MOLECULAR SYSTEMS OF BIOTRANSFORMATION AND METAL STORAGE OF BIVALVE MOLLUSK IN THE EXPOSURE TO NANOFORM OF ZINC OXIDE

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Engineering nanoparticles of Zinc oxide (nZnO) belong to most widely utilized metal-based nanoparticles in electronics and personal care products. Their expected biological effects could be related to particular properties of nanosized particles and/or bioavailability of Zn^{2+} from these structures. Recently, the study of the effect of nZnO on the march frog *Pelophylax ridibundus* detected bioavailability of Zn from nZnO that was not accompanied by increased cytotoxicity in the exposure to nZnO *per se*. However, the combine exposure to nZnO and elevated temperature oppressed this bioavailability in frog (Falfushynska et al., 2017; Myhalska et al., 2016). Freshwater bivalve mollusks are expected to be particularly at risk from pollution by nZnO because they are sessile and filter large amounts of surface water, including suspended particles. The specimens inhabiting the cooling reservoirs of fuel power plants are subjecting constant combine influence of elevated temperature and industrial pollution. The aim of this study was to elucidate the ability of bivalve mollusks from these reservoirs subjected to combine adverse effects to decompose nZnO at two different temperature regimes depending on their life history in the native habitat.

Freshwater mussels *Unio tumidus* from two cooling ponds, Burshtyn and Dobrotvir pover plants (B-pond and D-pond correspondingly) were exposed for 14 days to elevated temperature (T, 25^{o}C), n-ZnO (3.1 μM), Zn^{2+} (3.1 μM), or n-ZnO at 25 ^{o}C (n-ZnO+T). Control groups from both sites were held at 18^{o}C for 14 days. The metal-buffering thermostable and reach with thiol groups intracellular proteins metallothioneins were eluted from the thermostable extract by gel-exclusive chromatography on Sephadex-50, and the level of Zn in their pike was assessed by spectral analysis after digestion as described in (Falfushynska et al., 2014). Activity of multixenobiotic families of ABC transporter (p-glycoprotein) was determined as a rate of efflux of Rhodamine B in the presence or absence of MXR inhibitors verapamil and cyclosporine A (Ivanina and Sokolova, 2008). Genotoxicity was determined as the level of alkaline DNA precipitation (Olive, 1988) using Hoechst 33342.

Results demonstrate the prominent differences in the responses of mussels from two sites in the same exposures. Only mussels from D-pond were able to decompose nZnO and utilize an excess of Zn in the metallothioneins in the gills, whereas in the specimens from B-pond only exposure to Zn induced Zn-MT, and exposure to nZnO and nZnO+T decreased it. Besides, in the mussels from D-pond specific activation of p-glycoprotein was detected. In the mollusks from B-pond the up-regulation of p-glycoproteins was detected as non-specific response in each exposure. Additionally, in the D-mussels nZnO did not cause the elevation of DNA fragmentation, unlike in the mussels from B-pond. The differences in the bioavailability of nZnO were less prominent under the heat stress. The responses to nZnO and Zn were distinct. Since, the nZnO caused the biological effects both per se and as a source of Zn^{2+} depending on the origin of mussels and temperature of exposure.

Our findings indicate the limited capacity of cellular mechanisms to protect against nZnO in the mussels subjected to the combination of multiple stressors.

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