-MAP software (Green et al. 1990). The BamHI marker was positioned 9 cM telomeric to SW2126 and 15.6 cM centromeric to SW1943.

Sequence comparisons of the marker were performed using BLAST analysis of the GenBank database. As a result of the 3’-primer design, the fragment’s sequence had high identity with several swine intronic DNA repeat elements (e.g. GenBank accession nos AJ251914, AJ251829, AJ410870). Also, the isolated fragment showed sequence identity ranging between 75 and 83% to intronic, promoter, and 5’- and 3’-untranslated regions of several X-chromosome porcine genes: proteolipid protein (PLP), coagulation factor IX, androgen receptor (AR) and NADPH oxidase 1 (NOX1).

Association analyses of the BamHI marker with growth and meat quality traits in the BY reference family were performed using PROC MIXED in SAS (SAS Institute, Cary, NC, USA). For both growth and meat quality traits, sex, slaughter date and genotype were included as fixed effects with dam included as a random effect. For growth traits, live weight was also included as a covariate. The BamHI marker was found to be significantly associated with marbling ($P < 0.03$), 10th rib back fat ($P < 0.09$) and total lipid percentage ($P < 0.05$) in the BY reference family. It was also significantly associated with loin eye area ($P < 0.04$), average glycolytic potential ($P < 0.03$) and average lactate content ($P < 0.04$). The number of positive association results is greater than expected by chance alone for the 40 traits tested. Interestingly, BY females homozygous for allele 2 had significantly larger loin eye area and significantly less marbling than other female genotypes, while differences were not significant between males.

Additional studies should confirm these results and attempt to identify X-chromosome functional genes underlying growth and fat deposition in the pig.

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References


