



Genetic Analysis of the
Piceance-East Douglas HMA, CO

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June 1, 2010

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*This report specific to wild horses
gathered off of the East Douglas
portion of PEDHMA.*

The following is a report of the genetic analysis of the Piceance-East Douglas HMA, CO.

A few general comments about the genetic variability analysis based upon DNA microsatellites compared to blood typing. The DNA systems are more variable than blood typing systems, thus variation levels will be higher. Variation at microsatellite loci is strongly influenced by allelic diversity and changes in variation will be seen in allelic measures more quickly than at heterozygosity, which is why more allelic diversity measures are calculated. For mean values, there are a greater proportion of rare domestic breeds included in the estimates than for blood typing so relative values for the measures are lower compared to the feral horse values. As well, feral values are relatively higher because the majority of herds tested are of mixed ancestry which results in a relatively greater increase in heterozygosity values based upon the microsatellite data. There are no specific variants related to breed type so similarity is based upon the total data set.

METHODS

A total of 32 samples were received by Texas A&M University, Equine Genetics Lab on October 6, 2006. DNA was extracted from the samples and tested for variation at 12 equine 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; 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*); mean number of alleles per locus (*MNA*); the

number of rare alleles observed which are alleles that occur with a frequency of 0.05 or less (RA); the percent of rare alleles ($\%RA$); and estimated inbreeding level (Fis) which is calculated as $1-Ho/He$.

Genetic markers also can provide information about ancestry in some cases. Genetic resemblance to domestic horse breeds was calculated using Rogers' genetic similarity coefficient, S . This resemblance was summarized by use of a restricted maximum likelihood (RML) procedure.

RESULTS AND DISCUSSION

Variants present and allele frequencies are given in Table 1. No variants were observed which have not been seen in horse breeds. Table 2 gives the values for the genetic variability measures of the Piceance-East Douglas horse herd. Also shown in Table 2 are values from a representative group of domestic horse breeds. The breeds were selected to cover the range of variability measures in domestic horse populations. Mean values for feral herds (based upon data from 126 herds) and mean values for domestic breeds (based upon 80 domestic horse populations) also are shown.

Mean genetic similarity of the Piceance-East Douglas herd to domestic horse breed types are shown in Table 3. A dendrogram of relationship of the Piceance-East Douglas herd to a standard set of domestic breeds is shown in Figure 1.

Genetic Variants: A total of 56 variants were seen in the Piceance-East Douglas herd which is below the mean for feral herds and well below the mean for domestic breeds. Of these, 8 had frequencies below 0.05. This percentage of variants at risk of future loss is below average. Allelic diversity as represented by Ae also is below the average for feral herds as is MNA .

Genetic Variation: Genetic variation, as indicated by heterozygosity, in the Piceance-East Douglas herd is well below the feral mean. *He* is slightly higher than *Ho*. The higher *He* indicates that inbreeding could become an important factor for this herd.

Genetic Similarity: Overall similarity of the Piceance-East Douglas herd to domestic breeds was about average for feral herds. Highest mean genetic similarity of the Piceance-East Douglas herd was with North American Gaited breeds, followed very closely by the Light Racing and Riding breeds. As seen in Fig. 1, the Piceance-East Douglas herd fits most closely to the Fell pony and is near to the Dales pony and Shire, which probably does not reflect true ancestry but is more due to no close similarity with any particular breed or breed group. As with most trees involving feral herds, the tree is somewhat distorted.

SUMMARY

Genetic variability of this herd is somewhat low but not yet at a level to cause concern. The values related to allelic diversity are below the average as is heterozygosity. Genetic similarity results suggest a herd with mixed ancestry that primarily is North American. There is a possibility that this herd has seen a recent loss of population size and genetic diversity.

RECOMMENDATIONS

Current variability levels warrant monitoring of this population. Although variation levels are not yet at a critical level the risk of additional loss of diversity exists in due to small population size. If population size cannot be increased an introduction of horses from another area may need to be considered.

Table 1. Allele frequencies of genetic variants observed in the Piceance-East Douglas HMA feral horse herd.

VHL20															
I	J	K	L	M	N	O	P	Q	R	S					
0.234	0.000	0.000	0.000	0.250	0.234	0.172	0.000	0.047	0.063	0.000					
HTG4															
i	J	K	L	M	N	O	P	Q	R						
0.000	0.000	0.281	0.141	0.516	0.000	0.063	0.000	0.000	0.000						
AHT4															
H	I	J	K	L	M	N	O	P	Q	R					
0.063	0.219	0.344	0.000	0.000	0.000	0.000	0.375	0.000	0.000	0.000					
HMS7															
I	J	K	L	M	N	O	P	Q	R						
0.000	0.250	0.000	0.453	0.000	0.219	0.078	0.000	0.000	0.000						
AHT5															
I	J	K	L	M	N	O	P	Q	R						
0.000	0.672	0.000	0.031	0.203	0.094	0.000	0.000	0.000	0.000						
HMS6															
I	J	K	L	M	N	O	P	Q	R						
0.000	0.000	0.000	0.031	0.594	0.000	0.250	0.125	0.000	0.000						
ASB2															
B	I	J	K	L	M	N	O	P	Q	R					
0.000	0.000	0.000	0.141	0.000	0.000	0.328	0.422	0.000	0.109	0.000					
HTG10															
H	I	J	K	L	M	N	O	P	Q	R	S	T			
0.000	0.000	0.000	0.266	0.109	0.359	0.000	0.109	0.000	0.109	0.047	0.000	0.000			
HMS3															
H	I	J	K	L	M	N	O	P	Q	R	S				
0.000	0.047	0.000	0.000	0.000	0.000	0.063	0.000	0.875	0.016	0.000	0.000				
ASB17															
D	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T
0.000	0.406	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.094	0.000	0.078	0.000	0.297	0.000	0.125
ASB23															
G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	V
0.000	0.000	0.000	0.094	0.141	0.484	0.000	0.000	0.000	0.000	0.000	0.000	0.094	0.000	0.188	0.000
LEX33															
F	G	K	L	M	N	O	P	Q	R	S	T				
0.000	0.047	0.266	0.156	0.000	0.000	0.109	0.000	0.391	0.031	0.000	0.000				

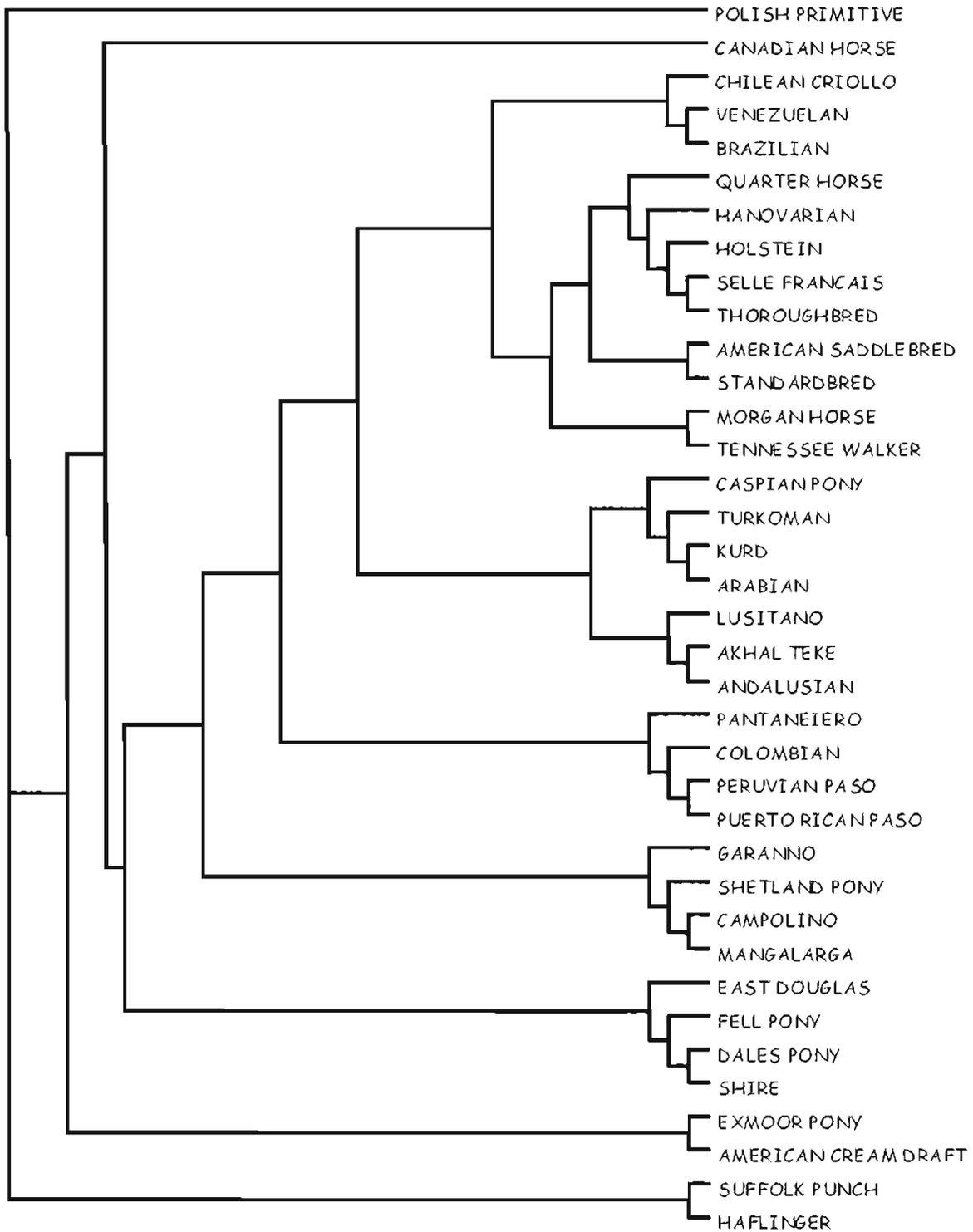
Table 2. Genetic variability measures.

	<i>N</i>	<i>H_o</i>	<i>H_e</i>	<i>F_{is}</i>	<i>A_e</i>	<i>T_{NV}</i>	<i>M_{NA}</i>	<i>R_a</i>	<i>%R_a</i>
EAST DOUGLAS CO	32	0.635	0.640	0.007	3.116	56	4.67	8	0.143
Cleveland Bay	47	0.610	0.627	0.027	2.93	59	4.92	16	0.271
American Saddlebred	576	0.740	0.745	0.007	4.25	102	8.50	42	0.412
Andalusian	52	0.722	0.753	0.041	4.26	79	6.58	21	0.266
Arabian	47	0.660	0.727	0.092	3.81	86	7.17	30	0.349
Exmoor Pony	98	0.535	0.627	0.146	2.87	66	5.50	21	0.318
Friesian	304	0.545	0.539	-0.011	2.56	70	5.83	28	0.400
Irish Draught	135	0.802	0.799	-0.003	5.19	102	8.50	28	0.275
Morgan Horse	64	0.715	0.746	0.041	4.19	92	7.67	33	0.359
Suffolk Punch	57	0.683	0.711	0.038	3.88	71	5.92	13	0.183
Tennessee Walker	60	0.666	0.693	0.038	3.66	87	7.25	34	0.391
Thoroughbred	1195	0.734	0.726	-0.011	3.92	69	5.75	18	0.261
Feral Horse Mean	126	0.716	0.710	-0.012	3.87	72.68	6.06	16.96	0.222
Standard Deviation		0.056	0.059	0.071	0.66	13.02	1.09	7.98	0.088
Minimum		0.496	0.489	-0.284	2.15	37	3.08	0	0
Maximum		0.815	0.798	0.133	5.25	96	8.00	33	0.400
Domestic Horse Mean	80	0.710	0.720	0.012	4.01	80.88	6.74	23.79	0.283
Standard Deviation		0.078	0.071	0.086	0.74	16.79	1.40	10.11	0.082
Minimum		0.347	0.394	-0.312	1.78	26	2.17	0	0
Maximum		0.822	0.799	0.211	5.30	119	9.92	55	0.462

Table 3. Rogers' genetic similarity of the Piceance-East Douglas HMA feral horse herd to major groups of domestic horses.

	Mean <i>S</i>	Std	Minimum	Maximum
Light Racing and Riding Breeds	0.699	0.020	0.669	0.723
Oriental and Arabian Breeds	0.685	0.036	0.650	0.731
Old World Iberian Breeds	0.673	0.023	0.653	0.699
New World Iberian Breeds	0.662	0.021	0.636	0.691
North American Gaited Breeds	0.701	0.016	0.671	0.713
Heavy Draft Breeds	0.657	0.048	0.579	0.710
True Pony Breeds	0.657	0.022	0.623	0.684

Figure 1. Partial RML tree of genetic similarity to domestic horse breeds.



Appendix 1. DNA data for the Piceance-East Douglas HMA, CO herd.

AID	VHL20	HTG4	AHT4	HMS7	AHT5	HMS6	ASB2	HTG10	HMS3	ASB17	ASB23	LEX33	LEX3
13086	IN	LM	HO	LO	MM	MM	NQ	KL	NP	RT	LS	GK	FL
13087	NO	LM	JO	LL	JJ	MM	KO	MQ	PP	FR	LL	KQ	FF
13088	MN	KM	IJ	JN	JM	MP	OO	KQ	PP	FR	LL	KL	FF
13089	NO	MM	JO	LN	JM	MP	KO	MQ	PP	RT	JL	KK	FF
13090	IM	LM	HO	LO	JJ	MM	NQ	LM	NP	NT	JK	OQ	FM
13091	OO	MM	IJ	JN	JM	OP	KN	OQ	PP	FT	JL	KQ	LL
13092	MO	KK	IJ	JL	JJ	MO	NQ	KM	IP	FF	LU	LQ	FF
13093	IO	MM	IJ	JL	JJ	OO	NN	KO	PP	FF	LU	OQ	FL
13094	MM	KK	IJ	JL	JJ	MM	NO	MQ	PP	FR	LL	LQ	FF
13095	MO	KL	IO	JL	JJ	LM	KO	KQ	PP	FR	LU	QQ	FF
13096	IN	MM	JJ	JL	JJ	MO	OO	MO	PP	FR	JL	KO	FL
13097	IM	KL	JO	JL	JJ	MO	OO	MM	PP	FR	LU	KQ	FF
13098	RR	MO	OO	NN	LN	MP	NQ	MR	PP	PP	KS	LQ	FF
13099	MO	KL	II	JL	JJ	LM	KN	KK	PP	FF	LU	QQ	FF
13100	MN	KM	IO	JL	JM	MM	OO	KM	PP	FR	JU	KQ	FF
13101	IM	KM	JJ	JL	JJ	OO	OO	KO	IP	FF	LU	LO	LL
13102	NR	MO	OO	NN	LN	MP	NN	MR	PP	NP	KK	LR	FF
13103	IM	KM	IJ	JL	JJ	MO	OO	KO	PP	FF	LU	LQ	FF
13104	MN	KM	JO	LN	JM	MM	OO	KM	PP	RT	LL	KL	FF
13105	IM	KM	IO	JL	JJ	MO	NO	KO	PP	FR	UU	QQ	FF
13106	NN	MO	OO	NN	NN	MM	NN	MM	PP	NP	KK	QR	FH
13107	IQ	LM	HO	OO	JM	MM	NN	LL	NP	NR	KS	GO	HM
13108	IN	MM	JJ	LL	JJ	OP	KO	MM	PP	RT	LL	KQ	FF
13109	IO	LM	JO	JL	JJ	MO	KO	MM	PP	FR	LL	KQ	FF
13110	MQ	KM	JO	LN	MM	MM	NQ	KL	PP	RT	LS	KQ	MM
13111	OO	MM	JO	LL	JM	OP	KO	MM	PP	RR	LL	KK	FF
13112	IQ	LM	HO	NO	JM	MM	NN	LL	NP	NR	KS	GQ	FL
13113	MN	KM	JO	LL	JM	MM	KQ	KM	PP	FT	JL	KQ	FF
13114	NN	KM	IO	JL	JJ	MP	OO	KQ	PP	FR	LL	KQ	FF
13115	NR	MO	OO	NN	NN	MM	NQ	MR	PQ	NP	KS	LQ	FH
13116	II	KM	IJ	JL	JJ	OO	NO	KO	IP	FF	LU	OQ	FL
13117	IM	KM	IJ	LL	JJ	MO	OO	KM	PP	FF	LU	LO	FF