

Appendix G

HMA Genetic Reports

Genetic Analysis of the
Adobe Town, WY HMA 2017

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The following is a report of the genetic analysis of the Adobe Town, WY HMA.

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 40 samples were received by Texas A&M University, Animal Genetics Lab on November 14, 2017. 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 (*H_o*) which is the actual number of loci heterozygous per individual; expected heterozygosity (*H_e*), which is the predicted number of heterozygous loci based upon gene frequencies; effective number of alleles (*A_e*) which is a measure of marker system diversity; total number of variants (*TWY*); 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 Adobe Town 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 Adobe Town herd to domestic horse breed types are shown in Table 3. A dendrogram of relationship of the Adobe Town herd to a standard set of domestic breeds is shown in Figure 1.

Genetic Variants: A total of 90 variants were seen in the Adobe Town herd which is much higher than the average for domestic breeds and for feral horses. Of these, 25 had frequencies below 0.05. This percentage of variants at risk of future loss is above the average for feral horse populations and slightly above that for domestic breeds. Allelic diversity as represented by Ae is high for feral herds as is MNA .

Genetic Variation: Genetic variation, as indicated by heterozygosity, in the Adobe Town herd is well above the feral mean. H_o is very slightly higher than H_e but the difference is compatible with genetic equilibrium.

Genetic Similarity: Overall similarity of the Adobe Town herd to domestic breeds was somewhat high for feral herds. Highest mean genetic similarity of the Adobe Town herd was with Light Racing and Riding breeds, followed very closely by Old World Spanish breeds and by the North American breeds. As seen in Fig. 1, the Adobe Town herd fits most closely to the Morgan Horse but the tree is somewhat distorted and there is evidence for some Spanish relationship shown in the tree as well. The combined evidence suggests a mixed origin and the variability measures are consistent with this.

The Adobe Town herd was previously analyzed in 2003 (using blood typing methods) and in 2012 using DNA based analysis. Variability levels were relatively high in both previous sampling periods and the values of variability measures were quite similar. Sample size was small in 2003 and the blood typing variation values are not directly comparable but they do reflect very similar levels. Both previous data sets suggest mixed ancestry, primarily from North American derived breeds with some Spanish component.

SUMMARY

Genetic variability of this herd is high. The values related to allelic diversity in particular suggest a herd with highly mixed ancestry. This view is consistent with the similarity values seen and the heterozygosity measures. The herd ancestry likely includes some Spanish component based upon this data and the data from 2003 and 2012.

RECOMMENDATIONS

Current variability levels are good and this herd has shown no obvious change in diversity levels since 2012. Re-sampling of the herd should be considered by 2022 to check for changes in variation. However, unless there is some serious change in population size resampling could be delayed for two to three additional years.

Table 1. Allele frequencies of genetic variants observed in Adobe Town HMA 2017 feral horse herd.

VHL20																	
I	J	K	L	M	N	O	P	Q	R	S							
0.336	0.013	0.125	0.063	0.174	0.063	0.050	0.100	0.063	0.013	0.000							
HTG4																	
I	J	K	L	M	N	O	P	Q	R								
0.000	0.000	0.188	0.087	0.563	0.025	0.025	0.112	0.000	0.000								
AHT4																	
H	I	J	K	L	M	N	O	P	Q	R							
0.325	0.137	0.112	0.063	0.013	0.112	0.013	0.225	0.000	0.000	0.000							
HMS7																	
I	J	K	L	M	N	O	P	Q	R								
0.000	0.150	0.000	0.187	0.200	0.200	0.175	0.025	0.063	0.000								
AHT5																	
I	J	K	L	M	N	O	P	Q	R								
0.000	0.387	0.125	0.075	0.013	0.250	0.150	0.000	0.000	0.000								
HMS6																	
I	J	K	L	M	N	O	P	Q	R								
0.000	0.000	0.000	0.112	0.125	0.075	0.150	0.538	0.000	0.000								
ASB2																	
B	I	J	K	L	M	N	O	P	Q	R							
0.000	0.050	0.000	0.250	0.000	0.175	0.163	0.075	0.000	0.262	0.025							
HTG10																	
H	I	J	K	L	M	N	O	P	Q	R	S	T					
0.000	0.063	0.000	0.013	0.200	0.125	0.336	0.062	0.013	0.038	0.150	0.000	0.000					
HMS3																	
H	I	J	K	L	M	N	O	P	Q	R	S						
0.000	0.112	0.000	0.000	0.000	0.162	0.025	0.275	0.213	0.038	0.175	0.000						
ASB17																	
D	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T		
0.000	0.100	0.000	0.000	0.013	0.000	0.050	0.000	0.187	0.212	0.062	0.175	0.013	0.175	0.013	0.000		
ASB2																	
G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	V		
0.038	0.000	0.125	0.250	0.100	0.100	0.000	0.000	0.000	0.000	0.000	0.000	0.237	0.000	0.150	0.000		
LEX33																	
F	G	K	L	M	N	O	P	Q	R	S	T						
0.000	0.013	0.050	0.475	0.050	0.000	0.125	0.000	0.175	0.087	0.025	0.000						

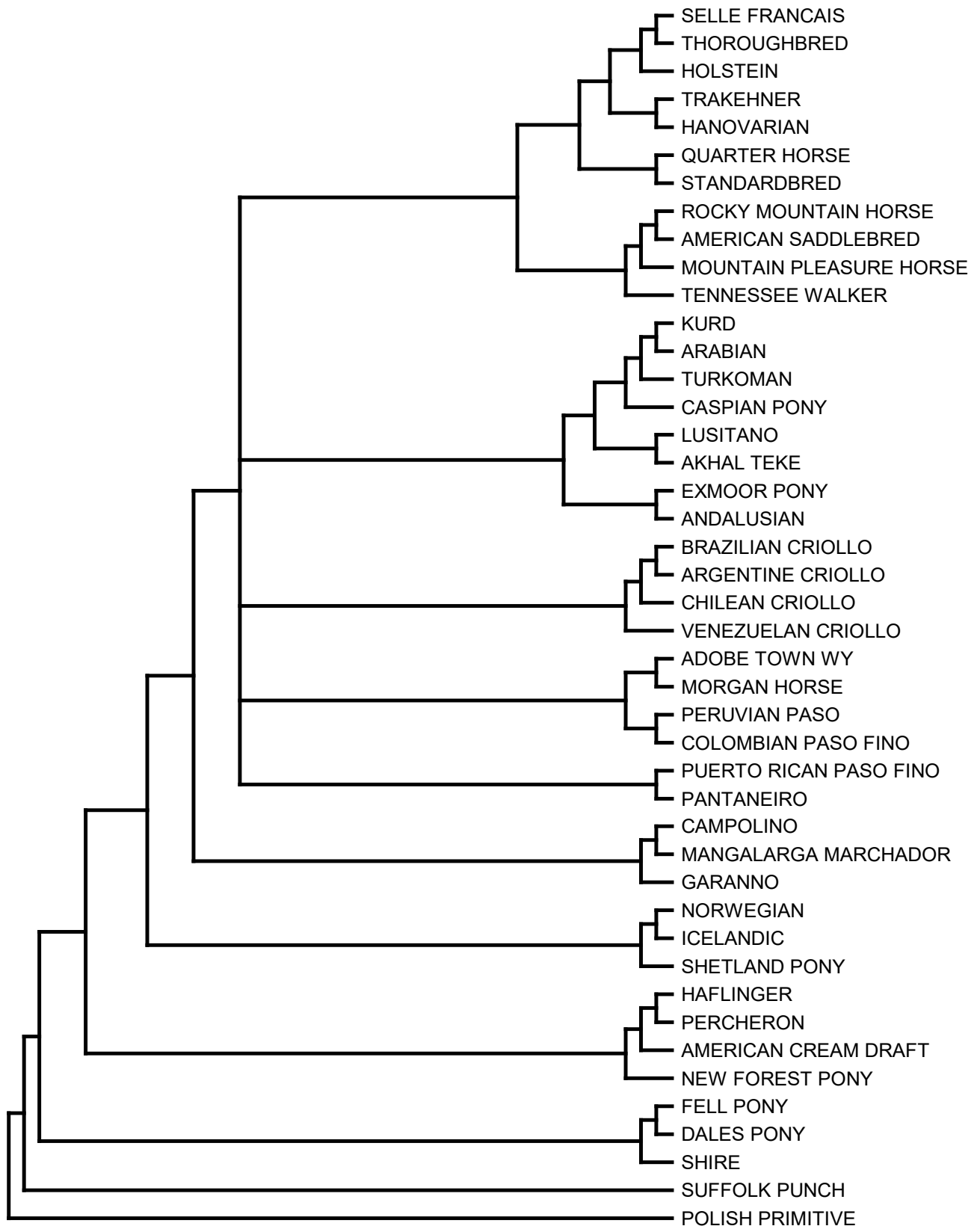
Table 2. Genetic variability measures.

	<i>N</i>	<i>Ho</i>	<i>He</i>	<i>Fis</i>	<i>Ae</i>	<i>TNV</i>	<i>MNA</i>	<i>Ra</i>	<i>%Ra</i>
Adobe Town, WY 2017	40	0.773	0.771	-0.003	4.68	90	7.5	25	0.278
Cleveland Bay	47	0.610	0.627	0.027	2.934	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	4.259	79	6.58	21	0.266
Arabian	47	0.660	0.727	0.092	3.814	86	7.17	30	0.349
Exmoor Pony	98	0.535	0.627	0.146	2.871	66	5.50	21	0.318
Friesian	304	0.545	0.539	-0.011	2.561	70	5.83	28	0.400
Irish Draught	135	0.802	0.799	-0.003	5.194	102	8.50	28	0.275
Morgan Horse	64	0.715	0.746	0.041	4.192	92	7.67	33	0.359
Suffolk Punch	57	0.683	0.711	0.038	3.878	71	5.92	13	0.183
Tennessee Walker	60	0.666	0.693	0.038	3.662	87	7.25	34	0.391
Thoroughbred	1195	0.734	0.726	-0.011	3.918	69	5.75	18	0.261
Feral Horse Mean	126	0.716	0.710	-0.012	3.866	72.68	6.06	16.96	0.222
Standard Deviation		0.056	0.059	0.071	0.657	13.02	1.09	7.98	0.088
Minimum		0.496	0.489	-0.284	2.148	37	3.08	0	0
Maximum		0.815	0.798	0.133	5.253	96	8.00	33	0.400
Domestic Horse Mean	80	0.710	0.720	0.012	4.012	80.88	6.74	23.79	0.283
Standard Deviation		0.078	0.071	0.086	0.735	16.79	1.40	10.11	0.082
Minimum		0.347	0.394	-0.312	1.779	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 Adobe Town, WY 2017 eral horse herd to major groups of domestic horses.

	Mean <i>S</i>	Std	Minimum	Maximum
Light Racing and Riding Breeds	0.790	0.025	0.755	0.819
Oriental and Arabian Breeds	0.770	0.038	0.732	0.836
Old World Iberian Breeds	0.787	0.026	0.750	0.818
New World Iberian Breeds	0.778	0.032	0.731	0.805
North American Gaited Breeds	0.786	0.025	0.752	0.818
Heavy Draft Breeds	0.713	0.060	0.629	0.810
True Pony Breeds	0.717	0.044	0.654	0.774

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



Appendix 1. DNA data for the Adobe Town, WY 2017 herd.

AID	VHL20	HTG4	AHT4	HMS7	AHT5	HMS6	ASB2	HTG10	HMS3	ASB17	ASB23	LEX33	LEX3
102412	IQ	MO	HM	LM	JK	MN	KM	IM	OP	MM	SU	KL	LO
102413	IM	MM	HO	OQ	JM	MO	KM	IN	IR	MP	GI	LQ	MN
102414	IK	MM	HM	LM	KN	LL	NQ	LN	IQ	MN	JK	OQ	FK
102415	IR	MM	JK	NN	JN	OP	IN	KL	MM	NR	LL	KK	LN
102416	IK	KK	HM	LN	JN	NP	MQ	NO	NO	MP	IS	LR	NP
102417	LP	MM	LM	MN	JJ	LP	KO	LN	IP	NR	JU	LQ	HK
102418	JM	KM	HO	JO	NN	NP	KM	MR	MQ	FM	JS	LL	LO
102419	MP	MM	HO	LO	JN	PP	MM	LN	PR	NP	GU	LL	HO
102420	KM	MM	MO	NQ	JO	MP	OR	LQ	MP	NN	IJ	LR	MM
102421	IM	MM	MO	LL	JJ	MP	OQ	LM	MO	MN	IS	LL	FM
102422	IK	LM	HH	MN	KL	MO	KO	MQ	PP	PS	JU	GL	FH
102423	KL	KM	HO	NO	JJ	OP	QQ	NN	OR	MP	LL	LL	MM
102424	IO	KL	KO	LQ	NO	PP	QQ	NR	PR	KN	SS	OS	LL
102425	KN	MM	KO	JL	JO	OP	MN	NN	MM	PR	UU	LQ	MO
102426	IQ	KP	HH	MN	NO	PP	KO	MN	OR	KN	IS	LR	PP
102427	IP	KO	HO	MO	JK	PP	KN	IQ	MR	MP	IS	LO	LM
102428	MN	MN	IO	OQ	JJ	NP	OQ	LR	MP	PR	JL	OQ	FO
102429	IQ	KM	HO	LL	JO	PP	KM	NN	MR	NR	GK	OS	LL
102430	IM	NP	IN	JO	JN	PP	KQ	NR	OP	NO	JS	LR	MM
102431	LM	LM	IK	MN	LO	PP	KQ	LL	OP	NR	IK	LQ	FL
102432	IM	MP	HK	JN	JK	OP	KM	LM	OO	OR	IK	LQ	FF
102433	IN	MM	HI	LN	JO	PP	NQ	MN	IP	FP	JS	LO	MM
102434	II	KM	HJ	JO	JN	LP	KN	NO	OR	KM	SU	LQ	KK
102435	IP	MP	HM	MN	JJ	PP	MQ	LM	OP	OR	KU	LL	HH
102436	II	MM	HM	NN	JO	PP	NN	MN	OP	NP	JS	LO	PP
102437	NQ	MM	HH	LM	JO	PP	KN	MN	II	FM	JS	LO	HH
102438	KM	KM	IO	MO	KO	LO	NR	LN	OP	MR	JK	LQ	MM
102439	MM	KP	IJ	JN	LN	OO	IQ	OO	MM	RR	IJ	LO	NN
102440	KQ	KL	HI	JL	JK	MP	KN	NR	OR	MN	LS	LL	HH
102441	IN	KL	JO	MO	KN	LL	II	IL	IO	NN	JU	LO	LL
102442	II	LM	HI	PP	JL	MP	KK	LR	OO	PP	JK	LL	LL
102443	KK	KM	IM	NO	JN	MM	KQ	NN	IN	FM	JL	LR	HH
102444	IP	MM	IO	JM	NN	LO	KN	PR	OR	OP	JK	QQ	LL
102445	MP	MP	HO	OO	NO	LP	QQ	RR	OR	OP	JS	LQ	KK
102446	LO	KM	HI	MQ	JN	PP	QQ	IL	MR	IM	JU	QR	NN
102447	LP	MM	JJ	MO	NN	MP	KQ	NR	OP	NQ	SU	LL	OO
102448	MO	MP	OO	LM	JJ	OP	MN	RR	OQ	FF	SS	KM	FF
102449	IO	MM	JJ	JJ	KL	NO	MQ	LN	OR	FF	IJ	LM	PP
102450	II	MP	HO	JJ	KL	NP	MQ	NO	PR	RR	JU	MM	PP
102451	IP	LP	HJ	LM	NO	PP	KM	NN	IP	KR	LS	QR	LL

Genetic Analysis of the
Salt Wells Creek HMA, Manuel Gap subgroup, WY

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April 11, 2011

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The following is a report of the genetic analysis of the Salt Wells Creek HMA, Marvel Gap, WY. The herd will be referred to as the Salt Wells Creek HMA MG in this report.

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 25 samples were received by Texas A&M University, Equine Genetics Lab on November 22, 2010. 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 Salt Wells Creek HMA MG 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 Salt Wells Creek HMA MG herd to domestic horse breed types are shown in Table 3. A dendrogram of relationship of the Salt Wells Creek HMA MG herd to a standard set of domestic breeds is shown in Figure 1.

Genetic Variants: A total of 80 variants were seen in the Salt Wells Creek HMA MG herd which is above the mean for feral herds and just slightly below the mean for domestic breeds. Of these, 15 had frequencies below 0.05 which is a low percentage of variants at risk of future loss. Allelic diversity as represented by Ae is somewhat the average for feral herds while MNA is greater than the mean.

Genetic Variation: Observed heterozygosity in the Salt Wells Creek HMA MG herd is well above the feral mean while He is only slightly higher than average. Ho is a good bit higher

than *He*. Differences such as this can indicate a recent reduction in population size, within the past few generations, but this not possible to confirm by DNA data alone.

Genetic Similarity: Overall similarity of the Salt Wells Creek HMA MG herd to domestic breeds was about average for feral herds. Highest mean genetic similarity of the Salt Wells Creek HMA MG herd was with Light Racing and Riding breeds, followed closely by the Oriental and Arabian breeds. As seen in Fig. 1, however, the Salt Wells Creek HMA MG herd pairs with the Morgan Horse within a group of South American breeds. These results indicate a herd with mixed origins with no clear indication of primary breed type. As with most trees involving feral herds, the tree is somewhat distorted.

SUMMARY

Genetic variability of this herd in general is high with exceptionally high *Ho*. Indeed, this was one of the highest *Ho* values seen for a feral herd. The rest of the estimates of variation were high but not exceptionally so and there was a low percentage of variation that is at risk. There is a possibility that this herd has seen a recent loss of population size which would explain the differences in *Ho* compared to *He* and would increase the risk to genetic diversity which would be considered slight based upon variation levels seen. Genetic similarity results suggest a herd with mixed ancestry.

RECOMMENDATIONS

Current variability levels are high enough that no action is needed at this point but the herd should be monitored closely if it is known that the herd size has seen a recent decline. If there has been a recent population decline, variability levels could drop quickly over the next 5-10 years.

Table 1. Allele frequencies of genetic variants observed in Salt Wells Creek HMA MG feral horse herd.

VHL20															
I	J	K	L	M	N	O	P	Q	R	S					
0.260	0.000	0.000	0.140	0.200	0.080	0.120	0.180	0.000	0.020	0.000					
HTG4															
I	J	K	L	M	N	O	P	Q	R						
0.000	0.000	0.260	0.200	0.440	0.020	0.020	0.060	0.000	0.000						
AHT4															
H	I	J	K	L	M	N	O	P	Q	R					
0.160	0.260	0.220	0.000	0.000	0.080	0.020	0.260	0.000	0.000	0.000					
HMS7															
I	J	K	L	M	N	O	P	Q	R						
0.000	0.280	0.000	0.220	0.160	0.200	0.080	0.040	0.020	0.000						
AHT5															
I	J	K	L	M	N	O	P	Q	R						
0.020	0.360	0.120	0.000	0.140	0.240	0.120	0.000	0.000	0.000						
HMS6															
I	J	K	L	M	N	O	P	Q	R						
0.000	0.000	0.020	0.180	0.280	0.100	0.120	0.300	0.000	0.000						
ASB2															
B	I	J	K	L	M	N	O	P	Q	R					
0.000	0.060	0.000	0.220	0.000	0.020	0.420	0.100	0.000	0.160	0.020					
HTG10															
H	I	J	K	L	M	N	O	P	Q	R	S	T			
0.000	0.020	0.000	0.020	0.200	0.180	0.220	0.140	0.000	0.120	0.100	0.000	0.000			
HMS3															
H	I	J	K	L	M	N	O	P	Q	R	S				
0.000	0.140	0.000	0.000	0.000	0.160	0.080	0.380	0.100	0.020	0.120	0.000				
ASB17															
D	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T
0.000	0.120	0.060	0.000	0.000	0.000	0.120	0.000	0.180	0.060	0.100	0.080	0.000	0.280	0.000	0.000
ASB23															
G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	V
0.000	0.000	0.220	0.180	0.160	0.060	0.000	0.000	0.000	0.000	0.000	0.000	0.300	0.000	0.080	0.000
LEX33															
F	G	K	L	M	N	O	P	Q	R	S	T				
0.000	0.120	0.020	0.480	0.020	0.000	0.000	0.000	0.240	0.120	0.000	0.000				

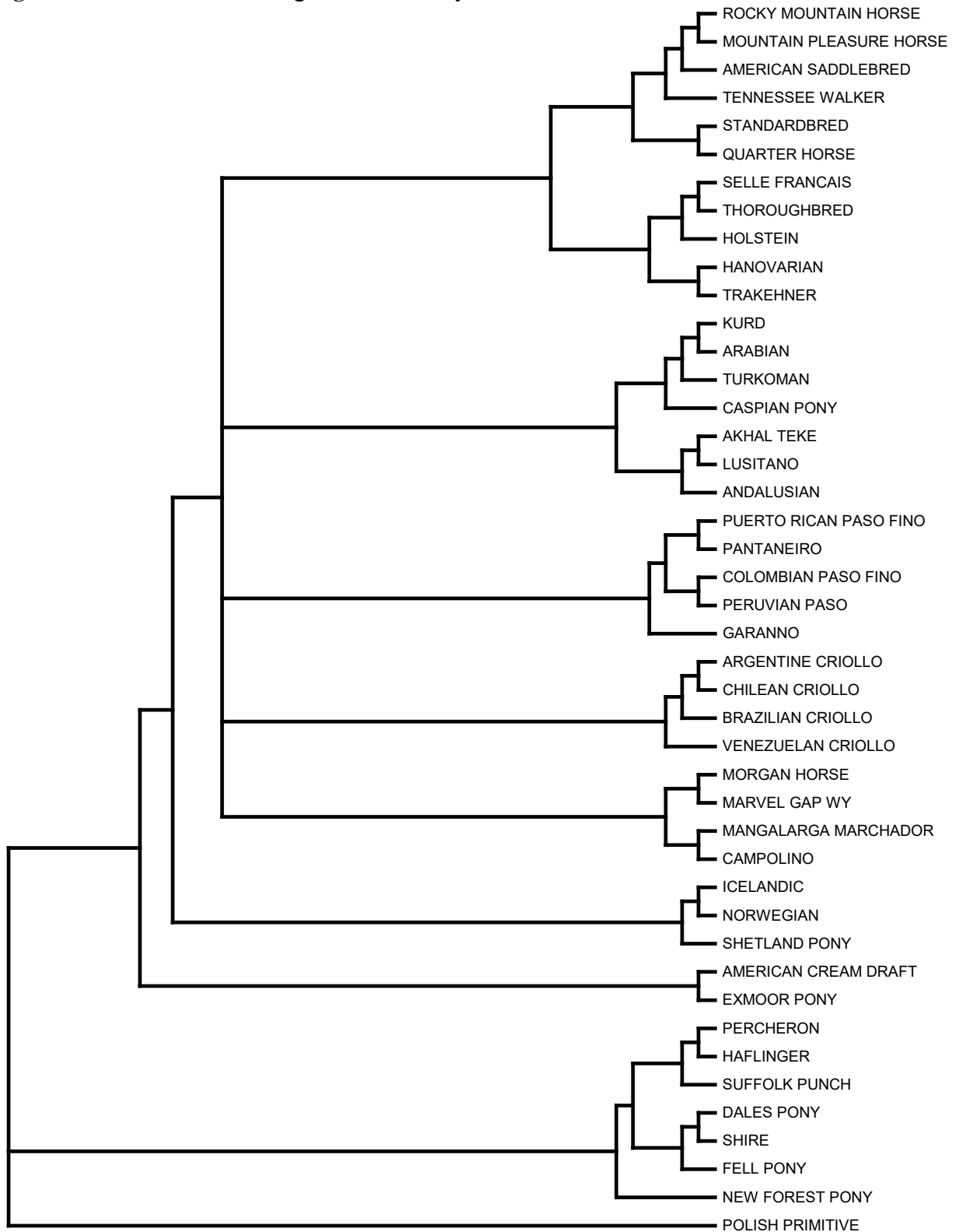
Table 2. Genetic variability measures.

	<i>N</i>	<i>Ho</i>	<i>He</i>	<i>Fis</i>	<i>Ae</i>	<i>TNV</i>	<i>MNA</i>	<i>Ra</i>	<i>%Ra</i>
MARVEL GAP WY	25	0.813	0.775	-0.050	4.63	80	6.67	15	0.188
Cleveland Bay	47	0.610	0.627	0.027	2.934	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	4.259	79	6.58	21	0.266
Arabian	47	0.660	0.727	0.092	3.814	86	7.17	30	0.349
Exmoor Pony	98	0.535	0.627	0.146	2.871	66	5.50	21	0.318
Friesian	304	0.545	0.539	-0.011	2.561	70	5.83	28	0.400
Irish Draught	135	0.802	0.799	-0.003	5.194	102	8.50	28	0.275
Morgan Horse	64	0.715	0.746	0.041	4.192	92	7.67	33	0.359
Suffolk Punch	57	0.683	0.711	0.038	3.878	71	5.92	13	0.183
Tennessee Walker	60	0.666	0.693	0.038	3.662	87	7.25	34	0.391
Thoroughbred	1195	0.734	0.726	-0.011	3.918	69	5.75	18	0.261
Feral Horse Mean	126	0.716	0.710	-0.012	3.866	72.68	6.06	16.96	0.222
Standard Deviation		0.056	0.059	0.071	0.657	13.02	1.09	7.98	0.088
Minimum		0.496	0.489	-0.284	2.148	37	3.08	0	0
Maximum		0.815	0.798	0.133	5.253	96	8.00	33	0.400
Domestic Horse Mean	80	0.710	0.720	0.012	4.012	80.88	6.74	23.79	0.283
Standard Deviation		0.078	0.071	0.086	0.735	16.79	1.40	10.11	0.082
Minimum		0.347	0.394	-0.312	1.779	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 Salt Wells Creek HMA MG feral horse herd to major groups of domestic horses.

	Mean <i>S</i>	Std	Minimum	Maximum
Light Racing and Riding Breeds	0.776	0.019	0.756	0.806
Oriental and Arabian Breeds	0.772	0.038	0.714	0.816
Old World Iberian Breeds	0.755	0.018	0.733	0.781
New World Iberian Breeds	0.764	0.022	0.723	0.795
North American Gaited Breeds	0.761	0.027	0.717	0.787
Heavy Draft Breeds	0.733	0.046	0.689	0.804
True Pony Breeds	0.704	0.044	0.647	0.752

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



Appendix 1. DNA data for the Salt Wells Creek HMA, Marvel Gap Subgroup, WY herd.

AID	VHL20	HTG4	AHT4	HMS7	AHT5	HMS6	ASB2	HTG10	HMS3	ASB17	ASB23	LEX33	LEX3
48679	MM	KP	JM	NN	JN	KP	NR	LL	MR	FM	IJ	LL	FM
48680	OO	KL	IJ	JJ	JM	LP	NN	LO	IP	RR	IK	GL	HO
48681	MM	KK	MO	NN	NN	MP	KN	LM	OR	FO	JK	LR	FM
48682	IP	LM	JO	LL	JJ	MN	KO	MM	MN	NR	IL	GL	HM
48683	MO	LM	IJ	JJ	JM	LP	NQ	LO	IP	PR	KL	GQ	NO
48684	LN	MM	HO	LM	JO	MM	QQ	IN	NO	MR	SS	LQ	MM
48685	IL	MN	IM	NO	JN	MP	KN	NR	MM	KK	KS	LL	FN
48686	IP	LM	JO	JL	JM	LP	NN	LN	IP	PR	IS	LQ	HO
48687	MP	MM	JO	LM	KO	MM	KN	MO	MN	NR	JS	GQ	HM
48688	IN	KL	IJ	MO	JN	MP	KK	LQ	NO	OR	KU	LQ	MM
48689	PP	LO	MN	MM	KN	LN	NO	KM	OQ	GR	II	LQ	FF
48690	IL	KM	IO	LN	JO	PP	IK	NQ	IP	FO	JJ	KL	OO
48691	IP	KM	HI	JL	JM	NO	KO	MQ	OO	FK	IU	LM	LL
48692	IP	KM	IO	LN	JN	NO	MQ	MN	OO	FG	JK	QR	FF
48693	OP	KM	IO	JM	KM	LP	KN	LO	OP	MR	KS	LQ	OO
48694	II	MP	HI	JN	JN	LP	NQ	MR	OO	FG	SU	LL	LL
48695	IL	MM	HI	JM	JN	MM	NQ	NN	OO	OR	SS	LQ	NN
48696	LP	KM	HJ	JP	KK	MP	KN	NQ	MO	MP	LU	LL	HH
48697	II	MM	JO	JL	JO	LO	IN	OR	IM	MM	IJ	LR	NN
48698	LL	LL	HO	JP	JO	PP	KN	NQ	OR	MP	SS	GR	OO
48699	MN	KM	HO	NQ	MN	LO	NO	QR	OR	KK	IK	LQ	OO
48700	IO	KL	IJ	JJ	KM	OP	NQ	LO	IO	MR	IJ	LQ	HH
48701	MN	LP	IO	LO	JJ	MO	NQ	LN	IR	KN	IS	LQ	LL
48702	MO	KM	HI	NO	IN	LM	NO	NR	OR	OR	JS	LR	FF
48703	MR	MM	JO	LM	NO	MN	IN	MO	MO	MR	SS	GR	NN

Genetic Analysis of the
Salt Wells Creek HMA, Miller Mountain Trap, WY

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April 11, 2011

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The following is a report of the genetic analysis of the Salt Wells Creek HMA, Miller Mountain Trap, WY. The herd will be referred to as the Salt Wells Creek HMA MMT in this report.

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 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 25 samples were received by Texas A&M University, Equine Genetics Lab on November 22, 2010. 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 Salt Wells Creek HMA MMT 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 Salt Wells Creek HMA MMT herd to domestic horse breed types are shown in Table 3. A dendrogram of relationship of the Salt Wells Creek HMA MMT herd to a standard set of domestic breeds is shown in Figure 1.

Genetic Variants: A total of 86 variants were seen in the Salt Wells Creek HMA MMT herd which is above the mean for feral herds as well as the mean for domestic breeds. Of these, 21 had frequencies below 0.05 which is just above the average percentage of variants at risk of future loss. Allelic diversity as represented by *Ae* is well above the average for feral herds while *MNA* is greater than the mean.

Genetic Variation: Observed heterozygosity in the Salt Wells Creek HMA MMT herd is above the feral mean while H_e is a good deal higher than average. H_o is a lower than H_e . Differences such as this can indicate population subdivision as the variation measures appear high for inbreeding to be a likely explanation for the positive F_{is} value.

Genetic Similarity: Overall similarity of the Salt Wells Creek HMA MMT herd to domestic breeds was just above average for feral herds. Highest mean genetic similarity of the Salt Wells Creek HMA MMT herd was with Light Racing and Riding breeds, followed by the Oriental and Arabian breeds with the North American Gaited breeds almost as close. As seen in Fig. 1, however, the Salt Wells Creek HMA MMT herd clusters within a group of “cold blood” horse breeds. These results indicate a herd with mixed origins with no clear indication of primary breed type.

SUMMARY

Genetic variability of this herd in general is on the high side but some of the diversity may be related to unrecognized population subdivision. Even if this is true, the H_o values indicated good levels of genetic variation. Genetic similarity results suggest a herd with mixed ancestry.

RECOMMENDATIONS

Current variability levels are high enough that no action is needed at this point.

Table 1. Allele frequencies of genetic variants observed in Salt Wells Creek HMA, Miller Mountain subgroup feral horse herd.

VHL20																		
I	J	K	L	M	N	O	P	Q	R	S								
0.167	0.015	0.000	0.030	0.167	0.167	0.045	0.197	0.076	0.136	0.000								
HTG4																		
I	J	K	L	M	N	O	P	Q	R									
0.000	0.000	0.121	0.182	0.545	0.015	0.061	0.076	0.000	0.000									
AHT4																		
H	I	J	K	L	M	N	O	P	Q	R								
0.242	0.030	0.152	0.106	0.091	0.015	0.000	0.242	0.121	0.000	0.000								
HMS7																		
I	J	K	L	M	N	O	P	Q	R									
0.000	0.121	0.152	0.318	0.076	0.227	0.106	0.000	0.000	0.000									
AHT5																		
I	J	K	L	M	N	O	P	Q	R									
0.000	0.303	0.136	0.061	0.197	0.167	0.136	0.000	0.000	0.000									
HMS6																		
I	J	K	L	M	N	O	P	Q	R									
0.000	0.000	0.061	0.136	0.197	0.015	0.212	0.379	0.000	0.000									
ASB2																		
B	I	J	K	L	M	N	O	P	Q	R								
0.000	0.000	0.000	0.258	0.000	0.167	0.061	0.061	0.152	0.182	0.121								
HTG10																		
H	I	J	K	L	M	N	O	P	Q	R	S	T						
0.000	0.045	0.000	0.000	0.182	0.303	0.015	0.258	0.000	0.000	0.182	0.015	0.000						
HMS3																		
H	I	J	K	L	M	N	O	P	Q	R	S							
0.000	0.061	0.000	0.000	0.000	0.106	0.303	0.015	0.288	0.197	0.030	0.000							
ASB17																		
D	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	V	W
0.000	0.091	0.000	0.000	0.061	0.030	0.106	0.000	0.136	0.333	0.015	0.076	0.045	0.091	0.000	0.000	0.000	0.000	0.015
ASB23																		
G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	V			
0.000	0.000	0.045	0.303	0.258	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.364	0.000	0.030	0.000			
LEX33																		
F	G	K	L	M	N	O	P	Q	R	S	T							
0.000	0.076	0.076	0.091	0.000	0.000	0.045	0.015	0.606	0.076	0.000	0.015							

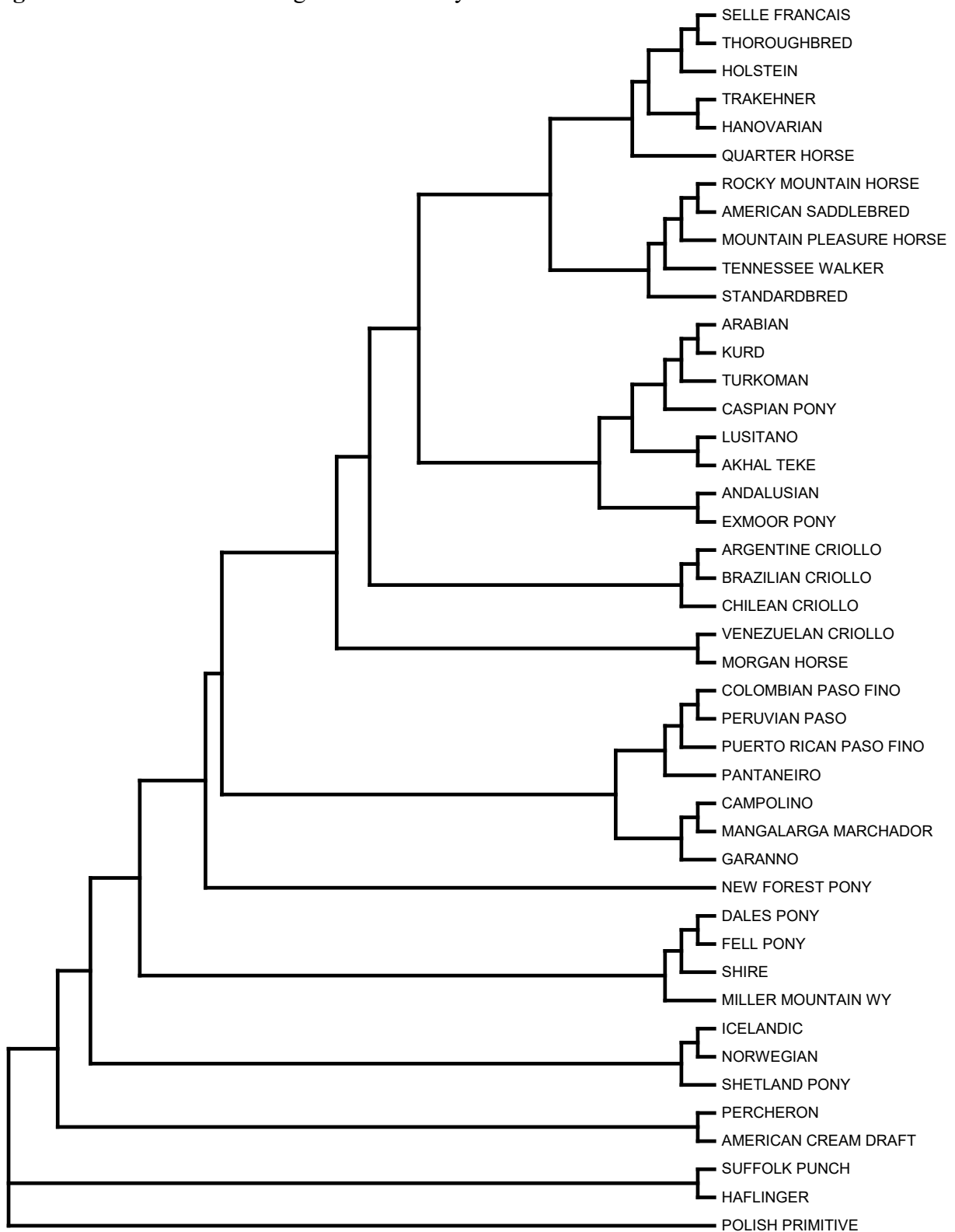
Table 2. Genetic variability measures.

	<i>N</i>	<i>Ho</i>	<i>He</i>	<i>Fis</i>	<i>Ae</i>	<i>TNV</i>	<i>MNA</i>	<i>Ra</i>	<i>%Ra</i>
Miller Mountain WY	25	0.730	0.765	0.045	4.62	86	7.17	21	0.244
Cleveland Bay	47	0.610	0.627	0.027	2.934	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	4.259	79	6.58	21	0.266
Arabian	47	0.660	0.727	0.092	3.814	86	7.17	30	0.349
Exmoor Pony	98	0.535	0.627	0.146	2.871	66	5.50	21	0.318
Friesian	304	0.545	0.539	-0.011	2.561	70	5.83	28	0.400
Irish Draught	135	0.802	0.799	-0.003	5.194	102	8.50	28	0.275
Morgan Horse	64	0.715	0.746	0.041	4.192	92	7.67	33	0.359
Suffolk Punch	57	0.683	0.711	0.038	3.878	71	5.92	13	0.183
Tennessee Walker	60	0.666	0.693	0.038	3.662	87	7.25	34	0.391
Thoroughbred	1195	0.734	0.726	-0.011	3.918	69	5.75	18	0.261
Feral Horse Mean	126	0.716	0.710	-0.012	3.866	72.68	6.06	16.96	0.222
Standard Deviation		0.056	0.059	0.071	0.657	13.02	1.09	7.98	0.088
Minimum		0.496	0.489	-0.284	2.148	37	3.08	0	0
Maximum		0.815	0.798	0.133	5.253	96	8.00	33	0.400
Domestic Horse Mean	80	0.710	0.720	0.012	4.012	80.88	6.74	23.79	0.283
Standard Deviation		0.078	0.071	0.086	0.735	16.79	1.40	10.11	0.082
Minimum		0.347	0.394	-0.312	1.779	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 Salt Wells Creek HMA MMT feral horse herd to major groups of domestic horses.

	Mean <i>S</i>	Std	Minimum	Maximum
Light Racing and Riding Breeds	0.790	0.032	0.748	0.836
Oriental and Arabian Breeds	0.773	0.043	0.719	0.826
Old World Iberian Breeds	0.755	0.007	0.746	0.762
New World Iberian Breeds	0.759	0.031	0.718	0.816
North American Gaited Breeds	0.770	0.036	0.719	0.804
Heavy Draft Breeds	0.751	0.025	0.719	0.784
True Pony Breeds	0.719	0.061	0.624	0.771

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



Appendix 1. DNA data for the Salt Wells Creek HMA, Miller Mountain subgroup, WY herd.

AID	VHL20	HTG4	AHT4	HMS7	AHT5	HMS6	ASB2	HTG10	HMS3	ASB17	ASB23	LEX33	LEX3
48654	PR	LL	HO	JO	NN	LP	KK	IL	PP	IP	IS	QQ	FF
48655	PP	KL	HJ	LL	JN	MO	PR	MO	MN	MP	JJ	QQ	PP
48656	PQ	LM	HO	JN	JK	LM	QR	MO	MN	NR	JS	OQ	FF
48657	PR	MM	HP	KL	JM	KP	KM	MM	QQ	KN	KS	QQ	IP
48658	NN	MM	HO	LN	NN	MM	MN	OR	IP	JP	JK	QQ	LP
48659	NN	NP	LO	KN	JJ	MO	NR	OR	PP	NQ	KK	KP	KM
48660	MR	OO	JK	LL	JJ	MM	NR	OO	NQ	FN	KK	QQ	FL
48661	IM	KM	HO	JM	KO	OP	NP	RR	NN	KM	JS	KR	FF
48662	MP	MM	HP	KL	MM	OO	KP	MO	QQ	KM	SS	GQ	PP
48663	NR	MM	JO	MO	JN	LP	MQ	IL	PP	NQ	IS	QR	LM
48664	MR	LP	LM	LL	KM	PP	PQ	LR	NP	MN	JJ	LQ	FF
48665	II	MP	LO	KM	JL	OP	MP	MN	NP	JM	JU	LQ	FN
48666	II	MM	JO	LN	JJ	NP	MM	RR	IP	NN	JS	OQ	KP
48667	NP	LM	HK	NO	NO	MP	KM	IR	IP	II	JS	QQ	FM
48668	JM	LM	HP	LL	MM	KO	KP	MO	MQ	KN	KS	QQ	IP
48669	RR	MM	HJ	LM	KN	KP	KM	LM	NN	FF	KS	GT	HL
48670	LO	KO	HO	JN	LO	PP	MR	OS	QR	NN	JS	QR	LL
48671	LP	MM	JJ	MO	JJ	PP	MQ	MM	MN	QR	JJ	QR	FP
48672	NN	MM	JO	LN	JJ	MM	OQ	OR	PP	NN	JK	KQ	LL
48673	IM	KM	HO	JN	JL	MP	MR	MR	IR	NN	JS	OR	LP
48674	IP	MM	HP	KO	JO	LO	KP	LM	MQ	MR	SS	QQ	PP
48675	IM	MM	OP	KL	MM	LP	KQ	MM	NQ	KM	KS	LQ	FF
48676	OP	MO	II	NN	JK	OP	KK	LL	OP	PP	KS	LQ	PP
48677	NQ	KL	JO	JO	JN	MP	OO	LR	MP	FN	KS	GQ	FF
48678	NN	KP	LO	KN	LN	LO	QQ	OO	PP	NN	JS	KQ	LL

Genetic Analysis of the
Great Divide Basin HMA, WY0002

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May 15, 2012

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The following is a report of the genetic analysis of the Great Divide Basin HMA, WY0002.

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 60 samples were received by Texas A&M University, Equine Genetics Lab on November 18, 2011. 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 Great Divide Basin HMA 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 Great Divide Basin HMA herd to domestic horse breed types are shown in Table 3. A dendrogram of relationship of the Great Divide Basin HMA herd to a standard set of domestic breeds is shown in Figure 1.

Genetic Variants: A total of 93 variants were seen in the Great Divide Basin HMA herd. This is only three less than the highest number observed for a feral herd and well above the mean for domestic breeds. Of these, 31 had frequencies below 0.05 which is a high percentage of variants at risk of future loss. Allelic diversity as represented by Ae and MNA is quite high as well.

Genetic Variation: Observed heterozygosity in the Great Divide Basin HMA herd is well above the feral mean as is He . Ho is almost identical to He indicating genetic equilibrium.

Genetic Similarity: Overall similarity of the Great Divide Basin HMA herd to domestic

breeds was about average for feral herds. Highest mean genetic similarity of the Great Divide Basin HMA herd was with Old World Iberian breeds followed closely by Light Racing and Riding breeds and New World Iberian breeds. As seen in Fig. 1, however, the Great Divide Basin HMA herd clusters with the Shetland Pony and outside any major group. These results indicate a herd with mixed origins with no clear indication of primary breed type. As with most trees involving feral herds, the tree is somewhat distorted.

SUMMARY

Genetic variability of this herd in general is high but understanding the diversity of this herd is somewhat complicated. This herd was previously sampled in 2003. At that time the sample consisted of two subdivisions of the herd area labeled North and South. Genetic variability levels of both groups were relatively high but not quite as high as seen here. Much of the high variability was attributed to mixing of the two groups and that would fit the herd now. However, the herds did not appear to be a single population but maintained some independence. This may not be the case now as the almost zero *F_{is}* is what would be expected for a single interbreeding group and not a subdivided population. The high percentage of variation that is at risk also is consistent with a formally subdivided population now interbreeding. Genetic similarity results suggest a herd with mixed ancestry.

RECOMMENDATIONS

Current variability levels are high enough that no action is needed, however, if population size drops below 150 breeding age animals, diversity levels can change quickly.

Table 1. Allele frequencies of genetic variants observed in Great Divide Basin HMA feral horse herd.

VHL20															
I	J	K	L	M	N	O	P	Q	R	S					
0.350	0.000	0.000	0.083	0.275	0.167	0.033	0.017	0.025	0.050	0.000					
HTG4															
I	J	K	L	M	N	O	P	Q	R						
0.000	0.000	0.092	0.192	0.483	0.000	0.233	0.000	0.000	0.000						
AHT4															
H	I	J	K	L	M	N	O	P	Q	R					
0.117	0.033	0.250	0.159	0.133	0.008	0.158	0.125	0.017	0.000	0.000					
HMS7															
I	J	K	L	M	N	O	P	Q	R						
0.000	0.008	0.008	0.283	0.108	0.359	0.217	0.017	0.000	0.000						
AHT5															
I	J	K	L	M	N	O	P	Q	R						
0.033	0.292	0.017	0.008	0.175	0.317	0.158	0.000	0.000	0.000						
HMS6															
I	J	K	L	M	N	O	P	Q	R						
0.000	0.000	0.183	0.225	0.075	0.142	0.158	0.217	0.000	0.000						
ASB2															
B	I	J	K	L	M	N	O	P	Q	R					
0.000	0.068	0.000	0.060	0.000	0.256	0.171	0.103	0.000	0.308	0.034					
HTG10															
H	I	J	K	L	M	N	O	P	Q	R	S	T			
0.000	0.025	0.000	0.050	0.108	0.225	0.025	0.258	0.008	0.008	0.293	0.000	0.000			
HMS3															
H	I	J	K	L	M	N	O	P	Q	R	S				
0.000	0.108	0.000	0.000	0.000	0.225	0.083	0.125	0.293	0.083	0.083	0.000				
ASB17															
D	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T
0.000	0.050	0.125	0.008	0.000	0.008	0.033	0.000	0.267	0.243	0.042	0.025	0.008	0.125	0.058	0.008
ASB23															
G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	V
0.083	0.025	0.100	0.117	0.233	0.158	0.000	0.000	0.000	0.000	0.000	0.000	0.167	0.000	0.117	0.000
LEX33															
F	G	K	L	M	N	O	P	Q	R	S	T				
0.000	0.000	0.192	0.208	0.008	0.008	0.243	0.008	0.150	0.183	0.000	0.000				

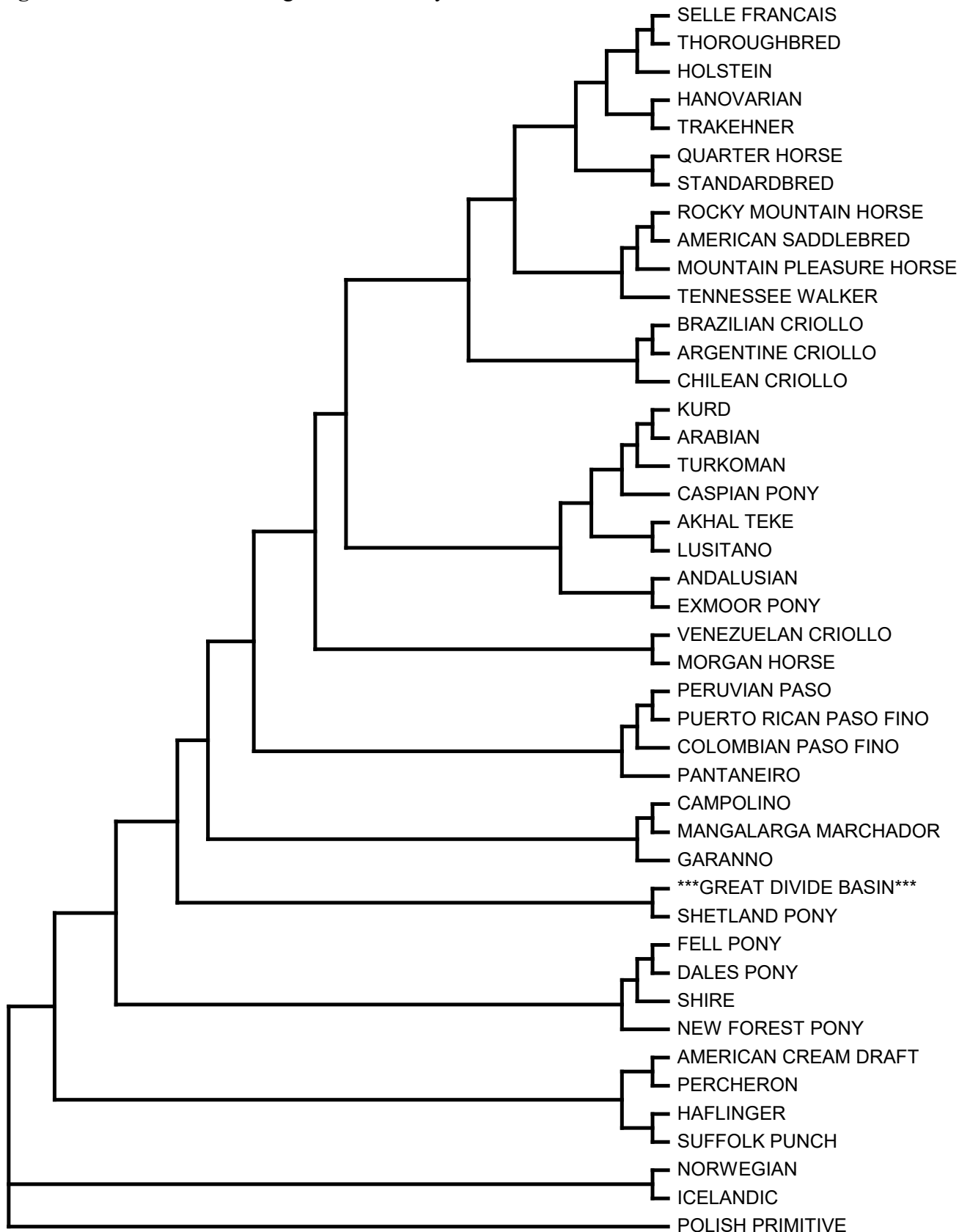
Table 2. Genetic variability measures.

	<i>N</i>	<i>Ho</i>	<i>He</i>	<i>Fis</i>	<i>Ae</i>	<i>TNV</i>	<i>MNA</i>	<i>Ra</i>	<i>%Ra</i>
GREAT DIVIDE BASIN WY	60	0.785	0.787	0.003	4.929	93	7.75	31	0.33
Cleveland Bay	47	0.610	0.627	0.027	2.934	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	4.259	79	6.58	21	0.266
Arabian	47	0.660	0.727	0.092	3.814	86	7.17	30	0.349
Exmoor Pony	98	0.535	0.627	0.146	2.871	66	5.50	21	0.318
Friesian	304	0.545	0.539	-0.011	2.561	70	5.83	28	0.400
Irish Draught	135	0.802	0.799	-0.003	5.194	102	8.50	28	0.275
Morgan Horse	64	0.715	0.746	0.041	4.192	92	7.67	33	0.359
Suffolk Punch	57	0.683	0.711	0.038	3.878	71	5.92	13	0.183
Tennessee Walker	60	0.666	0.693	0.038	3.662	87	7.25	34	0.391
Thoroughbred	1195	0.734	0.726	-0.011	3.918	69	5.75	18	0.261
Feral Horse Mean	126	0.716	0.710	-0.012	3.866	72.68	6.06	16.96	0.222
Standard Deviation		0.056	0.059	0.071	0.657	13.02	1.09	7.98	0.088
Minimum		0.496	0.489	-0.284	2.148	37	3.08	0	0
Maximum		0.815	0.798	0.133	5.253	96	8.00	33	0.400
Domestic Horse Mean	80	0.710	0.720	0.012	4.012	80.88	6.74	23.79	0.283
Standard Deviation		0.078	0.071	0.086	0.735	16.79	1.40	10.11	0.082
Minimum		0.347	0.394	-0.312	1.779	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 Great Divide Basin HMA feral horse herd to major groups of domestic horses.

	Mean <i>S</i>	Std	Minimum	Maximum
Light Racing and Riding Breeds	0.780	0.037	0.727	0.827
Oriental and Arabian Breeds	0.777	0.042	0.718	0.836
Old World Iberian Breeds	0.785	0.022	0.761	0.820
New World Iberian Breeds	0.779	0.030	0.729	0.807
North American Gaited Breeds	0.771	0.026	0.738	0.802
Heavy Draft Breeds	0.734	0.069	0.638	0.846
True Pony Breeds	0.747	0.028	0.713	0.777

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



Appendix 1. DNA data for the Great Divide Basin HMA, WY herd.

AID	VHL20	HTG4	AHT4	HMS7	AHT5	HMS6	ASB2	HTG10	HMS3	ASB17	ASB23	LEX3	LEX33
56831	IP	KM	HJ	LL	MO	PP	NQ	MR	IO	FR	KL	LM	KL
56832	IM	LM	KL	NO	JN	LN	OQ	LN	IP	RR	LU	FN	RR
56833	LM	MM	JM	LL	MN	LN	MN	MO	OP	FG	KS	NP	RR
56834	II	LO	JK	LL	IJ	KN	KM	MO	PR	RS	HS	NP	OR
56835	IM	OO	JJ	NN	JN	OO	QQ	MR	IO	FM	UU	FN	KL
56836	IM	MO	JN	NO	MN	OO	RR	QR	MO	GH	IL	HH	LO
56837	IN	MM	NO	NO	NO	KL	MQ	MO	MP	NN	IK	FM	LR
56838	IO	OO	JN	NN	JL	KO	NQ	MR	IN	FG	JU	MN	KL
56839	IN	OO	NN	LN	NN	KL	QQ	LR	IO	MN	II	FM	LL
56840	MM	LM	JO	NO	MN	LN	QQ	KM	MP	MT	UU	MQ	LO
56841	IN	MM	JJ	NO	JO	KL	NO	MO	IM	MN	GK	MP	RR
56842	NO	OO	OP	OO	JM	OP	QQ	KM	NP	MN	LS	FK	KR
56843	MN	LL	HL	LN	MM	MP	IN	MP	PQ	MR	LL	KM	KK
56844	MN	LL	JN	LN	JJ	MP	IN	LN	PR	NN	GU	KN	OQ
56845	IM	LM	HK	LN	JN	LN	MN	OR	MN	MM	JK	LN	OR
56846	IR	MM	HL	LM	OO	LN	MN	OR	MP	MM	GK	FP	OQ
56847	MM	MM	JJ	NP	JO	LM	IM	OO	PP	NR	GK	FP	QQ
56848	MN	MO	JN	NO	MN	LL	MR	MR	MN	GM	SS	NN	OQ
56849	IL	MM	JK	NO	JM	MM	II	MN	NR	MM	IJ	KM	LL
56850	NN	KM	HK	LN	JM	OO	CO	MR	OQ	GO	JU	MN	LQ
56851	II	MM	JO	LM	NO	NP	KM	OO	MP	GN	GK	FN	KR
56852	MM	MM	HL	NO	NN	KP	CM	OR	IM	MN	JU	MN	KL
56853	IL	KL	HL	LO	IN	KP	KM	MO	OP	NS	JS	KQ	KR
56854	II	MO	OO	LP	NO	LO	QQ	MO	QR	NS	LS	EN	LL
56855	IR	KM	JJ	LL	MO	KP	NO	RR	IO	FN	KS	KM	KQ
56856	MQ	MO	HO	JL	NN	NP	NN	OR	NP	RS	IK	MN	OR
56857	NN	LM	OO	MO	JN	NP	QQ	KK	PP	MM	UU	MQ	OQ
56858	MM	MM	KL	LN	NO	LP	MN	OR	MM	NN	JK	FN	QR
56859	IL	LM	HO	NO	JJ	KP	OQ	OR	MP	NN	IK	FN	OQ
56860	IM	KM	NN	LN	JJ	OO	MM	LR	OO	GM	LL	NQ	OQ
56861	II	LO	HP	LL	JN	LO	QQ	MO	NQ	GS	JS	LN	OQ
56862	IR	KM	JL	OO	JN	NP	MM	OR	IP	NR	IK	HN	KO
56863	MN	MM	IJ	NN	JJ	NO	QQ	RR	MP	JR	SU	KN	KL
56864	IR	LO	KL	LN	MN	KM	MO	LM	QR	MM	KS	FM	KO
56865	IM	MM	JK	MO	NO	LP	MQ	OO	MO	KN	KK	FP	KL
56866	MN	MO	KO	LO	JM	LM	NN	KM	MM	MN	LS	MM	KO
56867	MQ	LM	HL	LN	JN	NN	NN	OO	MP	MN	GJ	LL	OQ
56868	IM	MO	NN	OO	NO	KP	MN	RR	IO	NP	LS	MM	MR
56869	NN	KK	KK	LM	JJ	LO	OQ	MO	IP	NO	KL	NN	LO
56870	IN	MO	NO	OO	MO	KK	KQ	LR	PQ	NS	GI	FF	LR
56871	IP	MO	LN	MN	KM	KO	MQ	IM	OP	FG	KL	FF	LL
56872	IL	KL	NN	MN	JN	KP	MQ	LM	PR	MO	LL	NN	KO
56873	MR	LM	IL	LN	JO	LO	IM	MR	MQ	MR	KS	NN	KQ
56874	IM	LM	KL	MN	NN	KN	QR	LR	PR	KO	KS	NN	KO
56875	NQ	MO	JK	LN	MN	MO	KM	MR	MQ	MQ	SU	FF	LR
56876	MM	LM	KL	LN	NO	PP	NO	LR	NP	GP	JK	NN	KO
56877	IM	LM	HH	LN	IO	LL	IM	RR	MP	MR	GH	PP	NQ
56878	IL	LL	KK	LN	NN	KL	MN	LO	NP	GM	JK	NN	OO
56879	IM	MO	OO	MN	JN	NP	MO	IR	NR	OR	JL	NN	LP
56880	LR	LM	LO	LM	JO	LP	MM	MR	MM	NR	IL	KK	QR
56881	IN	MO	LN	NO	MN	KP	IQ	IL	MQ	KS	IK	LL	OR
56882	IL	KM	JK	KN	IN	KL	KQ	OO	IQ	MN	HS	MM	KL
56883	LO	MO	JJ	LO	JK	NP	NQ	KR	OP	GN	KL	LL	KO
56884	IM	MM	KL	NO	NN	KO	KQ	MR	PR	KM	IS	MM	OO
56885	NO	OO	JN	MN	JM	LO	OQ	MM	MP	MP	LS	NN	LO
56886	IM	MO	JN	NO	NO	LL	QQ	LO	OR	GN	JK	QQ	LQ
56887	IL	LM	HJ	LN	JO	KP	CM	OO	MP	MR	KK	FF	KO
56888	IM	KM	JN	NO	JM	PP	MO	OR	MP	GN	GS	NN	KR
56889	IN	MO	IK	MN	JM	LM	OQ	RR	MP	MR	GU	FF	RR
56890	IM	MO	IJ	MN	JN	KN	MQ	LO	IP	GM	JK	QQ	OQ

Genetic Analysis of the
White Mountain HMA, WY

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April 30, 2012

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The following is a report of the genetic analysis of the White Mountain HMA, WY.

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 60 samples were received by Texas A&M University, Equine Genetics Lab on September 7, 2011. 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 White Mountain HMA 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 White Mountain HMA herd to domestic horse breed types are shown in Table 3. A dendrogram of relationship of the White Mountain HMA herd to a standard set of domestic breeds is shown in Figure 1.

Genetic Variants: A total of 91 variants were seen in the White Mountain HMA herd which is far above the means for feral herds and domestic breeds. Of these, 34 had frequencies below 0.05 which is a high percentage of variants at risk of future loss. Allelic diversity as represented by Ae and MNA also have values much greater than the mean values.

Genetic Variation: Observed heterozygosity in the White Mountain HMA herd is slightly below the feral mean while He is only slightly higher than average. Ho is lower than He but not by a significant margin. The herd is very close to genetic equilibrium.

Genetic Similarity: Overall similarity of the White Mountain HMA herd to domestic breeds was about average for feral herds. Highest mean genetic similarity of the White Mountain HMA herd was with North American Gaited breeds, followed closely by the Light Racing and Riding breeds. As seen in Fig. 1, however, the White Mountain HMA herd clusters within a group Iberian horse breeds. These results indicate a herd with mixed origins with no clear indication of primary breed type. It is possible that there is some Spanish blood based upon the clustering but this is not strong evidence in this case. As with most trees involving feral herds, the tree is somewhat distorted.

SUMMARY

Genetic variability of this herd in general is on the high side but there is a high percentage of variation that is at risk and individual heterozygosity is below average by a small amount. The patterns seen here are very similar to what was seen in 2000 based upon blood typing. It was suggested then that there may be some gene flow into the population and that is consistent with the current data. The very high allelic diversity but high proportion of alleles at low frequency is just what would be expected if there was a small influx of horses into the herd at different times. It is possible there is introgression from the two neighboring HMAs with Salt Wells being the more probable based upon level of differentiation. Genetic similarity results suggest a herd with mixed ancestry.

RECOMMENDATIONS

Current variability levels are high enough that no action is needed at this point but the herd should be monitored closely due to the high proportion of rare alleles. This is especially true if it is known that the herd size has seen a recent decline.

Table 1. Allele frequencies of genetic variants observed in White Mountain HMA feral horse herd.

VHL20																
I	J	K	L	M	N	O	P	Q	R	S						
0.167	0.000	0.000	0.083	0.191	0.317	0.050	0.000	0.017	0.175	0.000						
HTG4																
I	J	K	L	M	N	O	P	Q	R							
0.000	0.000	0.025	0.125	0.700	0.017	0.033	0.100	0.000	0.000							
AHT4																
H	I	J	K	L	M	N	O	P	Q	R						
0.375	0.042	0.283	0.000	0.125	0.008	0.000	0.159	0.008	0.000	0.000						
HMS7																
I	J	K	L	M	N	O	P	Q	R							
0.000	0.141	0.117	0.341	0.067	0.250	0.067	0.000	0.017	0.000							
AHT5																
I	J	K	L	M	N	O	P	Q	R							
0.017	0.283	0.017	0.050	0.050	0.150	0.433	0.000	0.000	0.000							
HMS6																
I	J	K	L	M	N	O	P	Q	R							
0.000	0.000	0.067	0.025	0.108	0.008	0.050	0.742	0.000	0.000							
ASB2																
B	I	J	K	L	M	N	O	P	Q	R						
0.050	0.283	0.000	0.075	0.000	0.108	0.125	0.008	0.017	0.242	0.092						
HTG10																
H	I	J	K	L	M	N	O	P	Q	R	S	T				
0.000	0.025	0.000	0.017	0.017	0.292	0.117	0.125	0.000	0.000	0.382	0.025	0.000				
HMS3																
H	I	J	K	L	M	N	O	P	Q	R	S					
0.000	0.108	0.000	0.000	0.000	0.083	0.142	0.033	0.525	0.092	0.017	0.000					
ASB17																
D	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T	
0.000	0.108	0.000	0.000	0.000	0.000	0.191	0.000	0.117	0.050	0.067	0.025	0.017	0.200	0.217	0.008	
ASB2																
G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	V	
0.025	0.000	0.125	0.067	0.358	0.250	0.000	0.000	0.000	0.000	0.000	0.000	0.100	0.000	0.075	0.000	
LEX33																
F	G	K	L	M	N	O	P	Q	R	S	T					
0.000	0.017	0.083	0.350	0.125	0.000	0.092	0.108	0.133	0.017	0.025	0.050					

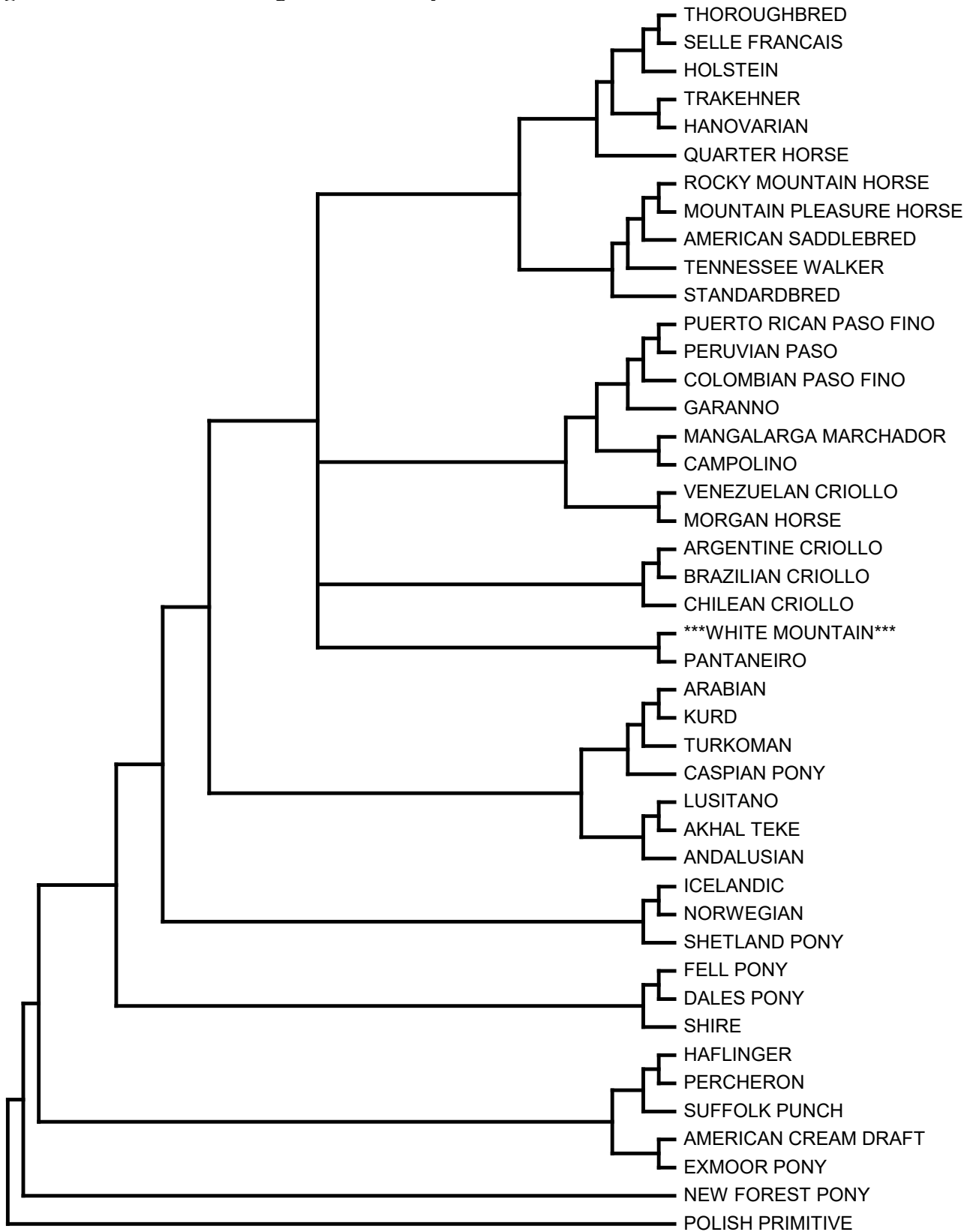
Table 2. Genetic variability measures.

	<i>N</i>	<i>Ho</i>	<i>He</i>	<i>Fis</i>	<i>Ae</i>	<i>TNV</i>	<i>MNA</i>	<i>Ra</i>	<i>%Ra</i>
WHITE MOUNTAIN WY	60	0.701	0.715	0.020	4.06	91	7.58	34	0.374
Cleveland Bay	47	0.610	0.627	0.027	2.934	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	4.259	79	6.58	21	0.266
Arabian	47	0.660	0.727	0.092	3.814	86	7.17	30	0.349
Exmoor Pony	98	0.535	0.627	0.146	2.871	66	5.50	21	0.318
Friesian	304	0.545	0.539	-0.011	2.561	70	5.83	28	0.400
Irish Draught	135	0.802	0.799	-0.003	5.194	102	8.50	28	0.275
Morgan Horse	64	0.715	0.746	0.041	4.192	92	7.67	33	0.359
Suffolk Punch	57	0.683	0.711	0.038	3.878	71	5.92	13	0.183
Tennessee Walker	60	0.666	0.693	0.038	3.662	87	7.25	34	0.391
Thoroughbred	1195	0.734	0.726	-0.011	3.918	69	5.75	18	0.261
Feral Horse Mean	126	0.716	0.710	-0.012	3.866	72.68	6.06	16.96	0.222
Standard Deviation		0.056	0.059	0.071	0.657	13.02	1.09	7.98	0.088
Minimum		0.496	0.489	-0.284	2.148	37	3.08	0	0
Maximum		0.815	0.798	0.133	5.253	96	8.00	33	0.400
Domestic Horse Mean	80	0.710	0.720	0.012	4.012	80.88	6.74	23.79	0.283
Standard Deviation		0.078	0.071	0.086	0.735	16.79	1.40	10.11	0.082
Minimum		0.347	0.394	-0.312	1.779	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 White Mountain HMA feral horse herd to major groups of domestic horses.

	Mean <i>S</i>	Std	Minimum	Maximum
Light Racing and Riding Breeds	0.755	0.025	0.715	0.784
Oriental and Arabian Breeds	0.739	0.039	0.680	0.798
Old World Iberian Breeds	0.749	0.031	0.716	0.786
New World Iberian Breeds	0.746	0.036	0.700	0.789
North American Gaited Breeds	0.762	0.035	0.723	0.806
Heavy Draft Breeds	0.684	0.056	0.591	0.743
True Pony Breeds	0.720	0.026	0.695	0.759

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



Appendix 1. DNA data for the White Mountain HMA, WY herd.

AID	VHL20	HTG4	AHT4	HMS7	AHT5	HMS6	ASB2	HTG10	HMS3	ASB17	ASB23	LEX33	LEX3
55250	IN	LM	JO	JN	JJ	MP	IM	MR	PQ	SS	KU	LL	FP
55251	NR	MM	IJ	JK	NO	PP	IM	MN	PP	KR	II	OP	FF
55252	IO	LM	HH	JL	OO	MP	MQ	NR	MP	KK	KK	OP	FL
55253	IM	LM	HJ	JN	JN	MP	IM	RR	PP	KS	SU	LL	FF
55254	NR	MM	HJ	JL	OO	PP	IR	MR	PP	KR	IS	LT	FH
55255	NN	MM	LO	KL	JJ	PP	II	MR	NN	OR	KL	MT	FF
55256	IM	MN	HJ	LO	NO	NP	KN	MR	MP	OP	IJ	KR	HP
55257	IN	MM	HJ	LN	NO	PP	IK	RR	PQ	QR	KL	MS	HL
55258	MN	MM	HH	LM	JO	PP	IR	MR	NP	FR	KK	LL	HP
55259	LL	MP	HH	JN	JJ	OP	IK	MR	PQ	FM	KU	LQ	KP
55260	IM	LM	JL	NN	NO	MP	IR	MR	PP	FS	KK	KP	FF
55261	MN	MP	HJ	LN	OO	PP	IN	MR	NP	FK	KL	MP	HP
55262	MR	MM	JO	KM	JO	PP	IM	MN	PP	MR	LL	KL	FP
55263	IO	LP	OO	LO	IO	PP	IK	MN	IP	KS	IK	PQ	FN
55264	NR	MM	HJ	LN	OO	PP	IQ	MM	NP	SS	KU	PT	FH
55265	NN	MM	HI	KL	NO	MP	QQ	MR	PQ	MN	KL	KL	FF
55266	NR	MM	LO	JN	NO	MP	IQ	NR	NP	KS	JS	LL	FF
55267	MR	MM	JJ	KN	JJ	PP	IP	MR	NP	MS	IK	PQ	FP
55268	MN	KP	HI	JK	NO	MP	IM	OR	PP	KR	LL	LM	FP
55269	RR	MM	HO	JL	NO	PP	MQ	RR	IP	KK	KL	LT	FP
55270	MM	MM	JJ	LL	NN	PP	QQ	MR	PP	SS	JK	LL	FH
55271	LN	MM	HJ	LO	JN	PP	BI	MO	MP	FR	LU	KQ	FO
55272	NR	LL	HM	LN	JM	OP	QQ	OR	NQ	QS	KL	LL	FP
55273	MN	MP	LO	LM	LO	PP	QR	MO	IN	MR	KS	QQ	FF
55274	LM	MM	HO	LN	JL	KK	NQ	MO	OQ	NS	KS	LO	FH
55275	MN	MM	HH	LL	OO	PP	IN	RR	PQ	KO	KL	OS	FF
55276	IN	MM	HH	LO	JM	KO	BQ	IO	IN	MM	GK	KL	FN
55277	LL	LL	HO	NO	LO	KP	KR	MO	OO	MO	KK	OR	KN
55278	IM	MM	JJ	JN	OO	MP	IR	MR	PP	FR	KK	LP	FF
55279	IN	KN	HJ	MO	JO	MP	NQ	KR	IN	MO	KS	LQ	FP
55280	IR	MM	JO	KL	NO	MP	MM	NO	PP	KM	KL	KQ	PP
55281	MN	MO	HL	LL	JJ	KP	BI	OR	IP	SS	IK	GM	FF
55282	IO	MP	HH	LN	JJ	LO	NN	KR	PR	NT	LS	LO	MM
55283	NO	MP	OP	LL	JO	LO	IN	IR	OR	NR	JL	LO	MM
55284	LR	LL	JL	NO	MO	PP	BQ	OR	IP	MS	LS	QQ	II
55285	NR	MP	HJ	LO	JO	PP	IQ	NN	II	RS	IK	LM	FF
55286	IR	KM	JO	KN	NO	PP	MQ	NO	PP	KM	LL	KQ	PP
55287	IL	MM	LO	NN	MM	KP	KN	MM	NP	FF	IS	MO	HP
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55289	II	OO	HH	QQ	KO	PP	QQ	LS	IN	OR	IL	LQ	PP
55290	OR	MM	HL	JN	JO	PP	IR	RR	PP	RR	LS	LM	FF
55291	IN	MM	HJ	JL	OO	PP	PQ	MR	PP	KO	KL	KM	FF
55292	IR	MM	JJ	LL	NO	MP	MQ	NO	NP	FM	KL	KM	PP
55293	OR	PP	HO	MN	MN	LM	KO	MM	MM	NR	LU	LQ	MM
55294	MN	MM	HO	LN	JO	PP	II	RR	NQ	KR	LL	OS	FF
55295	LQ	LM	LO	LN	LL	KO	BK	MS	MM	KM	JL	LQ	FF
55296	MN	MM	LO	LM	JO	PP	NR	NR	PP	RS	IK	LP	FF
55297	NR	MM	HJ	JN	OO	PP	QQ	NR	NP	NR	II	PP	FF
55298	RR	MM	JL	KN	JO	PP	IQ	MO	PQ	FK	KU	MT	PP
55299	MN	MM	IO	KL	OO	PP	IR	MR	PP	FK	IK	LO	FF
55300	NR	MM	HL	JN	JO	PP	IR	RR	PP	FR	KS	LL	FF
55301	LM	MM	HJ	LN	JJ	KP	BM	IO	MP	RS	KL	GM	PP
55302	MQ	LM	HH	MN	KL	PP	KN	LS	MP	PS	JK	LQ	FF
55303	MN	MM	JL	KM	JO	PP	QQ	NR	PQ	FS	KK	LT	FF
55304	NN	MP	HH	LL	IJ	PP	II	MO	IN	RS	GU	LL	LL
55305	NN	MM	JJ	JK	JO	PP	NQ	MR	PP	KS	JK	LM	FF
55306	MN	MM	JJ	LL	OO	PP	NN	MR	PQ	OS	JL	MM	PP
55307	IN	LM	HI	KL	NO	MP	IN	MR	MP	KK	IK	OP	FF
55308	NN	MM	HJ	JN	JO	PP	IQ	MR	PP	KR	KS	LL	FF
55309	IN	LO	LL	JL	JO	PP	QR	MR	II	PS	GL	LQ	PP

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May 7, 2012

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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 Little Colorado HMA 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 Little Colorado HMA herd to domestic horse breed types are shown in Table 3. A dendrogram of relationship of the Little Colorado HMA herd to a standard set of domestic breeds is shown in Figure 1.

Genetic Variants: A total of 80 variants were seen in the Little Colorado HMA herd which is above the mean for feral herds and right at the mean for domestic breeds. Of these, 15 had frequencies below 0.05 which is a low percentage of variants at risk of future loss. Allelic diversity as represented by Ae and MNA are both greater than the mean but not by a large margin.

Genetic Variation: Observed heterozygosity in the Little Colorado HMA herd is well above the feral mean as is He . Ho is slightly lower than He . The difference is not significant and the herd appears to be in genetic equilibrium.

Genetic Similarity: Overall similarity of the Little Colorado HMA herd to domestic breeds was a little above average for feral herds. Highest mean genetic similarity of the Little Colorado HMA herd was with Light Racing and Riding breeds, followed by the North American Gaited breeds. As seen in Fig. 1, however, the Little Colorado HMA herd clusters within the group of Nordic horse breeds. This same relationship was seen in the sample from this herd collected in 2007. This does not likely represent true relationship but instead is an artifact of a herd with mixed origins with changes in the genetic basis of the herd over time. The herd most likely is derived from riding or ranch stock of North American origin but possibly some draft blood which could be the cause of the placement in the tree. As with most trees involving feral herds, the tree is somewhat distorted. In comparison to other feral herds from Wyoming (Fig. 2), there is no herd that is closely similar.

SUMMARY

Genetic variability of this herd in general is on the high side but compared to 2007 there has been a loss of observed heterozygosity. Every other measure of genetic variation was essentially the same with what differences there were attributable to sampling variation. The change in H_o is accompanied by a change in F_{is} from a negative to a positive value. This could be an indication in a loss of population size which has resulted in greater inbreeding (positive F_{is}). However, this would not be expected in a period of only 5 years. Genetic similarity results suggest a herd with mixed ancestry.

RECOMMENDATIONS

Current variability levels are high enough that no action is needed at this point but the herd should be monitored closely due to the observed change in H_o . This is especially true if it is known that the herd size has seen a recent major reduction in size.

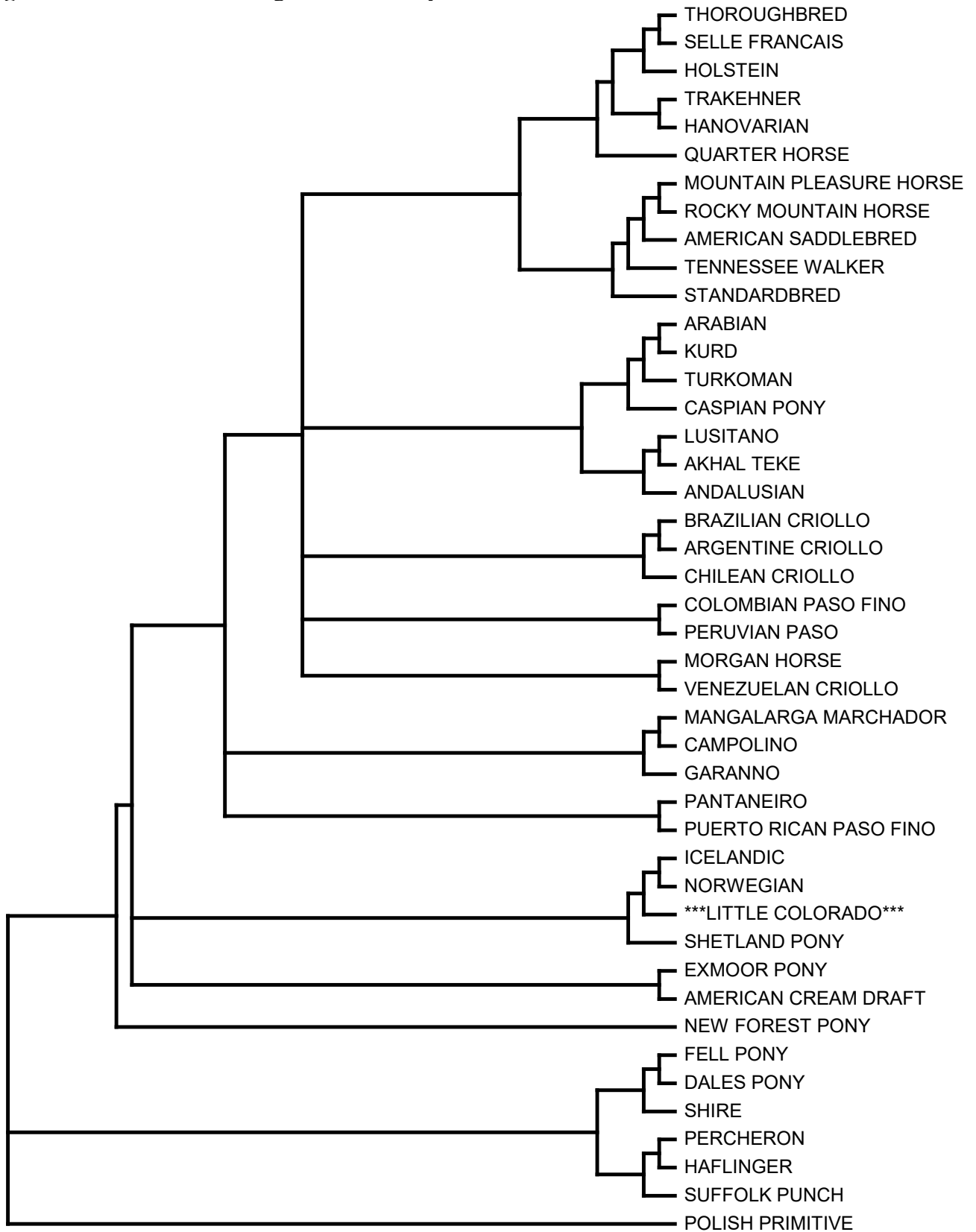
Table 2. Genetic variability measures.

	<i>N</i>	<i>Ho</i>	<i>He</i>	<i>Fis</i>	<i>Ae</i>	<i>TNV</i>	<i>MNA</i>	<i>Ra</i>	<i>%Ra</i>
LITTLE COLORADE WY	45	0.761	0.768	0.009	4.54	80	6.67	15	0.188
Cleveland Bay	47	0.610	0.627	0.027	2.934	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	4.259	79	6.58	21	0.266
Arabian	47	0.660	0.727	0.092	3.814	86	7.17	30	0.349
Exmoor Pony	98	0.535	0.627	0.146	2.871	66	5.50	21	0.318
Friesian	304	0.545	0.539	-0.011	2.561	70	5.83	28	0.400
Irish Draught	135	0.802	0.799	-0.003	5.194	102	8.50	28	0.275
Morgan Horse	64	0.715	0.746	0.041	4.192	92	7.67	33	0.359
Suffolk Punch	57	0.683	0.711	0.038	3.878	71	5.92	13	0.183
Tennessee Walker	60	0.666	0.693	0.038	3.662	87	7.25	34	0.391
Thoroughbred	1195	0.734	0.726	-0.011	3.918	69	5.75	18	0.261
Feral Horse Mean	126	0.716	0.710	-0.012	3.866	72.68	6.06	16.96	0.222
Standard Deviation		0.056	0.059	0.071	0.657	13.02	1.09	7.98	0.088
Minimum		0.496	0.489	-0.284	2.148	37	3.08	0	0
Maximum		0.815	0.798	0.133	5.253	96	8.00	33	0.400
Domestic Horse Mean	80	0.710	0.720	0.012	4.012	80.88	6.74	23.79	0.283
Standard Deviation		0.078	0.071	0.086	0.735	16.79	1.40	10.11	0.082
Minimum		0.347	0.394	-0.312	1.779	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 Little Colorado HMA feral horse herd to major groups of domestic horses.

	Mean <i>S</i>	Std	Minimum	Maximum
Light Racing and Riding Breeds	0.786	0.027	0.749	0.820
Oriental and Arabian Breeds	0.753	0.034	0.712	0.800
Old World Iberian Breeds	0.760	0.021	0.739	0.793
New World Iberian Breeds	0.747	0.015	0.728	0.769
North American Gaited Breeds	0.770	0.019	0.747	0.793
Heavy Draft Breeds	0.714	0.048	0.670	0.799
True Pony Breeds	0.717	0.044	0.647	0.765

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



Appendix 1. DNA data for the Little Colorado HMA, WY herd.

AID	VHL20	HTG4	AHT4	HMS7	AHT5	HMS6	ASB2	HTG10	HMS3	ASB17	ASB23	LEX33	LEX3
55205	IR	KP	HJ	LM	MN	PP	MN	KR	MN	MR	IK	GQ	NN
55206	LL	PP	IP	JL	MO	MP	OQ	MM	OP	OR	SU	OR	LL
55207	IM	KP	OP	LN	MO	LO	IK	OR	IN	MS	LU	OO	HH
55208	MR	MO	HO	JL	JN	PP	KO	MO	II	FR	KU	KK	MM
55209	MM	PP	JO	JL	LO	MP	IQ	OO	MN	NR	IU	KL	MM
55210	LP	PP	LO	NN	JO	LP	MN	KR	IO	SS	LU	KL	LL
55211	MM	OP	JJ	JN	JN	LM	NQ	MO	MP	GI	SS	KL	LL
55212	MQ	LO	LL	LN	MO	PP	NN	IK	NR	RS	JS	GK	LL
55213	NO	MO	HP	LN	MO	PP	MN	IK	IN	RT	JS	KQ	LL
55214	MR	KO	JJ	JL	MM	PP	IN	KM	MM	FR	KU	KQ	KK
55215	NN	MO	HJ	LL	MN	OP	KN	LO	NN	GR	LU	KR	NN
55216	NP	MO	HO	JL	MN	LO	MN	LR	NP	NR	LU	KQ	KK
55217	NN	LM	HJ	LQ	MN	MO	NN	IO	MR	KR	II	GO	PP
55218	IP	LO	IO	NN	KO	MP	OQ	IR	MP	KN	SU	LQ	KK
55219	NP	OP	HJ	JN	KO	MP	MM	LM	PR	NR	KS	KL	NN
55220	MN	KP	JO	LL	MN	PP	NN	KM	MN	FO	IK	KQ	NN
55221	MN	OP	HO	LN	JK	LO	NP	LO	MM	KN	IU	LL	FF
55222	MN	PP	JO	NN	JK	OP	NP	MR	MM	KN	IU	LL	KK
55223	MN	KP	JJ	MN	LM	PP	KO	MM	MN	RR	KK	KL	MM
55224	OO	PP	HO	NN	JO	PP	NO	RR	IO	IS	LS	LO	NN
55225	NN	MO	HJ	LL	MO	OP	NN	MM	NN	GR	II	GK	LL
55226	IP	KL	HO	JN	JK	MO	KQ	RR	PP	NS	SU	LO	LL
55227	II	KL	IO	NN	JK	PP	KQ	IR	PP	NO	LU	LO	LL
55228	NN	MO	JP	LN	MN	OP	NO	MM	NR	OR	IU	KK	LL
55229	OO	MO	KO	NN	JJ	PP	MO	KM	IR	IR	JS	KL	NN
55230	MP	OP	IO	LN	JO	LN	OO	IO	PP	MR	IU	LL	MN
55231	MO	PP	HO	JN	JO	LP	NN	IO	MM	FG	KS	LO	MP
55232	MM	PP	JJ	JL	JN	PP	NO	MO	MM	NR	SU	KL	MP
55233	IM	OP	HO	MN	NN	PP	OO	IM	NP	GR	KK	KL	HN
55234	NR	KP	HJ	JN	LN	MP	OO	MR	NN	GR	KK	LL	MP
55235	MN	OP	OO	NN	MN	OP	NO	LO	NR	IR	SU	KL	LM
55236	IR	MP	IO	MN	KN	PP	MQ	IP	IM	NP	KK	LS	FM
55237	MM	OO	JO	LN	LN	MM	IQ	IO	PR	IR	IS	KR	LM
55238	IN	MP	HO	LN	MN	NO	NO	MO	NO	GS	KS	KR	HN
55239	MN	MM	JO	LL	JN	MO	NO	IO	IM	MN	IL	GK	LN
55240	NP	MP	JJ	NN	JJ	MO	II	LO	PR	NR	LS	KR	FL
55241	MM	OO	JO	JN	JN	PP	IQ	MO	IP	MR	SS	KL	LM
55242	IO	MO	JO	NN	OO	PP	NQ	IO	PP	KM	UU	KQ	KN
55243	OP	LM	JO	NN	MO	MN	NO	IM	MP	NN	SU	KL	KN
55244	II	LM	HO	JN	JK	MP	NO	II	II	FG	LU	QQ	NN
55245	NO	OP	HO	LN	JK	OO	OP	LR	MN	KN	JU	LO	FK
55246	MR	KP	JO	JM	MN	MP	KO	MO	IN	FG	IK	GL	HM
55247	NR	PP	HJ	LM	MN	MP	NO	MM	MP	GR	IK	LQ	HN
55248	MN	MP	OO	JL	MO	NO	IN	OO	IO	MS	SS	KO	HO
55249	IQ	PP	LO	NN	JM	PP	NN	OO	IN	RS	LS	KQ	LM

Figure 2. Dendrogram of genetic similarity of Wyoming herds.

