

**ROE DEER (*CAPREOLUS CAPREOLUS*)
IN PARMA PROVINCE:
MORPHOLOGICAL CHARACTERIZATION OF ANIMALS
WITH KNOWN MITOCHONDRIAL HAPLOTYP**

**POPOLAZIONE DI CAPRIOLO (*CAPREOLUS CAPREOLUS*)
DELL'APPENNINO PARMENSE:
CARATTERIZZAZIONE MORFOLOGICA DI SOGGETTI
CON APLOTIPO MITOCONDRIALE NOTO**

Soffiantini Chiara Serena¹, Malacarne Massimo¹, Gandolfi Greta², La Fata Isabella², Pisani Giovanni Maria¹, Beretti Valentino¹, Sabbioni Alberto¹.

Key words:

Capreolus capreolus, Parma, body traits, skull measurements, D-loop.

Parole chiave:

Capreolus capreolus, Parma, misure biometriche, misure craniometriche, D-loop.

Abstract

The aim of the study was to characterize biometric features of the population of Parma roe deer (*Capreolus capreolus*) as related to their haplotypic pattern. Fifty-four male roe deer were analyzed for body traits (weight, body length, height at withers, chest circumference, ear length, fore leg length, hind leg length and circumference of neck's base). On 21 animals out of 54, 16 skull measurements were collected as well. The mitochondrial haplotypes of individuals (Central-Europe or Central-Southern Italy group) were known. All morphological measures were analysed with ANOVA considering as fixed factor the D-loop haplotype. No significant differences between the two haplogroups were detectable.

Riassunto

Lo scopo del lavoro è stato di caratterizzare la popolazione di capriolo (*Capreolus capreolus*) di una zona dell'Appennino parmense sotto il profilo morfologico, in relazione all'aplotipo mitocondriale presentato dagli individui considerati. Sono state analizzate le schede biometriche di abbattimento di 54 caprioli

¹ Dipartimento di Produzioni Animali, Biotecnologie Veterinarie, Qualità e Sicurezza degli Alimenti - Università degli Studi di Parma.

² Dipartimento di Biologia Evolutiva – Università degli Studi di Parma.

Indirizzo per corrispondenza - Corresponding Author: Dott.ssa Chiara Serena Soffiantini - Dipartimento di Produzioni Animali, Biotecnologie Veterinarie, Qualità e Sicurezza degli Alimenti – Via del Taglio 8, 43100 Parma – Tel: +39-521-032615 – e-mail: chiaraserena.soffiantini@nemo.unipr.it

maschi, attraverso le seguenti misure morfologiche: lunghezza totale, altezza al garrese, circonferenza toracica, lunghezza orecchio, lunghezza arto anteriore e posteriore e circonferenza base collo. Di 21 dei 54 campioni è stato possibile raccogliere anche 16 misure craniometriche. Di ciascuno degli individui era noto l'aplotipo mitocondriale (D-loop) e quindi l'appartenenza a uno dei due gruppi aplotipici principali: l'aplotipo dell'Europa centrale e l'aplotipo dell'Italia centro-meridionale. Il peso pieno e le misure lineari (corporee e del cranio) sono stati analizzati con ANOVA, considerando l'aplotipo D-loop come fattore fisso. Nessuna differenza significativa a carico delle misure somatiche è stata rilevata fra i due aplogruppi.

Introduction

The roe deer (*Capreolus, Artiodactyla, Cervidae*) is distributed in the Euro-Asiatic continent; it includes European roe deer (*Capreolus capreolus*), distributed in Western Europe, and Siberian roe deer (*Capreolus pygargus*), distributed in Asia and Eastern Europe (Randi *et al.*, 1998).

The European roe deer shows wide morphological variability (Sokolov & Gromov, 1990). The study of body traits of roe deer population of Castelporziano (Rome) (Festa, 1925) and Orsomarso (north of Calabria) (Lehmann, 1973) suggested the presence of a subspecies, defined *C.c. italicus*. The analysis of the control region of mitochondrial DNA (D-Loop mtDNA) substantially confirmed that roe deer populations in Southern Italy ("Italicus", from Castelporziano, Orsomarso, Gargano) are different from other Italian and Central European populations ("Central-European" group) (Randi *et al.* 1998; Vernesi *et al.*, 2002). Furthermore, according to Lorenzini *et al.* (2002), roe deer populations in many Italian areas have to be considered as an admixture of *C.c. italicus* and "Central-European" animals, in consequence of reintroductions, restocking and translocations of allochthonous subjects for hunting purposes. In Parma province as well, roe deer are historically believed to originate from subjects coming from the North of Italy and Eastern Europe. However, Soffiantini *et al.* (2006) found both "Central-European" and "Italicus" mtDNA haplotype in Parma Apennines.

The aim of the study was to characterize biometric features (body traits and skull parameters) of the population of Parma with relation to haplotypic pattern.

Material and Methods

The research was carried out on 54 male roe deer (*Capreolus capreolus*) shot in a Hunting District of Parma (ATC PR 4) during 2005 and 2006.

The animals were characterized for their haplotype by Gandolfi *et al.* (2007) and Gandolfi (2007). Those authors evidenced the presence of haplotypes corresponding to the two main mtDNA haplogroups reported by Randi *et al.* (1998): "Central-European" (West Alps and Central Europe) and "Italicus" (Central and southern Italy), and haplotype frequencies were 23% and 77%, respectively.

Body weight and linear parameters were measured on each animal: body length, height at withers, chest circumference, ear length, fore leg length, hind leg

length and circumference of neck's base (Figure 1). On 21 out 54 male roe deer, skull measurements (Figure 2) were registered with a sliding calliper.

ANOVA univariate GLM (package SPSS, vers.15.0, SPSS Inc., Chicago, IL, US) was employed to check the effect of mitochondrial haplotype on morphology (body weight, linear parameters and skull measurements), according to the following model:

$$y_{ijkl} = \mu + \alpha_i + \beta_j + \gamma_k + \varepsilon_{ijkl}$$

y_{ijkl} = dependent variable

μ = common mean

α_i = effect of factor "class of age" ($i = 0, \dots, 3$)

β_j = effect of factor "area" ($j = 1, \dots, 5$)

γ_k = effect of factor "mitochondrial haplotype" ($k = 1, 2$)

ε_{ijkl} = residual error

The factor "class of age" was divided into 4 levels:

0 = class 0 = 1st -11th month;

1 = class 1 = 12th -23rd month;

2 = class 2 = 2-7 years;

3 = class 3 = over 7 years.

As all the skulls were obtained from adults roe deer (>2 years), for the analysis of skull measurements the factor "class of age" was not included in the GLM model.

The factor "area" was divided into 5 levels according to the number of zones that constitute ATC PR 4.

The factor "mitochondrial haplotype" was divided into 2 levels according to the results of the mitochondrial analysis, performed by Gandolfi *et al.* (2007) and Gandolfi (2007):

1 = "Italicus" haplotype;

2 = "Central-European" haplotype.

Results and Discussion

Tables 1, 2 and 3 show the mean values of body traits and skull measurements of roe deer with "Italicus" and "Central-European" haplotypes. ANOVA univariate did not show any difference ($P > 0.05$) between the two groups, for none of the considered measurements.

Preliminary results of this research contradict the hypothesis of Festa about the possibility to distinguish *C.c. italicus* ("Italicus" haplotype) from *C.c. capreolus* ("Central-European" haplotype) on the basis of body traits.

According to Vernesi *et al.* (2002) and Lorenzini *et al.* (2002), populations of Castelporziano should be genetically similar to populations from Tuscany Maremma (*Capreolus capreolus italicus*), while populations of Eastern Alps should be distinguishable both from Castelporziano+Tuscany Maremma and from Tuscany Apennines populations. Montanaro *et al.* (2003) applying linear discriminant analysis to some skull measures were able to distinguish roe deer living in the area corresponding to Castelporziano and Tuscany Maremma (Siena and Grosseto

provinces) from animals living in eastern Alps or in Tuscany Apennine. Not so clear was the possibility to discriminate those two last populations between themselves. In relation to those observations, *Capreolus capreolus italicus* (Castelporziano+Tuscany Maremma) seems to show differences concerning skull measurements compared with *Capreolus capreolus capreolus*, when animals living in different areas are considered. The present research evaluates skull differences between animals with distinct haplotype (“Italicus” and ”Central-European”), living in the same *habitat* (Parma province, ATC PR 4). Results here observed suggest that, as regards animals living in the same habitat, skull measurements don’t show any difference (Table 3). This could mean that differences found by other authors between Alpine populations and Central Italy populations (Castelporziano+Tuscany Maremma) could partly depend on the habitat and not (or not only) on the haplotype.

On the definition of roe deer morphology, *habitat* seems to play a bigger role than the belonging to a certain haplogroup: concerning animals living in Parma province (ATC PR 4), despite the clear differentiation at mitochondrial DNA (Gandolfi *et al.*, 2007; Gandolfi, 2007), no morphological distinction was found.

In any case the results of the present investigation have to be considered as preliminary: sample dimension should be improved, most of all concerning “Central-European” roe deer. Furthermore, investigation at nuclear level should be carried out in order to evaluate the admixture degree between “Italicus” and “Central-European” population.

References

- 1) Aragon S, Blaza F, San Joe C, Fandos P (1997). *Variation in skull morphology of roe deer (Capreolus capreolus) in Western and Central Europe*, Journal of Mammology, 79: 131-140.
- 2) Douzery E, Randi E (1997). *The mitochondrial control region of Cervidae: evolutionary patterns and phylogenetic content*. Molecular Biology and Evolution, 14: 1154-1166.
- 3) Festa E (1925). *Il capriolo dell’Italia Centrale*. Bollettino del Museo di zoologia dell’Università di Torino, vol. XL, Nuova Serie, N. 37 (Napoli).
- 4) Gandolfi G (2007). *Origini e struttura di popolazioni di capriolo (Capreolus capreolus) dell’Appennino tosco-emiliano, attraverso marcatori mitocondriali e nucleari*. Tesi di laurea specialistica in ecologia, Università degli Studi di Parma.
- 5) Gandolfi G, Soffiantini CS, Malacarne M, Casagrande S, La Fata I, Apollonio M, Tagliavini J (2007). *Caratterizzazione molecolare di Capreolus capreolus Linneo, 1758, dell’Appennino parmense*. Lecce, IX Congresso Nazionale dell’Unione Zoologi Italiani (UZI).
- 6) Lehmann (von) E (1973). *Die Säugetiere der Hochlagen des Monte Caramolo (Lucanischer Apennin, Nordkalabrien)*. Suppl. Ricerche Biol. Selvaggina, Vol. V, N. 4 (Bologna).
- 7) Lorenzini R, Mattioli L, Rustoni M, Patalano M (1996). *Allozyme and cranio-metric variability in the Roe Deer (Capreolus capreolus L.) from Central Italy*.

- Zeitschrift für Säugetierkunde – International Journal of Mammalogy, 61: 7-24.
- 8) Lorenzini R, Lovari S, Masetti M (2002). *The rediscovery of the Italian roe deer: genetic differentiation and management implications*. Italian Journal of Zoology, 69: 367-379.
 - 9) Montanaro P, De Marinis AM, Riga F, Focardi S (2003). *Variabilità craniometrica in alcune popolazioni italiane di capriolo*. Hystrix, Italian Journal of Mammalogy, 14(s.i.): 169-170
 - 10) Randi E, Pierpaoli M, Danilkin A (1998). *Mitochondrial DNA polymorphism in populations of Siberian and European roe deer (Capreolus pygargus and C. capreolus)*. Heredity, 80: 429-437.
 - 11) Randi E, Alvez PC, Carranza J, Milosevic- Zlatanovic S, Sfougaris A, Mucci N (2004). *Phylogeography of Roe Deer (Capreolus capreolus) populations: the effects of historical genetic subdivision and recent nonequilibrium dynamics*. Molecular Ecology, 13: 3071-3083.
 - 12) Soffiantini CS, Pisani GM, Malacarne M, Gandolfi G, Sabbioni A & Tagliavini J (2006). *Genetic characterisation of roe deer (Capreolus capreolus) population of Parma Apennines*, Advances in Deer Biology, Atti del convegno “The 6th International Deer Biology Congress”, 88.
 - 13) Sokolov VE, Gromov VS (1990). *The contemporary ideas on roe deer (Capreolus Gray, 1821) systematization: morphological, ethological and hybridological analysis*. Mammalia, 54: 431-444.
 - 14) Vernesi C, Pecchioli E, Caramelli D, Tiedemann R, Randi E, Bertorelle G (2002). *The genetic structure of natural and reintroduced roe deer (Capreolus capreolus) population in the Alps and Central Italy, with reference to the mitochondrial DNA phylogeography of Europe*. Molecular Ecology, 11: 1285-1297.

Table 1 – Least square mean values of body traits (body weight, body length, height at withers, chest circumference) of roe deer shot in Parma province (ATC PR4): comparison between subjects with “Italicus” haplotype and subjects with “Central-European” (CE) haplotype.

Parameter	Body weight (kg)		Body length (cm)		Height at withers (cm)		Chest circumference (cm)	
	Italicus	CE	Italicus	CE	Italicus	CE	Italicus	CE
mtDNA haplotype*								
No.	41	13	41	13	41	13	41	13
Class of age**:								
1	21.7	24.2	112.4	117.5	67.4	69.0	63.2	64.0
2	25.3	24.2	117.2	119.3	68.7	69.0	67.2	64.3
3	26.0	26.5	123.5	118.2	72.0	67.5	68.0	63.5
ES	0.5	0.7	1.2	1.5	1.3	1.6	1.0	1.2

*mitochondrial DNA (mtDNA) haplotype was characterised analysing the control region (D-loop), Gandolfi *et al.* (2007) and Gandolfi (2007).

** class 1: from 12th to 23rd month; class 2: from 2nd to 7th year; class 3: over 7th year.

Table 2 – Least square mean values of body traits (ear length, fore leg length, hind leg length, circumference of neck's base) of roe deer shot in Parma province (ATC PR4): comparison between subjects with “Italicus” haplotype and subjects with “Central-European” (CE) haplotype.

*mitochondrial DNA (mtDNA) haplotype was characterised analysing the control region (D-

Parameter	Ear length (cm)		Fore leg length (cm)		Hind leg length (cm)		Circumference of neck's base (cm)	
	Italicus	CE	Italicus	CE	Italicus	CE	Italicus	CE
mtDNA haplotype*								
No.	41	13	41	13	41	13	41	13
Class of age**:								
1	14.6	16.5	26.4	27.0	34.7	35.5	33.6	38.0
2	15.8	15.0	26.6	26.7	34.6	34.7	35.3	36.0
3	15.5	16.0	27.0	27.2	37.0	35.2	36.0	38.5
ES	0.4	0.4	0.4	0.5	0.4	0.5	1.0	1.3

loop), Gandolfi *et al.* (2007) and Gandolfi (2007).

** class 1: from 12th to 23rd month; class 2: from 2nd to 7th year; class 3: over 7th year.

Table 3 – Least square mean values of skull measurements of roe deer shot in Parma province (ATC PR4): comparison between subjects with “Italicus” haplotype and subjects with “Central-European” (CE) haplotype.

	Italicus	CE	ES
N.	4	17	
Skull measurements (mm) ¹ :			
A	190.2	185.8	1.4
B	200.0	193.5	1.5
C	75.0	76.4	0.7
D	115.5	115.2	0.7
E	47.5	46.4	0.8
F	21.8	23.9	0.8
G	22.5	23.9	0.7
H	61.8	61.2	0.5
I	92.8	92.6	0.8
J	25.0	24.7	0.4
K	177.0	175.7	1.2
L	35.8	36.3	0.3
M	152.5	149.1	1.1
N	88.0	87.9	0.8
O	26.8	26.5	0.6
P	52.8	53.4	0.4

¹ See figure 2

Figure 1 – Body traits: p.p. = total weight; l.t. = body length; a.g. = height at withers; c.t. = chest circumference; l.a.a. = fore leg length; l.a.p. = hind leg length; c.c. = neck's base circumference.

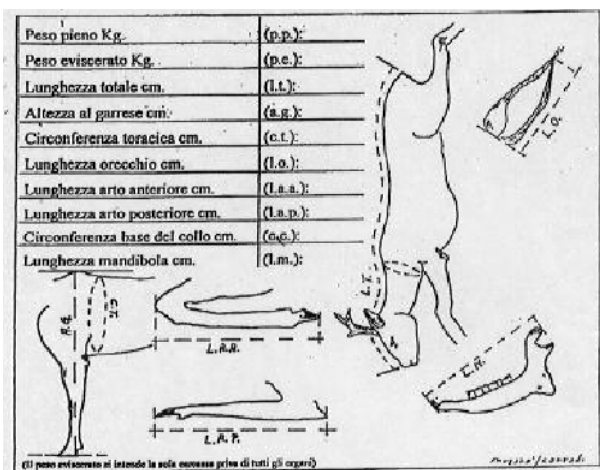


Figure 2 – Skull measurements (Lorenzini *et al.*,1996; Aragon *et al.*, 1997): A condylobasal length; B total skull length; C length from prosthion to P2; D length from prosthion to M3; E greatest prosthion length; F width of the right pedicle; G width of the left pedicle; H neurocranium width; I zygomatic width; J greatest width across the nasals; K basal length; L width of the occipital condyles; M length of the mandible from infradentale to condyle; N height of mandible; O length of the premolar row of the mandible; P neurocranium height.

