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Rapid communication: Linkage and physical mapping of the porcine basic fibroblast growth factor (FGF2) gene

D. R. Holz
Iowa State University

J. Helm
Iowa State University

Y. D. Zhang
Iowa State University

Max F. Rothschild
Iowa State University, mfrothsc@iastate.edu

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Abstract

Genus and species. Sus scrofa. Locus. Pig Fibroblast Growth Factor 2 (FGF2) gene. Source and Description of Primers. Primers were designed from human sequence (GenBank accession no. J04513.1) to amplify a 167-bp fragment within exon 1 of FGF2 from pig genomic DNA. This fragment was identified as FGF2 with 93% homology to the human FGF2 sequence.

Keywords

Genetic Polymorphism, Gene Mapping, Fibroblasts

Rapid communication: Linkage and physical mapping of the porcine *basic fibroblast growth factor* (*FGF2*) gene[1]

D. R. Holz, J. Helm, Y. D. Zhang, and M. F. Rothschild[2]

Department of Animal Science, Iowa State University, Ames 50011

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Source and Description of Primers. Primers were designed from human sequence (GenBank accession no. J04513.1) to amplify a 167-bp fragment within exon 1 of *FGF2* from pig genomic DNA. This fragment was identified as *FGF2* with 93% homology to the human *FGF2* sequence.

Primer Sequences. Primers designed from human sequence were as follows: forward primer: 5′ GCA GCC GGG AGC ATC ACC AC 3′; reverse primer: 5′ TCG CTC TTC TCC CGG ACC C 3′. Pig-specific primers were as follows: forward primer: 5′ TGA ATA TAA AAA ATC CTA AGC GGT TGC ACT 3′; reverse primer 5′ CGC TCT TCT CCC GGA CCC 3′.

Method of Detection. The PCR reaction using the pig-specific primers was performed using 12.5 ng of porcine genomic DNA, 1× PCR buffer, 1.5 m*M* $MgCl_2$, 100 μ*M* dNTP, 2.5 p*M* of each primer, and 0.35 units of *Taq* DNA polymerase (Promega, Madison, WI) in a final volume of 10 μL. The PCR program consisted of an initial 4 min 94°C denaturing, 35 cycles of 45 s at 94°C, 45 s at 57°C, 45 s at 72°C, and a final 5-min extension at 72°C in a MJ PTC-100 thermocycler (MJ Research, Watertown, MA). The PCR product was digested with *Bts*I at 37°C overnight and fragments were separated by electrophoresis on a 4% Nusieve gel (BMA, Rockland, ME).

Description of Polymorphism. A single nucleotide polymorphism (**SNP**), C/T base pair change (silent mutation), was detected in the pig sequence at the human *FGF2* nucleotide position 568 (GenBank accession no. J04513.1). A *Bts*I RFLP test was developed for this exon 1 SNP by designing pig-specific primers to amplify a 99-bp fragment and incorporate a *Bts*I restriction enzyme site. The *Bts*I digestion of the 99-bp exon 1 PCR fragment produced the following genotypes: homozygous 1/1 had a 99-bp fragment, heterozygous 1/2 genotype had 99-, 73-, and 26-bp fragments, and the 2/2 homozygous genotype had 73- and 26-bp fragments (Figure 1).

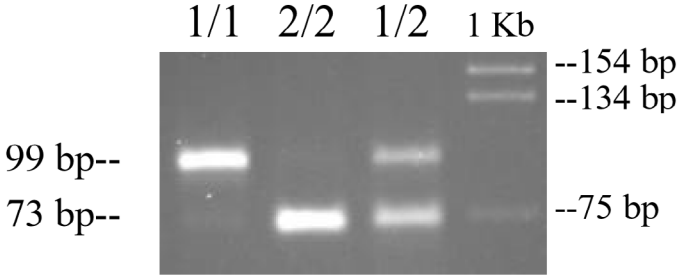

Figure 1. *FGF2 Bts*I PCR-RFLP allelic fragments are shown on a 4% Nusieve agarose gel. The 99-bp and 73-bp fragments represent alleles 1 and 2, respectively (26-bp fragment not shown).

Pattern of Inheritance. Mendelian inheritance was observed in the three-generation Swedish family of the European PiGMap family (Archibald et al., 1995).

Allele Frequencies. Allele 1 was identified in only 2 (one homozygous and one heterozygous individuals) of the 22 grandparents of the European PiGMaP families. A total of 312 unrelated animals from five breeds (Duroc, Hampshire, Meishan, Landrace, and Large White) were also genotyped and the combined gene frequency for allele 1 was 0.013.

Chromosomal Location. The *FGF2* gene was physically mapped to porcine chromosome 8 (SSC8) q23–27 using the pig/rodent somatic cell hybrid panel (Yerle et al., 1996). Two-point and multipoint linkage analysis were performed on PiGMaP family *FGF2 Bts*I genotypes using the CRI-MAP program (Green et al., 1990). The *FGF2* gene was most closely linked to *IL2*, *S0225*, *S0442*, and *S0447* with equal recombination fractions of 0.05 and LOD scores of 4.3, 4.3, 4.3 and 4.02, respectively. The most probable order of *FGF2* between linked markers (in Kosambi centimorgans) is *S0069*(9.0)-*S0225*(21.8)-*S0144*(26)-*S0442*(32.6)-*S0447*(32.6)-*FGF2*(37.7)-*SW61*(57.0)-*SPP1*(68.8).

Comments. The *FGF2* gene is multifunctional and has been associated with mitogenesis and angiogenesis (Florkiewicz et al., 1991). Mutations within *FGF2* could have strong implications in cell growth regulation and tissue repair (Abraham et al., 1986), making it a poten-

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[2]Correspondence: E-mail: mfrothsc@iastate.edu.

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tially important gene in muscle development association studies.

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