Acta Theriologica, Suppl. 3: 199–205, 1995.
PL ISSN 0001–7051

Genetic determination of cervid antlers in relation to their significance in social interactions

Günther B. HARTL, Marco APOLLONIO and Luca MATTIOLI

Hartl G. B., Apollonio M. and Mattioli L. 1995. Genetic determination of cervid antlers in relation to their significance in social interactions. [In: Ecological genetics in mammals II. G. B. Hartl and J. Markowski, eds]. Acta Theriologica, Suppl. 3: 199–205.

Antler traits (length of the main beam, circumference of the main beam, coronet circumference, and the number of antler points) in roe deer *Capreolus capreolus* (Linnaeus, 1758) from a population in Casentino (Northern Tuscany) were examined for associations with genotypes at loci coding for enzymes. Significant associations were found only in yearlings. Individuals homozygous for the allele Mpi^{120} had significantly larger antlers than carriers of other genotypes at *Mpi*. Individuals homozygous for $Pep\text{-}2^{100}$ had significantly smaller antlers than carriers of other genotypes at *Pep-2*. In adults the results were essentially the same, but only as a statistically insignificant trend. The data were interpreted in terms of one or more genetic components having a major influence on antler development only in yearlings. This corresponds with behavioural data suggesting that in adults antler size is not related to reproductive success, but in yearlings it is correlated with sexual maturity and the rate of being expelled from the home range of their mothers by territory holders. The situation in the roe deer was compared with previous results on the genetics of antler development in the red deer, where males are social rather than solitary, yearlings do not participate in reproduction, and some antler characteristics are related to reproductive success of adult stags.

Institut für Haustierkunde der Christian-Albrechts-Universität zu Kiel, Olshausenstraße 40, D-24118 Kiel, Germany (GBH); Dipartimento Scienze del Comportamento Animale e dell'Uomo, Università di Pisa, Via A. Volta 6, 56126 Pisa, Italy (MA); DREAM, Settore Fauna, Via Roma 167, Ponte a Poppi (Ar), Italy (LM)

Key words: *Capreolus capreolus*, *Cervus elaphus*, social interactions, antler development, allozyme electrophoresis, biochemical markers

Introduction

Antlers are the most obvious secondary sexual traits in most male cervids. Their significance in social interactions has been found to be chiefly related to inter-male competition for females (cf Clutton-Brock *et al.* 1980, Clutton-Brock 1982). On the one hand, antler size may be positively correlated with reproductive success of a male (eg Clutton-Brock *et al.* 1988). On the other hand, depending on the species, the annual rebuilding of antlers may require a considerable expenditure of energy. Taking into account the potential benefit of relatively large antlers in terms of increased reproductive success and their potential cost in terms

of increased metabolic effort, it would not be surprising to find some genetic mechanism responsible for balancing these components under variable environmental conditions. The genetic basis of antler development can be expected to differ from species to species, depending on the actual importance of male antlers for social interactions and ultimately for reproductive success, and may be elucidated by using molecular markers (eg Scribner and Smith 1990, Hartl *et al.* 1991a, 1995).

In the present study relationships between allozyme variation and antler development are examined in various age classes of roe deer *Capreolus capreolus* (Linnaeus, 1758) and compared with data on red deer *Cervus elaphus* Linnaeus, 1758 published by Hartl *et al.* (1991a, 1995). These two species show remarkable differences in social organization and male antler development. Thus, different patterns as to relationships between allozymes and antler traits may be expected.

Material and methods

Liver and kidney samples of 46 male and 72 female roe deer came from the Province of Arezzo in Northern Tuscany. This population is probably autochthonous and stems from the remaining nucleus of the Foreste Casentinensi (Lorenzini *et al.* 1993). All animals were shot during the hunting season of 1993. A total of 12 loci found polymorphic in European roe deer in previous investigations were examined for electrophoretic variation (cf Hartl *et al.* 1991b). Age determination was performed on the basis of dentition. Body weights of eviscerated specimens were measured immediately after shooting using a spring-balance. Mandible length served as an indicator of body size. As far as available, antler dimensions (number of antler points – NAP, length of the main beam – LMB, circumference of the main beam measured 1 cm above the coronet – CMB, and coronet circumference – CC) in the males studied electrophoretically were measured on skulls collected for trophy records.

Alleles were designated according to the relative mobility of the corresponding allozymes. All alleles found in the present study had been already defined by Hartl and Reimoser (1988) and Hartl *et al.* (1991b). Genotypic proportions were tested for agreement with Hardy-Weinberg expectations using the BIOSYS-1 programme package of Swofford and Selander (1989; release 1.7). Heterozygosity classes in yearlings and adults of both sexes were formed by grouping the specimens according to the number of heterozygous loci per individual). Standard statistical analyses were performed using SPSS for Windows (release 5.0).

Separately for yearlings and adults in each sex, differences in body weight and mandible length among heterozygosity classes were examined by the Kruskal-Wallis test. The same statistical procedure was used for examining antler traits in yearlings and adults (NAP, LMB, CMB, CC – measurements of both beams added) as to differences among heterozygosity classes. Antler traits were checked visually for differences among genotypic classes at *Pep-2* and *Mpi* using boxplots. Apparent differences were tested for statistical significance by *t*-test and Kruskal-Wallis test, respectively.

Results

Electrophoretic polymorphism was detected at *Dia-2*, *Ak-1*, *Pgm-1*, *Pgm-2*, *Acp-1*, *Pep-2*, *Mpi*, and *Gpi-1*. Allelic frequencies, observed (h_o) and expected (h_e) heterozygosities are given in Table 1. At none of the polymorphic loci genotypes were sex or age dependent (cross-tabs and chi square test). Both in males and in

Table 1. Allelic frequencies and heterozygosity at 8 polymorphic loci in male and female roe deer from Casentino. n – sample size, $h_o(h_e)$ – observed (expected) heterozygosity. Allelic frequencies were not significantly different between sexes (χ^2-test of Workman and Niswander).

Locus	Allele	Males	n	$h_o(h_e)$	Females	n	$h_o(h_e)$
Dia-2	*100*	0.924	46	0.065(0.141)	0.924	72	0.125(0.141)
	118	0.076			0.076		
Ak-1	*100*	0.054	46	0.109(0.103)	0.042	72	0.056(0.080)
	250	0.946			0.958		
Pgm-1	*100*	1.0	46	0.0(0.0)	0.993	72	0.014(0.014)
	–16	0.0			0.007		
Pgm-2	*100*	0.913	46	0.130(0.159)	0.887	71	0.169(0.200)
	70	0.087			0.113		
Acp-1	*100*	0.989	46	0.022(0.022)	0.993	72	0.014(0.014)
	200	0.011			0.007		
Pep-2	*100*	0.656	45	0.467(0.509)	0.529	69	0.377(0.609)
	115	0.200			0.254		
	107	0.144			0.217		
Mpi	*100*	0.685	46	0.239(0.432)	0.771	72	0.208(0.353)
	120	0.315			0.229		
Gpi-1	*100*	1.0	46	0.0(0.0)	0.993	72	0.014(0.014)
	500	0.0			0.007		

Table 2. Observed and expected genotypes at *Pep-2* and *Mpi* in male and female roe deer from Casentino. p-values in parentheses are from χ^2-tests with pooling of heterozygous genotypes and of homozygous genotypes, respectively, for rare alleles (cf Swofford and Selander 1989).

Locus	Geno-type	Males					Females				
		Observed frequency	Expected frequency	χ^2	df	p	Observed frequency	Excepted frequency	χ^2	df	p
Pep-2	*100/100*	19	19.225				25	19.182			
	100/115	14	11.933				14	18.650			
	100/107	7	8.618				9	15.985			
	115/115	2	1.719				9	4.343			
	107/115	0	2.629				3	7.664			
	107/107	3	0.876				9	3.175			
				8.485	3	0.037 (0.022)			24.494	3	<0.001 (0.005)
Mpi	*100/100*	26	21.462				48	42.692			
	100/120	11	20.077				15	25.615			
	120/120	9	4.462				9	3.692			
				9.680	1	0.002			12.689	1	<0.001

females, genotypic proportions at the loci *Pep-2* and *Mpi* deviated significantly from Hardy-Weinberg expectations (Table 2).

Table 3. Differences in antler dimensions (LMB – length of the main beam, CMB – circumference of the main beam, CC – coronet circumference) in carriers of different genotypes at *Mpi*. *n* – sample size, ns – statistically not significant.

	Yearlings			Adults		
	$Mpi^{non120/120}$	$Mpi^{120/120}$	*p* (*t*-test)	$Mpi^{non120/120}$	$Mpi^{120/120}$	*p* (*t*-test)
LMB	15.9 (*n*=13)	30.3 (*n*=2)	0.001	43.6 (*n*=13)	46.8 (*n*=4)	ns
CMB	8.8 (*n*=13)	11.7 (*n*=2)	0.077	14.2 (*n*=11)	14.9 (*n*=4)	ns
CC	10.8 (*n*=13)	17.0 (*n*=2)	0.000	24.0 (*n*=11)	26.9 (*n*=4)	ns

Antler traits (NAP, LMB, CMB, CC) were highly correlated in yearlings (r = 0.67–0.98, p = 0.007–0.000, n = 15), and somewhat less so in adults (r = 0.40–0.80, p = ns–0.000, n = 15). Mandible length was significantly correlated with body weight in yearlings (r = 0.67, p = 0.034, n = 10) but not in adults. In both age classes, antler traits were not correlated with either body weight or mandible length. In both sexes and age classes, body weight and mandible length were not related to individual heterozygosity. The same was the case for antler traits in males. Both for *Pep-2* and *Mpi* associations between genotypes and the development of antler traits became apparent. In yearlings, homozygous carriers of Mpi^{120} had significantly larger antlers than homo- or heterozygous carriers of Mpi^{100}. In adults, there was a statistically insignificant trend in the same direction (Table 3). Based on the same grouping of genotypes ($Mpi^{non120/120}$ vs $Mpi^{120/120}$), a two-way ANOVA revealed a significant influence of both age ($p < 0.001$ in all cases) and *Mpi* genotypes ($p_{LMB} = 0.05$, $p_{CC} = 0.031$) on antler dimensions. In yearlings, homozygous carriers of $Pep\text{-}2^{100}$ had significantly smaller antlers than carriers of any other genotype. There was the same trend in adults, but except for CMB, again statistically insignificant (Table 4). Based on the same grouping of genotypes ($Pep\text{-}2^{non100/100}$ vs $Pep\text{-}2^{100/100}$), a two-way ANOVA revealed a significant influence of both age ($p < 0.001$ in all cases) and *Pep-2* genotypes ($p_{LMB} = 0.015$, $p_{CMB} = 0.001$, $p_{CC} = 0.031$) on antler dimensions.

Table 4. Differences in antler dimensions (NAP – number of antler points, LMB – length of the main beam, CMB – circumference of the main beam, CC – coronet circumference) in carriers of different genotypes at *Pep-2*. *n* – sample size, ns – statistically not significant.

	Yearlings			Adults		
	$Pep\text{-}2^{non100/100}$	$Pep\text{-}2^{100/100}$	*p* (*t*-test)	$Pep\text{-}2^{non100/100}$	$Pep\text{-}2^{100/100}$	*p* (*t*-test)
NAP	2.9 (*n*=9)	2.0 (*n*=6)	0.021	6.0 (*n*=10)	5.4 (*n*=7)	ns
LMB	23.4 (*n*=9)	9.5 (*n*=6)	0.027	45.0 (*n*=10)	43.3 (*n*=7)	ns
CMB	10.4 (*n*=9)	7.4 (*n*=6)	0.015	15.1 (*n*=9)	13.4 (*n*=6)	0.019
CC	13.7 (*n*=9)	8.5 (*n*=6)	0.022	26.1 (*n*=9)	22.8 (*n*=6)	ns

Discussion

There are substantial differences between roe and red deer as to their respective social organization and reproductive behaviour. Roe deer males are generally solitary and often territorial from March to September (Kurt 1991). They do not form bachelor groups and yearlings may participate in reproduction. In mature males, access to females is a function of posessing a territory rather than of antler size and physical strength (Kurt 1991). Caused by the aggression of territory holders against possible competitors in reproduction, the dispersal of young males from the home ranges of their mothers may take place in their first year of life (Strandgaard 1972, Ellenberg 1978). It is the large-antlered yearlings, which are most frequently attacked and most likely to disperse (Wahlström, in press). Indeed antler size in yearlings was found to be positively correlated with the development of testes (Wahlström, in press), suggesting a relationship with sexual maturity (Møller 1989).

On the contrary, red deer males are generally social, and are often associated in bachelor gropus outside the rutting period. Their reproductive success may be correlated with the dominance rank they held in social groups in the previous winter (Appleby 1982). Yearlings are excluded from reproduction. The dispersal of young males from the home ranges of their mothers takes place in the second or third year of life and is not due to the aggression of older males. General antler characteristics of adult males may not be directly correlated with dominance or reproductive success, however, antler size, antler weight, and the number of antler points sometimes are. Moreover, these traits reflect general body condition and are often correlated with body weight, which is strongly linked to dominance and reproductive success (Clutton-Brock *et al.* 1979, Gibson and Guinness 1980, Appleby 1982, Clutton-Brock 1982).

In both species, these considerations are reflected by the data on relationships between allozymes and antler development. In roe deer, one or more marked genetic components seem to be responsible for differences in antler dimensions only among yearlings, and are chromosomally or functionally linked to *Mpi* and *Pep-2*. In contrast to many other allozyme polymorphisms in roe deer, those at *Mpi* and *Pep-2* are present for the same major alleles in virtually all populations examined so far (cf Hartl *et al.* 1991b, Lorenzini *et al.* 1993). Given the considerable human impact on population structure of large game animals, which may cause dramatic losses of allelic variation by genetic drift (cf Hartl and Pucek 1994, and references therein), the ubiquitous presence of particular alleles at an enzyme locus provides a strong argument in favour of their maintenance through balancing or countervailing selection. The deficiency of heterozygotes at *Mpi* in both sexes may be related to an increased dispersal of large-antlered yearlings. In an expanding population like Casentino (Lorenzini *et al.* 1993), many of these yearlings may have had the opportunity to establish new territories at the margins of its range. In roe deer territories females usually form stable mother clans which,

due to non-random mating, ultimately leads to some genetic substructuring of a population (Kurt *et al.* 1993). Thus, in some parts of the population, animals homozygous for Mpi^{120} (the otherwise rare allele) may have accumulated with time, and the non-random distribution of genotypes in the population may have become apparent as a Wahlund effect in the random sample of roe deer drawn for our study. At *Pep-2* there was a significant excess of homozygotes as well, but generally the large number of genotypes requires larger sample sizes of individuals to be investigated as to their detailed relationship with antler traits in the presence of different *Mpi* genotypes. At any rate, all of the large-antlered yearlings with $Mpi^{120/120}$ had a *Pep-2* genotype other than $Pep\text{-}2^{100/100}$.

In red deer at least two different components responsible for antler development could be identified (Hartl *et al.* 1991a, 1995). One of them was associated with *Idh-2*, where homozygous carriers of the allele $Idh\text{-}2^{125}$ had a significantly higher number of antler points from the second year of life on. However, the average difference from carriers of all other *Idh-2* genotypes was highest between 2 and 6 years of age, suggesting a more rapid antler development in the respective stags. The second genetic component was associated with *Acp-2*, where, from seven years on, homozygous carriers of $Acp\text{-}2^{100}$ had generally significantly larger antlers (NAP, LMB, CMB, CC) than carriers of the other *Acp-2* genotypes. Interestingly, homozygous carriers of $Acp\text{-}2^{100}$ had smaller antlers than average between 2 and 6 years of age, suggesting that the respective stags are slow in antler development during their first period of life, whereas they grow exceptionally large antlers once they are older. The presence of only three genotypes at each locus allowed us to check the combined effect of the component associated with *Idh-2* and that associated with *Acp-2* on antler development, and we found that both components can compensate or reinforce each other at the various stages of lifetime antler growth. In fact, by playing a role in balancing benefits and costs of male reproductive success, they may be part of a mechanism enabling the rapid adaptation of a population to various environmental and demographic conditions (Hartl *et al.* 1995).

Altogether, the data of the present study as well as those of Hartl *et al.* (1991a, 1995) suggest that the genetic control of antler development in cervids is clearly related to the respective significance of antlers in social interactions.

Acknowledgements: The authors are indebted to the local hunters at Casentino for their help in collecting samples. Part of this work was carried out at the Forschungsinstitut für Wildtierkunde und Ökologie der Veterinärmedizinischen Universität Wien. The excellent technical assistance of Anita Haiden is gratefully acknowledged.

References

Appleby M. C. 1982. The consequences and causes of high social rank in red deer stags. Behaviour 80: 259–273.

Clutton-Brock T. H. 1982. The function of antlers. Behaviour 79: 108–125.

Clutton-Brock T. H., Albon S. D., Gibson R. M. and Guinness F. E. 1979. The logical stag: Adaptive aspects of fighting in red deer (*Cervus elaphus* L.). Animal Behaviour 27: 211–225.

Clutton-Brock T. H., Albon S. D. and Guinness F. E. 1988. Reproductive success in male and female red deer. [In: Reproductive sucess – studies of individual variation in contrasting breeding systems. T. H. Clutton-Brock, ed]. The University of Chicago Press, Chicago and London: 325–343.

Clutton-Brock T. H., Albon S. D. and Harvey P. H. 1980. Antlers, body size and breeding group size in the Cervidae. Nature 285: 565–567.

Ellenberg H. 1978. Zur Populationsökologie des Rehes (*Capreolus capreolus* L., Cervidae) in Mitteleuropa. Spixiana, Suppl. 2: 1–211.

Gibson R. M. and Guinness F. E. 1980. Differential reproductive success in red deer stags. Journal of Animal Ecology 49: 199–208.

Hartl G. B., Klein F., Willing R., Apollonio M. and Lang G. 1995. Allozymes and the genetics of antler development in red deer (*Cervus elaphus*). Journal of Zoology, London 237: 83–100.

Hartl G. B., Lang G., Klein F. and Willing R. 1991a. Relationships between allozymes, heterozygosity and morphological characters in red deer (*Cervus elaphus*), and the influence of selective hunting on allele frequency distributions. Heredity 66: 343–350.

Hartl G. B. and Pucek Z. 1994. Genetic depletion in the European bison (*Bison bonasus*) and the significance of electrophoretic heterozygosity for conservation. Conservation Biology 8: 167–174.

Hartl G. B. and Reimoser F. 1988. Biochemical variation in roe deer (*Capreolus capreolus* L.): are *r*-strategists among deer genetically less variable than *K*-strategists? Heredity 60: 221–227.

Hartl G. B., Reimoser F., Willing R. and Köller J. 1991b. Genetic variability and differentiation in roe deer (*Capreolus capreolus* L.) of Central Europe. Genetics, Selection, Evolution 23: 281–299.

Kurt F. 1991. Das Reh in der Kulturlandschaft – Sozialverhalten und Ökologie eines Anpassers. Paul Parey, Hamburg, Berlin: 1–284.

Kurt F., Hartl G. B. and Völk F. 1993. Breeding strategies and genetic variation in European roe deer *Capreolus capreolus* populations. [In: Ecological genetics in mammals. G. B. Hartl and J. Markowski, eds]. Acta Theriologica 38, Suppl. 2: 187–194.

Lorenzini R., Patalano M., Apollonio M. and Mazzarone V. 1993. Genetic variability of roe deer *Capreolus capreolus* in Italy: electrophoretic survey on populations of different origin. [In: Ecological genetics in mammals. G. B. Hartl and J. Markowski, eds]. Acta Theriologica 38, Suppl. 2: 141–151.

Møller A. P. 1989. Ejaculate quality, testes size and sperm production in mammals. Functional Ecology 3: 125–130.

Scribner K. T. and Smith M. H. 1990. Genetic variability and antler development. [In: Horns, pronghorns, and antlers. G. A. Bubenik and A. B. Bubenik, eds]. Springer, New York, Berlin: 460–473.

Strandgaard H. 1972. The roe deer (*Capreolus capreolus*) population at Kalø and the factors regulating its size. Danish Review of Game Biology 7: 1–205.

Swofford D. L. and Selander R. B. 1989. BIOSYS-1. A computer program for the analysis of allelic variation in population genetics and biochemical systematics. Release 1.7. User's manual. Illinois Natural History Survey, Champaign, Illinois.

Wahlström K. (in press). The significance of male-male aggression for yearling dispersal in roe deer (*Capreolus capreolus*). Behavioral Ecology and Sociobiology.

Received 20 March 1995, accepted 1 September 1995.