

Linkage mapping of a single nucleotide polymorphism (SNP) in the porcine *QDPR* gene to chromosome 8^{1,2}

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Source/description of primers: Quinoid dihydropteridine reductase (*QDPR*) catalyzes the NADH-mediated reduction of quinonoid dihydrobiopterin and is an essential component of the pterin-dependent aromatic amino acid hydroxylating systems¹. A cDNA clone containing the full coding region of the porcine *QDPR* (GenBank accession no. AF526879) has been isolated from the 'Meat Animal Research Center (MARC) 2PIG' expressed sequence tag primary library². Primers were designed to amplify across the 3' untranslated region of the cDNA. The forward (F3) and reverse (R1) primers correspond to bases 744–763 and 1118–1098 of the porcine *QDPR* cDNA. Agarose gel electrophoresis and sequencing of the polymerase chain reaction (PCR) amplicons of porcine genomic DNA indicated that the size of this product was 375 bp as expected.

PCR Primer sequences and a flanking sequence for a single nucleotide polymorphism: *QDPR* F3: CCACGCAGGGAAAGACAGAG
QDPR R1: CATCCAGCAAATCTCTGACC
Sequence flanking polymorphism: ATGTGTCCCC(G/A)ATGGTGGCCG

PCR conditions: Polymerase chain reactions were carried out in a 25- μ l volume containing 100 ng genomic DNA, 1.5 mM MgCl₂, 20 pmol of each primer, 100 μ M dNTP, and 0.35 U *Taq* polymerase. Amplification was performed under the following PCR conditions; 2 min at 94 °C; 45 cycles of 30 s at 94 °C, annealing for 1 min at 61 °C, 1 min at 72 °C; and a final extension of 5 min at 72 °C. The amplified genomic DNA of eight parents (seven F1 sows and one white composite boar) from a subset of the MARC swine reference population³ were bidirectionally sequenced and evaluated for polymorphisms⁴.

Polymorphism and chromosomal location: An A/G single nucleotide polymorphism was detected at nucleotide 1075 in AF526879. This polymorphism was heterozygous in five of the seven F1 sows and two boars from the MARC swine reference population³. An assay was designed to genotype this polymorphism using primer extension with analyte detection on a

¹Mention of trade names or commercial products in this article is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the US Department of Agriculture.

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MALDI-TOF mass spectrometer (Sequenom Inc., San Diego, CA, USA)⁵. This marker generated 153 informative meioses in the MARC swine reference population. The *QDPR* gene was mapped to chromosome 8 position 25.7 cM, which is the same position as marker *PEPS*⁶ on the current MARC swine chromosome 8 linkage map (<http://www.marc.usda.gov/>) using CRI-MAP. The most significant two-point linkage detected was with *PEPS* (LOD = 29.50) at 0 recombination. The *QDPR* gene in human is located on chromosome 4p15.31, which shares homology with swine chromosome 8.

References

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Fishing *in silico*: searching for tilapia genes using sequences of microsatellite DNA markers

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Description: Genetic linkage maps of some of the main cultured fish species were constructed in recent years. Maps of tilapia^{1–3}, trout⁴ and catfish⁵ consist of hundreds of microsatellites and amplification fragment length polymorphism DNA markers, but only several genes.

Microsatellites DNA markers are short tandem repeats, usually dinucleotide (CA)_n, with unique flanking sequences, providing primer binding sites for PCR amplification⁶. Microsatellites are highly abundant throughout the genome and appear in coding and non-coding regions. Therefore, it is likely that the flanking sequences can be part of a gene, which can be identified by similarity searches against the GenBank sequence database. This *in silico* data mining approach has already been used to identify genes in mice⁷, cattle, pigs and chicken⁸. In this study we utilized a similar approach to add nine anchored genes to the current 14 genes in the tilapia³ map, and suggest seven additional genes that can be anchored by mapping to their matching microsatellites.

Methods: Microsatellites sequences were downloaded in FASTA format from the GenBank database using the Entrez nucleotide query webpage (<http://www.ncbi.nlm.nih.gov/Entrez>). The search string used to retrieve the tilapia microsatellites was