Plant-derived biosorbents for metal removal

M.Sc. research thesis by

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I hereby certify that this material, which I now submit for assessment on the programme of study leading to the award of M.Sc. is entirely my own work and has not been taken from the work of others save and to the extent that such work has been cited and acknowledged within the text of my own work.

Signed:.. ID No.: 97970794

Date: 23/9/99

Dedicated to my mother, Lylah Dean

Ye nymphs that reign o'er sewers and sinks,
The river Rhine, it is well known,
Doth wash your city of Cologne,
But tell me, nymphs, what power divine
Shall henceforth wash the river Rhine?

Samuel Taylor Coleridge
(1772-1834)

ABSTRACT

Plant – derived biosorbents for metal removal

The biosorption capacities of milled peat and a marine biomass, *Laminaria digitata* (a species of kelp), were investigated for removal of metal ions from solution and for the treatment of industrial wastewaters Because industrial solutions generally contain a mixture of more than one component, mixed metal solutions were also investigated In particular, adsorption of cationic Cr(III) and anionic Cr(VI) was studied as these two species often occur together in wastewater

Peat and kelp adsorbed chromium, copper, cadmium, calcium, magnesium and strontium from aqueous solution between pH2 and pH7 Biosorption by acid- and calcium-treated *L digitata* and peat is also reported Kelp was chosen as an inexpensive and readily available biomass as it is common in shallow coastal waters in the northwestern Atlantic Its biosorption characteristics are compared to peat which is recognised for its biosorptive properties *L digitata* was consequently used to treat tannery effluent

Untreated kelp outperformed peat and pre-treated kelp as a biosorbent Uptake of Cr(III) by *L digitata* at pH4 was significantly higher (1 12mmol/g biomass dry weight) than with other biomass as reported in the literature High uptake levels were also seen for Cr(VI) and copper Kelp can reduce Cr(VI) to Cr(III) at low pH These findings outline the suitability of *L digitata* as a biosorbent for industrial chromium solutions

Acknowledgements

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Thanks

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Sonia A Dean and John M Tobin (1999) Uptake of chromium cations and anions by milled peat *Resources, Conservation and Recycling* 27, 151-156

Sonia A Dean and John M Tobin (1999) Biosorption of metal ions from tanning wastewater by *Laminaria digitata* In preparation

CHAPTER 1

INTRODUCTION

CHAPTER 1: INTRODUCTION

1 01 Charged species in metal pollution

In aqueous systems, both cations and anions play an integral part in chemical and physical reactions In industrial wastewater streams, metal anions, as well as the more publicised cations, such as Hg, Pb, and Cd, pose the threat of environmental contamination Often, the anion is an essential nutrient in the ecosystem (Volesky, 1989) and only becomes hazardous at higher concentrations For example, in biological systems compounds of molybdenum have been shown to be essential, both in nitrogen fixation and, in conjunction with the enzyme, nitrate reductase (Sakaguchi *et al*, 1981) Chromium, in the trivalent state (Cr(III)), is recognised as essential to mammals for the maintenance of an effective glucose, lipid, and protein metabolism (Huheey *et al*, 1993) but in the anionic hexavalent state (Cr(VI), chromate), it can diffuse through cell membranes resulting in damage to the cell Cr(III) may be oxidised (Bartlett and Kimble, 1976) and then bound to other biological molecules with toxic results If an anion is not native to the environment however, it can be toxic at low concentrations It is not entirely possible to equate anion-exchangeable chromium with Cr(VI) species or cation-exchangeable chromium with Cr(III), since chromium can exist in various forms of different charge in solution and (Huheey *et al*, 1993) for example, $Cr(OH)_4$ is an anionic form of Cr(III)

1 02 Sources of Metal Pollution

Anions of molybdenum, antimony and vanadium occur along with nickel and manganese in wastewaters from the manufacturing of durable metal goods for household use (Wase and Forster, 1997) The manufacture of paints and varnishes is based on the use of pigments, which generally consist of metal oxides, sulphides and chromates These pigments are subjected to washings and rinsings in the manufacturing process resulting in high concentrations of metal compounds in the wastewater (Volesky, 1989) Chromium compounds, generally in the form of chromate, are found with cadmium, copper, zinc and lead in fertilisers and lubricants (Cary, 1982) Selenium, which forms anionic selenates, is used extensively as an additive in poultry and animal feeds Most of the arsenic used each year goes into organic pesticides, as arsenates, that are applied on agricultural land where the potential for seepage and leaching to groundwater is high (Onken and Hossner, 1996) Platinum also forms anions which are found in dilute concentrations in effluents resulting from catalysis processes (Guibal *et al*, 1998)

Waste produced by the plating and metal finishing industries arises from spent plating-bath solutions, acid and alkaline cleanings and rinse waters Approximately 90% of this effluent is rinse water, used to remove drag-outs from the metal surfaces Metals involved in electroplating include cadmium, chromium, zinc, nickel, lead, copper, tin and iron Various chromium compounds are corrosive to flesh and carcinogenic (Ajmal *et al*, 1996) The acid waste stream generally contains chromium, copper, and nickel from plating This discharge is usually mixed with alkaline effluent and lime is added to precipitate the metals as hydroxides for removal as sludge (Schoeman *et al*, 1992)

Metals are found in the atmosphere as a result of emissions from the burning of fossil fuels (Mahuli *et al*, 1997) Power plants and domestic, commercial and industrial burners are the main sources of atmospheric chromate, arsenate, thallium, antimony and manganese (Nriagu and Pacyna, 1988) Oil combustion is one of the largest sources of vanadium and nickel Molybdenum (Sakaguchi *et al*, 1981), cadmium (Holan *et al*, 1993), arsenic, copper and zinc emissions are also attributed to the non-ferrous metal industry and combustion of gasoline is a major contributor to lead emissions (Nriagu and Pacyna, 1988) The iron and steel industry contribute chromium and manganese to the atmosphere (Palmer and Wittbrodt, 1991) This release of metals is in addition to the natural chemical cycling of these elements (Loring, 1979) For example, a major source of atmospheric selenium is from marine biogenic origin (Maiers *et al*, 1988)

1 03 Metals in Soils

All soils contain various metals in trace amounts The type and amount depends on the parent rock from which the soil was formed and the processes involved in its formation Metals in soils may be present at elevated levels due to the addition of substances such as pesticides (As), swine manure (Cu) and some effluent sludges used as fertiliser (various metals)

Soils generally have an overall negative charge that facilitates cation adsorption Anion adsorption on soils can involve electrostatic forces but the process is generally characterised by bond formation between the anion and the soil particles Adsorption of chromate by some soils has been linked to positive charges on mineral coatings of iron oxides within these soils (Bartlett and James, 1979) Molybdate in soil can also bind to certain Fe or Al oxides (Parfitt, 1978) Elements undergo changes in oxidation state under the range of redox conditions found in soils Selenium may exist as selenide (Se^{2+}), elemental selenium, selenite (SeO_3^{2}) as Se^{4+} and selenate (SeO_4^{2}) as Se^{6+} in soils and sediments (Calderone *et al*, 1990)

Human activities can readily cause local conditions of elevated metal (or metalloid) concentrations in soil as seen in agricultural use of arsenic (Onken and Hossner, 1996) and selenium (Calderone *et al*, 1990) Selenate has a much higher bioavailability than the more reduced forms of Se Problems associated with Se contamination in soils include toxicity in livestock and wildlife (selenosis) and these occur frequently in areas of seleniferous sedimentary rocks Coal mining may also bring Se compounds to the surface Livestock can be susceptible to molybdenosis, a molybdate-induced copper deficiency Overabundance of trace elements and/or their substitution by non-essential ones (i e selenium for sulphur) can lead to serious disruption of normal physiological processes and may result in toxicity and death The effects of soil pH on bioavailability of oxyanions are variable As, Mo, Se and Cr become more available as pH increases but to different extents (Thompson-Eagle and Frankenberger, 1991)

Chromium may be present in soils in levels ranging from traces up to extremes of 1500ppm of total chromium (Cary, 1982) The majority is generally in insoluble form and only a minor fraction, (0 006 - 0 28%) of the total chromium is available to plants (Desmet *et al*, 1975) When Cr reaches the soil from an external (industrial) source, it will either be in solution, adsorbed or precipitated on sediment particles or complexed with organic matter The pH of Cr-containing soils normally lies between 4 and 8, with lower or higher values being exceptional (Cary, 1982) At very low pH, Cr may be present as Cr(III) while a combination of high pH and high redox potential will lead to the presence of chromate anions (Bartlett and James, 1979) As the Cr(III) cation will be strongly adsorbed on the cation exchange sites and complexed by organic matter, in most soils it will be the chromate anion that will stay in the soil solution i e the moisture surrounding each soil particle This may pose a toxicity problem for plants or the chromium may migrate down soil profile to the ground water table (Desmet *et al*, 1975) In acid solution, Cr^{2+} (chromous), Cr^{3+} (chromic) and Cr^{6+} (dichromate) in $Cr_2O_7^{2}$ are present while in basic solution $Cr(OH)_2$, CrO_2 (chromite) and CrO_4^{2} (chromate) are the dominant species (Cary *et al*, 1977b) The chromous ion is rapidly oxidised to Cr^{3+} by air and slowly by H^+ The most common natural source of chromium in soils is the mineral chromite and elemental Cr is not found in nature Cr(VI) is relatively stable and mobile in soils that are sandy or contain low organic matter concentrations In subsoil washed with dilute NaOH Cr(VI) sorption increases with decreasing pH A small amount of Cr(III) in soil may be oxidised to Cr(VI) and become mobile This chromium will either be sorbed, reduced or leached into ground or drainage water Naturally occurring Cr concentrations in water arise from mineral weathering processes, soluble organic Cr, sediment load and precipitation (Cary, 1982)

1.04 Metals and Plants

Plants may be used as indicators of trace element bioavailability They may also respond to trace elements in soils as accumulators or excluders Accumulators are plants that actively take up an element, out of necessity or with no apparent need, and which can tolerate high concentrations of the element without suffering any ill effects Legumes can accumulate Mo even though their metabolic requirements for Mo are quite low (Pierzynski *et al*, 1993) Species of *Astragalus* are Se accumulators, and their presence can be used to identify Se-rich soils Indicators are species that take up an element in proportion to the amount present in the soil (until phytotoxicity occurs) but do not necessarily require it for their growth Excluders actively exclude elements such that the concentration of the trace element in the plant will remain relatively low and constant at varying soil trace elemental levels (Pierzynski *et al*, 1993)

Although the chromium content of plants is generally low (0 01-0 1ppm) (Lyon *et al*, 1969), some species can accumulate an appreciable amount without presenting apparent toxic effects Among proposed mechanisms of tolerance is the formation of organic Cr complexes (Lyon *et al*, 1969) At low concentrations, Cr is considered to be an essential microelement and to have a stimulating effect either directly on the growth and the yield of the plants or indirectly by enhancing the assimilation of the nutrient elements A continuous flow of chromium into apples occurs throughout their growth This movement of chromium is similar to that of essential trace elements such as boron, zinc, iron, copper and manganese, into apples (Coahran *et al*, 1973) Toxicity of chromium is explained in relation to its chemical form and concentration as well as the presence of other metals It may interfere with the uptake of such necessary elements as calcium and iron, phosphorus or sulphur resulting in plant deficiencies Potassium chromate is more toxic to the growth of barley than chromic chloride, though apparent uptake of the latter is much faster Inhibitor studies demonstrated that CrO_4^2 uptake is active while Cr^{3+} uptake is passive (Skeffington *et al*, 1976) Cr-EDTA is less absorbed and metabolised by the roots of rice plants than CrO_4^2 or Cr^{3+} but its translocation rate is higher Chromium taken up by plants may be chelated by root proteins, and to a certain extent, its uptake is dependent on metabolism When chromium penetrates as chromate, it is reduced to the chromic state in the cytoplasm and organelles When it is present in the root environment as Cr(III) or CrO_4^{2-} absorbed chromium is mainly accumulated in the root system and only sparingly translocated to the aerial parts However, hexavalent organo-chromium compounds and even free chromate ions have been detected in the xylem of *Leptospermum scoparium* (Lyon *et al*, 1969)

Phytotoxicity poses major problems for food production The productivity of cropland may be reduced due to the presence of excessive amounts of metals Cadmium-enriched rice caused *itai-itai* disease and renal dysfunction in Japanese farmers The rice had been grown in paddies contaminated by lead and zinc mining and smelting activities Due to the geochemical link between cadmium and zinc, zinc-polluted sites often have high cadmium concentrations as well (Pierzynski *et al*, 1993)

1 05 Methods for Treating Metal-bearing Effluents

Removal of metals from wastewaters depends on the chemistry of the metals involved (Palmer and Wittbrodt, 1991) Treatments include flotation, coagulation and flocculation, precipitation as either hydroxides or sulphides (Volesky, 1989), ion exchange (Holan *et al*, 1993), oxidation and reduction (Von Gunten and Oliveras, 1998), membrane technology and adsorption (Mahuli *et al*, 1997)

Traditional treatment of chromium-containing wastewater involves the reduction of Cr(VI) to Cr(III), precipitation of Cr(III) as $Cr(OH)_3$ above pH5 5, collection of the precipitate and disposal of the dewatered sludge The problems with this method include sludge disposal and incomplete reduction of Cr(VI) (Kratochvil *et al*, 1998) Membrane filtration has been utilised in the treatment of tannery wastewater In conjunction with precipitation, membrane filtration can reduce Cr content in the effluent and improve the purity of recovered chromium Organic components, metals and other impurities are reduced or eliminated from the recycled Cr solution (Cassano *et al*, 1996)

Ion exchange has been used to treat iron and manganese in groundwater Membrane processes such as electrodialysis and reverse osmosis are presently used for desalination of large volumes of water (Wase and Forster, 1997) Chemical addition (to cause oxidation and reduction, for example) must be controlled in these processes to prevent excessive sludge production This sludge then has to be disposed of, usually to landfill sites This can lead to run-off and leaching to groundwater and nearby rivers (Wase and Forster, 1997) The volumes and characteristics of the sludge will vary between different industries and metals These methods for removing metals from wastewater are often costly and less efficient at lower metal concentrations or higher solution volumes These factors and problems associated with resulting sludge have forced industries to investigate other treatment options

Reverse osmosis has potential as a method for treating chromium-containing rinse waters It is claimed to have a lower operating cost than ion exchange or conventional evaporation for chemical recovery Chromium was concentrated by a factor of 13 3 for recycling by this process while water recovery was 91% (Schoeman *et al*, 1992)

Electrodialysis, normally used for desalination, can be utilised for the treatment of electroplating effluent The introduction of packed ion-exchange resins into the electrodialysis chamber results in higher ion recovery, utilising both adsorption and electrodialysis (Grebenyuk *et al*, 1998)

1 06 Chromium Tanning Wastewater

The use of chromium salts in leather tanning began in 1858 (Thorstensen, 1976) In 1985, 32% of the total world trade in chromium compounds was used in leather tanning (Walsh and O'Halloran, 1996a) Approximately 40 million cubic meters of Cr-containing tannery effluent is disposed of into watercourses worldwide each year (Macchi *et al*, 1991) Successful treatment of tanning effluent should result in reuse, in the tanning process, of recovered chromium and water without affecting the product quality (Landgrave, 1995)

The tanning process begins (see Table 1 01) with 'salting' where the fresh hides are treated with sodium chloride (Ronan Group Ltd Co Tipperary, personal communication) This is followed by fleshing before the hides are soaked in a sodium sulphide and lime solution This 'liming' hydrolyses and dissolves globular proteins and hair Bating and pickling acidifies the hides, preparing them for tanning using chromium 'Wet blue' is the name given to chromed leather The hides are treated with a Cr(III) solution between pH3-4 in rotating drums Cr(III) forms crosslinks between the collagen fibres This crosslinking is responsible for the durable finish of leather and prevents rotting of the skins (Walsh and O'Halloran, 1996a)

Tanning wastewater is characterised by three primary streams the chrome and sulphide streams and the final effluent (Landgrave, 1995) The sulphide stream contains dissolved protein, high BOD, salt and sulphide at pH greater than 11 The chrome stream mainly consists of Cr salts and fungicide at pH 4 - 5 Generally, the streams are mixed to neutralise the chrome stream and precipitate the Cr as hydroxides This final effluent is usually released with or without further treatment depending on the tannery (Walsh and O'Halloran, 1996b)

1 07 Use of Various Biomass in Metal Removal

An alternative to the conventional methods of metal removal is biosorption, the passive uptake of metal ions by natural materials of biological origin Biosorbent materials commonly include fungal biomass and yeast (Krauter *et al*, 1996, Blackwell *et al*, 1995), freshwater algae (Crist *et al*, 1990, Sandau *et al*, 1996), bacteria (Moore and Kaplan, 1992) and peat (Ho *et al*, 1994)

A desirable biosorbent must be cheap, readily available and easy to handle as well as having good uptake characteristics Metal binding to various types of biomass can occur by surface adsorption (biosorption) to viable and non-viable biomass or intracellular

Table 1 01 Chrome Tanning Process *adapted from* Cabeza *et al*, (1998)

Chemicals in				Wastewater	Solid Waste
		Brine Cured Hides			
		↓			
		Trimming and Sorting	→		Trimmings
		↓			
Water Wetting agents	→	Soaking	→	Salt Dirt Blood	
		↓			
		Fleshing	→		Fleshings
		↓			
Water Lime Sodium sulphide	→	Unhairing	→	Dissolved hair Alkaline solution	Lime trimmings
		↓			
Water Ammonium sulphate Enzymes	→	Bating	→	Unwanted hide constituents Alkaline solution	
		↓			
Water Sodium chloride Sulphuric Acid	→	Pickling Acidification of hides			
		↓			
Basic chromium sulphate Sodium bicarbonate	→	Tanning	→	Non-exhausted chromium Other salts	
		↓			
		Wringing	→	Non-exhausted chromium Other salts	
		↓			
		Splitting and Shaving	→		Splits Shavings
		↓			
Different chemicals depending on end use	→	Retanning Colouring and Fatliqoring	→	Non-exhausted chromium and chemicals	
		↓			
		Crust Leather			

accumulation by viable biomass only (bioaccumulation) The extent of binding mainly depends on the chemistry of the metal ion (Brady and Tobin, 1995)

The use of microorganisms as cationic metal biosorbents has been reviewed extensively (Hutchins *et al*, 1986, McHale and McHale, 1994) Their potential in the removal of anionic metals has also been demonstrated (Korenevsky and Karavaiko, 1990, Garnham *et al*, 1993, Lloyd-Jones *et al*, 1994) Bacterial species may exhibit anionic and cationic metal tolerance and uptake mechanisms in their natural environment and can be exploited for wastewater treatment (Lloyd-Jones *et al*, 1994, Macy *et al*, 1996, Moore and Kaplan, 1992, Wase and Forster, 1997) *Thauera selenatis* is a selenate-respiring bacterium which reduced the selenium oxyanion content of wastewater (230-640 g/l) in the San Joaquin Valley in California (Cantafio *et al*, 1996) Significant amounts of elemental selenium were recovered from the bacterium after contact while the selenium oxyanion content in the wastewater was reduced by 98% to an average of 12 ± 9 µg/l *Rhodobacter sphaeroides* has high level resistance to tellurite, selenite and at least fifteen other rare-earth oxides and oxyanions (Moore and Kaplan, 1992) This resistance enables the organism to accumulate high metal concentrations without deleterious effects Other members of the class Proteobacteria also exhibit arsenate, molybdate and tungstate resistance (Moore and Kaplan, 1992) Bacteria have also been utilised as a biosorbent for chromate from wastewaters (Wang and Shen, 1995) *Enterobacter cloacae* reduced Cr(VI) to the less toxic form of Cr(III) with complete reduction of as much as 2 mM chromate occurring within 24h (Komori *et al*, 1990) *Pseudomonas putida* also reduced chromate and the chromate reductase activity was associated with soluble protein rather than the membrane fractions of the cells (Ishibashi *et al*, 1990)

Fungi have metal sequestering properties and potential use in wastewater remediation Metals such as Ni, Mn, Co, Cu and Zn are actually micro-nutrients essential for growth in yeast and fungi Other metals such as Cd, Ag and Au are taken up even though they are not essential to the organism (Wase and Forster, 1997) *Saccharomyces cerevisiae* has been used in the removal of chromate from groundwater (Krauter *et al*, 1996) and in the treatment of other metals in wastewater (Volesky and May-Phillips, 1995) Molybdenum biosorption using *Candida scotti* and *Rhizopus arrhizus* biomass attained maximum adsorption capacities of 173 2 mg/g and 188 2 mg/g dry weight respectively (Korenevsky and Karavaiko, 1990) The influence of anions on cation uptake by *R arrhizus* varied between inhibition of uptake to no effect at all (Tobin *et al*, 1987)

Immobilised *Saccharomyces cerevisiae* removed 6 63 mg/g chromate from chrome-plating wastewater which initially contained 30 mg/l Cr(VI) at pH 7 1 (Zhao and Duncan, 1998) The final chromium concentration after successive treatment was 4 mg/l, which was still too high for discharge and contained predominantly Cr(III)

Algal biomass is also a potential biosorbent Metal interactions with algae range from relatively weak electrostatic bonding exhibited by alkali and alkaline earth metals to stronger coordination bonding for certain transition metals with oxygen, nitrogen and sulphur ligands (Crist *et al*, 1988) The accumulation of molybdenum by the green microalgae, *Chlorella regularis*, increased with higher Mo concentrations but decreased with increasing algal concentration Maximum uptake was 132 mg/g dry weight (Sakaguchi *et al*, 1981) *Chlorella vulgaris*, adsorbed gold, silver and mercury ions (Darnall *et al*, 1986) while the estuarine microalga, *Chlorella salina*, adsorbed cobalt (Garnham *et al*, 1991) The use of algal biomass has several attractive features with regard to the recovery and disposal of bound metals These include (1) the bound metals can often be eluted and the biomass reused for several cycles, (2) the metal-laden biomass can be greatly reduced in volume by an inexpensive drying step, and (3) the metal-laden biomass is more suitable for incineration than metal-laden precipitates or ion exchange resins derived from conventional metal removal processes (Wilde and Benemann, 1993)

Larger plants such as water ferns (Zhao and Duncan, 1997) and seaweed (Schiewer and Volesky, 1997) are potential biosorbents Three species of seaweed, *Sargassum natans, Fucus vesiculosus* and *Ascophyllum nodosum* have high cadmium biosorptive capacities (Holan *et al*, 1992) *A nodosum* accumulated the highest amount of cadmium, exceeding 100 mg/g, outperforming a commercial ion exchange resin, DUOLITE GT-73 Equilibrium metal uptake of cadmium, cobalt, nickel, lead and zinc by *Ascophyllum nodosum* and *Sargassum fluitans* ranged from 369 mg Pb/g to 77 mg/g for Zn and Ni by *S fluitans* and from 287 mg Pb/g to 73 mg Zn/g for *A nodosum* (Leusch *et al*, 1996) The maximum adsorption capacity for chromate uptake by the water fern, *Azolla filiculoides*, was between 70 6 mg/g and 120 2 mg/g, at pH 2 (Zhao and Duncan, 1997)

Aquatic macrophytes, such as waterferns and duckweed, have potential in the treatment of chrome-laden tanning waste Abundant and easily cultivated aquatic plants provide an economic and convenient source of biomass Although successful at low concentrations, a problem still arises in the disposal of the contaminated plant matter (Vajpayee *et al*, 1995) Seaweed has traditionally been used as a fuel and fertiliser so consequently, this may be a possible mechanism of disposal

1 08 Peat as a Biosorbent of Metals

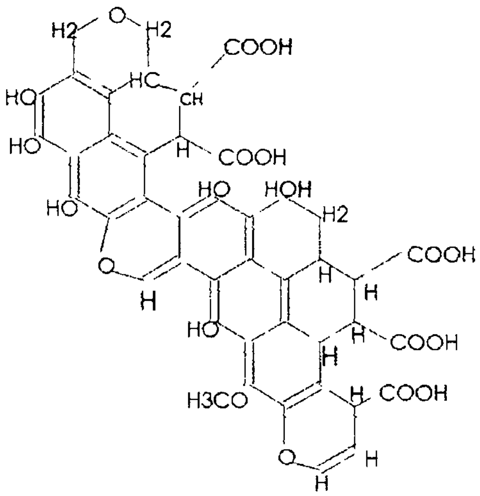
Peat is partially decayed plant matter formed in predominantly anaerobic conditions where the accumulation of plant material exceeds the decomposition rate It is usually formed in areas where water saturates the dead plant material Water restricts the action of aerobic bacteria and consequently inhibits the rate of decay of the plant matter (Wase and Forster, 1997), (Figure 1 01) Major components include cellulose and lignin (Figure 1 02) which bear polar functional groups such as aldehydes, ketones, alcohols, acids and phenolic hydroxides that can be involved in chemical bonding (Coupal and Lalancette, 1976)

Peat has a high adsorption capacity for polar organic molecules and transition metals (Couillard, 1992) This polar nature and its ion exchange capacity has led to investigation of its potential for the treatment of metal-contaminated wastewaters (Bloom and McBride, 1979, Chen *et al*, 1990, Couillard, 1994) The ability to adsorb metals is dependent on the solution pH Below pH 3, most metals will be leached from peat and above pH 8-8 5, the peat itself is not stable and will degrade (Coupal and Lalancette, 1976) The optimum pH for uptake of most metals can be found within this range

Lignin, one of the main components of peat, consists of aromatic rings with 3-carbon side chains and is structurally stable (Figure 1 02) Peat, by its nature, is compressed and has lost many of the lignin side chains resulting in the presence of charged groups which give rise to chelation properties (Wase and Forster, 1997)

Peat is a highly porous substance (Couillard, 1992) Partially decomposed peat has a specific area of 200m^2/g and a porosity of approximately 95% (Couillard, 1994) It can be considered as an organic soil with a capacity for cation exchange It adsorbs basic dye cations due to the negatively charged functional groups in its fulvic and humic acids and to exchange with hydrogen ions (Allen *et al*, 1988) Repulsion between anions and the negatively charged groups means that although cation exchange capacity of peat is high, anion exchange capacity is generally low (Valentin, 1986)

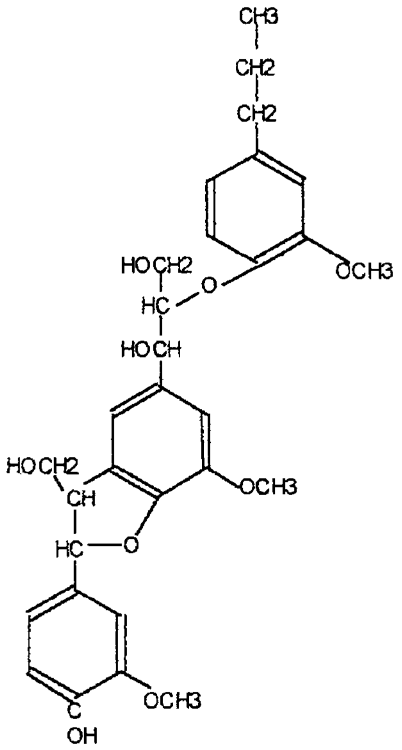
Figure 1 01 Structure of Peat



adapted from Biosorbents for Metal Ions, Wase & Forster, 1997

Environmental characteristics during formation and the extent of its decomposition determine most of the properties of natural raw peat These properties can also be affected by the manner of harvesting of the peat The distribution and concentration of Ca, Fe, Al, and Cu, Cd, Cr, and Sr, if present, vary with depth in the peat bed (Parkarinen *et al*, 1981)

Figure 1 02 Structure of Lignin



adapted from Biosorbents for Metal Ions, Wase & Forster, 1997

Peat that is partially decomposed has good adsorptive and hydraulic characteristics and is more fibrous Sphagnum peat, of low to moderate degree of decomposition, is acidic and porous, has a large surface area and provides a favourable environment for the growth of microscopic fungi It is capable of holding large quantities of water as it has a high pore volume (Van Quach *et al*, 1971) All water-borne particles larger than the interstitial channels within the peat are filtered out while other contaminants are removed by ion exchange and chelation (Bloom and McBride, 1979) The peat fibres can be teased apart by drying and this can increase surface area for adsorption (Couillard, 1992) Peat has been used as a biosorbent for different types of contaminants and effluents from oil to animal waste Since peat is polar, it can adsorb large amounts of metals (Chaney and Hundemann, 1979) Although most research has focused on cationic metal species, work with acidic dyes (overall negative charge) shows that peat could possibly be used to treat a range of contaminating metal anions (Allen *et al*, 1988)

The ash content of peat is very small so the combustion of the spent biomass will result in the formation and easy recuperation of metal oxides (Couillard, 1994)

1.09 Pre-treatment of Peat for Biosorption

While raw peat can be successfully used in adsorption applications to remove various components (metals, organic compounds, dyes etc) (Dubuc *et al*, 1986), efficiency may be restricted by a number of factors including chemical instability, shrinkage or swelling of the peat due to permeability and low mechanical strength (Chen *et al*, 1990) Consequently, peat is generally treated before use The most common treatment is sieving and washing followed by drying, usually at high temperatures Chemical treatment of the peat with phosphoric or sulphuric acids enhances its cation exchange capabilities, while addition of amines and ethylenediamine to previously acid-treated peat increases the anion exchange capacities (Smith *et al* , 1977) In some cases peat (in filter beds) has been used in conjunction with other treatments mechanism such as flocculation or precipitation to treat metal contaminated water (Coupal and Lalancette, 1976) For example, chromate-containing water required special treatment involving the addition of ferric chloride and sodium sulphide in the range of pH 5-7 This resulted in a 2 5 6 molar ratio of Cr(VI), ferric chloride and sodium sulphide in the effluent, which precipitated chromium in the trivalent form Chromate was reduced by the sulphide ion and coprecipitated as hydroxide with iron The supernatant was then contacted with a bed of peat and the Cr(VI) concentration in the final effluent was below detectable limits (Coupal and Lalancette, 1976) Effluent containing the cyanide anion has also been treated using this method A ferro-cyanide was formed by the addition of ferrous sulphate resulting in precipitation The treated water was then contacted with peat and the cyanide concentration reduced to below 1ppm

Peat is a potential biosorbent for a range of effluents Septic tank effluent was treated with peat (Brooks *et al* , 1983) as was beef extract and alkyl benzene sulphonate solution (Van Quach *et al* , 1971) Sphagnum peat adsorbed 12 29 mg/l copper from wastewater (Ho *et al* , 1994) and cadmium, from an initial concentration of 560 µg/l to 3 µg/l (Chaney and Hundemann 1979) The removal of various metals, phosphates and organic matter (oil, detergents and dyes) by peat has been investigated (Bloom and McBride, 1979, Pakarinen *et al*, 1981, Coupal and Lalancette, 1976) The adsorption of K^+, Na^+, Ba^{2+}, and Ca^{2+} on peat was faster than uptake of trivalent metallic ions (Tummavuori and Aho, 1980a) This was attributed to the difference in ionic radii of the metals The equilibrium time can also vary with the type of peat used, as peat from different areas will have distinct characteristics and biosorption properties When chromate was adsorbed on sphagnum peat it took several hundred hours to reach equilibrium (Couillard, 1992) However, adsorption equilibrium was attained in about 30 minutes with a chromium-containing wastewater (Van Quach *et al*, 1971)

1 10 Biosorption Mechanisms of Peat

The mechanism of uptake can vary with the type of peat used Ion exchange alone may account for metal sorption by sphagnum peat, i e that two H^+ ions are exchanged with one divalent metal ion (Aho and Tummavuori, 1984), although the occurrence of both complexation and ion exchange has been reported (Bloom and McBride, 1979) No anion release was observed no matter what type of peat was used during copper fixation by raw peat (Chen *et al*, 1990) The fixation of metal ions by untreated peat most likely involves cation exchange with H^+, Ca^{2+} and Mg^{2+} contained within the peat, and adsorption-complexation, i e uptake and fixation of the same equivalent of cations and anions

Assessing hydrogen ion release is insufficient to study the process of ion exchange since the peat may have adsorbed other metal cations in its natural environment This is especially true when using untreated peat (Chen *et al*, 1990) Raw peat possesses significant cation-exchange capacity due primarily to the carboxyl groups of its humic acid constituents The cation-exchange capacity of peat is increased by sulphuric acid treatment (Smith *et al*, 1976) Various types of peat were less effective in removing metals at pH 2-3 than at pH 4-8, reflecting the effects of hydrogen ion competition for exchange sites Peat-based anion-exchangers (prepared from sulphuric acid treated peat by refluxing with EDTA) have generally had small exchange capacities when compared to cation exchangers A total effective chloride capacity 25-50% of that of commercial anion exchangers with free amine groups was attained Peat anion-exchangers also have the economic disadvantage of requiring multistep preparations, using more than one reagent (Smith *et al*, 1977)

Peat behaves very much like synthetic cross-linked polycarboxylic acids in terms of metal ion adsorption Localisation of ions at carboxylate sites has been suggested (Bloom and McBride, 1979) Soil organic matter in the pH range of most soils (pH less than 8) can be considered a polycarboxylic acid The carboxylic acid groups participate in ion exchange reactions by binding metal ions with the release of H^+ ions (Bloom and McBride, 1979)

The sorption mechanism of peat may be explained as either a chemical reaction in which the surface functional groups form organic metal complexes or a cation exchange reaction For both processes, the diffusion of the ion from the aqueous medium to the peat particle is necessary The overall process is governed by diffusion because both reactions are considered instantaneous

There are essentially four stages in the adsorption process using porous peat

(i) transport from the bulk of solution to the exterior surface of the peat

(ii) movement of contaminant across the interface and adsorption onto external surface sites

(iii) migration of molecules within the pores of the peat, and

(iv) interaction of molecules with the available sites on the interior surfaces of the pore and capillary spaces of the peat (Couillard, 1994)

The affinity of peat for basic dyes (Basic yellow 21, also known as Astrazon yellow 7GL, has an overall positive charge) is not unexpected since the phenolic, hydroxyl and carboxyl groups in humic acid, fulvic acid and lignin will attract ionic dyes (Allen *et al*, 1988, 1989) Many dyes ionise in solution to give coloured cationic or anionic groups, which may then be attracted to the peat On the basis of charge, a basic dye will have a strong adsorption affinity (Couillard, 1994)

1 11 Biosorption by Marine Macrophytes

For economic reasons, biosorbents which are abundant, either generated as a waste by-product of large-scale industrial fermentations or naturally found in large quantities in the environment, are of particular interest Among the diversity of biomass available, marine algae have already proved to be promising for metal adsorption and recovery (Kratochvil and Volesky, 1998) Brown seaweeds of the genus *Sargassum* adsorbed Cd, Ni, Cu, Pb and Zn cations up to 20% of the biosorbent dry weight (Leusch *et al*, 1995) while *Ascophyllum nodosum* accumulated more than 65mg Cd/g biomass (Volesky and Prasetyo, 1994) Most of the seaweed biomass found on Irish coasts consists of a relatively small number of species of which one of the most common are the kelps, *Laminaria* and its relatives (genera Phaeophyta)

1.12 Characteristics of *Laminaria digitata*

Kelps are confined mainly to the subtidal zone of the shore In general, on the east coast of Ireland, kelps occur to depths of a maximum of 8m whereas in the optically clearer waters of the northwestern, western, and southwestern coasts, they may occur to 25m and, exceptionally, to 32m Kelp biomass in the subtidal zone consists of three species of *Laminaria* The most important is *Laminaria hyperborea*, the cast stipes of which are currently collected, dried and exported for alginate production The other two species are gathered in small quantities as food All three kelps occur right around the coast, in large quantities *L digitata* is the kelp commonly seen at low water, whereas *L hyperborea* occurs in deeper water *Laminaria saccharina* occurs sporadically amongst *L digitata* but often becomes the dominant species on wave-sheltered shores (Guiry, 1997)

Classification of seaweeds is primarily based on morphology, generally of the reproductive system Further division into classes depends on the photosynthetic pigments All marine algae contain chlorophyll *a* but in the phaeophyta it is masked by the xanthophyll, fucoxanthin A general feature of seaweeds is the presence of at least one polysaccharide linked with sulphate ester groups These substances resemble complex sulphated polysaccharides in animal tissues but are absent in land plants Most brown algae contain the (-1, 3- linked) glucan, laminaran The polyuronide, alginic acid, is also present (Bold *et al*, 1980)

1 13 Taxonomy of *L digitata*

Kelp, which can grow up to 2m in length, is a fast-growing marine macroalgae Its structure comprises of a claw-like holdfast, a smooth flexible stipe, and a laminate blade to 1 5 m long dividing into finger-like segments The brownish shades of the plant reflect the abundant presence in the plastids of the xanthophyll, fucoxanthin, which is dominant over chlorophylls *a* and *c*, the other xanthophylls, and β-carotene No starch occurs in Phaeophyta Instead the excess photosynthate accumulates as a carbohydrate, *laminarin*, as mannitol, or in the form of fat droplets The protoplast is bounded by a primary wall and middle lamella composed of a gummy substance, alginic acid This may represent 10-25% dry weight Alginic acid is a polymer of D-mannuronic and L-guluronic acids (Kaplan, 1988)

Growth of *L digitata* occurs in the meristematic cells at the junction of the stipe and the blade Growth is rapid for the first half of the year and very slow for the remainder The dry matter content of *Laminaria* is at a maximum in autumn and a minimum in spring Mannitol content is at a minimum during spring and laminarin is absent Alginic acid and proteins are at a spring maximum These changes reflect the fast and slow growth periods Laminarin and mannitol are the two photosynthetic products characteristic of the phaeophyta Alginic acid occurs in the cell membrane In brown seaweeds, the roles of mannitol and lamarin are analogous to that of sugar and starch in higher plants Alginic acid and cellulose are cell wall constituents (Bold *et al*, 1980)

Other metabolism products include fats, proteins and sterols Iodoamino acids are characteristic of Phaeophyta while the fatty acids are similar to those of higher plants and animals Fucosterol is the common sterol Fucosan vesicles are found in meristematic, reproductive and assimilatory cells of Phaeophytes and are comparable to tannins Iodine metabolism fluctuates seasonally in brown algae and iodo-volatilisation occurs on exposure to air Iodine, present in the form of iodide, is liberated by enzyme action when surface layers become damaged

The functional groups of components such as alginic acid, mannitol and lamarinin in the seaweed may provide the binding sites for the biosorption of metal ions. These include amine, carboxyl, phenol, ester and hydroxyl functional groups.

1 14 *L. digitata* as a biosorbent for metals

Brown seaweeds in general have been shown to be successful biosorbents of metal ions from solution (Volesky and Prasetyo, 1994, Leusch *et al.*, 1995, 1996). The biosorption properties of *L. digitata* have remained largely undiscovered as it has rarely been used in metal remediation. As *L. digitata* is a novel biomass, it is compared with peat, a biosorbent that has been characterised in detail (Coupal and Lalancette, 1976, Ho *et al.*, 1994, Couillard, 1994). This work is a comparative study of the capacities of *L. digitata* and peat biomass to adsorb metals from a range of single-metal solutions and from industrial wastewaters.

CHAPTER 2

MATERIALS AND METHODS

CHAPTER 2: MATERIALS & METHODS

2 01 *L digitata* biomass

Laminaria digitata blade sections were harvested under water from a rocky coastline at Portrane, Co Dublin The kelp was washed extensively with tap water followed by distilled water and dried for two days at 60°C Kelp dry weight was determined by drying 1g wet samples to a constant weight at 60°C The kelp was size-fractionated using a mechanical sieve shaker Size fractions in the ranges of 2-4 mm, 1 4-2 mm and 1-1 4 mm were used

2 02 Milled Peat biomass

Milled peat supplied by Bord na Mona was sieved using a mechanical sieve shaker to remove fines and large particles Particles in the range 2-4 mm were used in this work Pretreatment of the biomass was the same as that used for kelp (see section 2 07)

2 03 Metal solutions

Metal stock solutions (1000ppm) were made, of Cr(VI) (K_2CrO_4 ACS Reagent, Sigma chemicals), Cr(III) ($Cr(NO_3)_3$ $9H_2O$, Riedel de Haen), copper ($Cu(NO_3)_2$ $3H_2O$), cadmium ($Cd(NO_3)_2$ $4H_2O$ Riedel de Haen), calcium ($Ca(NO_3)_2$ $4H_2O$ Riedel de Haen), magnesium ($Mg(NO_3)_2$ Riedel de Haen), sodium ($NaNO_3$ Riedel de Haen) and strontium ($Sr(NO_3)_2$ Riedel de Haen) with distilled water

2.04 Industrial Effluents

Industrial tanning effluent was provided by Ronan Group Ltd , Clonmel, Co Tipperary The tanning waste was gravity-filtered through Whatman No 1 filter paper to remove excess hair and solid waste The filtered solution was extracted with isohexane (50 ml tanning waste to 100 ml hexane, mixing for 10 minutes in a separation funnel) to determine the presence, if any, of organically-bound chromium This organic filtrate was analysed for the presence of chromium by atomic absorption spectroscopy (see section 2 06)

The initial pH and chromium and sodium chloride concentrations of the inorganic filtrate were determined The solution was analysed for the presence of Cr(VI) by the diphenyl carabazide method (see section 2 06)

The pH of the tanning effluent was raised to pH 11 (with 1 M KOH) to precipitate most of the Cr(III) This simulated industrial conditions where the chrome wastewater is neutralised by the sulphate waste stream resulting in precipitation The final supernatant was filtered and the remaining chromium and sodium concentrations determined before contacting with biomass

2 05 Metal Uptake Experiments

Quantities of 0 1 g kelp and peat were contacted overnight, on an orbital shaker, with 100 ml volumes of a range of concentrations (up to 8 7 mM, equivalent to 10-200 ppm) of each metal or industrial solution The pH was adjusted to 2, 4, or 7 as required, with 1 M H_2SO_4 and 1 M NaOH (or KOH when measuring sodium), before the addition of the biomass Uptake of chromate was the only species investigated at pH 7 due to solubility restraints of metal cations above pH 5 Metal-free and biomass-free solutions were utilised as controls Following centrifugation, the metal concentration of the supernatant after contact was determined

2 06 Metal Analysis

Copper, cadmium, strontium, chromium, calcium, sodium and magnesium concentrations were measured using a 'Perkin Elmer 3100' Atomic Absorption Spectrophotometer (A A S) Cr(VI) concentration was determined by uv/vis spectrophotometry at 540 nm after colormetric reaction with 1,2-diphenylcarbazide (Greenberg *et al* 1992) The final pH was noted

Uptake values, in mmol/g biomass, were calculated from the change in solution concentration using the equation

$$q = V(C_0 - C_f) / M$$

where q is the uptake, C_0 and C_f are initial and final concentrations respectively, V is the volume of solution and M is the weight of biomass used

MINEQL, a chemical speciation program (Schecher and McAvoy, 1994) was used to predict the speciation in the metal solutions and the results were compared with the uptake results from the contacting experiments

2.07 Pretreatment of the biomass

For acid-treated kelp, the *L digitata* fractions were contacted with 1 M H_2SO_4 for 5 minutes and rinsed 3 times with distilled water before drying overnight at 60^0C Calcium-treatment of the kelp involved soaking the acid-treated biomass in 2 5 M calcium hydroxide for 10 minutes before rinsing and drying Untreated kelp was also contacted with a range of copper concentrations (see sections 2 03 and 2 05) before later contact with Cr(VI) This loading was performed in the same way as the uptake experiments, but after equilibrium had been reached, the copper-laden kelp was washed with distilled water adjusted to the experimental pH before subsequent contacting with chromium

2 08 Statistical analysis of experimental errors

Duplicate experiments were performed in all cases, and the results were found to agree to within limits of 5%

Eight replicate Cu^{2+} and Cr^{6+} uptake experiments, at an initial pH of 4 and 2 respectively, were performed for the purpose of statistical analysis Both A A S and the carbazide method were examined The standard deviation was found to be less than 4% These values are consistent with previously reported error analysis (Tobin *et al*, 1990)

CHAPTER 3

RESULTS

CHAPTER 3: RESULTS

3.01 Uptake of metal cations

Metal cation uptake by both types of biomass was greater at pH 4 than at pH 2 Biosorption at pH 7 was not investigated due to solubility constraints of cations above pH 5 *L digitata* proved to be a superior biomass for the adsorption of most metals (Figure 3 01) Uptake levels are quoted as experimental maximums but in many cases, saturation had not been reached Higher sorption values may be attained using higher initial concentrations

Table 3.01 Uptake of metal cations by various biomass at pH 4.

Metal	*L digitata*		Milled Peat	
	pH 2	pH 4	pH 2	pH 4
Chromium*	0 37 mmol/g	1 12 mmol/g	0 08 mmol/g	0 27 mmol/g
Copper	0 13 mmol/g	0 97 mmol/g	0 11 mmol/g	0 32 mmol/g
Cadmium	0 12 mmol/g	0 65 mmol/g	0 07 mmol/g	0 27 mmol/g
Strontium	0 08 mmol/g	0 46 mmol/g	0 05 mmol/g	0 26 mmol/g
Calcium	---	0 62 mmol/g	---	0 29 mmol/g
Magnesium	---	1 10 mmol/g	---	1 33 mmol/g

*Cr(III)

Maximum uptake of copper was 0 97 mmol/g kelp at pH 4 from an initial concentration of 3 14 mM (Figure 3 01) while at pH 2 (Figure 3 02) the maximum level was 0 13 mmol/g kelp from the same initial concentration (Figure 3 02) Cadmium uptake was lower than copper at pH 4 (0 65 mmol/g kelp) but similar at pH 2 with 0 12 mmol/g kelp (initial concentration 1 8 mM) Uptake of strontium from 2 3 mM was 0 46 mmol/g kelp, the lowest uptake value of cations at pH 4 (Figure 3 01)

Both calcium and magnesium are found in seaweed biomass as they are present in seawater at typical concentrations of ca 400 mg/l and ca 1300 mg/l respectively The maximum uptake of calcium by *L digitata* was 0 62 mmol/g at pH 4 (initial concentration 5 mM) while that of magnesium (initial concentration 8 2 mM) was 1 10 mmol/g (Figure 3 01) These uptake levels differed considerably for acid-treated biomass (see section 3 06)

At pH 4, uptake of cadmium was 0 27 mmol/g for milled peat while copper was adsorbed to 0 32 mmol/g Milled peat adsorbed strontium to a maximum of 0 27 mmol/g (Figure 3 03a)

Maximum calcium uptake by peat at pH 4, was 0 29 mmol/g biomass (Figure 3 03b) This is within the range of the adsorption levels of other cationic species, with the exception of magnesium, by peat (Figure 3 03a) but lower than uptake by kelp Conversely, magnesium uptake by peat was greater than by *L digitata* (1 33 mmol/g peat compared to 1 10 mmol/g kelp, Figure 3 01 and Figure 3 03b)

3 02 Biosorption of Cr(III)

The uptake of Cr(III) by untreated kelp biomass increased with increasing concentration at all experimental pHs Maximum uptake of Cr(III) by untreated *L digitata* occurred at pH 4 to a level of 1 12 mmol/g kelp from 3 84 mM (Figure 3 01) This was the highest uptake value recorded for any Cr species by kelp At pH 2, for Cr(III), 0 36 mmol/g dry weight kelp was the maximum uptake by kelp recorded (Figure 3 02) At pH 7, Cr(III) precipitated out of solution and consequently biosorption levels could not be determined

Uptake of Cr(III) by peat increased with increasing solution concentrations until onset of saturation at concentrations of approximately 2 mM and 3 mM at pH 2 and pH 4 respectively Maximum uptake at pH 4 was 0 27 mmol/g (Figure 3 03a) while uptake at pH 2 was 0 08mmol/g (Table 3 01)

3 03 Uptake of Cr(VI) by *L digitata*

Maximum adsorption of Cr(VI) by kelp decreased with increasing pH from pH 2 over a range of concentrations from 0 19 - 3 84 mM (Figure 3 04) Maximum biosorption occurred at pH 2 at 0 82 mmol/g dry weight while appreciable uptake of 0 46 mmol/g was observed at pH 4 The lowest adsorption levels for Cr(VI) by *L digitata* occurred at pH 7 to a level of 0 06 mmol/g

Uptake of Cr(VI) by milled peat increased markedly with decreasing solution pH (Figure 3 05) Highest uptake at pH 2 was found to be 0 58 mmol/g at a final concentration of approximately 3mM At pH 4 and 7, corresponding uptake values dropped to 0 15 mmol/g and 0 09 mmol/g respectively (Table 3 02)

Table 3 02 Uptake of chromate by various biomass

	L digitata	Milled Peat
pH 2	0 82 mmol/g	0 58 mmol/g
pH 4	0 46 mmol/g	0 15 mmol/g
pH 7	0 06 mmol/g	0 09 mmol/g

Figure 3 01 Uptake of cations by kelp at pH 4

(Symbols ▲ Cadmium, ◆ Calcium, ● Chromium,
▼ Copper, ⬢ Magnesium, ■ Strontium)

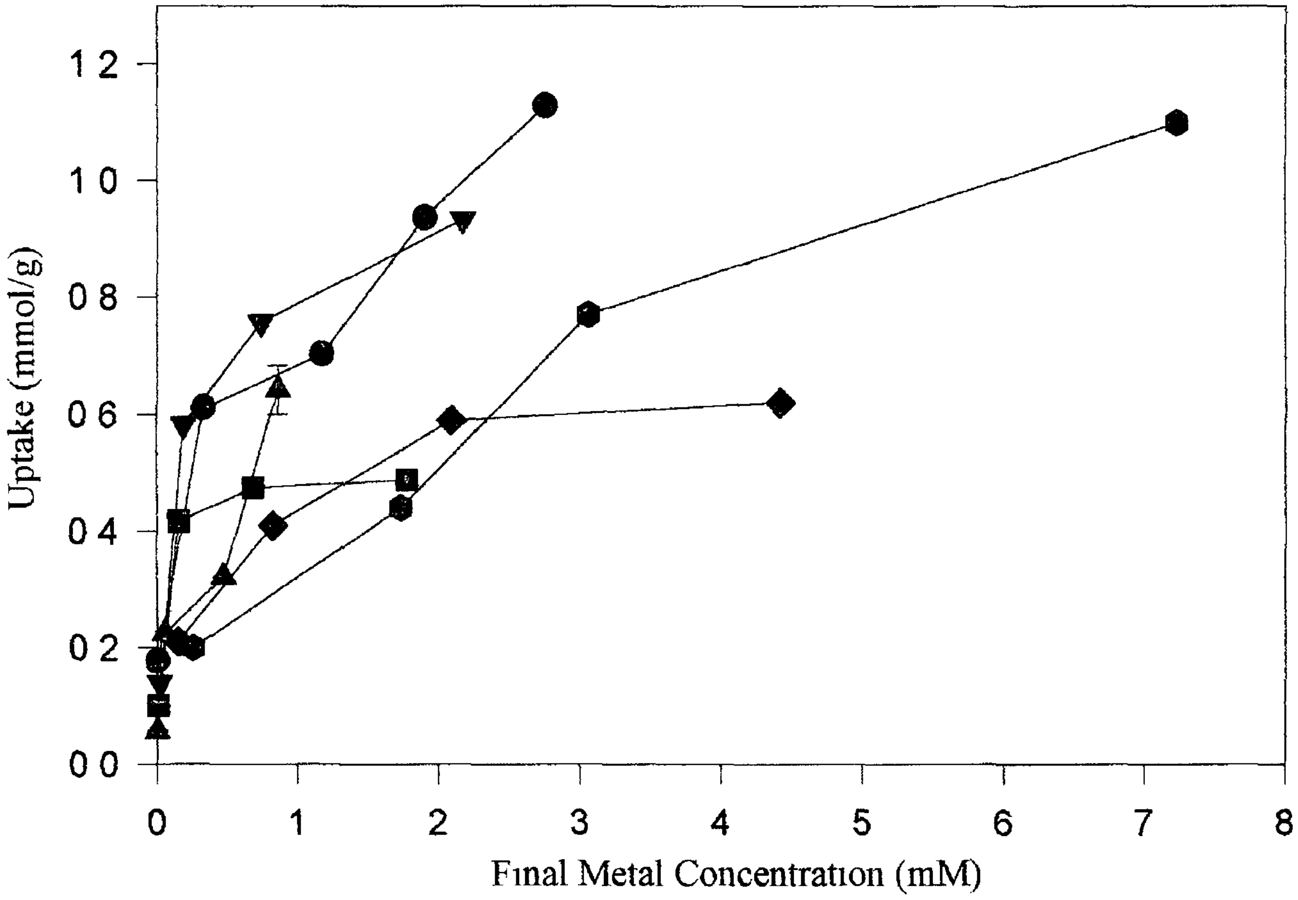


Figure 3 02 Uptake of cations by kelp at pH 2

(Symbols ▲ Cadmium, ● Chromium, ▼ Copper)

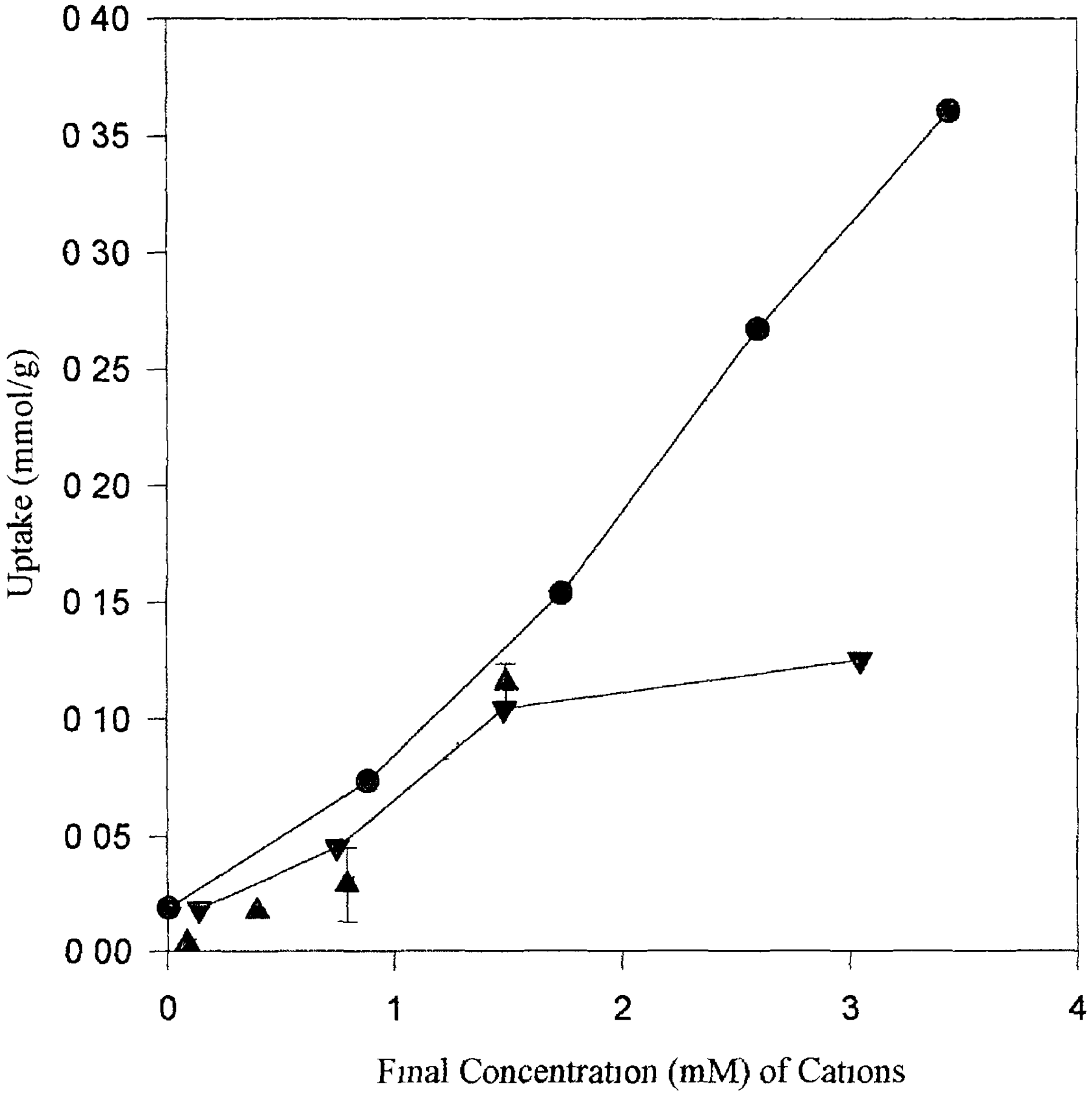


Figure 3 03a Uptake of cations by milled peat at pH 4

(Symbols ● Chromium, ▼ Copper, ■ Strontium, ▲ Cadmium)

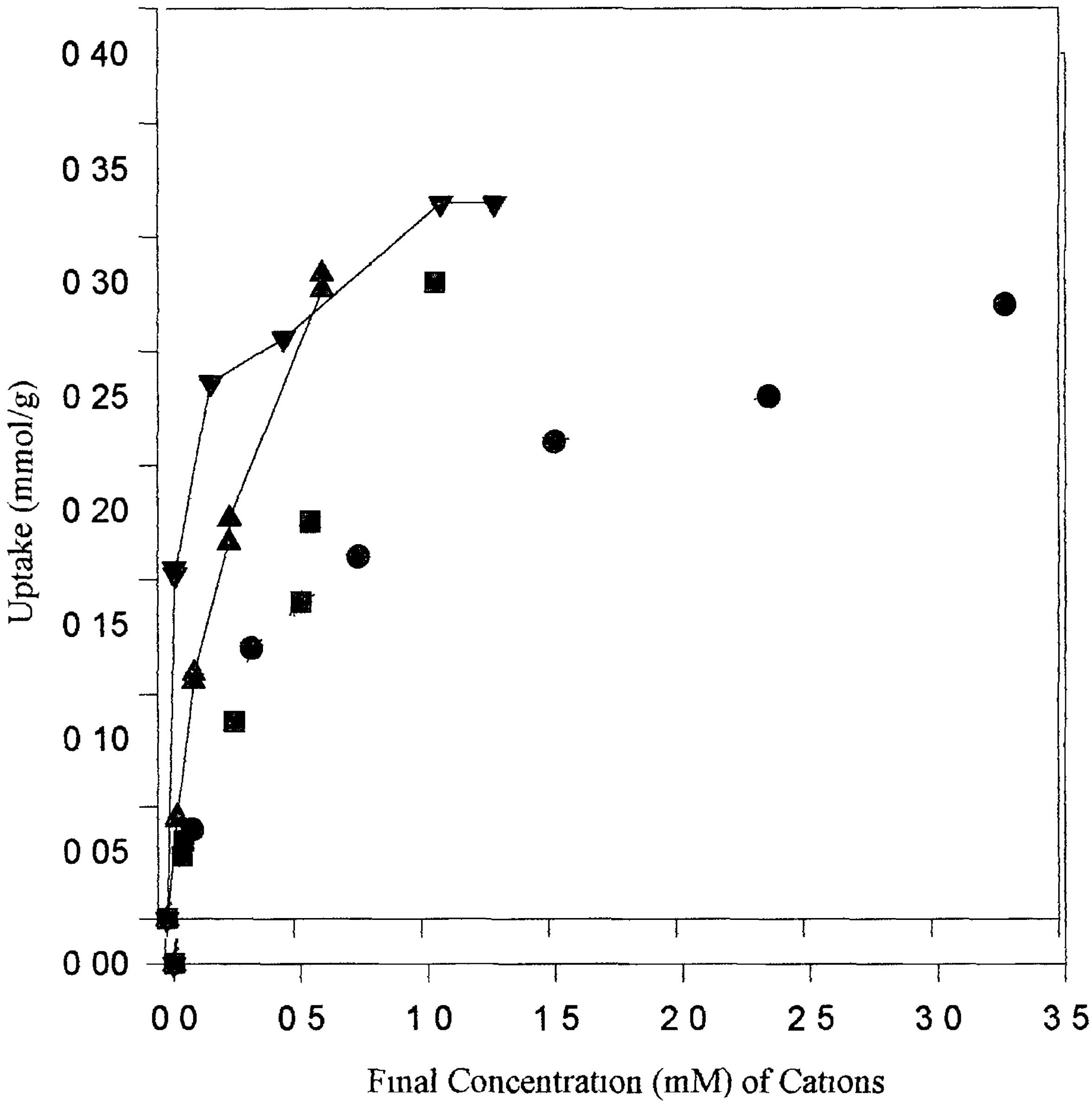


Figure 3 03b Uptake of calcium and magnesium by peat at pH 4

(Symbols ◆ calcium, ● magnesium)

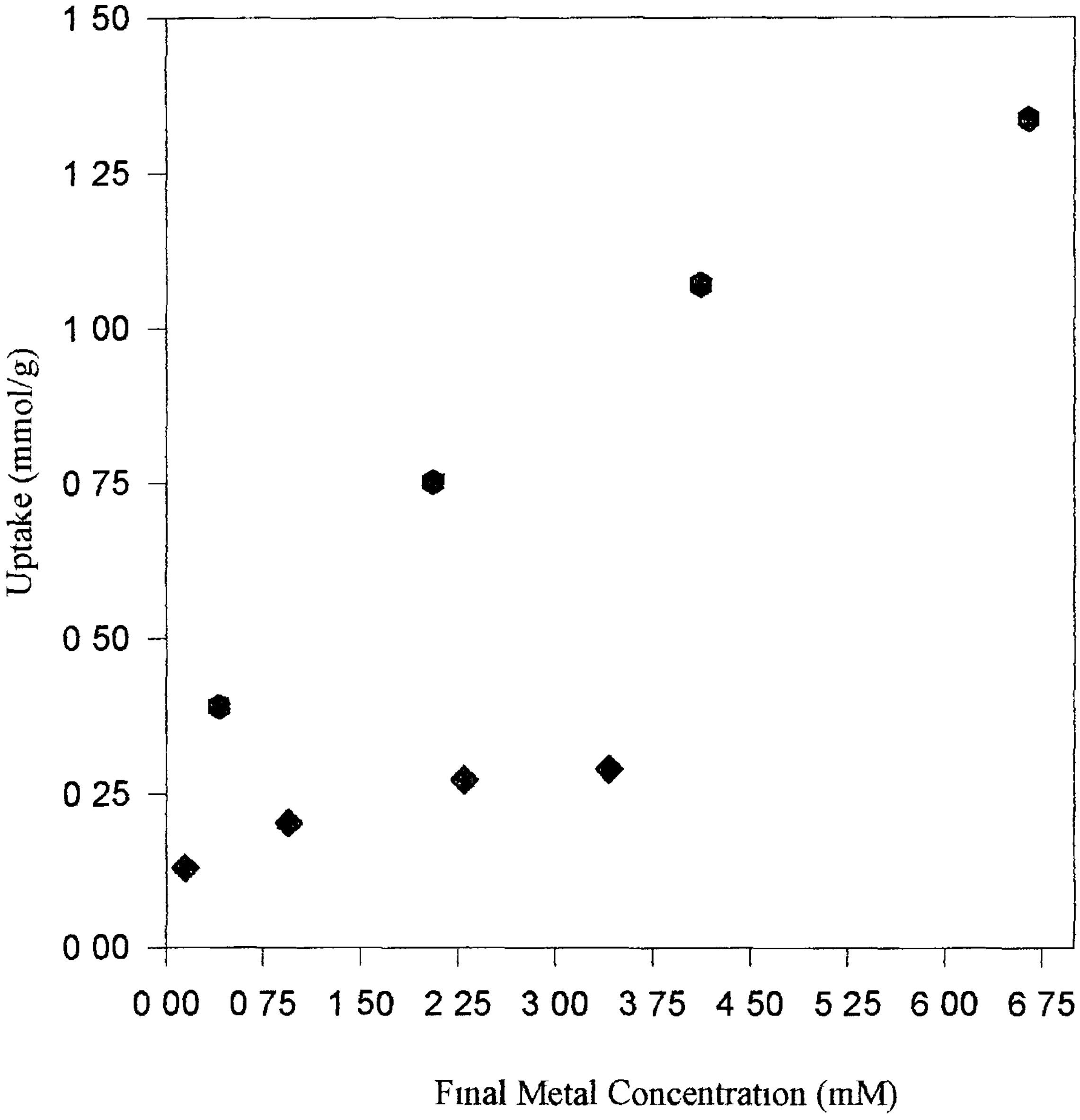


Figure 3 04 Uptake of Cr(VI) by kelp

(Symbols ● pH 2, ■ pH 4, ▼ pH 7)

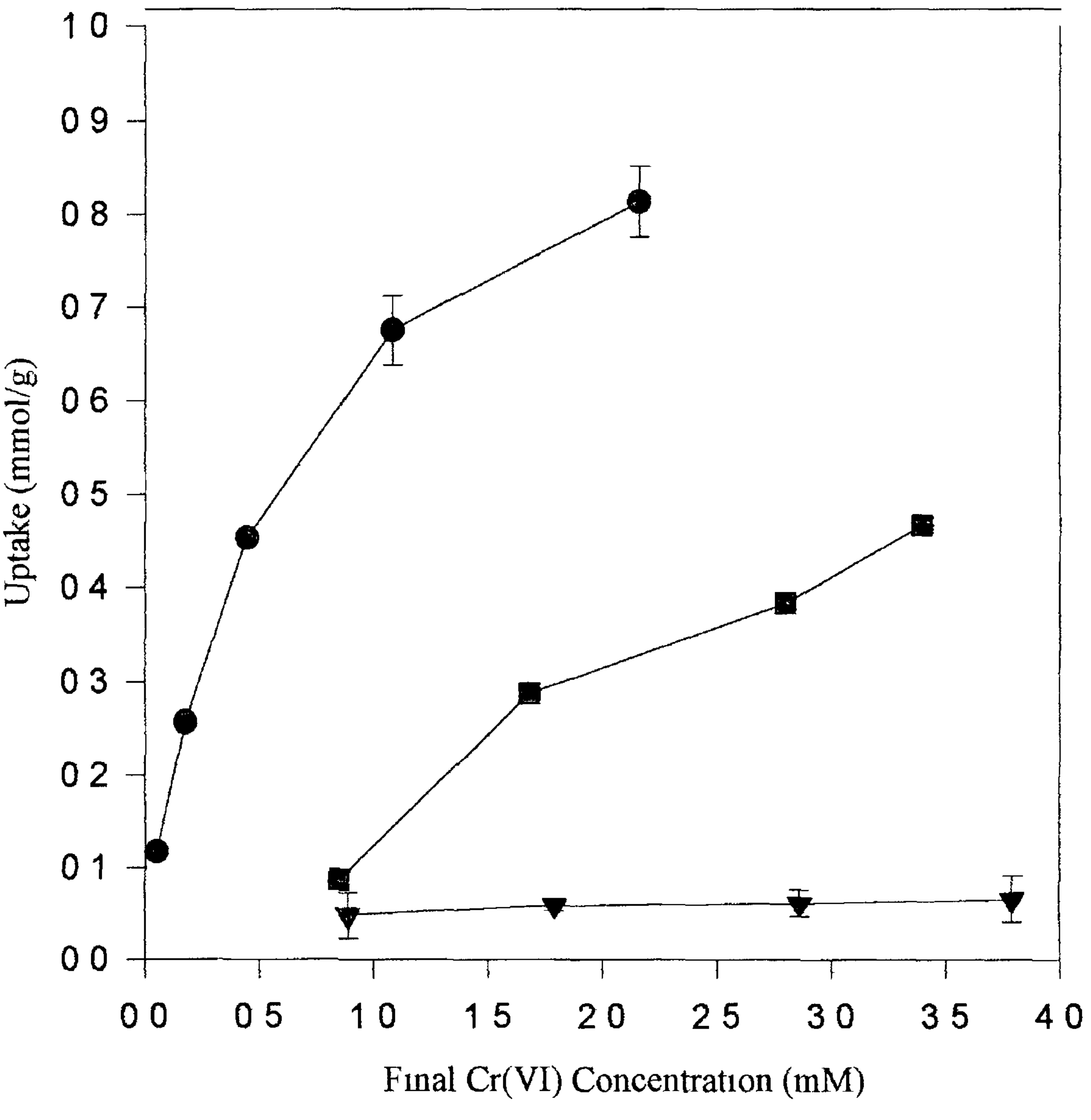
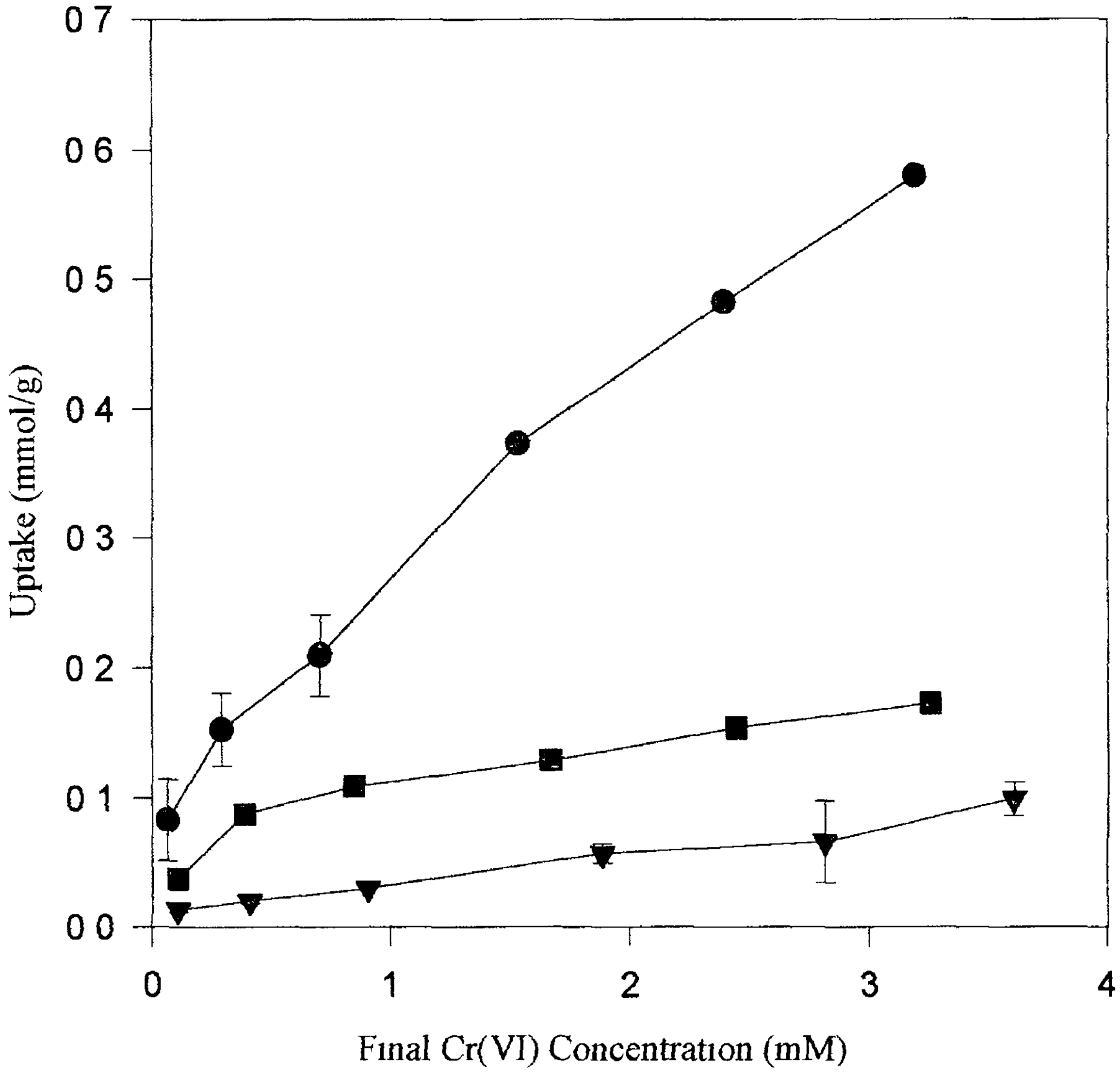


Figure 3 05 Uptake of Cr(VI) by milled peat

(Symbols ● pH 2, ■ pH 4, ▼ pH 7)



3 04 Cr(III) Speciation as predicted by MINEQL

According to MINEQL predictions (Figure 3 06), at pH 2 the predominant Cr(III) species present in solution was Cr^{3+} for all concentrations whereas at pH 4 approximately 49% is Cr^{3+} and 49% is $Cr(OH)^{2+}$ with less than 2% present as $Cr(OH)_2^{+}$ $Cr(OH)_2^{+}$ is the main species at pH 7, at 55% with 42% $Cr(OH)_3$ and less than 2% $Cr(OH)^{2+}$

3 05 Cr(VI) Speciation as predicted by MINEQL

The predominant species at both pH 2 and pH 4 is $HCrO_4$ but the concentration of $Cr_2O_7^{2}$ increases with increasing concentration from less than 2% to 19% (Figure 3 07) At pH 7 CrO_4^{2} accounts for approximately 76% of total chromium while 24% is $HCrO_4$

3.06 Uptake of chromium(III) by pretreated biomass

Both peat and *Laminaria* biomass were subjected to a variety of pretreatments in an attempt to enhance and select certain types of binding sites These included acid, calcium and copper treatments Overall results show that these pretreatments reduced uptake as compared to untreated biomass

Biosorption of Cr(III) by acid-treated kelp followed the same trend with increasing pH as untreated kelp (Figure 3 08) The maximum uptake of Cr(III) by acid-treated *L digitata* occurred at pH 4 to a level of 0 45 mmol/g for a 3 3 mM final solution

Uptake at pH 2 (0 39 mmol/g) by acid-treated kelp was similar to that of untreated kelp at the same pH (Figure 3 08)

The highest uptake of Cr(III) by acid-treated peat, occurred at pH 4, to a level of 0 24 mmol/g (Figure 3 09) At pH 2, the maximum uptake of Cr(III) was 0 06 mmol/g This compares to 0 45 mmol/g and 0 39 mmol/g acid-treated kelp at pH 4 and pH 2 respectively (Table 3 03)

The maximum biosorption of Cr(III) by calcium-treated kelp biomass at pH 4, was 0 75 mmol/g (Figure 3 10) This adsorption exceeded the uptake level of acid-treated kelp but was lower than the maximum uptake by untreated biomass (Table 3 03)

Table 3 03 Uptake of chromium(III) by pretreated biomass

	Untreated biomass		Acid-treated biomass		Calcium-treated biomass	
	kelp	peat	kelp	peat	kelp	peat
pH 2	0 27 mmol/g	0 14 mmol/g	0 39 mmol/g	0 06 mmol/g	0 55 mmol/g	0 15 mmol/g
pH 4	1 12 mmol/g	0 27 mmol/g	0 45 mmol/g	0 24 mmol/g	0 75 mmol/g	0 27 mmol/g

At pH 2, the highest uptake of Cr(VI) by acid treated kelp was 0 68 mmol/g biomass (Figure 3 11) Maximum adsorption at pH 4 occurred to a level of 0 16 mmol/g while 0 07 mmol/g uptake was recorded at pH 7 (Table 3 04) This is consistent with the trend of decreasing uptake from pH 2, with increasing pH, observed with untreated biomass

Maximum adsorption of chromate by acid-treated peat occurred at pH 2 to 0 53 mmol/g (Figure 3 12) This was lower than the uptake attained by untreated biomass At pH 4, uptake was 0 30 mmol/g while at pH 7, 0 19 mmol/g was the maximum uptake achieved (Table 3 04)

Table 3 04 Uptake of chromium(VI) by pretreated biomass

	Untreated biomass		Acid-treated biomass	
	kelp	peat	kelp	peat
pH 2	0 82 mmol/g	0 58 mmol/g	0 68 mmol/g	0 53 mmol/g
pH 4	0 46 mmol/g	0 15 mmol/g	0 16 mmol/g	0 30 mmol/g
pH 7	0 06 mmol/g	0 09 mmol/g	0 07 mmol/g	0 19 mmol/g

Uptake of calcium by acid-treated kelp was 1 05 mmol/g This value was higher than the uptake achieved by the untreated biomass (0 62 mmol/g) Conversely, magnesium was adsorbed to 0 36 mmol/g by the acid-treated biomass compared to 1 10 mmol/g by the untreated kelp (Table 3 05)

Table 3.05 Uptake of calcium and magnesium at pH 4 by treated biomass

	Untreated biomass	Acid-treated biomass
Calcium	0.62 mmol/g	1.05 mmol/g
Magnesium	1.10 mmol/g	0.36 mmol/g

Figure 3 06 Cr(III) speciation as predicted by MINEQL

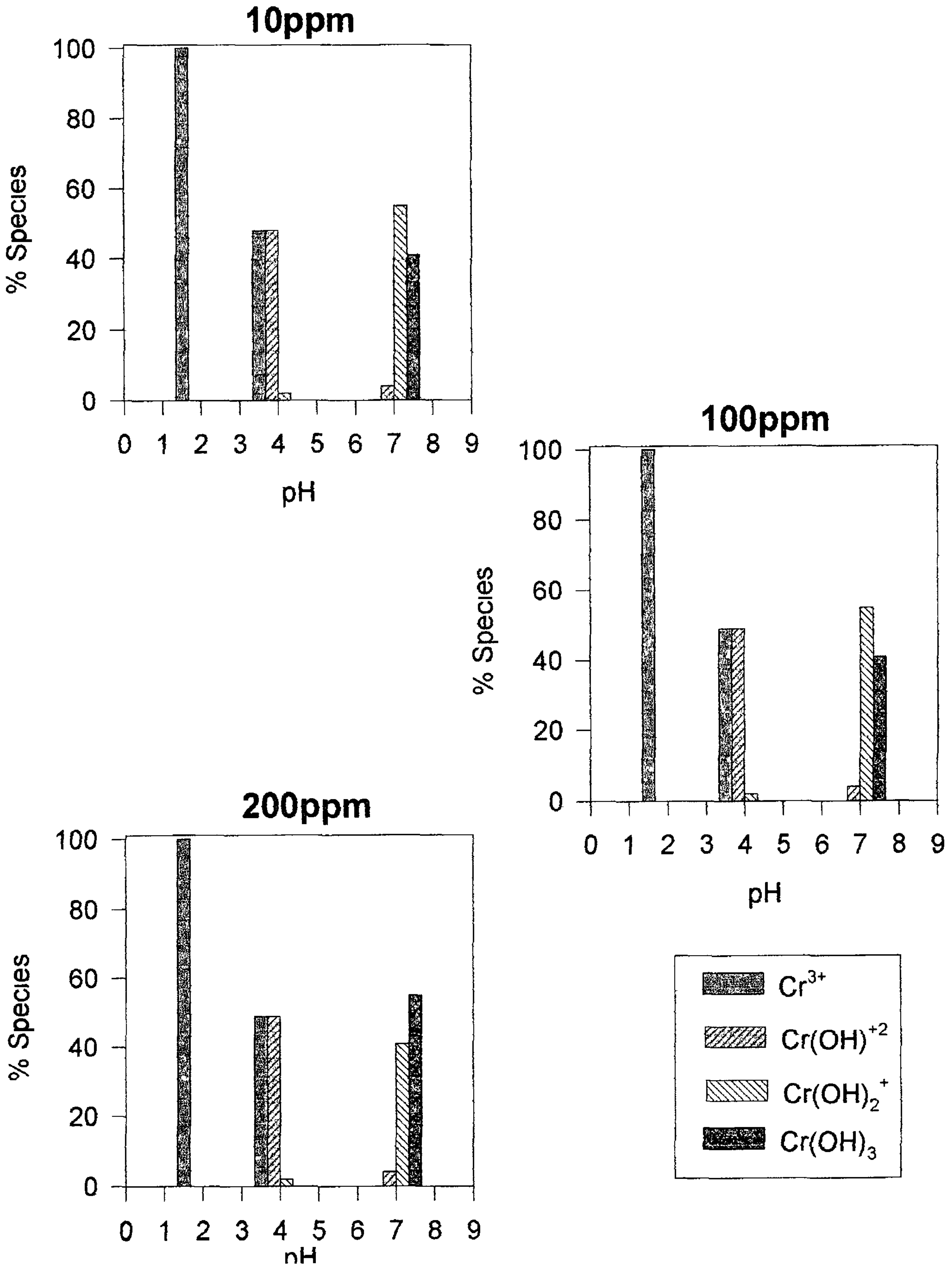


Figure 3 07 Speciation of Cr(VI) as predicted by MINEQL

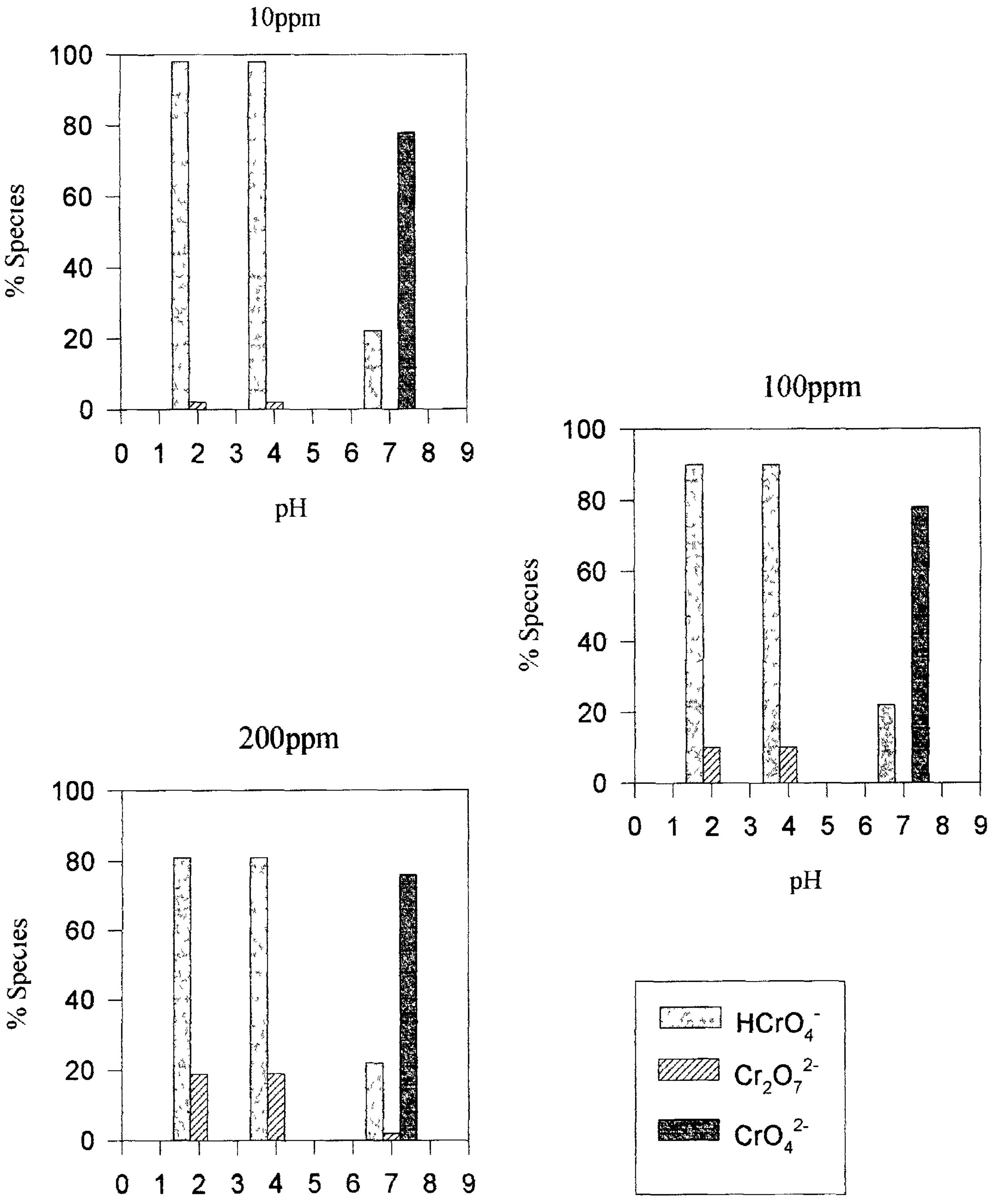


Figure 3 08 Uptake of Cr(III) by acid-treated kelp at pH 2 and 4

(Symbols ■ pH 4, ● pH 2)

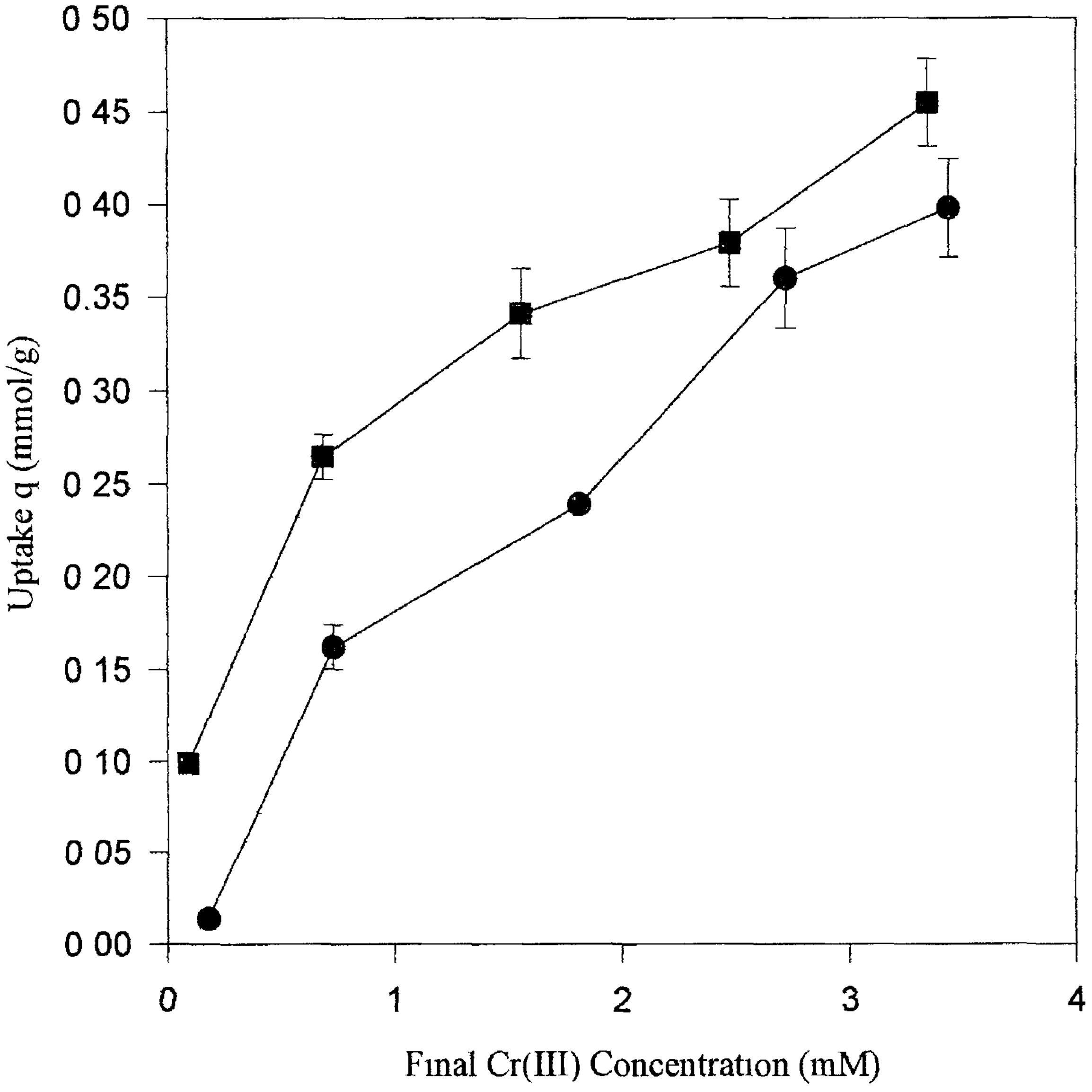


Figure 3 09 Uptake of Cr(III) by acid-treated peat

(Symbols ● pH 4, ○ pH 2)

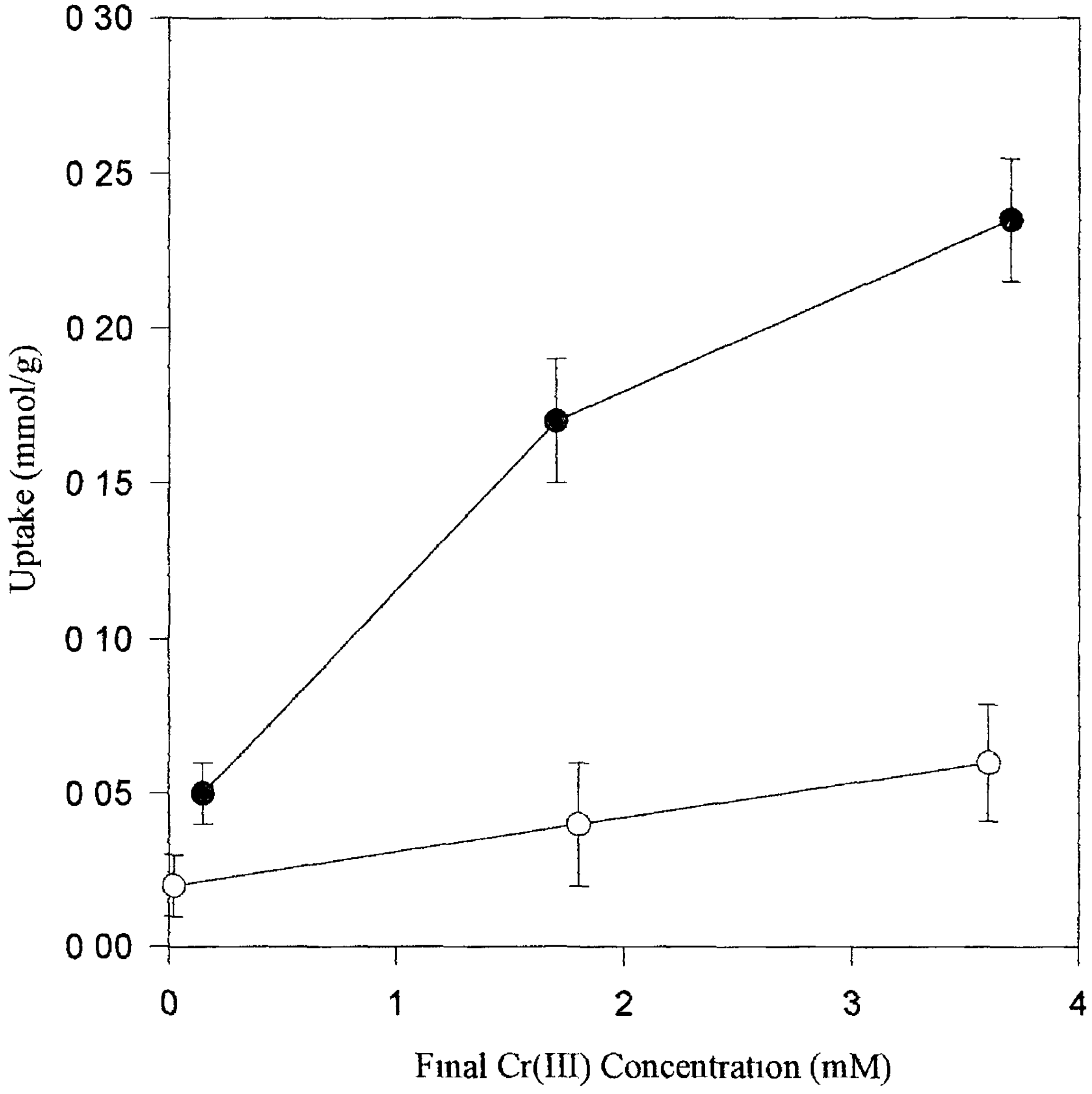


Figure 3 10 Uptake of Cr(III) by treated and untreated kelp at pH 4

(Symbols ● Untreated biomass, ■ Calcium-treated biomass,
▼ Acid-treated biomass)

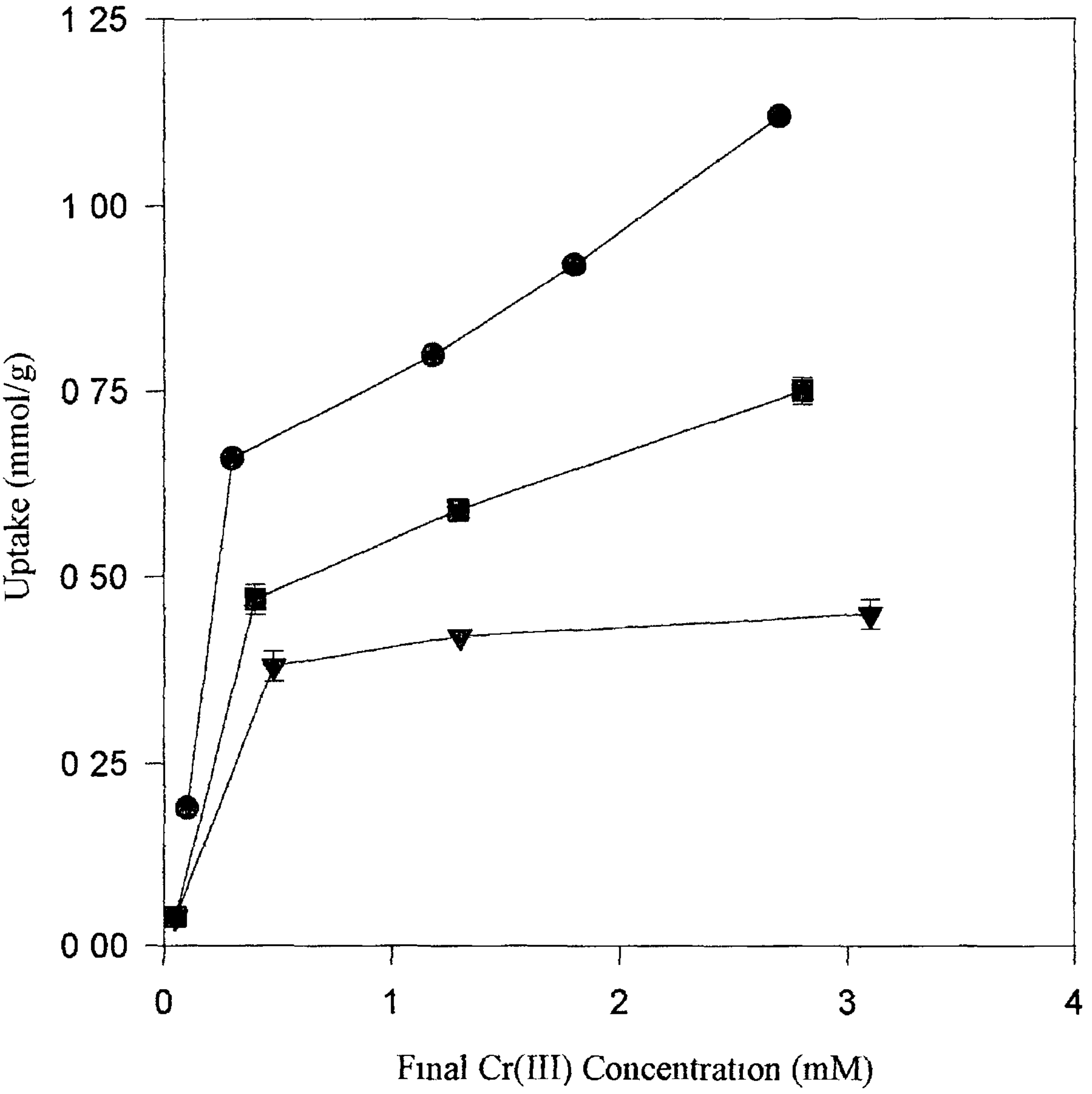


Figure 3 11 Uptake of chromate by acid-treated *L digitata*

(Symbols ● pH 2, ■ pH 4, ▼ pH 7)

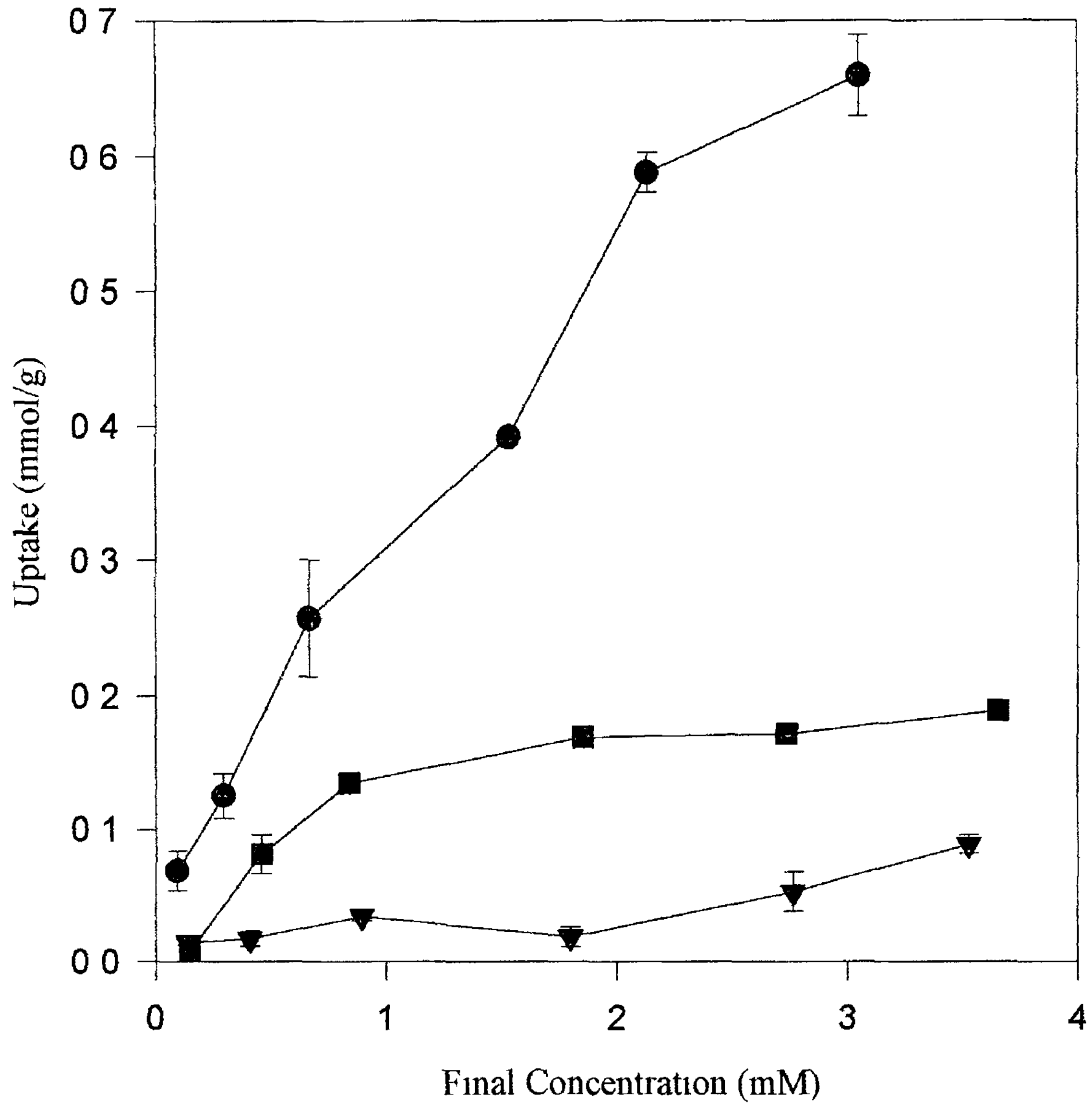
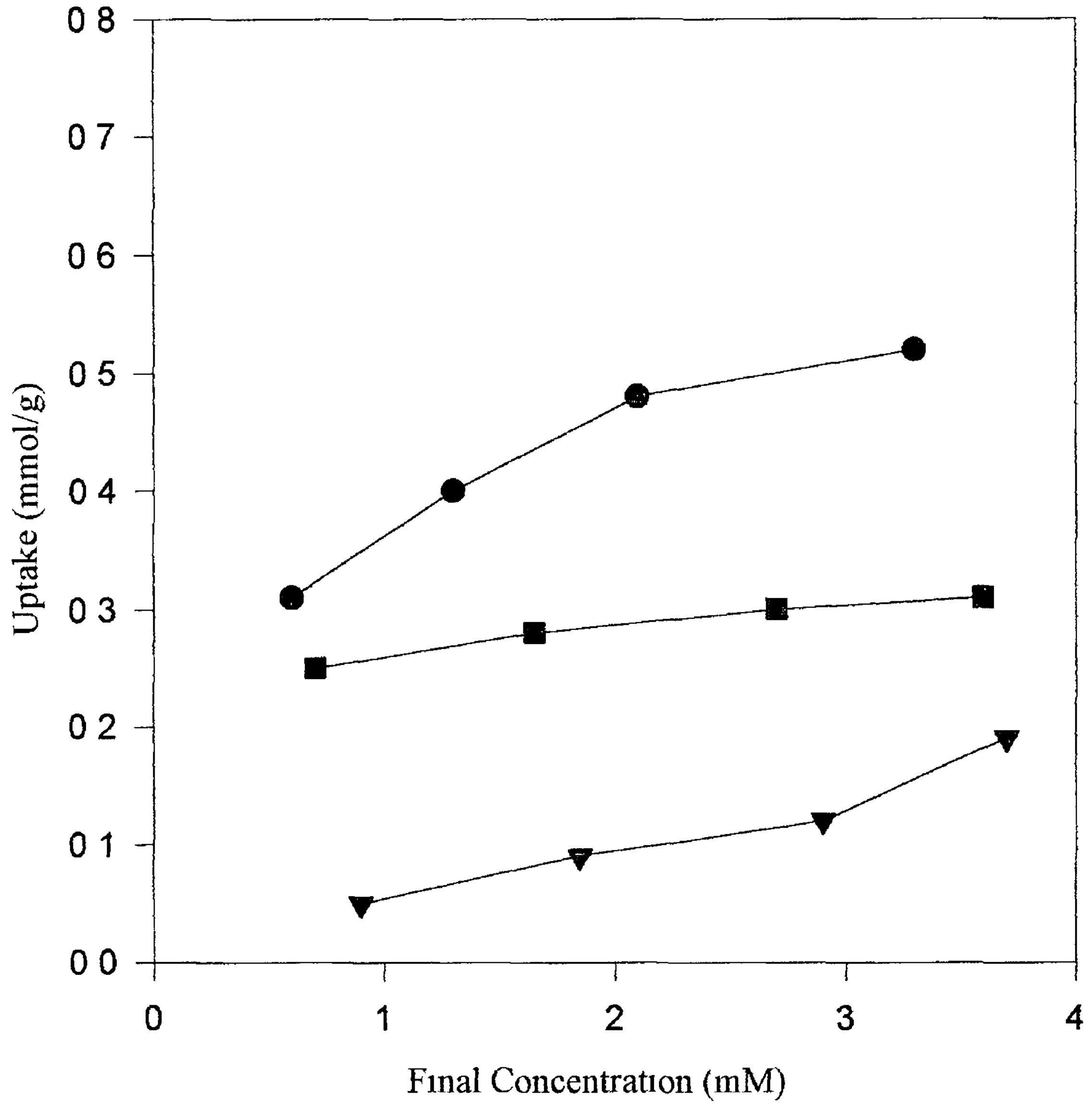


Figure 3 12 Uptake of chromate by acid-treated peat

(Symbols ● pH 2, ■ pH 4, ▼ pH 7)



3.07 The presence of counter-ions during biosorption

In the process of biosorption, uptake can occur by ion exchange This may involve hydrogen ions or other species already on the biomass *L digitata* is a marine macrophyte that, unlike many seaweeds, remains continuously submerged with the possible exception of the low spring tides Seawater is an ionic solution of dissolved substances including calcium, magnesium, potassium and sodium When *L digitata* was contacted with distilled water at pH 4, it releases ions until equilibrium is reached between the ions in solution and the ions on the biomass When *L digitata* was contacted with 100 ml of distilled water, sodium was released to a level of 1 8 mmol/g, calcium to 0 017 mmol/g, magnesium to 0 056 mmol/g and potassium to 0 079 mmol/g

Peat, under the same conditions, released 0 007 mmol/g magnesium and 0 004 mmol/g calcium

3 08 Uptake of chromium(VI) by metal-pretreated biomass

L digitata biomass was contacted with various concentrations of copper at pH 2 and pH 4 The biomass was then contacted with a range of concentrations of chromate The uptake of chromate was conducted at the same pH as the adsorption of copper to avoid metal displacement by pH effects

The uptake of copper by untreated kelp biomass at pH 4 reached a maximum value of 0 97 mmol/g from a 3 13 mM initial concentration At this pH, the maximum chromate uptake was 0 46 mmol/g At pH 4, the presence of copper decreased the uptake of chromate with increasing copper concentration (Figure 3 13a) The maximum chromate uptake was 0 29mmol/g with biomass treated with 0 15mmol copper (Table 3 06)

The optimum pH for chromate biosorption with untreated biomass was pH 2, where uptake was 0 82 mmol/g At pH 2, maximum copper uptake was 0 12 mmol/g from a 3 13 mmol initial solution Maximum adsorption of chromate by copper-treated kelp at pH 2 was 0 4 mmol/g (Figure 3 13b)

Table 3 06 Uptake of Cr(VI) by copper treated kelp

	Cr(VI) uptake untreated biomass	Cu uptake untreated biomass	Cr(VI) uptake Cu-treated biomass
pH 2	0 82 mmol/g	0 97 mmol/g	0 29 mmol/g
pH 4	0 46 mmol/g	0 12 mmol/g	0 40 mmol/g

Figure 3 13(a) Uptake of chromate by copper-treated kelp at pH 4

(Symbols ● 0 mM Cu, ■ 0 15 mM Cu, ▼ 1 5 mM Cu, ◆ 3 14 mM Cu)

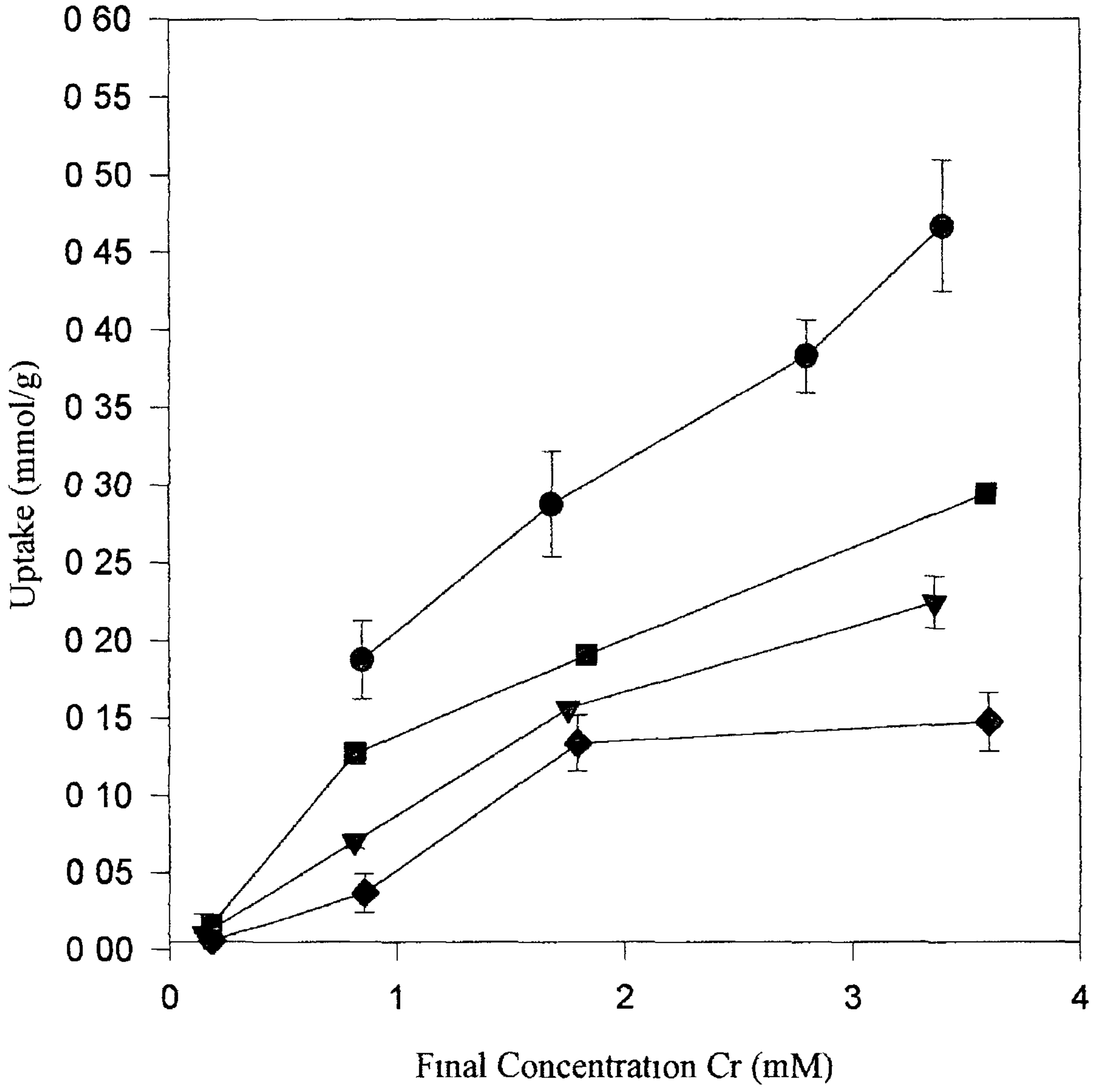
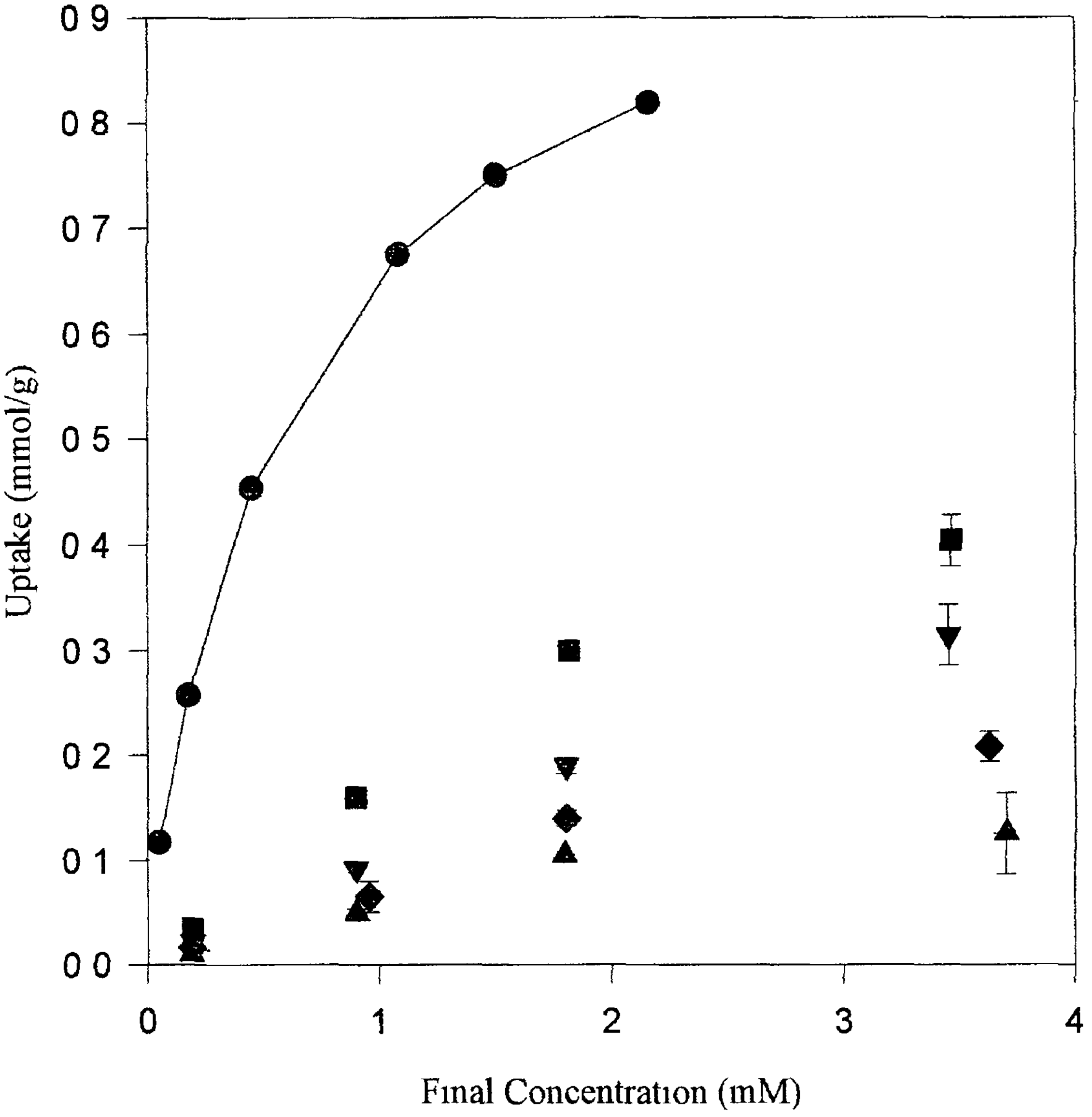


Figure 3 13(b) Uptake of chromate by copper treated kelp at pH 2

(Symbols ● 0 mM Cu, ■ 0 15 mM Cu,▼ 0 78 mM Cu,
◆ 1 5 mM Cu, ▲ 3 14 mM Cu)



3 09 Metal competition

Mixed metal solutions consisting of a range of copper and chromate concentrations were contacted with untreated kelp to investigate the order of uptake at pH 2 and pH 4 (Table 3 07)

Uptake of chromate at pH 2 (0 82 mmol/g untreated biomass) decreased with increasing copper concentration The maximum uptake was 0 25 mmol/g in the presence of 0 15 mM copper (Figure 3 14) Conversely, the presence of chromate had little effect on the uptake of copper at this pH

Copper uptake, by untreated biomass at pH 2, was 0 12 mmol/g In the presence of 0 19 mM chromate, copper adsorption reached a level of 0 085 mmol/g Lower chromate concentrations resulted in 0 045 mmol/g copper uptake (Figure 3 15)

Also at pH 4, the presence of chromate had little effect on the uptake of copper Maximum adsorption ranged from 0 93 mmol/g in the presence of 0 19 mM chromate to 0 78 mmol/g for 3 84 mM chromate This compares to the maximum value for the biosorption of copper by untreated kelp of 0 97 mmol/g (Figure 3 16)

The biosorption of chromate at pH 4 decreased with the presence of increasing copper concentrations The uptake ranged from 0 40 mmol/g for 0 15 mM copper to 0 17 mmol/g with 3 13 mM copper Maximum uptake of chromate at this pH by untreated biomass was 0 47 mmol/g (Figure 3 17)

Table 3 07 Competition of uptake for copper and chromium(VI)

pH 2	Cu conc	0 mM	0 15 mM	0 78 mM	1 5 mM	3 14 mM
	Cr(VI) uptake	0 82 mmol/g	0 25 mmol/g	0 20 mmol/g	0 14 mmol/g	0 08 mmol/g
pH 2	Cr(VI) conc	0 mM	0 19 Mm	0 96 mM	1 9 mM	3 84 mM
	Cu uptake	0 12 mmol/g	0 085 mmol/g	0 073 mmol/g	0 062 mmol/g	0 045 mmol/g
pH 4	Cu conc	0 mM	0 15 mM	0 78 mM	1 5 mM	3 14 mM
	Cr(VI) uptake	0 47 mmol/g	0 40 mmol/g	0 35 mmol/g	0 26 mmol/g	0 17 mmol/g
pH 4	Cr(VI) conc	0 mM	0 19 Mm	0 96 mM	1 9 mM	3 84 mM
	Cu uptake	0 97 mmol/g	0 93 mmol/g	0 86 mmol/g	0 80 mmol/g	0 78 mmol/g

Figure 3 14 Uptake of chromate from a mixed solution of copper and chromate by kelp at pH 2

(Symbols ● No Copper, ■ Cu 0 15 mM,▼ Cu 0 78 mM,
◆ Cu 1 5 mM, ▲ Cu 3 14 mM)

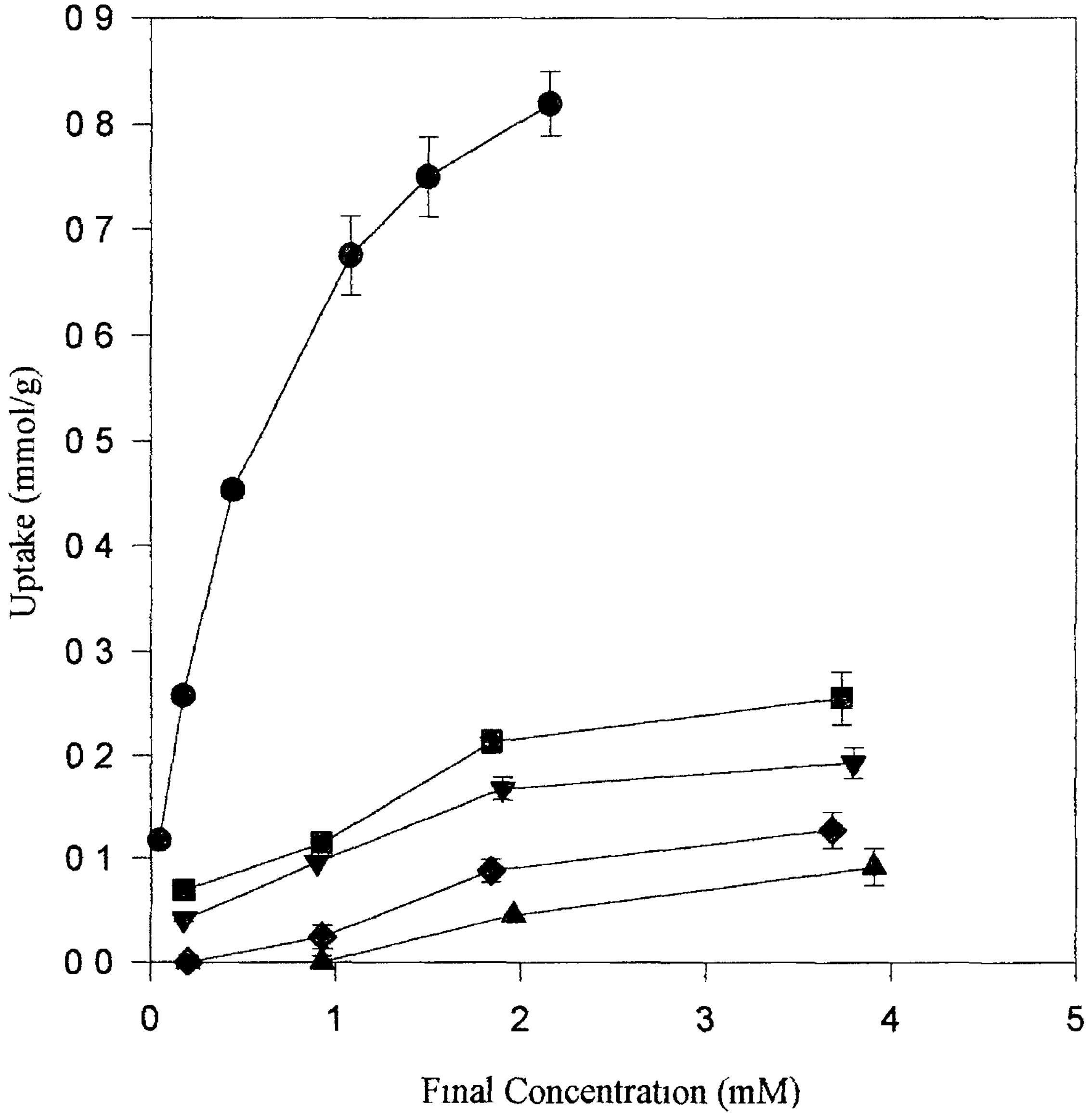


Figure 3 15 Uptake of copper from a mixed solution of copper and chromate by kelp at pH 2

(Symbols ● No chromate, ■ Cr 0 19 mM, ▼ Cr 0 98 mM,
◆ Cr 1 9 mM, ▲ Cr 3 84 mM)

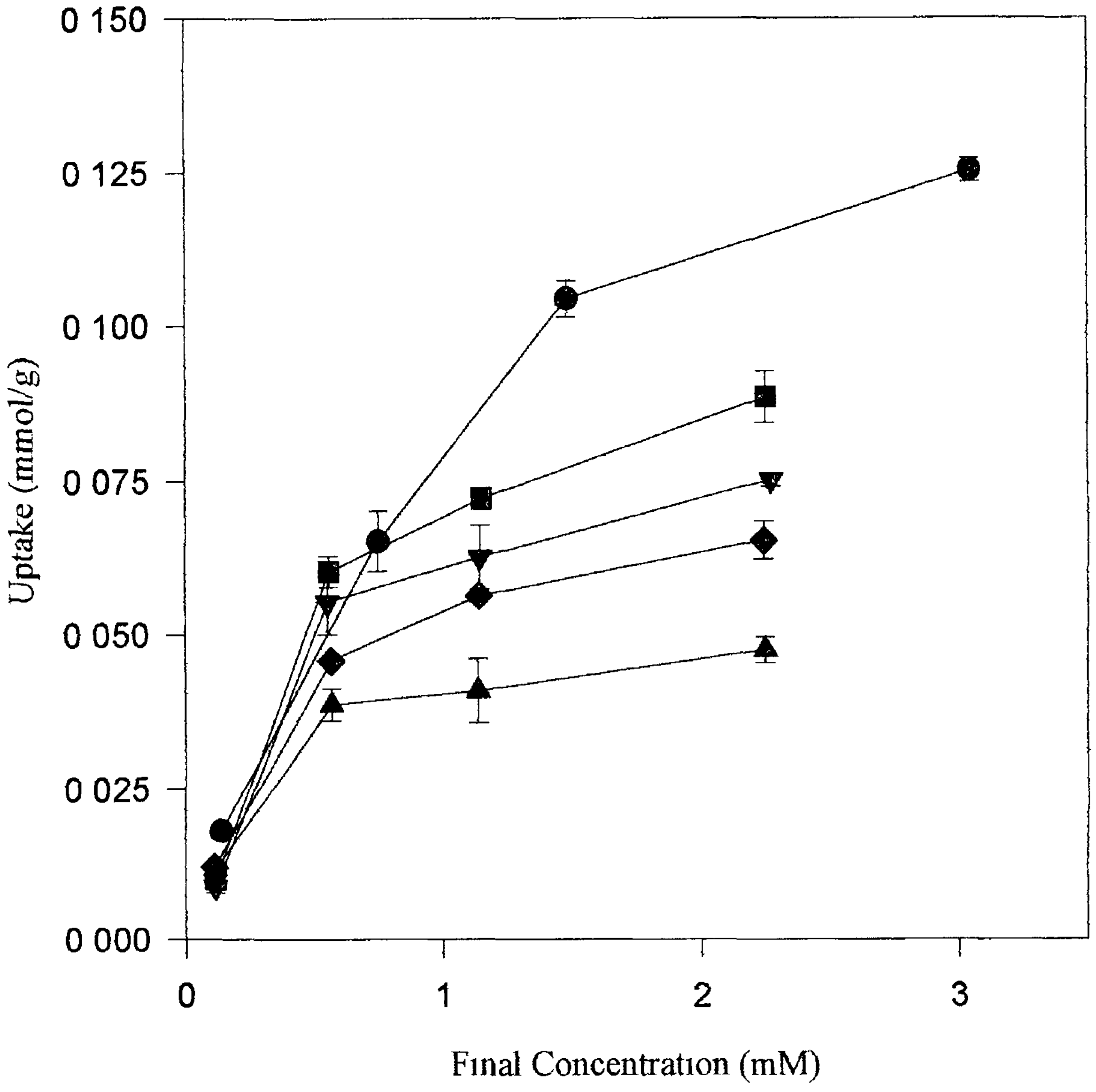


Figure 3 16 Uptake of copper by kelp at pH 4

Competition with Chromate

(Symbols ● No chromate, ● 0 19 mM Chromate, ▼ 0 98 mM Chromate,

◆ 1 9 mM Chromate, ■ 3 84 mM Chromate)

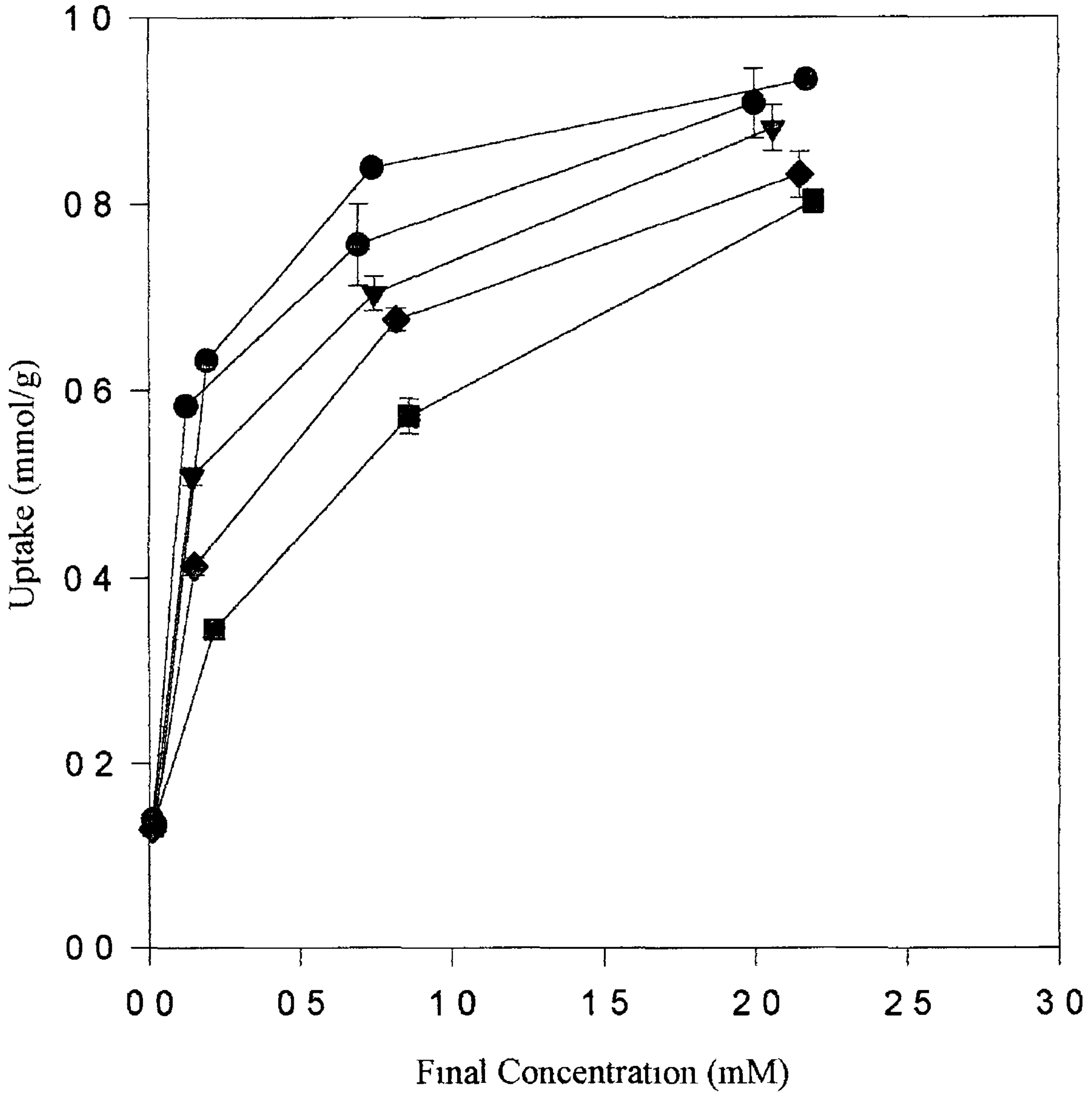
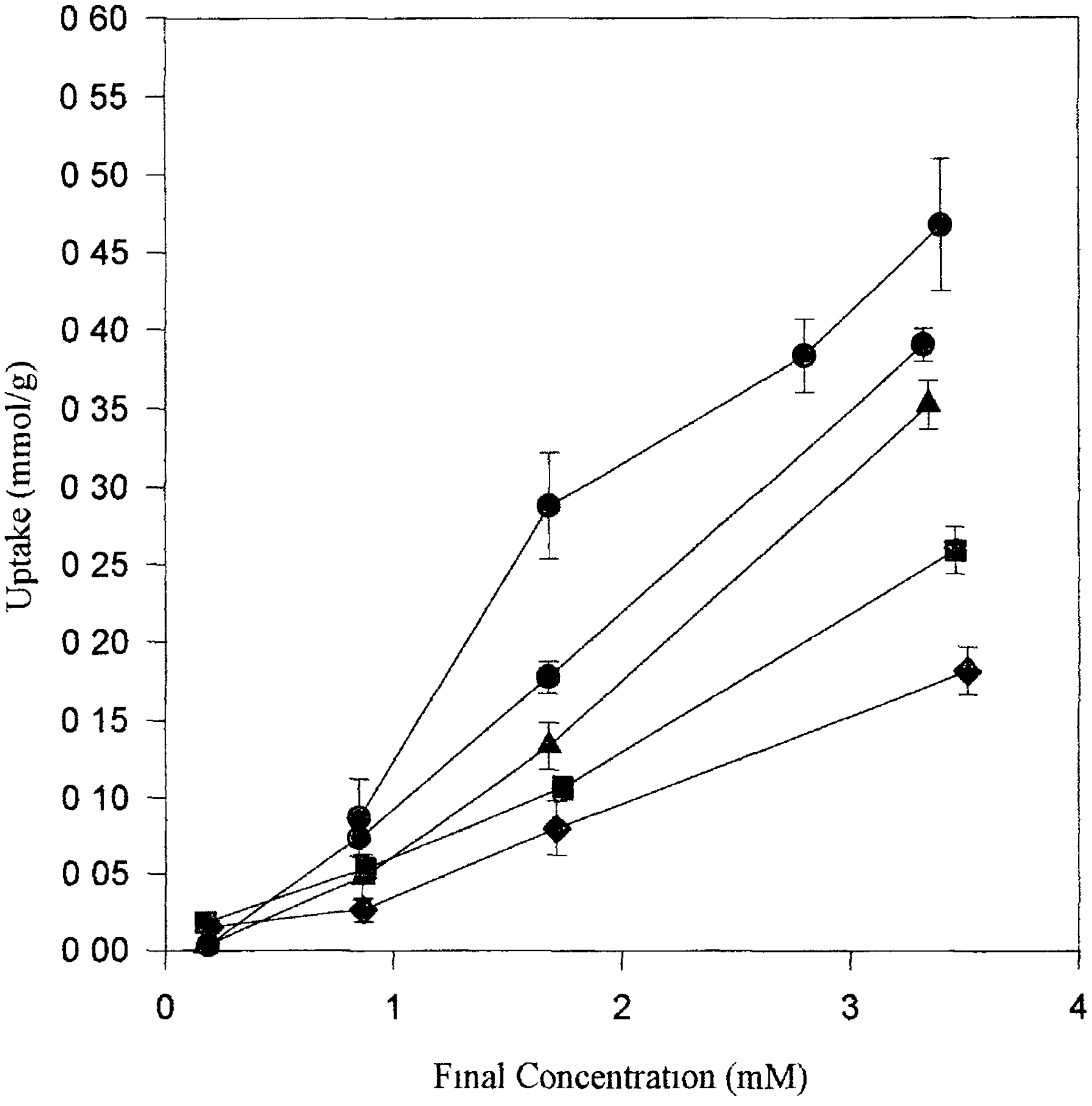


Figure 3 17 Uptake of chromate by kelp at pH 4

competition with copper

(Symbols ● 0 mM Cu, ● 0 15 mM Cu, ▲ 0 78 mM Cu,
■ 1 5 mM Cu, ◆ 3 14 mM Cu)



3 10 Competition with sodium

The presence of sodium inhibits the uptake of Cr(III) by kelp biomass The maximum uptake of chromium in the presence of sodium was 0 87 mmol/g for the lowest concentration of sodium at pH 4 This result was lower than uptake of Cr(III) by untreated biomass The biosorption decreased with increasing sodium concentration to 0 44 mmol/g for a solution containing 8 70 mM sodium The inhibitory effect is more pronounced at higher chromium and sodium concentrations (Figure 3 18)

3 11 Reduction of chromate at pH1 by *L digitata* and peat

At pH 1, 3 24 mM Cr(VI), from an initial concentration of 3 84 mM, was reduced to Cr(III) by contact with *L digitata* or peat biomass On visual inspection, the initial solution of only Cr(VI) was yellow while the final solution was blue indicating the presence of Cr(III) Reduction did not occur in biomass-free controls and so is dependent on the presence of kelp or peat The graph shows the same solutions measured for total chromium and for chromate for both biomass types (Figure 3 19) The initial solutions contained only hexavalent chromate whereas the final solutions contain predominantly Cr(III) after reduction Conditions at pH 1 did not favour biosorption because of competition with excess hydrogen ions (Table 3 08)

Figure 3 18 Cr(III) uptake in the presence of sodium

(Symbol ● 0 mM Na, ■ 0 43 mM Na, ▼ 2 17 mM Na,
◆ 4 35 mM Na, ▲ 8 70 mM Na)

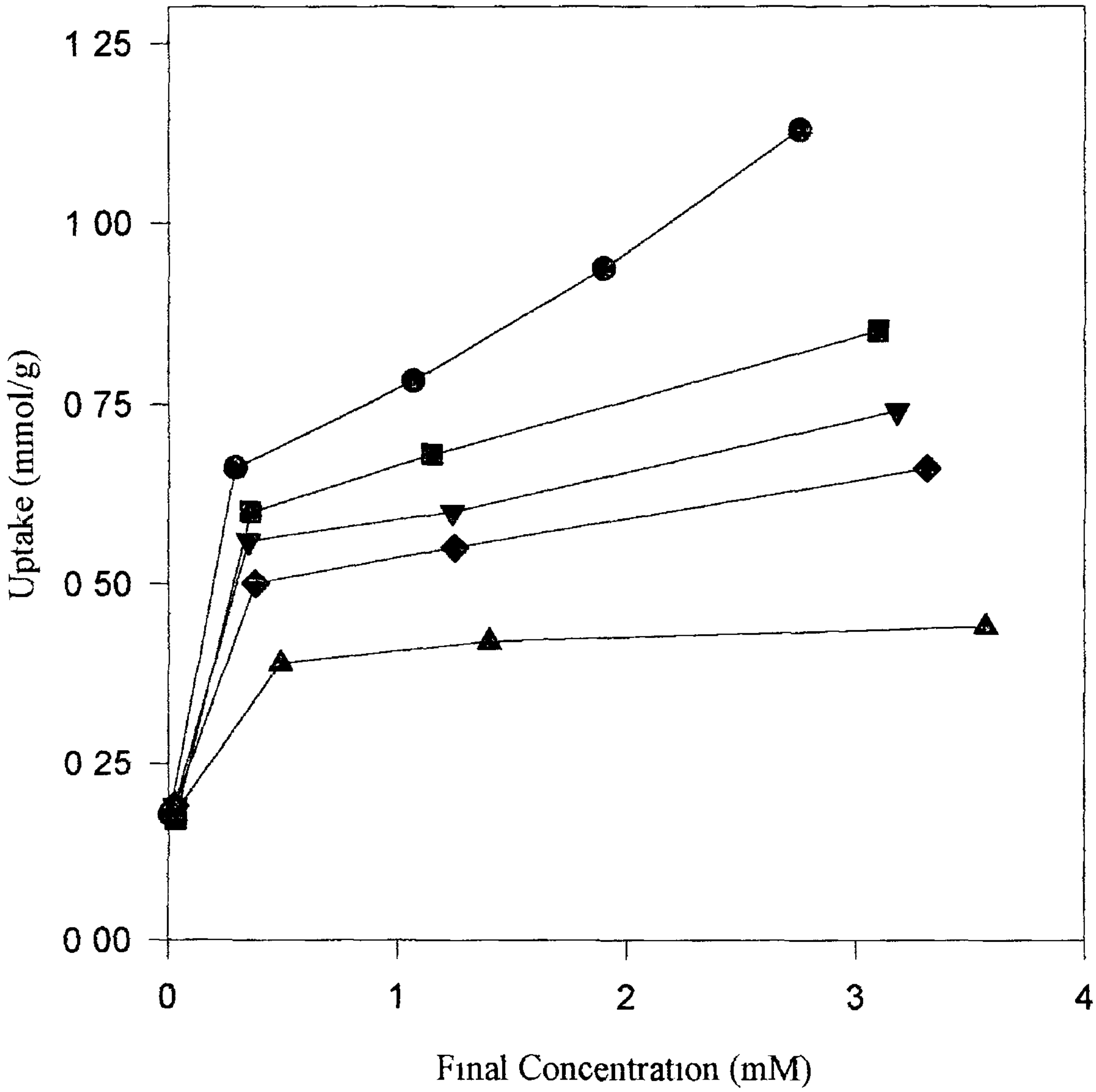


Figure 3 19 Chromate reduction at pH 1

(Symbols ● Kelp (chromate), ○ Total Cr Kelp,

■ Milled Peat (chromate), □ Total Cr Milled Peat)

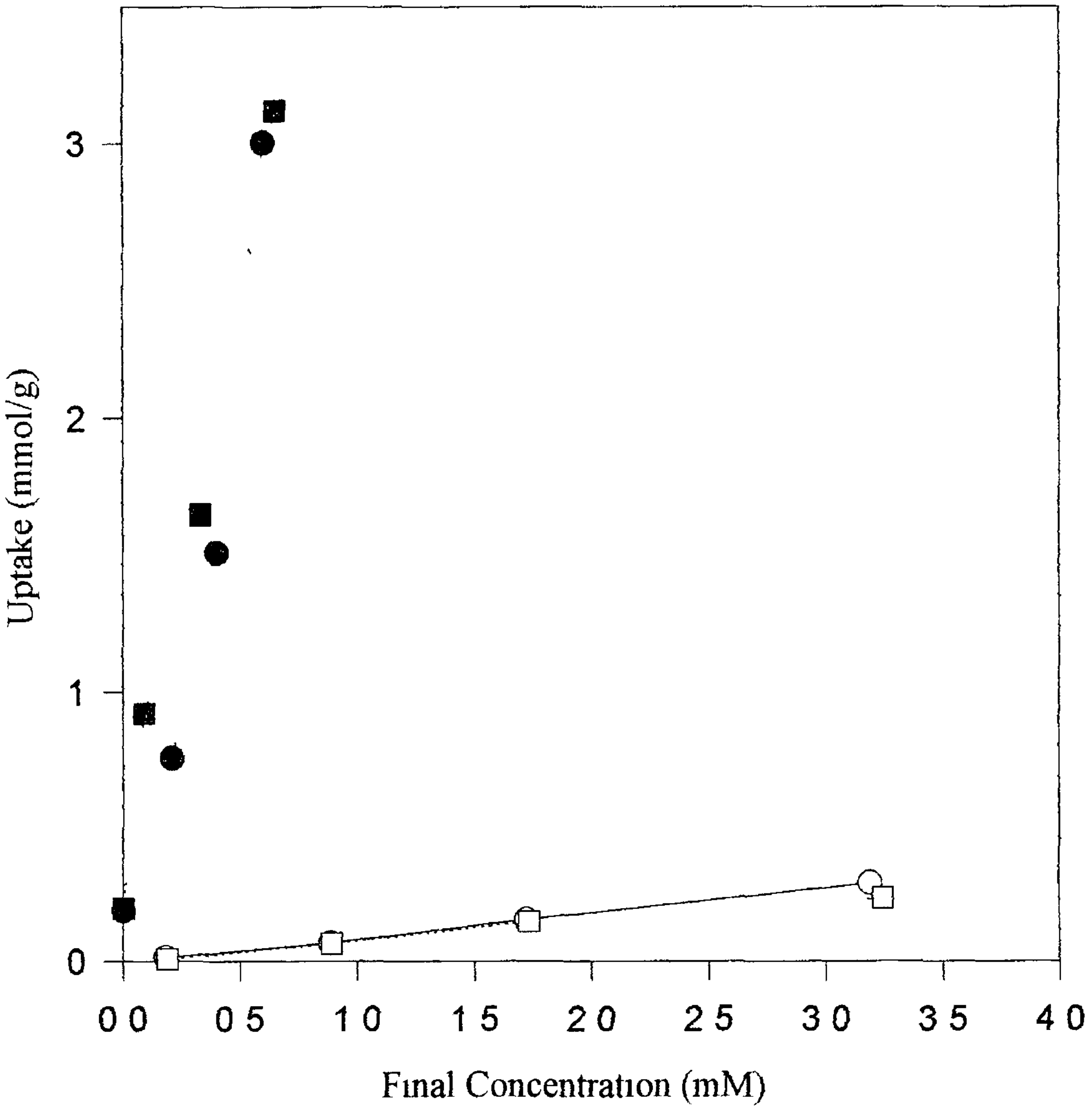


Table 3 08 Reduction of chromium(VI) at pH 1

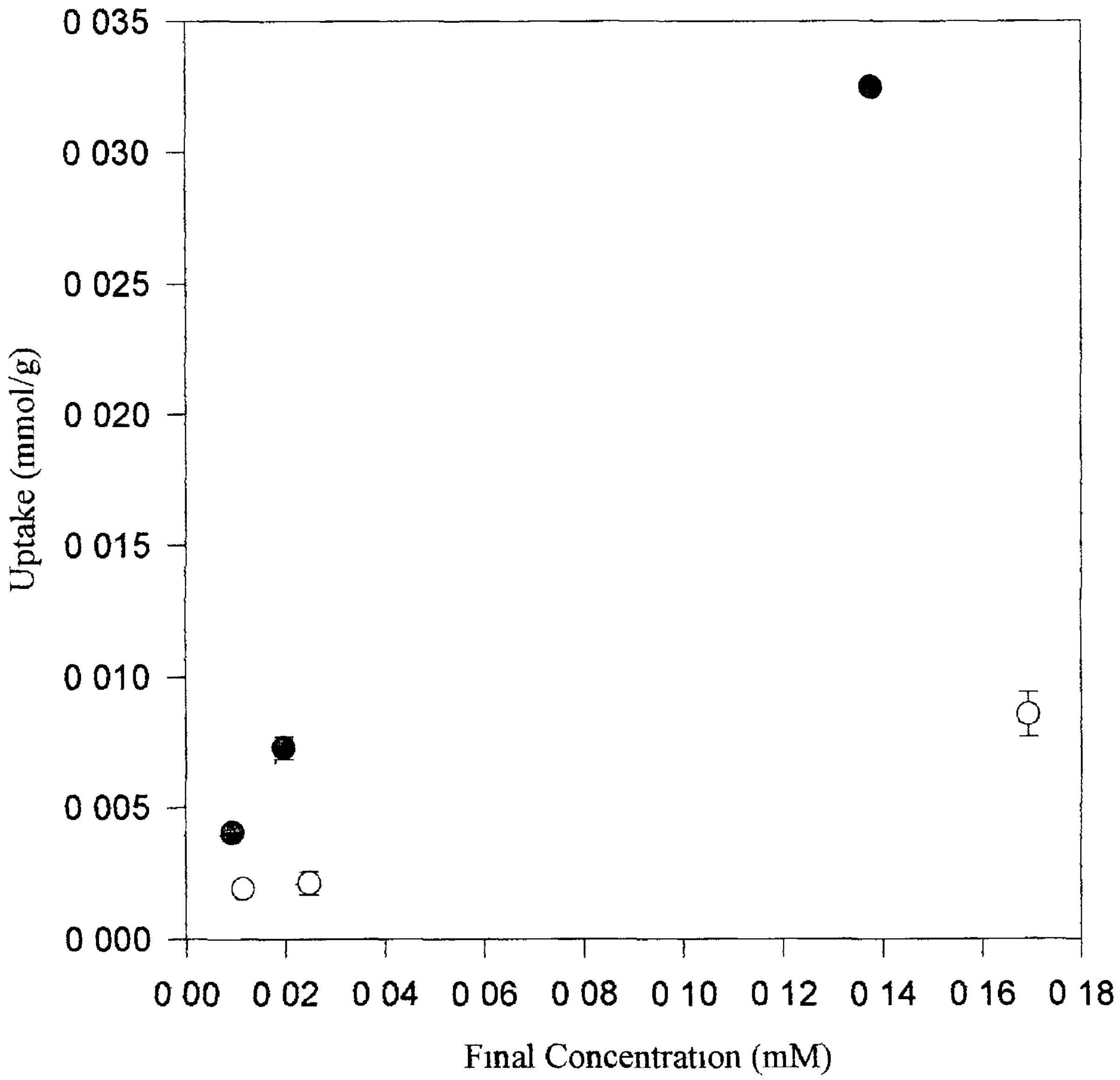
	Chromium (VI)	Chromium(III)
Initial concentration	3 84 mM	0 mM
Concentration after contact with kelp	0 60 mM	3 24 mM
Concentration after contact with kelp	0 65 mM	3 19 mM
Biomass–free control	3 84 mM	0 mM

3 12 Purification and uptake of chromium from tanning waste

After filtration, the pH of the effluent was pH 3 75 The initial concentration of chromium(III) in the tanning effluent was determined to be ca 117 mM (6100 ppm) The tanning effluent contained no chromium(VI) On gravity filtration and extraction with hexane, the chromium concentration in the aqueous layer did not change nor was there any chromium detected in the organic fraction Above pH 4 5, an insoluble precipitate settled out of solution The remaining clear solution contained 0 170 mmol (8 8 ppm) chromium On adjusting the pH to 3 75 again, the precipitate did not redissolve but remained as a gel in the bottom of the flask The sodium concentration of the filtered tanning waste was 16815 mM (386420 ppm) while the remaining solution after precipitation had a sodium concentration of 1502 mM (34516 ppm) Chromium uptake at pH 4 from the supernatant was 0 032 mmol/g while the sodium concentration was 925 5 mM in the remaining solution The kelp removed 577 3 mmol/g sodium from the original supernatant Corresponding results with peat showed less chromium uptake (0 008 mmol/g) and also lower sodium uptake (189 5 mmol/g) (Figure 3 20)

Figure 3 20 Uptake of Cr(III) from tanning waste

(Symbols, ● *L digitata*, ○ Peat)



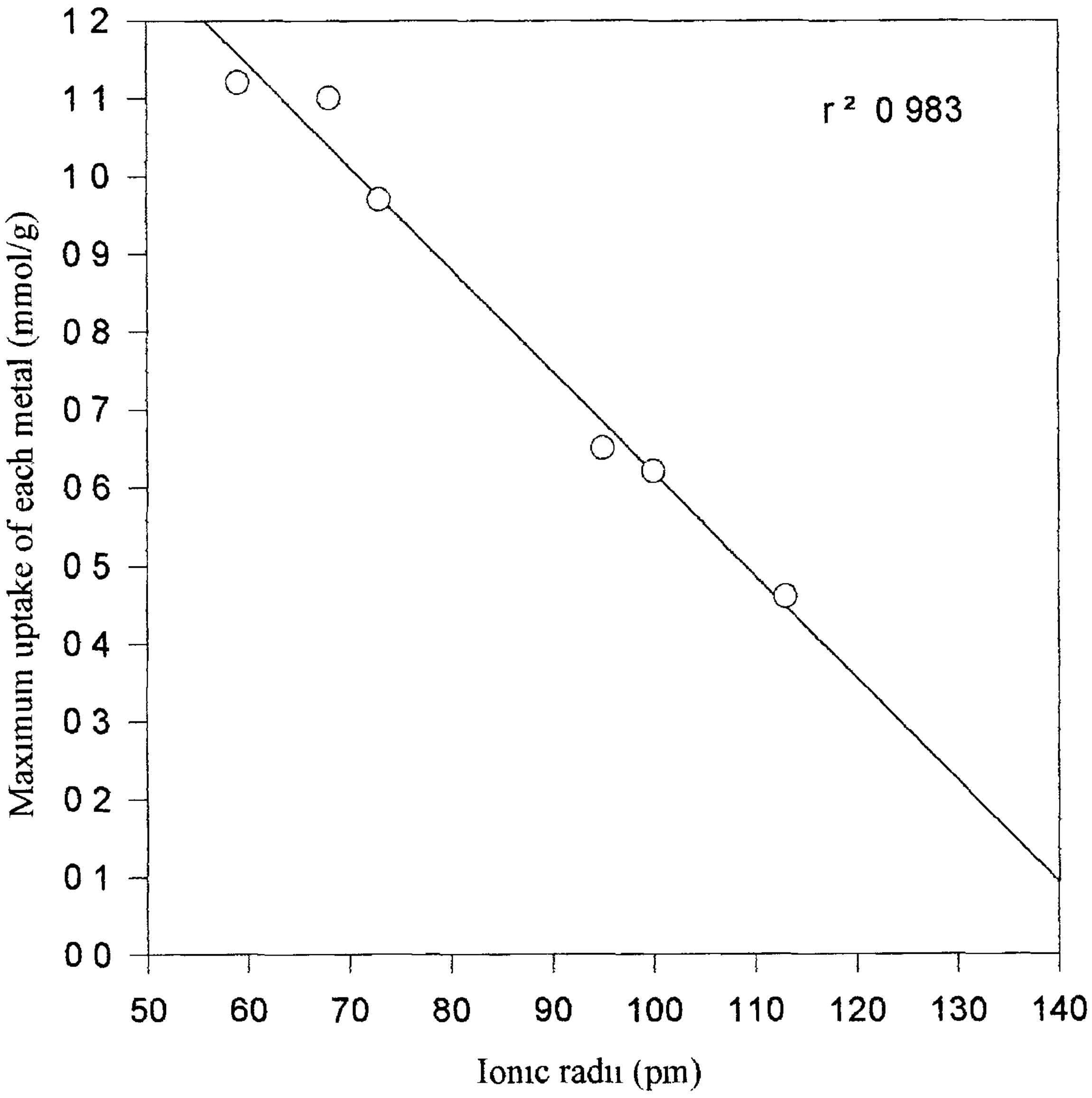
3 13 Metal uptake by kelp and ionic radii

Metal cations were adsorbed by *L digitata* in the order of increasing ionic radii i e the smallest ionic radius corresponded to the highest metal uptake at pH 4 (Table 3 09 and Figure 3 21) and pH 2 (Table 3 01 and 3 09)

Table 3 09 Ionic radii of cationic metals

Metal Cation	Ionic Radius (pm)	Uptake (mmol/g) pH 4
Magnesium	68 00	1 10 ± 0 04 mmol/g
Chromium	62 00	1 12 ± 0 05 mmol/g
Copper	73 00	0 97 ± 0 03 mmol/g
Cadmium	95 00	0 65 ± 0 03 mmol/g
Calcium	100 00	0 62 ± 0 03 mmol/g
Strontium	113 00	0 46 ± 0 02 mmol/g

Figure 3 21 The relationship between metal uptake by kelp and ionic radii



CHAPTER 4

DISCUSSION

CHAPTER 4: DISCUSSION

4 01 Uptake of metal cations

Untreated *L digitata* adsorbed cations at pH 4 in the order of maximum uptake, $Cr^{3+} > Mg^{2+} > Cu^{2+} > Cd^{2+} > Ca^{2+} > Sr^{2+}$, from 1 12 mmol/g for Cr^{3+} to 0 46 mmol/g for Sr^{2+} (see section 3 12) For each cation, the maximum uptake was higher than the corresponding values at pH 2, or adsorbed by milled peat, with the exception of magnesium (1 33 mmol/g compared to 1 10 mmol/g for peat) At pH 7, the metals precipitated out of solution *L digitata* adsorbed cations in order of increasing ionic radii, at pH 2 and pH 4 (Table 3 01), smallest first (Table 3 09) This may indicate the importance of steric effects in the binding of metals to kelp as the larger the ion, the less it binds This finding is in contrast with *R arrhizus* biomass, where uptake levels were in order of decreasing ionic radii (Tobin *et al*, 1984)

Peat adsorbed cations at pH 4 in the order of $Mg^{2+} > Cu^{2+} > Cr^{3+} > Ca^{2+} > Cd^{2+} > Sr^{2+}$ This follows no trend of ionic radii, electronegativity or charge Uptake of Mg was the highest (1 33 mmol/g) while all other cations were adsorbed to a level of 0 26 – 0 32 mmol/g) This pattern may be a consequence of the small mass of Mg compared to the other cations It is possible that mass may be a limiting factor in the biosorption of metal ions by peat

The difference in the order of maximum uptake of metal ions may be due to the individual nature of each biomass and the different types of binding sites on each Peat forms in an acidic environment, while kelp grows immersed in seawater – an ionic solution at approximately pH 8 The presence of excess H^+ on the peat may hinder cation uptake by this biomass, more so than by kelp, as H^+ will compete with metal cations for binding sites

Kelp, being native to a marine environment, has cations such as Ca^{2+} and Mg^{2+} already present in quantity on its surface while peat has substantially less (section 3 07) This may partially explain the high uptake of Mg^{2+} by milled peat, but uptake by kelp is also high The affinity of both biomass types for Mg^{2+} may be due to its small size and hence, its charge to mass ratio

Ca^{2+}, which would be expected to follow the same trend of high uptake by peat, does not This ion has a larger ionic radius than Mg^{2+} so, although peat may have a high affinity for Ca^{2+}, its size may cause steric effects and restrict binding

4 02 Biosorption of Cr(III)

Maximum uptake of Cr(III), 1 12 mmol/g biomass, by untreated *L digitata* occurred at pH 4 from a 3 84 mM (200 ppm) solution This uptake value greatly exceeds uptake of Cr(III) by milled peat to a level of 0 30 mmol/g under the same experimental conditions (Dean and Tobin, 1999) Biosorption of Cr(III) by a brown seaweed, *Sargassum*, at pH 4 attained a maximum value of 0 63 mmol/g for the same initial concentration (Kratochvil *et al* , 1998) while maximum adsorption by *Mucor* biomass was approximately 0 35 mmol/g (Tobin and Roux, 1998) *Rhizopus arrhizus* adsorbed 0 59 mmol/g of Cr(III) from the same concentration at a final pH of 3 7 (Tobin *et al* , 1984) while approximately 0 2 mmol/g uptake occurred at pH 4 by *Azolla filiculoides* (Zhao and Duncan, 1997)

Metal ions are adsorbed by brown seaweeds into the outer cell perimeters, lodging in the cell wall or extracellular polymers, which are mainly polysaccharide substances (Madgwick, 1994) The uptake of metals by members of the Phaeophyta is predominantly a consequence of the ion exchange behaviour of low solubility cell wall and cell-solid alginates (Madgwick, 1994)

Seaweed metal-binding capacities of *L japonica*, *S fluitans*, *A nodosum* and *F vesiculosis* were directly proportional to their respective total carboxyl group content and related to the electronegativity of the sorbed metal (Fourest and Volesky, 1996) The carboxyl groups of alginate play a major role in the complexation of lead and cadmium by *S fluitans* biomass Alginates are linear polysaccharides containing 1,4-linked β-D-mannuronic and α-L-guluronic acid residues The linear arrangement of the uronic acid residues varies among algal species (Fourest and Volesky, 1996) The surface functional groups of the brown algae are potentially superior binding sites for some metal ions compared to other biomass such as fungus and plants The different uptake capacities of members of the Phaeophyta may de due to the wide variation in cell wall composition of eukaryotic algae Only cellulose is common to all (Bold *et al*, 1980) Alginic acid is a major source of binding sites in brown seaweed There are many different forms of alginate, depending on the size and order of the mannuronic and guluronic fragment blocks This varies with species and may account for the preference in order and uptake ability of some seaweeds for particular metals (Table 4 01)

4.03 Uptake of Cr(VI)

In the present work, maximum uptake of Cr(VI) by *L digitata* occurred at pH 2, to a level of 0 82 mmol/g compared to 0 67 mmol/g by *Sargassum* as reported by Kratochvil *et al* 1998, under the same conditions Adsorption levels of chromate by kelp were appreciably in excess of uptake levels by milled peat (0 58 mmol/g) (Dean and Tobin, 1999) They also exceeded molybdate and vanadate uptake values for *Rhizopus arrhizus* (Tobin *et al*, 1984) where maximum uptake of 0 38 and 0 45 mmol/g respectively was reported at pH 4 5 Anion uptake by *R arrhizus* did not occur at pH 5 5, at which pH value it was suggested that the balance of positively and negatively charged species became unfavourable for anion binding (Tobin *et al*, 1984) In the present work, this effect was not observed although uptake diminished markedly at higher pHs At pH 4, the binding levels are significantly higher than corresponding values recently reported for chromate uptake by the water fern *Azolla filiculoides* (ca 0 34 mmol/g) at the same initial concentration (Zhao and Duncan, 1997)

Uptake of chromate by kelp may involve cell wall components, such as alginic acid or alginates, absent from peat and other biomass such as *Azolla* and *Rhizopus* This would explain the higher metal uptake by seaweed over these biomass The variation in alginate forms between species may account for the different uptake levels, of the same metal, by each seaweed

4.04 Cr(III) speciation predicted by MINEQL

As described in section 3 04, MINEQL predicted that, at pH 2, the predominant Cr(III) species present in solution was Cr^{3+} for all concentrations whereas at pH 4 approximately 49% is Cr^{3+} and 49% is $Cr(OH)^{2+}$ with less than 2% present as $Cr(OH)_2^+$ (Figure 3 06) $Cr(OH)_2^+$ is the main species at pH 7, at 55%, with 42% $Cr(OH)_3$ and less than 2% $Cr(OH)^{2+}$ In the case of 3 84 mM (200 ppm), there is 55% $Cr(OH)_3$ and 42% $Cr(OH)_2^+$ The increased Cr(III) uptake at pH 4, as compared to pH 2, reflects diminished H^+ competition for binding sites However, it is interesting that the altered speciation and decrease in Cr^{3+} concentration does not appear to affect uptake

4 05 Cr(VI) speciation predicted by MINEQL

The predominant species at both pH 2 and pH 4 is $HCrO_4$ but the concentration of $Cr_2O_7^{2}$ increases with increasing concentration from less than 2% to 19% At pH 7, CrO_4^{2} accounts for approximately 76% of total chromium while the remainder is primarily, $HCrO_4$

Biosorption of chromium(VI) is likely speciation-dependent as well as pH-dependent Uptake may be due to binding of $HCrO_4$ ions (the main species at pH 2 as predicted by MINEQL) to protonated sites on the kelp While chromium speciation at pH 4 is identical, lower but appreciable uptake of 0 46 mmol/g reflects the decrease in H^+ ions and consequent decrease in availability of protonated sites Lowest uptake occurred at pH 7 where the chromate anion is the predominant species This may represent steric hindrance to binding of this molecule as well as the expected exponential decrease in positively charged sites The trend of highest uptake at pH 2 and lowest at pH 7 is duplicated by milled peat

4 06 Uptake of Cr(III) by pretreated biomass

Acid-treated *L digitata* adsorbed 0 57 mmol/g at pH 4 compared with 0 62 mmol/g by *Sargassum fluitans* (Kratochvil *et al* , 1998) The kelp had been treated with 1M H_2SO_4 while 0 2M H_2SO_4 was used in the treatment of *S fluitans* When the treatment of *L digitata* involved 0 2 M acid, maximum uptake was 0 66 mmol/g indicating that adsorption may be dependent on the level of protonation

At both pH 2 and pH 4, the solution pH increased after contact with the biomass The trend of pH increase at pH 4 was inversely proportional to concentration with no pH change occurring at 3 8 mM initial concentration

Uptake of Cr(III) was higher with untreated *L digitata* than with calcium or acid-treated biomass Hence protonation and subsequent contact with calcium is deleterious to metal uptake This may be due to modification of the binding sites so that ion exchange cannot occur to the same extent Maximum uptake was 1 12 mmol/g by untreated compared to 0 75 mmol/g calcium-treated and 0 57 mmol/g acid-treated biomass These uptake levels exceed maximum values of 0 3 mmol/g Cr(III) using milled peat at pH 4 and 0 2 mmol/g *Azolla filiculoides* (Zhao and Duncan, 1997) Maximum uptake by *Rhizopus arrhizus* was 0 59 mmol/g at a final pH of 3 7 (Tobin *et al* , 1984) Maximum biosorption by Ca-treated kelp (0 75 mmol/g) was similar to the maximum uptake of Cr(III) by *Sargassum* biomass of 0 77 mmol/g (Kratochvil *et al* , 1998) Hence, it can be assumed that these pretreatments affect various species of brown algae in the same way, possibly due to a conformational change of binding sites or components common to all species

4.07 The presence of counter-ions during biosorption

The adsorption by peat of one divalent metal cation was coupled with the release of two H^+ ions (Bunzl *et al* 1976) All three ions, H^+, $Ca^{2+}$and Mg^{2+}, are involved in biosorption by *L digitata* This was illustrated by the highest uptake level of Cr(III), which was adsorbed by untreated kelp biomass, compared to calcium and acid-treated biomass The acid-treatment removed Ca^{2+} and Mg^{2+}, as evidenced by the release of low levels of these ions by acid-treated biomass This type of biomass adsorbed the lowest amount of chromium Metal uptake by acid-treated *S fluitans* was also less than the uptake by the untreated biomass (Schiewer and Volesky, 1997) Untreated kelp released higher levels of Ca^{2+} and Mg^{2+} ions than calcium or acid-treated biomass Ca-treated kelp released less calcium than untreated biomass This indicated that when ions were removed (including calcium) during the acid wash of the first stage in biomass preparation, they were replaced by a lesser amount of Ca^{2+} in the $Ca(OH)_2$ wash Ca-treated biomass released the least amount of magnesium ions indicating that most of the native magnesium was removed and not replaced during the treatment Although this lowered its biosorption potential, uptake of Cr(III) by Ca-treated biomass exceeded that of H-treated biomass This was most likely due to the presence of Ca^{2+} on the biomass and suggests that Ca^{2+} may be more important for ion exchange than H^+ and Mg^{2+} The brown seaweed, *Ecklonia maxima,* also demonstrated a corresponding calcium release on adsorption of nickel ions (Williams and Edyvean, 1997)

Vaucheria species of freshwater algae released Ca^{2+} and Mg^{2+} on adsorption of Sr^{2+} indicating that biosorption of alkali and alkaline-earth metals occurred by ion exchange and was based on electrostatic interactions Copper adsorption by this biomass also caused a release of H^+ ions and may have demonstrated additional covalent bonding for this transition metal (Crist *et al*, 1990)

4.08 Metal pretreatment of the biomass· copper

Biosorption of chromate by *L digitata* at pH 2 and 4 was reduced by the presence of previously bound copper on the biomass At the optimum pH for copper adsorption, pH 4, copper uptake reached a maximum value of 0 97 mmol/g Subsequent chromate adsorption was 0 32 mmol/g in comparison to 0 45 mmol/g uptake of chromate by untreated biomass at this pH The predominant chromate species at both pH 2 and pH 4 is the anion $HCrO_4$ Chromate uptake by untreated biomass is relatively low (0 45 mmol/g) at pH 4, because anion uptake is favoured at lower pH The chromate biosorption level of 0 32 mmol/g by copper-treated biomass shows that the presence of copper has a negative influence on uptake at this pH

Conversely, at pH 2, which is the optimum pH for the uptake of chromate (0 82 mmol/g), only 0 12 mmol/g of copper was adsorbed Corresponding chromate adsorption was 0 4 mmol/g, less than 50% of that taken up by untreated kelp This is probably due to steric hindrances caused by the already bound copper Copper and chromate should not compete for the same binding site because of the different respective charges The binding of copper may also cause a conformational change in the surface of the kelp resulting in loss of chromate binding sites (Table 3 06)

4.09 Metal competition

The presence of chromate had little effect on the uptake of copper at pH 2 and 4. Biosorption of copper by untreated kelp at pH 4 was 0.92 mmol/g. Uptake ranged between 0.8 mmol/g and 1.0 mmol/g in the presence of decreasing chromate concentration. At pH 2, maximum adsorption of copper was 0.13 mmol/g by untreated biomass compared to 0.14 mmol/g in the presence of chromate. This illustrates a preference of the biomass for copper. The copper cation is smaller than the chromate ion resulting in less steric effects to inhibit binding. Kelp predominantly contains anionic polysaccharides on its surface, providing cationic rather than anionic binding sites (Bold *et al.*, 1980).

The presence of copper was deleterious to chromate uptake. Maximum chromate uptake at pH 2 was 0.25 mmol/g in the presence of 0.15 mmol/g copper compared to 0.82 mmol/g by untreated biomass. At both pH 2 and 4, chromate uptake decreased with increasing copper concentration although less so at pH 4 where conditions are unfavourable for anion binding. As with previously bound copper, the presence of copper in solution may cause steric effects, in this case, by binding preferentially to the biomass (Table 3.07).

4.10 Competition with sodium

The presence of metal ions can influence biosorption by humic and fulvic acids as well as by alginate (Garnham *et al.*, 1991, Schiewer and Volesky, 1997). In the present work, the adsorption of Cr(III) by *L. digitata* biomass decreased with increasing concentration of sodium. The presence of sodium may balance the negatively charged binding sites on the surface of the kelp preventing the binding of chromium This effect was also discerned in the biosorption of zinc by *Sargassum fluitans* (Leusch *et al*, 1995) and in the uptake of cobalt by *Chlorella salina* (Garnham *et al*, 1991) where the presence of 0 5 M sodium reduced uptake by 60% and 1 M sodium inhibited biosorption altogether

4 11 Reduction of chromate at pH 1

Cr(VI) was reduced to the trivalent form on contact with both kelp and peat biomass at pH 1 The degree of reduction was greater with *L digitata* than with peat Reduction at pH 1 also occurred with *S fluitans* biomass (Kratochvil *et al*, 1998) This reduction has implications in the remediation of electroplating waste which is predominantly Cr(VI)

4.12 Purification and uptake of chromium from tanning waste

The biosorption of chromium from tanning waste was lower than the uptake of Cr from single-metal solutions of the same pH This is attributable to the high concentration of sodium (1500 mM) which inhibits metal uptake by seaweed biomass This finding confirms the results of the effect of sodium on uptake of chromium by kelp, from single-metal solutions and agrees with other reports (Leusch *et al*, 1995, Garnham *et al*, 1991) However, kelp removed high concentrations of sodium from the tanning solution Excess salinity in soils or freshwater systems presents a real environmental concern for agriculture and fisheries (Pierzynski *et al*, 1993) The removal of sodium by kelp was in contrast with work involving *Rhizopus arrhizus* biomass Sodium and the other alkali metals were not adsorbed by *R arrhizus*, as this group of metals does not form complexes with most ligands This agreed with the proposed mechanism of uptake by this biomass (Tobin *et al*, 1984) The biosorption of sodium by kelp may not involve complexes but ion exchange and may be dictated by its ionic radius (ca 102 pm, similar to that of calcium) Peat also adsorbed sodium from the tanning waste but to a lesser extent It may be that fungal biomass is unique in its binding sites and mechanisms and can be used as a selective biosorbent Kelp, on the other hand, may be more applicable to solutions containing a mix of contaminants

Table 4.01 Comparison of adsorption levels (mmol/g) by various biomass.

Biomass type	Cu^{2+}	Cd^{2+}	Sr^{2+}	Ca^{2+}	Mg^{2+}	Cr^{3+}	Cr^{6+}	Reference
L digitata	0 97	0 65	0 46	0 62	1 10	1 12	0 82$^{\backprime}$	Present work
Milled peat	0 36	0 27	0 21	0 29	1 33	0 30	0 58$^{>}$	Present work (also Dean and Tobin 1999)
E radiata	0 97	0 90$^{\Psi}$						Matheickal (1994)
FCAN$^{\circ}$	0 50*	0 39*						deCarvalho (1995)
FCAN2	0 90*	0 70*						Chong (1995)
R arrhizus	0 40	0 25	0 18					Brady (1994)
R arrhizus	0 25	0 27				0 59$^{\wedge}$		Tobin (1984)
Vaucheria sp			0 20*					Crist (1990)
S fluitans		0 91*				0 63	0 67$^{\prime}$	Fourest (1996)
Sargassum sp						0 76	0 76$^{>}$	Kratochvil (1998)
A nodosum	1 38$^{<}$	0 90$^{<}$						Leusch (1996)
F vesiculosis		0 57$^{\prime}$						Holan (1993)
Sphagnum	0 23							Ho (1994)
Mucor						0 35		Tobin (1998)
A filiculoides						0 20	0 34$^{\prime}$	Zhao (1997)

$^{\backprime}$ pH 2, $^{\prime}$ pH 3 5, $^{\wedge}$ pH 3 7, * pH 4 5, $^{\Psi}$ pH 5, ° formaldehyde cross-linked *Ascophyllum nodosum* Unless otherwise indicated, values are shown for pH 4

CHAPTER 5

CONCLUSIONS

Chapter 5 Conclusions

5 01 Concluding remarks

The kelp, *Laminaria digitata*, is a good biosorbent with considerable metal adsorbing abilities Uptake of both hexavalent and Cr(III) was higher than the corresponding biosorption levels achieved by a range of biomass types recently reported (Table 4 01) These high adsorption capacities show that this biomass has potential for use in industries such as electroplating and leather tanning, where the removal of Cr(III) can reduce the volume of toxic sludge to be disposed of The ability of kelp to reduce Cr(VI) to the less toxic Cr(III) can be utilised to replace $FeCl_2$ in conventional reduction processes When chromate is adsorbed onto kelp, the metal is reduced and the biomass is oxidised, thus removing and reducing the toxic chromate in one treatment Kelp can also be applied as a polishing treatment for wastewater and is particularly suitable for use with high volume, low concentration solutions

The application of kelp biosorption to the treatment of tanning effluent is, however, influenced by the high concentrations of sodium as well as chromium in solution This results in diminished chromium biosorption levels Nevertheless, high levels of sodium removal are obtained This has clear environmental implications as the disposal of high sodium effluent is an ongoing problem for many industries including leather tanneries

There is potential for the application of biosorption by kelp in the remediation of industrial metal-bearing wastewaters containing metal cations other than chromium

Uptake levels of copper, cadmium and strontium by kelp were among the highest achieved by any biomass reported (Table 4 01) The biosorption of copper is not notably inhibited by the presence of chromate, possibly as a result of the ionic radii difference between the cation and anion This may have potential in the remediation of mixed metal solutions such as electroplating wastewater

Kelp is a cheap and readily available source of biomass in most coastal areas but as of yet, is under-utilised Wild kelp forests can be managed and farmed or the seaweed can be grown artificially Depending on the characteristics of the spent biomass, kelp can be used as an agricultural fertiliser or incinerated as a fuel

Peat as a biosorbent was not as successful as kelp as it did not reach the same high uptake levels Biosorption of metal ions by peat was lower than uptake by other algal biomass (Table 4 01) Uptake levels were comparable, however, with fungal biomass such as *R arrhizus* and the plant biomass *Azolla filiculoides* Reduction of chromate by peat occurred to a lesser extent than with kelp

5.01 Direction for future work

For the successful application of biosorption to metal removal from wastewater, there is a need for the potential reuse of the biomass Reuse helps to reduce costs of materials, transport and disposal of spent biomass This is particularly important when using algal biomass such as kelp, as the biomass is oxidised on adsorption of the metal ions This oxidation can modify the surface components and may effect future biosorption performance The effects of this biomass oxidation need to be investigated as does the potential for reuse of the biosorbent over consecutive cycles

The ability to recover metals from spent kelp biomass can aid reuse and recycling of materials in industrial processes The quality and quantity of the recovered metals need to be determined before reuse so as not to compromise the final product standard If removal of the metals from the spent kelp is not viable, the characteristics of the spent biomass need to be investigated before deciding on the method of disposal

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