Does biodiversity of macroinvertebrates and genome response of Chironomidae larvae (Diptera) reflect heavy metal pollution in a small pond?

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Received: 20 August 2010 / Accepted: 9 February 2011 / Published online: 15 March 2011 © Springer Science+Business Media B.V. 2011

Abstract The investigation was carried out on a small pond situated on a recent mine spoil at Bolesław in the Olkusz region with Zn–Pb ore deposits. Water of the pond had pH 7.2–8.5 and low concentrations of heavy metals. Concentrations of Pb (487 μg g⁻¹) and Zn (1,991 μg g⁻¹) in the sediment were very high and potentially could lead to toxicological effects. In the pond, 48 taxa of macroinvertebrates belonging to Oligochaeta and water stages of Ephemeroptera, Odonata, Megaloptera, Trichoptera, Heteroptera, Coleoptera and Diptera (mainly Chironomidae family) were found. The influence of heavy metals on macroinvertebrates diversity was not found. Effect of heavy metal pollution was observed on the appearance of chromosome aberrations in the polytene chromosomes of Chironomidae larvae. It was manifested by two ways: (1) in Kiefferulus tendipediformis and Chironomus sp. chromosome rearrangements in fixed state (tandem fusion and homozygous inversions), indicated intensive process of speciation; (2) in Chironomus sp., K. tendipediformis, Glyptotendipes gripekoveni (Chironomidae) somatic chromosome rearrangements (inversions, deficiencies, specific puffs, polyploidy) affected few cells of every individual. The somatic functional and structural alterations in Chironomidae species are particular suitable as biomarkers—they can be easily identified and used for detecting toxic agents in the environment.

Keywords Water body · Pb · Zn · Macroinvertebrate · Diversity · Genotoxicity

Introduction

Contamination of the environment by chemical wastes poses one of the most serious threats to the quality of freshwater ecosystem. Pollution by trace metals as a result of mine drainage is a particularly serious problem in many parts of the world. It is known that heavy metals
are genotoxic and neurotoxic and affect many physiological and cellular processes in different invertebrates which have been studied so far (Ross et al. 2002; Bonacker et al. 2005; Florea and Büsselberg 2006). It is necessary to find sensitive and inexpensive ways of assessing the degree of environmental damage arising from such contaminants. Very important in this aspect is the identification of a good biological system which can be used as an indicator of pollution in the environment. In rivers and lakes, composition and abundance of benthic invertebrate fauna are important elements for classification of ecological status (Directive 2000/60/EC). Among the benthic invertebrate Chironomidae larvae are considered as a suitable biomonitoring model for ecotoxicological assessment (Warwick 1990). The larvae are widely distributed and inhabit every type and condition of aquatic habitat (Warwick 1990). They are included in biotic indices (De Pauw et al. 1992). They possess excellent salivary gland chromosomes which make them very suitable organisms for genotoxicological studies (Michailova et al. 2003, 2009a, b).

The aim of the study was to use biodiversity of macroinvertebrates and the appearance of chromosome aberrations of Chironomidae (Diptera) larvae in monitoring of small pond, which sediments are heavily contaminated by heavy metals. The genome response of the Chironomid species was studied for the first time. Investigation was carried out in a small pond within Olkus Industrial Region with Zn–Pb deposits near Kraków (Poland).

**Materials and methods**

**Study area**

The study was conducted in a small pond situated on a recent mine spoil in Boleslaw, in the area of zinc–lead ore deposits in Olkus Industrial Region (Southern Poland, geographical coordinates: 50°17′26.61″ E, 19°26′36.93″ N (Fig. 1). It is 52 m in length and 17 m in wide. Its maximum depth is 1.2 m. The bottom sediment contains both sand and mud. A large part of the pond (ca. 75%) is overgrown by emergent macrophytes, mainly *Glyceria maxima* (Hartm. Holmb.), *Phragmites australis* (Cav.) and *Typha latifolia* (L.). Open water occurred only as a narrow belt in the middle part of the pond. Now on the spoil recultivation processes are being conducted.

The mining of the Olkus Industrial Region started in the twelfth century and has lasted up to the present day. Many Zn–Pb mines, smelters and a sulphur acid plant have operated in this area (Cabała 2001; Cabała and Sutkowska 2006).

Investigations were conducted during the spring and summer of 2001, 2002 and 2008. Samples of water and sediment (upper layer 0–5 cm) for physicochemical analysis, macroinvertebrates for biodiversity analysis and larvae of (Chironomidae, Diptera) for cytogenetical analysis were collected.

**Physicochemical and biological analysis of samples**

Water temperature, conductivity and pH (also sediment) were measured in situ. Dissolved oxygen was determined according to the Winkler method. Chloride, sulphate, hydrocarbonate and nitrate anions were analyzed using ion chromatography (DIONEX, IC25 Ion Chromatograph), while ion chromatography (DIONEX, ICS-1000) was used for cations (Mg, Ca, K and P). Ammonia was analyzed with the nesslerization method, while P-tot (after mineralization) with the molybdenum blue method (APHA 1992).

Sediment samples were passed through a 0.063-mm sieve. For the study of total metal concentrations, sediment samples (three subsamples from each station) were digested with 65% HNO$_3$ using DigiPREP HT (Tusnovics Instruments). Fractionations of the heavy metals were analyzed using the operationally defined BCR procedure (Larner et al. 2006). The BCR procedure aims to fractionate metals into four operationally defined phases of (F1) acid extractable, (F2) reducible, (F3) oxidisable and (F4) residual, with steps targeting exchangeable and carbonate bound metals, iron and manganese oxide/hydroxide associated metals, metals bound to sulphide and organic phases and mineral phases, respectively.