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Optical research of biomaterials of Sorbulak

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Abstract. Within the framework of optical research it was established that on the unpolluted samples of sedge stems occurs structuring of material, whereas on contaminated and irradiated blurring of its structure takes place. Sampling of sedges and rushes for research was carried out in areas near the first dam Sorbulak. For comparison, samples of same materials were taken far away from populated areas. Irradiation was carried out with high-energy electrons with energy of 2 MeV and integral dose of 3•10⁵ Gr. Irradiation leads to a more pronounced structuredness of material. There is a significant difference in the structural elements (epidermis, vascular bundles, parenchymal cells, etc.). There are traced dark spots and bands associated with the presence of huge amounts of heavy metals against the background of a green matrix.

1. Introduction

It is known fact that one of the most important problems of environmental biotechnology for today is how to clean polluted water. Detailed analysis of the scientific and technical information indicates that the chemical and technological monitoring to ensure the normal operation of enterprises is not enough for environmental protection. It is necessary to conduct on-going biotechnological monitoring of flora, which includes biomaterials. In the environment, plants such as sedges, reeds and many others, are the most vulnerable to anthropogenic changes. Toxic contamination is a striking example that represents a dangerous threat of our time [1 – 4]. There are many environmental problems that our world is facing today. For instance, pollution mechanisms of reservoirs, the impact of toxic substances on their flora and fauna, purification of water bodies from pollutants. Thus, the attention is focused on the contaminated areas. The wastewater, which is subjected to special purification systems in water treatment facilities, is collected in so-called “lake reservoirs”. The study of environmental issues of reservoirs occurs extremely rare. Being a part of the environment of the regions aquatic environment determines to a large extent the well being of the entire population. For this reason ecological problems of region aquatic environment and the location of wastewater treatment systems require immediate solutions. It is necessary to believe that investigation of ecology of reservoirs will be successful in sequential comparison with already known positions of ecology of freshwater environments [5 – 9].

Heavy metals (HM) are among the most main polluting substances of reservoirs. Therefore, in the environmental and hydro biological monitoring of pollutants HM is given primary importance. Reduction of heavy metals may occur as a result of physical processes of mass transfer, physicochemical processes of complexation, sorption, and accumulation. Many HM are biologically active, do not undergo transformation in hydrobiont organisms, and slowly leave the biological cycle. They are absorbed by the cell walls and accumulate within the cells of microorganisms such as...
bacteria, mycelial fungi, yeasts, and algae. The sorption capacity of most of the HM is relatively high. For this reason, these organisms can be considered as effective and cost efficient sorbents for water purification from impurities. Sedges, reeds, various algae are the most promising indicators of changes in the environment due to the increased concentration of biogenic elements. Higher aquatic plants (macrophytes) absorb, concentrate and convert those chemical elements into a chelate form during the growth and development. Thus, they reduce the concentration of toxic metals in the water and improve its quality. The search for natural physical factors, which have the effect on improving the water quality, should remain.

2. The experimental procedure
Sampling of sedges and rushes for research was carried out in areas near the first dam Sorbulak. For comparison, samples of same materials were taken far away from populated areas. Irradiation was carried out with high-energy electrons with energy of 2 MeV and integral dose of 3•10^5 Gr. This large dose was taken for determining the responses of plant facilities to irradiation. The reflection and transparency of contaminated and non-contaminated samples of biomaterials were examined to study their structure by using optical-microscopic method. The measurements were performed on an automated digital microscope (ACM) Leica DM 6000 M. This microscope is a precision system for research with high-resolution digital cameras and software for analysing and storing images. Selection of different colours in the image is used to detect small or subtle elements of the sample by contrast. Studies of the anatomical structure of elaeagnus leaves were carried out according to the methodological guidelines of Prozinoy M.N. [10]. Surface and pressed samples along with cross-sections were manufactured. First, glycerol was used to lighten preparations. Then, microphotographs were taken on MS-300 microscope (magnification X63). A large number of temporary preparations were produced. Measurement of morphometric parameters was performed for quantitative analysis using ocular micrometre MOV-1-15 (with the lens X9, magnification x10.7). The concentration of heavy metals in the sample is calculated as follows,

\[ C = \frac{A V K}{P}, \text{ (mg/kg)}, \]

where \( A \) – concentration of the element in the sample solution, mcg/ml; \( V \) – volume of the sample solution, mL; \( K \) – dilution factor; \( P \) – mass of weighed portion sample, g; where: \( A = \frac{\gamma}{\ell_{\text{st.s.}}} \ell_{\text{p.l.s.}} \)

where \( \gamma \) – concentration of standard solution; \( \ell_{\text{st.s.}} \) – peak length of standard solution; \( \ell_{\text{p.l.s.}} \) – peak length of sample.

3. Experimental research
Optical microphotographs of unpolluted, polluted, and irradiated samples of sedge plants are shown in figures 1 – 3. As follows from the figures, changes in the morphology of objects clearly appear on contaminated samples. As a result, a huge number of dark spots were found. Structuring of the material is observed on non-contaminated samples of sedge stems; whereas, blurring of its structure is occurred on contaminated and irradiated samples. Thus, it was revealed that irradiation leads to a more pronounced structuring of material.

Analysis of leaf biometrics of check and contaminated samples of sedge (Figures 5 and 6) shows a substantial difference in the structural elements between check and contaminated materials (epidermis, vascular bundles, parenchymal cells, etc.). Dark spots and bands associated with the presence of a huge number of heavy metals are clearly seen on the background of green matrix. Figures 5 and 6 reveal that cells of the sedge epidermis have tabular shape without any intercellular spaces. They also have a thickened outer wall, which is covered on the surface by cuticle along with a waxy coating, living (check sample), and dyingoff (contaminated sample) hairs, which act as screen for reflecting part of the solar rays. Moreover, there are some important parts among the main cells of the same type of epidermis. Those are the guard cells of stomata, the ferruginous and highly sensitive hairs (trichomes), the hydathode, the motor cells etc. The water and nutrients, such as foliar application for
plants, penetrate through existing pores and pectin strands in the outer walls of cells of epidermis. This epidermis protects all the inner tissues of plants from desiccation, mechanical damage, infiltration of various infections. It also regulates gas exchange and transpiration of the whole plant through stomata. There is an accumulation of various substances, such as glycosides, tanning agents, alkaloids, as well as heavy metals with phytoncide properties, in the epidermal cells of contaminated leaf of elaeagnus. Glandular hairs form essential oils, resin, and mucus. Since covering hairs synthesise plant hormones, enzymes and substances necessary for normal functioning of plants, their existence and functioning is essential for the whole organism.

The analyses of the presence of heavy metals in non-contaminated and contaminated samples of sedges were conducted as described in methodology part of this work. The results indicate that sedge differently adsorb environmental elements, particularly heavy metal (Table 1).

![Figure 1. Optical microphotographs of sedge.](image)

![Figure 2. Optical microphotographs of the irradiated sedge.](image)

![Figure 3. The anatomical structure of sedge.](image)
Table 1. Heavy metals content in the material of sedge (Lake Sorbulak)

| (mg/kg)                  | Pb  | Cd  | Zn  | Cu  | Fe  | Ni  | Co  | Mn  | Cr  | Sr  |
|-------------------------|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Sedge (check sample)    | 6.40| 0.80| 32.00| 9.60| 648.0| 20.40| 0.40| 70.0| 25.20| 49.60|
| Sedge (contaminated sample) | 7.00| 1.00| 32.00| 6.40| 720.0| 12.60| 0.48| 3800.0| 8.40| 89.60|

The lowest concentration is observed for cadmium, from contaminated to non-contaminated materials of sedge, 0.8 and 1.0 mg/kg, respectively, and the highest is for manganese, 70 and 3800.0 mg/kg, respectively. The increase of concentration is also observed for iron from contaminated to non-contaminated materials of sedge 648.0 and 720.0 mg/kg, respectively. Selective adsorbing ability of certain heavy metals can be successfully used for cleaning various reservoirs from many harmful elements and compounds. We have found that industrial and domestic wastewater initially contains significant concentrations of metals. Thus, it is the main sources of heavy metals in the aquatic environment of Sorbulak.

As is known, the absolute content of heavy metals in plants are divided into four groups: the elements of increased (Sr, Mn, Fe, Zn), medium (Cu, Ni, Pb, Cr), low (Mo, Cd, Se, Co), and very low (Hg) concentration. In our case, to a first approximation, this pattern is confirmed for sedges as well (the first group).

4. Conclusion

1. It was established that changes in the morphology of objects clearly occur on the contaminated samples. A great number of dark spots were found. Structuring of material is observed on non-contaminated samples of sedge stems; whereas, blurring of its structure was revealed on contaminated and irradiated samples. It was discovered that irradiation leads to a more pronounced structuring of material.

2. Analysis of leaf biometrics of check and contaminated samples of sedge shows a substantial difference in the structural elements between check and contaminated materials (epidermis, vascular bundles, parenchymal cells, etc.). Dark spots and bands associated with the presence of a huge number of heavy metals are clearly seen on the background of green matrix.

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