Zeraatkar, A.K., Ahmadzadeh, H., Talebi, A.F., Moheimani, N.R. and McHenry, M.P. (2016) Potential use of algae for heavy metal bioremediation, a critical review. Journal of Environmental Management, 181 . pp. 817-831.

Potential use of algae for heavy metal bioremediation, a critical review

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Abstract

Algae have several industrial applications that can lower the cost of biofuel co-production. Among these co-production applications, environmental and wastewater bioremediation are increasingly important. Heavy metal pollution and its implications for public health and the environment have led to increased interest in developing environmental biotechnology approaches. We review the potential for algal biosorption and/or neutralization of the toxic effects of heavy metal ions, primarily focusing on their cellular structure, pretreatment, modification, as well as potential application of genetic engineering in biosorption performance. We evaluate pretreatment, immobilization, and factors affecting biosorption capacity, such as initial metal ion concentration, biomass concentration, initial pH, time, temperature, and interference of multi metal ions and introduce molecular tools to develop engineered algal strains with higher biosorption capacity and selectivity. We conclude that consideration of these parameters can lead to the development of low-cost micro and macroalgae cultivation with high bioremediation potential.

Keywords: Algae biomass, biosorption capacity, heavy metals, bioremediation, biofuel.

Introduction

The presence of heavy metal ions such as lead, copper, cadmium, zinc, and nickel as common contaminants in industrial wastewater leads to pollution of natural environment [1, 2]. Residual nutrients and heavy metal ions in domestic and agro-industrial wastewaters are also responsible for the pollution of rivers, lakes, and seas [3]. Biosorption and accumulation of heavy metal ions in aquatic food chains can pass to humans causing major health problems [4]. Heavy metal ions even at low concentrations can be toxic to humans. For example, lead is highly toxic and can cause damage to the nervous system, kidneys, and disturbance of vitamin D metabolism, especially in children [5]. Nickel compounds are known to be carcinogenic [6], and long-term exposure to cadmium is associated with kidney damage, bone mineral loss, increased risk of bone fractures, and reduced lung function [7]. Exploring innovative means to effectively treat wastewater can further protect global freshwater resources and aquatic ecosystems. Over five decades of research on algal-based wastewater treatment and environmental biotechnology has a potentially valuable role to play both in industrial pollution remediation and research [8, 9].

To reduce the cost of treatment, the recovery of precious metals such as gold and silver from processed waters, and extraction of radionuclides such as uranium from aqueous solutions, may have some economic benefits [10]. However, treating wastewater containing heavy metal ions is a major economic challenge. The main physicochemical approaches to remove heavy metal ions from wastewaters include chemical precipitation [11], ion exchange [12], electrokinetic [13], membrane processing [14], and adsorption [15, 16]. The high costs of chemicals at industrial scales, and incomplete removal of the heavy metal ions are among the main limiting factors in the development of physicochemical approaches. Moreover, increasingly stringent rules and restrictions on effluent discharge into the environment necessitate the use of alternative methods. Biosorption of heavy metal ions in wastewater using algae can offer an ecologically safer, cheaper, and more efficient means to remove metal ions from wastewater. Indeed algae can be used for sorption of toxic and radioactive metal ions [17], and also to recover precious metal ions like gold and silver [18, 19]. However, to achieve the desired level of treatment with live algal systems it is necessary to know the maximum autotrophic production, requiring detailed algal culture physiological characterization.

The biosorption of heavy metal ions by various mechanisms such as ion exchange, complex formation, and electrostatic interaction takes place at the micro-scale [20, 21]. Among these mechanisms, ion exchange is the most important mechanism in the biosorption of heavy metal ions by algal biomass [22, 23]. In this review article, we have focused on heavy metal ion bioremediation using algal biomass to treat wastewaters, and have critically assessed the potential venues of future research and application. We have also presented enhancements to the biosortion capacity of biosorbents and reviewed the effective parameters in the biosorption of specific heavy metal ions by algal biomass [24-26]. We have also discussed different approaches that can be used to reduce the cost of algae cultivation by linking biomass production with wastewater treatment in order to grow algae in wastewater for biological treatment of wastewater and simultaneous production of biofuel [27].

Industrial wastewater

According to global statistics the distribution of water usage is 22% in industry, 8% domestic and 70% in agriculture [28]. A big fraction of this water is discharged into the environment as wastewater. For example in Germany 1534.6 million m^3 wastewater was generated in 2010 [29]. Therefore, it is necessary to have a modern approach to treat the industrial effluents.

Disposal of such huge effluent volumes to surface waters has major implications for the environment and freshwater sources has forced authorities to regulate standards for discharging industrial wastewater (IW). The initial composition of the IW largely determines the technical and economic requirements for treatment to meet regulated discharge criteria. O’Connell *et al*. [1] published details of a number of industries that produce IW with different heavy metal ions. Some IWs can be considered an enriched medium to cultivate highly productive algal strains with high biosorption capacity in order to remove heavy metal ions. However, the presence of some heavy metal ions in IWs may interfere with the growth of algae, although their influence can be moderated with dilution or mixing of IW with organic compounds [30]. Hence, characterization of the IW in order to determine the type of pollution and available nutrients is important as it directly influences the algae growth and IW treatment [31]. In living algae cells, the ability to treat IW is dependent on the growth rate; growth rate directly determines the biomass concentration, and it in turn influences the total biosortion capacity of metal ions. However, this review focusses on the uniqueness of using algae biomass (live and non-living) for bioremediation. Detailed laboratory studies indicated that algae biomass (dead or alive) can actively remove various heavy metals. However, to date no detailed economical feasibility on such process has been conducted. It is to be noted that reliability of any process that must be tested at pilot and demonstration scale prior to any commercialization.

Bioremediation of heavy metal ions using algae

Biosorption is considered an innovative technology to remove heavy metal ions from wastewaters using predominantly inactive biomass and non-living algae. There are few reports [32] of using live algae with a limited sorption capacity as the heavy metal ions often poison the living cells. Moreover, the sorption process shows large variations based on the growth phase of algae. More specifically, living algae are affected by several environmental factors which directly influence the metal ion biosorption capacity. Absorption mechanisms in living algae are more complex than non-living algae since absorption takes place during the growth phase and intracellular uptake of heavy metal ions occur. In contrast, non-living algae cells absorb metal ions on the surface of the cell membrane and it is a kind of extracellular process [129]. Non-living algal biomass can be regarded as an assemblage of polymers (such as sugars, cellulose, pectins, glycoproteins, etc.) that are capable of binding to heavy metal cations as adsorbents with the potential for cost-effective wastewater treatment [43, 131]

The toxic level of heavy metal ions in variant algal species can be highly strain specific, which consequently determines the potential remediation capacity using a specific algal strain. In other words, a heavy metal ion may exhibit a selective interaction with one specific algal strain, in addition to differences between similar species. For example, Monteiro *et al*. [33] investigated cadmium ion removal using two strains of *Desmodesmus pleiomorphus* cells, and found a 25% difference between the capacity of cadmium biosorption using ‘L’ and ‘ACOI 561’ strains. In terms of species differences, Romera *et al*. [34] found the following macroalgal species possess differing copper sorption capacity: *Fucus spiralis* > *Ascophyllum nodosum* > *Chondrus crispus* > *Asparagopsis armata* > *Spirogyra insignis* > *Codium vermilara.* The physicochemical conditions affecting the maximum capacity of metal ion removal for different micro and macro algae strains are summarized in Table 1. This table shows that most metal ion uptake occurs at a low pH (3-5), and that dried algal biomass exhibits a greater metal ion biosorption capacity compared to live algae. The solution pH has a significant influence on dissociation of the surface functional groups of non-living algal biomass and the solution chemistry of the heavy metal ions [137, 138]. The impact of pH on metal uptake can be influenced by the surface functional groups on the biomass' cell walls, and the solution metal chemistry [139]. Table 1 also reports the optimal time for heavy metal ion sorption. Accordingly, biosorption capacity could usually reach to the acceptable level during the first 120 minutes.

Heavy metal ion accumulation by microorganisms generally occurs in two phases [38, 39]. The first phase occurs on the cell surface and consists of fast inactive biosorption, which is completely independent of cellular metabolism. The second phase consists of active sorption of heavy metal ions into the cytoplasm of algal cells. This phase is dependent on cell metabolism and is known as intracellular ion uptake [40]. Intracellular ion uptake has a large contribution in heavy metal ions biosorption and detoxification [41, 42].

Heavy metal ion biosorption capacity has been attributed to the presence of different types of binding groups on the algal cell surface i.e. hydroxyl, phosphoryl, carboxyl, sulphuryl, amine, imidazole, sulphate, phosphate, carbohydrate, etc. [35]. The availability of active sites for heavy metal ion uptake in algal cells can be probed by FTIR spectroscopy [36]. The sorption capacity of an algae cell surface to a specific ion also depends on factors such as the number of functional groups in the algae cells, the coordination number of the metal ion to be sorbed, the accessibility of binding groups for metal ions, the complex formation constants of metal ion with the functional group, and the chemical state of these sites. Usually the presence of binding groups make the net charge of the cell surface negative, which is related to the deprotonation of carboxyl and phosphate groups on the cell surface [37].

Figure 1 shows a schematic representation of the binding sites on the algal cell wall. Metal ions adsorbed by the algal cell wall acts as the first step in bioaccumulation. Different binding groups, such as OH^-, SH^-, COO^- , PO_4^{3-}, $NO3^-$, RNH_2^- , RS^- and RO^- promote the metal ion adsorption. These binding groups are present at the cell surface, in the cytoplasm, and especially vacuoles. If the mechanism of metal ion bioremediation is the uptake of ions by algal cells, cytosolic proteins mediate the transfer of metal ions into the cells [42]. Consequently, the vacuoles could be regarded as an organelle that accumulates metal ions. Table 2 presents a summary of the affinity between different metal ions and the cellular ligands, with R showing alkyl groups such as propyl, CH_3-CH_2CH_2-, and metal ions classified into classes A, B, and borderline. Class A tends to establish links with ligands in Group I through their oxygen atoms. Metal cations belong to class B tend to bridge with ligands in Groups II and III, and the borderline metal ions can be linked with different atoms of Groups I, II, and III[43]. Although metal-ligand complex formation is well classified into different Groups and Classes, but from the chemistry perspective it would have been more beneficial to include the complex formation constants between the metal ions and the different ligands at the cell surface. This will enable the researchers decide on preferential metal ion biosoprption and the effects of interfering ions. According to the pK_a of functional groups listed in Table 3, carboxyl groups, sulfonate, phosphate, and phosphodiester have the largest contribution in sorption capacity. Due to the relative abundance of each of these functional groups in different algal strains, each will exhibit a different capacity for metal ion biosorption.

Algae cell walls are the first barrier against the biosorption of heavy metal ions. Polysaccharides and proteins present in algae cell walls have the most metal binding sites [44]. Due to the different distribution and abundance of cell wall compositions in different algal strains, the capacity of metal ions biosorption by the variant algal strains will vary. Romera *et al*. [45] introduced brown algae as a very good candidate for biosorbents of heavy metal ions based on the comparison of different algal strains and biomass-metal ion affinity. Brown algae, with alginate in their cell wall composition has a high affinity for biosorption of lead ions [34]. Alginate polymers constitute the primary means of sorption of heavy metal ions in brown algae, and their biosorption capacity is directly related to the presence of binding sites on this polymer [45, 46].

The main factors influencing heavy metal ion biosorption

Biosorption of heavy metal ions by algae may be affected by several factors, including concentration of metal ions and algae biomass, pH, temperature, and the presence of competing ions. This section aims to review these factors and their possible effects on the metal ions biosorption.

The influence of initial metal ion concentration

Heavy metal ion removal by algal biomass depends largely on the initial concentration of metal ions in the solution phase. Biosorption initially increases as the initial concentration of metal ion increases. In following, no more considerable increase in metal sorption is observed by a tandem increase of metal ions concentration [47]. This phenomenon could be used to increase biosorption capacity. For example, Monteiro *et al*. [48] reported a 5-fold increase in initial concentrations of Zn (II) (from 10 to 50 ppm) boosted the metal ion sorption from 19 to 209.6 mg Zn (II)/g dry biomass of *Scenedesmus obliqus*. This leads to an increased biosorption capacity and a reduction in the removal yield of the metal ions. In other words, the higher the metal ion concentrations the lower the efficiency and removal yield would be [49]. At low metal ion concentrations removal takes place more efficiently than higher concentrations. For example, Mehta and Gaur [50] reported that *Chlorella vulgaris* biomass is able to remove 69% and 80% of Ni (II) and Cu (II) cations in concentrations of 2.5 ppm, respectively. While increase in the initial concentration of Ni (II) and Cu (II) to 10 ppm, the metal removal rate was reduced only to 37 and 42%, respectively. This clearly shows that the increase in metail ion concentration from 2.5 to 10 ppm reduced the bioremoval rates by about half. Due to the toxicity of some heavy metal ions for live algal strains metal ions uptake will be reduced by destruction of algal cells, and an optimization of metal ion concentrations is necessary for the efficient growth of algae. Shanab and Essa [51] investigated the effects of concentrations of mercury, cadmium, and lead ions on the growth of *Scenedesmus quadricauda*. They observed that low concentrations of lead and cadmium ions (5-20 ppm) enhanced algae growth through increased chlorophyll content, while mercury ions had a toxic effect on the algal cells in any concentration. Lamaia *et al.* [32] continually increased the exposure time and concentrations of lead and cadmium ions to explore the toxicity in a common filamentous live green algae, *Cladophora fracta*. The main toxicity symptom of Pb and Cd ions to *C. fracta* was a relative decrease in culture productivity, with total chlorophyll content loss, reduced number of chloroplasts, and disintegrated cell walls responsible for cell death and reduced cell growth.

To illustrate the interaction of live algal cells and toxic concentration of heavy metal ions, it is worth noticing that after biosorption of heavy metal ions to algal cells, they are transported to cell vacuole. During this step structural/binding proteins such as metallothioneins (MTs) bind to adsorbed ions and thus avoids inhibitory effects of accumulative concentration of metal ions in the host cells. This mechanism allows the normal biochemical activities to continue in the presence of toxic/lethal concentrations of heavy metal ions [39]. However, the presence of excessive toxicity of heavy metal ions could lead to protein structure denaturation, replacing essential elements or damage to the oxidative balance of the live algae. Intensity of the stress on algal cells depends on the content of oxidized proteins and lipids in the algae cells. The protection response of algae cells against heavy metal ions is extremely dependent on their resistance to the oxidative damages [40, 52].

The influence of pH on sorption selectivity

pH is one of the most important determining parameters of the capacity of metal ion uptake by algal biomass [19, 23, 53-56]. Dependence of metal ion uptake on pH is related to the metal ion complexation chemistry in solution, and behavior of many different functional groups present in the surface of algal cells as well as to complex formation constants [36, 53, 57]. Han *et al.* [53] investigated the Cr (III) uptake by *Chlorella miniata* biomass and found that biosorption capacity in pH 3, 4 and 4.5, was 14.17, 28.72 and 41.12 mg Cr (III)/g dried algae, respectively. Similar research by Gupta and Rastogi [58] on the uptake of Pb (II) by *Spirogyra* sp. biomass showed that biosorption of Pb (II) at the pH<3, is very low. When the pH increased in the range of 3-5, an increase in lead ions sorption was observed, with the maximum amount of sorbed ions being 140 mg/g at pH 5. Considering lead (II) hydroxide solubility product (K_{SP}) to be $1.4x10^{-20}$, and assuming 1.4 μM lead ion concentration, the hydroxide ions from K_{sp} calculations would be $K_{SP} = 1.4x10^{-20} = (1x10^{-6})(OH^{-})^{2}$, or hydroxyl ion concentration of 10^{-7}M and pH = 7. This implies that even at a micromolar concentration of lead ions, at pH 7 or higher, the lead(II) ions will precipitate as lead(II) hydroxide before biosorption by algae cells. In the case of biosorption using living algal cells, it can be inferred that during photosynthesis the inorganic carbon content of the culture medium was depleted, and consequently the pH increased. Concurrently, the biosorption of some metal ions such as Pb (II) might increase. Thus, injection of CO_2 can be used to control the acidity of the culture medium [59, 60].

The absence of H^+ ions increases the ability of establishing links between metal cations and ligands, leading to improved metal ion removal by algal biomass. Conversely, functional groups in acidic solutions are protonated and prevented from binding cations to functional groups [53, 58], resulting in a reduction of biosorption capacity. Therefore, finding the optimal pH for maximum metal ion removal by specific algae is paramount, as it strongly correlates with the biomass surface charge, degree of ionization, and absorbing sites.

The first step in the mechanism of biosorption and bioaccumulation of heavy metal ions is the diffusion of ions to the algae cell surface which is negatively charged from ionization of functional groups. The negatively charge surface will adsorb the counterions, ie heavy metal ions in this case, to have a double layer originated from the cell surface. The sorption of the metal ions causes the depletion of ions in media (growth media for live algae) and this depletion lowers the ionic strength of the media that causes the expansion of double layer thickness. Therefore, the biosorption of heavy metal ions is more efficient in dilute media [61] .

The tendency for selective metal ion uptake at an optimized pH is useful in targeted biosorption in multi metal ion solutions. Aksu *et al.* [62] in a study on *C. vulgaris* biomass, determined the optimal Cu (II) and Cr (VI) biosorption at pH 4 and 2, respectively. The optimal pH for these metal ions is related to their chemical interaction with the algal cells. In an investigation by Cimino et al. [63] the influence of pH on the distribution of Cr(VI) in solution showed that for pH values under 3.0 the $HCrO_4^-$ and $Cr_2O_7^{2-}$ ions species were predominant and efficiently absorbed on the protonated cell binding sites. At pH values over 5.0 the total chromium bioremoval was negligible since increasing pH shifted $HCrO_4^-$ to CrO_4^{2-}. Therefore, increasing pH negatively affected the final capacity of chromium bioremoval.

Due to the various chemical forms of metal ions found in IW, pH adjustment could play an important role in biosorption capacity [41]. Usually NaOH and HCl [64], H_2SO_4 [24], HNO_3 [26], or the buffer [65] are used for adjusting pH of IW solutions. Based on the properties of metal ions, suitable acids, bases, or buffers should be chosen to adjust the pH. For example, in biosorption of lead due to the formation of the $PbSO_4$ precipitates, H_2SO_4 should not be used. Buffering interferences with metal ions in the solution is also important and should be considered. For example, Ni (II) and Cd (II) concentrations when using a phosphate buffer to adjust the pH may result in the formation of phosphate precipitate.

The influence of biomass concentration

The amount of metal ions removed from a solution phase is dependent on the algae biomass concentration, and increasing biomass concentrations reduces metal ion uptake per gram of biomass [50, 66-68]. In practical terms, increased biomass concentrations positively increases final bioremoval, although it negatively affects biosortion capacity of heavy metal ions [69]. Electrostatic interactions between cells have a significant effect on metal ion uptake by algal biomass, with high biomass concentrations having a ‘shell effect’ on the outer structure of biomass and avoiding functional group binding to metal ions [34, 70]. The shell effect enables the control of complex formation by adjusting pH to the isoelectric point. Mehta and Gaur [69] found out that a 100-fold increase in biomass concentration of *C. vulgaris* is accompanied by a significant increase in removal of Ni (II) and Cu (II). In a similar study on *Scenedesmus abundans* by Terry and Stone [71], competition between Cu (II) and Cd (II) for binding sites was observed, and higher concentrations of biomass prevented such competition. There is also a variable effect of biomass concentrations on the metal ion biosorption capacity This was investigated by Romera *et al.* [34] using different algal strains and metal ions, and was reported that maximum biosorption efficiency could be obtained at the lowest biomass concentration.

The influence of temperature

Biosorption efficiency of each metal ion is different for each algae species with different response to the temperature [33, 72]. Although metal ligand complex formation constants are primarily a function of temperature, some previously published studies claimed that increased algal culture temperatures could potentially increase metal ion biosorption capacity [58, 73-77], with no consideration of formation constants changes by temperature. The possible reasons for increasing temperatures to result in increasing metal ion biosorption include: (1) An increased number of active sites involved in metal ion uptake; (2) an increased tendency of active sites to absorb metal ions [37]; (3) a reduction in mass transfer resistance in the diffusion layer by a reduction of the thickness of the diffusion boundary layer around the adsorbent groups [78], and (4) change of complex formation constant with temperature [79, 80]. However, other studies suggest that metal ion uptake by some algae is exothermic and uptake capacity increases with decreasing temperature [72, 81]. There is also observation that indicate temperature has no significant influence on the metal ion uptake by algal cells [56, 82, 83], and similarly several studies have determined temperature-linked changes in metal ion uptake by living algal cells [23, 84]. These seemingly incompatible results may be resolved by noting that optimum temperatures is usually a narrow range for active biological reactions in living cells, and temperature variations cause different biosorption behaviors in various algal strains with different metal ions. Most importantly is the change of complex formation constant with temperature which is apparently been neglected by most researchers. The biosorption capacity of cadmium ions increase with decreasing temperature for specific algae because of the exothermic nature of cadmium ion bioremoval [85-88]. Similarly, research by Aksu [85, 89] investigated the effect of temperature on the *C.vulgaris* biomass for biosorption of Cd

(II) and Ni (II). They observed the maximum biosorption for Cd (II) and Ni (II) occurred at 20 and 45 ^{0}C, respectively.

Temperature also influences biosorption of metals by non-living algal biomass as the adsorption equilibrium is determined by the exothermic or endothermic nature of the process. A number of studies on the effect of temperature on adsorption isotherms, metal uptake, and also biosorption thermodynamics parameters have been performed [90-92]. Due to intracellular absorption and enzymes in the transfer of ions into the living algae cell, increasing temperature might have a greater impact on the absorption capacity as compared with non-living algae. Altogether, these factors will lead to reduced absorption capacity of the living algae more than non-living algae.

The influence of contact time

Heavy metal ion biosorption is highly dependent on contact time. Based on the previously published reports discussing the kinetics of heavy metal ion biosorption on algae cell surface, the mechanism of biosorption is algae strain specific [26]. Biosorption takes place in two stages, where; (1) for algae biomass, ions adsorb to cell membrane passively and biosorption of metal ions occurs rapidly within the first minutes, and; (2) for live algae, active sorption occurs as heavy metal ions slowly uptake into the algal cell. Vogel *et al.* [93] investigated the uptake of uranium by non-living *C. vulgaris* and observed that more than 90% of the dissolved uranium adsorb during the first 5 minutes. In another study, Tüzün *et al.* [94] showed that the biomass of *Chlamydomunas reinhardtii* microalgae rapidly adsorbed free ions of Hg (II), Cd (II) and Pb (II), with the biosorption equilibrium achieved in 60 minutes. Mata *et al.* [19] reported the amount of Au (III) adsorbed at a pH of 7 on the biomass of *Fucus* *vesiculosus* macroalgae, after 1 and 8 hours were 28.95 mg/g and 74.05 mg/g of dry algae, respectively. This demonstrates that biosorption of heavy metal ions is a passive process that occurs relatively rapidly, even when algal cells are non-living. In living algae contact time has a greater effect on the biosorption capacity. For example, Lamai *et al*. [32] measured the uptake of cadmium and lead ions by *Cladophora fracta*, separately harvested after 2, 4, 6, and 8 days, and found while the algal growth rate decreased over time, a greater biosorption capacity was obtained in older cultures. These results suggest that while passive heavy metal biosorption commences swiftly in the first moments of contact, a greater level of IW heavy metal bioremoval can be achieved with longer contact times using living algae. The issue with 'older' cultures from chemistry point of view is the gradual depletion of nutrients and reduction of the ionic strength of the growth media with time. This will affect the biosorption capacity of heavy metal ions onto the algae cell surface.

The influence of multi metal ion systems

The type, combinations, and concentrations of heavy metal ions vary greatly among wastewaters. For example, electrolytic effluent contain a mixture of metal ions such as Hg, Mn, Ni, Pb, and Cu ions [95]. Bioremoval of multiple metal ions in solution is a common situation rather than relatively simple single metal ion solutions. Despite investigation of single metal ion solutions being routinely surveyed in the research literature, the real situation for IW treatment is more complicated due to the presence of multiple metal ions that needs further investigations. The presence of multiple heavy metal ions in the algal growth media imparts major physiological and biochemical consequences [96, 97]. In multi-metal ion systems metal ions compete for binding to algal ligands, and the presence of some cations significantly influence the uptake of other metal ions by algal cells [98, 99]. Aksu and Dönmez [100] studied the effect of cadmium ions on the removal of nickel ions and vice versa, and found simultaneous biosorption of nickel and cadmium ions significantly repressed the total biosorption capacity in comparison to the single ion solutions. Table 4 presents heavy metal ion uptake in binary solutions. In general all binary solutions show a decrease of metal ion biosorption. There are several studies showing that the role of light metal ions on the toxicity of heavier metal ions biosorption is very small [73, 74]. However, high concentrations of monovalent cations of Na^+ and K^+, could increase the ionic strength of wastewater, leading to a reduction in biosorption capacity of biomass [101, 102]. In water contaminated with multiple heavy metal ions, competition among the metal ions to bind to the active sites of cell surface is directly influenced by the concentration of each ion and their properties, primarily electronegativity and ionic radius [98, 103]. For example, aluminium ions can interfere with biosorption of copper ions preventing access to the binding sites at the cell surface[104], while the copper ions in the solution had no significant effect on Al^{+3} ion biosorption [105]. Similar research by Kaewsarn *et al.* [106] showed the effect of interfering anions including ethylenediaminetetraacetic acid (EDTA), SO_4^{2-}, PO_4^{3-} and CO_3^{2-} ions on the biosorption of Cu (II). They reported that the biosorption of copper relatively decreased in the presence of EDTA, SO_4^{2-}, PO_4^{3-}, and CO_3^{2-}, respectively.

The influence of other factors

Growth rates, level of dissolved nitrates, and light intensity can contribute to the removal of heavy metal ions by algae. Nitrate is a primary nutrient for algae growth, and changing in initial nitrate concentration can influence algae growth and biomass production [107]. Nitrate depression results in algae producing high amounts of lipids or low amounts of biomass, and therefore, low metal ion biosorption [108].

The effect of light intensity on metal ion uptake is largely unknown. The metal ion biosorption is proposed to be metabolism-independent for algal biomass and a two phase of metabolism-independent and metabolism-dependent for living algae [109], the former is slow and the latter is fast. The initial metabolism-independent step, commonly valid for biosorption of metal ions on biomass is indepdent of light and temperature. However, research by Subramanian *et al*. [110] found biosorption of Zn (II) in the dark regions is slightly higher than that in light regions. Culture medium dissolved gas concentration is also another factor that affects the growth rate of biomass and its contents. For example, Ota *et al*. [111] investigated the effect of dissolved oxygen on lipid synthesis in *Chlorococcum littorale*, and found that the lipid production can be limited by dissolved oxygen in photoautotrophic culture. The numerous variations in growth conditions affecting the availability of binding groups also influence the characteristics of the algal biomass, resulting in changes to relative heavy metal ion biosorption capacity.

Metal ion sorption by pretreated algae biomass

Increased heavy metal ion uptake by algal biomass can be enhanced by several physical/chemical treatments that change the algal cell surface properties to provide additional binding sites. Algal biomass physical treatments such as heating/boiling, freezing, crushing, and drying usually lead to an enhanced level of metal ion biosorption. These treatments influence the important role of the cell wall in biosorption of metal ions, as non-living cell membrane destruction provides more surface area to increase the biosorption capacity [112] and release the cell contents for possible increase in binding cell components to metal ions. The most common algal pretreatments are $CaCl_2$, formaldehyde, glutaraldehyde, NaOH, and HCl. Pretreatment by $CaCl_2$ causes calcium binding to alginate that plays an important role in ion exchange [113, 114]. Formaldehyde and glutaraldehyde help strengthening the crosslinking between functional groups, especially hydroxyl groups and amino groups [115, 116]. NaOH increases the electrostatic interactions of metal ion cations, and provides optimum conditions for ion-exchange, while HCl replaces light metal ions with a proton and also dissolves polysaccharides of cell wall [45], or denatures proteins [117], and increase bonding sites to improve biosorption.

Arica *et al*. [118] investigated the effect of heat and acid treatment on the uptake of Cr (VI) by the biomass of *Chlamydomunas reinhardtii*. The Cr (VI) biosorption capacity for the treated biomass was 25.6 and 21.2 mg/g, respectively; significantly higher than the untreated dried biomass (18.2 mg/g). Table 1 summarizes the effect of different physicochemical treatments on the biosorption capacity of different algal strains, enabling a comparison of implemented treatments on biosorption capacity. In order to increase biosorption of Cu(II) and Ni(II), Mehta and Gaur [50] treated *chlorella vulgaris* biomass by HCl, HNO_3, and NaOH, and observed Cu(II) and Ni(II) bioremoval were higher than the control sample. Several studies indicated that $CaCl_2$ is a cost-effective treatment to increase the metal ion sorption by algal biomass. For example, in order to increase biosorption of Pb (II), Rincon *et al*. [113] treated *Fucus vesiculosus* macroalgal biomass by $CaCl_2$, HCl, and formaldehyde, and observed the Pb (II) biosorption capacity of $CaCl_2$ treated biomass was higher than the control sample.

The effectiveness of implemented treatments in metal ion biosorption is directly dependent on the type of active sites present on the cell surface. In an study, Zhao *et al.* [119] investigated the effects of different treatments (HNO_3, HCl, NaOH, acetone and water, 60°C) on the biosorption of variant metal ions such as Pb, Cu, Zn, Cd, Cr, Mn, Ni, Co, Hg, Au, and Ag using six species of marine algae. They found that to varying extents all treatments successfully increased the ability of biomass to bind metal ions and improve biosorption capacity. Other chemical treatments (such as phosphorylation) can enhance the biosorption of radioactive ions from aquatic environments [120]. For example, Pohl and Schimmack [17] performed phosphorylation of *Laminaria japonica* and two species of cyanobacteria biomass to increase the biosorption capacity of radioactive nuclei (^{134}Cs, ^{85}Sr, ^{226}Ra, ^{241}Am). However, chemical pretreatments do not always produce predictable results, and may even cause opposite effects. For example, Zhang *et al.* [121] observed a decrease in uranium uptake by *Scenedesmus obliqus* after treatment with HCl, NaOH, NaCl, and diluted ethanol. Modification of the growth media (i.e. introducing supplements such as glucose, ammonium sulfate, phosphate, etc.) can potentially improve the metal ion uptake by the biomass [122]. The goal of all these growth media treatments are improving the conditions to favor contact between functional groups and metal ions through additional binding sites or improved linkage between the chains of biopolymers [113, 123].

Macro vs micro algae

Seaweed, green macroalgae and their alginate derivatives exhibit high affinity for many metal ions [124]. The passive removal of toxic heavy metals by biological materials is an emerging potential known as biosorbents.

To investigate the biochemical properties of the brown algae a comprehensive review was previously published [46]; A detailed description of cellular structure, storage polysaccharides, cell wall and extracellular polysaccharides were discussed in terms of their potential role in metal biosorption in brown macroalgal strains. Alginate plays a critical role in metal biosorption by brown algae. Alginate participate in ion-exchange and complexation result in binding of heavy metals by this polymer. The adsorption capacity of the brown algae is directly related to the alginate content, availability and its specific macromolecular conformation. Alginate comprises a significant component up to 40-45% of the dry weight of *Sargassum* biomass [125]. The affinity of alginates for divalent cations such as Pb^{2+},Cu^{2+},Cd^{2+} and Zn^{2+} donate 227, 51, 79 and 78 mg g^{-1} metal uptake [46]. *Sargassum* packed columns was investigated to be used in flow-through column systems. Implementation of such packed bed columns inactively adsorb and detoxify heavy metals bearing industrial wastewater [126]. Algal biomass immobilization techniques will be further discussed in the next section.

To reply to the question about finding suitable freshwater filamentous algae that possess a high metal ion removal capability, Lee and Chang [127] evaluate the Pb(II) and Cu(II) bioremoval capacity in two green macroalgae species, *Spirogyra* and *Cladophora*, the results indicated that although the functional groups of these two genera of algae were similar, but the adsorption efficiency of *Spirogyra* spp. for Pb(II) and Cu(II) were superior to those of *Cladophora* spp.. (87.2 and 38.2 mg g^{-1} for *Spirogyra* and 45.4 and 13.7 mg g^{-1} for *Cladophora*, respectively). Further example of biosorption capacity of different heavy metal ions using diverse macroalgal strains under varying physicochemical conditions are summarized in Table 1.

Comparison of living and non-living algal species will be comprehensively reviewed in the next section from other standpoints. Microalgae usually step further in contamination bioremoval. In more details, denitrification, dephosphorylation and COD reduction beside heavy metal biosorption are well established in microalgae wastewater treatment [128].

Living vs non-living algae

Although clear differences exist between accumulation of metal ions onto living algae cells and biosorption of metal ions onto non-living algae biomass, the process with the largest contribution in both living and non-living algae is the ion exchange process [39]. Since the influence of operating parameters such as pH, temperature and contact time have been previously discussed, herein the efficiency and also the applicability of living and non-living algae in the removal processes of metal ions will be introduced. While metabolic processes in living algae generally contribute to heavy metal bioremediation [130], using non-living algae has recently gained popularity for biosorption of heavy metal ions from solutions.. Non-living biomass biosorption advantages include a heavy metal biosorption several times greater in non-living algae as compared to living algae [37, 132, 133]. Moreover,the possibility to recycle non-living algal biomass is a unique characteristic dealing by dead biomass [134]. For example, metal ions bound to the algal cell wall may be removed by washing the biomass with deionised water and desorption agents (HCl, NaOH, $CaCl_2$) [135], whereas living algae have a low mechanical and chemical resistance to physical and chemical treatments for recycling. It is worth quoting that the non-living algae can be easily treated using physical and chemical protocols to enhance adsorption capacity

[118]. The use of non-living algal biomass also removes the risks of exposures to highly toxic environments, and do not require intensive management or addition of further growth nutrients [52, 136]. Nonetheless, several environmental factors influence non-living algae heavy metal ion biosorption. For example, changes in pH impact living algae to a greater extent than non-living algae as most algae grow in neutral or slightly alkaline mediums [140], and acidic media can affect the algae growth rate, and basic media might cause precipitation of the metal ions [141, 142]. Heavy metal removal in solutions with an extreme pH favors non-living algae over living algae, as using live algae adds complexity to culture medium chemistry management that might lead to unwanted metal ion precipitation and bioremediation interference.

Regarding to the summarized data in Table 1, meaningful differences could not be tracked among removal efficiency of living and non-living algae; in more details, different living and non-living samples of *C. vulgaris* presented the same ion removal efficiency for U^{4+} [93] and Ni^{2+} [151] biosorption. In summary, the living cells having metabolic activities possibly present higher uptake of metal ions compared to dead biomass. They could also adsorb more diverse range of ions (Doshi, 2007), However, non-living cells present faster uptake kinetics. The dead biomass materials could be successfully reused in successive adsorption–desorption cycles [64]. Finally, low cost and ease of use in non-living cells have developed this technology as a serious candidate for bioremediation of for IW in large scale. Consequently, to achieve the highest removal efficiency, interaction between algal strains, dead or live cells and pollutants should be optimized.

Immobilized algae

Techniques such as flocculation, adsorption on surfaces, covalent binding to carriers, crosslinking of algal cells, and entrapment of algae in polymeric matrix are used for cell/biomass immobilization [143, 144]. For immobilization of biomass, natural biopolymers (such as agar and alginate) or synthetic compounds (such as silica gel and polyacrylamide) can be used as supporting materials. Natural polymers are often preferred to synthetic polymers due to non-toxicity to biomass, and for this reason calcium alginate has been widely used for immobilization of algal cells and many other biomass sources [69, 145, 146]. Among synthetic polymers, polyacrylamide has been most extensively used [18, 147], as it is more resistant than calcium alginate, although its application for immobilization processes is limited by its high cost and toxicity to living cells. Table 5 presents research where immobilized algae has resulted in an increase in biosorption capacity relative to free algal cells, and prevented loss of biomass during the biosorption cycle [148]. Biomass immobilization enhances photosynthetic capacity [149] and reduces toxicity of some substances [150]. It also facilitates repetitive use of algal cells during successive sorption/desorption cycles of metal ions bioremoval from aqueous solutions. Enhanced surface sorption in the immobilized powdered algal cells result in a 2-fold increase in nickel removal in comparison to the free non-living *C. vulgaris* cells [151]. The same observation was reported by Murugesan *et al.* [152] on the potential of immobilized algal cells of *Spirulina platensis* in cadmium ion biosorption. While algae immobilization has a high potential for removing toxic metal ions from IW, it requires an ideal cost effective method.

Metal ion biosorption enhancement using molecular tools

Exploiting biological mechanisms at the molecular level to produce engineered organisms with higher biosorption capacity and selectivity for specific metal ions can be used to develop new biosorbents. The high cost of conventional technologies to reduce toxic metal ions concentrations in IW to acceptable regulatory standards has prompted exploitation of genetic and protein engineering approaches to produce cost effective ‘green’ biosorbents. One emerging area of research is the design and development of novel algae strains with increased affinity, capacity, and selectivity for biosorption of heavy metal ions. Many genes are involved in metal-uptake, detoxification, or tolerance [153]. Cysteine-rich peptides such as glutathione (GSH), some lipopolysaccharides, phytochelatins (PCs), and metallothioneins (MTs) bind metal ions (Cd, Cu, Hg etc.) and enhance metal ions bioaccumulation [154]. For example, tripeptide GSH as a typical low molecular weight thiol has a significant role in detoxification of metal ions. Moreover, it acts as a storage form of endogenous sulfur and nitrogen [155]. Cell surface treatment technologies have been recently used to improve the performance of biomass in metal ion removal from aqueous solutions, and cell surface MTs or PCs could increase metal ion accumulation capacity. For example Kuroda *et al.* [156] expressed a histidine hexapeptide on the cell surface of engineered yeast *S. cerevisiae*. Furthermore, the introduction of surface exposed MerR (a metalloregulatory protein with high affinity and selectivity toward mercury engineered to strains of *E. coli*) can increase the capacity of Hg (II) sorption six-fold higher than the wild-type [157].

Genetic and protein engineering can also create artificial proteins with fundamentally new molecular activities and/or imitative functions [158]. A novel protein with both high metal-binding and pre-programmed properties for heavy metal ion removal in theory can be located in any specific cellular compartment [159]. Bae *et* *al*. [157] researching recombinant *E. coli* strains harboring synthetic fusion genes encoded outer membrane peptides with the general structure of (Glu-Cys) nGly, resulting in a doubling of accumulated Cd (II). Outer membrane expression involves nonviable cells in metal ion accumulation with efficient metal ion bounding [160]. A recombinant *E. coli* strain expressing MT fused to the outer membrane of a maltose protein (LamB) showed a 15-20-fold increase in Cd (II) binding compared to the control sample [161]. The efficiency of MT heteroproteins could be enhanced according to the specific role of metal ion membrane transporters. For example, fusion of glutathione S-transfers to MT lead to a 3-fold increase in Ni (II) accumulation in comparison to cells expressing MT with no transporter in transgenic *E. coli* strains [162]. Cytoplasmic expression of metal-binding polypeptides such as PC were evaluated as an effective system for cellular detoxification of some metal ions [163]. A combined approach was investigated in a recombinant *E. coli* by a fusion plasmid harboring mercury transport system and strong intracellular accumulator system. Immobilized cells were able to remove mercurial contamination from wastewater repeatedly [164].

Transgenic plants which detoxify/accumulate cadmium, lead, mercury, arsenic, and selenium ions have been transformed by PCs, MTs, metal chelators, and transporter. For example, the MT-transformed plants can grow normally in the presence of 0.1 mM cadmium chloride [165]. The responsible genes for detoxification functions have their highest diversity in bacteria and fungi. Rhizosphere strongly participates in contaminant detoxification. Root exudates increase soil microbial growth and in turn translates into greater metal ion detoxification. Genetic manipulations of mycorrhizal communities associated with woody plants could improve the capacity of woody plants in remediation purposes [165].

To date little attention has been paid to investigate the recombinant microalgal strains for metal ion biosorption, and it remains highly prospective for engineered algae achieving higher sorption capacities and specificity for targeted metal ions. However, without detailed analyses and targeted strategies, wide-scale implementation of molecular tools has the potential for ecological harm that genetically modified algal strains could possibly threaten the sustainability of a host ecosystem. To mitigate the impacts of such risks one strategy may include further processing downstream from bioremediation activities, or the use of hybrid technologies to obtain a byproduct/biofuel from produced algal feedstock.

Coupling wastewater treatment and biofuel production

Costly chemical-based treatments to remove very high concentrations of nutrients and toxic metal ions from wastewater is the major problem with most wastewater applications [166]. The potential of algae to efficiently remove heavy metal ion, candidates them as an extremely promising tools for sustainable and low cost wastewater treatment [3, 167]. Capital, operation, and maintenance costs for microalgal biofuel production can be significantly reduced by using wastewaters for biomass production [168]. Hybrid wastewater treatment and algae cultivation systems could decrease unit costs of energy by 20-25%, and largely eliminate the cost of nutrient and freshwater supplementation [169]. Coupling of the production of biofuel-directed microalgae with bioremediation of wastewaters provides a pathway to combat eutrophication and industrial pollution in conjunction with the renewable energy production [27].

Bioremoval of heavy metal ions using microalgae has been considered as an environmentally and economically sustainable approach to remove toxic metals from wastewaters [170]. On the cost side, the need to reduce requirements for chemical remediation of wastewaters, minimizes freshwater consumption, enhances the suitability of algal introduction in the wastewater treatment process [27, 171-173]. Besides, a wide range of valuable by-products (such as bioethanol and biodiesel), valuable nutrients and bioactive compounds can be extracted from the produced biomass [168]. Integrated algal-based treatment of wastewater and biofuel production can not only reduce the inputs and costs of algal biomass production, but also efficiently remove potentially hazardous contamination such as residual nutrients, toxic metal pollutants, and even transgenic algae from wastewaters [3, 174]. The coupled system is a useful approach where nutrient and heavy metal ion removal is required prior to wastewater discharge. Moreover, production of biofuels could also decrease the final cost of CO_2 sequestration from industrial sources or power plants [60]. However, to achieve the proposed potentials of a coupled algal systems, maximizing autotrophic production is of primary importance. It could be applicable through using high rate algal ponds (HRAPs), which play an efficient and cost-effective role for the conventional wastewater treatment widely used in industrial scale globally [175]. HRAPs in comparison to the traditional wastewater methods has lowered the capital and operating costs, does not need advanced technology to operate, while providing all the benefits of coupled systems to produce biofuel [169, 175].

Conclusion

Low-cost cultivation, high metal ion uptake, and metal selectivity, and suitable mechanical properties for large scale production makes algae a suitable candidate for wastewater bioremediation. A complete characterization of biochemistry of microalgal substrates and its environmental benefits will be necessary to credibly emphasize the advantages of algal biosorption over conventional ion-exchange resins and routine chemical treatments. Further research at both fundamental and field-scales will assist optimization of final biosorption capacity to improve the economic sustainability and practicalities of large-scale implementation of algal heavy metal bioremediation. To achieve implementation of algal biosorption technology in industrial and environmental remediation requires a better understanding of influencing parameters, including initial concentrations, physico-chemical conditions, and also contact times, in addition to other parameters discussed in this review article. Successful biosorption processes require inexpensive biomaterials display high metal uptake and selectivity based on biochemical constitution, as well as suitable mechanical properties for applied remediation procedures. Based on the high biomass productivity of wastewater-grown algae, it is an attractive dual-use algae cultivation for wastewater treatment coupled with other downstream or hybrid production systems. However, life-cycle assessment, techno-economic analysis and energy intensity of any utility-connected algae systems should be precisely determined prior to implementation.

Acknowledgements

HA thanks ATF committee for the financial support of this work.

References:

1. D.W. O’Connell, C. Birkinshaw,T.F. O’Dwyer, Heavy metal adsorbents prepared from the modification of cellulose: A review. Bioresour. Technol., 99 (2008) (15) 6709-6724.
2. K. Tsekova, D. Todorova,S. Ganeva, Removal of heavy metals from industrial wastewater by free and immobilized cells of *Aspergillus niger*. Int. Biodeterior. Biodegrad., 64 (2010) (6) 447-451.
3. L.E. de-Bashan,Y. Bashan, Immobilized microalgae for removing pollutants: Review of practical aspects. Bioresour. Technol., 101 (2010) (6) 1611-1627.
4. N. Sridhara Chary, C.T. Kamala,D. Samuel Suman Raj, Assessing risk of heavy metals from consuming food grown on sewage irrigated soils and food chain transfer. Ecotoxicol. Environ. Saf., 69 (2008) (3) 513-524.
5. ATDR, Toxicological profile for Lead, Agency for Toxic Substances and Disease Registry, Public Health Services, US Department of Health and Human Services. (2007).
6. ATDR, Toxicological profile for Nickel, Agency for Toxic Substances and Disease Registry, Public Health Services, US Department of Health and Human Services. (2005).
7. ATDR, Toxicological profile for cadmium, Agency for Toxic Substances and Disease Registry, Public Health Services, US Department of Health and Human Services. (2012).
8. W. Oswald, My sixty years in applied algology. J. Appl. Phycol., 15 (2003) (2-3) 99-106.

9. J.P. Hoffmann, Wastewater treatment with suspended and nonsuspended algae. J. Phycol., 34 (1998) (5) 757-763.

10. J. Wang,C. Chen, Biosorbents for heavy metals removal and their future. Biotechnol. Adv., 27 (2009) (2) 195-226.

11. L. Charerntanyarak, Heavy metals removal by chemical coagulation and precipitation. Water Sci. Technol., 39 (1999) (10) 135-138.

12. A. Dąbrowski, Z. Hubicki, P. Podkościelny,E. Robens, Selective removal of the heavy metal ions from waters and industrial wastewaters by ion-exchange method. Chemosphere, 56 (2004) (2) 91-106.

13. C. Yuan,C.-H. Weng, Electrokinetic enhancement removal of heavy metals from industrial wastewater sludge. Chemosphere, 65 (2006) (1) 88-96.

14. H.A. Qdais,H. Moussa, Removal of heavy metals from wastewater by membrane processes: a comparative study. Desalination, 164 (2004) (2) 105-110.

15. S.-M. Lee, C. Laldawngliana,D. Tiwari, Iron oxide nano-particles-immobilized-sand material in the treatment of Cu(II), Cd(II) and Pb(II) contaminated waste waters. Chem. Eng. J., 195–196 (2012) (0) 103-111.

16. E.K. Goharshadi,M.B. Moghaddam, Adsorption of hexavalent chromium ions from aqueous solution by graphene nanosheets: kinetic and thermodynamic studies. Int. J. Environ. Sci. Technol. , 12 (2015) (7) 2153-2160.

17. P. Pohl,W. Schimmack, Adsorption of Radionuclides (134Cs, 85Sr, 226Ra, 241Am) by Extracted Biomasses of Cyanobacteria (*Nostoc Carneum, N. Insulare, Oscillatoria Geminata and Spirulina Laxis-Sima*) and Phaeophyceae (*Laminaria Digitata and L. Japonica*; Waste Products from Alginate Production) at Different pH. J. Appl. Phycol., 18 (2006) (2) 135-143.

18. D.W. Darnall, B. Greene, M.T. Henzl, J.M. Hosea, R.A. McPherson, J. Sneddon,M.D. Alexander, Selective recovery of gold and other metal ions from an algal biomass. Environ. Sci. Technol., 20 (1986) (2) 206-208.

19. Y.N. Mata, E. Torres, M.L. Blazquez, A. Ballester, F. Gonzalez,J.A. Munoz, Gold(III) biosorption and bioreduction with the brown alga *Fucus vesiculosus.* J. Hazard. Mater., 166 (2009) (2-3) 612-8.

20. Y.N. Mata, M.L. Blazquez, A. Ballester, F. Gonzalez,J.A. Munoz, Characterization of the biosorption of cadmium, lead and copper with the brown alga *Fucus vesiculosus.* J. Hazard. Mater., 158 (2008) (2-3) 316-23.

21. A. Demirbas, Heavy metal adsorption onto agro-based waste materials: a review. J. Hazard. Mater., 157 (2008) (2-3) 220-229.

22. I. Michalak,K. Chojnacka, Interactions of metal cations with anionic groups on the cell wall of the macroalga *Vaucheria sp.* Eng. Life Sci., 10 (2010) (3) 209-217.

23. S. Mehta, A. Singh,J. Gaur, Kinetics of adsorption and uptake of Cu2+ by *Chlorella vulgaris*: influence of pH, temperature, culture age, and cations. J. Environ. Sci. Health. Part A Toxic/Hazard. Subst. Environ. Eng. , 37 (2002) (3) 399-414.

24. D. Uzunoğlu, N. Gürel, N. Özkaya,A. Özer, The single batch biosorption of copper (II) ions on *Sargassum acinarum.* Desalin. Water Treat., 52 (2014) (7-9) 1514-1523.

25. D. Das, S. Chakraborty, C. Bhattacharjee,R. Chowdhury, Biosorption of lead ions (Pb2+) from simulated wastewater using residual biomass of microalgae. Desalin. Water Treat., (2015) 1-11.

26. A. Yaqub, M. Mughal, A. Adnan, W. Khan,K. Anjum, Biosorption of hexavalent chromium by *Spirogyra spp*.: equilibrium, kinetics and thermodynamics. J. Anim. Plant Sci., 22 (2012) (2) 408-415.
27. S. Lyon, H. Ahmadzadeh,M. Murry, Algae-Based Wastewater Treatment for Biofuel Production: Processes, Species, and Extraction Methods, in Biomass and Biofuels from Microalgae, Moheimani N.R., McHenry M.P., de Boer K.,Bahri P.A., Editors. 2015, Springer International Publishing. p. 95-115.
28. “Water for People, Water for Life” United Nations World Water Development Report, UNESCO, 2003 www.unesdoc.unesco.org.
29. Eurostat, the statistical office of the European Union, 2011 www.europa.eu.
30. S. Abinandan,S. Shanthakumar, Challenges and opportunities in application of microalgae (*Chlorophyta*) for wastewater treatment: A review. Renewable and Sustainable Energy Rev., 52 (2015) 123-132.
31. O. Komolafe, S.B. Velasquez Orta, I. Monje-Ramirez, I.Y. Noguez, A.P. Harvey,M.T. Orta Ledesma, Biodiesel production from indigenous microalgae grown in wastewater. Bioresour. Technol., 154 (2014) 297-304.
32. C. Lamaia, M. Kruatrachuea, P. Pokethitiyooka, E.S. Upathamb,V. Soonthornsarathoola, Toxicity and accumulation of lead and cadmium in the filamentous green alga *Cladophora fracta* (OF Muller ex Vahl) Kutzing: A laboratory study. Sci. Asia, 31 (2005) (2) 121-127.
33. C. Monteiro, P.L. Castro,F.X. Malcata, Cadmium Removal by Two Strains of *Desmodesmus pleiomorphus* Cells. Water Air Soil Pollut., 208 (2010) (1-4) 17-27.

34. E. Romera, F. González, A. Ballester, M.L. Blázquez,J.A. Muñoz, Comparative study of biosorption of heavy metals using different types of algae. Bioresour. Technol., 98 (2007) (17) 3344-3353.

35. D. Kaplan, Absorption and adsorption of heavy metals by microalgae, in Handbook of Microalgal Culture: Applied Phycology and Biotechnology, Richmond A. ,Hu Q., Editors. 2013, Blackwell Publishing. p. 439-447.

36. V. Gupta,A. Rastogi, Equilibrium and kinetic modelling of cadmium (II) biosorption by nonliving algal biomass *Oedogonium sp*. from aqueous phase. J. Hazard. Mater., 153 (2008) (1) 759-766.

37. S.K. Mehta,J.P. Gaur, Use of algae for removing heavy metal ions from wastewater: progress and prospects. Crit. Rev. Biotechnol., 25 (2005) (3) 113-152.

38. C. Monteiro, P.L. Castro,F.X. Malcata, Microalga-mediated bioremediation of heavy metal–contaminated surface waters, in Biomanagement of metal-contaminated soils, Khan M.S., Zaidi A., Goel R.,Musarrat J., Editors. 2011, Springer Netherlands. p. 365-385.

39. C.M. Monteiro, P.M.L. Castro,F.X. Malcata, Metal uptake by microalgae: Underlying mechanisms and practical applications. Biotechnol. Prog., 28 (2012) (2) 299-311.

40. A.F. Talebi, Tabatabaei, M., Mohtashami, S.K., Tohidfar, M.,Moradi, F., Comparative Salt Stress Study on Intracellular Ion Concentration in Marine and Salt-adapted Freshwater Strains of Microalgae. Not. Sci. Biol., 5 (2013) (3) 309-315.

41. K.L. Wilde, J.L. Stauber, S.J. Markich, N.M. Franklin,P.L. Brown, The effect of pH on the uptake and toxicity of copper and zinc in a tropical freshwater alga (*Chlorella sp*.). Arch. Environ. Contam. Toxicol., 51 (2006) (2) 174-185.

42. H.V. Perales-Vela, J.M. Peña-Castro,R.O. Cañizares-Villanueva, Heavy metal detoxification in eukaryotic microalgae. Chemosphere, 64 (2006) (1) 1-10.

43. B. Volesky, Biosorption and me. Water Res., 41 (2007) (18) 4017-29.

44. S. Schiewer,B. Volesky, Biosorption by Marine Algae, in Bioremediation, Valdes J., Editor. 2000, Springer Netherlands. p. 139-169.

45. E. Romera, F. Gonzalez, A. Ballester, M.L. Blazquez,J.A. Munoz, Biosorption with algae: a statistical review. Crit. Rev. Biotechnol., 26 (2006) (4) 223-35.

46. T.A. Davis, B. Volesky,A. Mucci, A review of the biochemistry of heavy metal biosorption by brown algae. Water Res., 37 (2003) (18) 4311-4330.

47. R. Singh, R. Chadetrik, R. Kumar, K. Bishnoi, D. Bhatia, A. Kumar, N.R. Bishnoi,N. Singh, Biosorption optimization of lead (II), cadmium (II) and copper (II) using response surface methodology and applicability in isotherms and thermodynamics modeling. J. Hazard. Mater., 174 (2010) (1-3) 623-634.

48. C. Monteiro, P.L. Castro,F. Xavier Malcata, Biosorption of zinc ions from aqueous solution by the microalga *Scenedesmus obliquus.* Environ. Chem. Lett., 9 (2011) (2) 169-176.

49. E. Malkoc,Y. Nuhoglu, The removal of chromium (VI) from synthetic wastewater by *Ulothrix zonata.* Fresenius Environ. Bull., 12 (2003) (4) 376-381.

50. S.K. Mehta,J.P. Gaur, Characterization and optimization of Ni and Cu sorption from aqueous solution by *Chlorella vulgaris.* Ecol. Eng., 18 (2001) (1) 1-13.

51. S. Shanab,A. Essa, Heavy metal Tolerance, biosorption and bioaccumulation by some microalgae (Egyptian isolates). New Egypt. J. Microbiol., 17 (2007) (1) 76-88.

52. K.K.I.U. Arunakumara,X. Zhang, Heavy metal bioaccumulation and toxicity with special reference to microalgae. J. Ocean Univ. China, 7 (2008) (1) 60-64.

53. X. Han, Y.S. Wong,N.F. Tam, Surface complexation mechanism and modeling in Cr(III) biosorption by a microalgal isolate, *Chlorella miniata.* J. Colloid Interface Sci., 303 (2006) (2) 365-71.

54. V.K. Gupta, A.K. Shrivastava,N. Jain, Biosorption of Chromium(VI) From Aqueous solutions by green algae *spirogyra species.* Water Res., 35 (2001) (17) 4079-4085.

55. Y. Zeroual, A. Moutaouakkil, F. Zohra Dzairi, M. Talbi, P. Ung Chung, K. Lee,M. Blaghen, Biosorption of mercury from aqueous solution by *Ulva lactuca* biomass. Bioresour. Technol., 90 (2003) (3) 349-351.

56. N. Rangsayatorn, E. Upatham, M. Kruatrachue, P. Pokethitiyook,G. Lanza, Phytoremediation potential of *Spirulina*(*Arthrospira*) *platensis*: biosorption and toxicity studies of cadmium. Environ. Pollut., 119 (2002) (1) 45-53.

57. P.X. Sheng, Y.P. Ting,J.P. Chen, Biosorption of heavy metal ions (Pb, Cu, and Cd) from aqueous solutions by the marine alga *Sargassum sp.* in single-and multiple-metal systems. Ind. Eng. Chem. Res., 46 (2007) (8) 2438-2444.

58. V.K. Gupta,A. Rastogi, Biosorption of lead from aqueous solutions by green algae *Spirogyra species*: kinetics and equilibrium studies. J. Hazard. Mater., 152 (2008) (1) 407-414.

59. M. Raeesossadati, H. Ahmadzadeh, M. McHenry,N. Moheimani, CO2 Environmental Bioremediation by Microalgae, in Biomass and Biofuels from Microalgae, Moheimani N.R., McHenry M.P., de Boer K.,Bahri P.A., Editors. 2015, Springer International Publishing. p. 117-136.

60. M.J. Raeesossadati, H. Ahmadzadeh, M.P. McHenry,N.R. Moheimani, CO2 bioremediation by microalgae in photobioreactors: Impacts of biomass and CO2 concentrations, light, and temperature. Algal Research, 6, Part A (2014) (0) 78-85.

61. H. Ahmadzadeh, M. Prescott, N. Muster,A. Stoyanov, Revisiting electroosmotic flow: an important parameter affecting separation in capillary and microchip electrophoresis. Chem. Eng. Commun., 195 (2007) (2) 129-146.

62. Z. Aksu, Ü. Acikel,T. Kutsal, Investigation of simultaneous biosorption of copper (II) and chromium (VI) on dried *Chlorella vulgaris* from binary metal mixtures: application of multicomponent adsorption isotherms. Sep. Sci. Technol., 34 (1999) (3) 501-524.

63. G. Cimino, A. Passerini,G. Toscano, Removal of toxic cations and Cr (VI) from aqueous solution by hazelnut shell. Water Research, 34 (2000) (11) 2955-2962.

64. M.M. Areco, S. Hanela, J. Duran,S. Afonso Mdos, Biosorption of Cu(II), Zn(II), Cd(II) and Pb(II) by dead biomasses of green alga *Ulva lactuca* and the development of a sustainable matrix for adsorption implementation. J. Hazard. Mater., 213-214 (2012) 123-132.

65. N. Tamilselvan, K. Saurav,K. Kannabiran, Biosorption of Cr (VI), Cr (III), Pb (II) and Cd (II) from aqueous solutions by *Sargassum wightii* and *Caulerpa racemosa* algal biomass. J. Ocean Univ. China, 11 (2012) (1) 52-58.

66. S.V. Gokhale, K.K. Jyoti,S.S. Lele, Kinetic and equilibrium modeling of chromium (VI) biosorption on fresh and spent *Spirulina platensis/Chlorella vulgaris* biomass. Bioresour. Technol., 99 (2008) (9) 3600-8.

67. I. Singleton,P. Simmons, Factors affecting silver biosorption by an industrial strain of *Saccharomyces cerevisiae.* J. Chem. Technol. Biotechnol., 65 (1996) (1) 21-28.

68. E. Finocchio, A. Lodi, C. Solisio,A. Converti, Chromium (VI) removal by methylated biomass of Spirulina platensis: The effect of methylation process. Chem. Eng. J., 156 (2010) (2) 264-269.

69. S.K. Mehta,J.P. Gaur, Removal of Ni and Cu from single and binary metalsolutions by free and immobilized *Chlorella vulgaris.* Eur. J. Protistol., 37 (2001) (3) 261-271.

70. A. Fraile, S. Penche, F. González, M.L. Blázquez, J.A. Muñoz,A. Ballester, Biosorption of copper, zinc, cadmium and nickel by *Chlorella vulgaris.* Chem. Ecol., 21 (2005) (1) 61-75.

71. P.A. Terry,W. Stone, Biosorption of cadmium and copper contaminated water by *Scenedesmus abundans.* Chemosphere, 47 (2002) (3) 249-255.

72. V.K. Gupta, A. Rastogi,A. Nayak, Biosorption of nickel onto treated alga (*Oedogonium hatei*): Application of isotherm and kinetic models. J. Colloid Interface Sci., 342 (2010) (2) 533-9.

73. L. Deng, Y. Su, H. Su, X. Wang,X. Zhu, Biosorption of copper (II) and lead (II) from aqueous solutions by nonliving green algae *Cladophora fascicularis*: Equilibrium, kinetics and environmental effects. Adsorption, 12 (2006) (4) 267-277.

74. L. Deng, Y. Su, H. Su, X. Wang,X. Zhu, Sorption and desorption of lead (II) from wastewater by green algae *Cladophora fascicularis.* J. Hazard. Mater., 143 (2007) (1–2) 220-225.

75. M.A. Khan, M. Ngabura, T.S. Choong, H. Masood,L.A. Chuah, Biosorption and desorption of Nickel on oil cake: batch and column studies. Bioresour. Technol., 103 (2012) (1) 35-42.

76. A. Sari,M. Tuzen, Equilibrium, thermodynamic and kinetic studies on aluminum biosorption from aqueous solution by brown algae (*Padina pavonica*) biomass. J. Hazard. Mater., 171 (2009) (1-3) 973-9.

77. Y. Sağ,T. Kutsal, Determination of the biosorption heats of heavy metal ions on *Zoogloea ramigera* and *Rhizopus arrhizus.* Biochem. Eng. J., 6 (2000) (2) 145-151.

78. A.K. Meena, G.K. Mishra, P.K. Rai, C. Rajagopal,P.N. Nagar, Removal of heavy metal ions from aqueous solutions using carbon aerogel as an adsorbent. J. Hazard. Mater., 122 (2005) (1–2) 161-170.

79. G. Bayes, S. Raut, V. Patil,R. Lokhande, Formation of Diazepam–Lanthanides(III) Complexes in the 50–50 Volume % Ethanol–Water Solvent System and Study of the Effect of Temperature on the Complex Formation Constants. J. Solution Chem., 41 (2012) (2) 241-248.

80. D.C. Harris, Quantitative chemical analysis. 8 ed. Clancy Marshall, 2010.

81. M. Tuzen, A. Sari, D. Mendil, O.D. Uluozlu, M. Soylak,M. Dogan, Characterization of biosorption process of As(III) on green algae *Ulothrix cylindricum.* J. Hazard. Mater., 165 (2009) (1-3) 566-72.

82. P. Lodeiro, J.L. Barriada, R. Herrero,M.E. Sastre de Vicente, The marine macroalga *Cystoseira baccata* as biosorbent for cadmium(II) and lead(II) removal: Kinetic and equilibrium studies. Environ. Pollut., 142 (2006) (2) 264-273.

83. R.J.E. Martins, R. Pardo,R.A.R. Boaventura, Cadmium (II) and zinc (II) adsorption by the aquatic moss *Fontinalis antipyretica*: effect of temperature, pH and water hardness. Water Res., 38 (2004) (3) 693-699.

84. S. Mehta, B. Tripathi,J. Gaur, Influence of pH, temperature, culture age and cations on adsorption and uptake of Ni by *Chlorella vulgaris*. Eur. J. Protistol., 36 (2000) (4) 443-450.

85. Z. Aksu, Equilibrium and kinetic modelling of cadmium(II) biosorption by *C. vulgaris* in a batch system: effect of temperature. Sep. Purif. Technol., 21 (2001) (3) 285-294.

86. C.C.V. Cruz, A.C.A. Costa, C.A. Henriques,A.S. Luna, Kinetic modeling and equilibrium studies during cadmium biosorption by dead *Sargassum sp.* biomass. Bioresour. Technol., 91 (2004) (3) 249-257.

87. A. Sarı,M. Tuzen, Biosorption of cadmium(II) from aqueous solution by red algae (*Ceramium virgatum*): Equilibrium, kinetic and thermodynamic studies. J. Hazard. Mater., 157 (2008) (2–3) 448-454.

88. A. Sarı,M. Tuzen, Biosorption of total chromium from aqueous solution by red algae (*Ceramium virgatum*): Equilibrium, kinetic and thermodynamic studies. J. Hazard. Mater., 160 (2008) (2–3) 349-355.

89. Z. Aksu, Determination of the equilibrium, kinetic and thermodynamic parameters of the batch biosorption of nickel(II) ions onto *Chlorella vulgaris.* Process Biochem., 38 (2002) (1) 89-99.

90. M. Dundar, C. Nuhoglu,Y. Nuhoglu, Biosorption of Cu(II) ions onto the litter of natural trembling poplar forest. J. Hazard. Mater., 151 (2008) (1) 86-95.

91. E. Malkoc,Y. Nuhoglu, Determination of kinetic and equilibrium parameters of the batch adsorption of Cr(VI) onto waste acorn of *Quercus ithaburensis.* Chem. Eng. Process., 46 (2007) (10) 1020-1029.

92. V.K. Gupta,A. Rastogi, Sorption and desorption studies of chromium(VI) from nonviable cyanobacterium *Nostoc muscorum* biomass. J. Hazard. Mater., 154 (2008) (1–3) 347-354.

93. M. Vogel, A. Gunther, A. Rossberg, B. Li, G. Bernhard,J. Raff, Biosorption of U(VI) by the green algae *Chlorella vulgaris* in dependence of pH value and cell activity. Sci. Total Environ., 409 (2010) (2) 384-395.

94. İ. Tüzün, G. Bayramoğlu, E. Yalçın, G. Başaran, G. Çelik,M.Y. Arıca, Equilibrium and kinetic studies on biosorption of Hg (II), Cd (II) and Pb (II) ions onto microalgae *Chlamydomonas reinhardtii.* J. Environ. Manage., 77 (2005) (2) 85-92.

95. F. Vegliò, R. Quaresima, P. Fornari,S. Ubaldini, Recovery of valuable metals from electronic and galvanic industrial wastes by leaching and electrowinning. Waste Manage., 23 (2003) (3) 245-252.

96. A. Bajguz, Suppression of *Chlorella vulgaris* Growth by Cadmium, Lead, and Copper Stress and Its Restoration by *Endogenous Brassinolide.* Arch. Environ. Contam. Toxicol., 60 (2011) (3) 406-416.

97. M.M. El-Sheekh, W.A. El-Shouny, M.E.H. Osman,E.W.E. El-Gammal, Growth and heavy metals removal efficiency of *Nostoc muscorum* and *Anabaena subcylindrica* in sewage and industrial wastewater effluents. Environ. Toxicol. Pharmacol., 19 (2005) (2) 357-365.

98. J. Bayo, Kinetic studies for Cd(II) biosorption from treated urban effluents by native grapefruit biomass (*Citrus paradisi L.*): The competitive effect of Pb(II), Cu(II) and Ni(II). Chem. Eng., 191 (2012) (0) 278-287.

99. M.H. El-Naas, F.A. Al-Rub, I. Ashour,M. Al Marzouqi, Effect of competitive interference on the biosorption of lead(II) by *Chlorella vulgaris.* Chem. Eng. Process., 46 (2007) (12) 1391-1399.

100. Z. Aksu,G. Dönmez, Binary biosorption of cadmium(II) and nickel(II) onto dried *Chlorella vulgaris*: Co-ion effect on mono-component isotherm parameters. Process Biochem., 41 (2006) (4) 860-868.

101. S. Schiewer,M.H. Wong, Ionic strength effects in biosorption of metals by marine algae. Chemosphere, 41 (2000) (1–2) 271-282.

102. V.J.P. Vilar, C.M.S. Botelho,R.A.R. Boaventura, Copper removal by algae *Gelidium*, agar extraction algal waste and granulated algal waste: Kinetics and equilibrium. Bioresour. Technol., 99 (2008) (4) 750-762.

103. A. Sulaymon, A. Mohammed,T. Al-Musawi, Competitive biosorption of lead, cadmium, copper, and arsenic ions using algae. Environ. Sci. Pollut. Res., 20 (2012) (5) 1-13.

104. H.S. Lee, J.H. Suh, I.B. Kim,T. Yoon, Effect of aluminum in two-metal biosorption by an algal biosorbent. Miner. Eng., 17 (2004) (4) 487-493.

105. H.S. Lee,B. Volesky, Interference of aluminum in copper biosorption by an Algal Biosorbent. Water Qual. Res. J. Can., 34 (1999) (3) 519-533.

106. P. Kaewsarn, Q. Yu,W. Ma, Interference of Co-ions in Biosorption of Cu2+ by Biosorbent from Marine Alga *Durvillaea potatorum.* Environ. Eng. Sci., 18 (2001) (2) 99-104.

107. M. Taziki, H. Ahmadzadeh,M.A. Murry, Growth of *Chlorella vulgaris* in High Concentrations of Nitrate and Nitrite for Wastewater Treatment. Current Biotechnology, 4 (2015) 1-7.

108. M. Taziki, H. Ahmadzadeh,M.A. Murry, Nitrate and Nitrite Removal from Wastewater using Algae. Current Biotechnology, 4 (2015) 1-15.

109. G.W. Garnham, G.A. Codd,G.M. Gadd, Kinetics of uptake and intracellular location of cobalt, manganese and zinc in the estuarine green alga *Chlorella salina.* Appl. Microbiol. Biotechnol., 37 (1992) (2) 270-276.

110. V. Subramanian, V. Sivasubramanian,K. Gowrinathan, Uptake and recovery of heavy metals by immobilized cells of *aphasocapsa polchra* (Kütz.) Rabenh. J. Environ. Sci. Health. Part A Toxic/Hazard. Subst. Environ. Eng. , 29 (1994) (9) 1723-1733.

111. M. Ota, Y. Kato, M. Watanabe, Y. Sato, R.L. Smith Jr, R. Rosello-Sastre, C. Posten,H. Inomata, Effects of nitrate and oxygen on photoautotrophic lipid production from *Chlorococcum littorale.* Bioresour. Technol., 102 (2011) (3) 3286-3292.

112. E. Lopez Errasquın,C. Vazquez, Tolerance and uptake of heavy metals by *Trichoderma atroviride* isolated from sludge. Chemosphere, 50 (2003) (1) 137-143.

113. J. Rincón, F. González, A. Ballester, M.L. Blázquez,J.A. Muñoz, Biosorption of heavy metals by chemically-activated alga *Fucus vesiculosus.* J. Chem. Technol. Biotechnol., 80 (2005) (12) 1403-1407.

114. N.R. Bishnoi, R. Kumar, S. Kumar,S. Rani, Biosorption of Cr(III) from aqueous solution using algal biomass *spirogyra spp.* J. Hazard. Mater., 145 (2007) (1-2) 142-7.

115. R. Dabbagh, M. Ebrahimi, F. Aflaki, H. Ghafourian,M.H. Sahafipour, Biosorption of stable cesium by chemically modified biomass of *Sargassum glaucescens* and *Cystoseira indica* in a continuous flow system. J. Hazard. Mater., 159 (2008) (2-3) 354-7.

116. M. Ebrahimi, R. Panahi,R. Dabbagh, Evaluation of native and chemically modified *Sargassum glaucescens* for continuous biosorption of Co(II). Appl. Biochem. Biotechnol., 158 (2009) (3) 736-46.

117. P. Srinivasa Rao, S. Kalyani, K.V.N. Suresh Reddy,A. Krishnaiah, Comparison of Biosorption of Nickel (II) and Copper (II) Ions from Aqueous Solution by *Sphaeroplea* Algae and Acid Treated *Sphaeroplea* Algae. Sep. Sci. Technol., 40 (2005) (15) 3149-3165.

118. M.Y. Arıca, İ. Tüzün, E. Yalçın, Ö. İnce,G. Bayramoğlu, Utilisation of native, heat and acid-treated microalgae *Chlamydomonas reinhardtii* preparations for biosorption of Cr(VI) ions. Process Biochem., 40 (2005) (7) 2351-2358.

119. Y. Zhao, Y. Hao,G.J. Ramelow, Evaluation of treatment techniques for increasing the uptake of metal ions from solution by nonliving seaweed algal biomass. Environ. Monit. Assess., 33 (1994) (1) 61-70.

120. S. Klimmek, H.J. Stan, A. Wilke, G. Bunke,R. Buchholz, Comparative analysis of the biosorption of cadmium, lead, nickel, and zinc by algae. Environ. Sci. Technol., 35 (2001) (21) 4283-4288.

121. X. Zhang, S. Luo, Q. Yang, H. Zhang,J. Li, Accumulation of uranium at low concentration by the green alga *Scenedesmus obliquus* 34. J. Appl. Psychol. , 9 (1997) (1) 65-71.

122. J. Wang,C. Chen, Biosorption of heavy metals by *Saccharomyces cerevisiae*: A review. Biotechnol. Adv., 24 (2006) (5) 427-451.

123. G. Yan,T. Viraraghavan, Effect of pretreatment on the bioadsorption of heavy metals on *Mucor rouxii.* Water SA, 26 (2000) (1) 119-124.

124. D. Mani,C. Kumar, Biotechnological advances in bioremediation of heavy metals contaminated ecosystems: an overview with special reference to phytoremediation. International Journal of Environmental Science and Technology, 11 (2014) (3) 843-872.

125. E. Fourest,B. Volesky, Contribution of sulfonate groups and alginate to heavy metal biosorption by the dry biomass of Sargassum fluitans. Environmental Science & Technology, 30 (1995) (1) 277-282.

126. C. Bertagnolli, A. Uhart, J.-C. Dupin, M.G.C. da Silva, E. Guibal,J. Desbrieres, Biosorption of chromium by alginate extraction products from Sargassum filipendula: Investigation of adsorption mechanisms using X-ray photoelectron spectroscopy analysis. Bioresource technology, 164 (2014) 264-269.

127. Y.C. Lee,S.P. Chang, The biosorption of heavy metals from aqueous solution by *Spirogyra* and *Cladophora filamentous* macroalgae. Bioresour. Technol., 102 (2011) (9) 5297-5304.

128. K. Larsdotter, A. Norström, L. Gumaelius, J.L.C. Jansen,G. Dalhammar. A small scale hydroponics wastewater treatment system under Swedish conditions. in *Water science and technology*. (2003).

129. B. Godlewska-Żyłkiewicz, Analytical applications of living organisms for preconcentration of trace metals and their speciation. Crit. Rev. Anal. Chem., 31 (2001) (3) 175-189.

130. G.M. Gadd, Accumulation of metals by microorganisms and algae. Biotechnology, 6 (1988) 401-433.

131. V.O. Arief, K. Trilestari, J. Sunarso, N. Indraswati,S. Ismadji, Recent Progress on Biosorption of Heavy Metals from Liquids Using Low Cost Biosorbents: Characterization, Biosorption Parameters and Mechanism Studies. Clean, 36 (2008) (12) 937-962.

132. C.K. Tsang, P.S. Lau, N.F.Y. Tam,Y.S. Wong, Biodegradation capacity of tributyltin by two *Chlorella* species. Environ. Pollut., 105 (1999) (3) 289-297.

133. N.F.Y. Tam, A.M.Y. Chong,Y.S. Wong, Removal of tributyltin (TBT) by live and dead microalgal cells. Mar. Pollut. Bull., 45 (2002) (1–12) 362-371.

134. Z. Aksu,G. Dönmez, Comparison of copper (II) biosorptive properties of live and treated *Candida sp.* J. Environ. Sci. Health. Part A Toxic/Hazard. Subst. Environ. Eng., 36 (2001) (3) 367-381.

135. C.Y. Chen, H.W. Chang, P.C. Kao, J.L. Pan,J.S. Chang, Biosorption of cadmium by CO(2)-fixing microalga *Scenedesmus obliquus* CNW-N. Bioresour. Technol., 105 (2012) 74-80.

136. N.M. Franklin, J.L. Stauber, R.P. Lim,P. Petocz, Toxicity of metal mixtures to a tropical freshwater alga (Chlorella sp.): The effect of interactions between copper,

cadmium, and zinc on metal cell binding and uptake. Environmental toxicology and chemistry, 21 (2002) (11) 2412-2422.

137. P. Pavasant, R. Apiratikul, V. Sungkhum, P. Suthiparinyanont, S. Wattanachira,T.F. Marhaba, Biosorption of Cu2+, Cd2+, Pb2+, and Zn2+ using dried marine green macroalga *Caulerpa lentillifera.* Bioresour. Technol., 97 (2006) (18) 2321-2329.

138. X. Guo, S. Zhang,X.-q. Shan, Adsorption of metal ions on lignin. J. Hazard. Mater., 151 (2008) (1) 134-142.

139. P.X. Sheng, Y.-P. Ting, J.P. Chen,L. Hong, Sorption of lead, copper, cadmium, zinc, and nickel by marine algal biomass: characterization of biosorptive capacity and investigation of mechanisms. J. Colloid Interface Sci., 275 (2004) (1) 131-141.

140. Q.X. Kong, L. Li, B. Martinez, P. Chen,R. Ruan, Culture of microalgae *Chlamydomonas reinhardtii* in wastewater for biomass feedstock production. Appl. Biochem. Biotechnol., 160 (2010) (1) 9-18.

141. D. Skoog, D. West, F. Holler,S. Crouch, Fundamentals of analytical chemistry. Cengage Learning, 2013.

142. F. Fu,Q. Wang, Removal of heavy metal ions from wastewaters: A review. J. Environ. Manage., 92 (2011) (3) 407-418.

143. N. Rangsayatorn, P. Pokethitiyook, E.S. Upatham,G.R. Lanza, Cadmium biosorption by cells of *Spirulina platensis* TISTR 8217 immobilized in alginate and silica gel. Environ. Int., 30 (2004) (1) 57-63.

144. A. López, N. Lázaro,A.M. Marqués, The interphase technique: a simple method of cell immobilization in gel-beads. J. Microbiol. Methods, 30 (1997) (3) 231-234.

145. G. Bayramoğlu, I. Tuzun, G. Celik, M. Yilmaz,M.Y. Arica, Biosorption of mercury(II), cadmium(II) and lead(II) ions from aqueous system by microalgae *Chlamydomonas reinhardtii* immobilized in alginate beads. Miner. Process., 81 (2006) (1) 35-43.

146. G. Bayramoglu,M. Yakup Arica, Construction a hybrid biosorbent using *Scenedesmus quadricauda* and Ca-alginate for biosorption of Cu(II), Zn(II) and Ni(II): kinetics and equilibrium studies. Bioresour. Technol., 100 (2009) (1) 186-93.

147. A. Nakajima, T. Horikoshi,T. Sakaguchi, Recovery of uranium by immobilized microorganisms. Eur. J. Appl. Microbiol. Biotechnol., 16 (1982) (2-3) 88-91.

148. E. Eroglu, S. Smith,C. Raston, Application of Various Immobilization Techniques for Algal Bioprocesses, in Biomass and Biofuels from Microalgae, Moheimani N.R., McHenry M.P., de Boer K.,Bahri P.A., Editors. 2015, Springer International Publishing. p. 19-44.

149. C. Bailliez, C. Largeau, C. Berkaloff,E. Casadevall, Immobilization of *Botryococcus braunii* in alginate: influence on chlorophyll content, photosynthetic activity and degeneration during batch cultures. Appl. Microbiol. Biotechnol., 23 (1986) (5) 361-366.

150. J. Bozeman, B. Koopman,G. Bitton, Toxicity testing using immobilized algae. Aquat. Toxicol., 14 (1989) (4) 345-352.

151. F.A. Al-Rub, M. El-Naas, F. Benyahia,I. Ashour, Biosorption of nickel on blank alginate beads, free and immobilized algal cells. Process Biochem., 39 (2004) (11) 1767-1773.

152. A. Murugesan, S. Maheswari,G. Bagirath, Biosorption of cadmium by live and immobilized cells of *Spirulina platensis.* Int. J. Environ. Res., (2008).

153. B.P. Rosen, Transport and detoxification systems for transition metals, heavy metals and metalloids in eukaryotic and prokaryotic microbes. Comp. Biochem. Physiol. A: Mol. Integr. Physiol., 133 (2002) (3) 689-693.

154. M. Ghosh, J. Shen,B.P. Rosen, Pathways of As(III) detoxification in *Saccharomyces cerevisiae.* Proc. Natl. Acad. Sci. U.S.A. , 96 (1999) (9) 5001-5006.

155. M.M. Gharieb,G.M. Gadd, Role of glutathione in detoxification of metal(loid)s by *Saccharomyces cerevisiae.* Biometals, 17 (2004) (2) 183-188.

156. K. Kuroda, M. Ueda, S. Shibasaki,A. Tanaka, Cell surface-engineered yeast with ability to bind, and self-aggregate in response to, copper ion. Appl. Microbiol. Biotechnol., 59 (2002) (2-3) 259-264.

157. W. Bae, C.H. Wu, J. Kostal, A. Mulchandani,W. Chen, Enhanced mercury biosorption by bacterial cells with surface-displayed MerR. Appl. Environ. Microbiol., 69 (2003) (6) 3176-3180.

158. C.M. Agapakis,P.A. Silver, Synthetic biology: exploring and exploiting genetic modularity through the design of novel biological networks. Mol. Biosyst., 5 (2009) (7) 704-713.

159. J. Kostal, G. Prabhukumar, U.L. Lao, A. Chen, M. Matsumoto, A. Mulchandani,W. Chen, Customizable Biopolymers for Heavy Metal Remediation. J. Nanopart. Res., 7 (2005) (4-5) 517-523.

160. M. Valls,V. de Lorenzo, Exploiting the genetic and biochemical capacities of bacteria for the remediation of heavy metal pollution. FEMS Microbiol. Rev., 26 (2002) (4) 327-338.

161. C. Sousa, P. Kotrba, T. Ruml, A. Cebolla,V. De Lorenzo, Metalloadsorption by *Escherichia coli* Cells displaying yeast and mammalian metallothioneins anchored to the outer membrane protein LamB. J. Bacteriol., 180 (1998) (9) 2280-2284.

162. R. Krishnaswamy,D.B. Wilson, Construction and characterization of an *Escherichia coli* strain genetically engineered for Ni (II) bioaccumulation. Appl. Environ. Microbiol., 66 (2000) (12) 5383-5386.

163. N.J. Robinson, S.K. Whitehall,J.S. Cavet, Microbial metallothioneins, in Advances in Microbial Physiology. 2001, Academic Press. p. 183-213.

164. M. Kiyono,H. Pan-Hou, Genetic Engineering of Bacteria for Environmental Remediation of Mercury. J. Health Sci., 52 (2006) (3) 199-204.

165. A.-M. Stomp, K.-H. Han, S. Wilbert, M.P. Gordon,S.D. Cunningham, Genetic Strategies for Enhancing Phytoremediation. Ann. N.Y. Acad. Sci., 721 (1994) (1) 481-491.

166. J. Gasperi, S. Garnaud, V. Rocher,R. Moilleron, Priority pollutants in wastewater and combined sewer overflow. Sci. Total Environ., 407 (2008) (1) 263-272.

167. J.K. Pittman, A.P. Dean,O. Osundeko, The potential of sustainable algal biofuel production using wastewater resources. Bioresour. Technol., 102 (2011) (1) 17-25.

168. J.B.K. Park, R.J. Craggs,A.N. Shilton, Wastewater treatment high rate algal ponds for biofuel production. Bioresour. Technol., 102 (2011) (1) 35-42.

169. R. Craggs, S. Heubeck, T. Lundquist,J. Benemann, Algal biofuels from wastewater treatment high rate algal ponds. Water Sci. Technol., 63 (2011) (4) 660-665.

170. Y.N. Mata, M.L. Blazquez, A. Ballester, F. Gonzalez,J.A. Munoz, Biosorption of cadmium, lead and copper with calcium alginate xerogels and immobilized *Fucus vesiculosus*. J Hazard Mater, 163 (2009) (2-3) 555-62.

171. J.W. Lee, Advanced biofuels and bioproducts. Springer, 2013.

172. X. Zeng, X. Guo, G. Su, M.K. Danquah, S. Zhang, Y. Lu, Y. Sun,L. Lin, Bioprocess considerations for microalgal-based wastewater treatment and biomass production. Renewable and Sustainable Energy Rev., 42 (2015) 1385-1392.

173. M. Kesaano,R.C. Sims, Algal biofilm based technology for wastewater treatment. Algal Research, 5 (2014) 231-240.

174. A. Ruiz-Marin, L.G. Mendoza-Espinosa,T. Stephenson, Growth and nutrient removal in free and immobilized green algae in batch and semi-continuous cultures treating real wastewater. Bioresour. Technol., 101 (2010) (1) 58-64.

175. R. Craggs, D. Sutherland,H. Campbell, Hectare-scale demonstration of high rate algal ponds for enhanced wastewater treatment and biofuel production. J. Appl. Phycol., 24 (2012) (3) 329-337.

176. J. Adhiya, X. Cai, R.T. Sayre,S.J. Traina, Binding of aqueous cadmium by the lyophilized biomass of *Chlamydomonas reinhardtii*. Colloids Surf., A, 210 (2002) (1) 1-11.

177. A. Sarı,M. Tuzen, Biosorption of Pb(II) and Cd(II) from aqueous solution using green alga (*Ulva lactuca*) biomass. J. Hazard. Mater., 152 (2008) (1) 302-308.

178. N. Akhtar, M. Iqbal, S.I. Zafar,J. Iqbal, Biosorption characteristics of unicellular green alga *Chlorella sorokiniana* immobilized in loofa sponge for removal of Cr(III). J. Environ. Sci. (China), 20 (2008) (2) 231-9.

179. D. Onyancha, W. Mavura, J.C. Ngila, P. Ongoma,J. Chacha, Studies of chromium removal from tannery wastewaters by algae biosorbents, *Spirogyra condensata* and *Rhizoclonium hieroglyphicum.* J. Hazard. Mater., 158 (2008) (2–3) 605-614.

180. G. Dönmez,Z. Aksu, Removal of chromium(VI) from saline wastewaters by *Dunaliella species.* Process Biochem., 38 (2002) (5) 751-762.

181. C.R. Jácome-Pilco, E. Cristiani-Urbina, L.B. Flores-Cotera, R. Velasco-García, T. Ponce-Noyola,R.O. Cañizares-Villanueva, Continuous Cr(VI) removal by *Scenedesmus incrassatulus* in an airlift photobioreactor. Bioresour. Technol., 100 (2009) (8) 2388-2391.

182. A. El-Sikaily, A.E. Nemr, A. Khaled,O. Abdelwehab, Removal of toxic chromium from wastewater using green alga *Ulva lactuca* and its activated carbon. J. Hazard. Mater., 148 (2007) (1–2) 216-228.

183. S. Mehta, B. Tripathi,J. Gaur, Enhanced sorption of Cu^{2+} and Ni^{2+} by acid-pretreated *Chlorella vulgaris* from single and binary metal solutions. J. Appl. Phycol., 14 (2002) (4) 267-273.

184. A. ÖZer, D. Özer,H. İbrahim Ekİz, The Equilibrium and Kinetic Modelling of the Biosorption of Copper(II) Ions on *Cladophora crispata.* Adsorption, 10 (2004) (4) 317-326.

185. S. Karthikeyan, R. Balasubramanian,C.S.P. Iyer, Evaluation of the marine algae *Ulva fasciata* and *Sargassum sp.* for the biosorption of Cu(II) from aqueous solutions. Bioresour. Technol., 98 (2007) (2) 452-455.

186. A. Singh, D. Kumar,J.P. Gaur, Copper(II) and lead(II) sorption from aqueous solution by non-living *Spirogyra neglecta.* Bioresour. Technol., 98 (2007) (18) 3622-3629.

187. A. Singh, D. Kumar,J.P. Gaur, Continuous metal removal from solution and industrial effluents using *Spirogyra* biomass-packed column reactor. Water Res., 46 (2012) (3) 779-788.

188. Y. Nuhoglu, E. Malkoc, A. Gürses,N. Canpolat, The removal of Cu(II) from aqueous solutions by *Ulothrix zonata.* Bioresour. Technol., 85 (2002) (3) 331-333.

189. J.P.K. Wong, Y.S. Wong,N.F.Y. Tam, Nickel biosorption by two chlorella species, *C. Vulgaris* (a commercial species) and *C. Miniata* (a local isolate). Bioresour. Technol., 73 (2000) (2) 133-137.

190. N. Akhtar, J. Iqbal,M. Iqbal, Removal and recovery of nickel(II) from aqueous solution by loofa sponge-immobilized biomass of *Chlorella sorokiniana:* characterization studies. J. Hazard. Mater., 108 (2004) (1–2) 85-94.

191. F. Luo, Y. Liu, X. Li, Z. Xuan,J. Ma, Biosorption of lead ion by chemically-modified biomass of marine brown algae *Laminaria japonica.* Chemosphere, 64 (2006) (7) 1122-1127.

192. M. Tuzen,A. Sarı, Biosorption of selenium from aqueous solution by green algae (*Cladophora hutchinsiae*) biomass: Equilibrium, thermodynamic and kinetic studies. Chem. Eng. J., 158 (2010) (2) 200-206.

193. D. Özer, A. Özer,G. Dursun, Investigation of zinc (II) adsorption on *Cladophora crispata* in a two staged reactor. J. Chem. Technol. Biotechnol., 75 (2000) (5) 410-416.

Table 1. Biosorption capacity of 14 different heavy metal ions using variant micro and macroalgal strains under optimal conditions. (The potential of macroalgal vs. microalgal strains and living vs. non-living cells are summarized).

Metal	**Algae species**	**Treatment**	**Max. sorption (mgg^{-1})**	**Optimal pH**	**Initial metal conc. (mgL^{-1})**	**Biomass conc. (gL^{-1})**	**Temp (C°)**	**Time (hour)**	**References**
Al(III)	Laminaria japonica #	$CaCl_2$	75.27	4.5		1		30	[104]
As(III)	*Ulothrix cylindricum*		67.2	6	10		20	1	[81]
Au(III)	*Fucus vesiculosus #*		74.05	7	100	1	23	8	[19]
Cd(II)	*Ascophyllum nodosum #*		87.7	6	50	0.5		2	[34]
	Asparagopsis armata #		32.3	6	50	0.5		2	[34]
	Chlorella vulgaris		85.3	4	200	0.75	20	2	[85]
	C. vulgaris		86.6	4	150	1	25		[100]
	Chondrus crispus #		75.2	6	50	0.5		2	[34]
	*Cladophora fracta**		4.08	5	8		25	192	[32]
	*Chlamydomonas reinhardtii**		42.6	6			25	1	[94]
	C. reinhardtii		145	7	989.21		23		[176]
	Codium vermilara		21.8	6	50	0.5		2	[34]
	Laminaria japonica #	$CaCl_2$	136.1	4.5		1		30	[104]
	Fucus spiralis #		114.9	6	50			2	[34]
	F. vesiculosus #		125.9	6		0.25		2	[113]
	Spirogyra insignis		22.9	6	50	1		2	[34]
	Ulva lactuca #		29.2	5	10		20	1	[177]
Cr(II)	*Laminaria japonica #*	$CaCl_2$	94.103	4.5		1		30	[104]

Table 1. continued

Metal	Algae species	Treatment	Max. sorption (mgg^{-1})	Optimal pH	Initial metal conc. (mgL^{-1})	Biomass conc. (gL^{-1})	Temp (C°)	Time (hour)	References
Cr(III)	*Chlorella miniata**		41.12	4.5	100		25	24	[53]
	C. sorokiniana		58.8	4		1	25		[178]
	Rhizoclonium heiroglyphicum #	HCl	11.81	4				2	[179]
	Spirogyra condensate	HCl	14.82	5				2	[179]
	Spirogyra sp.	HCHO	28.81	5	50		25	3	[114]
	Spirogyra sp.	NaOH	29.15	5	50		25	3	[114]
	Spirogyra sp.	$CaCl_2$	30.21	5	50		25	3	[114]
Cr(VI)	*Chlorella vulgaris*		140	1.5	250	1	25		[66]
	*Chlamydomonas reinhardtii**		18.2	2		0.6	25	2	[118]
	C. reinhardtii		25.6	2		0.6	25	2	[118]
	C. reinhardtii	HCl	21.2	2		0.6	25	2	[118]
	Dunaliella sp.1*		58.3	2	100	1	25	72	[180]
	Dunaliella sp.2*		45.5	2	100	1	25	72	[180]
	*Scenedesmus incrassatulus**		4.4	8.9			25	24	[181]
	Spirogyra sp.		14.7	2	5		18	2	[54]
	Spirogyra sp.	HNO_3	265	4		1	30	2	[26]
	Ulva lactuca #		10.61	1		2	25	2	[182]
Cu(II)	*Ascophyllum nodosum #*		58.8	4	50	0.5		2	[34]
	Asparagopsis armata #		21.3	5	50	0.5		2	[34]
	*Chlorella vulgaris**		89.19	3.5		0.005	25	0.5	[50]
	C. vulgaris		14.48	3.5		0.1	25	0.5	[50]
	C. vulgaris		420.67	3.5	31.77		25	3	[183]
	C. vulgaris	HCl	714.892	3.5	31.77		25	3	[183]

Table 1. continued.

Metal	Algae species	Treatment	Max. sorption (mgg^{-1})	Optimal pH	Initial metal conc. (mgL^{-1})	Biomass conc. (gL^{-1})	Temp (C°)	Time (hour)	References
	Chondrus crispus #		40.5	4	50	0.5		2	[34]
	Cladophora fascicularis #		102.309	5		2	25		[73]
	C. crispate #		57.5	4.5	200	1	25	0.5	[184]
	Cladophora sp. *#*		13.7	5	100		25	1	[127]
	Codium vermilara #		16.9	5	50	0.5		2	[34]
	Fucus. spiralis #		70.9	4	50	0.5		2	[34]
	F. vesiculosus #		61.63	5		0.25		2	[113]
	F. vesiculosus #		105.48	5			23	2	[20]
	F. vesiculosus #	$CaCl_2$	85.15	5		0.25		2	[113]
	Laminaria japonica #	CaCl2	101.038	4.5		1		30	[104]
	Sargassum sp. *#*		72.5	5.5		1	22	3	[185]
	Sphaeroplea sp.		140.43	4		1	33	1.5	[117]
	Sphaeroplea sp.	HCl	216.535	4		1	33	1.25	[117]
	Spirogyra insignis		19.3	4	50			2	[34]
	S. neglecta		115.3	4.5	100	0.1	25	0.16	[186]
	S. neglecta	urea-HCHO	30.17	4.5	50		25		[187]
	Spirogyra sp		38.2	5	100		25	1	[127]
	Ulothrix zonata		176.2	4.5		0.1	20	2	[188]
	Ulva fasciata #		73.5	5.5		1	22	3	[185]
Hg(II)	*U. lactuca #*		149.25	7			25	2	[55]
	*Chlamydomonas reinhardtii**		72.2	6			25	1	[94]

Table 1. continued

Metal	Algae species	Treatment	Max. sorption (mgg^{-1})	Optimal pH	Initial metal conc. (mgL^{-1})	Biomass conc. (gL^{-1})	Temp (C°)	Time (hour)	References
Ni(II)	*Ascophyllum nodosum #*		43.3	6	50	0.5		2	[34]
	Asparagopsis armata #		17.1	6	50	0.5		2	[34]
	*Chlorella miniata**		1.367	7.4				24	[189]
	C. sorokiniana		48.08	5	200	1	25	0.33	[190]
	*C. vulgaris**		0.641	7.4				24	[189]
	*C. vulgaris**		15.4	5	100	2.5	25	2	[151]
	*C. vulgaris**		23.47	5.5		0.005	25	0.5	[50]
	C. vulgaris		15.6	5	100	2.5	25	2	[151]
	C. vulgaris		20.23	5.5		0.1	25	0.5	[50]
	C. vulgaris		58.4	4.5	150	1	25		[100]
	C. vulgaris		59.29	4.5	5			1	[69]
	C. vulgaris		264.7	5.5	29.34	0.1	25	3	[183]
	C. vulgaris	HCl	437.84	5.5	29.34		25	3	[183]
	Chondrus crispus #		37.2	6	50	0.5		2	[34]
	Codium vermilara #		13.2	6	50	0.5		2	[34]
	Fucus spiralis#		50	6	50	0.5		2	[34]
	F. vesiculosus #		46.95	5		0.25		2	[113]
	Sphaeroplea sp		199.55	6		1	33	1.16	[117]
	Sphaeroplea sp	HCl	244.85	6		1	33	1	[117]
	Spirogyra insignis		17.5	6	50	1		2	[34]

Table 1. continued

Metal	Algae species	Treatment	Max. sorption (mgg^{-1})	Optimal pH	Initial metal conc. (mgL^{-1})	Biomass conc. (gL^{-1})	Temp (C°)	Time (hour)	References
Pb(II)	*Ascophyllum nodosum #*		178.6	3	50			2	[34]
	Asparagopsis armata #		63.7	4	50	0.5		2	[34]
	Chondrus crispus #		204.1	4	50			2	[34]
	Cladophora fascicularis#		198.5	5		2	25	1.5	[74]
	C. fracta*		61.400	5	80		25	192	[32]
	Cladophora sp#		45.4	5	100		25	1	[127]
	*Chlamydomonas reinhardtii**		96.3	5			25	1	[94]
	Codium vermilara#		63.3	5	50	0.5		2	[34]
	Fucus spiralis #		204.1	3	50			2	[34]
	F. vesiculosus #		211.34	5			23	2	[20]
	F. vesiculosus #		215.48	5		0.25		2	[113]
	F. vesiculosus #	$CaCl_2$	259	5		0.5		2	[113]
	Laminaria japonica #		250.71	5.3			25	2	[191]
	L. japonica #	$KMnO_4$	319.08	5.3			25	2	[191]
	L. japonica[1]*#*	$C_3H_5ClO^2$	335.66	5.3			25	2	[191]
	L. japonica[3]*#*	C_3H_5ClO	346.02	5.3			25	2	[191]
	L. japonica#	$CaCl_2$	348.09	4.5		1		30	[104]
	Spirogyra insignis		51.5	5	50	0.5		2	[34]
	S. neglecta		116.1	5	100	0.1	25	0.33	[186]
	Spirogyra sp		87.2	5	100		25	1	[127]
	Spirogyra sp		140	5	200	0.5	25	1.66	[58]
	Ulva lactuca		34.7	5	10		20	1	[177]
Se(IV)	*Cladophora hutchinsiae#*		74.9	5		8	20	1	[192]
U(VI)	*Chlorella vulgaris**		14.3	4.4	23.8	0.76		0.08	[93]
	*C. vulgaris**		26.6	4.4	23.8	0.76		96	[93]
	C. vulgaris		27	4.4	23.8	0.76		96	[93]

Table 1. continued

Metal	Algae species	**Treatment**	**Max. sorption (mgg^{-1})**	**Optimal pH**	**Initial metal conc. (mgL^{-1})**	**Biomass conc. (gL^{-1})**	**Temp (C°)**	**Time (hour)**	**References**
Zn(II)	*Ascophyllum nodosum #*		42	6	50	0.5			[34]
	Asparagopsis armata #		21.6	6	50	0.5		2	[34]
	Chondrus crispus #		45.7	6	50	0.5		2	[34]
	Cladophora crispate#		31.06	5	100	1	25	2	[193]
	Codium vermilara#		23.8	6	50	0.5		2	[34]
	Fucus spiralis#		53.2	6	50	0.5		2	[34]
	laminaria japonica #	$CaCl_2$	56.88	4.5		1		30	[104]
	*Scenedesmus. obliqus(ACOI598)**		429.6	6-7	75	0.02	25	24	[48]
	*S. obliqus(L)**		836.5	6-7	75	0.02	25	24	[48]
	S. obliqus(L)		209.6	6-7	50	0.02	25	1.5	[48]
	Spirogyra insignis		21.1	6	50	1		2	[34]

*: living algae #: seaweed/macroalgae

(1): washing with 2-propanol 20% (2): Epichlorohydrin (3): washing with 2-propanol 70%

Table 2. Functional groups in biological systems and three types of metals.

Ligand class	Ligands	Metal classes
I: ligands Preferred to Class A	F^-, O^{2-}, OH^-, H_2O, CO_3^{2-}, SO_4^- $ROSO_3^-$, NO_3^-, HPO_4^{2-}, PO_4^{3-}, ROH $RCOO^-$, C=O, ROR	Class A: Li, Be, Na, Mg, K, Ca, Sc, Rb, Sr, Y, Cs, Ba, La, Fr, Ra, Ac, Al, Lanthanides, actinides
II: Other Important ligands	Cl^-, Br^-, N_3^-, NO_2^-, SO_3^{2-}, NH_3, N_2, RNH_2, R_2NH, R_3N, =N–, –CO–N–R, O_2, O_2^-, O_2^{2-}	Borderline ions: Ti, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, Ga, Cd, In, SN, Sb, As
III: Ligands Preferred to Class B	H^-, I^-, R^-, CN^-, CO, S^{2-}, RS^-, R_2S, R_3AS	Class B: Rh, Pd, Ag, Lr, Pt, Au, Hg, Ti, Pb, Bi

(Adapted from Wang and Chen (2009), with permission)

Table 3. Important functional groups involved in metal ion biosorption.

Binding group	**Structural formula**	**pKa**	**Ligand atom**	**Occurrence in selected biomolecules**
Hydroxyl	–OH	9.5–13	O	PS, UA, SPS, AA
Carbonyl (ketone)	>C=O	–	O	Peptide bond
Carboxyl	–C=O | OH	1.7–4.7	O	UA, AA
Sulfhydryl (thiol)	–SH	8.3-10.8	S	AA
Sulfonate	O ‖ –S=O ‖ O	1.3	O	SPS
Thioether	>S	–	S	AA
Amine	–NH2	8–11	N	Cto, AA
Secondary amine	>NH	13	N	Cti, PG, Peptide bond
Amide	–C=O | NH2	–	N	AA
Imine	=NH	11.6–12.6	N	AA
Imidazole	–C-N-H ‖ >CH H-C-N	6.0	N	AA

PHospHonate	OH | –P=O | OH	0.9–2.1 6.1–6.8	O O	PL PL
Phosphodiester	>P=O | OH	1.5	O	TA, LPS

PS = polysaccharides; UA = uronic acids; SPS = sulfated PS; Cto = chitosan; PG = peptidoglycan; AA = amino acids; TA = teichoic acid; PL = phospholipids; LPS = lipoPS. (Adapted from Ref. Volesky 2007 with permission)

Table 4. Comparison of biosorption capacity of metal ions using algal biomass in the binary solution vs. sole systems**.**

Type of algae	**Metal ion**	**Binary solution**	**Maximum sorption (mgg^{-1})**		**Reference**
			Sole	**Binary**	
Chlorella vulgaris	Cd (II)	Cd, Ni Cd(II)	86.60	68.5	[100]
C. vulgaris	Ni(II)	Cd, Ni	58.4	28.3	
C. vulgaris	Ni(II)	Cu, Ni Ni(II)	264.69	25.82	[183]
C. vulgaris	Cu(II)	Ni, Cu	420.67	84.17	

Table 5. Comparison of biosorption capacity of metal ions using immobilized algal biomass vs. living algae.

Algae species	**Immobilization system**	**Initial metal ion conc. (mgL^{-1})**	**Metal ion**	**Max. sorption (mgg^{-1})**		**References**
				Living algae	Immobilized algae	
Chlamydomonas reinhardtii	Ca - alginate	500	Cd(II)	28.9	79.7	[145]
Chlorella sorokiniana	Loofa spong	300	Cr(III)	58.80	69.26	[178]
Scenedesmus quadricauda	Ca - alginate	600	Cu(II)	35.9	75.6	[146]
C. reinhardtii	Ca - alginate	500	Cu(II)	35.9	106.6	[145]
C. sorokiniana	Loofa spong	200	Ni(II)	48.08	60.38	[190]
C. vulgaris	Blank alginate	100	Ni(II)	15.6	28.6	[151]
S. quadricauda	Ca – alginate	600	Ni(II)	9.7	30.4	[146]
C. reinhardtii	Ca – alginate	500	Pb(II)	230.5	308.7	[145]
S. quadricauda	Ca – alginate		Zn(II)	20.2	55.2	[146]

Figure 1.

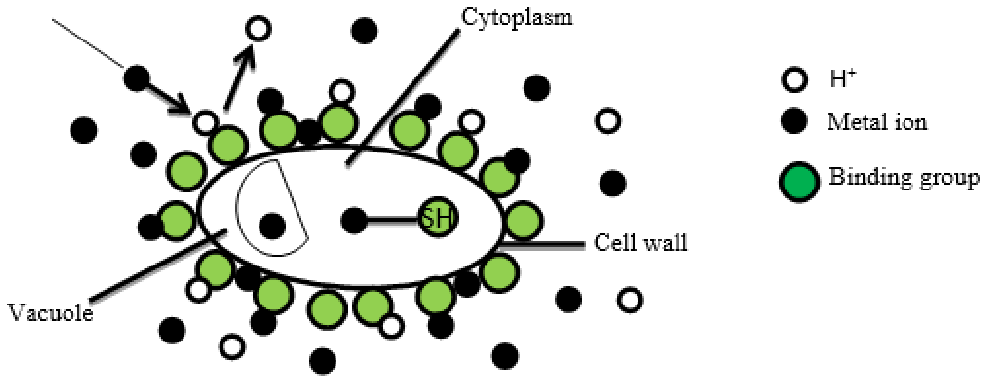

Figure Legends

Figure 1. Metal ion sorption by the algal cells. Different binding groups, i.e. OH^-, SH^-, COO^-, PO_4^{3-}, NO3$^-$, RNH_2^-, RS^-, RO^- and etc. promote the metal ion biosorption.