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# Investigation of Single Nucleotide Polymorphisms in Porcine Chromosome 2 Quantitative Trait Loci for Meat Quality Traits

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**ABSTRACT :** Several studies have reported quantitative trait loci (QTL) for meat quality on porcine chromosome 2 (http://www.animalgenome.org/QTLdb/pig.html). For application of the molecular genetic information to the pig industry through marker-assisted selection, single nucleotide polymorphism (SNP) markers were analyzed by comparative re-sequencing of polymerase chain reaction (PCR) products of 13 candidate genes with DNA from commercial pig breeds such as Berkshire, Yorkshire, Landrace, Duroc and Korean Native pig. A total of 34 SNPs were identified in 15 PCR products producing an average of one SNP in every 253 bp. PCR restriction fragment length polymorphism (RFLP) assays were developed for 11 SNPs and used to investigate allele frequencies in five commercial pig breeds in Korea. Eight of the SNPs appear to be fixed in at least one of the five pig breeds, which indicates that different selection among pig breeds might be applied to these SNPs. Polymorphisms detected in the PTH, CSF2 and FOLR genes were chosen to genotype a Berkshire-Yorkshire pig breed reference family for linkage and association analyses. Using linkage analysis, PTH and CSF2 loci were mapped to pig chromosome 2, while FOLR was mapped to pig chromosome 9. Association analyses between SNPs in the PTH, CSF2 and FOLR suggested that the CSF2 MboII polymorphism was significantly associated with several pork quality traits in the Berkshire and Yorkshire crossed F2 pigs. Our current findings provide useful SNP marker information to fine map QTL regions on pig chromosome 2 and to clarify the relevance of SNP and quantitative traits in commercial pig populations. (**Key Words :** Pig, Quantitative Trait Loci, Pig Chromosome 2, Single Nucleotide Polymorphism, Meat Quality)

## INTRODUCTION

In recent years the swine industry has improved production efficiency related traits, such as growth rate, reduced back fat thickness, and feed efficiency, through selective breeding of superior pigs based on available phenotypic information. Recently, the consumer's demand for improved pork quality has increased, but the improvement of pork quality traits has been difficult with breeding schemes using phenotypic information because genetic improvement of meat quality traits requires extensive and expensive measurements of traits at slaughter on relatives of animals considered for selection.

Early investigations to elucidate genetic variation of pork quality have discovered two major genes primarily involved in pale, soft, and exudative (PSE) meat condition (*HAL*) and cured-cooked ham yield (*RN*), respectively (Le Roy et al., 1990; Fujii et al., 1991). More recent developments of quantitative trait loci (QTL) studies have detected major chromosomal regions affecting various meat and eating quality traits in commercial pigs (Malek et al., 2001; Kim et al., 2005; Rohrer et al., 2005; Jin et al., 2006; van Wijk et al., 2006; Wimmers et al., 2006). According to these studies, pig chromosome 2 microsatellite markers are associated with several meat quality traits, such as marbling, lipid percent, drip loss, muscle pH, tenderness, meat color and muscle fiber diameter.

Many of the meat quality QTL were mapped to the intermediate region of SSC2 (*SW2445-S0565*). This identified chromosomal region spans about 100 cM, which contains four different human chromosomal fragments, HAS11, HSA19, HSA1 and HSA5 (Meyers et al., 2005). Previously, Jungerius and coworkers (2003) have identified over 300 single nucleotide polymorphisms (SNPs) on SSC2

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**Table 1.** PCR primers and conditions used for amplification and sequencing

| Gene | STS name* | Accesion no* | Primer* | | Annealing Temp. | Product size |
|---|---|---|---|---|---|---|
| | | | Forward (5'→3') | Reverse (5'→3') | | |
| *PTH* | PTHsts1 | BV079397 | ACCAGGAAGAGATCTGTGAGTG | TGCCCTATGCTGTCTAGAGC | 56 | 311 |
| *CSF2* | CSF2sts1 | BV079385 | CAGCATGTGGATGCCATC | GTACAGCTTCAGGCGAGTCTG | 56 | 974 |
| *FOLR1* | FOLR1sts1 | BV012577 | AGACGGTCCTTCTGCCTGT | TTGAGGAGGAGCCTATGGTTT | 58 | 356 |
| | P006C12sts1 | BV079398 | TGAGTACTCGTTATGGACGC | CTGTGCCTTTAGGACTGAGG | 56 | 506 |
| | P006D12sts1 | BV079401 | CCAAGATACAGAAGTAGGAGC | TGCAGTCTTCTTGGTGCAGG | 56 | 393 |
| *BDNF* | P006A04sts1 | BV079400 | ATATCAGGTGCTCACAGTGC | GACTTAACTCTAGGAGTTCC | 56 | 612 |
| *LDHA* | LDHAsts2 | BV012579 | TTTCACTGTCTAGGCTACAACAAGA | AGCTGGATAGTTGGCTGCAT | 56 | 517 |
| *RPS13* | P005E11sts1 | BV079399 | CTTCCCCTAATGTCAGTG | ATTAAGAGACAGTAGAGTCC | 45 | 799 |
| *ADM* | ADMsts2 | BV079374 | ATTGAGAGACCGAGAGTCCG | TTGCTACTTCGCATATCACCC | 56 | 646 |
| *CAT* | CATsts1 | BV079378 | TGCCTCTGAAACAAAACGTG | TTCAAAAGACCCCAAAGCAT | 58 | 458 |
| *WT1* | WT1sts1 | BV079371 | TTAACATTCCTCCTGGCTCG | GCCTTGCCCTCTGATTTATTT | 60 | 425 |
| *FSHB* | FSHBsts2 | BV079389 | GCCAGCTTCAGGCTAACATT | GACTTCATCTTGGGGTGGAA | 58 | 1,101 |
| *MYOD1* | MYOD1sts3 | BV012581 | GGTGACTCAGACGCATCCA | ATAGGTGCCGTCGTAGCAGT | 60 | 599 |
| *IL4* | IL4sts1 | BV079417 | GATCCCCAACCCTGGTTCTGCT | GGCAGAAAGACGTCGTCAC | 56 | 434 |
| *ADRB2* | ADRB2sts1 | BV079375 | CAAGTACCAGAGCCTGCTGACC | TAGAGAAGGGCAGCCAGC | 62 | 455 |

* The STS name, accession number and primer information used in the Table 1 is reported by Jungerius et al. (2002).

with a DNA panel of eight pigs which were one Meishan, one Pietrain, one Wild Boar and five Large Whites. These SNPs will certainly have a potential for fine mapping of the observed QTL region on pig chromosome 2. Therefore, the purpose of this study was to further characterize and implement the SNPs for identification of candidate genes associated with meat quality QTL on SSC2 in commercial pig breeds.

## MATERIALS AND METHODS

### Animals

The sequencing DNA panel for SNP detection consisted of two individuals from each of five commercial breeds in Korea, such as Berkshire, Duroc, Large White, Landrace, and Korean Native Pig (KNP). The Berkshire×Yorkshire population developed at Iowa State University was used to localize the SNP and study phenotypic association of the SNPs (Malek et al., 2001).

### Primer design and polymerase chain reaction

A total of 15 primer pairs were tested for amplification and sequencing of 13 candidate genes (*PTH, CSF2, FOLR1, BDNF, LDHA, RPS13, ADM, CAT, WT1, FSHB, MYOD1, IL4* and *ADRB2*). These genes were selected for their known biological roles in skeletal muscle development and metabolism and probable locations within the QTL region. Primer sequences of these candidate genes were obtained from Jungerius et al. (2003). Polymerase chain reactions were performed in 10 μl volumes contained 12 ng of genomic DNA, 10 pmol of each primer, 200 μM of each dNTP, 2.5 units of *Taq* DNA polymerase (Enzynomics$^{TM}$, Korea), and reaction buffer with 1.5 mM $MgCl_2$. Thermocycling reaction was performed in a PTC-200 thermocycler (MJ Research, Watertown, MA, USA) with a 10 min initial denaturation at 94°C, 35 cycles of 94°C for 30 s, 45-62°C for 40 s, 72°C for 30 s, and a final extension at 72°C for 10 min. The result of the PCR reaction was identified by 1% agarose gel electrophoresis at 100 mV for 20 min. The information for each primer sequence, annealing temperature and fragment size are given in Table 1.

### Sequencing, polymorphism identification and genotyping

A total of 15 PCR products were sequenced with both forward and reverse amplification primers at Genotech Co. (Daejeon, Korea). Sequencher software (Gene Codes, version 4.6, Ann Arbor, MI) was used to assemble the sequences and to identify DNA polymorphisms. Polymorphic sites were analyzed for putative restriction fragment length polymorphisms (RFLPs) using the NEBcutter program (http://tools.neb.com/NEBcutter2/index.php). Genotyping of the putative RFLPs was performed on individual DNA samples from five different pig breeds which included Duroc, Landrace, Large White, Berkshire and Korean Native pig. All restriction enzymes were supplied by New England BioLabs (Ipswich, MA, USA) and restriction digestions were performed according to manufacturer's recommendations. Digested PCR products were analyzed on 2.5-4% agarose gels and each allele was scored manually. The restriction enzymes and polymorphic fragment sizes used for SNP genotyping were given in Table 2.

### Statistical analysis

The SNPs of three candidate genes (*PTH, CSF2, FOLR1*) were chosen to linkage map on pig chromosomes with Berkshire×Yorkshire (B×Y) family (Malek et al., 2001) using the CRIMAP (version 2.4) mapping program (Green et al., 1990). Associations between the SNPs of the three candidate genes and meat quality traits in the B×Y F2

**Table 2.** PCR primers and restriction enzymes used for SNP genotyping.

| Gene | Primer sequences (5'→3') | Fragment size (bp) | $T_A$(°C) | Restriction enzyme | Size (bp) of the allelic polymorphism |
|---|---|---|---|---|---|
| *CAT* | TGCCTCTGAAACAAAACGTG | 458 | 56 | *Msc* I | 458,353 |
| | TTCAAAAGACCCCAAAGCAT | | | | |
| *PTH*-1 | ACCAGGAAGAGATCTGTGAGTG | 311 | 56 | *APek* I | 218,136 |
| | TGCCCTATGCTGTCTAGAGC | | | | |
| *PTH*-2* | ACCAGGAAGAGATCTGTGAGTG | 201 | 56 | *Tsp509* I | 201,108 |
| | ATGGCTCTCAACCAGGACAT | | | | |
| *WT1*-a | TTAACATTCCTCCTGGCTCG | 425 | 60 | *Hpy188* I | 137,74 |
| *WT1*-b | GCCTTGCCCTCTGATTTATTT | | | *Aci* I | 425,251 |
| *MYOD1* | GGTGACTCAGACGCATCCA | 599 | 60 | *Dde* I | 599,340 |
| | ATAGGTGCCGTCGTAGCAGT | | | | |
| *IL4* | GATCCCCAACCCTGGTTCTGCT | 434 | 56 | *Alu* I | 308,194 |
| | GGCAGAAAGACGTCGTCAC | | | | |
| *FOLR1*-1 | CCAAGATACAGAAGTAGGAGC | 393 | 58 | *Dde* I | 161,126 |
| | TGCAGTCTTCTTGGTGCAGG | | | | |
| *FOLR1*-2* | CACCTGTGGAAGCAGAGTTC | 273 | 56 | *Dde* I | 273,155 |
| | CTGTGCCTTTAGGACTGAGG | | | | |
| *CSF2*-1 | CAGCATGTGGATGCCATC | 974 | 56 | *Hha* I | 870,588 |
| | GTACAGCTTCAGGCGAGTCTG | | | | |
| *CSF2*-2* | GCTGTGATGGTGAGTGAGGA | 362 | 56 | *Mbo* II | 362,276 |
| | CCCTTGAATGCTAGGACTGC | | | | |

* The three SNPs were genotyped in the ISU Berkshire×Yorkshire cross F2 animals for linkage and association analyses.

**Table 3.** Comparison of SNPs between two independent studies

| Gene | STS name | Accesion no. | Product size (bp) | No. of SNPs in Jungerius et al. | Site of SNPs in this study |
|---|---|---|---|---|---|
| *PTH* | PTHsts1 | BV079397 | 311 | 5 | 4: 111(C/T), 229(A/G), 246(A/G), 249(C/T) |
| *CSF2* | CSF2sts1 | BV079385 | 974 | 10 | 7: 103(C/T), 104(A/G), 155(C/T), 192(A/G), 459(G/T)*, 650(C/G), 691(C/T) |
| *FOLR1* | FOLR1sts1 | BV012577 | 356 | 2 | 2: 75(C/T)*, 256(A/G) |
| | P006C12sts1 | BV079398 | 506 | 18 | 1: 209-212(ATAC)**, 219(G/T) |
| | P006D12sts1 | BV079401 | 393 | 8 | 5: 26(C/T)*, 31(A/G), 39(A/G)*, 69(A/C), 313(C/T) |
| *BDNF* | P006A04sts1 | BV079400 | 612 | 5 | 0 |
| *LDHA* | LDHAsts2 | BV012579 | 517 | 1 | 1: 471(A/G) |
| *RPS13* | P005E11sts1 | BV079399 | 799 | 1 | 1: 115(T)**, 487(A/C), 768(A)** |
| *ADM* | ADMsts2 | BV079374 | 646 | 4 | 3: 157(A/G), 452(A/G) , 552~553(TG)**, 562(A/T)* |
| *CAT* | CATsts1 | BV079378 | 458 | 1 | 2: 243(A/G)*, 356(A/G) |
| *WT1* | WT1sts1 | BV079371 | 425 | 2 | 3: 64(A/G), 237(C/T), 252(C/T)* |
| *FSHB* | FSHBsts2 | BV079389 | 1,101 | 5 | 2: 335(A/G), 447(A/G), 518(G)** |
| *MYOD1* | MYOD1sts3 | BV012581 | 599 | 2 | 2: 343(A/C), 345(G/T)* |
| *IL4* | IL4sts1 | BV079417 | 434 | 1 | 1: 321(C/T) |
| *ADRB2* | ADRB2sts1 | BV079375 | 455 | 0 | 0 |
| Total | | | 8,586 bp | 62 SNPs | 34 SNPs |

* New SNPs. ** Different nucleotide with public sequences.

population were performed using SAS mixed model procedure (SAS procedure MIXED; SAS institute, Gary, NC, USA) with a model that included sex, slaughter date and marker genotype as fixed effects and dam as a random effect.

## RESULTS

### PCR amplification, sequencing and SNP detection

A total of 15 primer sets for 13 candidate genes successfully amplified a single band, and used for subsequent sequencing of the amplicons. Ten animals from five different commercial pig breeds representing Duroc, Large White, Landrace, Berkshire and Korean Native pigs were sequenced for the 15 primer sets. A total of 34 SNPs were detected in the 13 primer sets (Table 3). A total of 11 SNPs were used to determine allelic frequencies of SNP among commercial pig populations using RFLP methods. Jungerius et al. (2003) detected 62 SNPs from the 15 amplicons of eight animals made of one Meishan, one

**Table 4.** Allele 1* frequencies of SNPs on pig chromosome2 in the five pig breeds

| Gene | Berkshire (N = 10) | Duroc (N = 10) | Landrace (N = 10) | Yorkshire (N = 10) | KNP (N = 10) |
|---|---|---|---|---|---|
| *CAT* | 0 | 0.25 | 0.16 | 0.25 | 0 |
| *PTH*-1 | 1 | 0 | 0.2 | 0.13 | 0.67 |
| *PTH*-2 | 0 | 1 | 0.8 | 0.87 | 0.33 |
| *WTI*-a | 0.33 | 0 | 0.6 | 0.13 | 0.5 |
| *WTI*-b | 0 | 0 | 0.17 | 0 | 0 |
| *MYOD1* | 0.49 | 0.5 | 0.46 | 0.57 | 0.5 |
| *IL4* | 0.2 | 0.16 | 0.3 | 0.4 | 0.1 |
| *FOLR*-1 | 0 | 0 | 0.16 | 0 | 0.35 |
| *FOLR*-2 | 0.42 | 1 | 0.95 | 1 | 1 |
| *CSF2*-1 | 0.05 | 0.21 | 0.28 | 0.25 | 0 |
| *CSF2*-2 | 0.25 | 0.25 | 0.63 | 0.65 | 0.14 |

* Uncut fragment.

**Table 5.** Association of 3 candidate genes (*PTH*, *CSF2* and *FOLR1*) genotypes and phenotypic traits from Berkshire×Yorkshire cross F2 pigs

| Gene | Trait[2] | Genotypic least squares means[1] (SE) | | | |
|---|---|---|---|---|---|
| | | 11 | 12 | 22 | p-value |
| *PTH* | Cholesterol (mg/100 g) | 57.99 (1.03) | 59.83 (0.7)[c] | 57.14 (0.9)[d] | 0.019 |
| (n = 299) | 24-h loin Minolta L value | 21.02 (0.71) | 20.27 (0.54)[a] | 21.68 (0.64)[b] | 0.04 |
| *CSF2* | Average back fat (cm) | 3.33 (0.07)[a] | 3.19 (0.08)[b] | 3.34 (0.13) | 0.1 |
| (n=374) | Cholesterol (mg/100 g) | 57.96 (0.6)[a] | 59.16 (0.74) | 61.36 (1.51)[b] | 0.07 |
| | Lumber back fat (cm) | 3.62 (0.08)[a] | 3.46 (0.09)[b] | 3.57 (0.15) | 0.12 |
| | Average daily gain to weaning (kg/day) | 0.236 (0.008) | 0.249 (0.009)[a] | 0.218 (0.159)[b] | 0.07 |
| | Fiber type II ratio | 1.027 (0.064)[e] | 1.085 (0.081)[c] | 1.591 (0.181)[d,f] | 0.01 |
| | 48-h loin Minolta L value | 22.10 (0.24)[a] | 21.96 (0.3)[a] | 20.56 (0.66)[b] | 0.09 |
| *FOLR1* | Chewiness score (1-10) | 2.34 (0.12)[a] | 2.63 (0.11)[b] | 2.31 (0.19) | 0.07 |
| (n = 166) | Flavor score (1-10) | 3.00 (0.22) | 2.77 (0.18)[a] | 3.55 (0.33)[b] | 0.1 |
| | Carcass weight (kg) | 87.47 (0.32)[a] | 87.07 (0.26)[a] | 86.02 (0.49)[b] | 0.04 |
| | Drip loss (%) | 5.78 (0.19)[a] | 5.69 (0.16) | 5.10 (0.28)[b] | 0.11 |
| | 24-h Semimembranosus Hunter L values | 41.26 (0.39)[a] | 41.83 (0.33) | 42.70 (.59)[b] | 0.11 |
| | 24-h Semimembranosus Minolta L values | 17.13 (0.35)[a] | 17.58 (0.29) | 18.38 (0.52)[b] | 0.11 |

[1] Significance levels: [a, b] 0.05; [c, d] 0.01; [e, f] 0.005.

[2] Detailed information on these traits is reported in Malek et al. (2001).

Pietrain, one Wild Boar and five Large White pigs, while 34 SNPs were found from the five pig breeds used in this study (Table 3).

**Allelic variation of SNP in pigs**

A total of 11 SNPs were genotyped in five different commercial pig breeds and a summary of the frequencies is presented in Table 4. Only three SNPs were polymorphic in all five pig breeds. In Berkshire pigs, five SNPs out of 11 SNPs were fixed although the number of tested animals is relatively small. Six out of 11 SNPs were monomorphic in Duroc pigs, and two of the monomorphic SNPs were common ones between Berkshire and Duroc pigs. It is interesting to note that alternative alleles in PTH SNPs were also fixed between Berkshire and Duroc pigs. In Yorkshire pigs, three SNPs were fixed, but only one SNP was fixed in Landrace pigs. Korean Native pigs had a similar pattern of SNP genotypes to that of the animals from Berkshire breed, which had similar allelic frequencies between the Korean Native pigs and Berkshire breeds tested.

**Linkage mapping and association with pork quality**

Three candidate genes (*PTH*, *CSF2* and *FORL1*) were chosen as anchor loci to determine the precise linkage with QTL on SSC2, and their SNPs were genotyped at a three-generation Berkshire×Yorkshire reference family (Malek et al., 2001). Two- and multi-point linkage analyses of the *PTH* and *CSF2* loci mapped both *PTH* and *CSF2* to SSC2 as follow: *SW2445*-10.7 cM-*LXRA*-13.1 cM-*SW1686*-12.1 cM-*PTH*-1.9 cM-*RETN*-7.3 cM-*SW766*-2.3 cM-*CAST*-14.5 cM-*SW1408*-3.4 cM-*SW2157*-10.3 cM-*CSF2*-7.8 cM-S0565. Association analyses of *PTH* and *CSF2* genotypes revealed that significant effects of a *PTH* polymorphism on cholesterol content ($p<0.02$) and 24-h loin Minolta L value ($p<0.05$), and a significant *CSF2* genotypic effects on fiber type II ratio ($p<0.02$) and several closely significant effects on cholesterol content ($p = 0.07$), lumbar backfat ($p = 0.12$), average daily gain to weaning ($p = 0.075$), 48-h loin Minolta L value ($p = 0.089$) (Table 5). Linkage analyses mapped the *FOLR1* to SSC9 with the following map order: SWR68-3.4 cM-*FOLR1*-11.5 cM-SW21-18.6 cM-S0024.

Closely significant fixed effects were found between *FOLR1* genotype and meat quality traits, but significant phenotypic variation were found between *FOLR1* genotypes on several meat quality traits (Table 5).

## DISCUSSION

SNP markers have high potential for detailed haplotype analayses and applications in association studies to identify the underlying genes responsible for the observed QTL effects. In this study, we report re-identification of SNPs on pig chromosome 2, SNP allelic frequencies between commercial breeds, and meat quality association of SNP alleles.

In total, 15 primers pair amplicons were sequenced for SNP identification from Jungerius et al. (2003). In the total contig length of 8,586 bp, 34 polymorphic positions were identified in our study, while Jungerius et al. (2003) found 62 SNPs. The SNP density difference between the two data sets was largely due to three STSs (BV079398, BV079400 and BV079389). In Jungerius et al. (2003), BV079398 (*FOLR1*), BV079400 and (BDNF) STSs contained 18 and 5 SNPs respectively, but we found only one SNP in the BV079398 STS and no SNP in BV079400 STSs (Table 3). This result revealed that the number of SNP might be variable among the pig breeds in comparison, and sequence data sources used for SNP identification.

Knowing allelic distribution of SNPs is important for commercial application of these polymorphisms. Although a small sample size was used in this study, it was possible to determine to some extent that the pattern of SNPs differed for each population. Of 11 SNPs tested for allelic distribution, 9 SNPs were fixed in at least one commercial population. Based on the information available in this study, it was suggested that between Duroc and Berkshire pigs might be sharing a higher genomic similarity than Yorkshire or Landrace pigs on pig chromosome 2. However, in case of the *PTH* gene polymorphisms, the two SNPs appear to be alternatively fixed between Berkshire and Duroc pigs, which suggest a selection force for the *PTH* gene may be acted on either Berkshire or Duroc pigs. In addition, Korean native pigs showed a high similarity in allelic frequencies to Berkshire pigs, which agree with the previous microsatellite loci analysis of Korean native pigs (Kim et al., 2005).

Fresh pork quality is controlled by several factors including processing and sensory characteristics, which can be measured as intramuscular fat contents, ultimate pH, color, and water holding capacity etc. Berkshire breed was demonstrated with very positive meat quality from these measurements (Goodwin and Burroughs, 1995). Using Berkshire and Yorkshire crossed pigs, several studies (Ciobanu et al., 2004; Otieno et al., 2005; Yu et al., 2006) mapped candidate genes, *LXRA*, *RETN* and *CAST* respectively. One *CAST* haplotype was significantly associated with lower Instron force, cooking loss and higher juiciness (Ciobanu et al., 2004), a *RETN* allele was significantly associated with more marbling, total lipids, firmness, WHC and loin pH (Otieno et al., 2005), and *LXRA* allele was significantly associated with loin eye area and total lipid (Yu et al., 2006). However, some of these effects were not detected in other commercial pig populations probably due to differences in linkage disequilibrium across pig populations.

From our results, there were several significant phenotypic differences between individual genotypes (Table 5). The 12 genotype of *PTH* polymorphism was associated with more cholesterol content and darker meat color. While the 12 genotypes of *CSF2* polymorphism was associated with lower back fat and higher growth rate to weaning, the 22 *CSF2* genotypes was associated with more type IIA fiber and darker meat color. Fiber type II ratio is the ratio of density of the myosin IIA to the density of the myosin IIB within muscle fiber composition. Muscle fiber type composition contribute to variation in eating quality of meat, but its direct effect is not clear because meat quality is associated with other components such as sarcoplasmic proteins, muscle enzymes, intramuscular fat, and connective tissues. However, more fiber type IIA are red, and increasing the proportion of IIB fiber increased the rate and extent of post mortem pH decline, leading to a higher cooking loss (Larzul et al., 1997). The favorable association of *CSF2* allele 2 with meat quality was not surprising, given the fact that the allele 2 is at a high frequency in Berkshire breed (Table 4). Therefore our results might be useful for the further fine mapping of the QTL region on SSC2.

Sato et al. (2003) found both intramuscular fat and muscle moisture QTL on SSC9 where the *FOLR1* is closely assigned. In addition, the 22 genotype of *FOLR* polymorphism was associated with more flavor and less drip loss in meat, but lighter meat color (Table 5). Previously, uncoupling protein 2 and 3 (UCP2 and 3) were mapped at the same regions on SSC9 and considered as candidate genes for the intramuscular fat QTL (Werner et al., 1999).

In conclusion, comparative re-sequencing of 15 PCR products has identified 34 SNP markers which might be important for commercial pork production in Korea. More work is necessary with more genes and animals to utilize the DNA marker information on SSC2 for superior pork production, and the identified SNPs from this study would be a primary step identifying individual pigs with high pork quality as well as determining origin of the pig breed in commercial populations.

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