Allozymic variation in the European Polecat Mustela putorius from western France

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Allozymic variation was surveyed in 21 European polecats *Mustela putorius* Linneaus, 1758 from western France, using starch gel electrophoresis. Fourteen loci were success-fully examined and genetic variation was detected at 28.6% of the loci at the 0.05 level. Heterozygosity level averaged 0,082. European polecats from western France clearly showed significant levels of genetic variability. This result contrasts with previously reported analysis from Danish populations.

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Introduction

Comparative studies on genetic variability among mammals reported a supposed lower heterozygosity level in carnivores than in other groups (cf Nevo 1978, Allendorf *et al.* 1979, Sage *et al.* 1982, Simonsen 1982, Wooten and Smith 1985). One extreme example is the African cheetah *Acinonyx jabatus* which is known to show very little genetic variation (O'Brien *et al.* 1985).

Nevertheless, most of the studies only deal with some species or limited number of relatively localised populations. For instance, the Danish population of Carnivores examined by Simonsen (1982) revealed no allozymic variation within any studied species. By contrast, Lidicker and McCollum (1997) detected 19.4% of polymorphic loci in Sea otter *Enhydra lutris* populations from Alaska and California. In the same way, Mitton and Raphael (1990) surveying only 10 American marten *Martes americana* from Wyoming found genetic variations at 33% of the loci.

Although the chromosome number remains generally constant within the different carnivore families, Mustelidae exhibit a certain karyotypic variation which may result from Robertsionian processes (Wurster and Bennirschke 1968). From a phylogenetic point of view, Mustelidae form a primitive but rather diverse family. Thereby, the "polecat" syngameon, according to the Templeton's definition (1989), may include together the European polecat *Mustela putorius*, the domestic ferret *M. furo*, the steppe polecat *M. eversmanni* and probably the European mink *M. lutreola* all of which are at least partially interfertile (Grafodatsky *et al.* 1978, Ternovskaya 1990) despite karyological differences. In this syngameon, the divergence of the species tends to be correlated with different ecological niches

(Youngman 1982). Polecat populations are widespread in the Palearctic region and live in various habitats; therefore polecats may exploit diverse trophic resources (cf Blandford 1987, Lodé 1997). Thus, it seemed reasonable to suspect the existence of genetic variations paralleling such diversity among polecats.

I report here allozymic variations in a polecat *Mustela putorius* Linneaus, 1758 population from western France

Material and methods

The genetic analysis is based on 21 European polecats *Mustela putorius* (14 females and 7 males) from western France sampled between Brittany and Poitou. The studied animals were road-killed polecats; only fresh individuals were opportunistically collected since 1995 (authorisation DPN 95-97) and stored -22°C in the laboratory.

Muscle tissue samples were removed from back limbs and analysed by horizontal starch gel electrophoresis using standard techniques (Selander *et al.* 1971). About 1 g of tissue was homogenised and centrifuged. I successfully investigated 11 enzymatic protein encoded by 14 putative loci : adenylate kinase (E.C. 2.7.4.3, *Ak.*); esterases (EC 3.1.1.1, *Est, Est-2*); fructose diphosphatase (EC 3.1.3.11, *Fdp*); glucose-phosphate isomerase (EC 5.3.1.9, *Gpi*); lactate dehydrogenase (EC 1.1.1.27, *Ldh-1, Ldh-2*); malate dehydrogenase (EC 1.1.37, *Mdh-1, Mdh-2*); malic enzyme (EC 1.1.1.40, *Me*); mannose phosphate isomerase (E.C. 2.4.2.1. *Np*); superoxide dismutase (E.C. 2.7.5.1. *Pgm-2*); purine nucleoside phosphorylase (E.C. 2.4.2.1. *Np*); superoxide dismutase (E.C. 1.15.1.1, *Sod*). The continuous buffer systems employed were Tris-citrate pH6 and Tris-EDTA Borate. Zones of enzymatic activity detected on the gels were called F (fast) or S (slow) considering their mobility from the most anodal and numbered sequentially for each enzyme.

Genotypic and allelic frequencies were determined by direct counts from allozyme phenotypes. The mean observed heterozygosity level was averaged over loci and the mean unbiased heterozygosity level was calculated as $H' = \sum [2n (1 - \sum p_i^2) / (2n - 1)] / r$, where *n* is the number individuals, p_i the frequency *i*th allele at a locus, and *r* the number of successfully investigated loci (Nei 1978).

Results and discussion

Among 21 polecats from western France, 5 of 14 loci examined 5 were polymorphic loci whereas 9 appeared to be monomorphic but only four loci were polymorphic at the 0.05 level. Thus the proportion of polymorphic loci was 35.7% of studied loci but only 28.6% at the 0.05 level.

Smith *et al.* (1978) documenting 42 species found an average of 20% of polymorphic loci (range of 3% to 62.5%). Thus, this polecat population showed levels of allozymic variability comparable or slighty above that of other mammals. The polecat polymorphism averaged a similar level to that of one population of American marten *Martes americana* from Wyoming (33.3%, 24 loci, Mitton and Raphael 1990). Of 31 loci scored, only 19.4% were polymorphic in Sea otter *Enhydra lutris* populations (Lidicker and McCollum 1997), and the Italian population of gray wolf *Canis lupus* only showed 10% of polymorphic loci (Randi *et al.* 1993). Nevertheless, since only 14 loci were considered here, it is possible that the pattern observed among polecats might be different with more loci examined. This estimate provides however new information about unsuspected levels of allozymic variations in polecats.

Locus	Genotype	п	Observed frequencies	Heterozygosity $(1 - \sum p_i^2)$	Allelic frequencies	Standart deviation
Est-2	SS SF FF	0 1 20	$0.0000 \\ 0.0476 \\ 0.9524$	0.0465	p = 0.976 q = 0.024	0.007
Mdh-1	SS SF FF	7 11 3	$\begin{array}{c} 0.3333 \\ 0.5238 \\ 0.1429 \end{array}$	0.4819	p = 0.405 q = 0.595	0.023
Me	SS SF FF	14 6 1	$0.6667 \\ 0.2857 \\ 0.0476$	0.3084	p = 0.191 q = 0.809	0.018
Np	SS SF FF	7 3 11	$\begin{array}{c} 0.3333 \\ 0.1429 \\ 0.5238 \end{array}$	0.4819	p = 0.595 q = 0.405	0.023
Pgm-2	SS SF FF	18 3 0	$0.8571 \\ 0.1429 \\ 0.0000$	0.1326	$\begin{array}{l} p = 0.071 \\ q = 0.929 \end{array}$	0.012

Table 1. Genotypic and allelic frequencies for 5 polymorphic loci in 21 polecats from western France (S — slow, F — fast), (p and q respectively refer to fast and slow electrophoretic mobilities).

Here, polymorphic loci were *Est-2*, *Mdh-1*, *Me*, *Np* and *Pgm-2* and two alleles were found in each locus (Table 1). Enzymatic activity revealed two independent isozymes for the *Mdh* and only the fastest one (*Mdh-1*) was found polymorphic. This locus was also polymorphic in American marten (Mitton and Raphael 1990). Similarly, *Est-2* and *Pgm-2* were polymorphic both in polecat and in marten (Mitton and Raphael 1990).

Genotypic frequencies are given for the five polymorphic loci in Table 1. Only one individual was polymorphic for the *Est-2* showing the lowest level of heterozygosity. At this locus, the FF homozygotes prevailed in populations. By contrast, observed heterozygosity reached 0,524 for the *Mdh-1* locus. The SS genotype was predominant at the *Pgm-2* locus and the other allele was only observed in heterozygotes.

The mean heterozygosity level for the 14 studied loci was H = 0.082 and the standardised heterozygosity level was H' = 0.106. Both estimate of heterozygosity clearly exceeded the mean heterozygosity value of 0.039 reported by Wooten and Smith (1985) for 138 mammalian species. However, the *H* estimate is below the mean heterozygosity averaging 0,167 in *Martes americana* (Mitton and Raphael 1990).

Although our sample originated from a restricted geographical area, the polecats showed a high level of allozymic variability in western France. This result underlines that Mustelids are able to reveal higher levels of genetic variability than previously suggested from Danish populations (Simonsen 1982). In terms of biological conservation, maintenance of high level of genetic variability might be an essential element to preserve polecat populations.

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