

CELLS IMMOBILIZATION OF SOME MICROORGANISMS AS A TOOL FOR BIOREMEDIATION : A- *CHLORELLA VULGARIS*

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(Received 27 June 2019, Revised 18 September 2019, Accepted 25 September 2019)

ABSTRACT : The present study included the use of immobilization technique for some microorganisms (Algae, Fungi and Bacteria) and studying their efficiency in treating some pollutants such as heavy metals. Standard solutions of lead and cadmium at concentrations (10, 20 and 30) ppm. The bioremediation was analysis by using atomic absorption device, FTIR technique and SEM. *Chlorella vulgaris* algae was selected to immobilized and test its ability to treatment heavy metals in aqueous solutions. The lead concentrations after treatment algae were (4,703, 4,863 & 5.798) ppm with a removing ratio (53, 76 & 81)% for each concentration, respectively, and for cadmium were:3.151 ppm and 6.935 ppm finally 7.264 ppm, and emoval Efficiency was 68%,65% and 76%. Results show the immobilization of algae will in general lead to increment trace elements gathering by biomass and finally increase the removal efficiency to remove the trace elements from aqueous solution.

Key words : Immobilization, *Chlorella vulgaris*, heavy metals.

INTRODUCTION

Enormous scale generation of sewageare outcome of every modern society. Generality of sewage is generally unsafe for human beings populates and earth and must manage before transfer for all surface water (Zhou *et al*, 2008; Bashan and Bashan, 2010). Physical and concoction treatment techniques have constrained achievement when connected to the treatment of recalcitrant natural toxins in light of the fact that these procedures prompted auxiliary gushing issues because of development of poisonous materials. In this way, concentrated consideration to the corruption for most poisons by micr-organisms also change, it to mixes tamely through arrangement of cells bulk (Karigar *et al*, 2006; Cozma *et al*, 2012). Trace elements are broadly utilized in the manufacture such as, material, cowhide, paper, electroplating, chrome plating, oil refining, paint, converting factories. The manufactures release huge amounts of dangerous material and the untreated discharge to the earth and cause a genuine environmental sullyng (Wang, 2002). Immobilized cells display more various suitable state of affairs than liberated cells, for example, reuse of biocatalysts, rise quantity profitability, amended method monitor and diminished defenselessness of cells to sullyng (Ksungur *et al*, 1999). Among the distinctive cell immobilization strategies, ensnarement in calcium alginate gel has been one of the most utilized

lattices for entire cell capture because of its effortless and non-dangerous character. This straightforward and gentle immobilization method includes the drop-wise expansion of cells suspended in sodium alginate onto an answer of calcium chloride whereon the cells are immobilized in encouraged calcium alginate gel as globules (Rosevear, 1984). Here, we have been keen on get together of explicit cells on surfaces from the perspective of utilizing the cells as wellsprings of compounds for biotransformations and combination of new materials.

MATERIALS AND METHODS

Algae selection

Chlorella vulgaris was isolated from the Biology Department Laboratories and cultivation it in the CHU 10 culture medium (Chu, 1942) and modified by Kassim (Al-Asady, 2014). The medium was prepared in Stock Solutions. Algae isolates were cultured by Batch culture method in a 250 ml from cultured media in 500 ml conical flasks and 50 ml of pure cultures were incubated in Plant Cabinet at 25 ±2 and with light intensity 50 ìm / m² / s and 16: 8 hours light:dark.

Beads formation

About 100 ml from studied organisms culture in the stationary phase had been taken and concentrated by filtering by Whattman 20 filter paper, then the concentrated organisms were mixed with an equal volume

of the 2% sodium alginate soluble solution and well shaken to homogenize this ingredients and put the mixture in a medical syringe. At this time, calcium chloride solution (1M) was prepared in a separate beaker and the contents of the syringe are gradually dropped in the calcium chloride solution. A drop of solution (organisms and sodium alginate) is solidify and become immobilized in the form of beads in the beaker and left for 5-10min. Then the beads (immobilized organisms) are separated from calcium chloride solution by the tea strainer and wash gently with tap water and rinse thoroughly with distilled water (Adlercreutz and Mattiasson, 1982).

Measurement by atomic absorption

Lead and cadmium were selected to test the efficiency of immobilized organisms with sodium alginate in their treatment, three standard concentrations of each metal (10, 20, 30) mg / L from (Sigma-Aldrich) and from each standard concentration of 10 ml for use in the experiment.

A bioreactor was constructed as shown in Fig. 1. The heavy metals sample was placed in the reactor and left for 15 minutes for each concentration/per metal. The sample was then collected and measured by atomic absorption.

Removal efficiency (R.E.) was calculated as below:

$$R.E. = \frac{C1 - C2}{C1}$$

Where, R.E: Removal efficiency, C1: Heavy metal concentration before treatment, C2 : Heavy metal concentration before treatment.

Diagnostic of treatment according to the functional



Fig. 1 : Bioreactor used in research.

group by FT-IR and according to surface binding between immobilized algae and heavy metals by FE-SEM.

RESULTS AND DISCUSSION

Results of the present study, immobilized algae (*Chlorella vulgaris*) were used to show their ability to treat lead concentrations by using Standards concentrations (10, 20 & 30) ppm. The lead concentrations after treatment with these immobilized algae were (4,703, 4,863 & 5.798) ppm. Respectively (Fig. 2), with a removing ratio (53, 76 & 81%) for each concentration, respectively (Fig. 3). Results of this study shows that the concentrations of cadmium after treatment by immobilized *Chlorella vulgaris* were 3.151 ppm and 6.935 ppm finally 7.264 ppm when the initial concentrations before treatments were (10, 20 & 30) ppm respectively (Fig. 4).

Removal Efficiency was 68% for initial concentration

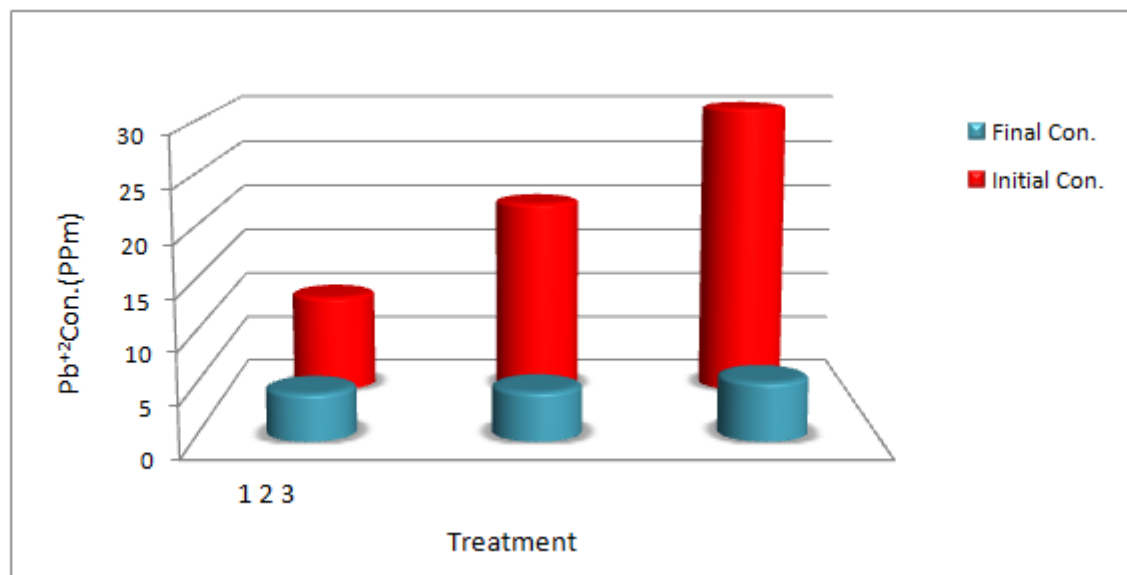


Fig. 2 : Lead Concentration before and after treatment.

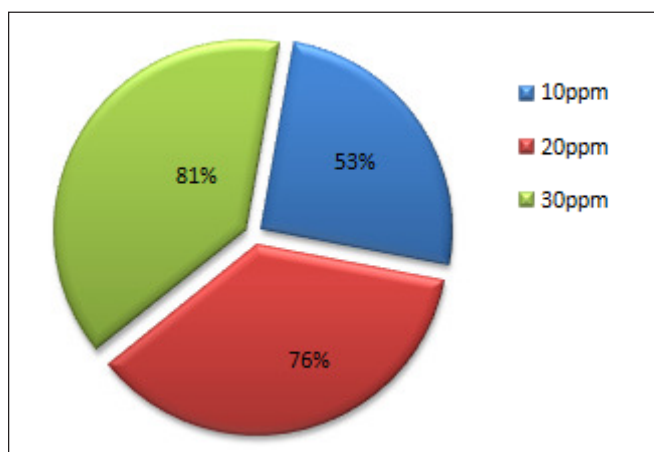


Fig. 3 : Removal efficiency of Immobilized *Chlorella vulgaris* for lead.

10ppm, 65% and 76% for initial concentration 20 ppm and 30 ppm, respectively (Fig. 5).

From the result, *Chlorella vulgaris* has been able to adsorb and remediate the trace element and decrease its concentration in aqueous solution containing it because *C. vulgaris* has particular and non-particular positions had been bound trace elements (Mehta and Gaur, 2001) and in this manner, sensibly be understand that the immobilized algae have more No. of negatively charged locales/identity organisms for elements bio-adsorption (Knauer *et al*, 1997). Ionic charge and covalent bonds are engaged with bio-adsorption and the wall of cell components are likewise known to assume a critical function in bio-adsorption (Crist *et al*, 1981; Mahan *et*

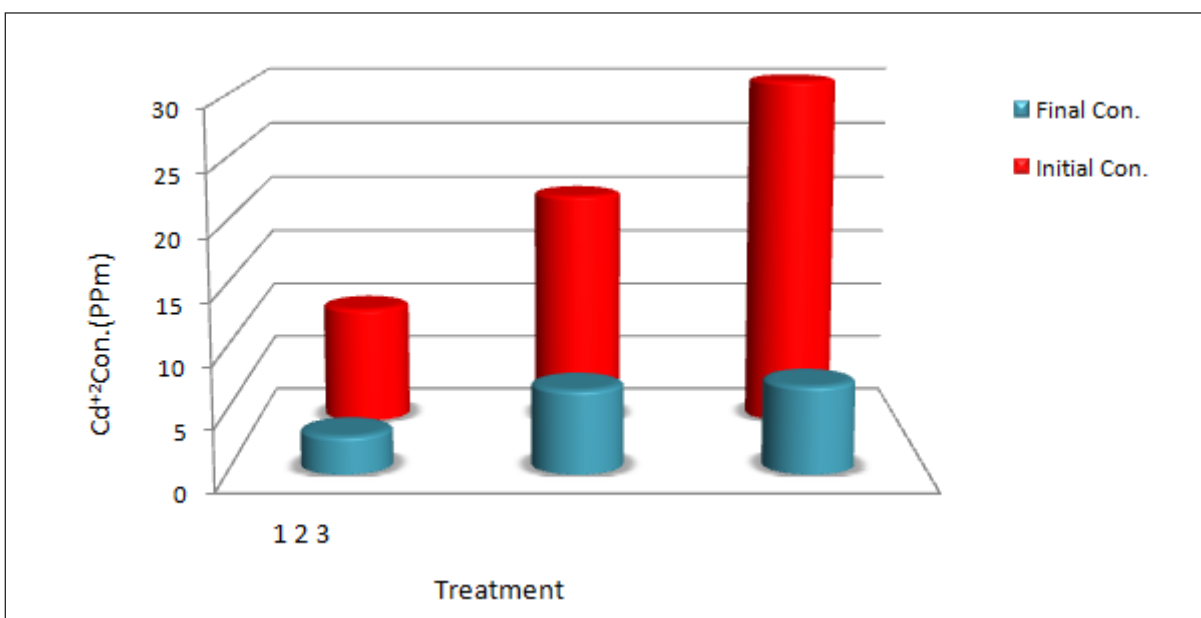


Fig. 4 : Cadmium concentration before and after treatment.

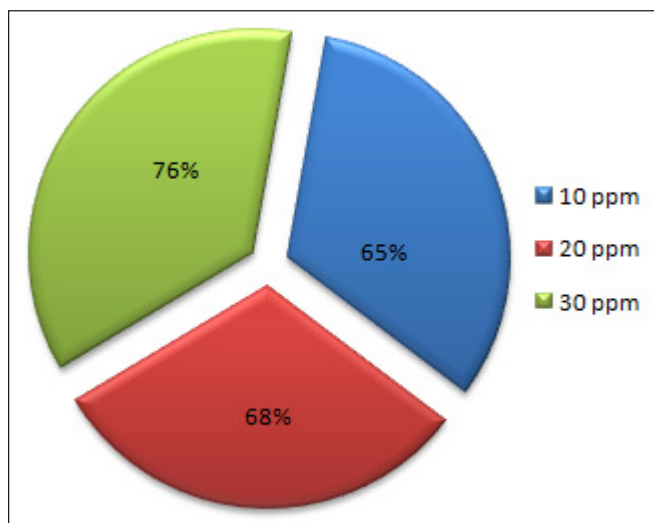


Fig. 5 : Removal efficiency of Immobilized *Chlorella vulgaris* for cadmium.

al, 1998; Knauer *et al*, 1998). These results are identical to Chong *et al* (2000) and Hamdy (2000).

Curves of the FT-IR examination (Figs. 6, 7) found a clear change in the values of wavelengths resulting from the process of linking of trace element bonds found after treatment by calcium alginate and Algal cell surface demonstrates the task of various groups to particular practical gatherings that have a place with contrast class of macromolecules in the control tests just as annoyance in the band structure after Pb & Cd treatment. The variation in FTIR peaks and their transmission were because of the reaction with the active sites on the *Chlorella vulgaris* and trace elements through bio-adsorption operation. The Scanning Electron Microscope (Figs. 8, 9) showed the aftereffects of treatment procedure. So as to check whether the layer was as yet

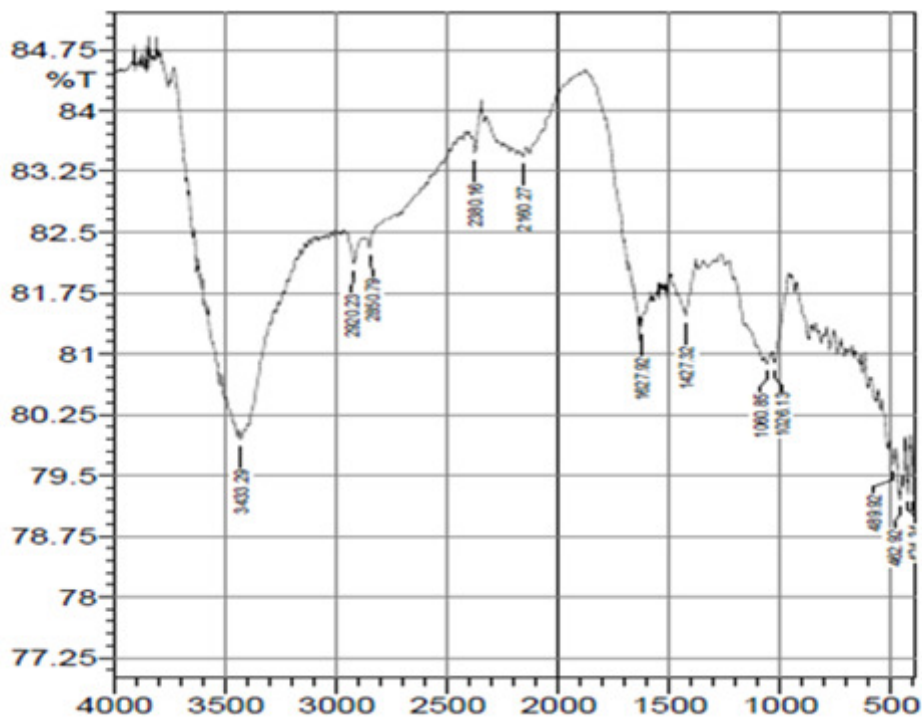


Fig. 6 : FT-IR for Immobilized *Chlorella vulgaris* that treated Pb-10 ppm.

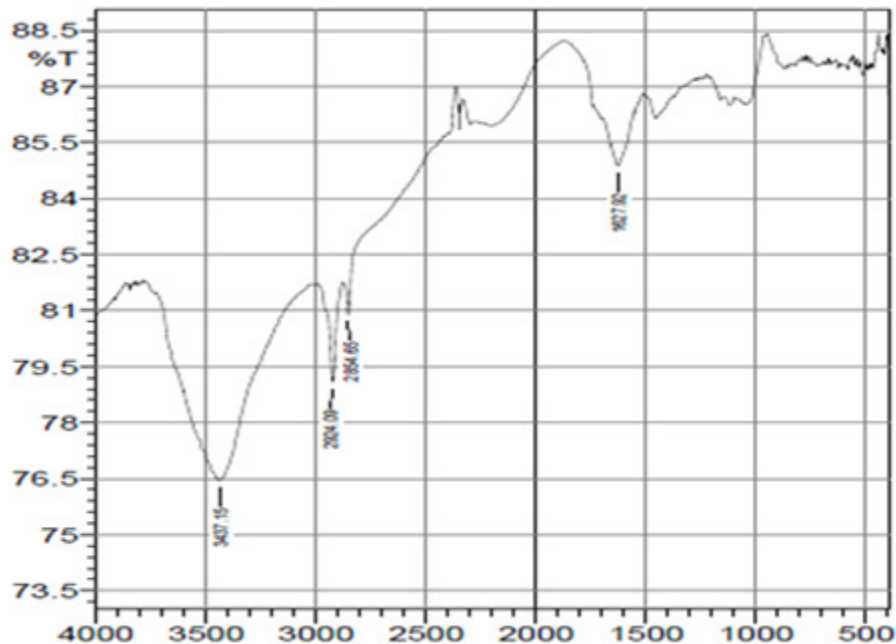


Fig. 7 : FT-IR for Immobilized *Chlorella vulgaris* that treated Pb-20 ppm.

adsorbed of lead and cadmium on the surface of beads and some concentrations were penetrate inside beads, backscattered examining of and photos investigation were with un-wet beads. With the assistance of this innovation, components with high nuclear No. can be pictured. Because of this technique scattering they showed up as candid sites on photos, in contrast previous results that demonstrated the SEM for Algal beads without any treatment. This photo explain the raw material of Calcium Alginate without any white site of adsorbed lead and show

the porosity of beads that mean the ability of beads to adsorbed lead and penetrate, it to inside beads to increase the removal efficiency results of adsorption showing in Figs. 8 and 9.

CONCLUSION

The immobilization of algae will in general lead to increment trace elements gathering by biomass and finally increase the removal efficiency to remove the trace elements from aqueous solution.

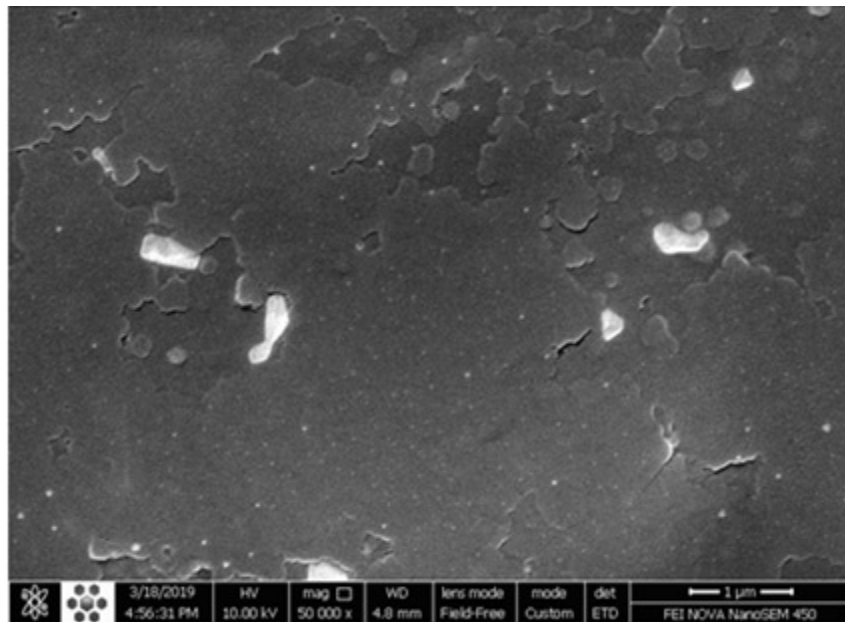


Fig. 8 : SEM of immobilized *Chlorella vulgaris* surface with treat Pb-10 ppm.

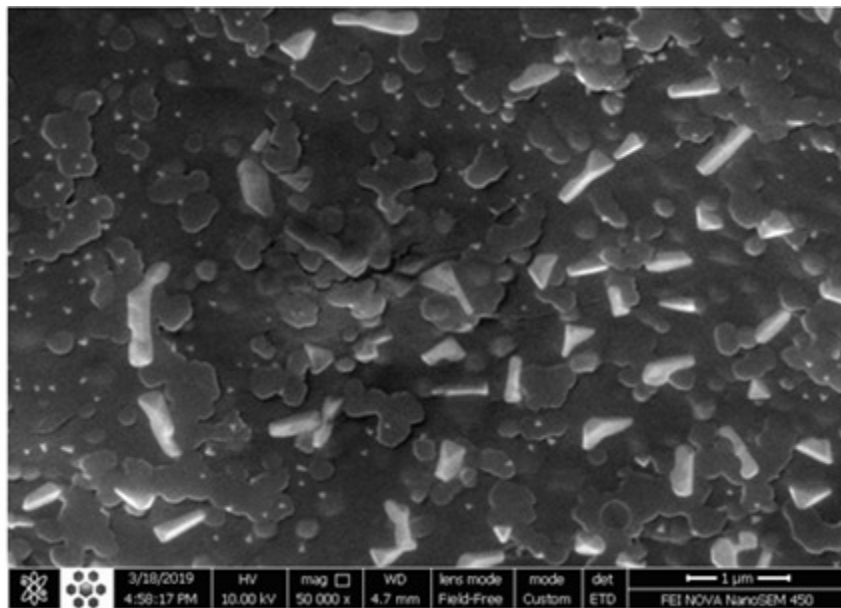


Fig. 9 : SEM of immobilized *Chlorella vulgaris* surface with treat Pb-20 ppm.

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