Rapid Communication: Mapping the Pig VCAM1 Locus to Chromosome 4 Using a Double-Stranded Conformation Polymorphism Marker (VCAM1-2)

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Abstract
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Keywords
Pigs, Gene Mapping

Disciplines
Agriculture | Animal Sciences | Genetics and Genomics

Comments
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Rapid Communication: Mapping the Pig VCAM1 Locus to Chromosome 4 Using a Double-Stranded Conformation Polymorphism Marker (VCAM1-2)1

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Source and Description of Primers. We previously identified a SacI polymorphism by using a pig VCAM1 cDNA probe on Southern blots (VCAM1-1; Helm et al., 1994). This polymorphism was not informative enough to map VCAM1. To develop PCR-based genotyping, we sequenced the 3’ untranslated region of pig VCAM1. Subsequently, a pig VCAM1 cDNA was deposited in Genbank; our data agree completely with that reported by Tsang et al. (Accession: U08351). The PCR primers were designed (forward, 5’-TATCAGCCCTCATAATGCATCAT 3’ and reverse, 5’-GAAATTGTTGTCCATGACCTTATA 3’).

Method of Detection. The primers were used to PCR amplify a 193-bp segment (25 μL reactions, 1.5 mM MgCl2, 0.5 pmol of primer) under the following program: 95°C 4 min; 5 cycles of 94°C 1 min, 48°C 1 min, and 72°C 1 min; then 35 cycles of 90°C 1 min, 48°C 1 min, and 72°C 1 min; and a final 72°C 5-min incubation. The PCR fragments were sequenced to confirm VCAM1 was amplified.

Description of Polymorphism. Three VCAM1 alleles were identified based on double strand conformation polymorphisms (DSCP) and heteroduplex analysis of PCR products (see Figure 1).

Inheritance Pattern. Most F1 crosses within the PiGMaP gene mapping families were informative for this marker (142 informative meioses). No deviation from expected Mendelian segregation was observed.

Frequency. Analysis of 53 unrelated pigs across five breeds (Duroc [17], Landrace [10], Large White [12], Meishan [12], and Wild Boar [2]) shows that the allele frequency in commercial breed type animals is allele A, 6%; allele B, 67%, and allele C, 27%. Allele A was seen only in Landrace (25% frequency) and in Meishan (100%).

Chromosomal Location. The VCAM1 was mapped to chromosome 4 with two-point LOD scores ranging from 3.11 to 13.73 for several SSC4 markers. The best multipoint map indicates the gene order (cM distance) for these loci is S0073-(9)-S0214-(14)-TSHB-(9)-S0067-(11)-VCAM1-(11)-S0161.

Comments. The VCAM1 locus maps in humans to HSA1p3.1-3.2 (Cybulsky et al., 1991), between two conserved syntenic groups found on SSC4 and SSC6. Our new result places VCAM1 within the syntenic group on SSC4.

Literature Cited


Key Words: Pigs, Gene Mapping

Figure 1. The PCR products of VCAM1 3’ untranslated region show six genotypes. The PCR products were electrophoresed on 15% gels (37.5:1 acrylamide:bis ratio). (A) Two single bands of differing size and three patterns of multiple bands were initially observed. (B) Pedigree information allowed definition of the A, B, and C alleles. In the first pedigree (B1), even though parentals look identical, a new pattern in the offspring is observed (called BC), indicating two indistinguishable alleles are present. The second pedigree (B2) confirms this as the mating between an AA animal and a BC animal creates two new patterns (AC and AB); the third (B3) and fourth (B4) pedigrees show patterns consistent with AA × CC and AC × AB, respectively. (C) Results of mixing experiments used to confirm DSCP and to distinguish the B and C homozygote genotypes: equal amounts of unknown DNA were mixed with known AA, BB or CC DNA. One PCR cycle was run as above to denature/renature the DNA, and electrophoresis was then performed to determine the band pattern obtained. Known genotypes: Lanes 1–4, AA; Lane 9 and 11, AB; Lane 14, AC; Lane 17, BC. Lanes 5–8 were unknown BB/CC genotype. Lane 10, mix of lanes 3 and 6; lane 12, mix of lanes 7 and 1; lane 13, mix of lanes 3 and 7; lane 15, mix of lanes 1 and 8; lane 16, mix of lanes 3 and 8; lane 18, mix of lanes 7 and 8.