INTRODUCTION

Insulin-like growth factors (IGFs) belong to a family of peptide hormones with a broad range of metabolic and mitogenic actions. The structure of the two peptides, insulin-like growth factor (IGF)-I and IGF-II, share an extensive homology with insulin, suggesting that a functional analogy exists between them. However, in contrast to insulin, IGFs are synthesized in many tissues, both muscular and follicular (Lahbib et al., 1995). The biological effects of IGFs arise through interaction with specific cell-membrane receptors that have a high affinity for these two peptides.

The IGF-I receptor plays a key role in the function of the IGF axis (Jones and Clemmons, 1995). It has maximum affinity for IGF-I, but also binds the closely related, and largely homologous IGF-II, thus mediating the actions of both IGFs. The growth factors and receptors, together with at least six specific binding proteins, play a central role in the regulation of fetal and postnatal growth and development (Jones and Clemmons, 1995) and malignant transformation (Baserga, 1996). The IGFIR is a heterotetrameric transmembrane glycoprotein composed of two extracellular α subunits (135 kDa), which bind IGF, and two transmembrane β subunits (90 kDa), which possess tyrosine kinase activity (Giudice, 1992; Thomas, et al., 1998). By inactivating the IGFIR gene in mice, Holzenberger et al. (2003) found that IGFIR regulates lifespan and resistance to oxidative stress. In humans, the IGFIR gene was localized in chromosome 15q25-q26. Porcine IGFIR was assigned to 1q17-21 by radioactive in situ hybridization (Lahbib et al., 1995; Kopečný et al., 2002) and mapped by linkage analysis (Hu et al., 1997). Harumi et al. (2001) cloned the porcine IGFIR gene and confirmed that the sequence of the putative porcine IGFIR has an open reading frame (ORF) of 1367 amino acids. The translated amino acid sequence from the porcine IGFIR cDNA has 98.1% and 95.2% similarity to that of human and rat IGFIR, respectively.

Growth rate and body composition are two important characteristics in pig production. Efficient pig production...
systems demand lean individuals with high growth rate and a low conversion of feed to meat. These important economical traits are often controlled by many genes and modified by environmental factors. As mentioned above, the growth factors and receptors together with at least six specific binding proteins play a central role in the regulation of fatal and postnatal growth and development (Jones and Clemmons, 1995), but little information currently exists regarding the relationship between the \textit{IGFIR} genotype and performance traits in swine. Consequently, in this study, we investigate one \textit{IGFIR} restriction fragment length polymorphism (RFLP) (as described by Kopečný et al. (2002)) in 12 pig breeds and its relationship with performance traits in two pig breeds.

**MATERIALS AND METHODS**

**Animals**

The ear notches of 12 pig breeds (total 593 samples) were collected, including 4 European breeds and 8 Chinese native breeds. They were Landrace (n = 65, Dongxiang County Pig Farm and Jiangxi Seed Pig Farm), Duroc (n = 52, Dongxiang County Pig Farm and JAU Seed Pig Farm), Piétrain (n = 30, Daxing County Pig Farm), Large White (n = 46, Jiangxi Seed Pig Farm), Erhualian (n = 48, Chanshu Pig Farm of Jiangsu Province), Jinhua Pig (n = 50, Jinhua County Pig Farm), Ningxiang Pig (n = 56, Ningxiang County Pig Farm), Jiaxing Pig (n = 50, Jianxing County Pig Farm), Shangdi Pig (n = 50, Hefeng County Pig Farm), Daweizi Pig (n = 53, Changsha County Daweizi Pig Farm), Shangyuhua Pig (n = 48, Shengxian Pig Farm), Taoyuan Pig (n = 45, Taoyuan County pig Farm). All of the collected pigs were unrelated within 3 generations. In order to detect the relationship of the IGF-1 receptor genotype to growth performance, we also investigated two other pig breeds, Wanbai pigs (n = 156, Anhui Province Seed Pig Farm) and Large Yorkshire (n = 202, Jiangxi Provincial Seed Pig Farm). Growth related variables including body weight at birth, 2-, 4- and 6-mo ages, the pigs were weighted individually, and backfat thickness and lean percentage estimated by ultrasonography at 6-mo age were recorded for analyzing the association between \textit{IGFIR} gene polymorphism and growth traits.

**IGF1R genotypes defined by PCR-RFLP**

Genomic DNA was extracted from ear notches, using a phenol/chloroform extraction method followed by ethanol precipitation. Working dilutions of extracted DNA were prepared for each individual at a concentration of 50 ng/µg. Primers 5′-AGC TAT CTC TAC CGG CAT AA -3′ and 5′-TCT CGA AGA CCT TGC GGT ACT-3′ were used for polymerase chain reaction (PCR) amplification of the intron 9 of the \textit{IGFIR} gene (GenBank accession number: AJ491314) according to Kopečný et al. (2002). The PCR mixture contained 50 ng genomic DNA, 25 pmol of each primer, 25 nmol of each dNTP, 1 unit of Taq DNA Polymerase (TAKARA) and 1× reaction buffer in a 25 µL reaction volume. PCR was performed on a PE9600 (PERKIN ELMER) according to the following procedure: 95°C for 300s then 35 cycles of: 94°C for 30 s, 53°C for 30 s, 72°C for 45 s, and finally 72°C for 480 s. The 379 bp PCR products (parts of exons 9 and 10 of the porcine \textit{IGFIR} gene) were subsequently digested by \textit{Sac} II and revealed allele A (fragment of 379 bp) or allele B (fragments of 235 bp and 144 bp). The restriction digests were separated using 2.5% agarose gel in 1×TAE at a constant current of 50 mA. The gels were stained with ethidium bromide and the fragments were visualized using a UV transilluminator (Figure 1).

**Statistics**

Polymorphisms Information Content (PIC) : Expected PIC value of each locus was calculated by using the method of Bostein et al. (1980).

\[
PIC = 1 - \sum_{i=1}^{n} p_i^2 - \sum_{i=1}^{n} \sum_{j=i+1}^{n} 2p_i^2p_j^2
\]

with, \(p_i\) = frequency of the \(i\)th allele
\(p_j\) = frequency of the \(j\)th \((= 1+1)\) allele
\(n\) = number of alleles

Association test between genotype and production traits : The \(\chi^2\) test was used for estimation of Hardy-Weinberg equilibrium in the population. The least square means (LSM) was used for statistical analysis for data

**Figure 1.** Agarose gel electrophoresis (2.5%) indicating genotypes of the porcine IGFIR after digestion with \textit{Sac}II. The genotypes (AA, AB, BB) are indicated at the top of each lane. M, 100 bp DNA Ladder.
collected. Associations between IGFIR genotypes and growth traits were analyzed using the SAS (1989) and difference between genotypes was tested using the General Linear Model of SAS. The model included sex, environment, genotypes of IGFIR gene and fixed errors effects.

**RESULTS**

After the PCR products digested by Sac II and separated with Agarose gel electrophoresis (2.5%), three genotypes were found, and we defined 379 bp as AA genotype, 235+144 bp as BB genotype, and 379+235+144 bp as AB genotype (Figure 1).

The genotype frequencies and allele frequencies, PIC value and $\chi^2$-test of IGFIR in 12 pig breeds are listed in Table 1.

From Table 1, it can be seen that the AA genotype frequency in European pig breeds was higher than those of Chinese native breeds; all were over 50%, with the Duroc possessing the highest (96.15%). For the allele frequency, the A frequency in European pig breeds was higher, especially in Duroc, with a frequency of 98.08. On the other hand, in Chinese native pig breeds, the B frequency was higher (above 50%), with the higher frequency occurring in Jinhua pigs (97.00%). In all the investigated 12 pig breeds, only Large White has a middle PIC value (0.256), and all pig breeds were at Hardy-Weinberg disequilibrium.

Table 2 lists the observed genotypes and their frequencies in Wanbai and Large Yorkshire and the relationship between different genotypes and production performances. The genotype AA in both pig breeds was high (76.92% in Wanbai pigs and 79.72% in Large Yorkshire, respectively).

In Wanbai pigs, birth weight, 2-mo and 6-mo body weights (BW) showed a significant association with the IGFIR genotypes. The pigs with AA genotype had higher birth weight, 2 months BW and 6 months BW than those pigs with BB genotype (p<0.05), but no significant difference was observed among the three genotypes for 4 months BW.

In Large Yorkshire pigs, pigs with AA genotype had higher birth weight, 2 months BW and 6 months BW than those pigs with BB genotype (p<0.05), but no significant difference was observed among the three genotypes for 4 months BW (p>0.05). For hinder-back-fat thickness and

<table>
<thead>
<tr>
<th>Breed</th>
<th>n</th>
<th>AA</th>
<th>AB</th>
<th>BB</th>
<th>PIC value</th>
<th>$\chi^2$-test</th>
</tr>
</thead>
<tbody>
<tr>
<td>Duroc</td>
<td>52</td>
<td>96.15 (50)</td>
<td>3.85 (2)</td>
<td>0.00 (0)</td>
<td>98.08</td>
<td>0.037</td>
</tr>
<tr>
<td>Large White</td>
<td>46</td>
<td>73.91 (34)</td>
<td>15.22 (7)</td>
<td>10.87 (5)</td>
<td>81.52</td>
<td>0.256</td>
</tr>
<tr>
<td>Landrace</td>
<td>65</td>
<td>86.15 (56)</td>
<td>9.23 (6)</td>
<td>4.62 (3)</td>
<td>90.77</td>
<td>0.153</td>
</tr>
<tr>
<td>Piétrain</td>
<td>30</td>
<td>80.00 (24)</td>
<td>13.33 (4)</td>
<td>6.67 (2)</td>
<td>86.67</td>
<td>0.228</td>
</tr>
<tr>
<td>Ningxiang pig</td>
<td>56</td>
<td>7.14 (4)</td>
<td>10.71 (6)</td>
<td>82.15 (46)</td>
<td>12.50</td>
<td>0.194</td>
</tr>
<tr>
<td>Shangdi pig</td>
<td>50</td>
<td>2.00 (1)</td>
<td>8.00 (4)</td>
<td>90.40 (4)</td>
<td>6.00</td>
<td>0.110</td>
</tr>
<tr>
<td>Jinhuia pig</td>
<td>50</td>
<td>0.00 (0)</td>
<td>6.00 (3)</td>
<td>9.00 (47)</td>
<td>3.00</td>
<td>0.057</td>
</tr>
<tr>
<td>Jiaxing pig</td>
<td>50</td>
<td>4.00 (2)</td>
<td>10.00 (5)</td>
<td>86.00 (43)</td>
<td>7.00</td>
<td>0.149</td>
</tr>
<tr>
<td>Daweizi pig</td>
<td>53</td>
<td>5.66 (3)</td>
<td>15.10 (8)</td>
<td>79.25 (42)</td>
<td>13.21</td>
<td>0.204</td>
</tr>
<tr>
<td>Taoyuan pig</td>
<td>45</td>
<td>2.22 (1)</td>
<td>13.33 (6)</td>
<td>84.45 (38)</td>
<td>8.89</td>
<td>0.149</td>
</tr>
<tr>
<td>Erhualian pig</td>
<td>48</td>
<td>4.17 (2)</td>
<td>15.06 (7)</td>
<td>81.25 (39)</td>
<td>11.46</td>
<td>0.183</td>
</tr>
<tr>
<td>Shangyuha pig</td>
<td>48</td>
<td>6.25 (3)</td>
<td>12.50 (6)</td>
<td>81.25 (39)</td>
<td>12.50</td>
<td>0.194</td>
</tr>
</tbody>
</table>

Table 1. The genotype frequencies and allele frequencies, PIC value and chi-square test of pig IGFIR gene in 12 pig breeds

<table>
<thead>
<tr>
<th>Genotype</th>
<th>AA</th>
<th>AB</th>
<th>BB</th>
<th>PIC value</th>
<th>$\chi^2$-test</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number</td>
<td>120</td>
<td>23</td>
<td>13</td>
<td>169</td>
<td>33</td>
</tr>
<tr>
<td>PIC value</td>
<td>0.230</td>
<td></td>
<td></td>
<td>0.194</td>
<td></td>
</tr>
<tr>
<td>$\chi^2$-test</td>
<td>65.78</td>
<td></td>
<td></td>
<td>28.71</td>
<td></td>
</tr>
</tbody>
</table>

Birth weight 1.35±0.23 a 1.20±0.09 ab 1.09±0.12 1.46±0.18 a 1.31±0.21 ab 1.22±0.10 b
2 mon. BW (kg) 21.74±2.29 a 20.35±1.78 ab 19.14±2.75 b 25.43±2.10 a 23.99±2.38 ab 22.47±2.65 b
4 mon. BW (kg) 58.46±5.43 57.87±5.22 56.87±4.32 62.45±7.30 61.39±6.54 59.74±5.68
6 mon. BW (kg) 85.79±7.98 a 82.47±6.69 ab 80.14±9.20 b 103.45±11.84 a 98.43±9.43 ab 94.22±9.19 b
Hind-back-fat thickness (cm) 1.92±0.12 b 1.98±0.19 b 2.23±0.31 a
Mid- back-fat thickness (cm) 1.70±0.29 b 1.83±0.23 ab 2.20±0.18 a
Lean percentage (%) 61.63±2.23 a 59.43±2.43 Ab 55.67±3.05 B

Means with different character on superscript within the same row in the same breed are significantly different (p<0.05 or p<0.01).

Values are LSM±SE.

Table 2. Effects of different genotypes on some performance traits in two pig breeds

<table>
<thead>
<tr>
<th>Breeds</th>
<th>Wanbai pig</th>
<th>Large Yorkshire</th>
</tr>
</thead>
<tbody>
<tr>
<td>Genotype</td>
<td>AA</td>
<td>AB</td>
</tr>
<tr>
<td>Number</td>
<td>120</td>
<td>23</td>
</tr>
<tr>
<td>PIC value</td>
<td>0.230</td>
<td></td>
</tr>
<tr>
<td>$\chi^2$-test</td>
<td>65.78</td>
<td></td>
</tr>
</tbody>
</table>
mid-back-fat thickness, pigs with AA genotype were found to be significantly thinner than those with the BB genotype (p<0.05). For lean percentage, pigs with AA genotype had a higher percentage than those with the BB genotype (p<0.01).

**DISCUSSION**

A significant QTL effect in swine was found for post-weaning average daily gain (ADG) between 5.5 and 56 kg of body weight that mapped between markers SW373 and SW1301 near the telomere of Chromosome 1 q arm, but the QTL was 60 cM away from the IGFIR (Paszek et al., 1999). QTLs for the deposition of backfat as well as loin eye area and carcass length were also located within the same marker interval on SSC1 (Rohrer and Keele 1998a, 1998b). Also as Taylor and Phillips (1996) showed that a QTL concerning body weight in mouse is on the mouse chromosome 7, and one QTL affecting adiposity index, which accounted for 12.3% of phenotypic variance in gender-merged data and IGFIR is as a candidate gene. Furthermore, the Mouse Genome Database at The Jackson Laboratory (http://www.informatics.jax.org/homology.html) reports one of the three candidate genes from regions of mouse and human genomes homologous to SSC1 is IGFIR, IGFIR therefore seemed a logical choice to try and identify major genetic factors affecting mammalian growth (Paszek et al., 1999). But in porcine, does the QTL affect this gene IGFIR, or IGFIR might be a possible positional candidate for the Paszek's QTL, is the problem that we should do more work to identify.

The present results are consistent with those of Kopečný et al. (2002), in which they found western pigs carried BB genotype with a low frequency, while Chinese native pig breeds had higher frequencies. In our study, the allelic frequency of Landrace was similar to Kopečný et al. (2002), whose result (n = 11) was 0.91 (A) and 0.19 (B), but had a higher frequency than the results of Kopečný et al. (2002) in Large Yorkshire (n = 14, allele A frequency is 0.71) and Pietrain (n = 24, allele A frequency is 0.83), while Chinese native pig Meishan (n = 12) carried 0.21 frequency of allele A.

The results of the comparison of genotype and performance traits for one European and one Chinese hybrid pig breed indicated that different genotype affected the growth traits and lean percentage variedly. From Table 2, we observe that all the performance traits in Wanbai pigs were lower than those in Large Yorkshire. This is because Wanbai is a hybrid (Huaizhu×Large Yorkshire), which has only been bred for 3 generations. The pigs contain 12.5% blood of Huaizhu, and Huaizhu is a Chinese native pig breed, and so this may affect the function of allele A.

As interaction with IGF1 gene, and more works were done on the effects of IGF1 genotype on performance traits in swine (Cass-Carrilo et al., 1997; Wang et al., 2002). Our results suggest that the selection of A-allele pigs may help increase the growth rate and carcass lean percentage, which mean that the IGFIR gene plays an important role in the growth and fat deposition process via its interactions with the IGF1 gene. However, samples size in this study was small, and the study should be repeated, either with a larger sample size or in a large commercial population to verify our results before our findings are incorporated into a production facility on a large scale.

**ACKNOWLEDGEMENTS**

This research was supported by Educational Department of Jiangxi Province (Gancaijiao 2007-73). The authors deeply appreciated the critical suggestions from Dr. Simon Rayner in the State Key Laboratories for Agrobiotechnology, China Agricultural University; also the authors thank the two anonymous referees for their instructive suggestions.

**REFERENCES**


