DETECTION OF IMPRINTED QUANTITATIVE TRAIT LOCI AFFECTING GROWTH AND MEAT QUALITY IN KOREAN NATIVE PIGS

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INTRODUCTION

Due to availability of large numbers of polymorphic markers, it is possible to scan a whole genome for genes affecting quantitative traits of interest, so-called quantitative trait loci (QTL). QTLs underlying the genetic variance of economic traits of livestock have been mapped in several chromosomal regions. On the basis of genetic linkage maps and data from F₂ breed cross resource populations, several recent studies have reported a number of discoveries for QTL in the pig on a variety of chromosomes (Anderson et al., 1994). Accordingly, whole genome scans have derived a number of genomic regions containing QTLs and also provided a better insight into the mode of inheritance for those traits. Recently, several studies have shown that non-Mendelian form of gene expression can be searched through a genome scan. There are several reports of QTLs exhibiting non-Mendelian inheritance in pigs. An imprinted QTL affecting muscle mass and fat deposition was identified near the IGF-2 locus on chromosome 2 by Nezer et al. (1999) and Jeon et al. (1999). De Koning et al. (2000) have presented the results of a genome-wide scan aiming at detecting imprinted regions for multifactorial traits. They detected gametic imprinting for QTL in swine F₂ cross by comparing likelihood of model with paternal and maternal imprinting effects against a model with no linked QTL. If imprinting is a more common phenomenon than usually thought, it is needed to investigate whether imprinting occurs or not in previously published QTL for application of marker assisted selection. To develop and apply tests that identify deviations from a purely Mendelian model, it is proposed that imprinting acts to maximize the genetic contribution of one parent at the expense of the other in polygamous species. This study presents the results of a partial genome-wide scan to detect imprinted regions for multifactorial traits in an F₂ cross between outbred lines of pigs.

MATERIAL AND METHODS

Experimental population and performance traits. Five Korean native pig sires and eleven Landrace dams were mated to produce nine F₁ litters. From the F₁ litters, 11 boars and 36 gilts were chosen to produce F₂ animals. A resource population with 141 F₂ progeny was established.

F₂ pigs were genotyped for 24 microsatellite markers on chromosome 2 (8 markers), chromosome 6 (8 markers) and chromosome 7 (8 markers).

Body weight was measured at birth, 3 weeks, 5 weeks and 12 weeks of age. The number of nipples was recorded. Pigs were weighed and slaughtered at 210 days of age. Average daily
gain was calculated from birth to 3 weeks of age, from 3 to 5 weeks of age, from 5 to 12 weeks of age, and from 12 to 30 weeks of age. After slaughtering, hot carcass weight and backfat thickness were recorded. Meat pH was measured at twenty-four hours after slaughtering. Trimmed wholesale cut was recorded. Six additional meat quality traits i.e., meat color, muscle sheaf force, drip loss, cooking loss, intramuscular fat content and water holding capacity were also recorded. Meat color was measured with Hunter reflectometer using the L, a and b coordinate system, where L is a general indication of lightness, a represents the degree of green-redness and b represents the degree of blue-yellowness.

**Statistical analysis.** In order to provide evidence of imprinting and identify QTL that are paternally or maternally expressed in the genome, the imprinting model was reparameterized so as to enable a direct test for the contribution of the paternally and maternally inherited effects (de Koning et al., 2000). Under the traditional line cross approach, offspring performances can be modelled in terms of the additive \( (a) \) and dominance \( (d) \) components that can be estimated using the regression of the phenotypes on the \( P_a \) and \( P_d \) (Haley et al., 1994):

\[
Y_j = m + a P_{aj} + d P_{dj} + e_j 
\]

(Mendelian model)

Accounting for the grandparental origin of the alleles by using the multiple marker information, it is possible to calculate probabilities of the two alleles in an offspring according to the four possible genotypes in the \( F_2 \) generation conditional on marker information. On the basis of this concept, we developed full imprinting model to test imprinting effects \( (P_{imp} = P_{pat} - P_{mat}) \):

\[
Y_j = m + a P_{aj} + d P_{dj} + i P_{imp}(j) + e_j 
\]

(Full Imprinting model)

Maximum likelihood analysis was used for ordering marker loci with the CRIMAP program (Green et. al., 1990). For QTL analysis, an interval mapping procedure using least squares regression method was applied under line cross concept. Founder breeds or lines are assumed to be fixed for alternative alleles at the QTL affecting the traits of interest. The statistical model included sex and slaughtering date as fixed effects and live weight as a covariable. A single QTL was fitted in all cases by regressing on additive and dominance coefficients for the QTL at each putative position of the QTL (every \( cM \)). Additive and dominance coefficients at a given position of the QTL were derived from the marker data with the procedure of Haley et al. (1994). To test the presence of a QTL, different tests were performed Mendelian vs/no QTL model (Men / Null), full imprinting vs/no QTL model (Full / Null).

The method proposed by Churchill and Doerge (1994) was used to empirically determine significance thresholds for the \( F \) statistic to control Type I error rate at the chromosome-wide level using data permutation.

**RESULTS AND DISCUSSION**

Thresholds (5% chromosome-wide) against the no QTL model for the traits analyzed ranged from 4.39 to 4.89 for the Mendelian model and from 4.09 to 4.77 for the imprinting model. A partial genome scan was performed for chromosomes 2, 6 and 7 in this study. Under the Mendelian model, the presence of 23 QTL was detected on the basis of a critical value estimated by permutation test at the single test significance level (5%). Twenty QTL were
shown as putative QTL under the full imprinting model. Of the 23 QTLs that exceeded single test threshold, 12 QTLs were significant at the 5% chromosome-wide level and two QTLs were significant at the 1% chromosome-wide level under the Mendelian model. With the model including imprinting effect, twelve QTLs were evidenced at the 5% chromosome-wide level and one at 1% chromosome-wide level were found.

Table 1 presents the QTLs detected at the 5% chromosome-wide level under the most likely genetic model. Genetic effects (additive and dominance) estimated in this study correspond to genotype values of +\(a\), \(-d\) and \(-\alpha\), respectively, for individuals having inherited two Korean native alleles, heterozygotes and individuals with two Yorkshire alleles. Positive additive effects indicate that Korean native alleles increase the phenotypic value of traits, negative values indicate additive that the Korean native pigs alleles decrease it. Dominance effects are relative to the mean of the two homozygotes.

### Table 1. Evidence for QTL significant at the 5% chromosome-wide level for various economic traits, estimated significance levels (\(F\)-Value), location and genetic effect

<table>
<thead>
<tr>
<th>SS C</th>
<th>Trait</th>
<th>Genetic model</th>
<th>Test statistics (Position, cM)</th>
<th>Additive (s.e.)</th>
<th>Dominance (s.e.)</th>
<th>Imprinting (s.e.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>Bwt</td>
<td>Mendelian</td>
<td>4.88 (25)</td>
<td>-0.08 (0.03)</td>
<td>-0.12 (0.06)</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Postwadg</td>
<td>Mendelian</td>
<td>6.03 (155)</td>
<td>0.02 (0.02)</td>
<td>0.10 (0.02)</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Carcwt</td>
<td>Mendelian</td>
<td>6.55 (78)</td>
<td>-2.34 (1.87)</td>
<td>9.34 (2.77)</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>24-PH</td>
<td>Imprinting</td>
<td>5.04 (72)</td>
<td>-0.03 (0.03)</td>
<td>-0.14 (0.04)</td>
<td>0.05 (0.03)</td>
</tr>
<tr>
<td></td>
<td>FAT</td>
<td>Mendelian</td>
<td>4.78 (98)</td>
<td>-1.50 (0.72)</td>
<td>2.81 (1.11)</td>
<td>-</td>
</tr>
<tr>
<td>6</td>
<td>Heat loss</td>
<td>Mendelian</td>
<td>5.43 (109)</td>
<td>-1.54 (0.56)</td>
<td>1.84 (0.83)</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Start wt</td>
<td>Imprinting</td>
<td>5.87 (80)</td>
<td>-1.30 (0.85)</td>
<td>4.53 (1.33)</td>
<td>1.92 (0.86)</td>
</tr>
<tr>
<td></td>
<td>Total-teat</td>
<td>Imprinting</td>
<td>4.47 (142)</td>
<td>-0.36 (0.13)</td>
<td>0.09 (0.06)</td>
<td>0.32 (0.14)</td>
</tr>
<tr>
<td></td>
<td>Wt 5wk</td>
<td>Mendelian</td>
<td>5.58 (77)</td>
<td>-0.0490 (0.24)</td>
<td>1.23 (0.37)</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Postwadg</td>
<td>Imprinting</td>
<td>4.16 (0)</td>
<td>-0.00 (0.01)</td>
<td>-0.06 (0.02)</td>
<td>-0.04 (0.02)</td>
</tr>
<tr>
<td>7</td>
<td>24-PH</td>
<td>Mendelian</td>
<td>6.45 (9)</td>
<td>-0.14 (0.05)</td>
<td>-0.15 (0.09)</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>FAT</td>
<td>Mendelian</td>
<td>9.81 (25)</td>
<td>1.86 (0.52)</td>
<td>-3.28 (1.01)</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Hun-a</td>
<td>Mendelian</td>
<td>8.14 (107)</td>
<td>0.81 (0.32)</td>
<td>1.50 (0.51)</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Hun-b</td>
<td>Mendelian</td>
<td>6.14 (103)</td>
<td>0.27 (0.22)</td>
<td>0.998 (0.33)</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Shear</td>
<td>Mendelian</td>
<td>5.82 (59)</td>
<td>0.55 (0.15)</td>
<td>-0.06 (0.24)</td>
<td>-</td>
</tr>
</tbody>
</table>

* Significant at the 5% chromosome-wide level.
** Significant at the 1% chromosome-wide level.

In this study the line-cross analyses for twelve traits revealed a total of 15 QTLs with evidence of suggestive linkage (chromosome-wide significance) with the 2 models used. For 24h_PPH, FAT and PSTWADG, two QTLs were detected on chromosome 2, 6 and 7, respectively. Of the detected QTLs there were 4 putative QTLs that showed significant imprinting effects for 24h_PPH, STARTWT, Total-teat and PSTWADG on chromosome 6.
CONCLUSION
In this study, a partial genome scan of data from a cross between the Korean native and Landrace breeds was performed using a least-squares regression approach for outbred crosses, including a systematic test for imprinting and thereby some evidence for QTLs affecting growth and meat quality traits obtained.

Until now genomic imprinting was regarded as a rare phenomenon and, consequently, was ignored in most studies. As several results showing significant imprinted QTLs, have recently been reported, it appears that genomic imprinting might be a more common phenomenon than previously expected. We detected fifteen QTLs of which four were subject to imprinting at a suggestive significance level. A similar imprinting effect was reported for this same chromosomal region by de Koning et al. (2000). Outbred crosses between two pig breeds with different genetic backgrounds, are the ideal resource population for detecting imprinted regions. Yet, it remains necessary to test larger numbers of data or joint data sets produced from several resource populations and to construct fine maps in target regions such as these we found in this study to better understand the underlying phenomena. For the practice of animal breeding, identification of major imprinted loci affecting economic traits has several implications. If much more strongly imprinted genes affecting economic traits are found and confirmed as a real phenomenon in livestock performance, a revision of methods for genetic evaluation that currently ignore non-Mendelian gene expression is necessary. The identification of imprinted loci also opens new perspectives for crossbreeding, which is common a practice in pig breeding.

REFERENCES
Green, P., Fall, I. and Crook, S. (1990) "Documentation for CRI-MAP version 2.4". Washington University School of Medecine, St-Louis.