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Association of prolactin polymorphism with milk fat content in Iranian Sarabi cows

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Introduction Prolactin (PRL) is a polypeptide hormones produced by cells of the anterior pituitary. The bovine PRL gene consists of 4 introns and 5 exons, located on chromosome 23. Different biological functions of PRL were subdivided into five broad categories: reproduction, osmoregulation, growth, integument, and synergism with steroids. Also, It has been shown to be important for control of mammary growth, lactogenesis and lactation (Skinkyte *et al.,* 2005). Chung et al, (1996) showed that PRL-*Rsa*I locus had a significant effect on milk fat percent in dairy cattle. Therefore the PRL gene was chosen as a candidate gene for milk traits in dairy cattle. The objective of this study was to evaluate the association of genetic differences in bovine PRL gene and milk fat content in Iranian Sarabi cows.

Materials and methods Genomic DNA samples were obtained from 95 Sarabi cattle. DNA was extracted from whole blood by guanidinium thiocyanate-silica gel method (Boom *et al*., 1990). In a volume of 20 µl which consisted of 50 ng of template DNA, PCR reaction contained: 2.5 µl PCR buffer 10-X (200 mM $(NH_4)_2SO_4$, 0.1 mM Tween 20, 750 mM Tris-HCl, pH=8.8), 2.5 mM $MgCl_2$, 200 µM dNTPs, and 10 pM of oligonucleotids, 1 u *Taq* DNA polymerase. Thermal condition consisted with thirty-five cycles of 95°C (1 min), 62°C (1 min), and 72°C (2 min) followed by 72°C (8 min). Exon 4 of the bovine PRL gene was amplified to produce a 156 bp fragment using the primers described by Skinkyte *et al.* (2005). PCR products (4 µl) of each PCR reaction were incubated for 5 h at 37 °C with 4 µl *Rsa* I enzyme. Digestion products were separated by electrophoresis on 3% agarose gel and visualized after staining with etithium bromide. The frequencies of genotypes, alleles and Hardy-Weinberg equilibrium test were calculated using PopGene32 (ver. 1.31) program. Data from previous lactations on milk fat content were analyzed by analyzing Standard Least Square within mixed models using JMP software (version 5.1; SAS Institute Inc, NC. USA). Animal was fitted as a random effect. Statistical significance was declared at $P < 0.05$.

Results A 156 bp fragment of the bovine PRL gene from the exon 3 was amplified successfully. Digestion with *Rsa* I differentiated alleles A and B. The digested AA genotype revealed a fragment of 156, AB genotype exhibited three fragments of 156, 82 and 74 and BB genotype had two fragments of 82 and 74 bp. The frequencies of A and B alleles were 0.73 and 0.27. The $\chi 2$ test confirmed the Hardy–Weinberg equilibrium in this population. The milk fat percent was similar among the genotypes ($P = 0.17$) but tended to be lower in the BB cows compare with the AA and AB cows (Figure 1).

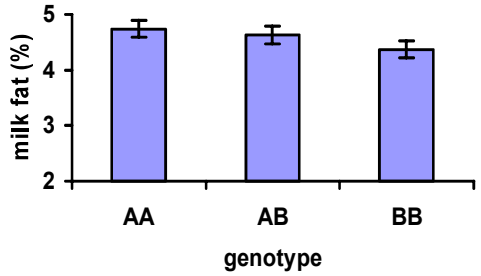

Figure 1 Effect of the prolactin genotypes on milk fat percent using previous lactation records in Sarabi cows.

Conclusions The results of this study demonstrate that the prolactin polymorphism in the exon 3 had no effect on milk fat content in Sarabi cows and hence the polymorphism cannot be a suitable marker in selection programs for improving milk fat content in Sarabi cows.

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