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Abstract: Genetic analyses are performed using microsatellites in order to address following questions: Do the two populations (Alps and Jura Mts) and other re-introduced populations have a reduced genetic variability compared to the Slovakian source population and other autochthonous populations in Europe (Scandinavia, Finland, Baltics)? Do the two geographically separated populations of Switzerland differ genetically? In the re-introduced populations, we have observed a tendency for smaller values (0.42-0.61). The population in the Alps had with 0.42 by far the smallest level of heterozygosity. The allelic diversity in the lynx population in the Jura Mts and in the Alps was smaller than in the source population in the Carpathian Mts of Slovakia. The strong genetic drift has led to a significant difference between the lynx population in the Alps and the Jura Mts, and between the two populations and the source population in the Carpathian Mts of Slovakia.

## **Population and conservation genetics of two re-introduced lynx (*Lynx lynx*) populations in Switzerland – a molecular evaluation 30 years after translocation**

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Lynx went extinct in most of Central and Western Europe at the end of the 19<sup>th</sup> century. In the 1970s, re-introduction programs started in the Alps and in adjacent mountain ranges of Switzerland, Slovenia, Italy, Austria and France. In all projects, only very few founder individuals were released. All came from the same founder population, the Carpathian Mountains of Slovakia, and some of them were even closely related (siblings, parent-offspring). The two populations in Switzerland - Jura and Alps - are still small and isolated. They consist today of not more than 40 - 50 and 60 - 80 reproducing individuals, respectively. From this situation, the following questions arise: Do the two populations and other re-introduced populations have a reduced genetic variability compared to the Slovakian source population and other autochthonous populations in Europe (Scandinavia, Finland, Baltics)? Do the two geographically separated populations of Switzerland differ genetically? Additionally, in the context of future re-introductions of lynx in Europe, the taxonomic status of the species in the various populations in Europe is of interest. To address these questions, genetic analyses are performed using microsatellites, which were developed in domestic cats (Menotti-Raymond & O'Brien 1995, Menotti-Raymond et al. 1997, Menotti-Raymond et al. 1999), Canada lynx (Carmicheal et al. 2000) and Sumatra tigers (Williamson 2002). For preliminary analyses results of 15 microsatellites and 350 samples from 11 populations were available. Additionally, we have analysed samples from Swiss zoos. The allele length was determined with an ABI 3100 sequencer with ABI Genescan and Genotyper software. Statistical analysis was performed using the programs GENEPOP (Raymond & Rousset 1995) and GENETIX (Belkhir et al. 1996-1997).

### **Genetic variability**

Levels of heterozygosity varied in autochthonous populations between 0.54 (Sweden) and 0.68 (Latvia; Fig. 1). Striking is the small value for Sweden. The lynx population there experienced a bottleneck at the end of the 19<sup>th</sup> and early 20<sup>th</sup> century that left its genetic tracks (Hellborg *et al.* 2002).

In the re-introduced populations, we have observed a tendency for smaller values (0.42-0.61). The population in the Alps had with 0.42 by far the smallest level of heterozygosity. The allelic diversity in the lynx population in the Jura Mts and in the Alps was smaller than in the source population in the Carpathian Mts of Slovakia. For the other re-introduced populations, the samples size is still too small for a comparison.

In addition to the loss of alleles and the reduction of the level of heterozygosity, the re-

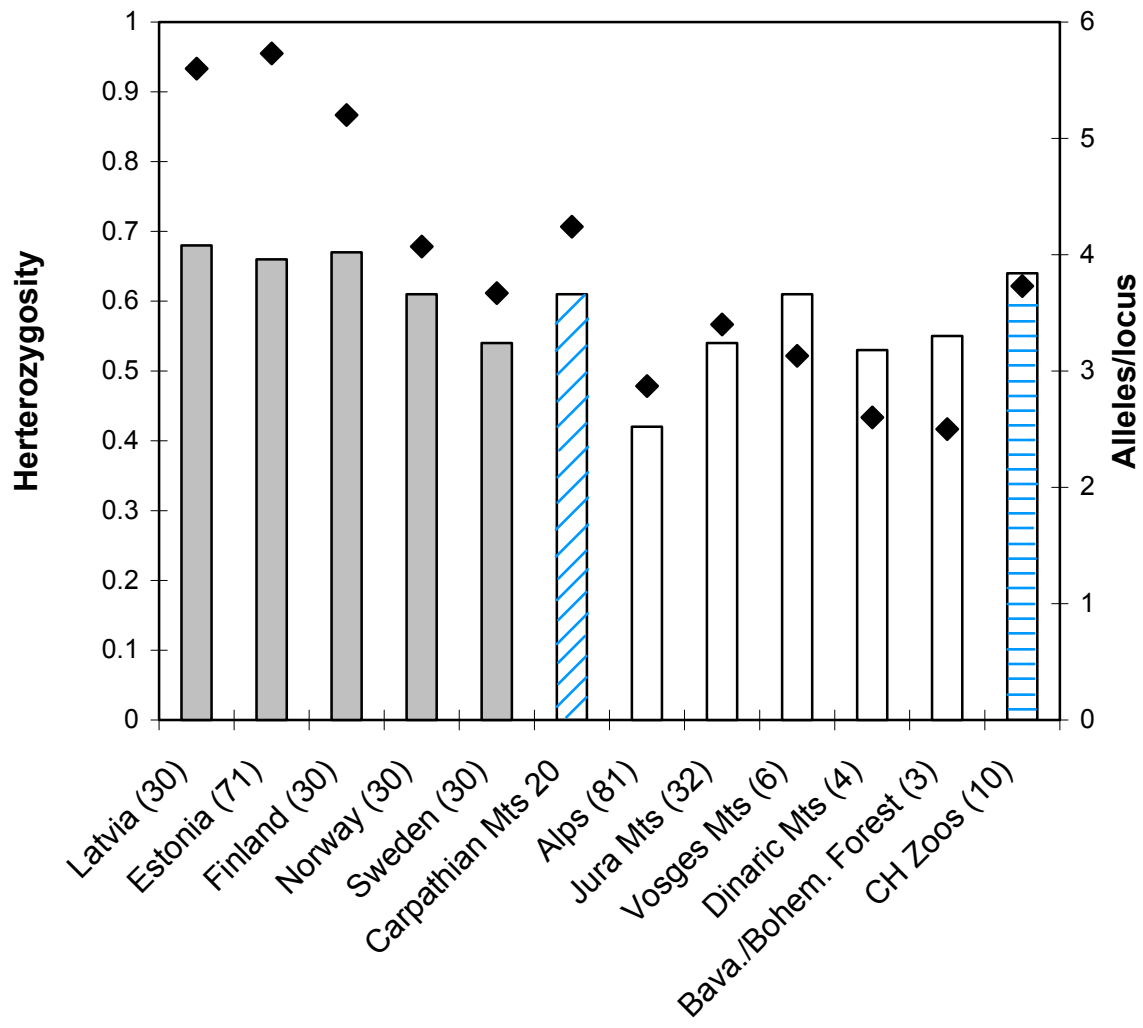


Fig. 1. Genetic variability of lynx populations in Europe. The columns present the level of heterozygosity (left y-axis), the symbols the mean number of alleles per locus (right y-axis). Grey columns = autochthonous populations, diagonal hatched column = source population (Carpathian Mts of Slovakia) for the re-introductions. White columns = re-introduced populations. Horizontal hatched column = Swiss zoo population. Number in parentheses refer to the sample size.

introduced populations in the Jura Mts and in the Alps have experienced a strong genetic drift. For example for microsatellite Fca 115, seven alleles went lost, and the frequencies of the remaining ones have changed drastically, from almost disappearing to becoming very frequent.

## Genetic differentiation

The strong genetic drift has led to a significant difference between the lynx population in the Alps and the Jura Mts, and between the two populations and the source population in the Carpathian Mts of Slovakia (Fig. 2). The re-introduced population in the Dinaric Mts (Slovenia, Croatia and Bosnia & Herzegovina) also drifted away from the source. The two populations in the Vosges Mts and the Bavarian (D)/Bohemian (CZ) forest on the other side are less distinct from the source population. They were founded with a larger number of individuals that were released over several years.

Lynx from the Nordic populations seem to be clearly distinct from those in the Carpathian Mts. To decide if they can be called subspecies as proposed from morphology further analyses are needed.

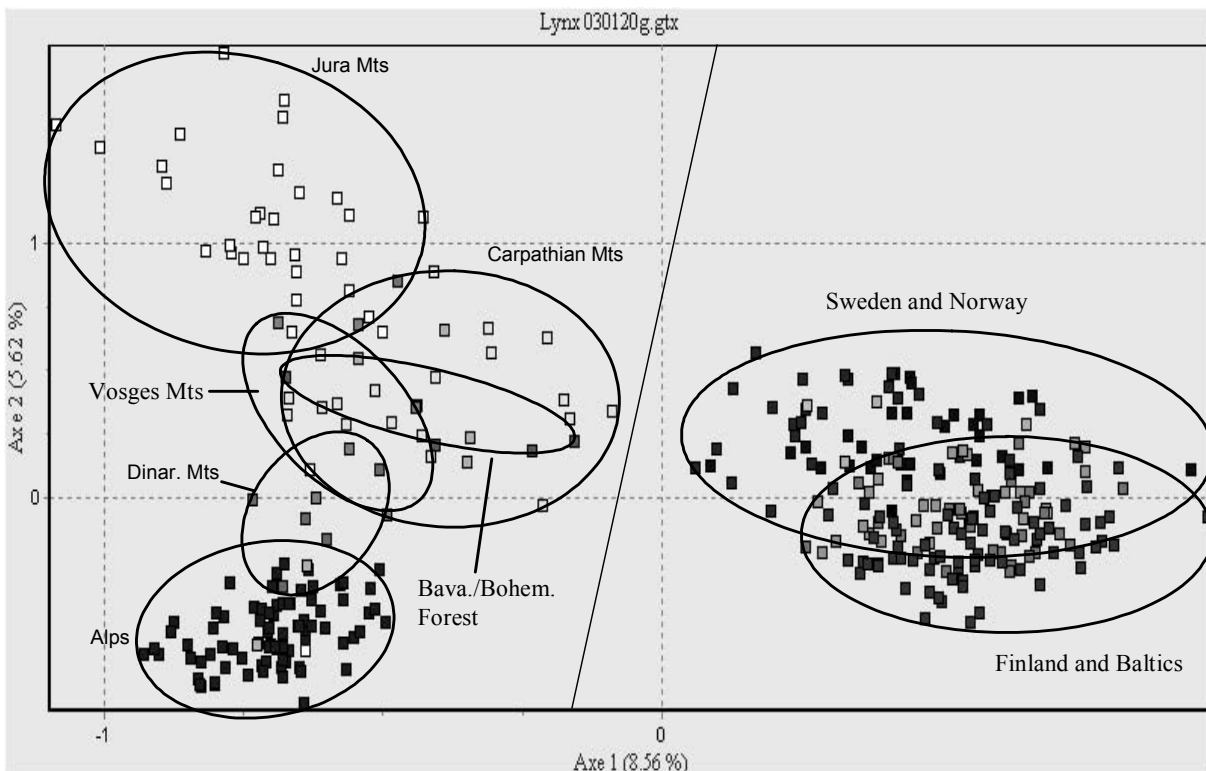


Fig. 2. Principle component analysis based on allele frequencies of 15 microsatellites for lynx populations in Europe.

## References

Belkhir et al. GENETIX, logiciel sous Windows TM pour la génétique des populations. Laboratoire Génome et Populations, available at <http://www.univ-montp2.fr/~genetix/genetix.htm>. 1996-1997.

- Carmicheal, L.E., Clark, W. and Strobeck, C. (2000). Development and characterization of microsatellite loci from lynx (*Lynx canadensis*), and their use in other felids. *Molecular Ecology*, 9: 2155-2234.
- Hellborg, L., Walker, C. W., Rueness, E. K., Stacy, J. E., Kojola, I., Valdmann, H., Vilà, C., Zimmermann, B., Jakobsen, K. S., Ellegren, H. (2002). Differentiation levels of genetic variation in Northern European lynx (*Lynx lynx*) populations revealed by microsatellites and mitochondrial DNA analysis. *Conserv. Genetics*, 3: 97-111.
- Menotti-Raymond, M. and O'Brien, S.J. (1995). Evolutionary conservation of ten microsatellite loci in four species of Felidae. *J.Hered.* 86: 319-322.
- Menotti-Raymond, M., David, V.A., Stephens, J.C., Lyons, L.A. and O'Brien, S.J. (1997). Genetic individualisation of domestic cats using feline STR loci for forensic applications. *J. Forensic Sci.* 42: 1039-1051.
- Menotti-Raymond, M., David, V.A., Lyons, L.A., Schaffer, J.F., Hutton, M.K. and O'Brien, S.J. (1999). A genetic linkage map of microsatellites in the domestic cat (*Felis catus*). *Genomics* 57: 9-23.
- Raymond, M. and Rousset, F. (1995). GENEPOP (version 1.2) - population genetics software for exact tests and ecumenicism. *J.Hered.* 86: 248-249.
- Williamson, J. E., Huebinger, R. M., Sommer, J. A., Louis, E. E., Barber, R. C. (2002). Development and cross-species amplification of eighteen microsatellite markers in the Sumatran tiger (*Panthera tigris sumatrae*). *Mol. Ecol. Notes* 2: 110-112.

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