

Genetic differentiation of the edible dormouse *Glis glis* in the Stolowe Mountains (Poland)

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This is a continuation of our study started in 2013, where we dealt with genetic diversity of the edible dormouse in the Stolowe Mountains National Park (south-western Poland). The study was conducted in 2014 (from the beginning of August to the end of October). A total of 48 dormice from 5 localities were studied. Molecular study involved 12 microsatellite sequences. Eleven out of them were polymorphic. Low genetic variability of the studied population was found. The average allelic richness (A) of each sample ranged from 2.10 to 2.98. The observed heterozygosity (H_O) over all loci amounted to 0.30 whereas an average value of the expected heterozygosity (H_E) was 0.45. The striking genetic difference between populations IV, V and I, II, III were discovered.

Keywords: genetic diversity, *Glis glis*, Stolowe Mountains

1 Introduction

The edible dormouse *Glis glis* is an arboreal European rodent. In Poland this rare mammal is generally more abundant in the southern and eastern parts of the country while in central and northern Poland is sparse (Pucek and Jurczyszyn 2001). In many sites, environmentally isolated populations with no gene flow between one population to another are found (Bartmańska et al. 2010; Trzop and Moska 2013). A main cause of this state is probably the forest fragmentation.

The aim of our study was to reveal the genetic diversity of dormice living in the Stolowe Mountains in south-western Poland.

2 Material and methods

The study was conducted in 2014 (from the beginning of August to the end of October) in the Stolowe Mountains National Park (SMNP). In total 48 individuals captured in bird nest boxes set up in 5 study sites (Permission No. DOPpn-4102/165/10997/11/RS) (Fig. 1) were investigated. The distribution of captured dormice among the study sites was as follows: I (n = 20), II (n = 9), III (n = 10), IV (n = 6) and V (n = 3). Each individual was marked with a unique ear *tattoo*.

DNA was isolated from hair using Sherlock AX Kit. Twelve polymorphic autosomal microsatellite loci known in *G. glis* served for the analysis of nuclear DNA (*Gg3*, *Gg8*, *Gg9*, *Gg12*, *Gg13*, *Gg14*, *7pilch*, *20pilch*, *23pilch*, *24pilch*, *30pilch*, *36pilch*) (Hürner et al. 2009; Dabert et al. 2013). In order to improve the estimation of the amplification product size, one primer of each pair was labeled with a fluorescent dye (FAM, JOE or TAMRA) on the 5'-end, allowing analysis with the use of an ABI 3100 Avant automated sequencer (Applied Biosystems).

The population genetic analyses were performed using R package (R Core Team 2015). The allelic richness and observed (H_O) and expected (H_E) heterozygosity and inbreeding coefficient (F_{IS}) for each of the localities were calculated in hierfstat package (Goudet and Jombart 2015). The observed (H_O) and expected (H_E) heterozygosity and inbreeding coefficients (F_{IS} , F_{st}) for each of the microsatellite

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markers were estimated using pegas package (Paradis 2010). Hardy – Weinberg Equilibrium tests were performed in pegas package. Genetic distances for Principal Coordinates Analysis (PCoA) were calculated using ape package (Paradis et al. 2004).

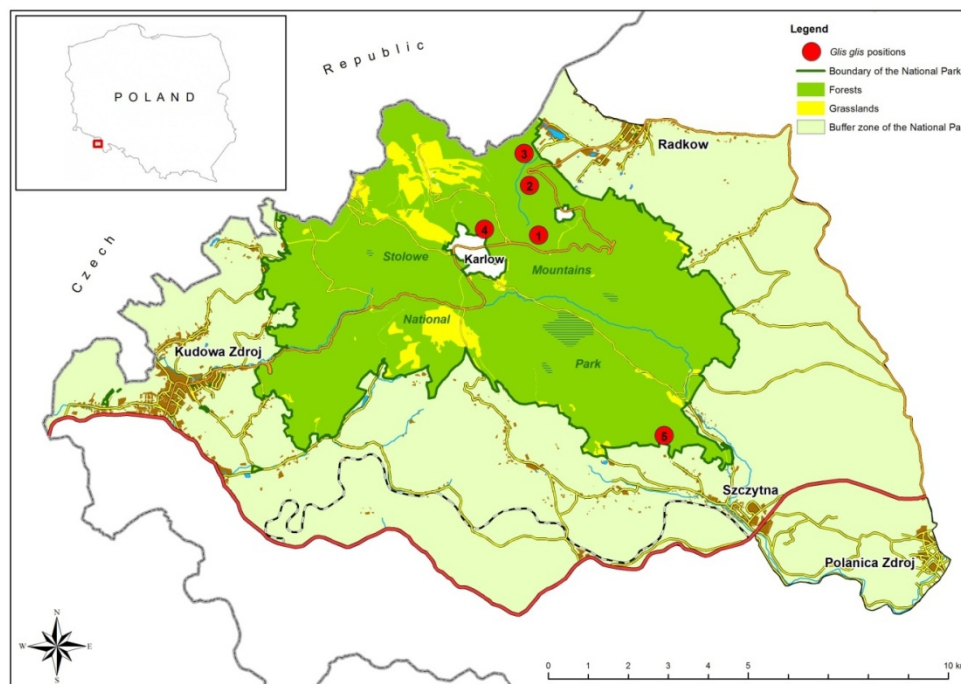


Figure 1 Map of the Stolowe Mountains National Park with indicated (circles) studied areas

3 Results and discussion

Eleven out of twelve microsatellite loci were polymorphic. Only locus *23pilch* was monomorphic and thus was excluded from our study. The average allelic richness (A) of each sample ranged from 2.10 to 2.98. Forty five alleles were found at 11 autosomal microsatellite loci. The most variable locus (*7pilch*) had 7 alleles (Tab. 1). The observed heterozygosity (H_o) per locus ranged from 0.03 to 0.88 and over all loci amounted to 0.30. Expected heterozygosity (H_E) ranged from 0.10 to 0.75 with an average value of 0.45 (Tab. 1). The average H_o in different localities ranged from 0.23 to 0.63, whereas H_E fluctuated from 0.42 to 0.70.

Table 1 Basic parameters of genetic variability estimated for studied *G. glis*

Locus	n_a	H_E	H_o	F_{IS}	F_{ST}	HWE p-value
Gg3	3	0.50	0.23	0.54	0.01	0.004
Gg8	2	0.11	0.12	-0.09	0.15	1.000
Gg9	4	0.57	0.45	0.21	-0.03	0.018
Gg12	2	0.15	0.03	0.80	0.28	0.006
Gg13	4	0.54	0.56	-0.04	0.04	0.041
Gg14	2	0.10	0.11	-0.10	0.26	1.000
7pilch	7	0.69	0.13	0.81	0.15	0.000
20pilch	5	0.75	0.54	0.28	0.09	0.070
24pilch	4	0.64	0.38	0.41	0.13	0.000
30pilch	6	0.74	0.88	-0.19	0.02	0.196
36pilch	6	0.55	0.14	0.75	0.15	0.000
Average	4.09	0.45	0.30	0.31	0.11	-

n_a – number of alleles, H_E – expected heterozygosity, H_o – observed heterozygosity, F_{IS} – inbreeding coefficient, F_{ST} – fixation index

Our results demonstrated very low genetic variability of the studied population and confirmed low level of diversity of European dormouse population. Low, comparable average values of H_o and H_E were observed in the German and Spain populations (0.45, 0.48 and 0.47, 0.53, respectively) (Hürner et al. 2009, Segelbacher et al. 2010).

Improper forest management, which has been carried out in the country for decades, is considered a main cause of this state. It is well known that tree-dependent species that rarely venture to the ground are highly sensitive to forest fragmentation.

The genetic distances between pairs of individuals representing considered populations I, II, III, IV and V were illustrated with PCoA plot (Fig. 2). The striking genetic difference between populations IV, V and I, II, III were discovered. However, a few individuals belonging to population I, II and III were found to be genetically closer to representatives of populations IV and V, than to other representatives of pre-defined populations (I, II and III). In addition, several individuals belonging to population I appeared to be genetically distant from the other representatives of their population and also from individuals of the other considered populations.

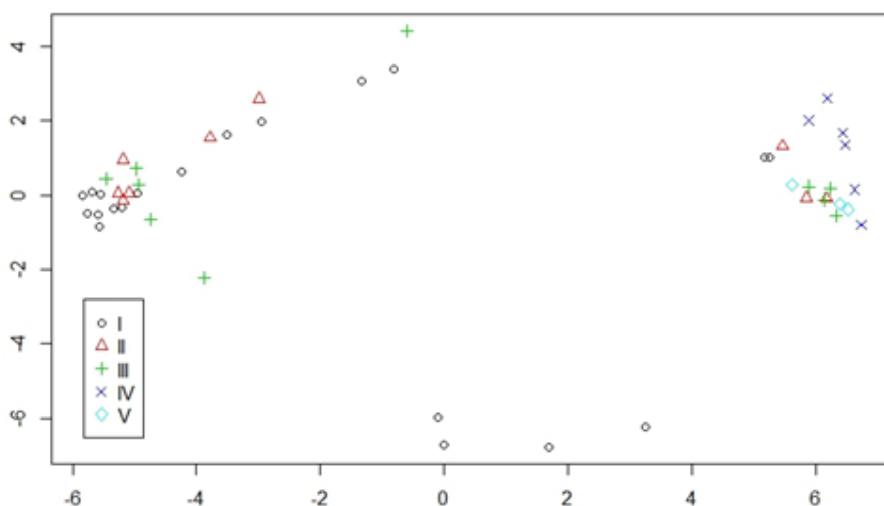


Figure 2 PCoA plot of genetic distances between pairs of individuals from considered populations

4 Conclusions

Low genetic variability of the studied population confirms low level of diversity of European dormouse population. Therefore, besides being extremely important for dormouse habitat conservation activities, there is a need to elucidate the genetic diversity reflecting the history of the species and predicting its future.

Acknowledgments

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References

- BARTMAŃSKA, J., MOSKA, M. and GOTTFRIED, T. (2010) Recent range and distribution of dormice (*Gliridae*, *Mammalia*) in the Sudetes (Poland). *Acta Zool Cracov*, vol. 53, pp. 65–78.
- DABERT, M. et al. (2013) New fourteen highly polymorphic STR markers developed and characterized from edible dormouse (*Glis glis*). *Conserv. Genet Resour*, vol. 5, pp. 429–431. doi: <http://dx.doi.org/10.1007/s12686-012-9820-5>
- GOUDET, J. and JOMBART, T. (2015). *hierfstat: Estimation and Tests of Hierarchical F-Statistics*. R package version 0.04-22 [Software]. Retrieved January 10, 2016 from <https://CRAN.R-project.org/package=hierfstat>

HÜRNER, H. et al. (2009) Isolation, characterization and PCR multiplexing of polymorphic microsatellite markers in the edible dormouse *Glis glis*. *Mol Ecol Resour*, vol. 9, pp. 885–887. doi: <http://dx.doi.org/10.1111/j.1755-0998.2008.02365.x>

PARADIS, E., CLAUDE, J. and STRIMMER, K. (2004) APE: analyses of phylogenetics and evolution in R language. *Bioinformatics* 20: 289-290. doi: <http://dx.doi.org/10.1093/bioinformatics/btg412>

PARADIS, E. (2010) pegas: an R package for population genetics with an integrated-modular approach. *Bioinformatics*, vol. 26, pp. 419–420.

PUCEK, Z. and JURCZY SZYN, M. (2001) *Glis glis* (Linnaeus, 1766). In: GŁOWACIŃSKI, Z. (ed) *Polish Red Book of Animals*. Warszawa: Państwowe Wydawnictwo Rolnicze i Leśne, pp 79–81.

R CORE TEAM (2015) *R: A language and environment for statistical computing*. R Foundation for Statistical Computing, Vienna, Austria. Retrieved January 10, 2016 from <https://www.R-project.org/>

SEGELBACHER, G. et al. (2010) Isolation of di- and tetranucleotide microsatellite loci in the edible dormouse (*Glis glis*). In: AURELLE, D. et al. (eds) *Molecular ecology resources primer development consortium*. *Mol Ecol Resour*, vol. 10, pp. 751–754.

TRZOP, K. and MOSKA, M. (2013) Dormice (Gliridae) of Ślęza Landscape Park. *Chroń Przyr Ojcz*, vol. 69, pp. 409–414.