Effect of Industrial Pollution in Some Biochemical Parameters of the Garden Snail (Helix Pomatia L.) in the Region of “Trepça” smelter in Mitrovica, Kosovo

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Abstract. The aim of this research was to evaluate the effect of industrial pollution from “Trepça” smelter in Mitrovica in some biochemical parameters of snail hemolymph. For research purpose were used about 30 individuals of natural populations of the Garden snail (Helix pomatia L.), taken in two regions with different level of pollution. The first group (test) is taken near the “Trepça” smelter in Mitrovica, while the control group from the vicinity of Peja, a locality unaffected by industrial pollution. Each of individuals was extracted an approximately equal amount of hemolymph, in which were analyzed: glucose (Glc), total protein (TP), albumin (Al), triglycerides (Tgl) and total lipids (TL). Biochemical parameters were analyzed by standard enzymatic methods and ready reagents of the "HUMAN" firm. Research results shown for a significant decrease (P<0.001) in the concentration of glucose, total proteins, albumins, total lipids and triglycerides in the hemolymph of the snails taken near the locality “Trepça” smelter, compared to the control group taken from the vicinity of Peja.

Statistical calculation was conducted using different statistical software such MINITAB®16 and Microsoft Excel 2007.

Keywords: Snail, hemolymph, biochemical parameter, pollution

1. Introduction

Based on the available data on air quality, emissions and other statistical data, it is estimated that the air quality in Kosovo is unsatisfactory. Primary air pollution problem in Kosovo are pollutants with PM10 particles, including other pollutants that affect the human health, flora and vegetation, animals and materials as well. The Kosovo institutions, in the post-war period, have made great efforts in order to reduce the air pollution sources and to improve the air quality. However, even greater commitment is needed to improve the overall situation in sector of air quality assessment [1]. Many researches as well showed that for overall of its history, “Trepça” mining and metallurgical activities presented a health hazard [2].

Actually, the most serious environmental problems arose from the malfunctioning of the “Zvečan Lead Roaster Complex” of “Trepça”. The plant was built in 1967 and has never worked satisfactorily. While a well-operated modern lead smelter emits 0.2 kg lead per ton of crude lead smelted, lead emissions from the “Zvečan” smelter were as high as 14.8 kg lead per ton of crude lead smelted. Also, production waste was dumped anywhere around the site, making it look more like a dumpsite than a smelter [3].

The smelter was closed down by UNMIK, with KFOR assistance, in summer 2000 in order to reduce environmental and health risks caused by the pollution. Also other operations, such as the “Mitrovica Industrial Park” was closed also. Reportedly, their shut down was done in a hurry, without proper decommissioning and safe closure of the facilities. As a consequence some equipment like bins, roaster, conveyors, leaching and purification tanks, pipes and pumps as well as induction furnaces are still filled with
solutions, solid materials or metallic zinc until recently. Today, uncontained waste piles and tailings dams, eroding under wind and water as well as contaminated buildings and polluted soils left behind by previous operations are found to be the main source of contamination. Locally grown foodstuff and poultry exceed maximum limits for various elements such as lead, cadmium, copper and arsenic [4].

Results indicate that soil surface (about 50 cm) is significantly contaminated (exceeding international safety standards) and that lead contamination peaks in the most densely populated areas [5].

2. Material and Methods

2.1. Material

The investigation was conducted with natural populations of garden snail Helix pomatia L. For this research were used about 30 Garden snails, 15 tests and 15 as control. The test groups of snails were taken near the locality of “Trepça” smelter, in Mitrovica while the control group was taken in a region far away from the polluted area, respectively in Peja. The reason why exactly garden snail is used as biomonitor is because the molluscs generally have a wide distribution, easy to be determined, and are sufficient in the study area and have adequate size to provide an appropriate sample for analysis [6]. Another advantage is that, because of their special relation with the ecosystem (soil and plants), of the terrestrial gastropods, effects of pollutants in the body can be identified for a very short time [7]. To avoid changes in the content of hemolymph and tissues depending on the age or weight [8, 9] or by the season or implications of circadian rhythms in results [10] the snail of the approximately same body weight were selected and analyzes were performed at the same time also.

2.2. Methods

For laboratory analyses, were used standard methods and reagents prepared by HUMAN firm. Hemolymph centrifugation was performed on centrifuge (10 min/3000rpm), while biochemical parameters were determined using apparatus for automatic analysis Humalyzer 2000 Human.

Snail hemolymph glucose concentration was determined by enzymatic method, GOD-PAP method, enzymatic colorimetric test [11]. Total proteins were measured by Biuret’s method [12], albumins were estimated by the method of Doumas et al. [13], whereas for estimation of total lipids concentration the method of Zoeliner [14] was used and for triglycerides the method of Schettler and Nussel [15].

The results of this investigation were expressed as the arithmetical average values ± (plus-minus) standard deviation (SD). For statistical analysis were also used software MINITAB 16 and Microsoft Excel 2007.

3. Results and Discussions

The biochemical analysis of the snail’s hemolymph from the polluted locality of “Trepça” smelter in Mitrovica shows a high significant decrease (P<0.001) for all parameters compared with the control group of snails from Peja region (Table 1 and Fig. 1).

Table 1: Biochemical parameters values of in the hemolymph of Garden snail (Helix pomatia L.) from two localities (Mitrovica near “Trepça” smelter and Peja).

<table>
<thead>
<tr>
<th>Region</th>
<th>Glucose (mmol)</th>
<th>Total proteins (g/L)</th>
<th>Albumins (g/L)</th>
<th>Triglycerides (mmol/L)</th>
<th>Total lipids (g/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mitrovica (A)</td>
<td>0.16 ± 0.04 N=15</td>
<td>18.31 ± 3.64 N=15</td>
<td>2.26 ± 0.46 N=15</td>
<td>0.08 ± 0.03 N=15</td>
<td>1.68 ± 0.05 N=15</td>
</tr>
<tr>
<td>Peja (B)</td>
<td>0.382 ± 0.05 N=15</td>
<td>24.026 ± 5.17 N=15</td>
<td>3.2 ± 0.72 N=15</td>
<td>0.15 ± 0.03 N=15</td>
<td>1.760 ± 0.06 N=15</td>
</tr>
</tbody>
</table>

Significance (A:B) P<0.001 P<0.001 P<0.001 P<0.001 P<0.001

A – Test locality, B – Control locality, N=15 – Number of individual per sample
There is evidence of recent years that confirm that Kosovo is considered as one of the ten most contaminated regions of Europe [16, 17]. Based on the data of different authors concerning the environmental situation in Mitrovica region, it is a unique conclusion that contamination with heavy metals in this area is very present and especially the concentration of lead and zinc in this region is at a high level. This situation is very worrying for ecosystems and public health [18].

Researches in the snails, in view of bioaccumulation of heavy metals has a practical importance since they are used as food and can serve as a source of human intoxication with heavy metals [19]. Otherwise molluscs are used as indicators of environmental pollution caused by heavy metals [20]. It should note that trace metal accumulation in snails is known to be affected by the environmental and biological factors including season, animal size and reproductive cycle [21].

The research results show for decrease in the concentration of all parameters investigated, even in high significant degree. These results are consistent with the results of Nedjoud Grara et al. [22], who found a decrease in the concentration of glucose and lipids in the snails taken in a region polluted with heavy metals.

Our previous research results for the snails of the region of Power Plants in Obiliç (Kosovo) also indicate a significant decrease in the concentration of glucose, total protein and cholesterol compared with control group [23].

The results concerning the lipid level in the hemolymph have shown a significant decrease in the lipids concentration in treated snails. According to Aurousseau [24] this is as consequence of the free oxygenated radicals that are toxic via the degradation of the lipids of which the β-oxidation. Decrease in the concentration of carbohydrates could be as a result of the presence of metal dust in the body, metal ions leading to the release of aldehyds and hydrogen peroxide, under the condition of stress, the reserves of carbohydrates are exhausted to satisfy the energy demands. These results are similar to the El-wakil and Radwan [25], who suggested that animals exposed to the pesticides such endosulfane, methyl parathion, quinalphos and nuvan (pesticides) would be the consequence of the direct use of glycogen for the generation of energy.

Also some research conducted by Rao and Jayashree [26] shown for the changes in the glucose, glycogen, total lipid and total protein levels in foot, mantle and digestive gland of the adult Bellamya dissimilis exposed to 96 hours LC 50 concentrations of copper sulphate and zinc sulphate. They showed a marked decrease in all the four biochemical parameters in the treated animals.

In this context shouldnt also mentioned the research results of [27] who also found homeostatic and glycemic disorders in the snails taken from the region contaminated with heavy metals on mining complex “Zvecan” in Mitrovica and to the snails intoxicated with Pb-acetate also.

4. Conclusions
Based on the results obtained from the snails of the region "Trepça" contaminated with heavy metals compared with the control group from Peja could be concluded that:

All biochemical parameters, glucose, albumins, total proteins, triglycerides and total lipids are decreased in a high significant degree (P <0.001) and

The snail Helix pomatia L. may serve as a good biomonitor for evaluation of heavy metal pollution

5. Acknowledgements

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6. References

[19] Annette Gomot-de Vaufleury and François Pihan. Methods for toxicity assessment of contaminated soil by oral or


