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The Pony Express



Genetic analysis of the Lac La Croix Pony

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I have examined genetic variation in the Lac La Croix Indian Pony (LLCIP) from Canada using both blood typing genetic markers and equine DNA microsatellite markers. A total of 43 ponies were used for the analysis. Seventeen blood typing marker systems were analyzed. Seven systems were red blood cell alloantigen loci (the *A*, *C*, *D*, *K*, *P*, *Q* and *U* horse blood groups) tested by standard serological methods of agglutination and complement mediated hemolysis. The other 10 systems were biochemical polymorphisms detected by electrophoretic techniques. These systems were Albumin (*ALB*), Alpha-1-beta Glycoprotein (*A1B*), Serum Cholinesterase (*ES*), Vitamin D Binding Protein (*GC*), Glucose Phosphate Isomerase (*GPI*) Alpha Hemoglobin (*HB*), Phosphoglucomutase (*PGM*), Phosphogluconate Dehydrogenase (*PGD*), Protease Inhibitor (*PI*), and Transferrin (*TRF*). In addition to the above genetic systems, DNA was extracted from the blood samples and tested for variation at 12 microsatellite (mSat) systems. These were (*AHT4*, *AHT5*, *ASB2*, *ASB17*, *ASB23*, *HMS3*, *HMS6*, *HMS7*, *HTG4*, *HTG10*, *LEX33*, and *VHL20*). These systems were tested using an automated DNA sequencer to separate

Polymerase Chain Reaction (PCR) products.

A variety of genetic variability measures were calculated from the gene marker data. The measures were observed heterozygosity (*Ho*) which is the actual number of loci heterozygous per individual and is based upon biochemical loci only; expected heterozygosity (*He*) which is the predicted number of heterozygous loci based upon gene frequencies; effective number of alleles (*Ae*) which is a measure of marker system diversity; total number of variants (*TNV*); and estimated inbreeding level (*Fis*) which is calculated as $1 - Ho/He$. These same measures were calculated for the mSat data.

Table 1 gives the genetic variability measures based upon blood typing data for the LLC as well as for a number of domestic breeds. The breeds were selected to demonstrate the range of variation in horses. The individual variation (*Ho*) of the LLC was relatively high at 0.384. This is greater than the mean value for horses. However, populational variation as shown by *He* was lower than average for horses. There are probably two components to this result. First, there is a statistically significant excess of *Ho* compared to *He* at the *ALB*

system. If the expected level of *Ho* for *ALB* was used in calculation of overall *Ho* instead of what was actually observed, overall *Ho* would have been 0.366. It is not possible to determine the reason for this difference although such excesses of variation at the *ALB* system have been seen for a number of horse populations. The second component to the difference in overall *Ho* and *He* could be due to the reduction in size of the LLC population in recent times. This is known as a population bottleneck and in the first few generations after a bottleneck *Ho* will usually exceed *He*. This is usually an indication of a loss of overall genetic diversity in the population. The level of genetic diversity as measured by *Ae* is fairly low.

The DNA variation (Table 2) was relatively lower when compared to the average values for horses. *Ho* again was higher than *He* for the LLCIP but not by as great of a margin as with the blood typing results. The level of variation at the DNA marker is still well above that seen for a number of breeds, including some with large population sizes (such as the Arabian). Variation in DNA markers for the LLCIP is high enough that for parentage analysis the probability of de-