

EFFICIENCY OF SOME SERUM PROTEIN SYSTEMS IN PARENTAGE CONTROL IN YUGOSLAV TROTTER HORSES

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A total of 85 blood samples, obtained from Yugoslav trotter horses were analysed for serum protein polymorphism at the following loci: albumin (Al), protease inhibitor (Pi), transferrin (Tf), esterase (Es) and Xk protein by standard starch gel electrophoretic procedures. From the results obtained the homogeneity index and parentage exclusion probability were calculated.

The characteristic gene frequencies of the investigated Al, Pi, Tf, Es and Xk protein systems were established as: AIA and AIB (0.424 and 0.576); PiF, PiL, PiG, Pil, PiV and PiS (0.135, 0.318, 0.123, 0.100, 0.259 and 0.576); TiD, TiF, TiH and TiO (0.359, 0.529, 0.036 and 0.076), EsF, EsI and EsS (0.265, 0.570 and 0.165); and XkK and XkS (0.912 and 0.088), respectively. The Homogeneity index of the tested population was 0.0049, 0.5755, 0.2209, 0.1336 and 0.6790 for the Al, Pi, Tf, Es and Xk, loci, respectively. The joint paternity exclusion probability was 83.40% for the population of Yugoslav trotters.

Key words: horse, protein polymorphism, parentage control

INTRODUCTION

All breeding groups of horses show a high degree of variability, which is visual. The biological significance of the wealth of horse polymorphism is presently unknown, and the factors which maintain this phenomenon are still unclear (Nei, 1975). In the last few hundred years, breed societies throughout the world have defined characteristics of the breeds and established studbooks in an attempt to preserve a unique collection of traits within each population.

Scientific researchers, breeders, and breeding societies express a constant interest in improving knowledge about the extent of variation in all defined breeding groups of horses (Braend, 1972; Trommershausen Bowling and Clark, 1985; Bengtsson et al., 1968.). Blood protein systems can be applied as genetic markers for the characterization of the breed, in the evaluation of the origin of breeds and species, gene mapping, identification and parentage control. The distribution of allelic genes can be used as a screen for checking various selec-

tive programs that can influence the gene pool in a population (Weitkamp et al., 1988).

The genetic variability of some local pig, cattle, sheep and goat breeds have been evaluated in our country (Jovanović, 1982; 1988; Reljić-Savić, 1989), but the protein polymorphism of the horse has been neglected so far. The increasing need for precise characterization and the demand for scientific verification of parentage led to this evaluation of protein polymorphism in the Yugoslav trotter. Therefore the polymorphism at five serum protein loci: albumin (Al), protease inhibitor (Pi), Xk protein (Xk), transferrin (Tf) and esterase (Es), was determined and the gene frequencies for the separate loci were calculated. The results obtained were applied in parentage testing, and the probability of false paternity exclusion was calculated.

All the tested proteins can be considered as closed codominant systems, with each phenotype defining a unique genotype. However, silent alleles with little or no detectable protein product have been reported in the Tf and Es systems (Scott, 1970; Patterson and Bell., 1987). Silent alleles behave as recessives to those with detectable protein products. Since these recessive genes were rarely reported in the literature, it is generally assumed that they can be applied in parentage control without limits. At this stage of evaluation there is no evidence either for or against the presence of recessive Es and Tf genes in the population of the Yugoslav trotter horse.

MATERIAL AND METHODS

Blood samples, collected from 11 stallions, 30 mares and 44 foals and fillies of the Yugoslav trotter breed, forming 17 complete and 17 incomplete families, were tested for serum protein polymorphism. All 85 samples were obtained from animals over 6 months of age.

The Yugoslav trotter is a local horse breed with the disposition to perform a single gait (–) racing trot. The origin of the breed can be traced to local light harness mares whose racing ability was developed by crossing with the Orlov trotter and Standardbred horse. Although this breed has been selected from cross-bred origins for highly heritable traits, the extent of genetic polymorphism within the population could not be predicted. Therefore, evaluation of polymorphism at five serum protein loci: albumin (Al), protease inhibitor (Pi), Xk protein (Xk), transferrin (Tf) and esterase (ES), has been performed, and the gene frequencies for separate loci calculated.

Standard methods of starch gel electrophoresis were used to identify inherited variants at the following protein and enzyme loci: Al, Pi, Xk, Tf and Es. Albumin variants were typed according to the method described by Gahne (1966), while Pi and Xk were simultaneously typed as described by Braend (1970). Tf variants were detected in a discontinuous system of buffers (Gahne, 1966). After completing the electrophoresis, Pi/Xk and Tf gels were horizontally sliced into two slices, one of which was stained in amido black solution and the other one was differentially stained for determination of esterase phenotypes by Fast Garnet GBC salt (Scott, 1970, Trommershausen-Smith and Suzuki, 1978).

Allelic frequencies were estimated by direct counting from the phenotypes and the homogeneity index (HI) was calculated. Paternity exclusion probability (PE) was established according to Jamieson and Wiener (Avers, 1984).

RESULTS AND DISCUSSION

As presented in Table 1, evaluation by starch gel electrophoresis revealed the presence of three albumin phenotypes controlled by two codominant alleles AIA and AIB (Sandberg, 1972). The products of six (from a total of at least 18) alleles Pi^F, Pi^L, Pi^G, Pi^L, Pi^U and Pi^S have been identified in the protease inhibitor zone, while two of three electrophoretic variants controlled by the K and S genes were simultaneously revealed in the Xk zone of the electrophoresograms. Four of 11 genes in the transferrin locus (Ti^D, Ti^F, Ti^H and Ti^O) were segregated, while products of the Es^F, Es^I and Es^S (three of 6) alleles were identified in the tested population of Yugoslav trotter horses. The observed and expected phenotypes (data not presented) showed that the population of Yugoslav trotters was not in genetic equilibrium at Pi and Al loci. The observed disequilibrium could be the consequence of constant (–) cross matings with imported stallions of different origin (Bowling and Clark, 1988).

Table 1. – Gene frequencies of five serum protein systems in the Yugoslav trotter

Locus	Allele	Frequency
Al	Al ^A Al ^B	0.424 0.576
Pi	Pi ^F Pi ^L Pi ^G Pi ^L Pi ^U Pi ^S	0.135 0.318 0.123 0.100 0.259 0.065
Xk	Xk ^K Hk ^S	0.912 0.088
Tf	Tf ^O Tf ^F Tf ^H Tf ^O	0.359 0.529 0.036 0.076
Es	Es ^F Es ^I Es ^S	0.265 0.570 0.165

The homogeneity index (HI) was calculated from the number and frequencies of alleles identified at each loci. Homogeneity indexes for the separate loci in the Yugoslav trotter are presented in table 2.

Table 2. – Homogeneity indexes in the Yugoslav trotter

System	Al	Pi	Xk	Tf	Es
No of alleles	2	6	2	4	3
HI	0.0049	0.5755	0.6790	0.2209	0.1336

The most heterogeneous system tested was Al, then came Es and Tf, while Xk protein showed extreme homogeneity in the Yugoslav trotter breed. Since a total of 17 allelic genes and a high degree of heterogeneity were detected in the Yugoslav trotter horse, the effectiveness of the 5 serum genetic markers in parentage exclusion was expected to be high (Grobet and Hanset, 1993). The parentage exclusion probability is presented in table 3.

Table 3. — Parentage exclusion probability obtained by use of the five serum protein systems in the Yugoslav trotter

		System			Joined probability
Al	Pi	XkK	Tf	Es	
0.1846	0.5834	0.0159	0.3079	0.3084	0.8340

The Pi system, alone could theoretically solve 58.34% of the doubtful paternity cases in the Yugoslav trotter. The esterase and Tf locus were also very effective, while the Xk locus could solve only 1.59% of the cases. The biallelic Al system could be effective in 18.46% of the cases only because of the high heterogeneity within the locus. All five loci together could theoretically solve 83.40% of the incorrect paternity cases. The established parentage exclusion probability in the Yugoslav trotter was within the range from 82.0% to 91.0% which has been theoretically estimated for warmblooded horse breeds (Trommershausen Bowling and Clark, 1985; Meriaux, 1992). The obtained results showed that five tested loci, taken together as a battery test, could be efficiently applied for parentage control in the Yugoslav trotter horse.

According to data from the literature the effectiveness of parentage control obtained for 8 to 11 protein polymorphism loci was around 95% in the tested breeds of warm-blooded horses (Trommershausen Bowling et al., 1985; Meriaux, 1992). If a similar degree of polymorphism existed in the Yugoslav trotter, application of the additional protein loci would enable very efficient pedigree control.

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EFIKASNOST PRIMENE NEKIH POLIMORFNIH PROTEINA U KONTROLI RODITELJSTVA KOD JUGOSLOVENSKOG KASAČA

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SADRŽAJ

Polimorfizam serumskih proteina: albumina (Al), inhibitora proteazne aktivnosti (Pi), transferina (Tf), esteraze (Es) i Xk proteina (Xk) je analiziran standardnim metodama elektroforeze na skrobnom gelu, tako da je ukupno ispitano 85 uzoraka krvi, dobijene od konja rase jugoslovenski kasač. Na osnovu dobijenih rezultata izračunat je indeks homogenosti, kao i verovatnoća isključenja pogrešnog roditeljstva.

Utvrdene su karakteristične frekvence gena u ispitanim Al, Pi, Tf, Es i Xk sistemima proteina: AIA i AIB (0.424 i 0.576), PiF, PiL, PiG, PiI, PiU, PiS (0.135, 0.318, 0.123, 0.100, 0.259 i 0.576); TiO, TiF, TiH i TiO (0.359, 0.529, 0.036 i 0.076), EsF, EsI i EsS (0.265, 0.570 i 0.165); kao i XkK i XkS (0.912 i 0.088), prema redosledu, Indeks homogenosti u testiranoj populaciji iznosio je 0.0049, 0.5755, 0.2209, 0.1336 i 0.6790 za AL, Pi, Tf, Es and Xk lokus, po redosledu. U populaciji jugoslovenskog kasača, verovatnoća isključenja pogrešnog roditeljstva iznosila je 83.40%.