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Quantitative inheritance of the coat greying process in horse

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Abstract

The quantitative inheritance of grey level was analyzed in 706 Lipizzan horses born in five state studs (Austria, Croatia, Hungary, Slovakia and Slovenia). A total of 1191 measurements (one to four per horse) of coat grey level, defined as L* parameter of the CIE L*a*b system, were taken by Minolta Chromameter CR210 during a period of four years (2000-2003). After analysis of the greying dynamics (the four parameter Richards growth equation provided the best fit) and variance heterogeneity, three data sets were formed; a) horses younger than seven years (377), b) horses older than six years (352) and c) all horses (706). On all three data sets we estimated (co)variance components by REML animal models with repeated records. The estimated heritabilities were 0.790±0.087 (young horses), 0.579±0.029 (old horses) and 0.494±0.055 (all horse) while the estimated permanent environmental effect was 0.107±0.083 (young horses), 0.000±0.000 (old horses) and 0.345±0.050 (all horses), respectively. The obtained estimates demonstrate that the speed and level of greying involve a very large heritable component. Genetic relationships with melanoma and vitiligo are investigated.

Introduction

The inheritance of coat colour in horses has been always studied from a qualitative (Mendelian) view (Sponenberg 1996; Bowling 2000) where phenotypes are defined by distinctive categories (bay, black, brown, etc.) and are controlled by few genes showing epistatic inheritance (Rieder et al. 2001). The change of coat colour in which the "dark (non-grey)" colour present in foals is progressively replaced by grey is a known phenomenon in horses. A similar process is present in humans. The grey coat colour is inherited as qualitative trait controlled by a single grey gene whose mode of inheritance is dominant. Although it has been assigned to the chromosome 25 (Henner et al. 2002, Locke et al. 2002 and Swinburne 2002), the grey gene is still not determined. Inheritance of the change in grey level over time and the variability of grey level within grey horses may not be covered by the qualitative genetics approach. Thus, the aim of this study was to quantify the inheritance and dynamics of the coat greying process in horses.

Material and Methods

Grey Lipizzan horses (706) were repeatedly measured (1191 records, up to four per horse) in five national studs (Djakovo – Croatia; 89 horses, Piber – Austria; 310 horses, Szilvesvarad – Hungary; 77 horses, Topol'cianky – Slovakia; 82 horses and Lipica - Slovenia; 148 horses) during a period of four years (2000-2003).
To deduce the grey level, coat colour was measured on four places (neck, shoulder, belly and croup) for each horse with a Minolta Chromameter CR210 using the CIE L*a*b* colour system where colour is quantified according to three axes: white-black (L*), red-green (a*) and yellow-blue (b*). As our

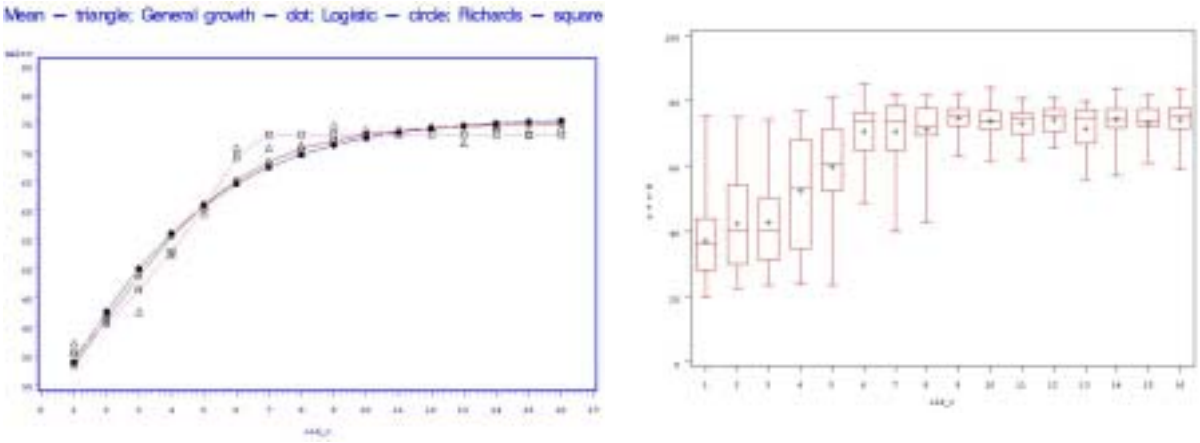
intention was to quantify the grey level, this analysis was related only to the parameter L* (higher L* values correspond to a more "white" - grey coat colour).
The greying dynamics was analyzed by fitting nonlinear functions to individual records or mean values of age groups (in years, except for year 16 which included all older horses). The following nonlinear functions were applied: "general growth" [grey level = a*(1-exp(-k*old_c)/s)**s], "Brody" [grey level = a*(1-s*exp(-k*old_c))], "Logistic" [grey level = a/(1+s*exp(-k*old_c))], "Gomperz" [grey level = a*exp(-exp(-k*old_c))], "Bertalanffy" [grey level = a*(1-(exp(-k*old_c)/3))**3] and "Richards" [grey level = a/((1+s*exp(-k*old_c))**m)] where a = asymptote, s = shape coefficient, k = growth rate; m = parameter defining relative position of the inflection point. Fitting was performed by the NONLIN procedure of the SAS/STAT module (SAS Institute, Cary, 1999-2001) while graphical presentations were done with the same software using the BOXPLOT and GPLOT procedures from the SAS/STAT and SAS/GRAPH module, respectively.
After analysis of the greying dynamics and variance heterogeneity (see Figure 1), three data sets were formed; a) horses younger than seven years (377), b) horses older than six years (352) and c) all horses (706). On all three data sets we estimated (co)variance components and their ratios (heritability, repeatability and genetic correlations) with the REML VCE package Version 4.2.5., which optimises the log likelihood by analytical gradients for covariance matrices of different sizes (Groeneveld 1998). Univariate (mean grey level as dependent variable) and multivariate (grey level of the neck, shoulder, belly and croup as dependent variables) animal models with repeated measurements were applied. Thus, in all models phenotypic variance (V_P) was partitioned into additive (V_A), permanent environment (V_{PE}) and residual (V_R) variance. Fixed effects in the models depended on the data set analysed. For the data set with horses older than six years only a stud - sex - year of measurement (20 classes) effect was used as fixed effect. In the other data sets linear, quadratic and cubic terms of age at measurement (16 classes corresponding to the age at measurement in years except for class 16 which consisted of all horses older than 15 years) were included in addition to the stud - sex - year of measurement effect. The corresponding pedigree file consisted of 4560 horses in which most of the founding horses were born in the 18th and early 19th centuries and some pedigrees were up to 32 generations long.

Results and discussion

For individual values as well as for mean values, the best fit was, according to the coefficient of determination (R^2), obtained with the Richards "growth" function, see Table 1 and Figure 1. In the Richards function the asymptotic value was reached at L* value 73.34 (individual values) and 73.05 (mean values), approximately at the age of seven years. Individual variation of the grey level presented in Figure 1b graphically also demonstrates heterogeneity of variances in young versus old horses.

Table 1. Parameters obtained by non-linear fitting of the grey level in Lipizzan horses by different non-linear functions (R^2 = coefficient of determination)

Function	Individual grey level					Mean grey level				
Parameters	a	s	k	m	R^2	a	s	k	m	R^2
General growth	97.42	$.71*10^8$	.11	.	.55	75.93	4.24	.30	.	.94
Brody	77.85	0.70	.22	.	.61	76.59	.73	.25	.	.93
Logistic	75.78	1.79	.39	.	.62	74.98	1.92	.42	.	.95
Gomperz	76.78	.	.28	.	.62	76.00	.	.29	.	.93
Bertalanffy	76.65	.	.29	.	.62	75.85	.	.31	.	.93
Richards	73.34	$1.82*10^8$	2.90	.045	.64	73.05	6.01	26.11	0.005	.98



a b

Figure 1. Greying dynamics of L* (mmltv) through aging (old_c) of Lipizzan horses;
a) Comparison of mean grey levels with values fitted by "General growth", "Logistic" and "Richards" function
b) Box-plot presentation of the individual grey levels.

Table 2. Estimated genetic parameters with standard errors for the grey level (mean of four points of measurement).

Data	Horses	Records	h^2	V_{PE} / V_P	R
Young	377	559	.79 ±.09	.11 ±.08	.90
Old	352	632	.58 ±.03	.00 ±.00	.58
All	706	1191	.49 ±.06	.35 ±.05	.84

R = repeatability = $(V_A + V_{PE}) / V_P$

Table 3. Estimated genetic parameters with standard errors for the grey level of different body parts.

Data	Trait	Horses	Records	h^2	V_{PE} / V_P	R
Young	Neck	377	559	.65 ±.04	.19 ±.04	.84
	Shoulder	377	559	.75 ±.04	.11 ±.04	.86
	Belly	377	559	.80 ±.03	.07 ±.02	.88
	Croup	377	557	.77 ±.04	.11 ±.04	.88
Old	Neck	352	632	.39 ±.07	.09 ±.04	.49
	Shoulder	352	632	.30 ±.07	.21 ±.07	.50
	Belly	352	632	.47 ±.04	.06 ±.02	.53
	Croup	352	632	.55 ±.04	.06 ±.02	.61
All	Neck	706	1190	.37 ±.05	.42 ±.04	.78
	Shoulder	706	1191	.42 ±.05	.37 ±.04	.79
	Belly	706	1191	.53 ±.05	.29 ±.04	.81
	Croup	706	1188	.51 ±.04	.31 ±.04	.81

R = repeatability = $(V_A + V_{PE}) / V_P$

Estimated genetic parameters (heritability, ratio of the permanent environment and phenotypic variance and repeatability) obtained from univariate and multivariate models are shown in Tables 2 and 3, respectively. In both, univariate and multivariate models the estimated heritabilities and repeatabilities were "high" for data with old horses (>= seven years old) and "very high" for data with young horses (< seven years old). Higher heritability estimates were obtained for belly and croup than for neck and shoulder. One possible explanation is that differences in neck and shoulder building

(surface measured is less even) did cause extra variability which went to permanent environment. The difference in the grey level (L*) between horses that are homozygous (GG) and horses that are heterozygous (Gg) for the grey gene is still not known (Sponenberg 1996). As the grey gene is still undetermined, although assigned to chromosome 25 (Henner et al. 2002, Locke et al. 2002 and Swinburne 2002), we do not know to what extent the obtained heritability estimates are influenced by the grey gene as well as if there is any non-linear difference in progressive greying between homozygous (GG) and heterozygous (Gg) horses.
The quantitative approach to the inheritance of the greying process happens to be useful in research related to the inheritance of melanoma and vitiligo (Curik et al. 2002 and Sölkner et al. 2004) in grey horses. On the other side the approach opens new perspectives for research to the gerontobiology of the coat (hair) pigmentation in mammals. In both cases, results are likely to be, as an animal model, transferable to some extent to humans.

Conclusions

The large phenotypic variability of level of Grey in one to six year old horses is well known. The estimates of genetic parameters (heritability, ratio of the permanent environment and phenotypic variance and repeatability) obtained in this study indicate an unusually strong heritable component of the grey level (L*). Further research will be related to the relationship with the grey gene as well as to the relationship of grey level with melanoma and vitiligo.

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