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Full description of copper uptake by algal biomass combining an equilibrium NICA model with a kinetic intraparticle diffusion driving force approach

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Abstract

In this work kinetic and equilibrium studies related to copper binding to the protonated macroalga *Sargassum muticum* are reported.. An intraparticle-diffusion linear driving force (LDF) model has been chosen for the quantitative description of the kinetics at several initial metal concentrations. Copper intraparticle homogeneous diffusion coefficient (D_h) obtained is in the range 0.2-0.9x10^{-10} $m^2 \cdot s^{-1}$. NICA isotherm is demonstrated to constitute a substantial improvement with respect to a simpler Langmuir competitive equation. The binding parameters were chosen to provide the best simultaneous description of the equilibrium experiments. Values of $\log \tilde{K}_{Cu}$ (4.3), n_{Cu} (1) and p (0.31) in NICA isotherm, and $\log K_{Cu}$ (3.5-5) in Langmuir competitive model, have been obtained. These parameters have been also used to predict the competition between copper and cadmium for binding sites. Two acids, HNO_3 and HCl, have been tested to evaluate their effectiveness to release copper from the metal-laden biomass.

Keywords: copper; *Sargassum muticum*; kinetics; LDF; equilibrium; NICA.

1. Introduction

The incidences of different pollution sources, that are harmful to the environment, have been gradually increased over time due to rapid urbanization and industrialization. Industrial activities have been recognized as a major contributor to a variety of water pollution problems. Moreover, their rapid growth has produced an increase in the volume and toxicity of residues, among which, liquid effluents containing metals are of special interest.

Metals have a high degree of toxicity, which can be harmful for human beings and the environment. According to EPA (Environmental Protection Agency), copper is an abundant trace element present in earth's crust and surface waters. Copper is not regarded as highly toxic, and only at elevated concentrations may become hazardous to some forms of aquatic life. However, treatment of wastewaters containing these metals is of importance both for environmental quality and for water reuse. The effluents from mining, leather, fabricated metal products, and electric equipment represent the most important sources of copper pollution. The European Pollutant Emission Register (EPER), which comprises 50 substances that have to be reported by industrial facilities if their emissions exceed certain threshold values, establishes for copper that the limits for air, water and land are, respectively, 0.1, 0.05 and 0.05 t. per year (Parliament and Council, 2006). As an example, these values are ten times higher than those for cadmium discharges.

Although various conventional methods, as ion-exchange, liquid extraction, precipitation, electrodialysis, etc., could be applied to treat wastewaters, most of these available physicochemical technologies are expensive or ineffective when they are applied to metal ions removal at low concentrations. Alternative low-cost technologies

are needed to reduce heavy metal concentrations in the environment to acceptable levels (Srivastava and Majumder, 2008).

Biosorption, the passive non-metabolically mediated process of metal binding by dead biomass, has a great potential to reach these objectives (Lodeiro et al., 2006; Volesky, 2003). Among the advantages offered by this technique are the high purity achieved by treated wastewaters or the use of inexpensive materials as biosorbents. Waste products from other industries or natural abundant biomass can be quoted as an example (Demirbas, 2008). The brown macroalga *Sargassum muticum*, an invasive species in Europe, has been the biosorbent employed in this work. Its native habitats are Japanese and Chinese waters, where its presence is much smaller than in the European coasts. This alga is an alien species that interferes with recreational use of waterways, blocking propellers and intakes. It is also a fouling organism in oyster beds and a nuisance to commercial fishermen.

The algal cell wall plays an important role in metal binding, due to its high content in polysaccharides with acid functional groups. In brown algae, the cell wall is mainly comprised of alginates, which usually constitute about 20-40% of the total dry weight, in addition to fucoidans (Davis et al., 2003). The carboxyl groups of alginates are likely to be the main functionalities involved in metal binding reactions because of their abundance with regard to both carboxyl and amine groups of the proteins.

The present work reports a study of Cu adsorption by non-living biomass of the brown marine macroalgae *Sargassum muticum*. The alga was previously protonated in order to increase the retention capacity of the raw biomass, and to achieve a further stabilization of the biomaterial. The process has been analysed through batch experiments with regard to the influence of initial metal concentration, pH and the presence of cadmium as competing cation. Both kinetic and equilibrium aspects have been discussed.

Desorption process has also been tested. Mathematical models for the quantitative description of the biosorption process have been employed in order to predict the dynamics and equilibrium behaviour.

2. Experimental Methods

Materials

Fresh samples of brown marine alga *Sargassum muticum* were collected from the coast of A Coruña (NW Spain). The samples were washed extensively with running and deionised water to removed adhering particles, and oven-dried at 60 ºC overnight.

Dried samples were then crushed with an analytical mill (IKA A 10) and sieved to a size range of 0.5-1 mm. After being sieved, the following pre-treatment was performed (Figueira et al., 2000). The biomass was protonated in 0.1 M HNO_3 (10 g of biomass/L) for 4 hours at room temperature, washed with deionised water, filtered and dried overnight at 60 ºC. This biomass was stored in polyethylene bottles until use.

Chemicals

Analytical grade $Cu(NO_3)_2 \cdot 3H_2O$, $NaNO_3$, HNO_3, $Cd(NO_3)_2 \cdot 4H_2O$ and HCl (Merck), NaOH (Panreac) were used in this work. Cellulose nitrate membrane filters were purchased from Whatman and Albet; N_2 C-55 (99.9995 %) was from Carburos Metálicos.

2.1 Methods

2.1.1 Kinetic studies

A copper ion selective electrode (ISECu, Radiometer Analytica) with a Ag|AgCl reference electrode, previously calibrated in copper concentration, was employed to analyse the copper evolution during the kinetic experiments. The experiments were monitored potentiometrically using a homemade program. This technique allows a great

126 number of experimental points to be obtained easily and quickly without need to
127 withdrawn solution for the measurements.
128 Experiments were carried out in a glass cell furnished with a thermostated jacket and a
129 nitrogen stream to remove dissolved O_2 and CO_2. The ionic strength was adjusted to
130 0.05 M with $NaNO_3$. All measurements were performed at least in duplicate.
131 *Effect of initial metal concentration*. Protonated *S. muticum* samples (0.25 g) were
132 added to 100 mL of Cu(II) solution at constant temperature (25.0 ± 0.1°C) and natural
133 pH (around 3). The initial concentration of tested copper solutions were 0.25, 0.50, 1.00
134 and 3.00 mmol·L^{-1}.
135 **2.1.2 Equilibrium studies**
136 All batch equilibrium studies were carried out in 100 mL Erlenmeyer flasks containing
137 0.1 g of the alga to which 40 mL of Cu(II) solutions were added. The mixtures were
138 agitated on a rotator shaker at 150 rpm for 3 hours, in order to ensure that equilibrium
139 was reached. The solution pH was adjusted by using HNO_3 or NaOH during the
140 equilibrium period; from these additions and the quantity of $Cu(NO_3)_2$ in solution, the
141 ionic strength was calculated for each sample. The experiments were performed at room
142 temperature. The algal biomass was removed by filtration through a 0.45 μm membrane
143 filter. In all equilibrium experiments the presence of organic matter was avoided by UV-
144 digestion of the aliquots for 75 minutes at 90°C (705 UV Digester, Metrohm);
145 afterwards aliquots were analyzed for Cu(II) released into solution by differential pulse
146 anodic stripping voltammetry (DPASV) using a 757 VA Computrace (Metrohm). All
147 batch experiments were carried out at least in duplicate.
148 *Influence of pH on metal biosorption*. The effect of solution pH was studied by using
149 eight Cu(II) solutions (40 mL, 2.22 mmol·L^{-1}) added to flasks containing 0.1 g of dry

biomass. The solution pH was adjusted to 1.5, 2.5, 3, 3.5, 4.0, 4.5, 5.0 and 5.5 by addition of HNO_3 or NaOH solutions.

Adsorption isotherms. 40 mL of nine Cu(II) solutions of several concentrations (0.1, 0.2, 0.5, 1.0, 1.5, 2.0, 2.5, 3.5 and 5.0 $mmol \cdot L^{-1}$) were placed in Erlenmeyer flasks containing 0.1 g of alga. The solution pH was adjusted to 4.0 by addition of NaOH solution.

Effect of Cd(II) ions on Cu(II) biosorption. The competition effect of cadmium ion was tested through batch sorption experiments carried out with 0.1 g of protonated *S. muticum* in contact with binary mixtures (40 mL) of several copper ion concentrations (the same used in adsorption isotherms) and the competitor metal ion at two different initial concentrations (0.1 and 5.0 $mmol \cdot L^{-1}$). The solution pH was adjusted to 4.0 by addition of NaOH solution.

Desorption experiments. For batch desorption experiments, copper-loaded *S. muticum* samples (obtained from previous adsorption process with 0.1 g of biomass, at a fixed pH of 4 and initial copper concentration of 2.5 $mmol \cdot L^{-1}$) were placed in 100 mL Erlenmeyer flasks and contacted with 10 mL of two different desorbents (HCl and HNO_3) at three different concentrations (0.05, 0.1 and 0.5 $mol \cdot L^{-1}$). The mixtures were agitated on a rotary shaker for 2 hours at room temperature.

From these experiments, the optimal eluent concentration was selected (0.05 $mol \cdot L^{-1}$) and then, the effect of contact time was studied. In this case, the volume of HCl or HNO_3 solutions was increased to 20 mL, in order to ensure better contact between algae and solution and to facilitate the samples extraction. Aliquots of 100 µL of solution were analyzed after 2 and 4 hours of contact.

3. Results and discussion

175 **3.1 Kinetic studies**

176 *3.1.1 Mathematical model*

177 Kinetic experiments are the first necessary stage in every biosorption study. They are
178 needed to determine the time required for sorption equilibrium to be reached and,
179 moreover, the resulting data may be used to extract kinetic parameters for the modelling
180 of column biosorption experiments. Several independent processes, including transport
181 phenomena and chemical reaction kinetics, which usually act in conjunction, determine
182 the dynamics of metal biosorption. In the case of porous sorbents, the following steps
183 may be present (Garcia-Reyes and Rangel-Mendez, 2010; Volesky, 2003): transport of
184 sorbate within the bulk solution (advection and diffusion), transfer of sorbate from bulk
185 solution to the sorbent surface through the boundary layer of fluid immediately adjacent
186 to the particle (external film diffusion), diffusion of sorbate within the particle
187 (intraparticle diffusion), and chemical reaction of the sorbate with the binding sites of
188 the biomass.

189 The experimental conditions and setup are very often chosen so that mass transfer
190 resistance, due to metal transport in the bulk solution, and film diffusion through the
191 boundary layer of biosorbent are minimized. In particular, an adequate mixing, created
192 by proper agitation, allows a fast transport of metal in bulk solution, and, at the same
193 time, it can contribute to suppress the boundary layer surrounding the particles. In these
194 cases, intraparticle diffusion and/or chemical reaction may be the rate-limiting steps in
195 the sorption kinetics. However, in biosorption of metal ions from aqueous solution it is
196 common to assume that the overall rate of uptake is controlled mainly by the diffusivity
197 of the sorbate within the particle, whereas the chemical reaction of binding with the
198 sorbent sites is assumed to be relatively faster.

In the present work an intraparticle-diffusion linear driving force model (LDF) was chosen for the quantitative description of the copper uptake kinetics. Further details about this model can be found in bibliography (Tien, 1994; Vilar et al., 2006). In summary, this model is based on the following assumptions:

-The rate of metal uptake is controlled by the homogeneous diffusion of the sorbed species within the biomass particles. The external film diffusion resistance is assumed negligible in the actual conditions selected for the experiments (agitation rate: 180 rpm, biomass particle size: 0.5-1 mm). Similar conditions were already tested in bibliography for *Sargassum* biomass (Yang and Volesky, 1999) and they were proved to ensure the exclusion of these kinetic limitations.

-The particles are modelled as homogeneous thin plates of thickness 2L. Therefore, the uni-dimensional diffusion of the sorbed metal ion along the direction normal to the particle surface determines the overall diffusion rate (Volesky, 2003).

-The equilibrium concentration of sorbed copper at the particle interface is described by the Langmuir isotherm (see section 3.2.1 below).

Based on the model assumptions, the mass balance equations for copper in the batch reactor are as follows:

$$V \cdot \frac{dC_{Cu,t}}{dt} + m_s \cdot \frac{dq_{Cu,t}}{dt} = 0 \qquad (1)$$

$$\frac{dq_{Cu,t}}{dt} = \frac{D_h}{L}\left(\frac{dq_{Cu,t,z}}{dz}\right)_{z=L} \qquad (2)$$

where $q_{Cu,t}$ represents the average metal concentration in the particle, $C_{Cu,t}$ the metal concentration in solution at any time t, and D_h is the intraparticle homogeneous diffusion coefficient of the sorbed species within the algae.

From the assumption that the sorbed copper concentration profile, $q_{Cu,t,z}$, is parabolic, the following expression (known as *linear driving force* approximation) can be derived

221 for the variation of the copper uptake with time (Tien, 1994; Vilar et al., 2006):

$$\frac{dy}{dt} = k \cdot \left(y_{eq} - y\right) \tag{3}$$

222 where $y = \frac{q_{Cu,t}}{Q_{\max,Cu}}$, $y_{eq} = \frac{Q_{Cu}}{Q_{\max,Cu}}$ and k is a rate constant; $Q_{\max,Cu}$ is the maximum copper

223 sorption capacity and Q_{Cu} is the equilibrium sorption capacity of the alga.

224 From the mass balance (Eq. 1) at every instant we get:

$$V \cdot C_{Cu,t} + m_s \cdot q_{Cu,t} = V \cdot C_{Cu,i} \tag{4}$$

225 Reordering this equation and introducing the definition of $x = \frac{C_{Cu,t}}{C_{Cu,i}}$, where, $C_{Cu,i}$ is the

226 initial copper concentration in solution, and $n_0 = \frac{V \cdot C_{Cu,i}}{m_s \cdot Q_{\max,Cu}}$ (the ratio between the

227 initial total amount of copper and the maximum quantity that can be sorbed in a mass,

228 m_s, of sorbent), the following expression is obtained:

$$y = n_0 \cdot (1 - x) \tag{5}$$

229 Substituting this equation in eq. 3 and using Langmuir isotherm, we obtain the

230 following expression:

$$\frac{dy}{dt} = k \cdot \left[1 - x - \frac{b \cdot C_{Cu,i} \cdot x}{n_0 \cdot \left(1 + \cdot b \cdot C_{Cu,i} \cdot x\right)}\right] \tag{6}$$

231 where b is the Langmuir affinity constant (see Eq. 7). The initial conditions are: t=0,

232 x=1, y=0. This ordinary differential equation was solved numerically using *ode45*

233 routine from Matlab (MATLAB® v.2008b, The Mathworks Inc.), which is based on the

234 Runge-Kutta algorithm.

235 Experimental kinetic data were fitted to this model by a least-squares minimization

236 procedure using *fminsearch* routine from Matlab. Only one parameter (k) was adjusted

237 to fit the four experimental data series simultaneously. The $Q_{\max,Cu}$ (0.32±0.02 $mmolg^{-1}$)

238 and *b* (0.9 ± 0.1 L·mmol^{-1}) parameters were taken from fitting results of equilibrium
239 isotherm data obtained at pH 3 using Langmuir model (r^2= 0.994).
240
241 *3.1.2 Copper kinetic sorption rate description*
242 Experimental data show that the equilibrium is attained within 1 hour, and no further
243 significant adsorption is noted beyond this period (Figure 1). In addition, it can be
244 observed that 50% of the total copper uptake occurred within 10 minutes, showing that
245 the rate of copper uptake is rather fast. It can also be observed that the percentage of
246 copper removed from solution diminishes as the initial copper concentration increases
247 (Table 1). The evolution of solution pH was also followed. As it is shown in Figure 2,
248 the pH was around 3 in all the kinetic experiments, and only a slight increase was
249 observed with time.
250 Figure 1 shows the copper kinetic data at four different initial metal concentrations and
251 their adjustment using the linear driving force model, as described above. As it is
252 denoted, this simple model could describe kinetic data accurately. The calculated
253 determination coefficients (r^2), used as an indication of model goodness, are showed in
254 Table 1.
255 The rate constant, k, regressed from the LDF model was 0.0642 min^{-1}. The validity of
256 this model is also demonstrated by the fact that the same rate constant is valid to
257 describe the experimental points for different initial metal concentrations. The rate
258 constant can be related with the intraparticle homogeneous diffusion coefficient, D_h, as:
259 $k= 3D_h/L^2$. Since L (half of the particle thickness) has an estimated average value
260 between 0.25 and 0.5 mm, then D_h must be in the range 0.2-0.9x10^{-10} m^2·s^{-1}. This
261 parameter is independent of concentration and, as expected, much lower than the
262 molecular diffusion coefficient for Cu in water (7.2x10^{-10} m^2·s^{-1}) (Heyrovsky and Kuta,

263 1966). The fact that *S.muticum* seaweed material is a porous gel-like particle makes D_h
264 for Cu to be about ten times lower than the corresponding diffusion coefficient in pure
265 water or aqueous salt solutions.
266
267 **3.2 Equilibrium studies.**
268 *3.2.1 Mathematical models*
269 The development of a technology based on biosorption implies the use of adequate
270 models for the metal ion binding to biomaterials. These models can be employed to
271 analyse equilibrium data and to compare quantitatively different biosorbents under
272 several conditions. Ideally, they would constitute a useful tool to predict the metal
273 biosorption, to deduce the binding mechanism and to determine the influence on
274 biosorption of variables such as pH, ionic strength or the presence of competing species
275 (Lodeiro et al., 2006).
276 The most common isotherms used in sorption studies, Langmuir-Freundlich (Eq. 7) and
277 Langmuir (Eq. 7 with n'=1), are able to accuracy reproduce equilibrium experimental
278 data if environmental parameters, such as pH, are controlled carefully during
279 experiments (Carro et al., 2009; Lodeiro et al., 2004).

$$Q_{Cu} = \frac{Q_{\max,Cu}\left(b\,C_{Cu}\right)^{1/n'}}{1+\left(b\,C_{Cu}\right)^{1/n'}} \qquad (7)$$

280 where b represents the affinity for the sorbate, which can be used to compare the
281 adsorption performance, n' is an empirical parameter that varies with the degree of
282 heterogeneity, and C_{Cu} is the copper concentration in solution at equilibrium.
283 In order to account for stoichiometry and pH effects, a modified competitive Langmuir
284 sorption model, Eq. 8, was proposed by Schiewer *et al.* (Schiewer and Wong, 1999).

285 The metal binding at equilibrium is described as a function of pH and free metal ion
286 concentration in solution.

$$Q_{Cu} = n\,Q_{\max,H}\,\frac{(K_{Cu}\,C_{Cu})^n}{1 + K_H C_H + (K_{Cu}\,C_{Cu})^n} \qquad (8)$$

287 where K_{Cu} and K_H are the equilibrium constants for the binding of copper and protons,
288 respectively; C_H is the proton concentration in solution, the parameter n defines the
289 stoichiometry ratio, 1:1 (n=1) or 1:2 (n=0.5), and $Q_{max,H}$ is the maximum binding
290 capacity for protons, which has been calculated from the equivalence point of the acid-
291 base titrations in absence of heavy metal.
292 However, another aspects that characterize algal biomass, such as chemical
293 heterogeneity, polyelectrolytic effects and conformational changes (Buffle, 1988), are
294 not considered by these equations. As a consequence, new models with additional
295 parameters, that reflect the complexity of the system, would be required.
296 The NICA model, developed by Kinninburg (Kinniburgh et al., 1999) for the
297 description of metal adsorption in heterogeneous materials, addresses to binding site
298 heterogeneity, interactions between ionic species and reaction stoichiometry. It is a
299 semi-empirical, competitive, non-ideal and thermodynamically consistent model, whose
300 application is fairly simple. This model, which is able to describe different types of
301 experiments (acid-base titrations, influence of pH on biosorption, metal sorption and
302 competition with other metals in solution), constitutes a powerful tool to describe
303 biosorption processes with both great accuracy and relatively small number of
304 parameters (see below) (Herrero et al., 2006; Lamelas et al., 2005; Pagnanelli et al.,
305 2005). However, despite the obtained encouraging results, the knowledge of the
306 geometric parameters that determine the electrostatic description of the system would be
307 required in order to derive the intrinsic binding parameters (i.e., independent of the bulk

ionic strength) (Li and Englezos, 2005; Rey-Castro et al., 2004; Rey-Castro et al., 2003).

The basic NICA equation for the overall binding of species *i* in the competitive situation is:

$$\theta_i = \frac{\left(\tilde{K}_i c_i\right)^{n_i}}{\sum_i \left(\tilde{K}_i c_i\right)^{n_i}} \times \frac{\left[\sum_i \left(\tilde{K}_i c_i\right)^{n_i}\right]^p}{1+\left[\sum_i \left(\tilde{K}_i c_i\right)^{n_i}\right]^p} \tag{9}$$

where θ_i is the coverage fraction of the species *i*, $\tilde{K}_i$ is the median value of the affinity distribution for species *i*, *p* is the width of the distribution (usually interpreted as a generic or intrinsic heterogeneity seen by all ions), n_i is an ion-specific non-ideal term and c_i is the local concentration of species *i* at the binding site.

The following normalization condition is used to calculate the amount of species *i* bound, Q_i:

$$Q_i = \theta_i \left(n_i / n_H \right) Q_{\max,H} \tag{10}$$

The ratio n_i/n_H has been interpreted by Kinniburgh *et al.* (Kinniburgh et al., 1999) in terms of stoichiometry and cooperativity. When this ratio is less than one, then the maximum binding of species *i* is lower than the total amount of sites (defined as the amount of titratable protons), which would be a consequence of certain degree of multidentism. On the other hand, a value of n_i/n_H greater than one would reflect some degree of cooperativity. Finally, if $n_i/n_H=1$, it can be demonstrated that the maximum proton/metal exchange ratio is one.

If only the proton binding is considered (i.e., absence of competing ions), Equations 9 and 10 simplify to the Langmuir-Freundlich (LF) isotherm:

$$Q_H = Q_{\max,H} \frac{\left(\tilde{K}_H c_H\right)^{m_H}}{1+\left(\tilde{K}_H c_H\right)^{m_H}} \quad (11)$$

where this time the heterogeneity parameter m_H describes the combined effect of n_H and p ($m_H = n_H \cdot p$). In the case of a homogeneous system (where all the binding sites behave as independent, chemically equivalent sites) n_i and p are 1, and then the mono or multicomponent Langmuir isotherms are obtained.

The development of a physico-chemical model to describe metal ion binding to seaweed biomass needs a previous description of proton binding as a function of pH in 1:1 electrolytes. Rey-Castro et al. (Rey-Castro et al., 2003) reported proton binding data from potentiometric titrations of biomass from different seaweed species (*Sargassum*, *Cystoseira* and *Saccorhiza sp.*). Moreover, in a previous work, Lodeiro *et al.* (Lodeiro et al., 2005) obtained the maximum amount of acid functional groups of the algae *Sargassum muticum* (2.61 mmol·g^{-1}), through acid-base titrations of protonated biomass samples. The fit of the proton binding data to an isotherm model allowed the estimation of an average acid constant K_H, referred to $NaNO_3$ 0.05 mol·L^{-1}, with a value of $10^{3.8}$, and the heterogeneity parameter, 0.54, calculated from least-squares fit of the LF isotherm. These results were used in this work for the interpretation of both proton and metal binding in terms of competitive adsorption isotherms.

3.2.2 Copper uptake and the effect of pH

The copper uptake capacity of the biomass was tested using the empirical Langmuir-Freundlich (Eq. 7) and Langmuir isotherms (Eq. 7 with n'=1). As it is showed in Figure 3 (pH 4), both models describe with great accuracy the experimental points and identical fitted parameters are obtained ($Q_{max,Cu}$= 1.12±0.02 mmol·g^{-1} and Log b= 1.4±0.3), since the heterogeneity parameter (n') has a value about the unity, which turns

Langmuir-Freundlich equation into Langmuir one. However, one must be aware that the validity of these models or their underlying assumptions are not proved.

The maximum uptake capacity obtained for copper is around 1.1 $mmol \cdot g^{-1}$, which corresponds to 71 $mg \cdot g^{-1}$ (equivalent to 7% of the total dry weight of the alga). This value is comparable with maximum capacities obtained with other marine algae, such as *Fucus serratus* (1.60 $mmol \cdot g^{-1}$) (Ahmady-Asbchin et al., 2008), *Sargassum filipendula* (1.30 $mmol \cdot g^{-1}$) (Luna et al., 2007) or *Posidonea oceanica* (1.35 $mmol \cdot g^{-1}$) (Izquierdo et al., 2010). Moreover, there are a great variety of useful materials proposed for copper removal from solution with different maximum metal uptake capacities, such as: spent-grain (0.165 $mmol \cdot g^{-1}$) (Lu and Gibb, 2008), activated poplar sawdust (0.085-0.21 $mmol \cdot g^{-1}$) (Acar and Eren, 2006), *Trametes versicolor* (0.63 $mmol \cdot g^{-1}$) (Sahan et al., 2010), Mansonia wood sawdust (0.67 $mmol \cdot g^{-1}$) (Ofomaja et al., 2010) or chitosan (1.59 $mmol \cdot g^{-1}$) (Paulino et al., 2008).

The effect of pH on Cu(II) ion adsorption capacity of *S. muticum* was studied at 2.22 $mmol \cdot L^{-1}$ of initial Cu(II) concentration. Heavy metal sorption studies have shown that pH is one of the most important parameters affecting the process (Schiewer and Volesky, 2000). Both algae structure and copper speciation are affected by solution pH. The structure of the alga can be damaged by extremely acidic pH, due to the acid ability to dissolve certain groups of polysaccharides found on the surface of the biomass. The pH can also change the state of the active binding sites of the algae, mainly the carboxyl groups of alginates. Moreover, the MINEQL+ speciation programme shows that insoluble CuO appears at pH values greater than 5.5, decreasing free Cu(II) ion concentration.

As seen from Figure 4, the copper uptake capacity is almost negligible at pH values less than 2.5, the metal adsorption increases sharply between pH values 2.5 and 4.0, whereas

375 a plateau is reached around pH 4.5. To explain this behaviour, it is important to consider
376 both the metal speciation in solution and the ionic state of cell wall functional groups,
377 mainly carboxyl groups, at various pH values (Davis et al., 2003). Since Cu(II) is
378 present in its free ionic form (Cu^{2+}) at pH values lower than 5, copper biosorption
379 depends on the protonation or deprotonation state of the cell wall polymer functional
380 groups, which have a p*K* value of 3.8 (Lodeiro et al., 2005).
381 At pH values less than 2.0, these functional groups are clearly associated with hydrogen
382 ions, restricting the approach of Cu(II) cations; so low uptake capacities could be
383 explained by the metal ion binding to strongly acidic groups that do not become
384 protonated at these pH, like sulfonic groups from fucoidans, that are known to be
385 present in *Sargassum* biomass (Davis et al., 2003). As the pH increases, more
386 carboxylate groups would be exposed and the available negative charges would lead to
387 a rise in the binding of Cu(II) ions. Above pH 4.5, the increase in the Cu(II) sorption is
388 almost negligible and the uptake reaches a plateau. Similar behaviour has been reported
389 for Cd(II) ion biosorption by protonated *Sargassum* (Lodeiro et al., 2004; Lodeiro et al.,
390 2005).
391 The single component (proton) model was then extended to the general multi-
392 component case (interpretation of competitive ion binding). The conditional proton
393 binding parameters obtained in Eq. 11 ($Q_{\max,H}$, $\log \tilde{K}_H$ and m_H) were assumed to apply
394 also in the presence of copper. The values of $Q_{\max,H}$, and m_H were taken as fixed in all
395 subsequent calculations using the NICA model.
396 The binding parameters for the copper ion were chosen to provide the best simultaneous
397 description of the isotherm at constant pH (4) and the data of copper adsorption *vs*. pH.
398 The values of $\log \tilde{K}_{Cu}$, n_{Cu} and p, in the NICA isotherm, or $\log K_{Cu}$, in the Langmuir
399 competitive models, were first optimized by least squares fit for each data set, and then

average values (see Table 2) were used to fit the experimental data according to the different models, as shown in Figures 5 and 6. In the NICA model, the separation of n_H and p was made using the constraint $m_H = n_H \cdot p$.

The fits to NICA and Langmuir competitive models of the copper binding data at different pH values are shown in Figure 4. It can be observed that the NICA equation is able to reproduce experimental data satisfactory. On the other hand, Langmuir competitive 1:2 model describes with accuracy the "S-shaped" curve in Figure 4, but it does not reproduce the plateau reached at pH values higher than 4, whereas the Langmuir competitive 1:1 model is not able to reproduce the pH effect.

Experimental data from isotherm at pH 4 were also fitted using NICA and Langmuir competitive models (Figure 5). The obtained results demonstrate, like in the study for the adsorption of copper as a function of pH, that only NICA model can explain experimental data properly, employing the same constants attained through proton binding studies.

3.2.3 Cadmium competition on copper uptake

The values listed in Table 2, together with the obtained in a previous article for cadmium adsorption ($\log \tilde{K}_{Cd}$ = 3.1 and n_{Cd}= 1.8) (Lodeiro et al., 2005) were also used to predict the competition between copper and cadmium ions for the biosorbent binding sites. The comparison between experimental results from copper isotherms (at two different initial cadmium concentration) and NICA model prediction is shown in Figure 6. Note that there is good agreement between model and experimental data, using the same model parameters, estimated from the batch sorption experiments and acid-base titrations in the absence of metal.

One must be aware that the use of the initial concentration of the competitor metal (cadmium) does not reflect the sorption equilibrium; however, for modelling purposes only final equilibrium concentrations were considered. In general terms, it can be observed that as copper initial concentration is incremented, the effect of cadmium competition decreases. So that, little difference between the two isotherms is observed at the highest initial copper concentration. Moreover, the copper isotherm is practically identical to the respective one in absence of the competitive cation at any copper concentration for the lowest initial cadmium concentration (compare experimental points in Figure 5 and filled circle data in Figure 6). This result demonstrates a greater affinity of the active binding sites in the alga for copper ions than for cadmium ions. This fact is also supported by the median values of the affinity distribution for cadmium and copper determined with the NICA model, $10^{3.1}$ (Lodeiro et al., 2005) and $10^{4.3}$ (Table 2), respectively. This average Cu binding affinity reported lies between the values of 5.0 and 3.6 for the formation constant of Cu-alginate complexes (assuming langmuirian complexation) reported by De Stefano et al. (De Stefano et al., 2010) at infinite dilution and 0.1 M ionic strength, respectively. It is also probe that the main responsible for the metal sorption in brown algae is alginate.

3.3 Desorption studies

In general, the application of biosorption as a useful alternative in wastewater treatment implies the sorbent regeneration, in order to recover the bounded metal and to reduce process costs.

Desorption studies require a great amount of experimental work to determine both the ideal desorbent and its best conditions of use. In batch studies, it must be taken into account that the metal, once desorbed, remains in solution and it continues in contact

449 with the biosorbent, so a new adsorption equilibrium could be established, affecting
450 desorption process.

451 An important parameter that requires special consideration is the solid/liquid ratio
452 (biosorbent mass/solution volume). Ideally, this relation should be as high as possible,
453 which implies low desorbent volume and/or a great amount of biomass.

454 Two acids, HNO_3 and HCl, were both tested in batch studies and evaluated according to
455 their effectiveness to release copper from the metal-laden biomass, estimating the
456 percentage of metal desorbed (Table 3). The acid elution efficiency was based on the
457 competition between protons and the heavy metal ions bound to active sites, which will
458 be released if eluant concentration is high enough and there is not steric impediment.
459 Both acids were found to be very powerful metal-desorbing agents (Lodeiro et al., 2006;
460 Vilar et al., 2007), with the added advantage that both the release of metal and the
461 regeneration of the alga can be achieved just in one step.

462 First of all, three acid concentrations (0.05, 0.1 and 0.5 $mol \cdot L^{-1}$) were tested at fixed
463 both contact time (2 hours) and solution volume (10 mL). As it is shown in Table 3, the
464 percentage of desorbed copper was similar for the different concentrations of both acids,
465 although not as high as it was desirable (around 75% of copper removal). So that, the
466 lowest acid concentration was selected and the solution volume was increased to 20 mL
467 in order to improve the mix, making that the solution was not so dense. In this way, the
468 percentage of desorbed copper was raised up to 87%. As the contact time was
469 augmented to 4 hours, , percentages very close to the complete copper desorption were
470 achieved (Table 3).

471 To summarize, it can be said that the effectiveness of HNO_3 and HCl acids is very
472 similar. Their optimal conditions of use are an acid concentration of 0.05 $mol \cdot L^{-1}$, a
473 solid/liquid ratio of 5 g/L and a biomass-desorbent contact time of 4 hours.

4. Conclusions

Kinetic experiments showed that equilibrium was attained within 1 hour. An intraparticle-diffusion linear driving force model (LDF) was able to correctly explain these dynamic experiments.

A simple Langmuir or Langmuir-Freundlich isotherm can be used to accuracy describe equilibrium experiments. However, only the application of a model that takes into account the complexity of macromolecular systems, e.g. NICA model, allows a good description of all equilibrium experiments tested (isotherm, pH influence and competition between copper and cadmium) employing the same constants attained through proton binding studies.

Batch studies proved the high efficiency of HNO_3 and HCl acids as copper desorbing agents.

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FIGURE CAPTIONS

Figure 1

Sorption of copper as a function of contact time at different initial metal concentrations, for aqueous suspensions of the protonated *S. muticum* in 0.05 $mol \cdot L^{-1}$ $NaNO_3$ (temperature of 25.0 ± 0.1 ºC, alga dose 2.5 $g \cdot L^{-1}$). The symbols correspond to the experimental points at natural pH (see Figure 2) and different copper initial concentrations: 0.25 $mmol \cdot L^{-1}$ (squares), 0.50 $mmol \cdot L^{-1}$ (up triangles), 1.00 $mmol \cdot L^{-1}$ (circles) and 3.00 $mmol \cdot L^{-1}$ (down triangles). Lines represent modelled results calculated using Equation 6.

Figure 2

Evolution of solution pH values with time at different initial metal concentrations: 0.25 $mmol \cdot L^{-1}$ (dash line), 0.50 $mmol \cdot L^{-1}$ (solid line), 1.00 $mmol \cdot L^{-1}$ (dot line) and 3.00 $mmol \cdot L^{-1}$ (dash-dot line), for aqueous suspensions of the protonated *S. muticum* in 0.05 $mol \cdot L^{-1}$ $NaNO_3$ (temperature of 25.0 ± 0.1 ºC, alga dose 2.5 $g \cdot L^{-1}$).

Figure 3

Copper biosorption isotherms for suspensions of protonated *S. muticum* (alga dose: 2.5 gL^{-1}) at pH 4.0 ± 0.1 (open triangles) at 25 ºC. Lines represent the fits to the Langmuir model, Eq. 7 with *n*'=1, (dashed line) and Langmuir-Freundlich (solid line) model, Eq. 7.

Figure 4

Effect of pH on copper adsorption by 2.5 $g \cdot L^{-1}$ of protonated *S. muticum* at 25ºC, with initial copper concentrations of 2.22 $mmol \cdot L^{-1}$ (open squares). Lines represent the fit of the data to different equations: NICA isotherm, Eqns. 9 and 10 (solid line), competitive Langmuir isotherm assuming 1:1 stoichiometry (dotted line) and assuming 1:2 stoichiometry (dashed line), Eq. 8.

Figure 5

Copper binding by *S. muticum* at pH= 4.0 ± 0.1. Symbols represent experimental points (the same showed in Figure 3), solid line is the fitted NICA isotherm, Eqns. 9 and 10, dotted line is the competitive Langmuir isotherm assuming 1:1 stoichiometry and dashed line assuming 1:2 stoichiometry, Eq. 8.

Figure 6

Effect of cadmium ions competition on copper elimination by *S. muticum* at pH= 4.0 ± 0.1 at 25ºC. Symbols represent experimental points at two different cadmium initial concentrations: 0.1 mmol·L^{-1} (filled circles) and 5 mmol·L^{-1} (open circles). Solid lines represent the fitted NICA isotherm.

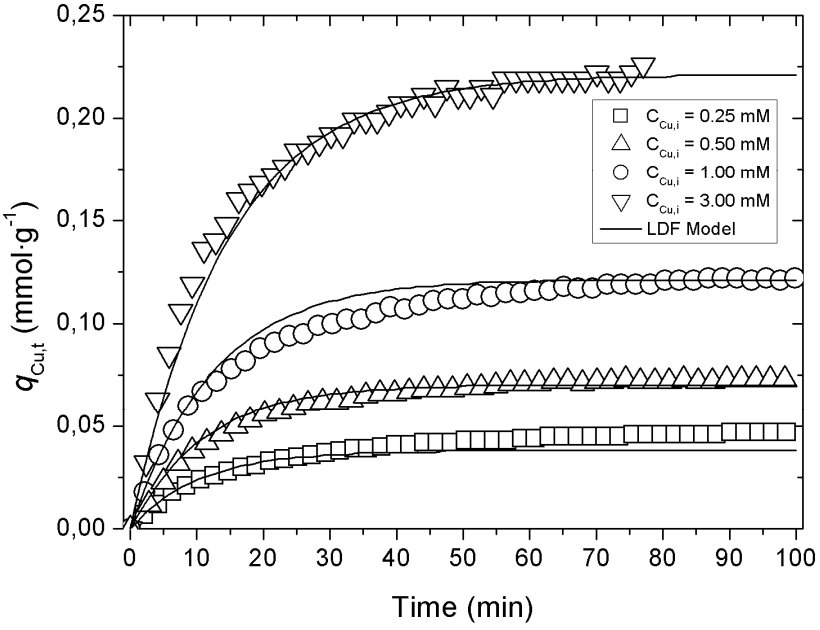

Figure 1

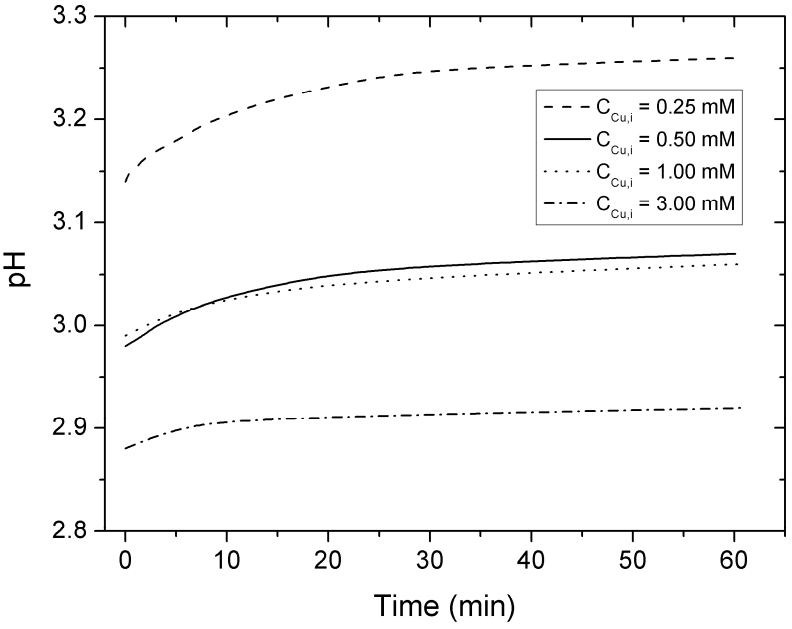

Figure 2

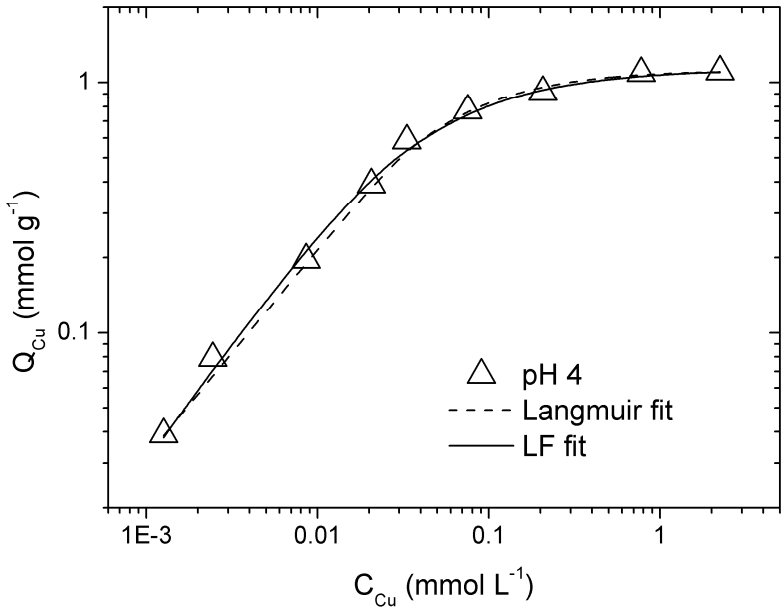

Figure 3

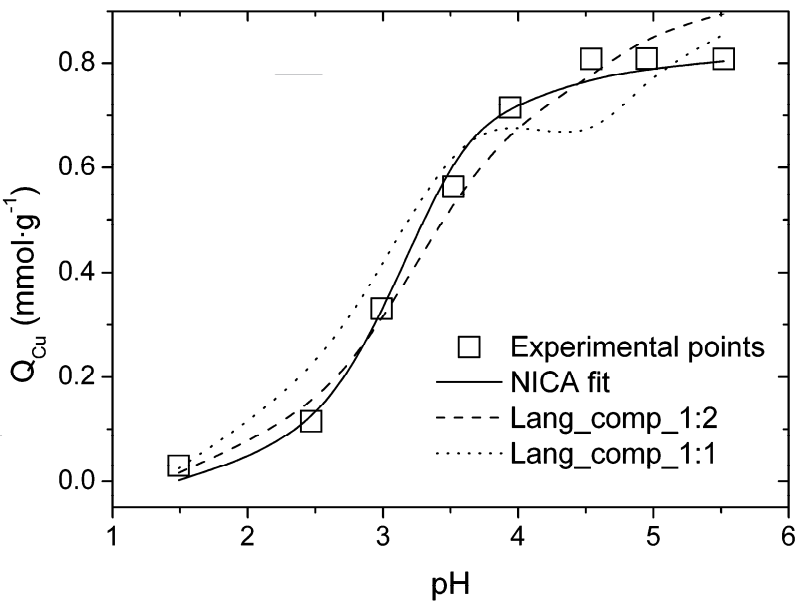

Figure 4

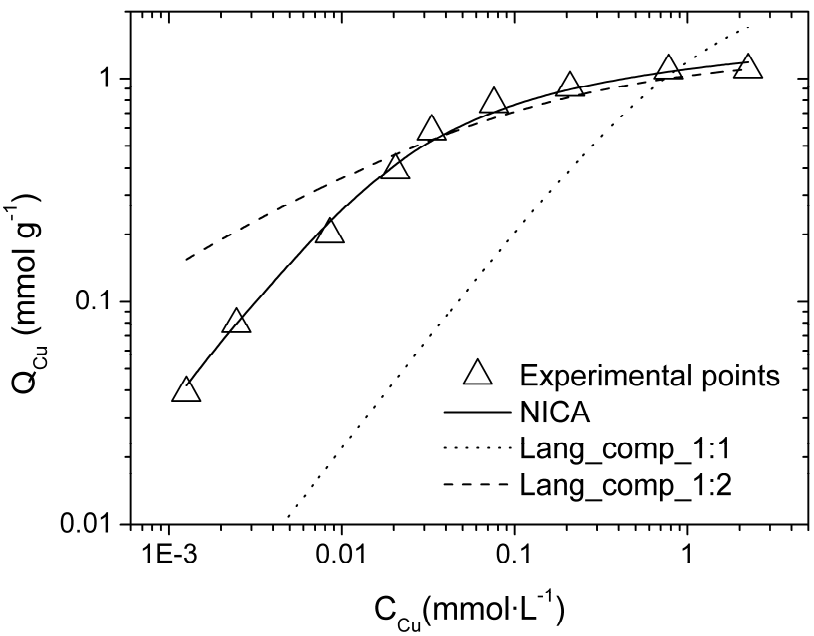

Figure 5

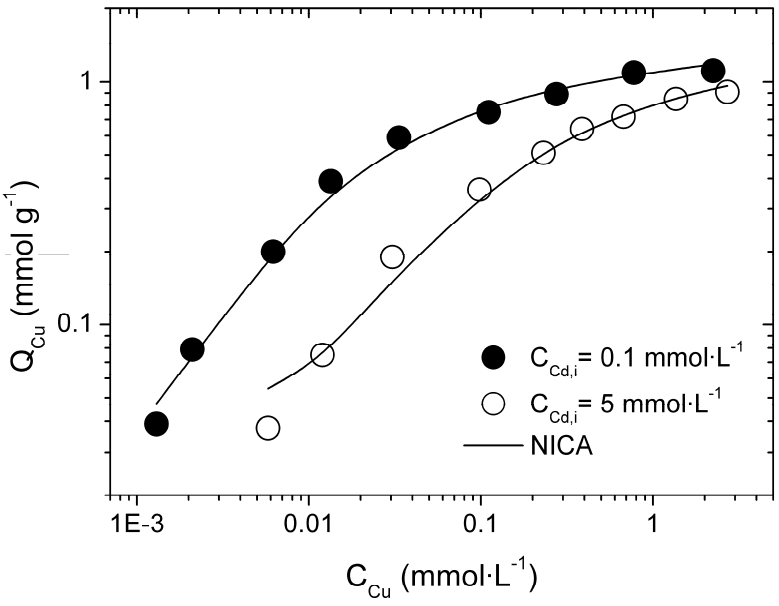

Figure 6

621 **TABLES**

622 **Table 1**. Kinetic rate constant for copper uptake by protonated *Sargassum muticum* at
623 several initial metal concentrations (T=298 K, pH= 3), obtained by fitting experimental
624 data to Equation 6 (LDF model). The percentage of Cu removed from solution at the
625 end of the kinetic process is also included.

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C_i (mmol·L^{-1})	%Cu removed	k (min^{-1})	r^2
0.25	43	0.0642	0.91
0.50	35	0.0642	0.98
1.00	30	0.0642	0.96
3.00	18	0.0642	0.992

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641 **Table 2.** Optimal parameters estimated for copper binding by the acid-treated biomass.

	Copper binding parameters		
	log $\tilde{K}_{Cu}$ /logK_{Cu}	**n_{Cu} /n**	**Heterogeneity parameter, p**
NICA fit	4.3 ± 0.1	1.05 ± 0.05	0.31 ± 0.02
Lang. fit	3.5 ± 0.1	1	
Lang. fit	5.0 ± 0.5	0.5	

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Table 3. Percentage of copper released from protonated *S. muticum* biomass (previously metal-loaded), employing HNO_3 and HCl acids as desorbent agents at different concentrations, solution volumes and contact times.

Concentration (mol·L^{-1})	Contact time (h)	Volume (mL)	%Cu removed
HCl			
0.05	2	10	78
0.1	2	10	73
0.5	2	10	76
0.05	2	20	86
0.05	4	20	94
HNO_3			
0.05	2	10	69
0.1	2	10	75
0.5	2	10	80
0.05	2	20	87
0.05	4	20	95

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