Rapid Communication: Restriction Fragment Length Polymorphisms in the Bovine *Calpastatin* Gene[1]

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Polymorphisms. *Bam*HI, *Eco*RI, and *Taq*I restriction fragment length polymorphisms at the bovine *calpastatin* locus.

Source and Description of Probe. A 2.2-kb cDNA insert coding for Domains 2 through 4 plus a 3′ untranslated region of the bovine *calpastatin* gene was excised from the pBluescript vector (Stratagene, La Jolla, CA) with *Eco*RI and *Xho*I.

Method of Detection. Nylon filters were prehybridized in 5× Denhardt's, .5% SDS with .1 μg/μL of sheared salmon sperm DNA for 2 h at 60°C in a hybridization oven. Random-primed labeled probe (1.0×10^8 cpm/mL) was added to 10 mL of .5 *M* $NaPO^4(H^3PO^4)$, pH 7.4, 7% SDS, 1% BSA, .65% PEG and hybridized at 60°C overnight. Filters were washed twice, 15 min each, in prewarmed (60°C) 2× SSC and once in 2× SSC, .1% SDS for 30 min at 60°C. If needed, filters were rewashed in 1× SSC, .1% SDS for 15 min at 60°C.

Description of Polymorphism. Two codominantly inherited fragments were detected using *Bam*HI with sizes of 9.0 and 5.0 kb plus one monomorphic band at approximately 15 kb (Figure 1). Fragment sizes for *Eco*RI and *Taq*I were 6.0 and 4.0 kb, and 1.9, 3.5, 4.0, and 5.0 kb, respectively.

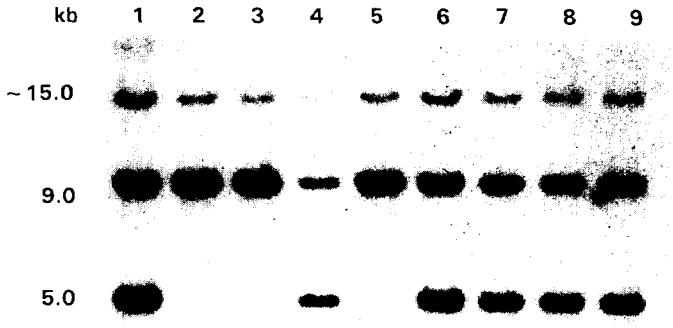

Figure 1. Restriction fragment patterns of nine cattle after genomic DNA was digested with *Bam*HI and hybridized to the bovine calpastatin probe. Fragment sizes in kilobases are given at left. Animal 1 is the dam and Animal 2 is the sire of animals represented in Lanes 3 to 9.

Inheritance Pattern. Codominant inheritance of the *Bam*HI fragments was established in 10 paternal half-sib families, including five heterozygous dams, two homozygous sires, and 63 progeny.

Frequency. Frequency of the *Bam*HI alleles were estimated using 40 unrelated cattle representing Hereford, Angus, Piedmontese, Simmental, Gelbvieh, Red Poll, Braunvieh, Limousin, Pinzgaur, Brahman, and Charolais breeds. The frequencies of the 9.0- and 5.0-kb alleles were .38 and .62, respectively.

Chromosomal Location. By linkage analysis to bovine synteny group U22 located on Chromosome 7.

Probe Availability. Mohammad Koohmaraie, U.S. Meat Animal Research Center, P.O. Box 166, Clay Center, NE 68933.

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