Christine Helsen
Stefan Prekovic
Bojan D. Petrovic
Thomas Van den Broeck

Editors

PROCEEDINGS

Arandjelovac and Belgrade, Serbia
February 02-08, 2014

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Christine Helsen, PhD
Stefan Prekovic, MSc
Bojan D. Petrovic, BSc
Thomas Van den Broeck, MD

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JOINT DANUBE SURVEY 3: MICROBIOLOGICAL QUALITY AND GENOTOXICITY ANALYSIS

Stoimir Kolarević[1], Margareta Kračun-Kolarević[2], Momir Paunović[2], Zoran Gačić[3], Jovana Kostić[3], Jelena Knežević-Vukčević[1], Andreas Fanleitner[4], Alexander Kirschner[5], Georg Reicher[4], Stefan Jackwert[5], Branka Vuković-Gačić[1]

[1] University of Belgrade, Faculty of Biology, Chair of Microbiology, Centar for Genotoxicology and Ecogenotoxicology, Belgrade, Serbia
[2] University of Belgrade, Institute for Biological Research Siniša Stanković, Belgrade, Serbia
[3] University of Belgrade, Institute for Multidisciplinary Research, Belgrade, Serbia
[4] Vienna University of Technology, Institute for Chemical Engineering, Research Group Environmental Microbiology and Molecular Ecology, Interuniversity Cooperation Centre Water & Health, Vienna, Austria
[5] Medical University Vienna, Institute for Hygiene and Applied Immunology Unit for Water Hygiene, Centre for Pathophysiology, Infectiology and Immunology Vienna, Austria

1. INTRODUCTION

The Joint Danube Survey 3 (JDS3) is the world's biggest river research expedition of its kind in 2013. The JDS is carried out every six years – JDS1 was in 2001 and JDS2 in 2007. For six weeks between 13 August and 26 September, the JDS3 ships travelled 2,375 km downstream the Danube River, through 10 countries, to the Danube Delta. An international Core Team of 20 scientists consisted from experts for microbiology, macrophytes, phytoplankton, phytobentos, macrozoobentos, fish and chemistry [1].

To be safe for consumption, water must be free of pathogenic bacteria among which enteric pathogens are the ones most frequently encountered. While total coliforms only arise suspicion on faecal pollution in aquatic environments, the faecal coliforms indicate presence of faecal pollution with high probability; they are mainly represented by *Escherichia coli* and together with faecal streptococci are used widely in examination of the water quality.

The simple detection of pollutants in environment provides only limited data on the substances present in the environment and gives no information on the relationship between contaminant exposure and biological effects in aquatic organisms; therefore a proper evaluation of the impact of pollutants by biomarkers becomes essential. Mussels and fish are commonly employed in biomonitoring; nevertheless our previous studies confirmed their applicability for studying DNA damage as biomarker [2,3,4].

The comet assay is a relatively simple procedure which evaluates DNA damage by the tail fluorescence intensity of single gel-embedded cells following alkaline electrophoresis. The ability to make a measurement of damage at the genetic level

provides information relevant to determining the health of the environment, particularly in the context of investigative monitoring.

2. EXPERIMENT

Indicators of faecal pollution were isolated using the Colilert® and the Quanti-Tray/2000. The Colilert® simultaneously detects total coliforms and *E. coli* density using the nutrient indicators *o*-nitrophenyl-β-D-galactopyranoside (ONPG) and 4-methylumbelliferyl-β-glucuronide (MUG), which is metabolized by total coliforms and *E. coli*, respectively. Detection of enterococci was performed with Biorad kits according to ISO 7899-1 (1998; multiwell plates, MPN technique).

Comet assay was performed on haemocytes of mussels *Unio pictorum*, *U. tumidus* and *Sinanodanta woodiana* and erytrocytes of fish *Alburnus alburnus* and *Neogobius melanostomus*. Total of 217 specimens of mussels and 98 of fish were analysed.

3. RESULTS AND DISCUSSION

The results of microbiological analyses are still being processed. Currently, results indicated similarities with the results of JDS2 in presence of hotspots of faecal pollution and influences of pollution in tributaries on the level of pollution of the Danube River. The results of genotoxicity analysis indicated presence of genotoxic pollution at some parts of the river. The highest levels of DNA damage were detected in animals collected in upper and middle course, while the lowest level of DNA damage was detected in animals from lower course of the Danube River.

4. CONCLUSION

To conclude, our results indicated presence of faecal and genotoxic pollution in the Danube River. However, it should be kept in mind that our results are obtained by single measurement and represent snapshot of the Danube River current condition.

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