

## Analysis of Genetic Diversity in the Puerto Rico Paso Fino

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The purpose of this study was to analyze genetic variability in the Puerto Rican Paso Fino in order to determine its conservation status and to understand its place in the history of the horse in the New World.

To start out I will give a little background. First, what is genetic diversity? Genetic diversity primarily refers to variation within the genetic make up of an individual. It also is variation in genetic make up among individuals within a population, genetic variation among different populations of a species, and genetic differences among species. For this work I am focusing on individual and populational variation.

Why is Genetic Diversity Important? It gives individuals flexibility to respond to different environmental conditions, it increased disease resistance, it gives populations the ability to adapt to environmental changes over time (for domestic animals this also can mean the ability to make selective improvements to a breed), and it buffers against deleterious recessive genes.

What factors act to change genetic diversity? The number one factor is population size, the smaller the size the lower the diversity. Also, inbreeding reduces individual variation. Out crossing increases diversity when different groups are crossed. Selection, which will usually decrease variation, is another factor as is mutation which increases variation but is unimportant over short time scales.

How do we measure diversity? Individual diversity is measured as Heterozygosity (Observed  $H_o$ ) which is the proportion of the genome that varies within an individual. The Inbreeding coefficient also is a measure of basically the same thing but on a theoretical level. Both these measures are independent of sample size. For estimating genetic diversity of a whole population or breed we again use Heterozygosity but it is Expected Heterozygosity ( $H_e$ ) which is the average proportion of the genome that varies based upon population genetic theory. Another important measure is simply the number of variants observed in the population. These measures are correlated with sample size. We determine these different measures by typing of genetic markers. A gene marker is a variable spot in the genome that we can test. The different forms the marker can take are called alleles. Each marker can have two possible alleles in an individual but many across the population. For an individual, if the alleles are the same, the marker is homozygous. If different, the marker is heterozygous. Heterozygosity is the proportion of markers out of all tested that are heterozygous. This can be calculated as the average of those actually observed as heterozygous ( $H_o$ ) or by the predicted proportion based upon theory calculated from the frequency of each allele in the population ( $H_e$ ).

Measures of genetic diversity for selected breeds.

| Breed                    | <i>N</i> | <i>Ho</i> | <i>He</i> | <i>Ae</i> |
|--------------------------|----------|-----------|-----------|-----------|
| ANDALUSIAN               | 52       | 0.722     | 0.753     | 4.26      |
| ARABIAN                  | 47       | 0.660     | 0.727     | 3.81      |
| CLEVELAND BAY            | 504      | 0.566     | 0.608     | 2.85      |
| FRIESIAN                 | 304      | 0.545     | 0.539     | 2.56      |
| IRISH DROUGHT            | 135      | 0.802     | 0.799     | 5.19      |
| LIPPIZANNER              | 37       | 0.657     | 0.711     | 3.58      |
| PUERTO RICAN PASO FINO 1 | 95       | 0.640     | 0.742     | 4.02      |
| PUERTO RICAN PASO FINO 2 | 17       | 0.657     | 0.659     | 3.06      |
| THOROUGHBRED             | 1195     | 0.734     | 0.726     | 3.92      |
| Domestic Horse Average   | 75       | 0.706     | 0.718     | 3.99      |

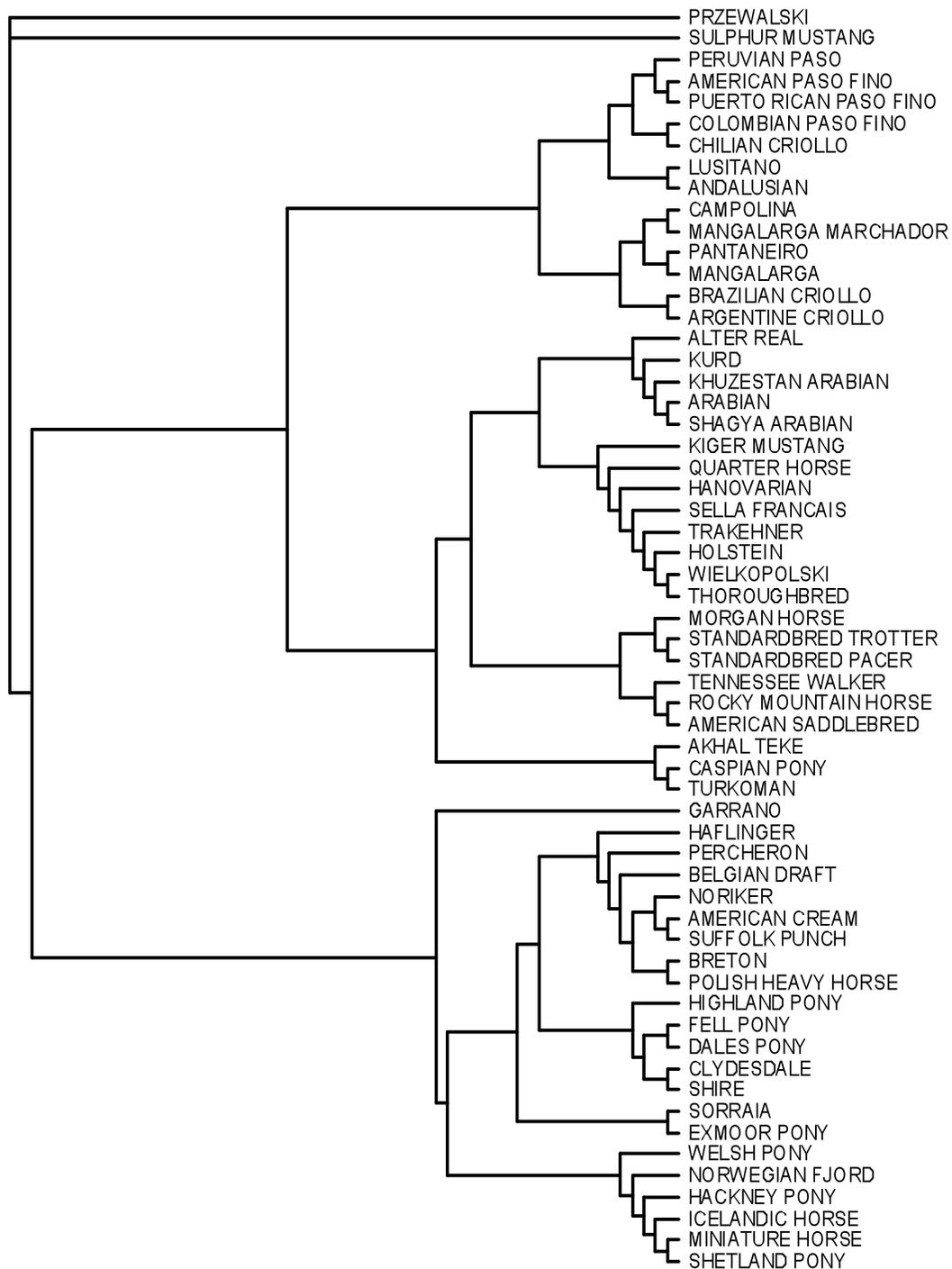
1 Horses sampled between 1992 and 2004, includes US animals.

2 Horses received in March of 2007, all from Puerto Rico.

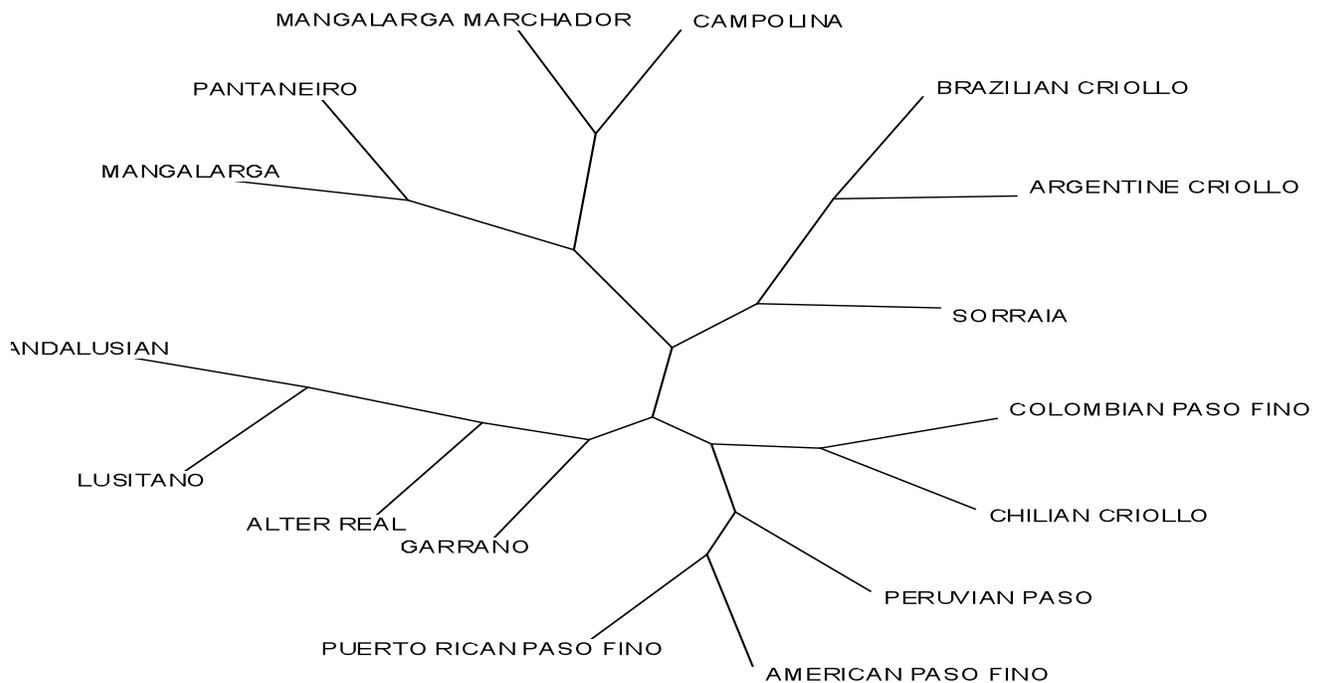
The results of the diversity analysis of the breed show that genetic diversity in the Puerto Rican Paso is slightly below the domestic horse mean but not seriously so. Allelic diversity is uninformative at this time due to sample size. Earlier testing done with horses in the US appears to have sampled across populations with some differences (either other types of Paso or there are differences between the US mainland and Puerto Rican gene pools). Although current levels of genetic variation are not low, the population should be monitored due to the relatively low population size.

What can the Puerto Rican Paso Fino tell us about horses in the Americas? *Equus caballus* (the species of the domestic horse) originated in North America but was extinct in the Americas by about 10,000 years ago. Domestication of the horse occurred in Central Asia about 6,000 years ago and there also is evidence for a domestication center in the Iberian peninsula. The horse first returned to the Americas with the second voyage of Christopher Columbus in 1493. The type of horse brought to the New World by the Spanish was a type known as the Jennet. This type of horse no longer exists in the Iberian Peninsula.

Comparison of allele frequencies can give measures of genetic resemblance. These are not true measures of ancestry but are approximations. Genetic similarity measure can be summarized by tree diagrams that give two dimensional representations of estimated relationships. The Puerto Rico Paso Fino clearly falls within the group of horses that are of Iberian ancestry, grouping with other Paso breeds. When only the Iberian breeds are analyzed the position is similar. However, closer analysis (not shown here) also shows the Puerto Rico Paso Fino has closer affiliations with the Old World breeds than do other New World breeds and has relative strong resemblance to the Sorraia, a Portuguese breed that is believed to be much like the old Jennet horse.



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One final note regarding the study of the genetics of the Puerto Rican Paso Fino is that this breed has been important in studies of the ancestry of American Mustangs. Because it is the North American breed that most resembles the horses that were brought to the Americas by the early Spanish, the presence of specific variants that are present in the Puerto Rico Paso Fino and some mustang herds but not in most North American breeds indicates that they likely have old Spanish horse ancestry.

My conclusions at this point are these. First, genetic diversity in the Puerto Rican Paso Fino is good but shows evidence that it is reduced so should be monitored. Also, the

Puerto Rico Paso Fino appears to be clearly related to Old World Iberian horses and may be one of the closest representatives left of the type of horse the Spanish brought to the Americas 500 years ago.