ADSORPTION OF HEAVY METALS ON MARINE ALGAE

by

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ABSTRACT

Biosorption is a property of certain type of inactive, microbial biomass to bind and concentrate heavy metals from even very dilute aqueous solutions. Biomass exhibits this property, acting just as a chemical substance, as an ion exchanger of biological origin. It is particularly the cell wall structure of certain algae that is found responsible for this phenomenon. In these experiments, the rate and extent for removal of copper is subjected to parameters such as pH, initial metal concentration, biosorbent size, contact time, temperature and the ability of the biomass to be regenerated in sorption-desorption experiments. The metal adsorption was found to be rapid within 25 minutes. The maximum copper uptake of 30 mg of copper / g of biomass has been observed, in the following conditions: 100 mg / L, 0.1 g of biomass, pH 4 and at temperature of 25^0C. From this study, it was found that copper uptake is increasing with increase in pH, with optimum being pH 4. Copper uptake increases substantially from 0 to 25 minutes.

Metal biosorption behaviour of raw seaweed *Sargassum* in six consecutive sorption-desorption cycles were also investigated in a packed-bed column, during a continuous removal of copper from a 35 mg/l aqueous solution at pH 4. The sorption and desorption was carried out for an average of 85 and 15 hours, respectively, representing more than 40 days of continuous use of the biosorbent. The weight loss of biomass after this time was 13.5%. The column service time decreased from 25 hrs in the first cycle to 10 hrs for the last cycle.

Nomenclature

Symbol	Description	Units
b	Ratio of adsorption and desorption rates	
C	Concentration of metal	mg/L
C_f	Equilibrium or final metal concentration	mg/L
C_i	Initial metal concentration	mg/L
C_{eq}	Metal residual concentration	mg/L
C_o	Metal concentration in the feed to a flow through column	mg/L
K_F, n	constants	
q	Metal uptake	mg/g
V	Volume of liquid sample	L
S	Amount of biosorbent on dry basis	g
q_{MAX}	Maximum metal uptake	mg/g
C_A, C_B	Divalent metal concentration	mg/g
C_{AB}	Total concentration of metal A, B in the feed to a flow-through column	mg/L
K_{AB}	Equilibrium ion exchange constant	
Q	Total concentration of metal binding sites in the biosorbent	mg/g
x_A	Equilibrium equivalent fraction of species A in the liquid	
y_A	Equilibrium equivalent fraction of species A in the sorbent	
T	Dimensionless time	
C/C_i	Dimensionless metal concentration	
C_e	Exit concentration	mg/L
t_b	Breakthrough time	hr
t_e	Exhaustion time	hr

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CHAPTER 1

INTRODUCTION

Aqueous heavy metal pollution represents an important environmental problem due to its toxic effects and accumulation throughout the food chain. The main sources of heavy metals pollution are mining, refining ores, sludge disposal, fly ash from incinerators, the processing of radioactive materials, metal plating, or manufacture of electrical equipment, paints, alloys, batteries, pesticides or preservatives, milling and surface finishing industries, discharging a variety of toxic metals such as cadmium, chromium, copper, zinc, lead, mercury, selenium, arsenic, gold, silver and nickel into an environment. Heavy metals such as zinc, lead and chromium have a number of applications in basic engineering works, paper and pulp industries, leather tanning, organochemicals, petrochemicals fertilizers, etc. Major lead pollution is through automobiles and battery manufacturers. As a result, removal of these toxins from industrial effluents has become an important priority that is reflected in tightening and enforcement of environmental regulations.

Since copper is a widely used material, there are many actual or potential sources of copper pollution. Copper may be found as a contaminant in food, especially shellfish, liver, mushroom, nuts, and chocolate. Briefly, any processing or container using copper material may contaminate the product, such as food, water or drink. Copper is essential to human life and health but, like all heavy metals, is potentially toxic as well.

While the removal of toxic heavy metals from industrial wastewaters has been practiced for several decades, the effectiveness, and particularly the cost effectiveness of the most common physical-chemical processes is limited. Biological materials have shown potential for heavy metal removal, but only low-cost biological materials with sufficiently high metal-binding capacity and selectivity for heavy metals are suitable in a full-scale biosorption process [2, 3].

Various bio-materials have been examined for their biosorptive properties and different types of biomass have shown levels of metal uptake high enough (in the order of 1mmol/g) to warrant further research [5]. Among the most promising types of biosorbents studied is the algal biomass. The abundance of algae can hardly be overestimated. Biosorption in algae has mainly been attributed to the cell wall, composed of the fibrilar skeleton [1, 6].

Due to the increasing awareness of the ecological effects of heavy toxic metals, a number of studies of metal accumulation from the view point of their removal from aqueous solution have been launched [3]. Unlike organic pollutants, which in most cases can eventually be destroyed, metallic species released into the environment tend to persist indefinitely, circulating and eventually accumulating throughout the food chain posing thus serious threat to animals and man [3, 8].

Large portions of heavy metals are released into the environment from industrial waters through inefficiencies built into the technological activities used directly in the processing of metals or through other routes. Virtually any industrial activity using metals has a metal disposal problem [8, 9, 10].

Conventional methods for removing metals from aqueous solutions before they are disposed of include chemical precipitation and sludge separation, chemical oxidation or reduction, ion exchange, reverse osmosis, electrochemical treatment, and evaporation. While the cheaper of these processes are becoming inadequate with progressively more stringent regulatory effluent limits, methods that are more effective are invariably prohibitively costly. Alternative processing methods are considered more seriously as their understanding is developing [3].

Considering the number of metals and different biosorbent materials of interest, in the absence of theoretical apparatus, experimental testing of these effects requires a large volume of laboratory work [6]. Although copper is not considered as a major environmental problem, its ubiquitous presence in the solutions makes it an obvious investigation target as to its effect on biosorbent uptake of any other metals of

interest. Among them, cadmium is of a primary interest for its well-recognized acute toxicity [13].

Brown seaweeds (*Phaeophyceae*) constitute an algal group containing the characteristic pigment fucoxantine, responsible for their brown color. Floating masses of *Sargassum* constitute the Sargasso Sea, being also very common in the Brazilian Coast. All *Sargassum* species contain floating bubbles, responsible for their decreased density, thus contributing for their presence in the marine environment. Quantitatively, the most abundant polysaccharide in the cell wall of brown seaweeds is alginic acid. Alginic acid is a polymer constituted by two uronic acids (b-1,4-D-mannuronic and a-1,4-L guluronic), with molar ratio between the acids ranging from 0.25 to 2.5. Alginic acid is present in these seaweeds usually as calcium, magnesium, sodium and potassium salts, mainly in the cell wall. It is a structural polysaccharide with strong ion-exchange properties. Beyond its high metal uptake capacity, this algal genus has been selected for study due to their wide distribution in most tropical countries, being available at high quantities as a waste biomaterial. In this work, the use of *Sargassum* species as a biosorbent for copper ions from aqueous solution was studied. The influence of different parameters on copper uptake such as sorption time, initial pH, temperature, biomass sizes, biosorption kinetics and initial copper concentration was investigated.

Simple sorption isotherms curves in this investigation were constructed as a result of studying equilibrium batch sorption behavior of different biosorbent material characteristics, which enable quantitative evaluation of biosorption performance. The aim of this study is to investigate the adsorption characteristics of marine algae on copper with emphasis on its efficiency, stability and regeneration.

CHAPTER 2

LITERATURE REVIEW

2.1. ENVIRONMENTAL THREAT

By far the greatest demand for metal sequestration comes from the need of immobilizing the metals mobilized by and partially lost through human technological activities. It has been established beyond any doubt that dissolved particularly heavy metals escaping into the environment pose a serious health hazard [3, 5].

Nowadays, with the exponentially increasing population the need for controlling heavy metals emission into the environment is even more pronounced. This is best done right at the source of such emissions, before toxic metals enter the complex ecosystem [3].

The danger multiplies and humans eventually tend to receive the problems associated with the toxicity of heavy metals pre-concentrated and from many different directions. The resulting health problems demonstrate themselves on the acute as well as chronic levels and are reflected in the well-being of individuals and in society's spiraling health care cost [3,15]. Controlling heavy metal discharges and removing toxic heavy metals from aqueous solutions has become a challenge for 21st century [17].

2.1.1 Environmental pressures

- Under the public and media pressure, governments introduce and progressively enforce stricter regulations with regard to the metal discharges particularly for industrial operations.
- The compounding toxic effects of heavy metals in the environment are being recognized and their dangerous impacts better understood.

- The currently practiced technologies for removal of heavy metals from industrial effluents appear to be inadequate, creating often-secondary problems with metal-bearing sludges, which are extremely difficult to dispose of. Due to their classification as "toxic substances", they require special handling, disposal methods and sites. The governments in most instances closely monitor their handling and disposal.
- The currently available '' best treatment technologies'' for metal-bearing effluents are either not effective enough or are prohibitively expensive and inadequate considering the vast wastewater quantities [3]

2.1.2 Conventional metal-removal technologies

Municipal sewage treatment plants are not designed and equipped for handling toxic wastes. Metals and their toxicity persist even in the sludges and by-product streams of municipal sewage treatment plants. Heavy metals need to best be removed with a specific kind of treatment. This specific treatment needs to be cheap because it most often deals with large volumes of effluents [3].

Over the few decades, several methods have been devised for the treatment and removal of heavy metals. The commonly used procedures for removing metal ions from aqueous streams includes:

Reverse osmosis: It is a process in which heavy metals are separated by a semi-permeable membrane at a pressure greater than osmotic pressure caused by dissolved solids in wastewater. The disadvantage of this method is that it is expensive.

Electrodialysis: In this process, the ionic components (heavy metals) are separated through the use of semi-permeable ionselective membranes. Application of an electrical potential between the two electrodes causes a migration of cations and anions towards respective electrodes. Because of the alternate spacing of cation and anion permeable membranes, cells of concentrated and dilute salts are formed. The disadvantage is the formation of metal hydroxides, which clog the membrane.

Ultrafiltration: They are pressure driven membrane operations that use porous membranes for removal of heavy metals. The main disadvantage of this process is the generation of sludge.

Ion-exchange: In this process, metal ions from dilute solutions are exchanged with ions held by electrostatic forces on the exchange resin. The disadvantages include: high cost and partial removal of certain metals.

Chemical precipitation: Precipitation of metals is achieved by addition of coagulants such as alum, lime, iron salts and other organic polymers. The large amount of sludge containing toxic compounds produced during the process is the main disadvantage.

Phytoremediation: Phytoremediation is the use of certain plants to clean up soil, sediment, and water contaminated with metals. The disadvantages include that it takes a long time for removal of metals and a regeneration of the plant for a further biosorption is difficult.

Hence the disadvantages like incomplete metal removal, high reagent and energy requirements, generation of toxic sludge or other waste products that require careful disposal has made it imperative for a cost-effective treatment method that is capable of removing heavy metals from aqueous effluents.

2.1.3 Advantages of Adsorption on biomass

The search for new technologies involving the removal of toxic metals from wastewaters has directed attention to adsorption, based on metal binding capacities of various biological materials. Adsorption can be described as the ability of biological materials to accumulate heavy metals from wastewater through metabolically mediated or physico-chemical pathways of uptake [9].

The biomass of marine alga Sargassum species has demonstrated a good capacity of

heavy metals biosorption, highlighting its potential for effluent treatment processes [27]. Under the term of metal 'adsorption' a passive process of metal uptake and sequestering is understood whereby chemical sites naturally present and functionally sequester the metal even when the biomass is dead. Heavy metals need to be best removed at the source in a specially designed 'pretreatment' step, which has to feature low costs to be feasible. The search is on for efficient and particularly cost-effective remedies. Adsorption promises to fulfill the requirements.

The advantage adsorption is in using raw biomass materials, which are abundant (seaweed). The metal-sorbing performance of certain types of biomass can be more or less selective for heavy metals [14, 24]. It is mainly dependent on several factors, e.g.

- The type of the biomass,
- The metal concentration in the solution,
- The type of biomass preparation,
- The chemico-physical environment,
- The favorable pH,
- The temperature.

It is to note that the concentration of a specific metal could be achieved either during the sorption uptake by manipulating the properties of a biosorbent, or upon desorption during the regeneration cycle of the biosorbent [19]. The main attraction of adsorption is its cost effectiveness. While the ion exchange can be considered the 'mature' technology, adsorption is in its early development stages and further improvements in both performance and costs can be expected [19,20].

Yes, adsorption can become a good weapon in the fight against toxic metals threatening our environment [3]. While adsorption process could be used even at low degree of understanding of its metal-binding mechanisms, better understanding will make for its more effective and optimized applications. That poses a scientific challenge and continued research and developments efforts.

In addition, even the same type of industrial activity can produce effluents, which

differ from each other a great deal. Close collaboration in industrial operations is absolutely essential, e.g. a consulting engineering type approach. Necessary industrial skills become quite important in process operations. Adsorption does offer a competitive wastewater treatment alternative, the basis of which needs to be well understood in order to prevent application failures [7]. In addition to the advantages of adsorption:

- Minimization of chemical and no biological sludge,
- No additional nutrient requirement,
- High efficiency at low metal / radionuclide concentration,
- Operates successfully over a wide range of pH and temperature,
- Calcium and magnesium ions do not compete for binding sites as it happens in ion exchange resins [7],
- Recovery is easy,
- Regeneration of biosorbent is possible,
- Microbial biomass required for biosorption may be available as wastes from fermentation plants, or by growing organisms on cheap substrate,
- Biosorptive processes are less costly than other physico-chemical processes, Biomaterials also are naturally abundant and can be inexpensively produced; their rapid binding rates make it possible to process large volumes of metal-bearing wastes in a timely manner,
- Biosorptive processes may be used as polishing step,
- The high selectivity of various organisms enables the removal and recovery of specific metal ions.

2.2 ADVANTAGES OF ALGAE FOR BIOSORPTION OF METALS.

An inexpensive source of biomass where it is available in copious quantities is in oceans as seaweeds, representing many different types of marine macro-algae. Most of the contributions studying the uptake of toxic metals by live marine and to a lesser extent fresh water algae focused on the toxicological aspects, metal accumulation, and pollution indicators by live, metabolically active biomass. Focus on technological aspects of metal removal by algal has been rare. Strong biosorbent behavior of certain

algal species towards metallic ions is a function of chemical make-up of the microbial cells. In this study, algae was used as a biomass.

Some types of biosorbents would be broad range, binding and collecting the majority of heavy metals with no specific activity, while others are specific for certain metals. Some laboratories have used easily available biomass whereas others have isolated specific strains of microorganisms and some have also processed the existing raw biomass to a certain degree to improve their biosorption properties.

Recent biosorption experiments have focused attention on waste materials, which are by-products or the waste materials from large-scale industrial operations. For example, the waste mycelia available from fermentation processes, olive mill solid residues, activated sludge sewage treatment plants, biosolids and aquatic macrophytes [29, 30, 31, 32].

The mechanism of biosorption is complex and occurs through physical forces, mainly ion exchange, chelation and adsorption. This is due to entrapment of intrafibrilliar capillaries and spaces of structural polysaccharide network which results in concentration gradient and diffusion through cell walls and membranes.

There are several chemical groups that would attract and sequester the metals in biomass: acetamido groups of chitin, structural polysaccharides of fungi, amino and phosphate groups in nucleic acids, amido, amino, sulphhydryl and carboxyl groups in proteins, hydroxyls in polysaccharide and mainly carboxyls and sulphates in polysaccharides of marine algae that belong to the divisions Phaeophyta, Rhodophyta and Chlorophyta. However, it does not necessarily mean that the presence of some functional group guarantees biosorption, perhaps due to steric, conformational or other barriers.

Choice of metal for biosorption: The appropriate selection of metals for biosorption studies is dependent on the angle of interest and the impact of different metals, on the basis of which they would be divide into four major categories:

(i) Toxic heavy metals

(ii) Strategic metals

(iii) Precious metals

(iv) Radio nuclides

In terms of environmental threats, it is mainly categories (i) and (iv) that are of interest for removal from the environment and/ or from point source effluent discharges. Apart from toxicological criteria, the interest in specific metals may also be based on how representative their behavior may be in terms of eventual generalization of results of studying their biosorbent uptake, e.g. copper. The toxicity and interesting solution chemistry of such elements as chromium, arsenic and selenium make them interesting to study. Strategic and precious metals though not environmentally threatening are important from their recovery point of view.

Adsorption Mechanisms: The complex structure of microorganisms implies that there are many ways for metal to be taken up by microbial cell. The adsorption mechanisms are various. They may be classified according to various criteria.

According to the dependence on the cell's metabolism, biosorption mechanisms can be divided into:

1. Metabolism dependent and
2. Non-metabolism dependent.

According to the location where the metal removed from solution is found, biosorption can be classified as:

1. Extra cellular accumulation/ precipitation
2. Cell surface sorption/ precipitation and
3. Intracellular accumulation

Transport of metal across the cell membrane yields intracellular accumulation, which is dependent on the cell's metabolism. This means that this kind of biosorption may

take place only with viable cells. It is often associated with an active defense system of the macroorganism, which reacts in the presence of toxic metal.

During non-metabolism dependent biosorption, metal uptake is by physico-chemical interaction between the metal and the functional groups present on the microbial cell surface. This is based on physical adsorption, ion exchange and chemical sorption, which is not dependent on the cell's metabolism. Cell walls of microbial biomass, mainly composed of polysaccharides, proteins and lipids have abundant metal binding groups such as carboxyl, sulphate, phosphate and amino groups. This type of biosorption, i.e., non-metabolism dependent is relatively rapid and can be reversible [2].

In the case of precipitation, the metal uptake may take place both in the solution and on the cell surface [33]. Further, it may be dependent on the cell's metabolism if, in the presence of toxic metals, the microorganism produces compounds that favour the precipitation process. Precipitation may not be dependent on the cells' metabolism, if it occurs after a chemical interaction between the metal and cell surface [34].

Transport across cell membrane: Heavy metal transport across microbial cell membranes may be mediated by the same mechanism used to convey metabolically important ions such as potassium, magnesium and sodium. The metal transport systems may become confused by the presence of heavy metal ions of the same charge and ionic radius associated with essential ions. This kind of mechanism is not associated with metabolic activity. Basically biosorption by living organisms comprises of two steps. First, a metabolism independent binding where the metals are bound to the cell walls and second, metabolism dependent intracellular uptake, whereby metal ions are transported across the cell membrane [18].

Physical adsorption: In this category, physical adsorption takes place with the help of van der Waals' forces. Researchers have hypothesized that uranium, cadmium, zinc, copper and cobalt biosorption by dead biomasses of algae, fungi and yeasts takes place through electrostatic interactions between the metal ions in solutions and cell

walls of microbial cells [2]. Electrostatic interactions have been demonstrated to be responsible for copper biosorption by bacterium Zoogloe *ramigera* and alga *Chiarella vulgaris*, for chromium biosorption by fungi *Ganoderma lucidium* and *Aspergillus niger* [4].

Ion exchange: Cell walls of microorganisms containing polysaccharides exchange bivalent metal ions with counter ions of the polysaccharides. For example, the alginates of marine algae occur as salts of K^+, Na^+, Ca^{2+} and Mg^{2+}. These ions can exchange with counter ions such as, Cu^{2+}, Cd^{2+} and Zn^{2+} resulting in the biosorptive uptake of heavy metals [2]. The biosorption of copper by fungi *Ganoderma lucidium* and *Aspergillus niger* was also up taken by ion exchange mechanism [34].

Complexation: The metal removal from solution may also take place by complex formation on the cell surface after the interaction between the metal and the active groups. It is hypothesized that biosorption of copper by C. *vulgaris* and Z. *ramigera* takes place through both adsorption and formation of coordination bonds between metals and amino and carboxyl groups of cell wall polysaccharides [2]. Complexation was found to be the only mechanisms responsible for calcium, cadmium, copper, magnesium, zinc and mercury accumulation by *Pseudomonas syringae.* Microorganisms may also produce organic acids (e.g. citric, oxalic, gluonic, fumaric, lactic and malic acid), which may chelate toxic metals resulting in the formation of metallo-organic molecules. These organic acids help in the solubilisation of metal compounds and their leaching from surfaces. Metals may be biosorbed or complexed by carboxyl groups found in microbial polysaccharides and other polymets.

Precipitation: Precipitation may be either dependent on the cellular metabolism or independent of it. In the former case, the metal removal from solution is often associated with active defense system of the microorganisms. They react in the presence of a toxic metal producing compounds, which favour the precipitation process. In the case of precipitation not dependent on the cellular metabolism, it may be a consequence of the chemical interaction between the metal and the cell surface.

The various biosorption mechanisms mentioned above can take place simultaneously [35, 37].

Use of Recombinant bacteria for metal removal: Metal removal by adsorbents from water and wastewater is strongly influenced by physico-chemical parameters such as ionic strength, pH and the concentration of competing organic and inorganic compounds. Recombinant bacteria are being investigated for removing specific metals from contaminated water. The presence of chelating agents Na^{+}, Mg^{2+} and Ca^{2+} does not affect bioaccumulation [39].

Factors affecting Biosorption : The investigation of the efficiency of the metal uptake by the microbial biomass is essential for the industrial application of biosorption, as it gives information about the equilibrium of the process which is necessary for the design of the equipment.

The following factors affect the biosorption process:

1. Temperature seems not to influence the biosorption performances in the range of 20-35 ^{0}C [2].

2. pH seems to be the most important parameter in the biosorptive process: it affects the solution chemistry of the metals, the activity of the functional groups in the biomass and the competition of metallic ions [35].

3. Biomass concentration in solution seems to influence the specific uptake: for lower values of biomass concentrations there is an increase in the specific uptake [9]. It is suggested that an increase in biomass concentration leads to interference between the binding sites [36]. The hypothesis attributing the responsibility of the specific uptake decrease results to metal concentration shortage in solution is invalidated. Hence this factor needs to be taken into consideration in any application of microbial biomass as

biosorbent [9].

4. Biosorption is mainly used to treat wastewater where more than one type of metal ions would be present; the removal of one metal ion may be influenced by the presence of other metal ions. For example: Uranium uptake by biomass of bacteria, fungi and yeasts is not affected by the presence of manganese, cobalt, copper, cadmium, mercury and lead in solution [37]. In contrast, the presence of Fe^{2+} and Zn^{2+} was found to influence uranium uptake by *Rhizopus arrhizus*, and cobalt uptake by different microorganisms seemed to be completely inhibited by the presence of uranium, lead, mercury and copper [37, 38].

Biosorption equilibrium models - Assessment of sorption performance: Examination and preliminary testing of solid-liquid sorption system are based on two types of investigations: (a) equilibrium batch sorption tests and (b) dynamic continuous flow sorption studies.

The equilibrium of the biosorption process is often described by fitting the experimental points with models usually used for the representation of isotherm adsorption equilibrium [36]. The two widely accepted equilibrium adsorption isotherm models for single solute system are given by the following:

$$q = \frac{q_{\max} b C_{eq}}{1 + b C_{eq}} \qquad \textbf{Langmuir} \qquad (1)$$

This is a theoretical model for monolayer adsorption.
Another empirical model for adsorption is:

$$q = K_F C_{eq}^{1/n} \qquad \textbf{Freundlich} \qquad (2)$$

These models at a constant pH are used in literature for modeling of biosorption equilibrium in the presence of one metal. The values are of the specific uptake q are plotted as a function of the metal concentration C_{eq}. But the above said adsorption

isotherms may exhibit an irregular pattern due to the complex nature of both the sorbent material and its varied multiple active sites, as well as the complex solution chemistry of some metallic compounds [45]. Evaluation of equilibrium sorption performance needs to be supplemented by process-oriented studies of its kinetics and eventually by dynamic continuous flow tests.

Biosorption by immobilized cells: Microbial biomass consists of small particles with low density, poor mechanical strength and little rigidity. The immobilization of the biomass in solid structures creates a material with the right size, mechanical strength and rigidity and porosity necessary for metal accumulation. Immobilisation can also yield beads and granules that can be stripped of metals, reactivated and reused in a manner similar to ion exchange resins and activated carbon. Various applications are available for biomass immobilization. The principal techniques that are available in literature for the application of biosorption are based on adsorption on inert supports, on entrapment in polymeric matrix, on covalent bonds in vector compounds, or on cell cross-linking [45, 46].

Adsorption on inert supports: Support materials are introduced prior to sterilization and inoculation with starter culture and are left inside the continuous culture for a period of time, after which a film of microorganisms is apparent on the support surfaces. This technique has been used for the immobilization of *Rhizopus arrhizus* fungal biomass in reticulated foam biomass support particles [39]. Also this technique is used immobilise the bacterium *Citrobacter sp* [40]. Some researchers used activated carbon as a support for *Enterobacter aerogens* biofilm, immobilized *Rhizopus nigricans* on polyurethane foam cubes and coconut fibres [41, 47, 48].

Entrapment in polymeric matrices: The polymers used are calcium alginate, polyacrylamide, and polysulfone. The materials obtained from immobilization in calcium alginate and polyacrylamides are in the form of gel particles. Those obtained from immobilization in polysulfone and polyethyleneimine are the strongest [42, 43, 44].

Covalent bonds to vector compounds: The most common vector compound (carrier) is silica gel. The material obtained is in the form of gel particles. This technique is mainly used for algal immobilization [45, 46].

Cross-linking: The addition of the cross-linker leads to the formation of stable cellular aggregates. This technique was found useful for the immobilization of algae [45]. The most common cross linkers are: formaldehyde, glutaric dialdehyde, divinylsulfone and formaldehyde - urea mixtures.

Desorption: If the biosorption process were to be used as an alternative to the wastewater treatment scheme, the regeneration of the biosorbent may be crucially important for keeping the process costs down and in opening the possibility of recovering the metals extracted from the liquid phase. For this purpose it is desirable to desorb the sorbed metals and to regenerate the biosorbent material for another cycle of application. The desorption process should:

- Yield the metals in a concentrated form,
- Restore the biosorbent to close to the original condition for effective reuse with undiminished metal uptake and
- create no physical changes or damage to the biosorbent.

While the regeneration of the biosorbent may be accomplished by washing the metal-laden biosorbent with an appropriate solution, the type and strength of this solution would depend on the extent of binding of the deposited metal. Dilute solutions of mineral acids like hydrochloric acid, sulphuric acid, acetic acid and nitric acid can be used for metal desorption from the biomass [36].

Polysulphone immobilized *Rhizopus nigricans* were subjected to Cr (VI) recovery experiments using 0.01 N solutions of mineral acids, salt solutions, alkalis, deionised distilled water and buffer solutions. A few experiments were conducted to desorb the metal ions from the loaded waste fungal biomass of *Aspergillus species* as a function of HCl concentration in the case of iron, calcium and nickel. The results revealed that with increase in HCl concentrations, the desorption of the metal ions increased and at 5M HCI, complete removal of calcium and iron would be achieved while about 78% Nickel would be desorbed [49].

The desorption of the adsorbed Hg (II) from the biosorbent - immobilized and heat inactivated *Trametes versicolor* and *Pleurotus sajur-caju* were studied in a batch system [50]. Hg (II) ions adsorbed onto the biosorbents were eluted with 10 mmol dm^{-3} HCl and the results showed that more than 97% of the adsorbed Hg (II) ions were desorbed from the biosorbents.

Effect of Pre-treatment on the biosorption of heavy metals: Metal affinity to the biomass can be manipulated by pretreating the biomass with alkalies, acids, detergents and heat, which may increase the amount of the metal sorbed. The bioadsorption capacity of autoclaved *Mucor rouxii* decreased as compared to the live fungus, attributed to the loss of intracellular uptake [51]. It is reported that the heat treatment could cause a loss of amino-functional groups on the fungal surface through the non-enzymic browning reaction. Amino functional groups in the polysaccharides contribute to the binding of heavy metals [52, 53]. However, *Pencillium* biomass pretreatment at 100°C for 5 minutes increased the bioadsorption of lead, cadmium, nickel and zinc and the increase was attributed to the exposure of latent binding sites after pre-treatment [54]. In the case of alkali pre-treatment, bioadsorption capacity of *Mucor rouxii* biomass was significantly enhanced in comparison with autoclaving [54]. However, NaOH treated *Pencillium digitatum* also showed enhancement of cadmium, nickel and zinc biosorption [54]. Removal of surface impurities, rupture of cell-membrane and exposure of available binding sites for metal bioadsorption after pre-treatment may be the reason for the increase in metal bioadsorption [55]. However, studies showed that alkali treatment of biomass might destroy autolytic enzymes that cause putrefaction of biomass and remove lipids and proteins that mask reactive sites [55, 56, 57]. The cell wall of *Mucor rouxii* was ruptured by NaOH treatment. Besides, the pre-treatment could release polymers such as polysaccharides that have a high affinity towards certain metal ions [53, 58].

Acid pretreatment of *Mucor rouxii* significantly decreased the bioadsorption of heavy metals, which is in agreement with the observation found in the case of *A.niger* and that was attributed to the binding of H^+ ions to the biomass after acid treatment which was responsible for the reduction in adsorption of heavy metals [51, 59]. The

polymeric structure of biomass surface exhibits a negative charge due to the ionization of organic and inorganic groups [60]. However, it is suggested that the higher the biomass electronegativity, the greater the attraction and adsorption of heavy metal cations [60]. Thus the remaining H^+ ions on the acidic pretreated *M.rouxii* biomass may change the biomass electronegativity, resulting in a reduction in bioadsorption capacity.

However, studies reported that acid pretreatment could strongly enhance the adsorption capacity of *Aspergillus.oryzae* mycelia [61]. In case of *A.oryzae,* live biomass after acid pre-treatment was directly used in bioadsorption of heavy metals instead of being autoclaved and dried. The difference in results after a specific pretreatment may be attributed to the different strains of fungi used and whether the biomass was live or dead when it is used in biosorption of metal ions. For example, pre-treatment of *A.oryzae* by $HClO_4$ increased the bioadsorption of lead, cadmium and nickel, but it was not the case for the species of *R.oryzae* [61, 62]. When non-viable biomass is used in the removal of heavy metals, alkali pretreatment is an effective method to improve the bioadsorption capacity for metal ions [51]. Hence, the bioadsorption efficiency of dead biomass may be greater, equivalent to, or less than that of live biomass depending on the pre-treatment method applied. It is necessary to carry out more detailed studies to understand why enhancement or reduction in adsorption capacity occurs under specific pre-treatment conditions.

2.3. ECOLOGICAL EFFECT OF HEAVY METALS

Over the past two decades, there has been an increasing awareness of the ecological effects of toxic heavy metals. This has been accompanied by a number of studies on the subject. Unlike organic pollutants, which can eventually be destroyed, metallic species released into the environment tend to persist indefinitely. They accumulate throughout the food chain, thus posing a serious threat to man and animals [3, 9].

Large portions of heavy metals species are released into the environment from industrial wastewater through inefficiencies built into the technological activities used

directly in the processing of metals or through other routes. Almost every industrial activity using metals has a metal disposal problem. There are also many technology-related activities, which result in the release of metallic species. An example of such a large-scale metal release of metallic species includes metal plating industries and battery manufactures. The presence of heavy metals in wastewater has many detrimental ecological effects. Concentrations in excess of 1 mg/l have been known to affect the fish life of the river. The effect of the discharge is dependent, however, on the factors such as dissolved oxygen, hardness and temperature [5].

2.4 THEORY OF ADSORPTION

Recently, several researchers have independently concluded that the major mechanism of heavy metal uptake by algae is ion exchange [8, 9]. Furthermore, it has been demonstrated that algal biosorbents, similar to ion exchange resins, can be prepared in different ion forms such as H-form and Ca-form [12]. Consequently, ion exchange models have been introduced to fit and interpret the data obtained from both equilibrium and dynamic biosorption experiments [11, 13]. A binary ion exchange system containing divalent metal ions A and B may be described by the following exchange reaction:

$$A^{2+} + (B - biomass) \Leftrightarrow B^{2+} + (A - biomass) \tag{3}$$

The corresponding equilibrium constant is defined as

$$K_{AB} = \frac{q_A C_B}{q_B C_A} \tag{4}$$

The overall binding capacity Q is given by the density of the functional groups in the sorbent and can be expressed as:

$$Q = q_A + q_B \tag{5}$$

The total concentration of the solution is given by:

$$C_{AB} = C_A + C_B \tag{6}$$

By substituting equation (5) into (4), q_B can be eliminated from (4) and the following expression for (q_A/Q) can be obtained:

$$\frac{q_A}{Q} = \frac{1}{1 + \frac{C_B}{K_{AB} C_A}} \tag{7}$$

Since (q_A/Q) represents the fraction of the binding sites occupied by A, equation (7) may be used to evaluate the decrease of the equilibrium uptake of the species A by the biosorbent due to the presence of the species B. Equation (7) shows that when $C_B = 0$, (q_A /Q) ~ 1, regardless of the absolute value of the final concentration of A, C_A. This distinguishes ion exchange from chemisorption and/or physical sorption known to occur on activated carbon whereby (q_A/Q) is a hyperbolic function of C_B. Equation (7) may be transcribed using the following dimensionless variables:

$$x_A = \frac{C_A}{C_0}; \quad x_B = \frac{C_B}{C_0}; \quad y_A = \frac{q_A}{Q} \tag{8}$$

yielding equation (9) which represents the binary ion exchange isotherm for the system:

$$y_A = \frac{1}{1 + \frac{x_B}{K_{AB} x_A}} \tag{9}$$

Since $y_A, y_B,$ and K_{AB} are all dimensionless, equation (9) represents the most generalized description of the equilibrium for binary systems.

2.5 SELECTION OF THE BIOMASS

The effectiveness of a biomass to adsorb heavy metals is related to the chemical make-up of the biomass [8, 9, 10].

There are various types of biosorbents as well as sources. Since cost is the huge factor when dealing with wastewater, the cheaper the source of the biosorbent the better. An inexpensive source of biomass is the one produced naturally [10, 12]. This variety of biomass was chosen as it fulfils the following criteria:

- The uptake and the release of the metal should be both rapid and efficient i.e. the metal should be easily adsorbed and desorbed.
- The biosorbent should be also obtained at a low cost and should preferably be reusable so that the process becomes as cost effective as possible.
- The particle size, shape and mechanical properties of the biosorbent should be suitable to use in a continuous flow system in freely mixed, packed or fluidized bed configurations.
- The seaweed should be metal selective in order to separate target metals from the solution.

2.6 Evaluation of Adsorption Performance

2.6.1 Sorption Equilibrium

The case of a sorption process considered here involves a solid phase (sorbent) and a liquid phase (solvent, normally water) containing a dissolved species to be sorbed (sorbate, e.g. metal ions). Due to the higher 'affinity' of the sorbent for the sorbate species the latter is attracted into the solid and bound there by different mechanisms. This process takes place until equilibrium is established between the amount of solid-bound sorbate species and its portion remaining in solution (at a residual, final or equilibrium concentration C_f). The degree of the sorbent 'affinity' for the sorbate determines its distribution between the solid and liquid phases. The quality of the sorbent material is judged according to how much sorbate it can attract and retain in an 'immobilized' form. For this purpose, it is customary to determine the metal uptake ***(q)*** by the biosorbent as the amount of the sorbate bound by the unit of solid

phase (weight, volume, etc.) [12, 15, 19].

2.6.2 Comparison of sorption performance

Performance of sorbing materials often needs to be compared. The simplest case is only one sorbate species in the system. The comparison of single-sorbate sorption performance is the best based on a complete single-sorbate sorption isotherm curve [19]. In order for the comparison of two or more sorbents to be clearly understood, comparison must always be done under the same conditions. These may be restricted by the environmental factors under which sorption may have to take place (pH, temperature, ionic strength, etc). They may not necessarily be widely adjustable. It is important to compare sorption performance e.g. under the same pH since isotherms could vary with pH [19, 23].

By 'performance' of the sorbent is usually meant its uptake $\boldsymbol{q}$. The sorbents can be compared by their respective q_{MAX} sorption performance plateau (the maximum sorbent saturation). Some isotherms might not exhibit it in an asymptotic manner, which is easily represented by the Langmuir equation of hyperbole. Obviously, in general, one is looking for a sorbent with a high sorption uptake capacity [20, 21].

2.6.3 Effect of temperature

Temperature was not studied as an important parameter in our biosorption experiments. The tests were performed at approximately 25-30^{0}C. However, a slight increase in cation uptake by seaweed in the range of 40 to 55^{0}C was reported. [3, 4, 9].

CHAPTER 3

EXPERIMENTAL

3.1 PROPERTIES OF SARGASSUM

Sargassum, seaweed biomass was collected during low tide on the North Coast of Kwa-Zulu Natal (Umdhloti Beach). After the seaweed biomass was collected a pre-treatment stage was applied as described in section 3.2.

Systematic Position

Class: Phaeophyceae (Brown Algae)
Order: Fucales
Family: Sargassaceae
Genus: Sargassum

Occurrence

Sargassum, with its about fifty species, is common in the warmer seas of Southern Hemisphere [16]

Structure

The plant body of Sargassum looks like an angiospermic plant. It can be studied as follows.

External Morphology

The main body of Sargassum is diploid or a Sporophyte and is distinguishable into a hold fast, main axis and primary and secondary laterals.

3.2 SAMPLE PREPARATION

After the seaweed biomass was collected it was sun-dried for two days. The biosorbent was prepared by washing it with 0.1N HCl and then rinsing it with distilled water. The sargassum was then dried in an oven for the duration of 24 hours at a temperature of 60^0C. The algae were then put in a grinder, to prepare for different sizes of biomass. In order to separate the different sizes, the load was placed in a shaker and collected through differently sized sieve trays.

3.3 SORPTION DYNAMICS EXPERIMENTS

In order to determine the contact time required for the sorption equilibrium experiments, the sorption dynamics experiments were conducted first. 0.1g of biomass was mixed with 50ml of different concentrations of Cu as $CuSO_4.5H_2O$ solution in a 100 ml Erlenmeyer flasks on a rotary shaker at 3Hz and room temperature. The pH value of the solution was controlled. The 0.1N HCl or 0.1N NaOH solution was added to maintain the pH value of the reacting solution at the level of the designed end-point.

The metal concentration were analysed before and after the experiment using Atomic Absorption Spectrophotometer (AAS). Figure 1 represents the schematic diagram for the experimental procedure. It took 3 hours to attain sorption equilibrium. A simple preliminary sorption kinetics test established the exposure time necessary for the given sorbent particles to reach the equilibrium state characterized by the unchanging sorbate concentration in the solution. This was determined by time-based analysis. Enough time was allowed for the sorption system to reach equilibrium.

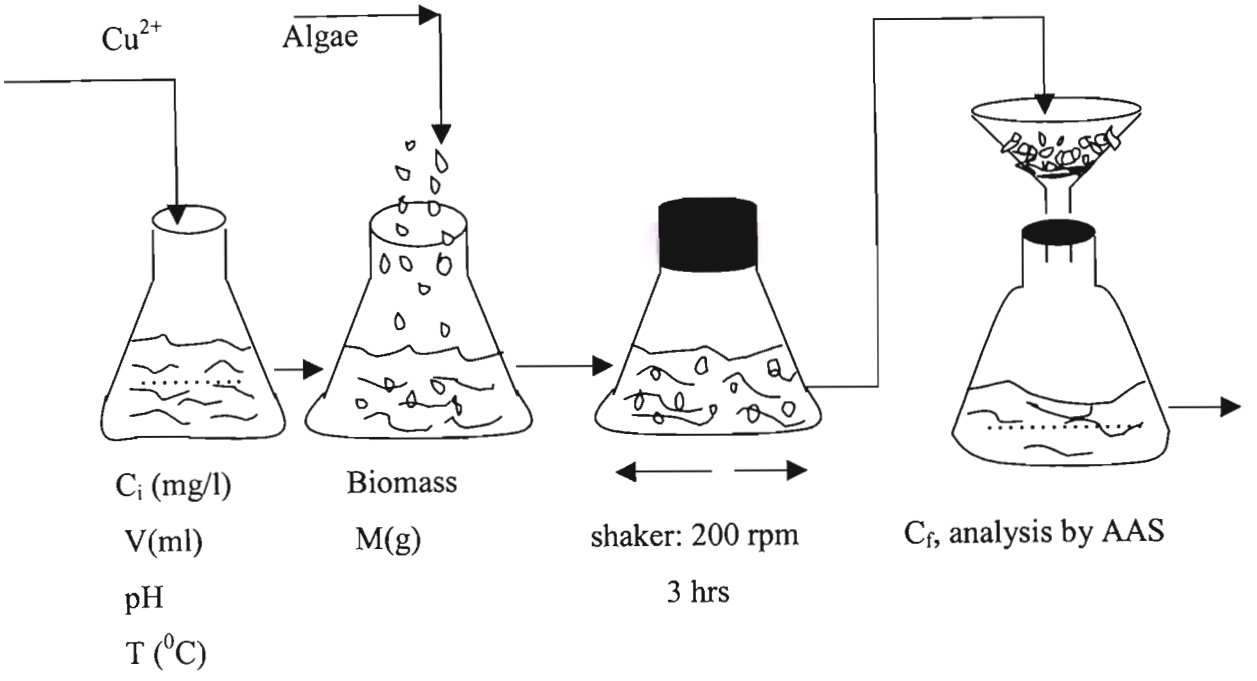

Figure 1: Outline of an experimental procedure for determining the equilibrium data.

3.4 SORPTION AND DESORPTION COLUMN EXPERIMENTS

Separation processes based on what is widely labeled as adsorption, followed by desorption are widely utilized [25]. Due to its inherent effectiveness in adsorption processes the packed-bed reactor is generally preferred [25, 26]. Its advantage is the highest possible packing density of the sorbent, yielding a high volumetric productivity. The performance of packed-bed adsorbers is analyzed using the effluent concentration versus time curves. For adsorption the plot is usually referred to as the breakthrough curve, and for desorption it is the elution curve. Both curves are a function of the column flow parameters, sorption equilibrium and mass transport factors [27, 28].

The column experiments were performed in a packed bed column of inner diameter 30 mm and length of 500 mm (Figure 2) uniformly packed with 38 g (dry basis) of acid treated biomass (see section 3.2). During the column sorption operation, an aqueous solution containing 35 mg/l copper (from $CuSO_4.5H_2O$), at pH 4 was pumped through the column at a constant flowrate (15 ml/min) continuously. At pH 4

it was shown (section 4.1) that the rate of rate of adsorption was high. Experiments were conducted at different pH as shown in figure 3. The work has been done and proven that at pH 5 there was no Cu adsorption and a Cu precipitate was observed. The samples were collected from the outlet of the column at different time interval and analyzed for the copper effluent concentration by atomic absorption spectrophotometer. After the biomass in the column became saturated, the column was washed at the same flowrate by distilled water for several hours, before a subsequent copper elution with 0.1N HCl acid. The reason for washing the column for several hours was to ensure that all solution with Cu ions were removed. This forms first step of biomass regeneration before elution step. The outlet sample collection and analysis was the same as that used in the biosorption uptake run.

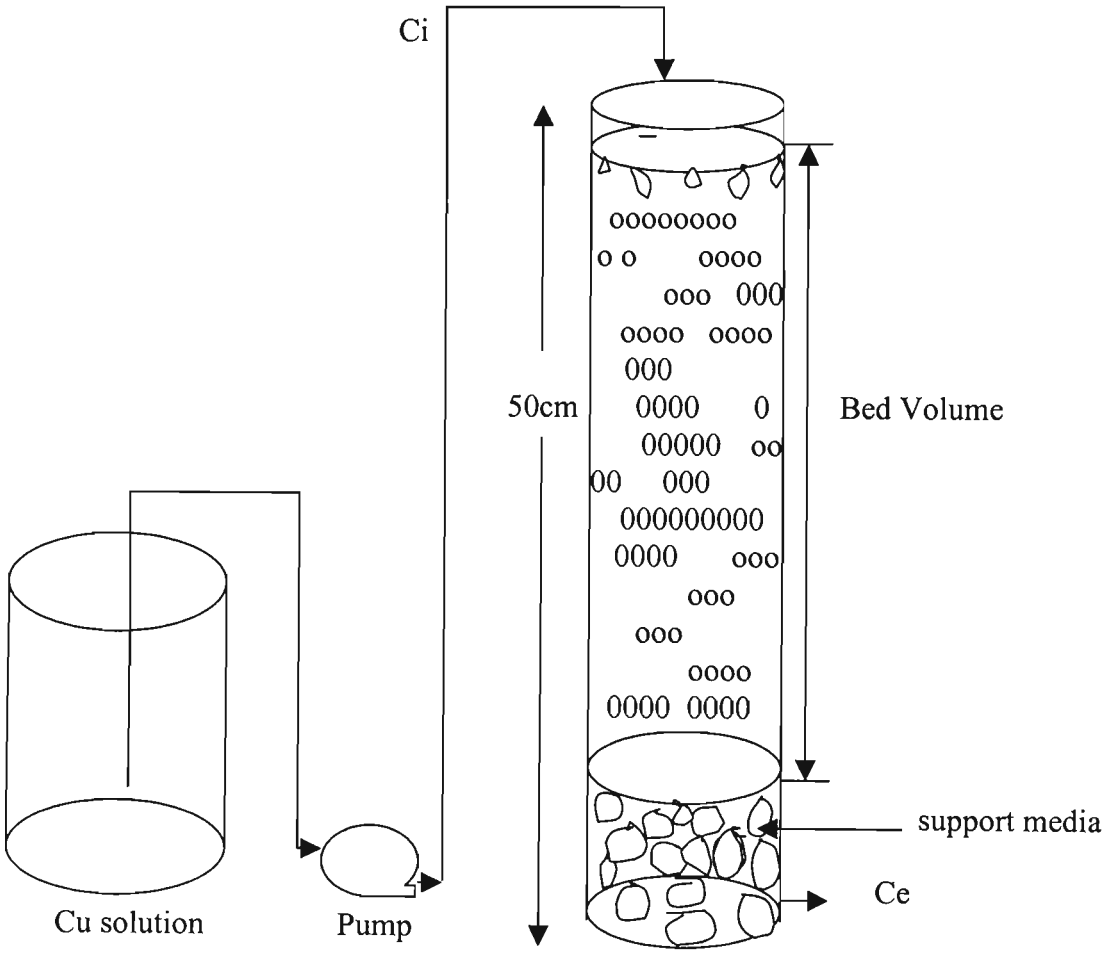

Figure 2: Sorption and desorption experiments

3.5 PROCEDURE FOR OBTAINING THE EXPERIMENTAL ADSORPTION ISOTHERM

1. Sorbate in solution at the highest concentration of interest was prepared.
2. Dilutions to cover the entire concentration range (from 0 –blank to the maximum) were made.
3. Parameters such as pH and ionic strength were adjusted.
4. The sorbate initial concentrations (***Ci***) in all liquid samples were determined.
5. The amount of the (bio) sorbent solids (S) to be used were accurately weighed for each contact test and recorded. It is helpful to be able to roughly estimate the anticipated sorption uptake so that there is a well detectable sorbate final concentration left in the solution at equilibrium in each sample. If there is too much solids added there may be virtually no sorbate left in the solution for a reliable analysis [22].
6. The sorbent solids were added into each sample solution and gentle mixing was provided over the sufficiently long contact period.
7. Parameter such as pH was controlled at a constant value during the contact period, using appropriate acid. In order to do that, the sorption system was not diluted by adding excessive volume.
8. At the end of the contact period, solids from liquids were separated by decantation or filtration.
9. The liquid portion was analyzed for the residual, final sorbate concentration (C_f).
10. The sorbate uptake was calculated as:

$$q = V(C_i - C_f)/S \tag{8}$$

11. The sorption isotherm (q vs. C_f) was plotted. In the above procedure, it was the initial sorbate concentration C_i that was varied

CHAPTER 4

RESULTS AND DISCUSSION

4.1 SORPTION DYNAMICS AND ISOTHERMS AT DIFFERENT pH VALUES IN A BATCH SYSTEM.

In order to determine the minimum contact time for the equilibrium experiments, the sorption dynamics was examined first. Figure 3 shows the profiles of dimensionless copper concentration against time.

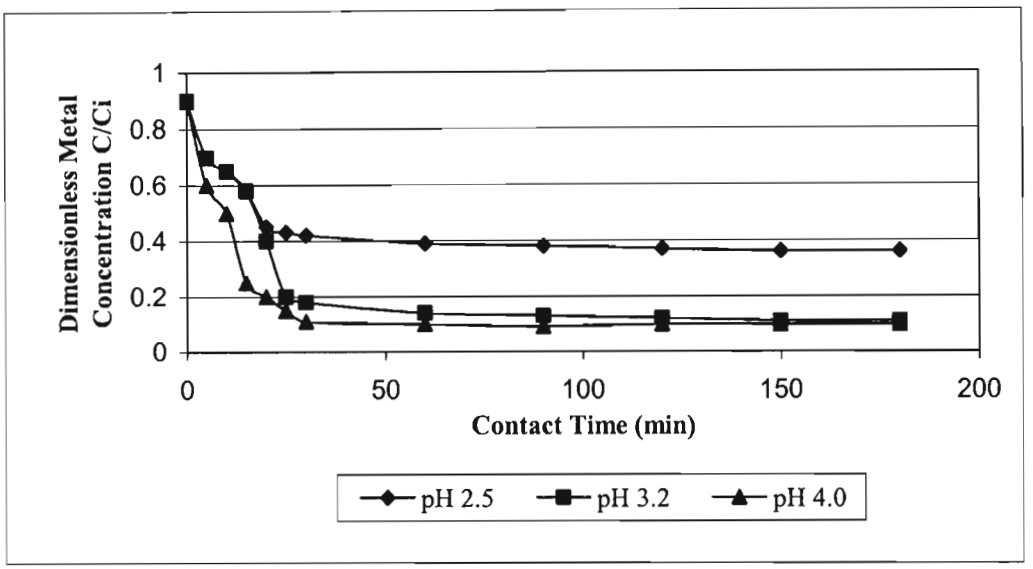

Fig. 3: Dimensionless copper concentration against time for determination of the minimum contact time for equilibrium experiments at different pH values.

The copper biosorption rate was strongly influenced by the sorption system pH value, the copper solution concentration in the solution decreased with contact time faster at higher pH values. At various pH values, approximately 60-70% of the copper present originally in the solution was sorbed onto biomass in about 25 minutes after the start of biosorption and the equilibrium could be reached within 3 hours. This provided a guide for the biosorption contact time to be used in the equilibrium experiments.

Same conditions were also applied to determine the contact time at pH 4 and it was observed that 3 hours is sufficient for the system to reach equilibrium (Figure 4). Best adsorption was observed at pH 4 as illustrated in figure 4 by approximately 75% adsorption rate.

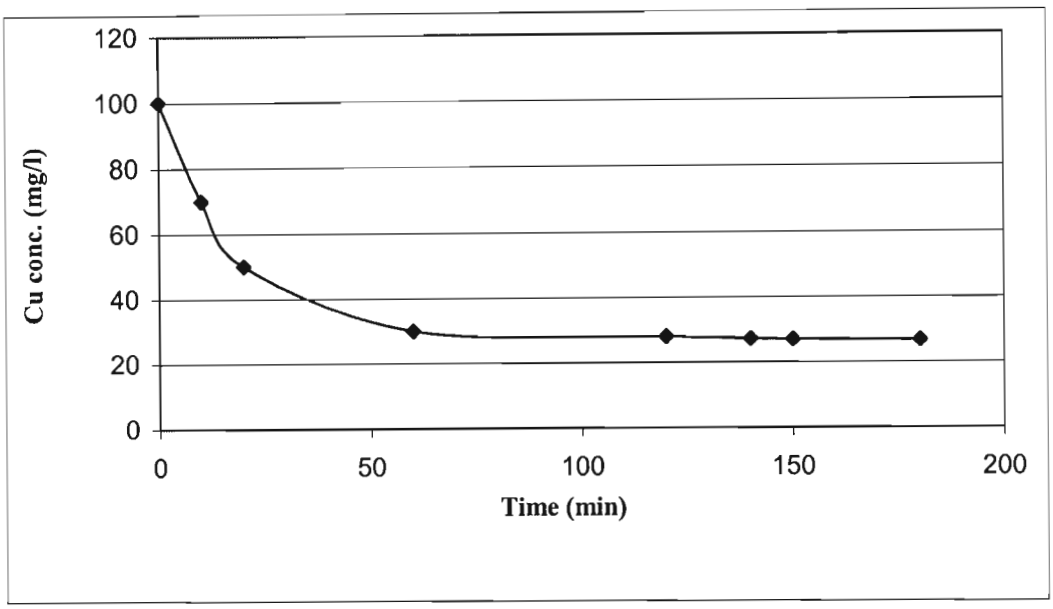

Figure 4:Copper concentration against time (batch studies).

COPPER BIOSORPTION KINETICS

The purpose of these experiments was to determine the contact time required to reach the equilibrium between dissolved and solid-bound sorbate. Equilibrium time is a function of many factors, such as type of biomass (number and kind of biosorption sites), size and form of biomass, physiological state of biomass (active or inactive, free or immobilized), as well as the metal involved in the biosorption system. Reported values for equilibrium time are in the range from 15 minutes [4]

Figure 5 presents the results at two different initial concentrations of 92 and 162 mg/l of copper with a contact time of 3 hours was enough for the system to reach equilibrium. So, this time was used to obtain the isotherms in all experiments. Figure 5 clearly indicates that sorption can be divided into two stages: one in which the

sorption rate is very high (60% of biomass saturation capacity in a contact time of 25 minutes), followed by a second stage with a much lower sorption rate. This behaviour has often been reported by other researches [14], observed that proton uptake by algal cells consists of two processes, a fast surface reaction and a slow diffusion of protons into the cells. The fast biosorption kinetics observed is typical for biosorption of metals involving no energy-mediated reactions, where metal removal from solution is due to purely physico-chemical interactions between biomass and metal solution [64].

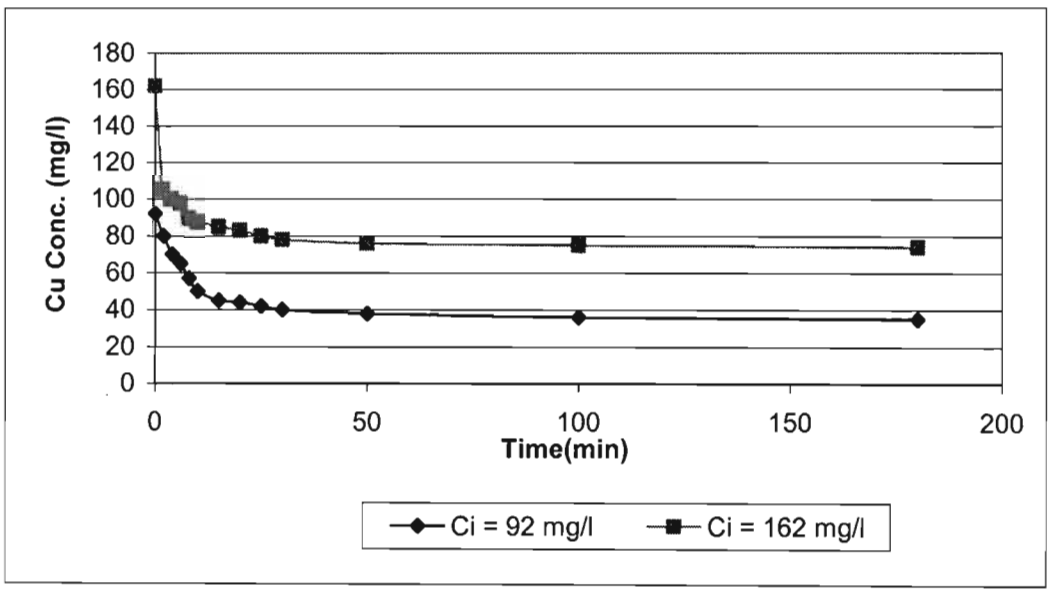

Figure 5: Copper biosorption kinetics at two different initial concentrations.

4.2 INFLUENCE OF BIOSORBENT SIZE ON COPPER BIOSORPTION

The influence of biosorbent size on copper biosorption can be evaluated from figure 6. The experimental results indicate that the biosorbent size did not influence the capacity and rate of copper biosorption.

Although this is contrary to expected for an intraparticle diffusion controlled process, it is necessary to point out that the two sizes of biomass are actually of the same thickness (dimension which determines the diffusion distance). This is so because size grading of ground biomass particle by standards sieves works on the length and width dimensions and the particle shape is flake.

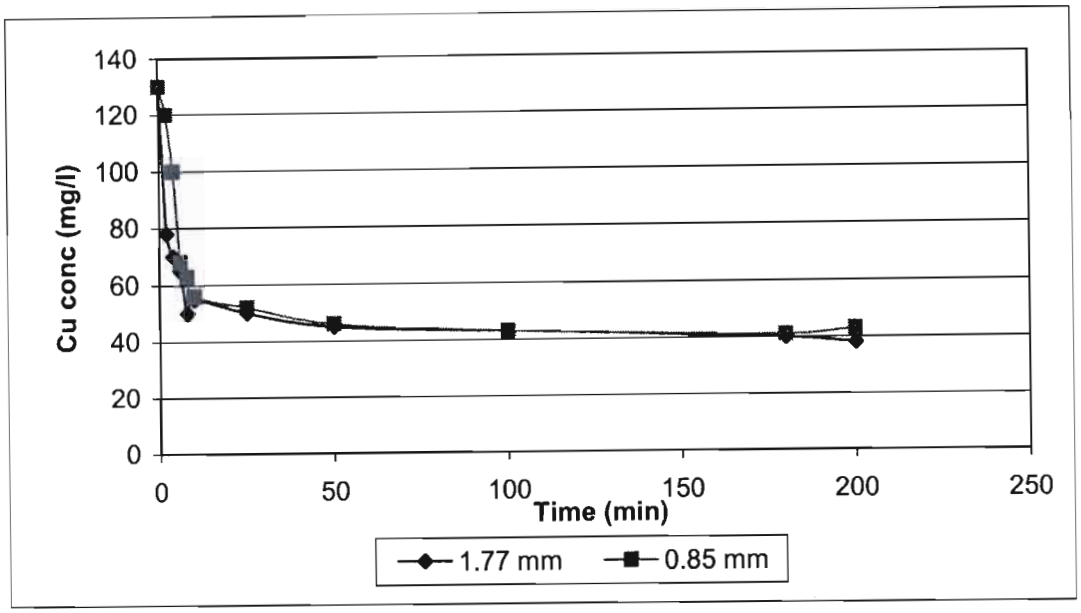

Figure 6: Influence of biosorbent size on copper biosorption by *Sargassum* species.

This behaviour has been reported by other researchers, although it has been showed that larger biomass particles of *Sargassum fluitans* and *Ascophylum nodosum* had higher metal uptake than smaller particles in the case of Cadmium, nickel, lead and zinc [2, 27].

4.3 Effect of pH

It is now well established that heavy metals are taken up from water predominantly by ion exchange [27]. Carboxyl and sulphate groups have been identified as the main metal-sequestering sites in seaweed and, as these groups are acids, its availability is pH dependent [15]. At pH in the range 3.5-5.0 these groups generate a negatively charged surface, and electrostatic interactions between cationic species and this surface is responsible for metal uptake [19]. As the pH increased, the ligands such as carboxylate groups in *Sargassum* sp. would be exposed, increasing the negative charge density on the biomass surface, increasing the attraction of metallic ions with positive charge and allowing the biosorption onto the cell surface.

Figure 7 shows the effect of pH on the biosorption capacity of the marine algae *Sargassum* species. It is shown that as the pH increases, the rate of adsorption also increases with the optimum pH of 4 for copper biosorption. Copper precipitated at pH higher than 4 and no adsorption was observed. The effect of pH on metal biosorption has been studied by many researches, and the results demonstrated an increase in cation uptake with increasing pH values, both on fungi and algae biomass [8, 10, 19]. At pH values lower than 2.5, copper (II) removal was inhibited, possibly as a result of the competition between hydrogen and copper ions on the sorption sites, with an apparent preponderance of hydrogen ions, which restricts the approach of metal cations as in consequence of the repulsive force [63]

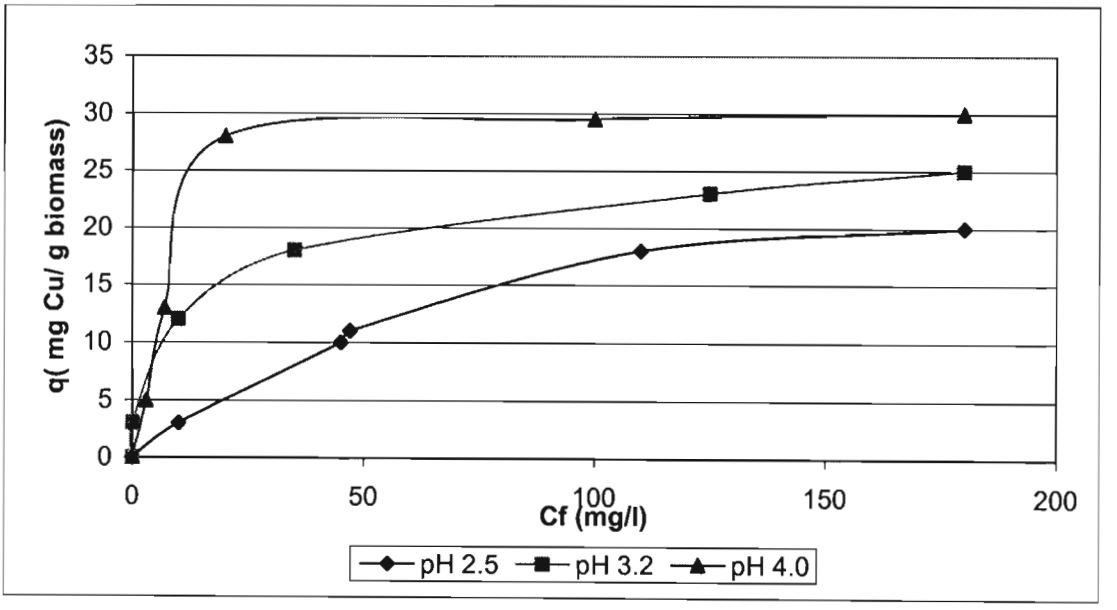

Figure 7: Effect of pH on the biosorption capacity of marine algae *Sargassum* species at pH 2.5, 3.2 and 4.0

4.4 MULTIPLE SORPTION-DESORPTION CYCLES

After it was established that biomass has a characteristic of adsorbing copper, the activeness of biomass in a multiple sorption-desorption was tested. The main reason was to see if the biomass is regenarable or it must be used once. The multiple sorption-desorption cycles were performed in a continuous flow biosorption process

with the aim to determine:

- the efficient utilization of biomass,
- checking its performance,
- change in mechanical properties,
- efficiency of biosorption and elution,
- biomass damage and stability over a prolonged operation time

Figure 8 shows six sorption and desorption cycles, which were carried out with the column packed with raw *Sargassum* biomass. The packed bed contained seaweed fragments of approximately 1.77mm diameter and bladders, leaves and complete branches with a length of up to 4 cm. A copper bearing feed solution of 35 mg/l Cu with pH 4 at a flow rate of 15 mL/min was passed through the column. The sorption process was stopped after reaching 35 mg Cu/l in the effluent and the regeneration solution of 0.1N HCl was pumped through the bed. Table 1 shows the results of the breakthrough (t_b) exhaustion time (t_e) for each cycle.

Table 1: Breakthrough parameters for six sorption-desorption cycles

Breakthrough number	t_b (hr)	t_e (hr)	Time of elution (hr)
1	25	90	15
2	23	80.5	15
3	21	70	15
4	18	85	15
5	17	95	15
6	10	95	15

The breakthrough point is the time (t_b) when the sorbate appeared in the effluent stream at some predetermined concentration, which was 1 mg Cu/l. The time t_e is the time when the whole column sorption bed became totally saturated by the sorbate at its inflow concentration and the bed was no longer effective and was 35mg Cu/l corresponding to the initial concentration. The time interval between t_b and t_e corresponds to the length of the mass-transfer zone of the bed as shown in figure 8.

To determine the weight loss after the 6th cycle, the biomass was washed with distilled water and dried in the oven at 45^{0}C overnight. From the initial 38 g of raw *Sargassum*, 29 g remained, with a weight loss of 23.7%. Process conditions used in this study included use of raw biomass, analytical reagents and distilled/deionised water. While a shortening breakthrough time from cycle to cycle was observed, the uptake capacity exhibited no decreasing trend.

The reason for shortening breakthrough time was apparently not the diminishing equilibrium uptake capacity, but rather a slight change in the column overall adsorption rate. This means that while adsorbing sites were still available, they became less accessible. A loss of sorption performance during the long-term use may have a variety of reasons. It may be caused by changes of the chemistry and of the structure of the biosorbent, as well as by changes of the flow within the column. Deteriorating sorption properties may be due to chemical changes of the cell wall components such as alginate and sulphated polysaccharides, which play a major role in adsorption by marine algae [25, 28].

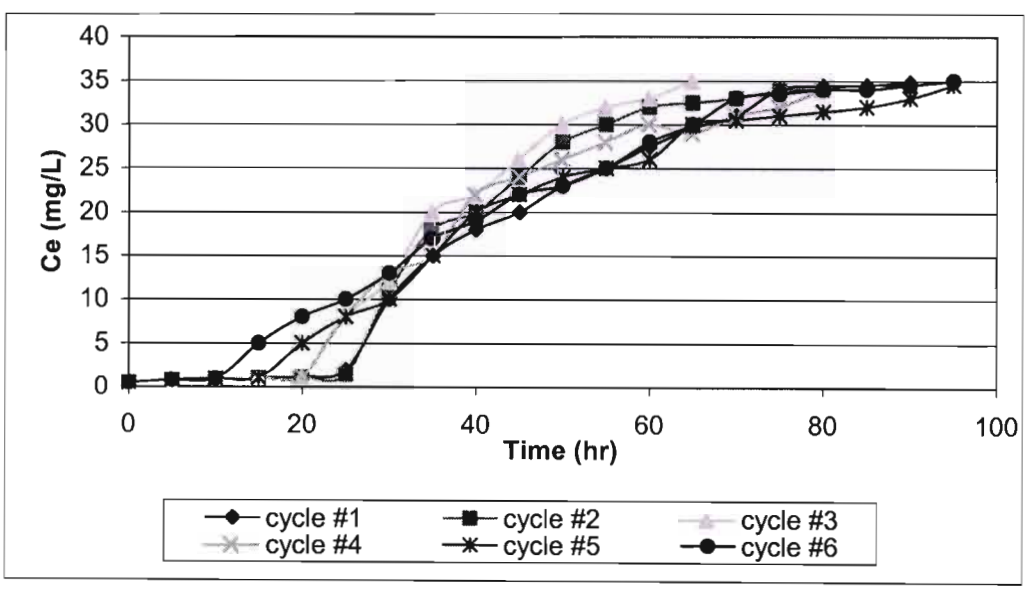

Figure 8: Breakthrough curves for all six biosorption cycles

CHAPTER 5

CONCLUSION

- The biomass of the marine algae *Sargassum* species demonstrated a good capacity of copper biosorption, highlighting its potential for effluent treatment processes. This biosorbent is widely available and easy to find.
- The kinetics of copper biosorption by inactive biomass of marine algae *Sargassum* species was fast, reaching 60% of the total biosorption capacity in twenty-five minutes.
- The biosorbent size had no influence on copper biosorption rate for flake-shaped particles.
- pH had a strong effect on copper biosorption capacity. The capacity of copper biosorption by biomass increased with pH up to pH 4.0.
- From the sorption-desorption experiments, a weight reduction of 24% of the biomass was observed after the 6^{th} cycle.
- The length of the packed bed after the 6^{th} desorption decreased from 38 to 35 cm.
- The high efficiency of biosorption and elution, low biomass damage and stability over a prolonged operation make a new biosorption process an effective alternative for copper pollution control.
- Therefore, marine algae can be used to treat effluents with heavy metals, with an advantage that it can be regenerated and widely available along the coast of seashores.

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