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To cite this article: D. Bigi, P. Zambonelli, G. Perrotta & M. Blasi (2007) The Ventasso Horse: genetic characterization by microsatellites markers, Italian Journal of Animal Science, 6:sup1, 50-52, DOI: [10.4081/ijas.2007.1s.50](https://doi.org/10.4081/ijas.2007.1s.50)

To link to this article: <https://doi.org/10.4081/ijas.2007.1s.50>



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Published online: 15 Mar 2016.



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The Ventasso Horse: genetic characterization by microsatellites markers

D. Bigi^{1,2}, P. Zambonelli^{1,2}, G. Perrotta³, M. Blasi³

¹ Dipartimento di Protezione e Valorizzazione Agroalimentare. Università di Bologna, Italy

² RARE – Razze Autoctone a Rischio di Estinzione, Italy

³ Laboratorio Genetica e Servizi. Cremona-Potenza, Italy

Corresponding author: Daniele Bigi. Dipartimento di Protezione e Valorizzazione Agroalimentare, Sezione Allevamenti Zootecnici. Università di Bologna. Via F.lli Rosselli 107, 42100 Reggio Emilia, Italy - Tel. +39 0522 290514 - Fax: +39 0522 290523 - Email: daniele.big@unibo.it

ABSTRACT: The genetic structure of Ventasso Horse (VH) was investigated using 12 microsatellites. The analyses were carried out on 117 VH individuals and the results were compared with those obtained analysing 11 other breeds reared in Italy. All microsatellites were polymorphic in VH and in the other breeds. A total of 124 alleles (from 6 to 19 alleles per microsatellite) were detected. Average heterozygosity was 0.743 in VH and ranged from 0.613 to 0.759 in the other breeds. The mean F_{ST} value had an average value of 0.0932. Genetic distances were calculated using Nei's standard genetic distance (Ds). The smallest Ds values were found between VH and Anglo-Arab, Thoroughbred, Maremmano and Lipizzan horse breeds. Phylogenetic trees constructed using neighbour-joining method showed two clear separate clusters: the first includes Bardigiano, Haflinger and Italian Heavy Draught Horse, the second contains the other 9 breeds.

Key words: Italian horse breeds, Microsatellites, Genetic diversity.

INTRODUCTION – The Ventasso Horse (VH) is a native Italian breed selected for saddle, and its name comes from Mount Ventasso, located in Province of Reggio Emilia, in the Center of Italy. The breeding area is a mountainous territory located for the most part in the Appennino Tosco-Emiliano National Park. In the last decades, the decreasing of economic interest in maintaining a pure VH line caused the reduction of its effective number and the larger utilization of stallions of different breeds. In particular, during the 1950s and '60s some Thoroughbred, some Maremmano and one Lipizzan stallions were introduced, while in the last twenty years different Anglo-Arab stallions were utilized to improve the athletic performances. According to the FAO-UNEP (2000), the small number of VH population, of about 300 individuals, place it in the list of endangered breeds. Genetic characterization is an important step in breed conservation and can be useful for future breeding strategies. Microsatellites have been successfully utilized in many studies for estimating genetic distances among closely related horse populations (Aberle *et al.*, 2004; Bjornstad *et al.*, 2000; Solis *et al.*, 2005; Marletta *et al.*, 2006). The aim of this study is the genetic characterization of Ventasso Horse based on variation in microsatellite loci and the determination of its genetic relationship with 11 horse breeds reared in Italy.

MATERIAL AND METHODS – A total of 117 Ventasso Horses, belonging to 49 different stud farms, representing about the half of the total population, were analyzed. Comparison with 11 other breeds reared in Italy (64 Anglo-Arab – AA, 202 Arab – AR, 46 Bardigiano - BA, 73 Haflinger - HF, 128 Italian Trotter - IT, 36 Lipizzan - LP, 50 Maremmano - MA, 42 Murgese - MU, 15 Sanfratellano - SA, 82 Italian Heavy Draught Horse - HD, 53 Thoroughbred – TB), typed for the same microsatellite markers, was carried out.

DNA extraction from hair roots, using proteinase k digestion, was performed for all samples.

The evaluation of the genetic structure of VH was investigated using 12 microsatellites (HTG10, VHL20, HTG7, HTG4, AHT5, AHT4, HMS3, HMS6, HMS7, LEX03, HMS2, ASB2), including the 9 markers recommended for parentage testing by the International Society of Animal Genetics. Gel electrophoresis and genotype determination were performed on ABI Prism 3100 DNA Sequencer, equipped with Genscan and Genotyper software.

Estimation of allele frequencies was made using GENEPOP package, version 3.4 (Raymond and Rousset, 1995). Observed (H_O) and expected (H_P) heterozygosity, mean number of alleles (MNA), F statistics (F_{IS} and F_{ST}) (Weir

and Cockeram, 1984), Nei's genetic standard distance (Ds) (Nei, 1972), using all markers but the sex-linked microsatellites LEX03, were calculated with GENETIX software, version 4.03 (Belkhir *et al.*, 2001). The neighbour-joining tree topology was obtained with PHYLIP package, version 3.65 (Felsenstein, 1989). For the validation of the tree topology, 1000 bootstrap resample were performed.

RESULTS AND CONCLUSIONS – All the microsatellites were polymorphic in VH and in the other breeds. A total of 124 alleles (from 6 to 19 alleles per microsatellite) were detected. Observed heterozygosity was 0.743 in VH and ranged from 0.613 (LP) to 0.759 (HD). The MNA per horse population varied between 4.83 (LP) and 7.50 (VH). The relatively high level of heterozygosity and the high MNA found in VH, in spite of the small size of this horse population, are comparable to those found in non endangered breeds, such as Haflinger or Maremmano and are probably due to the recent history of VH characterized by the introduction of stallions of other breeds during the last fifty years. The mean values for F_{ST} is 0.0932 and for F_{IS} is 0.0171. The F_{ST} value obtained indicates that about 9% of the total genetic variation is explained by differences between breeds, with the remaining 91% corresponding to differences among individuals. The F_{IS} calculated for each breed are in agreement with Hardy-Weinberg equilibrium.

Table 1. Sample size (N), observed (H_O) and expected (H_E) heterozygosity (\pm standard error), mean number of allele (MNA) and F_{IS} , calculated for each breed.

Population		N	H_O	H_E	MNA	F_{IS}
Ventasso Horse	VH	117	0.743 \pm 0.087	0.742 \pm 0.075	7.50	0.0032
Anglo-Arab	AA	64	0.721 \pm 0.110	0.720 \pm 0.090	6.50	0.0068
Haflinger	HF	73	0.704 \pm 0.099	0.714 \pm 0.99	6.66	0.0210
Bardigiano	BA	46	0.671 \pm 0.095	0.683 \pm 0.084	5.92	0.0319
Maremmano	MA	50	0.726 \pm 0.117	0.733 \pm 0.085	7.00	0.0201
Murgese	MU	42	0.681 \pm 0.224	0.684 \pm 0.220	6.00	0.0168
Arab	AR	202	0.665 \pm 0.108	0.697 \pm 0.110	6.83	0.0484
Thoroughbred	TB	53	0.670 \pm 0.107	0.704 \pm 0.088	5.83	0.0581
Sanfratellano	SA	15	0.713 \pm 0.171	0.693 \pm 0.138	5.66	0.0055
Italian Heavy Draught Horse	HD	82	0.759 \pm 0.083	0.712 \pm 0.093	6.50	- 0.0499
Italian Trotter	IT	128	0.731 \pm 0.102	0.724 \pm 0.105	7.25	- 0.0048
Lipizzan	LP	36	0.613 \pm 0.088	0.627 \pm 0.060	4.83	0.0387

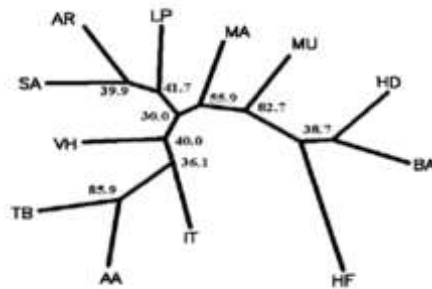
Table 2 shows Nei's standard genetic distances (Ds) among the 12 Italian horse populations. Ds between the VH and the other breed ranged from 0.059 between VH and AA to 0.320 between VH and HF.

Table 2. Nei's standard genetic distances (Ds) among 12 Italian horse populations.

	VH	AA	HF	BA	MA	MU	AR	TB	SA	HD	IT
AA	0.059										
HF	0.320	0.367									
BA	0.311	0.352	0.279								
MA	0.146	0.160	0.242	0.282							
MU	0.217	0.301	0.368	0.364	0.261						
AR	0.214	0.240	0.396	0.511	0.277	0.422					
TB	0.116	0.059	0.453	0.415	0.247	0.381	0.381				
SA	0.234	0.273	0.372	0.523	0.171	0.343	0.255	0.365			
HD	0.315	0.447	0.281	0.273	0.280	0.368	0.438	0.506	0.404		
IT	0.208	0.219	0.408	0.344	0.272	0.408	0.380	0.269	0.404	0.435	
LP	0.190	0.210	0.558	0.463	0.289	0.402	0.307	0.314	0.302	0.426	0.431

The smallest genetic distance observed was between VH and AA; this result was expected because of the recent use of AA stallions for mating. The introduction of TB, MA and LP stallions in the past decades could explain the small genetic distance between VH and these three breeds.

Figure 1. Neighbour-joining tree of the analysed breeds obtained using Ds.



The research was supported by ex 60% funds, Province of Reggio Emilia, Fondazione Manodori of Reggio Emilia. The phylogenetic tree shows the relationship among the horse population (Figure 1). The 12 breed are grouped in two main clusters (supported by a bootstrap value of 82.7%): one includes HD, BA and HF breeds, the other includes the remaining 9 breeds. In this second group VH form a separate cluster with IT, AA and TB, but with lower bootstrap scores.

Comparative analysis of genetic structure and genetic distance between VH and 11 other horse breed shows consistency with the recent history of VH, characterized by the mating with stallions of other breeds in the last fifty years. This practice has been useful to avoid excessive inbreeding but could be prejudicial to the conservation of genetic uniformity and identity of this small endangered Italian native population.

REFERENCES – Aberle, K.S., Hamman H., Drogemuller C., Distl O., 2004. Genetic diversity in German draught horse breeds compared with a group of primitive, riding and wild horses by means of microsatellite DNA markers. *Anim. Genet.* 35:270-7. **Belkhir**, K., Borsa, P., Chikhi, *et al.*, 2002. GENETIX, Version 4.03 logiciel sous WindowsTM pour la genetique des populations. Université Montpellier II, Montpellier, France. **Bjornstad**, G., Gunby, E., Roed, K.H., 2000. Genetic structure of Norwegian horse breeds. *J. Anim. Breed. Genet.* 117:307-17. **FAO – UNEP**, 2000. World watch list for domestic animal diversity. 3rd Edition. **Felsenstein**, J., 1989. PHYLIP (phylogeny inference package), version 3.65. Seattle: University of Washington. **Marletta**, D., Tupac-Yupanqui, I., Bordonaro, S., *et al.*, 2006. Analysis of genetic diversity and the determination of relationships among western Mediterranean horse breeds using microsatellite markers. *J. Anim. Breed. Genet.* 123:315-25. **Nei**, M., 1972. Genetic distances between populations. *Am. Nat.* 106:283-92. **Raymond**, M., Rousset, F., 1995. GENEPOP (version 1.2): population genetics software for exact tests and ecumenicism. *J. Hered.* 86:248-9. **Solis**, A., Jugo, B.M., Merieaux J.C., *et al.*, 2005. Genetic diversity within and among four south European native horse breeds based on microsatellite DNA analysis: implication for conservation. *J. Hered.* 96:670-8. **Weir**, B.S., Cockeram, C.C., 1984. Estimating F-statistics for the analysis of population structure. *Evolution* 38:1358-70.