

Bioindication of the metal-containing wastewater treatment process

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Abstract. The influence of Nickel (II) and Chronic (VI) ions on the redox processes in activated sludge community is discussed. Toxicity definition is made on the basis of fermentation activity (measurement dehydrogenase activity of sludge)

1 Introduction

In the modern world, more and more attention is paid to ecology and environmental protection, including water resources. The main source of pollution is wastewater from industrial enterprises, as well as municipal facilities. The greatest danger is posed by industrial effluents containing various toxic substances, as well as accidental emissions. Their composition is varied and depends on the nature of production.

Among other branches of industrial production in the Republic of Tatarstan, a complex of heavy industry enterprises related to mechanical engineering and tool making is developed. Their activity is associated with the use of heavy and other metals, which become a source of environmental pollution and are highly toxic [1, 2].

With a high bioaccumulation potential, heavy metals are rapidly incorporated into food chains and accumulate in organisms of species at high trophic levels, including humans. For this reason, even low content of heavy metals in comparison with other pollutants makes polluted water bodies unsuitable for use in various economic and household purposes. At the ecosystem level, heavy metal pollution manifests itself in the degradation of communities, changes and decreases in the number of species and animal units [3]. One of the results of such changes in ecosystems is a decrease in the capacity of water bodies for self-cleaning. In connection with the above, it is obvious that there is a need for constant monitoring of the content of heavy metals in reservoirs for various purposes. The possibilities of analytical chemistry, even under the condition of significant improvement of the existing methods and creation of a complex of new ones, in carrying out effective control of water pollution with harmful substances are by no means unlimited. Therefore, tests on various living organisms are used to assess the toxicity of natural waters, industrial waste, soil, feed and other environmental objects, as well as new chemicals and internal environments of humans and animals. In this regard, there is an objective need for the development and application of indirect methods to establish the presence of biologically hazardous chemicals in a reservoir based on observations and experiments with 'living

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reagents’ - aquatic organisms (hydrobionts) whose vital activity is disturbed in a toxic environment.

At the present stage of assessing the quality of the environment, the most actively developing direction is biological control of the state of fresh waters, based on systems of bioindication and biotesting [4].

With regard to ecological biotechnology of wastewater disposal, the results of biotesting of the composition and contamination of wastewater, as well as bioindication of their treatment processes are the most important conditions for ensuring high efficiency and reliable operation of biological treatment facilities. The priority direction of research in the field of biomonitoring of the wastewater treatment process containing metal ions is to identify the patterns of reaction of biotest systems and indicator organisms to the presence of metals in various concentrations individually, as well as in mixtures with each other.

In addition, determination of the values of parameters of the wastewater biological treatment process, such as inhibition by metals of hydrobionts in the aquatic environment, limitation by other components of wastewater as substrates for microorganisms of microbial communities will provide a full-fledged industrial implementation of biotechnology for wastewater treatment.

Thus, studies aimed at the use of various biotest systems, as well as the analysis of microbial communities of treatment facilities in order to bioindicate the process of purification of chemically contaminated wastewater are relevant [1].

2 Research objects and experimental conditions

The objects under study were the bioindication systems of the wastewater treatment process based on adapted microorganisms of activated sludge from the treatment facilities of OJSC ‘Kazan Optical and Mechanical Plant’.

Table 1 shows the composition and characteristics of the model wastewater solution, typical for domestic wastewater entering biological treatment.

Table 1. Composition and physicochemical characteristics of the model effluent stream

| <i>Component</i> | <i>Amount</i> |
|--|---------------|
| Na ₂ CO ₃ , mg/dm ³ | 40 |
| (NH ₄) ₂ SO ₄ , mg/dm ³ | 60 |
| NH ₄ Cl, mg/dm ³ | 30 |
| KH ₂ PO ₄ , mg/dm ³ | 15 |
| CH ₃ COONa, mg/dm ³ | 50 |
| Sucrose, mg/dm ³ | 125 |
| Glycerol, mg/dm ³ | 50 |
| Starch, mg/dm ³ | 50 |
| Physical and chemical indicators | |
| pH | 6.1 – 7.3 |
| COD, mg/dm ³ | 250 |
| BOD, mg/dm ³ | 125 |
| ammonium ions, mg/dm ³ | 40 |
| nitrite ions, mg/dm ³ | 0.3 |
| nitrate ions, mg/dm ³ | 1.3 |

The model solution was prepared considering presence in its composition of the most important macro- and microelements (Table 1). The absence of organic sources of nitrogen and phosphorus excluded additional formation of ammonium nitrogen in the course of ammonification and phosphates in the process of enzymatic decomposition [5]. To prepare the model solution, tap water was used, which explains the presence of nitrogen nitrates.

The temperature of the water entering treatment was not specially regulated and amounted to 18-26 °C.

A model wastewater solution (Table 1) was mixed in certain ratios with solutions of chromium (VI) and nickel (II) ions to provide specified concentrations of metal ions to study their effect on activated sludge microorganisms.

Ni (II) and Cr (VI) ions were used as toxicants, and solutions consisting of a mixture of ions of the studied metals in ratios of 1:2, 2:1, 1:3, and 3:1, respectively, were used.

A biosystem without added metals was used as control.

3 Experiment results

Experimental studies of the bioindication process of metal-containing wastewater were carried out under conditions of aerobic cultivation of suspended microorganisms of activated sludge in a fill and draw mode. For cultivation, model reactors with a capacity of up to 0.5 dm³ were used with the use of aeration devices for saturating the medium with oxygen and mixing it. The fill and draw process of cultivation of microorganisms of activated sludge assumed the systematic replacement of 1/3 of the volume of the culture liquid with an equivalent amount of the nutrient medium - a model solution of waste water with a frequency of 1 every 2 days.

The effect of metals was studied on the basis of toxicity assessment based on changes in the dehydrogenase activity of microorganisms. All measurements were carried out in triplicate.

Diagram of introducing toxicants into a biological purification system with a model wastewater solution (Table 2).

Table 2. Diagram of introducing toxicants into the model wastewater solution

| Time, day | Concentration of toxicant in incoming waste water, mg/dm ³ |
|-----------|---|
| 1 | 1 |
| 3 | 6 |
| 5 | 8 |
| 13 | 12 |
| 16 | 18 |
| 22 | 24 |

Changes in dehydrogenase activity of sludge (DAS) and the amount of biomass in the control sample depending on the cultivation time are shown in Fig. 1

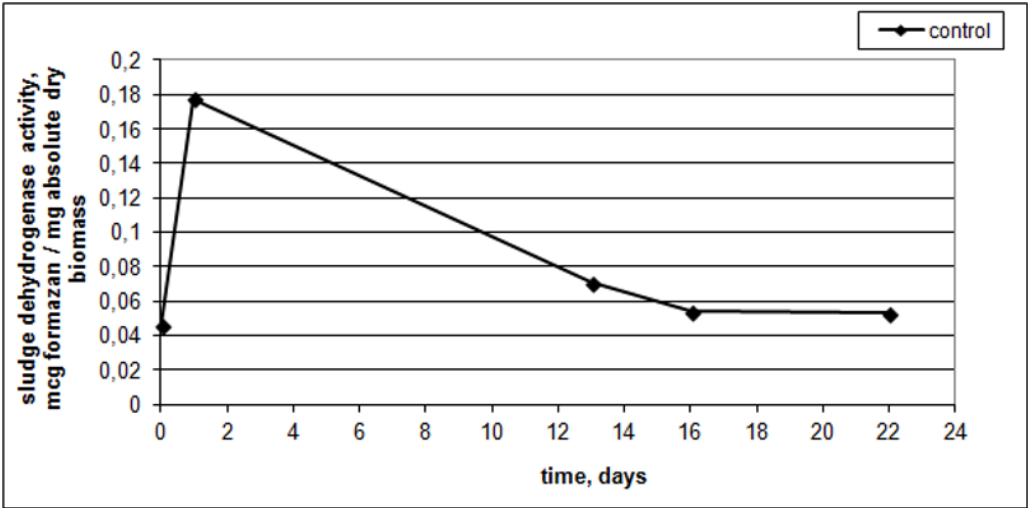


Fig. 1. Change in dehydrogenase activity in the control sample

In the study of dehydrogenase activity of sludge (DAS), the change in the biomass of microorganisms by dry weight was determined. According to the data obtained, over time, the amount of biomass in the cultivator decreased. Fig. 1 clearly shows that at the initial moment of time in the activated sludge, adaptation of microorganisms to the conditions of the laboratory cultivation system takes place, since, despite high concentration of activated sludge, the enzymatic activity of the cells is relatively low. Further, the adaptation of the sludge microflora and the development of cell activity occurs, which is illustrated by the fact that a decrease in biomass does not lead to a decrease in enzymatic activity. A further decrease in dehydrogenase activity is associated with both a decrease in biomass and an increase in the age of activated sludge.

It is known that toxic substances can exhibit both general and specific targeted effects. The examples of the latter include inhibition of enzyme systems (enzymatic poisons), blood corpuscles (hemolytic poisons), the central and peripheral nervous systems of animals (neutrotropic, paralytic poisons), etc. Moreover, the action of any substance on a living cell, as well as on a living organism as a whole, obeys the law of phase reactions, i.e. low concentrations act in the direction of strengthening the function (stimulation), higher concentrations in the direction of oppression (slowdown, inhibition), even higher ones lead to death. Accordingly, the entire process of toxic effects is divided into four phases: indifference, stimulation (arousal), inhibition (depression), death [6].

The values of dehydrogenase activity of the activated sludge samples shown in Fig. 2, are relative values and are obtained from the difference in measured values for activated sludge samples from experimental and control bioindication systems, according to the formula:

$$\Delta DAS = DAS_{(exper.)} - DAS_{(contr.)} \tag{1}$$

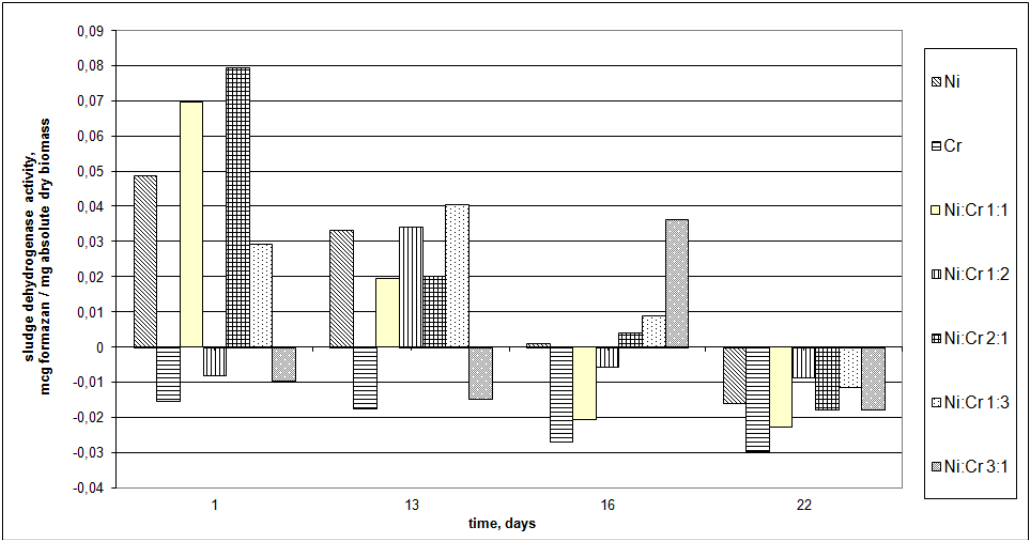


Fig. 2. Kinetics of dehydrogenase activity of metal-containing biosystems in the process of fill and draw cultivation

Note: 1st day - the concentration of toxicants in the model wastewater is 6 mg/l; 13th day - the concentration of toxicants in the model wastewater is 12 mg/l; 16th day - the concentration of toxicants in the model wastewater is 18 mg/l; 22nd day - the concentration of toxicants in the model wastewater is 24 mg/l;

Addition of a solution containing Ni (II) ions causes a more active response of the enzymatic system than a solution of chromium (VI). This is due to the fact that nickel is metabolized much more actively as coenzymes.

The lower toxicity of nickel in comparison with chromium, apparently, can be due to the stability of the complexes formed during its interaction with exopolymer substances of the cell in microbial aggregates, such as activated sludge. This fact is confirmed by the known series of stability of metal complexes with ethylenediaminetetraacetic acid (EDTA) (1) and amino acids (2)

1. $\text{Fe} > \text{Cu} > \text{Ni} > \text{Pb} > \text{Zn} > \text{Cd} > \text{Co} > \text{Mn} > \text{Ca}$
2. $\text{Hg} > \text{Cu} > \text{Ni} > \text{Pb} > \text{Zn} > \text{Co} > \text{Cd} > \text{Mn} > \text{Ca}$

At the same time, a correlation was found between the stability of complexes with EDTA and the stability of metal complexes with various ligands in biological media [7].

A high value of DAS was also noted for biosystems, in the model effluent stream of which Ni and Cr solutions were added in ratios of 1:1, 2:1. Based on the above, it can be assumed that nickel has a predominant influence in these mixtures of metals, however, metals act as antagonists, reducing the toxic effect of each other. This is due to the fact that against the background of the action of one metal-inducer of the biosynthesis of the so-called metallothioneins, the toxic effect of the other can be weakened. Thus, the biosynthesis of metallothioneins is stimulated under the influence of certain metals. Metallothioneins are small, cysteine-rich proteins responsible for binding toxic metals.

This, in particular, is one of the probable mechanisms of toxicological antagonism [8]. A similar mechanism is proposed, for example, as a reason for the antagonistic action of nickel and manganese: if nickel is an inhibitor of ATPase, then manganese, activating some other enzymes that play an important role in energy metabolism, thereby promotes

oxidative phosphorylation. A similar mechanism underlies the antagonistic combined action of chromium and manganese, etc.

Further, on the 13th day of cultivation, a decrease in DAS for the biosystem containing Ni:Cr = 3:1 was noted, and on the 16th day - its sharp increase. There was a violation of the biological balance in the ecosystem of activated sludge with deflocculation of flocks, which was visually manifested in the turbidity of the environment. Further lysis of cells led to the enrichment of the medium with intracellular organic substances. For a biosystem, the composition of the model wastewater of which included a mixture of metal solutions in a ratio of 1:2, an increase in the AIM value was noted on the 13th day, which was also explained by cell lysis with further absorption of the substrate.

4 Conclusions

Based on the results of the study, the following conclusions are drawn:

1. According to the results of changes in the dehydrogenase activity of activated sludge microorganisms under the influence of heavy metals, chromium has a stronger toxic effect than nickel during the entire experiment.
2. The effect of nickel on activated sludge microorganisms is experimentally investigated and it is shown that nickel ions in a concentration of up to 12 mg/l have a stimulating effect on the microorganism community.
3. With the combined presence of Ni (II) and Cr (VI) in the medium in ratios of 1:1 and 2:1 with a total concentration of metals of 6 mg/l, an antagonistic nature of their effect on the biosystem is observed.

References

1. Khalilova A.A., Yakovleva A.V., Sirotkin A.S. Comparative assessment of the toxicity of wastewater containing chromium and nickel ions using various biotest objects // Bulletin of Kazan Technological University. - 2010. - No. 10. - pp. 392-401.
2. Salin A.A., Galeev A.D., Ponikarov S.I. Study of the evaporation of hydrochloric acid: Modeling and experiment (2014) Journal of Engineering Physics and Thermophysics 87 (3), pp. 753-762.
3. Eichenberger E. Relationship between the need and toxicity of metals in aquatic ecosystems / E. Eichenberger; transl. from English; ed. Kh. Zigel, A. Zigel // *Nekotorye voprosy toksichnosti ionov metallov* [Some questions of the toxicity of metal ions]. - M.: Mir Publ., 1993. - pp. 63 - 87.
4. Semchenko V.P. Principles and systems of bioindication of flowing waters / V.P. Semchenko. - Minsk: Orekh Publ., 2004. - 125 p.
5. Kirilina, T.V. Bioconversion of nitrogen and phosphorus compounds in the process of biofiltration of wastewater and their additional treatment with submerged macrophytes: dis. cand. tech. sciences. - Kazan, 2011. - 126 p.
6. Interim methodological guidance on the regulation of levels of chemical substances in bottom sediments of surface water bodies (on the example of oil). - M.: REFIA, NIA – Priroda Publ., 2002.
7. Garipova, R.F. Reaction of microbial and plant systems to environmental pollution with heavy metals / R.F. Garipova // *Izvestiya OGAU* [Bulletin of OSAU]. - 2009. - No. 3. - pp. 192-195.
8. Microelements - biological regulators / N. Kaletina, G. Kaletin // *Nauka v Rossii* [Science in Russia], No. 1, 2007.