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Summary

Loss of biodiversity is a global concern. Next to the diversity of species and ecosystems, biodiversity includes the genetic diversity. Therefore, to protect biodiversity we should also protect the genetic diversity. It is especially important nowadays, when due to human activities the environment is changing at an unprecedentedly high rate and genetic diversity may allow species to adapt to these quickly changing, thus stressful, conditions. One of the factors that may affect genetic diversity of populations is environmental pollution with toxic chemicals. Although scientists have been studying already for decades to what extent pollution constitutes a threat for genetic diversity of natural populations, the results of the existing studies are inconclusive. Strikingly, there is a lack of large scale studies involving genome-wide analyses of numerous species from the same polluted area.

In this thesis, I tested the hypothesis that long-term metal pollution impacts the level of genetic diversity of natural populations, and that the scale of the impact depends on the species dispersal abilities. The study was conducted along a metal pollution gradient in the vicinity of the zinc and lead smelter “Bolesław”, close to Olkusz in Southern Poland. Populations of three invertebrate species with different dispersal abilities: the earthworm *Lumbricus rubellus*, the centipede *Lithobius forficatus* and the rove beetle *Staphylinus erythropterus*, were sampled along the studied gradient. Their genetic diversity and structure were assessed with the use of RAD tags and mtDNA molecular markers.

Firstly, as toxicity of metals is determined by their bioavailability, I applied a toxicokinetics approach to estimate bioavailability of metals along the studied gradient of pollution (Chapter 1). Exposing the earthworms *L. rubellus* to soils originating from the Olkusz gradient I demonstrated that, despite the high soil organic matter content, metals were bioavailable to soil organisms. Earthworms intensively accumulated nonessential metals (Cd, Pb) with very little elimination, but showed very efficient regulation of body concentrations of the essential metals (Cu, Zn).

Consequently, I assessed population genetic diversity of selected invertebrates. The earthworm *L. rubellus*, the least dispersive species, was shown to be highly polymorphic, with a population nucleotide diversity of 0.7-0.8% (Chapter 2). Opposite to the prediction of genetic diversity decrease

with increased pollution level, among the *L. rubellus* populations from the Olkusz sites the highest genetic diversity was observed at the most polluted site. Within the analyzed individuals I identified five highly divergent mtDNA lineages of *L. rubellus* in sympatry. However, the clustering of the nuclear RAD tags was not according to the mtDNA lineage but matched the population origin. This suggests no reproductive isolation between the mtDNA lineages of *L. rubellus* and violates an additional hypothesis about cryptic species of *L. rubellus* in continental Europe.

When analyzing the moderately dispersive species *L. forficatus* (Chapter 3) I discovered that this centipede constitutes in Poland a complex of at least three highly differentiated genetic groups with little admixture, probably being cryptic species. A low level of admixture, differences in the level of population genetic diversity and the presence of fixed polymorphisms among the genetic clusters implied their reproductive isolation. However, I did not find an impact of metal pollution on the observed genetic structure of *L. forficatus* populations.

The most mobile species, *S. erythropterus*, showed very little genetic differentiation, suggesting intensive gene flow between populations (Chapter 4). The populations showed very similar diversity at the nuclear genome ($\pi \sim 0.5\%$) and some differences in mtDNA diversity. None of the measures of genetic diversity was correlated with concentrations of metals in soil. However, rove beetles from the site with the highest bioavailability of metals showed significantly higher mtDNA diversity than other populations from the Olkusz area. At the nuclear level the number of haplotypes and haplotype diversity of this population also were the highest, but not significantly different from those observed at the other sites.

Overall, the results of the thesis did not confirm the hypothesis that metal pollution causes stronger decrease of the population genetic diversity in species with lower dispersal potential. In both the least dispersive species, *L. rubellus*, and the most dispersive species, *S. erythropterus*, the highest genetic diversity was found in populations from the site characterized by the highest bioavailability of metals. This might result from the increased mutation rate due to high oxidative stress at this site or, in case of rove beetles, intensified immigration from neighboring populations into a population acting as 'ecological sink'. The study resulted also in discovering cryptic species in the centipede, *L. forficatus*, indicating the importance of cryptic diversity in ecotoxicological studies.