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RESEARCH ARTICLE

Investigating the ability of chromium (VI) adsorption by *Nitzschia palea*

Mehrani Adl M [1], **Iranbakhsh A** [1]*, **Noroozi M** [2], **Asri Y** [3], **Saadatmand S** [1]

[1] Department of Biology, Science and Research Branch, Islamic Azad University, Tehran, Iran; * iranbakhsh@iau.ac.ir

[2] Department of Biology, Alzahra University, Tehran, Iran

[3] Department of Botany, Research Institute of Forests and Rangelands, Agricultural Research, Education and Extension Organisation (AREEO), Tehran, Iran

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Abstract

Recently, one of the effective methods for the removal of heavy metals and water purification is the use of algae and especially diatoms. In this study, diatoms *Nitzschia palea*) were cultured in F2 medium and after purification was exposed to various concentrations (2-5-10 ppm) of chromium VI for 14 days and chromium absorption was measured by atomic absorption. The maximum and minimum absorbance at 5 ppm and 10 ppm were observed respectively. Diatoms absorbed chromium at a concentration of 2 ppm and higher absorption at a concentration of 5 ppm, but with increasing concentrations up to 10, absorption decreased. Diatoms have the ability to exchange cations of some elements other than hexavalent chromium since chromium six has a very low exchange in diatoms. Diatoms are not able to perform anion exchange, but with sufficient activation, they are used as anionic converters. The results of ANOVA-Oneway showed that the effect of the date on chromium absorption was significant.

Keywords: Diatom, heavy metals, chromium, *Nitzschia palea*

Introduction

Diatoms, microscopic single-celled algae distributed widely in salty and fresh water, are characterized by having a silica cell wall (Naghavian 2012) and as tolerant species against ecological changes (Akar and Şahin 2017). Diatom is very sensitive to changes in the water environment (Zhang et al. 2017). Diatoms are ecologically important, playing crucial roles in biogeochemical cycles (Amato 2010). They are bioindicators of an environmental condition and have been introduced as a water contamination index (Garrido et al. 2013). For microbes, everything is everywhere, but the environment selects" (Patterson, 2009) and the environment being either natural or artificial (Supriya and Ramachandra 2011).

Heavy metals are naturally occurring elements and are present in varying concentrations in all ecosystems (Ilyin et al. 2004). Heavy metals are toxic to many of the microorganisms include algae which formula iniquitous components of the biosphere. Metals/metalloids include lead (Pb), cadmium (Cd), mercury (Hg), arsenic (As), chromium (Cr), copper (Cu), selenium (Se), nickel (Ni), silver (Ag), and zinc (Zn) and other less common metallic contaminants include aluminum (Al), cesium (Cs), cobalt (Co), manganese (Mn), molybdenum (Mo), strontium (Sr), and uranium (Singh et al. 2011).

Recently, heavy metal biosorption using biological material has emerged as a potential alternative instead of the existing conventional physicochemical methods (Sbihi et al. 2012).

Diatoms are sometimes exposed to environmental pollution to which they are highly susceptible (Anantharaj et al. 2011). Heavy metal can affect the growth of diatom or even affect its genetic composition. (Zhang et al. 2017). Chromium is present in different types of industrial effluents, being responsible for environmental pollution. Chromium (VI) is highly soluble in water and can cause cancer for humans. The removal of chromium is traditionally done to reduce its concentration to the extent necessary for the usual biological activities, but not enough (Sala Cossich et al, 2002).

According to the Indian Standard Bureau, the volume of industrial materials that can freely enter chromium and nickel into the environment are 0.1 mg/L and 3 mg/L respectively. Scientists based on biological findings know that they use a different cleansing method to pollute heavy metals. Recently, microbial systems such as fungi, algae, and bacteria have been successfully used as absorption agents for the removal of heavy metals (Juwarkar and Jambhulkar 2008). Some diatom species such as *Planothidium lanceolathum*, has the ability to remove the hexavalent chromium Cr (Cherifi et al. 2016).

There are few pieces of research related to the use of diatoms for the elimination of heavy metals from water and few studies have compared the behavior of several metals and several algal species simultaneously (Fisher et al. 1984). The target of this study was to evaluate the ability of *Nitzschia palea* to absorb heavy metal (chromium VI).

Materials and Methods

Diatom samples collection and identification

Water samples were taken from Kordan, Alborz, Iran. The samples were investigated looking for diatoms, and those containing such materials were chosen to cultivate and use in the study. To identify the live samples of diatom, they were studied by a microscope OLYMPUS-CX31 having 100 magnifying power (Fig. 1).

Then the diatomaceous suspension was poured on to the slurry to evaporate water for one hour at 80°C with H_2SO_4 and a fixed slide was prepared. The precipitates were separated and investigated using a light microscope (Palmer 1980).

Preparing cultivation medium and samples

F2 medium was used for cultivating diatom specimens. The F2 medium for the cultivation of coastal algae, especially diatoms, is widely used (Guillard and Ryther 1962). The culture medium F2 contains NaNO3, $NaH_2PO_4H_2O$, Na_2SiO_3 $9H_2O$, trace metal solution, vitamin solution. Trace metal solution contains $FeCl_3$ $6H_2O$, Na_2EDTA $2H_2O$, $CuSO_4$ $5H_2O$, Na_2MoO_4 $2H_2O$, $ZnSO_4$ $7H_2O$, $CoCl_2$ $6H_2O$, $MnCl_2$ $4H_2O$. Vitamin solution contains thiamine HCl, biotin (H) and cyanocobalamin (B_{12})(Olaizola et al. 1991).

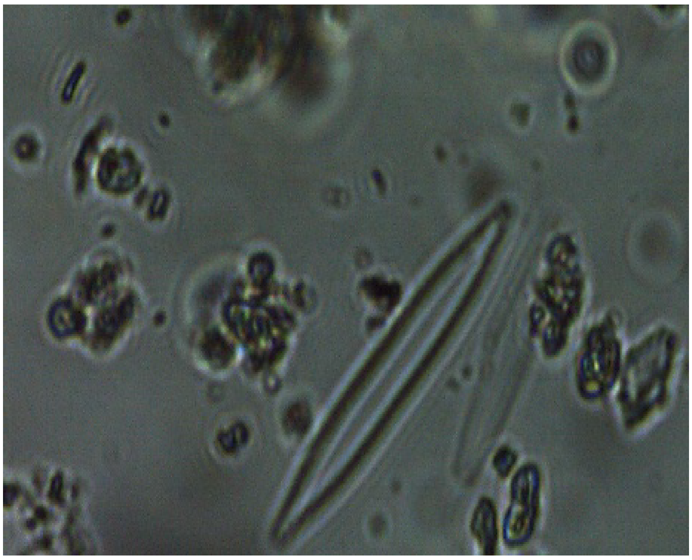

Figure 1. Diatom's microscopic image (100 times magnified) *Nitzschia palea*.

After preparing the culture medium, 950 ml of filtered river water, suitable for the life of diatoms, was added to the culture. Then, 10 ml of the stock was added to the river water sterilized with an autoclave (Andersen 2005). The dilution process with 4 flasks was performed under the sterile laminar hood. Samples were prepared with about 1 microlitre of vitamins according to the above ingredient. The specimens were placed on a shaker illuminated with fluorescent light for 8 and 16 hours of the light cycle and air-pumps and for 20 days. Light intensity was 40 µE $m^{-2}s$ adjusted with the luxi meter. In aerated conditions, the crops were continuously and uniformly aerated by the air pump. The cultivation of diatoms continued for 14 days. The laboratory temperature was fixed between 12°C and 10°C. After 10 days, the specimens were transferred to the new culture medium. The solution was checked several times to ensure that no other organisms were present. Then, at the end of the work, the samples were centrifuged and washed three times with distilled water.

Treatment with heavy metals

Three concentrations of $K_2Cr_2O_7$ as chromium VI (Hörcsik 2006) were used to treat diatoms with heavy metals (2-5-10 ppm). MOPS was used to fix the pH of the suspension. The absorption of metal was determined by atomic absorption spectrometry (Fig. 2).

Results and Discussion

The maximum chromium absorption in the first to 14th day was observed at 5PPM concentration (Fig. 3). Our results showed that Mean and standard deviation of control and treatment of different concentrations related to the first day and 14th in Fig. 4 and 5 and Tab. 1.

ANOVA-Oneway of three different concentrations on day 1 and 14th. Considering the sig column, we find that the effect of the date on chromium absorption was significant.

The F2 is a common medium for the growing algae, especially diatoms (Guillard and Ryther 1962). Chromium is vital for carbohydrate metabolism (Naghash Hosseini 2009). As the results indicated the highest absorption rate was recorded at the chromium's 5 ppm treatment, indicating that chromium can improve the algae growth to some extent (Fig. 3).

One of the factors that affect the absorption of metal ions is the solubility of metal in acid. The pH is effective on metal binding sites to the cell wall and on the chemistry of water-soluble ions. The absorption of heavy metal

Figure 2. Treatment with heavy metals. After the preparation tube was inserted into the samples for aeration and placed on the shaker.

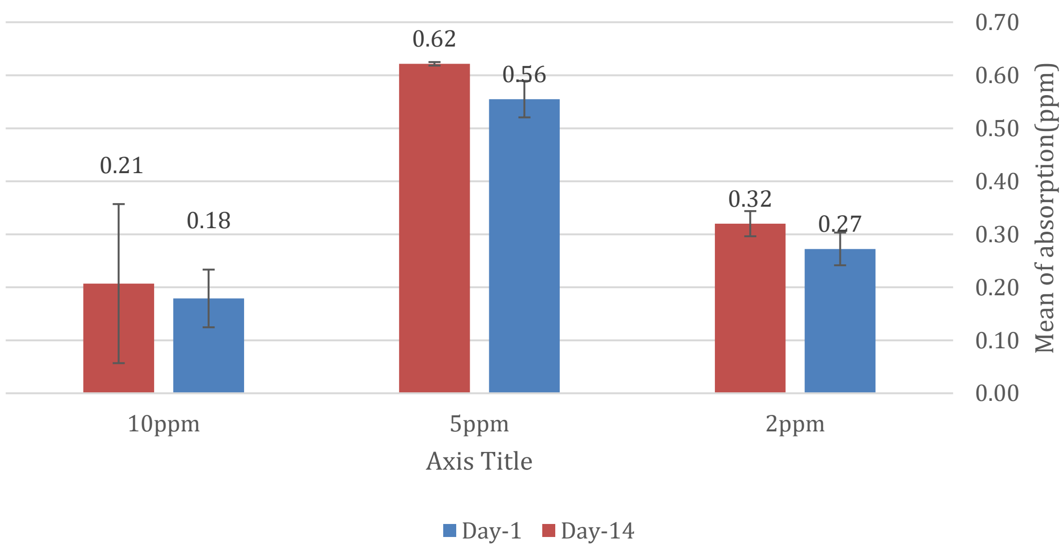

Figure 3. The absorption rate of chromium measured by atomic absorption spectroscopy. Both day 1 and day 14 had the highest adsorption at 5 ppm.

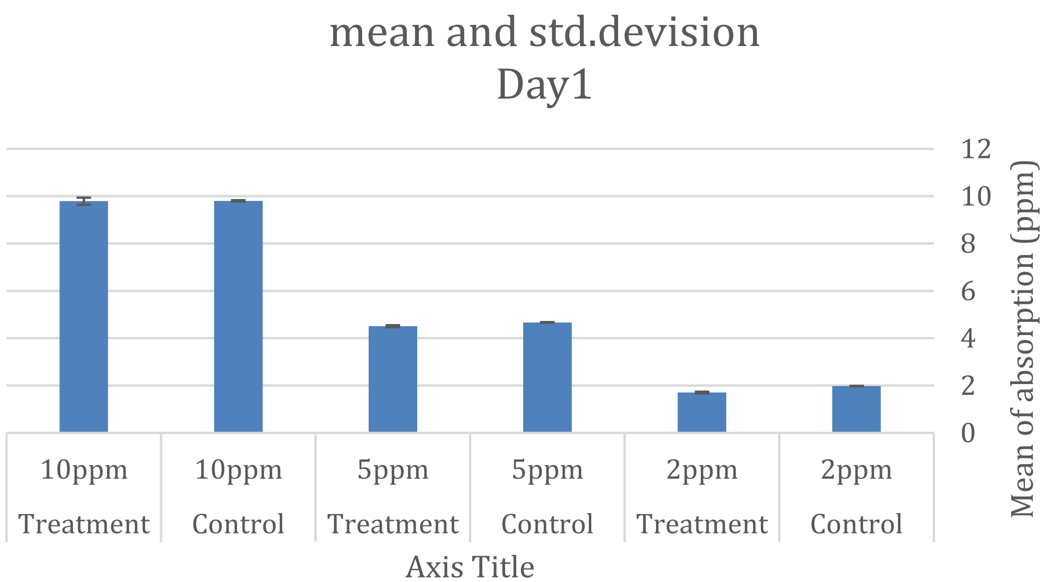

Figure 4. Mean and standard deviation of control and treatment of different concentrations related to the first day.

ions by microorganisms is divided into three groups: cell surface transplantation, intracellular accumulation, extracellular density. In the process of bioavailability, a chemical-physical reaction occurs between metal ions

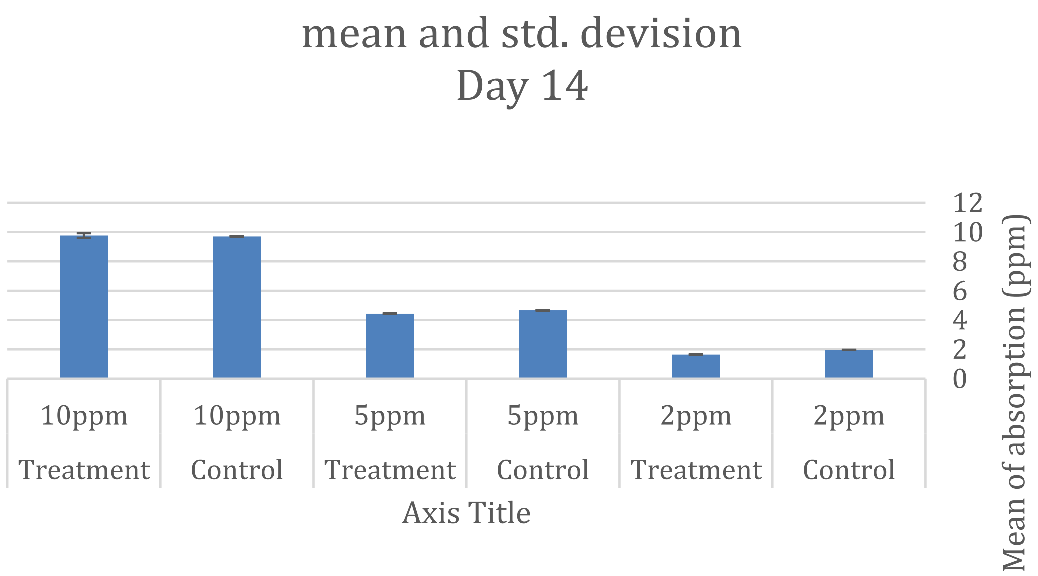

Figure 5. Mean and standard deviation of control and treatment of different concentrations related to the 14th day.

Table 1. ANOVA-Oneway of three different concentrations on day 1 and 14th.

		Sum of Squares	df	Mean Square	F	Sig.
Day.1	Between groups	101.068	2	50.534	5945.514	0.000***
	Within groups	0.051	6	0.008		
	Total	101.119	8			
Day.14	Between groups	101.981	2	50.991	6044.411	0.000***
	Within groups	0.051	6	0.008		
	Total	102.032	8			

and cellular constituents of living or dead cells. (Naghash Hosseini 2009), also, the concentration of the elements in the cell depends on its amount in the substrate (Favero et al. 1990).

Many elements combine to the carrier for transferring the cell membrane, which are proteins and some pass through pores (Williams 1981).

In a study (Hervey R.J. 2006) of the effect of chromium VI on green algae *Chlorella pyrenoidosa*, Hörcsik showed that a concentration of 20 mg dm^{-3} was toxic and deadly for this algae. The toxic properties of Cr (VI) can be due to the possibly free diffusion across cell membranes and strong oxidative potential. The toxicological effect of Cr (VI) originates from the action of this form as an oxidizing agent as well as from the formation of free radicals during the reduction of Cr (VI) to Cr (III) occurring inside the cell.

He showed that Chromium and Calcium changes in parallel with each other. Higher calcium concentrations can be observed with increasing chromium concentration, both in the cell wall system and in the whole of the cell (Hörcsik 2006). Heavy metals such as Pb, Ni and Cr are toxic for plants, fungi, and algae. In this study, chromium at a concentration above 5 ppm could not absorb well. Diou et al. (2003) reported that high chromium concentrations affect the absorption of essential elements such as potassium, and also Paris and Van den Hyde proved that there is the competition between the absorption of certain metals such as antimony and cadmium, sealant, nickel and mercury. The reason for the presence of similar ligands for these chelated compounds is competition, which may be due to the loss of potassium uptake by diatoms was observed in the presence of chromium due to competition for cell receptors for these two elements (Naghash Hosseini 2009). Saadi and Neyasi showed that the highest growth rate in diatoms is in the presence of copper ions at a concentration of 5 ppm in the presence of cadmium ion at a concentration of 0.7 ppm. Also, copper ion adsorption was highest at 16 ppm concentration and, as well with increasing copper ion concentration, the adsorption rate also increased. According to the results of this study, chromium uptake was decreased at concentrations above 5 ppm. In 2006, Hörcsik and colleagues in a study on *Chlorella pyrenoidosa* algae showed that chromium and calcium concentrations exhibit parallel changes and increase the concentration of chromium (both in the wall and inside the cell) by increasing the concentration of calcium, They proved lethal concentrations for the growth of this algae at 20 mg dm^{-3} (Hörcsik 2006). Hernandez and his colleagues in 2017 noted that diatoms have the ability to exchange cations of some elements other than hexavalent chromium since chromium six has a very low exchange in diatoms. Diatoms are not capable of performing anion exchange, but with sufficient activation, they are used as anionic converters. As mentioned above, in this study, the highest chromium uptake was at 5 ppm and at a lower concentration than other elements such as copper and cadmium absorbed. (Hernández-Ávila et al. 2017).

Naghash Hosseini 2009 study on fungi *Pleurotus ostreatus* showed that increased chromium concentration significantly reduced potassium uptake. There are ligands on the cell membrane that transfer the compounds from the environment to the cell, there is a competition between the absorption of some metals such as Sb, Ni, Cd and Hi due to the availability of similar ligands. Chromium concentrations may affect the absorption of essential elements, such as potassium, decreasing or increasing chromium absorption due to competition for cell surface receptors for these two elements (Naghash Hosseini 2009).

Studies by Torres and colleagues (1995) on the *Phaeodactylum tricornutum* diatom showed that there was a heterogeneous complex in the peptide composition of the membrane of diatoms that contains labile sulfur in addition to a heavy element. In this combination, the heavy element is sequestered, and its free presence in the cytoplasm is prevented (Torres et al. 1996).

Hervey's (1949) in a study of the effect of chromium on Chlorophyceae and diatoms showed that chlorococals were more tolerant than diatoms for chromium uptake. He showed that diatoms do not tolerate high concentrations of 0.32-0.032 ppm (Hervey 1949). In our study, the absorption was decreased by increasing chromium concentration above 5 ppm.

Conclusion

Due to the industrialization of life and the presence of pollutants and heavy metals in sewage, the use of diatoms as biological removers is considered. This study shows Nitzschia's role in chromium absorption. It needs to mention that *Nitzschia palea* does not have too high adsorption rate of chromium, However, it has been introduced in many sources as a resistant species which makes is a good candidate, for removal of other heavy metals.

References

1. **Akar B., Şahin B. 2017.** Diversity and ecology of benthic diatoms in karagöl lake in karagöl-sahara national park (Şavşat, Artvin, Turkey). *Turk J Fish Aquat Sci.* **17:** 15-24. http://www.trjfas.org/uploads/pdf_967.pdf
2. **Amato A. 2010.** Diatom reproductive biology: Living in a crystal cage. *Int J Plant Reprod Biol.* **2:** 1-10. https://www.academia.edu/7766233/Diatom_Reproductive_Biology_Living_in_a_Crystal_Cage
3. **Anantharaj K., Govindasamy C., Natanamurugaraj G., Jeyachandran S. 2011.** Effect of heavy metals on marine diatom *Amphora coffeaeformis* (Agardh. Kutz). *Glob J Environ Res.* **5:** 112-117. https://www.semanticscholar.org/paper/Effect-of-Heavy-Metals-on-Marine-Diatom-Amphora-Anantharaj-Govindasamy/bdb36bfec247c782459af4199f639d67cbc05e54
4. **Andersen R.A. (Ed.). 2005.** Algal culturing techniques. Elsevier. https://www.academia.edu/28995029/Andersen_Algal_Culturing_Techniques
5. **Cherifi O., Sbihi K., Bertrand M., Cherifi K. 2016.** The removal of metals (Cd, Cu and Zn) from the Tensift river using the diatom *Navicula subminuscula* Manguin: A laboratory study. *Int J Adv Res Biol Sci.* **3:** 177-187. http://dx.doi.org/10.22192/ijarbs.2016.03.10.024
6. **Favero N., Bressa G., Costa P. 1990.** Response of *Pleurotus ostreatus* to cadmium exposure. *Ecotoxicol Environ Saf.* **20:** 1-6. https://doi.org/10.1016/0147-6513(90)90039-8
7. **Fisher N.S., Bohe M., Teyssie J.L. 1984.** Accumulation and toxicity of Cd, Zn, Ag, and Hg in four marine phytoplankters. *Mar Ecol Prog Ser.* **19:** 201-213. https://doi.org/10.3354/meps018201
8. **Garrido B.A., Romo S., Villena M.J. 2013.** Diatom species composition and indices for determining the ecological status of coastal mediterranean spanish lakes. In *Anales del Jardín Botánico de Madrid.* **70:** 122-135. https://doi.org/10.3989/ajbm.2373
9. **Guillard R.R.L., Ryther J.H. 1962.** Studies of marine planktonic diatoms. I. Cyclotella nana Hustedt and Detonula confervacea Cleve. *Can J Microbiol.* **8:** 229-239. https://doi.org/10.1139/m62-029
10. **Hernández-Ávila J., Salinas-Rodríguez E., Cerecedo-Sáenz E., Reyes-Valderrama M., Arenas-Flores A., Román-Gutiérrez A., Rodríguez-Lugo V. 2017.** Diatoms and their capability for heavy metal removal by cationic exchange. *Metals.* **7:** 169. https://doi.org/10.3390/met7050169
11. **Hervey R.J. 1949.** Effect of chromium on the growth of unicellular Chlorophyceae and diatoms. *Botanical Gazette.* **111:** 1-11. https://www.jstor.org/stable/2472872?seq=1
12. **Hörcsik Z. 2006.** Effect of chromium (VI) on growth, element and photosynthetic pigment composition of *Chlorella pyrenoidosa*. *Acta Biologica Szegediensis.* **50:** 19-23. http://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.318.5629&rep=rep1&type=pdf
13. **Ilyin I., Berg T., Dutchak S., Pacyna J. 2004.** EMEP assessment part I European perspective. *Norwegian Meteorol Institute, Oslo, Norway.* 107-128. https://emep.int/publ/reports/2004/assessment/Part1_001-010_00a-Title.pdf
14. **Juwarkar A.A., Jambhulkar H.P. 2008.** Phytoremediation of coal mine spoil dump through the integrated biotechnological approach. *Bioresource Technol.* **99:** 4732-4741. https://doi.org/10.1016/j.biortech.2007.09.060
15. **Naghash H., Hadith Al-Sadat. 2009.** Evaluation and identification of tolerance of edible fungus *Pleurotus ostreatus* to different concentrations of Cr^{3+} MSc Master's Degree in Biology, Ferdowsi University of Mashhad.
16. **Naghavian N. 2012.** Experimental zinc stress on benthic diatoms assemblage growth in Fyrisan water. *Swedish Uni Agri Sci.* https://stud.epsilon.slu.se/4994/11/naghavian_n_121107.pdf
17. **Olaizola M., Duerr E.O., Freeman D.W. 1991.** Effect of CO_2 enhancement in an outdoor algal production system using Tetraselmis. *J App Psychol.* **3:** 363-366. https://doi.org/10.1007/BF00026099
18. **Palmer C.M. 1980.** Algae and water pollution: The identification, significance, and control of algae in water supplies and in polluted water. Castle House Publications.
19. **Sala Cossich E., Granhen Tavares C.R., Kakuta Ravagnani T.M. 2002.** Biosorption of chromium (III) by *Sargassum* sp. biomass. *Electron J Biotechnol.* **5:** 6-7. http://www.bioline.org.br/pdf?ej02024
20. **Sbihi K., Cherifi O., Bertrand M. 2012.** Toxicity and biosorption of chromium from aqueous solutions by the diatom *Planothidium lanceolatum* (Brébisson) Lange-Bertalot. *Am J Scienti and Indus Res.* **3:** 27-38. https://doi.org/10.5251/ajsir.2012.3.1.27.38

21. **Singh R., Gautam N., Mishra A., Gupta R. 2011.** Heavy metals and living systems: An overview. *Indian J Pharmacol.* **43:** 246-253. https://doi.org/10.4103/0253-7613.81505

22. **Supriya G., Ramachandra T.V. 2011.** Chronicle of marine diatom culturing techniques. *Indian J Fundamental and App life Sci.* **1:** 282-294. http://wgbis.ces.iisc.ernet.in/energy/paper/marine_diatom_culturing/isolation.htm

23. **Torres E., Cid A., Fidalgo P., Herrero C., Abalde J. 1996.** Tolerance and detoxification mechanisms in marine diatom *Phaeodactylum tricornutum* exposed to cadmium. *J Marine Biotechnol.* **9:** 944. https://core.ac.uk/download/pdf/61913724.pdf

24. **Williams R.J.P. 1981.** Physico-chemical aspects of inorganic element transfer through membranes. *Philos Trans R Soc Lond B Biol Sci.* **294:** 57-74. https://doi.org/10.1098/rstb.1981.0089

25. **Zhang Y., Tang Y., Yi R. 2017.** Research on the characteristics of benthic diatom in the longjiang river in guangxi and its relationships with heavy metals ions pollution. *Chem Eng Transactions.* **62:** 37-42. https://doi.org/10.3303/CET1762007