Bioscience and Bioengineering
Vol. 1, No. 2, 2015, pp. 17-21
http://www.aiscience.org/journal/bio

Biosorption of Zinc (Zn^{2+}) and Iron (Fe^{2+}) from Wastewater Using *Botrydium Granulatum* and *Euglena Texta*

B. O. Olusola, M. N. Aransiola*

Department of Microbiology, Faculty of Science, University of Ibadan, Ibadan, Nigeria

Abstract

One of the most serious environmental problems being faced by the world today is heavy metal pollution. Biosorption, being a cost-effective and efficient, has emerged as alternative technology for removal of heavy metals. The potential of two immobilized algae species for removal of Zinc (Zn^{2+}) and Iron (Fe^{2+}) from industrial wastewater was analysed through batch studies. Algal bloom water samples were obtained from fish ponds in Prof. Olu Odeyemi's Farm, Ilesa. Wastewater was collected from industrial discharge system around Ile–Ife (Nigeria). Algae were isolated from the water samples using pour plate technique on modified Allen agar medium at pH 7.1. The identity of the algae isolates was established using Identification guide to Freshwater and Terrestrial algae. Algae were immobilized with 3% sodium alginate and 2% calcium chloride dihydrate. The beads of approximately 4mm were formed by allowing the algae/alginate mixture to drip through a syringe barrel in to a beaker of calcium chloride dihydrate solution. Biosorption batch studies was carried out by the addition of 100 beads into 100ml solution (50 ml of wastewater and 50 ml of media) in 250ml conical flask at 150rpm, room temperature and pH 3, 5 and 7. Concentration of Zinc (Zn^{2+}) and Iron (Fe^{2+}) was determined in 2 days interval using atomic absorption spectrophotometer at their respective detection limit. The algal isolates were identified as *Botrydium granulatum* and *Euglena texta*. The percentage removal of Zinc (Zn^{2+}) and Iron (Fe^{2+}) (at their respective detection limit) by these algae varies with pH and time. At pH 5, *Botrydium granulatum* had the best percentage removal rate (100%) (that is not detectable in the wastewater) for the two metals while *Euglena texta* performed best (100%) at pH 3 and 5 for the two metals over the period of time. At pH 7, the percentage removal of the two metals decreases with time for both algae species. At pH 5, *Euglena texta* performed better by complete (100%) removal of both metals starting from day 1 while *Botrydium granulatum* completely (100%) remove Iron (Fe^{2+}) and Zinc (Zn^{2+}) starting from day 7 and 5 respectively. Zinc (Zn^{2+}) was more effectively removed than Iron (Fe^{2+}) by *Botrydium granulatum* at pH 5. Since the algae isolates showed appreciable sorption of the metals, they can be used for biosorption of metals from industrial wastewater prior to its discharge to avoid environmental pollution.

Keywords

Biosorption, *Botrydium Granulatum*, Immobilization, Wastewater, Zinc (Zn^{2+})

Received: April 9, 2015 / Accepted: April 25, 2015 / Published online: May 22, 2015

1. Introduction

Removal of heavy metal from aqueous solutions is an important issue faced by industries discharging waste water containing heavy metals. Thousands of tons of heavy metals are discharged from industrial processes such as plastics manufacturing, mining, electroplating, and metallurgical processes. A number of physicochemical methods, such as chemical precipitation, adsorption, ion exchange, membrane separation, etc., have been commonly employed for toxic

* Corresponding author
E-mail address: aransiolaunique@yahoo.com (M. N. Aransiola)

metals removal from wastewaters [1]. However, these methods have several disadvantages, such as incomplete metal removal, expensive equipment and monitoring system requirements, high reagent or energy requirements and generation of toxic sludge or other waste products that require disposal. Furthermore, they may be ineffective or extremely expensive when metal concentration in wastewater is in the range 10-100 mg l -1 [2]

Biosorption of heavy metals from aqueous effluents is a process that has proven very promising. The major advantages of biosorption technology are its effectiveness in reducing the concentration of heavy metal ions to very low levels. The advantages of biosorption over conventional treatment methods include: low cost, minimization of chemical and/or biological sludge regeneration of biosorbent, no additional nutrient requirement, high efficiency and the possibility of metal recovery [3].

One of the main interests for microalgae is focused on their use for heavy metal removal from effluents.

Many efforts have been made to test the efficacy of immobilized algae and cyanobacteria for the removal of heavy metals from aqueous solution. Immobilization is a general term that describes many different forms of cell attachment or entrapment [4]. Various techniques, such as flocculation, adsorption on surfaces, covalent binding to carriers, crosslinking of cells, encapsulation in polymer gel and entrapment in polymeric matrix, are used for cell or biomass immobilization. Immobilization generally tends to increase more metal accumulation by biomass. Size of the immobilized beads is a crucial factor for use of immobilized biomass in biosorption. More than 15 microalgal species have been already studied for their potential in heavy metal removal [5]. Immobilized Chlorella salina cells in alginate were used to remove Co, Zn, and Mn heavy metals [6]. Chlorella vulgaris beads were more efficient in heavy metal removal from sewage than free cells. The efficiency in Iron, Nickel, and Zinc removal was higher in the immobilized cells than free cells by 27, 23 and 25% respectively [7].

Objective of the study was to analyse the potential of immobilized algae *Botrydium granulatum* and *Euglena texta* (in alginate) for removal of Zinc (Zn^{2+}) and Iron (Fe^{2+}) from industrial wastewater through batch studies.

2. Materials and Methods

2.1. Materials

Algal bloom water samples were obtained from fish ponds in Prof. Olu Odeyemi's Farm, Ilesa, Osun State (Nigeria). Wastewater (effluent) was collected from industrial discharge system around Ile–Ife, Osun State (Nigeria). All chemicals were purchased from Sigma Aldrich and were of analytical grade.

2.2. Methods

2.2.1. Algae Isolates

Algae were isolated from algal bloom water sample by serial dilution and pour plate technique on modified Allen agar medium at pH 7.1. The plates were inverted and incubated for about 1 week under steady illumination at room temperature. After incubation period, isolated colonies were selected, carefully picked with a sterile inoculating needle and then re-suspended inside sterile modified Allen medium at pH 7.1. The flasks were incubated for about 1 week under steady illumination at room temperature inside a laminar flow hood. From this, a drop was taken and placed on a glass slide, covered with cover slip and then observed under light microscope with X40 objective for observation of morphology and size of the algal sample. Several observations were done for each flask to ensure pure culture. The shape, morphology and possible extracellular structures observed were compared with those documented in Identification guide to Freshwater and Terrestrial algae [8]. Biomass was then harvested by filtration, washed with generous amounts of deionized water, resuspended and washed again. A mixed culture flask was further purified by serial dilution and pour plate technique.

2.2.2. Immobilization of Algae with Calcium Alginate

Sodium alginate solution (3%) was prepared by the addition of 3g of sodium alginate powder in 100ml of pure distilled water. Calcium chloride solution (2%) was prepared by adding 4g of calcium chloride dihydrate powder in 200ml of pure distilled water. The two solutions were autoclaved for 15mins at 121^{o}C. After the solutions were cooled to room temperature, 10ml of the concentrated algae culture was added to the sodium alginate solution and mixed well. A 10ml syringe barrel of algae/alginate mixture was clamped above a beaker of calcium chloride dihydrate solution. The algae/alginate mixture was allowed to drip through and form beads in the beaker while the beaker was gently swirled. The beads were allowed to harden for few minutes before straining them out of the beaker. The beads were rinsed with sterile distilled water and then kept in a stoppered bottle of distilled water in refrigerator. The beads were approximately 4mm in diameter. Blank beads were prepared without adding the culture.

2.2.3. Batch Studies

The batch adsorption studies were carried out in 250ml Erlenmeyer flasks at room temperature. Experiments were conducted for different pH conditions (3, 5 and 7) and known initial concentration of Zinc (Zn^{2+}) and Iron (Fe^{2+}) in the industrial wastewater. The flasks were placed in orbital shaker with continuous shaking at 150rpm at room temperature. 100 beads were counted (Blank and Algal beads/cubes) and mixed with 100 ml solution in 250ml conical flask. The 100ml solution contains 50ml of wastewater and 50ml of media. At regular intervals (once in 2 days) 5ml of sample was taken and concentration of Zinc (Zn^{2+}) and Iron (Fe^{2+}) was checked using atomic absorption spectrophotometer at their respective detection limits.

2.2.4. Sample Digestion and Analysis

Sample digestion was carried out according to standard analytical methods using Nitric acid (HNO_3), Hydrochloric acid (HCl) and Sulphuric acid (H_2SO_4) at relatively low temperature as reported by [9] and [10]. For the analysis, Atomic Absorption Spectrophotometer (Pye Unicam Model) was calibrated with standard solutions and the digested sample was analyzed for the presence and concentrations of Zinc (Zn^{2+}) and Iron (Fe^{2+}) at their respective detection limits. The percentage of heavy metal removed was calculated as:

$$\%\ heavy\ metal\ removal = \frac{Initial\ heavy\ metal\ conc - Final\ heavy\ metal\ conc}{Initial\ heavy\ metal\ concentration}\ X\ 100$$

3. Results

The ability of *Botrydium granulatum* (Fig. 1) and *Euglena texta* (Fig. 2) to remove Zinc (Zn^{2+}) and Iron (Fe^{2+}) from an industrial wastewater was studied. At each metal's respective detection limit, the results indicated that the two heavy metals were removed by these algae species. The percentage removal of Zinc (Zn^{2+}) and Iron (Fe^{2+}) by *Botrydium granulatum* and *Euglena texta* varies with pH and time. At pH 5, *Botrydium granulatum* had the best percentage removal rate for the two metals while *Euglena texta* performed best at pH 3 and 5 for the two metals (Fig. 3 to 6). At pH 7, the percentage removal of the two metals decreases with time for both algae species (Fig. 3 to 6). At pH 5, *Euglena texta* performed better by complete (100%) removal of both metals starting from day 1 while *Botrydium granulatum* completely (100%) remove Iron (Fe^{2+}) and Zinc (Zn^{2+}) starting from day 7 and 5 respectively (Fig. 7 and 8). Zinc (Zn^{2+}) was more effectively removed than Iron (Fe^{2+}) by *Botrydium granulatum* at pH 5 (Fig. 9).

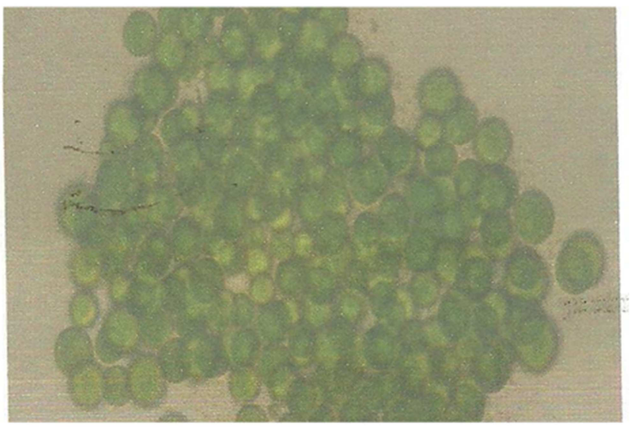

Fig. 1. *Botrydium granulatum.*

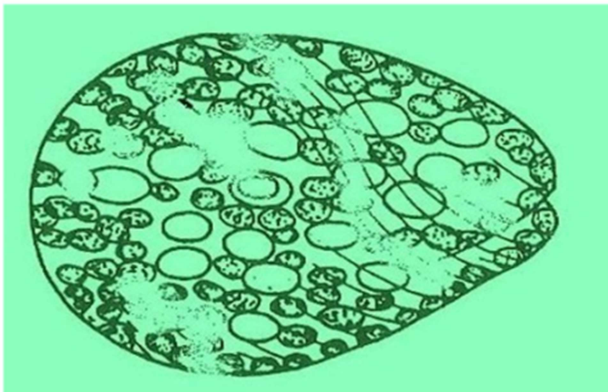

Fig. 2. *Euglena texta.*

Table 1. Initial metal concentration and their detection limit.

Metal	Initial Concentration (ppm)	Detection Limit (mg/l)
Iron (Fe^{2+})	0.52	0.05
Zinc (Zn^{2+})	0.363	0.005

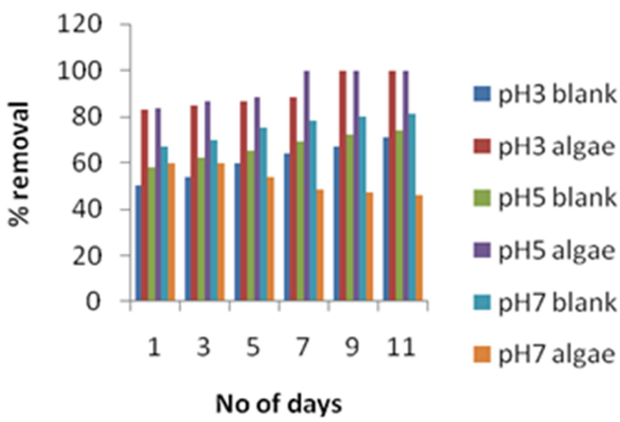

Fig. 3. Percentage removal of Iron (Fe^{2+}) by *Botrydium granulatum* with contact time.

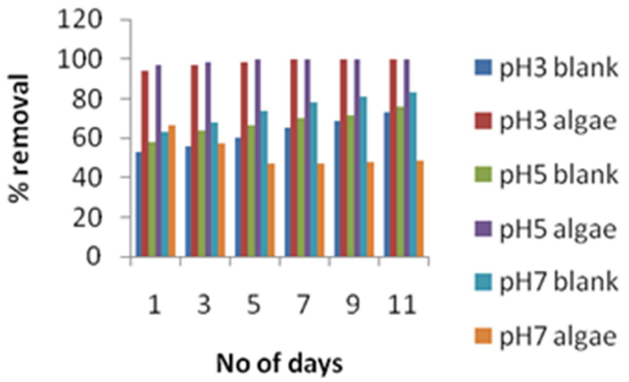

Fig. 4. Percentage removal of Zinc (Zn^{2+}) by *Botrydium granulatum* with contact time.

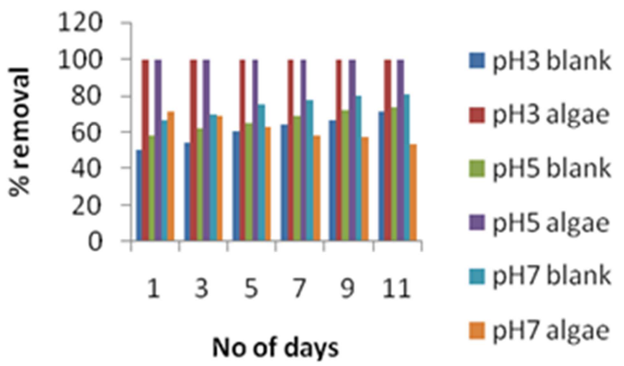

Fig. 5. Percentage removal of Iron (Fe^{2+}) by Euglena texta with contact time.

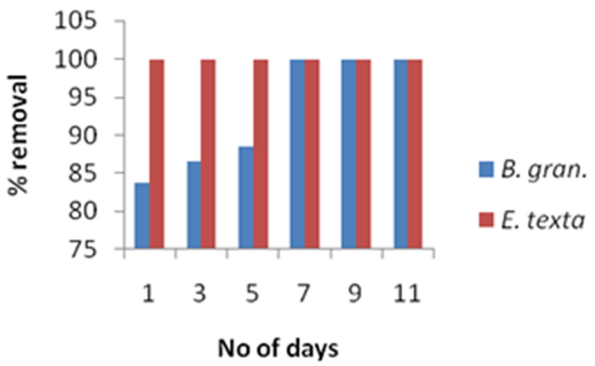

Fig. 6. Percentage removal of Zinc (Zn^{2+}) by *Euglena texta* with contact time.

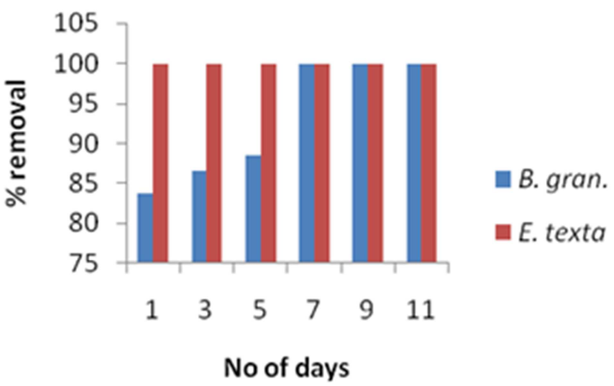

Fig. 7. Percentage removal of Iron (Fe^{2+}) by *Botrydium granulatum* and *Euglena texta* at pH 5 with contact time.

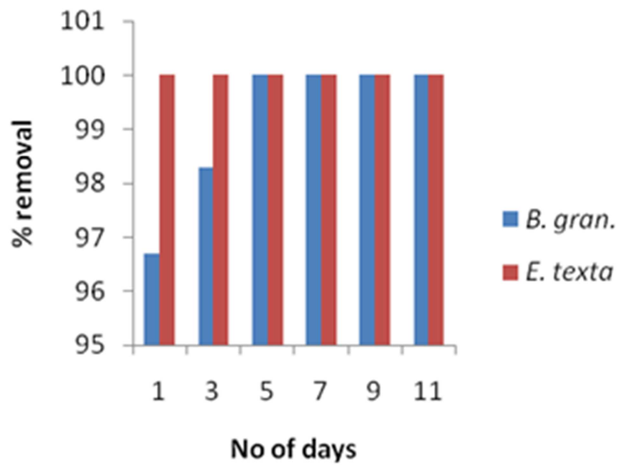

Fig. 8. Percentage removal of Zinc (Zn2+) by *Botrydium granulatum* and *Euglena texta* at pH 5 with contact time.

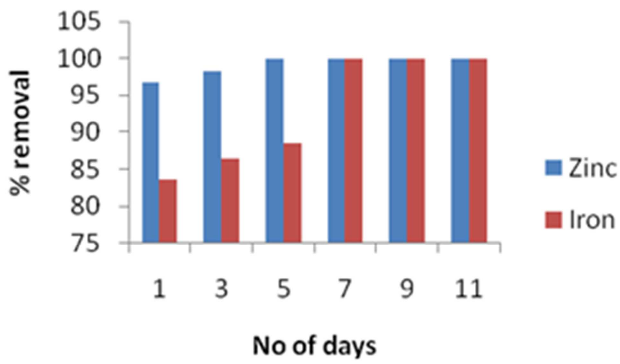

Fig. 9. Percentage removal of Zinc (Zn^{2+}) and Iron (Fe^{2+}) by *Botrydium granulatum* at pH 5 with contact time.

4. Discussion

The result of this investigation showed that *Botrydium granulatum* and *Euglena texta* (Fig. 1 and 2) were able to remove both Zinc (Zn^{2+}) and Iron (Fe^{2+}) from industrial wastewater. This was possibly due to their ability to bind metals by chemical sorbents such as Carboxylic groups, Phosphoryl groups and Amino groups on their cell surface and to intracellular ligands. This correlates with the observation of previous work reported, that algae accumulate high concentrations of metals [11]. The varied metal sorption ability of these algae species could be attributed to the fact that algae have varied cell wall composition. This is in agreement with the report of [2]. From Fig. 3 to 6 presented, *Botrydium granulatum* performed best at pH 5 while *Euglena texta* performed best at pH 3 and 5. All the algae species performed least at pH 7. This is in accordance with the report of [2] that acidic pH (3-5) is most favorable for the sorption of metal ions by algae. At pH 5, *Euglena texta* was able to remove both Iron (Fe^{2+}) and Zinc (Zn^{2+}) better (completely at their respective detection limit) than *Botrydium granulatum* (Fig. 7 and 8). This could be attributed to the fact that metal sorption ability of algae varies greatly from species to species as reported [2]. The ability of *Botrydium granulatum* to remove Zinc (Zn^{2+}) better than Iron (Fe^{2+}) (Fig 9) could be attributed to the differences in their initial concentration (TABLE 1) with Zinc (Zn^{2+}) having lower initial concentration. This is in agreement with previous report that the removal of a metal generally decreases with its increasing concentration in the solution [12]; [13].

5. Conclusion

The results of this present study have shown that algae are able to sorption heavy metals from industrial wastewater. This ability is largely dependent on pH, length of time and type of algae species.

Biosorption of Zinc (Zn^{2+}) and Iron (Fe^{2+}) by the algae species means that these algae can be used by industries for heavy metal removal from their wastewater before being discharged into the environment. This will reduce environmental pollution and improve economic value.

References

[1] H. Eccles, Treatment of metal-contaminated wastes: Why select a biological process?, Trends Biotechnol., 17, 1999, 462-465.

[2] S. K Mehta and J.P. Gaur, Use of algae for removing heavy metal ions from wastewater: progress and prospects, Crit. Rev. Biotechnol., 25, 2005, 113-152. DOI:10.1080/07388550500248571

[3] K. Ramakrishna, Ligy Philip, Bioremediation of Cr (VI) in contaminated soils, Journal of Hazardous Materials, B121, 2005, 109-107.

[4] A. Lopez, N. Lazora, A. M. Marques, The interphase technique: a simple method of cell immobilization in gel-beads. J. Microbiol. Meth, 30, 1997, 231-234.

[5] L. Brinza, M. J. Dring, M. Gavrilescu, Marine Micro and Macro-algal species as biosorbents for heavymetals, Environment Engineering Management Journal, 6, 2007, 237-51.

[6] W. G. Garnham, G. A. Codd, and M. G. Gadd, Accumulation of Cobalt, Zinc and Manganese by esturine green Chlorella salina immobilized in alginate microbeads, Environmental Science Technology, 26, 1992, 1764-70.

[7] M. S. Abdel Hameed, Continuous removal and recovery of metals by alginate beads, free and alginate immobilized chlorella vulgaris, Africal Journal of Biotechnology, 5, 2006, 1819-23.

[8] D. M. John, B. A. Whitton, A. J. Brook, The freshwater Algal Flora of the British Isles. An identification guide to Freshwater and Terrestrial Algae (Cambridge University Press and Natural History Museum, Cambridge, 2002)

[9] I. J. Tinsely, Chemical concept in pollutants behavior (John wiley and sons Inc. New York, 1979, 56 – 58).

[10] I. Walinga, V. Van, and J. J. Vanderlee, "Plant Analysis procedures" (Wageningen Agricultural Units, Soil and Plant Part 7, A Series syllabi, Netherlands. 1989, 10 – 167).

[11] L. E. Chojnacka, R. O. Canizares, S. Baena, Efficiency of ammonia and phosphorus removal from a Colombian agroindustrial wastewater by the microalgae Chlorella vulgaris and Scenedesmus dimorphus, Bioresour. Technol., 60, 2004, 259–262.

[12] S. K. Mehta, and J. P. Gaur, Characterization and optimization of Ni and Cu sorption from aqueous solution by Chlorella vulgaris, Ecol. Engin., 18, 2001, 1–13.

[13] S. K. Mehta, B. N. Tripathi, J. P. Gaur, Enhanced sorption of Cu^{2+} and Ni^{2+} by acid-pretreated Chlorella vulgaris from single and binary metal solution, J. Appl. Phycol., 14, 2002, 267-273.