Rapid Communication: A Restriction Fragment Length Polymorphism in the Ovine *Prolactin* Gene[1]

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Polymorphism. A *Hae*III PCR-RFLP was identified in the ovine *prolactin* (*PRL*) gene.

Source and Description of Primers. Human genomic (Truong et al., 1984) and pig cDNA (GenBank accession no. X14068) sequences were compared to design primers to span the second intron of the prolactin gene.

Primer Sequences. Forward primer: 5′-ACCTCTCTTCGGAAATGTTCA-3′; reverse primer: 5′-CTGTTGGGCTTGCTCTTTGTC-3′.

Method of Detection. PCR amplification (25 μL final volume) was performed using 25 ng of genomic DNA, 350 μM each dNTP, .3 μM each primer, 1.1 mM $MgCl_2$, 1 × *Taq* Extender Buffer, 1 unit *Taq* Extender, and 1 unit *Taq* polymerase. The thermal cycler profile was 92°C for 2 min; 35 cycles of 92°C for 45 s, 56°C for 45 s, and 72°C for 3 min, followed by a final extension at 72°C for 7 min.

Description of Polymorphism. Twenty microliters of the 2.5-kb product was digested with *Hae*III to produce bands of approximately 1,400, 530, 360, and 150 bp for the A allele and bands of approximately 1,400, 510, 360, and 150 bp for the B allele (Figure 1).

Inheritance Pattern. The PRL *Hae*III polymorphism was observed to have a Mendelian inheritance pattern in nine two-generation sheep families of the AgResearch International Mapping Flock (Crawford et al., 1995).

Frequency. Frequencies for the A allele were .30 for crossbred whitefaced (n = 10), .25 for Suffolk (n = 6), .375 for Coopworth (n = 4), and .50 for Texel (n = 1).

Chromosomal Location. Unknown.

Comments. The ends of the sheep PCR product were sequenced to confirm that the product was PRL. The coding portion was 100% homologous to the corresponding region of the ovine PRL cDNA (GenBank accession no. M27057). Two Suffolk animals had a differing restriction pattern with *Hae*III, but this was not observed in any other animals. A polymorphism was also identified with the restriction enzyme *Mse*I. This second polymorphism was observed to be in complete phase with the *Hae*III polymorphism in the families typed. Prolactin is an anterior pituitary hormone involved in many reproductive pathways.

Literature Cited

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Key Words: Sheep, PCR-RLFP, Prolactin Gene

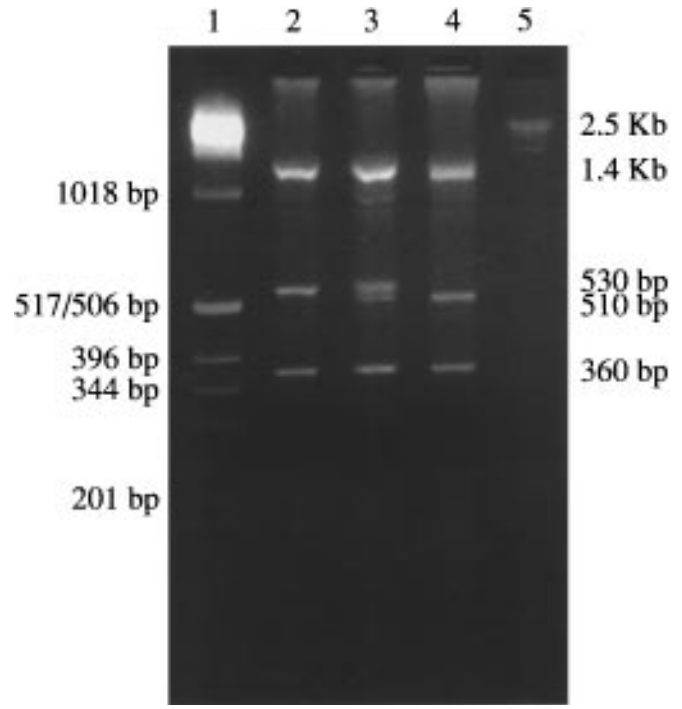

Figure 1. *Hae*III RFLP in the ovine prolactin PCR fragments separated on a 3% NuSieve gel (FMC). Lane 1: 1-kb ladder (Promega); lane 2: AA genotype; lane 3: AB genotype; lane 4: BB genotype; lane 5: uncut PCR product.

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