Rapid Communication: Molecular Cloning of the Porcine Corticotropin-Releasing Factor Gene

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Name of the Sequence. Porcine corticotropin-releasing factor gene.

Genus and Species. Sus scrofa.

Origin of Clone. A 664-bp DNA fragment encoding the complete corticotropin-releasing factor (CRF) gene was amplified from pig genomic DNA using the following primers: forward 5′-CATGGGCTGCCGCTGCTBGTGTCC-3′; reverse 5′-GCACGCTATGAGTACAGAATACTG-3′. The primers were derived from a multiple sequence alignment of human, rat, and mouse sequence data. The PCR product was subcloned into PCR-Script™ (Stratagene, La Jolla, CA) and both strands of the subcloned DNA fragment were characterized by automatic DNA sequencing using an ABI 373A DNA sequencer. The cloning and sequencing procedure was performed on two recombinant plasmids derived from two independent amplification products to generate an overall consensus sequence.

Comparison with Related Sequences. The pig CRF coding region nucleotide sequence (Figure 1) is 80% identical to human (Shibahara et al., 1983), 84% identical to sheep CRF (Roche et al., 1988), and 85% identical to rat CRF (Jingami et al., 1985), supporting the CRF character of the clone. The putative amino acid sequence of the porcine CRF is 83% similar to human and 74 and 75% similar to sheep and rat, respectively. The coding region for pig CRF is 12 nucleotides longer than that of the human sequence (Shibahara et al., 1983) and 15 nucleotides shorter than the rat nucleotide sequence (Jingami et al., 1985).

Sequence Data. A 664-bp genomic DNA fragment contains the complete porcine CRF gene sequence. The pig CRF gene contains no introns and has an open reading frame of 573 nucleotides encoding a protein of 191 amino acid residues. Nucleotides (1–25) may not accurately represent the porcine CRF gene sequence due to their incorporation into the final product by the forward primer annealing to porcine genomic DNA.

EMBL/GenBank Accession Number. Y15159.

Comments. The neuropeptide CRF plays an important role within the central nervous system in controlling neuroendocrine, physiological, and behavioral responses to stress. Identification of the gene sequence for porcine CRF will permit nucleotide probes to be developed for the study of gene expression in the brains of pigs exposed to environmental stressors. Knowledge thus obtained is likely to contribute to improvements in the welfare of intensively housed swine.