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USE OF GREEN ALGAE TO REDUCE HEAVY METALS FROM INDUSTRIALLY POLLUTED WATERS

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Abstract

A number of different physicochemical and biological processes are commonly used to remove heavy metals from industrial wastewater before being discharged into the environment. Conventional physicochemical methods, such as electrochemical treatment, ion exchange, precipitation, osmosis, evaporation, are not cost-effective and some of them are not environmentally friendly. On the other hand, bioremediation processes show promising results for metal removal, even when they are present in very low concentrations if the physicochemical elimination methods fail to function. Moreover, this is a cross-compatible and economically feasible option. The bioremediation strategy is based on the high metal binding capacity of biological agents, which can remove heavy metals from contaminated sites with high efficiency. In this sense, micro-organisms can be considered as a biological tool for metal removal as they can be used to concentrate, remove and recover heavy metals from contaminated aquatic environments.

***Key words**: heavy metals, water, pollution, bioremediation, green algae.*

INTRODUCTION

Today, heavy metal pollution is one of the major problems in the environment (Jerold et al., 2016). Heavy metal pollution is a serious environmental problem and poses a threat to human beings and the ecosystem.

Unlike organic contaminants, heavy metals such as chromium, cadmium, copper and lead are the main pollutants of water because of their carcinogenic and persistent nature (Wael et al., 2016).

Removing heavy metals is considered an important environmental issue and economic considerations (Azza et al., 2013). Romania has a long tradition in mining, especially in the Apuseni Mountains. Unfortunately, there are various negative consequences in this area that arise as a direct and indirect result of mining processes, such as mining water drainage, heavy metals pollution and environmental and life degradation (Babau et al., 2017). Heavy metal pollution has a cumulative and residual character, which means that pollutants accumulate slowly (Sur et al., 2016). The city of Baia Mare has been identified as one of the most polluted areas in Romania and even in Europe. This area presents the historical pollution due to the long period in which the mining industry was the main branch of the industry (Sur et al., 2017). Negative effects have influenced both the environment and people's health (Sur et al., 2012). Green algae play an important role in mitigating contaminating effects on other trophic levels through sorption or biodegradation.

Algae bioassays are now an indispensable part of water monitoring processes because first of all, algae are the main food chain producers and secondly they are more susceptible to contaminants than some species of fish or invertebrates (Wong, 1995). Unicellular planktonic algae, which are the main organics and oxygen producers in aquatic environments, occupy vital space not by increasing body volumes but by generating new individuals, so determining the rate of multiplication by successive cell divisions is the main means of assessing the dynamics changing the population density in order to bioindicate the direct effects of aquatic habitats pollution and to evaluate the treatment processes of heavy metals (Fodorpataki et al., 2010).

The relevance of the biological response of the bioindicator is directly proportional to the systemic level at which the pollutant acts, whether cellular, or organ, body or even population. Bioindication can be considered an induced anthropogenic molecular and biochemical response, manifested by changes in physiological parameters, the effects being perceived at one or more levels of the biological system (Fodorpataki et al., 2015). The present study focused on demonstrating the ability of some microorganisms, green algae, to accumulate a very toxic heavy metal for the environment, Cr^{6+} and the use of these microorganisms in bioremediation processes. Concentrations of metals absorbed by algae are influenced by many environmental factors (Shanab et al., 2012).

Biosorption is an innovative technology that uses living or dead biomass to remove toxic metals from water (Ghoneim et al., 2014).

Removing metals from wastewater through traditional treatments is expensive. Because of this, the research aims at developing more cost-effective alternatives and methods. And in this respect, I refer to the bioremediation processes that represent the method of elimination of environmental pollutants by microorganisms by transforming them from toxic compounds into non-toxic compounds, without affecting the environment. The viability of the process has been demonstrated in laboratory conditions by analysing various experimental parameters such as exposure time, initial metal concentration and pH that can significantly influence bioremediation.

MATERIALS AND METHODS

The test organism used is *Scenedesmus opoliensis*, green algae with unique propagation by spores, which excludes the hereditary variability of individuals, ensuring uniformity of responses to pollutants. In the experiments were used monoalgal axenic cultures of *Scenedesmus opoliensis*, strain AICB 141, a native cell line from Stiucilor Lake, Sacalaia, Cluj County.

The following steps have been taken:

1) Preparation of BBM culture medium (Bold's Basal Medium) with different concentrations of Cr^{6+};
2) Sterilization of the culture medium;
3) *In vitro* inoculation of algae;
4) Growth of static culture in growing room under fluorescent light emitting white light.

Algal cultures were periodically monitored at 3, 6, and 14 days by making observations using a microscope and a cytometer. The growing medium used for algae incubation is a standard nutrient medium containing a wide range of inorganic nutrients to ensure the natural growth conditions of the body. We prepared the stock solution required for the growing environment as recommended by the European standard ISO 8692 on water quality.

After preparing the growth medium, the choice of Cr6+ concentrations to which algae were exposed was made. We used 3 different concentrations of Cr^{6+}: a concentration of 5 μM, 50 μM and 500 μM, respectively chromium-free control samples.

The test was carried out over several weeks in which three Cr^{6+} departed concentrations were tested at a basic pH of 9 and an acidic pH of 5. Finally the demineralisation of the remaining chromium in the aqueous medium was carried out by filtration of algal cultures; the aqueous substance was subjected to spectrometric analysis in an atomic absorption spectrometer: SHIMADZU AA-6800.

RESULTS AND DISCUSSIONS

The pH of the aqueous media was adjusted to baseline of 9, baseline, with 30% KOH solution and 5% acid, respectively, with H_2SO_4. The total volume of the test samples was kept identical in all vessels. pH determination was performed according to methodological rules.

The table 1 shows the pH changes in the aqueous medium.

The effects of Cr^{6+} on algal cultures were more or less pronounced, depending on the type of concentration, the exposure time and the pH of the medium.

Experimental data show that this alga has a promising potential to extract heavy metals, including chrome from various polluted environments.

Table1. Changes in pH from the aqueous medium during the development of algal populations (expressed as difference in pH units versus control

Variants	Basic pH 3 days	Basic pH 6 days	Basic pH 14 days	Acidic pH 3 days	Acidic pH 6 days	Acidic pH 14 days
control	increase with 0.230±0.08	increase with 0.240±0.08	increase with 0.200±0.08	increase with 0.250±0.08	increase with 0.230±0.08	increase with 0.190±0.08
5 µM Cr^{6+}	decrease with 0.155±0.015	decrease with 0.130±0.045	decrease with 0.130±0.045	decrease with 0.160±0.007	decrease with 0.090±0.004	decrease with 0.015±0.005
50 µM Cr^{6+}	decrease with 0.330±0.021	decrease with 0.090±0.008	decrease with 0.090±0.008	decrease with 0.020±0.005	decrease with 0.210±0.010	decrease with 0.024±0.010
500 µM Cr^{6+}	decrease with 0.440±0.015	decrease with 1.460±0.004	decrease with 1.460±0.004	decrease with 0.140±0.005	decrease with 0.360±0.007	decrease with 0.024±0.005

The presence of chromium in the aquatic environment is a major concern for the industry.

At present, chemical precipitation methods are used to remove chrome from wastewater but lead to the formation of solid chromium waste. By the ability of these microorganisms to accumulate different amounts of chromium, bioremediation using algae can be accomplished successfully.

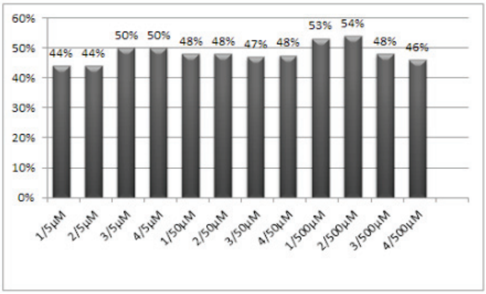

Figure 1.The quantity of Cr^{6+} extracted from algal cultures from aqueous solutions after the first week of exposure, basic pH (n = 4)

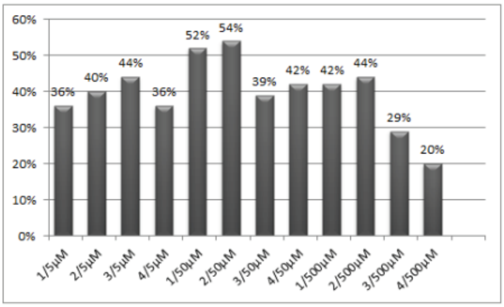

Figure 2. The quantity of Cr^{6+} extracted from the algal cultures from the aqueous solutions after the second week exposure, basic pH (n = 4)

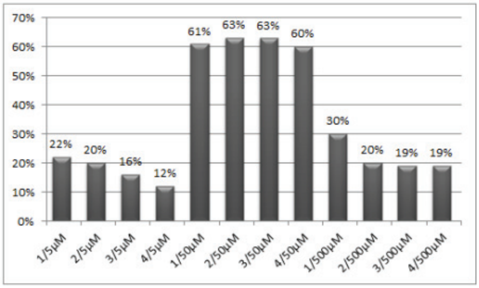

Figure 3. The quantity of Cr^{6+} extracted from the algal cultures from the aqueous solutions after the first week of exposure, acidic pH (n = 4)

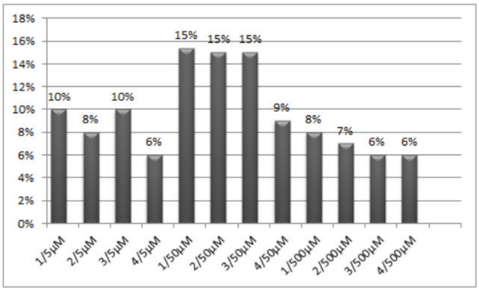

Figure 4. The quantity of Cr^{6+} extracted from the algal cultures from the aqueous solutions after the second week of exposure, acidic pH (n = 4)

Research suggests that *Scenedesmus opoliensis* has proven to be a good bioacumulator of metals, and can serve as a model for treating chrome-contaminated waters.

Scenedesmus opoliensis is one of the most abundant algae used in the treatment of heavy metals polluted waters and with a high potential for removing chrome. It is also true that the absorption/removal of metals is influenced by several factors, in a complex manner, to be evaluated for any program that targets large-scale applications to remove chrome from water.

It can be seen that the bioremediation process decreases with increasing chromium concentration, exposure time and pH environment (Figure 1).
Experiments show that when the pH is basic (Figure 2), the activity of the algae increases and in reverse, in the acidic environment, (Figure 3 and Figure 4), their activity decreases.
The optimal bioremediation period can be achieved over a short period of time in an acidic and long-term environment in the basic environment.

CONCLUSIONS

Natural and anthropic activities generate large amounts of wastewater containing toxic metals, which can generate various ecological problems.
The obtained results show the differences in growth between the control samples and the sample test.
A strong increase in algal density in the control samples and an inhibition of growth in the test samples were observed in both growth media.
This type of algae well tolerates concentrations of 5 μM, 50 μM and in some cases even those of 500 μM at a basic pH, and its use in bioremediation processes can be achieved over a longer period of time.
If polluted water has an acidic pH, bioremediation can be more effective over a shorter period.

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