**Non-metabolic uptake of $Al^{3+}$ by dead leaves of *Rubus ulmifolius*: comparison with metabolic bioaccumulation data**

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**Abbreviations**: **FTIR**, Fourier transform infrared; **pzc**, point of zero charge

**Abstract**

This paper reports a physicochemical study (thermodynamic and kinetic data) describing the ability of *Rubus ulmifolius* biomass (dead leaves) for metal uptake. The toxicity of aluminium is a major problem for crops in acidic soils and therefore, aluminium has been selected. The results obtained indicate that dead *R. ulmifolius* leaves uptake up to 10 000 mg/kg on its surface in less than 60 min. This suggests that *R. ulmifolius* can be an excellent component with adsorbent properties for aquatic environments and in particular for amendments to be used in acidic soils in order to control aluminium levels, thus its toxicity. The results obtained have been critically analysed and compared with literature on aluminium bioaccumulation. The application of a pseudo-second order kinetic equation, not previously used in toxicity studies, is discussed. Moreover, a good linear correlation between stability constants for $Al^{3+}$ complexes with several defined ligands and the Langmuir affinity constants obtained from the corresponding adsorption isotherm has been found. Therefore, in addition to its ethno-botanical relevance, applications of *R. ulmifolius* as a detoxifier for aluminium in a simulated acidic gastrointestinal fluid, as phytostabilization agent in amendments or in natural attenuation cycles or as biomass for wastewater treatment containing aluminium, are suggested.

**Keywords**: Adsorption, Aluminium, Amendments, Biomass, Phytoremediation

## 1. INTRODUCTION

Many cultures, without knowledge of the particular chemical agents, have observed medicinal properties in *Rubus* that have been applied in different ways [1]. *Rubus ulmifolius* Schott Rosaceae (elmleaf blackberry) is a wild shrub native to the Mediterranean area. The importance of *R. ulmifolius* as a traditional medicine was highlighted in different ethno-botanical studies conducted in rural communities of southern Italy [2]. An analysis of traditional knowledge about food applications of *R. ulmifolius* in northwest of the Iberian peninsula [3], western Spain [4] or central Italy [5], has also been performed. Although medicinal plants discovered by traditional societies are proving to be an important source of potentially therapeutic drugs [6], research in this field (treatment with *R. ulmifolius*) is very scarce. Only a few studies demonstrating the antioxidative [7--9], antimicrobial [9--12], or hypoglycaemic activity [13] of *R. ulmifolius* leaves can be found in the literature.

Phytoremediation techniques are characterised by using plants to reduce heavy metals or other contaminants in the environment [14]. They are used to mitigate toxic effects that produce variable disorders affecting the development of plants [15]. This methodology uses the variable response of plants, in terms of tolerance, to the presence of contaminants, such as metals. Therefore, immobilization of metals, present even at very low concentrations, is exploited in phytostabilization technologies. In contrast, hyperaccumulation, which is exhibited by different parts of plants accumulating large quantities of metals, characterizes the best phytoextraction activity [16]. Phytoremediation is mainly based in bioaccumulation processes of living plants, biomaterials and different phytotecnologies, and is a useful technique to clean polluted environment.

Adsorption technologies, using materials of biological origin for contaminants removal, are enclosed in the term biosorption. Therefore, biosorption can be considered a passive process, without metabolic assistance, and therefore is not subjected to toxicity problems. Both techniques present advantages and disadvantages, however comparative research analysing bioaccumulation vs. biosorption processes is scarce with only few examples in the literature [17, 18]. The aim of this work is also to contribute filling in some aspects of this lack.

Aluminium ion ($Al^{3+}$) that predominates in soils at pH < 5 (acidic environment) is toxic to plants at micromolar concentrations. Aluminium toxicity has been recognized as a major factor limiting crop productivity on acidic soils which comprises about 40% of the arable land worldwide. Micromolar concentrations of $Al^{3+}$ can inhibit root growth within minutes or hours in important plant species, such as wheat, maize or oats [19, 20].

The addition of organic amendments is considered a useful way to help in soil reclamation [21]. It has often been shown that the addition of organic amendments to soils increases the immobilization of metals essentially through adsorption reactions. The retention of metals in this case is attributed to an increase of charge and/or to the presence of metal binding compounds [22]. Due to the abundance of aluminium in

the earth´s crust and it´s widely use in modern technology, the general population is exposed to relatively high levels of this metal. The safety of $Al^{3+}$ in humans has been controversial for a century, including its use in cookware and food storage. Aluminium toxicity is observed when protective mechanisms, mainly through blood and kidneys, fail. This was most clearly demonstrated in the 1970s by the appearance of severe neurological disorders among long-term dialysis patients (dialysis dementia) [23]. Moreover, there has been considerable debate over a possible role for aluminium in the development of Alzheimer's disease [24, 25].

To the authors' knowledge, this is the first paper reporting a physicochemical study (thermodynamic and kinetic data) describing the ability of *R. ulmifolius* biomass (dead leaves) for metal uptake. Previously we presented discrete data for the removal of Cr(VI) [26], Cr(III) [27] and Hg(II) [28] using *R. ulmifolius*. Here, the selected metal is aluminium, which toxicity is a major problem for crops in acidic soils. The results obtained indicate that *R. ulmifolius* leaves uptakes up to 10 000 mg/kg on its surface. This suggests that *R. ulmifolius* leaves can be an excellent component with adsorbent properties for soil amendments, to be used in acidic soils to control $Al^{3+}$ levels, thus its toxicity. This adsorption power could be also responsible for the fixation of this metal, mainly in the roots of *R. ulmifolius* in some acidic environments, suggesting a phytostabilization feature of this plant. The kinetic study of $Al^{3+}$ fixation on *R. ulmifolius* leaves, in particular the proposed pseudo-second order equation and the rate constants obtained, may also be of some utility in other fields such as the toxic kinetics of $Al^{3+}$ in the brain, which is a high interest research [29].

## 2. MATERIALS AND METHODS

### 2.1. *Biomass characterization*

*Rubus ulmifolius* was collected in Galicia (43°19'35.7"N 8°24'28.2"W) from a non-contaminated soil. After washed with tap water, the biomass was dried in an oven at 60 °C overnight. Thereafter, it was grounded with an analytical mill (IKA A 10) and then sieved to particles of 0.5--1 mm diameter. Inductively coupled plasma-mass spectrometry (ICP-MS Thermo Finnigan X Series) was used, after microwave assisted acidic digestion, to assess the metals initially present in the biomass.

The pH at the point of zero charge (pzc) of the *R. ulmifolius* biomass was obtained by potentiometric mass titration [30, 31]. 50 mL 0.03 M $NaNO_3$ was used as supporting electrolyte and blank. The potentiometric titrations were done in duplicate. Two different biomass concentrations, 0.75 and 1.5 g/L, were used in temperature controlled experiments at 25.0 ± 0.1 °C, under magnetic agitation and $N_2$ bubbling. Potentiometric titrations were implemented using a Crison GLP 22 pH-meter and a Crison microBu 2031 automatic burette (Barcelona, Spain). The proton concentration was measured using a glass electrode (Ingold, Wilmington, USA) and an Ag/AgCl reference electrode (Metrohm, Herisau, Switzerland).

A spectrophotometer (Bruker Vector 22) equipped with an attenuated total reflection device from Specac (Golden Gate ATR) was used to obtain Fourier transform infrared (FTIR) scans within the range of 400--4000 $cm^{-1}$.

### *2.2 Chemicals*

The chemicals used were: $AlK(SO_4)_2 \cdot 12\ H_2O$ (Merck, p.a.), $NaNO_3$ (Panreac, p.a.), NaOH (Merck, p.a.), HCl (Merck, p.a.), $H_2SO_4$ (Panreac p.a.), $CH_3COONa \cdot 3\ H_2O$ (Panreac, PRS), $CH_3COOH$ (Panreac, PRS), $N_2$ C-55 (99.9995 %) from Carburos Metalicos, and Eriochrome cyanine R. (Sigma-Aldrich)

### *2.3 Kinetic and thermodynamic data*

#### *2.3.1 Kinetic studies*

Kinetic experiments were done for initial aluminium concentrations of 10 and 100 mg/L using 2.5 g/L of the biomass without pH adjustment. The equilibrium pH of the solutions varied between 4.1 and 3.3, for 10 and 100 mg/L $Al^{3+}$. A rotary shaker was used to agitate the solutions at 175 rpm and room temperature. The samples were taken at different times, centrifuged (Nahita centrifuge) at 10 000 rpm for 5 min, and thereafter the aluminium concentration was measured following the Eriochrome cyanine R method [32].

The aluminium uptake at time *t*, $Q_t$ (mg/kg), was calculated according to the following equation:

$$Q_t = \frac{V\ ([Al]_i - [Al]_t)}{m} \quad (1)$$

where *V* is the volume of solution (L), $[Al]_i$ and $[Al]_t$ are the initial $Al^{3+}$ concentration in solution and at time *t* (mg/L), respectively, and *m* is the mass of *R. ulmifolius* (kg, dry weight)

#### *2.3.2 Adsorption isotherms*

Adsorption isotherm studies were carried out using different initial aluminium concentrations between 5 and 150 mg/L and a *R. ulmifolius* concentration of 2.5 g/L. The pH of the solutions was adjusted to obtain final equilibrium pH values of 2.5 and 4.0. A rotary shaker was used to agitate the solutions for 3 h at 175 rpm and room temperature. The aluminium sorbed at equilibrium, $Q_e$ (mg/kg), was calculated as shown in Eq. (1), but referred to equilibrium conditions, where $Q_t$ and $[Al]_t$ were substituted by $Q_e$ and $[Al]_e$, the aluminium concentration at equilibrium.

## 3. RESULTS AND DISCUSSION

### 3.1 Biomass characterization

Initial concentrations of aluminium and other metals in native biomass of *R. ulmifolius* are shown in Table 1. That table also contains values for heavy metals present in leaves of *R. ulmifolius* taken from studies carried out in different contaminated mine soils [33-35]. It can be observed that, except for Ca, all the initial metal concentration values present in *R. ulmifolius* leaves (Table 1) are below 1000 mg/kg which is actually considered an hyperaccumulation threshold [36]. According to these values, *R. ulmifolius* cannot be considered as aluminium or other metal accumulator.

Another criterium to indicate the presence of Al stress in a plant is the calcium/aluminium ratio. According to Cronan and Grigal [37], a Ca/Al molar ratio of <12.5 would indicate a 50% risk of $Al^{3+}$ toxicity, and a ratio of <6.2 a 75% of risk. From Table 1 a Ca/Al ratio of approximately 110 was estimated. According to the criteria referenced above, the biomaterial selected for this study is not affected by aluminium toxicity or stress. The root cell wall is considered to be the major site of aluminium accumulation [36]. In fact, most of the plants growing in acidic environments (e.g. Galicia

region, NW of Spain) do not accumulate aluminium in their foliage. This is a form of exclusion of aluminium from plant tissue as an external resistance mechanism. Therefore, data from Table 1 suggest the potential use of *R. ulmifolius* in phytostabilization studies, in which plants are used to stabilise the soil surface by retaining metals in the roots, reducing mobility and bioavailability of environmental pollutants, and consequently decreasing erosion and leaching of metals to the soil deeper layers. However, this suggestion must be substantiated with more detailed studies about distribution of aluminium and other metals in different parts (leaves, shoots and roots) of *R. ulmifolius*.

The characterization of the *R. ulmifolius* surface was carried out by potentiometric mass titration and FTIR analysis. The former allows to calculate the pH at which the surface charge of different oxides [30], vegetable waste [31] or edible seaweeds [38] is zero ($pH_{pzc}$). This value is related to surface ionization, so it is suitable to interpret the interactions between the charged groups present in the material and the species in solution. Therefore, at solution pH values above $pH_{pzc}$ the *R. ulmifolius* surface presents a negative charge, favouring the interaction with negative charged species. In contrast, the *Rubus* surface is considered to be positively charged when solution pH values are lower than $pH_{pzc}$.

The $pH_{pzc}$ value for *R. ulmifolius* leaves, 3.7, was calculated from the intersection of the biomass titration curves with a control at two different biomass doses, obtained under identical conditions (Figure 1). Regarding metal removal as a function of pH, aluminium elimination takes place either at pH lower than $pH_{pzc}$ (pH 3.3), when the biomass surface presents positive charge, or pH greater than $pH_{pzc}$ (pH 4.1, obtained for an initial Al concentration of 10 mg/L) with a negatively charged surface.

Thus, taking into account that aluminium is present in solution as $Al^{3+}$ as speciation studies demonstrate [38], it can be postulated the existence of other mechanisms alternative or complementary to adsorption: e.g ion exchange, complexation, surface precipitation, etc. [39]. Metal adsorption appears to be independent of pH for *R. ulmifolius* leaves (Fig. 2), probably as a result of either the interaction of $H^+$ and $Al^{3+}$ with different chemical groups of the biosorbent cell walls, or the interaction of the metallic ion with other groups that were protonated at higher pH values (e.g. alcohols)

The *R. ulmifolius* leaves present a low metal adsorption capacity, c.a. 0.37 mmol/g, if it is compared for example with typical brown marine algae, a very good adsorbent biomaterial for metals (Table 2). In fact, it is observed a very similar behaviour of *R. ulmifolius* and *Gelidium sesquipedale*, a red alga [38]. In contrast to brown algae, which present a large number of carboxylic groups, in these materials alcoholic and amino groups predominant. These groups are not deprotonated at pH < 7--11 [40], and then, the metal interaction with alcoholic and amino could explain the small effect of pH observed.

Fourier Transform Infrared spectrum of *R. ulmifolius* leaves (Fig. 3) was used to identify in a qualitative way the different chemical entities present in the materials. The technique employed allowed analysing the samples directly without requiring KBr pellet preparation. The bands at 3300 $cm^{-1}$ can be ascribed to -OH and –NH groups present in the material's surface. The carboxyl ions show two bands: an asymmetrical stretching band at 1615--1630 $cm^{-1}$ and a weaker symmetrical band at 1410--1440 $cm^{-1}$ [41]. The strong band at 1030 $cm^{-1}$ is due to the –C–O stretching of alcoholic groups. –CH stretch can be ascribed to the band at 2920 $cm^{-1}$, while the bands at 1250 $cm^{-1}$ represent $–SO_3$ stretching.

**3.2 Kinetic studies and comparison with bioaccumulation data**

The batch experiments showed that aluminium uptake is relatively fast, reaching equilibrium in around 60 minutes (Fig. 4). The contact time significantly affects the aluminium uptake. The sorption rate increases sharply in the first minutes, followed by a slower uptake as equilibrium is approached. This fast aluminium uptake indicates that the sorption occurs mostly on the surface of the adsorbent, as was also proposed by other authors [42--44].

We have compared the kinetic results obtained in this work with those described in a classic work by Zhang and Taylor [45] on the kinetics of aluminium uptake by excised roots of Al-tolerant and Al-sensitive cultivars of *Triticum aestivum.* The aluminium uptake was considered by these authors to be biphasic, with a rapid phase of uptake in the first 30 minutes followed by a linear phase of uptake up to 180 min. At the end of the uptake period, higher concentrations of $Al^{3+}$ were found in roots of the Al-sensitive cultivars (Neepawa and Scout-66) than in the Al-tolerant cultivars (Atlas-66 and PT-741), but these differences were small. Uptake experiments were performed using a physiologically relevant concentration of aluminium (75 µmol/L) and pH 4.5

Dual kinetics, similar to the pattern of $Al^{3+}$ uptake reported here, have commonly been interpreted as representing uptake by passive accumulation into the apoplasm (rapid phase) and uptake across the plasma membrane (linear phase) into the symplasm. This constitutes the classical interpretation of the kinetics of the Al uptake. Similar results were described by Zhang and Taylor, and McDonald-Stephens and Taylor [46, 47]

However, none of these works propose a kinetic equation to quantitatively describe the experimental data. Here the kinetic data was successfully fitted to a pseudo-second order kinetic equation (Fig. 4):

$$Q_{\mathrm{t}} = \frac{q_{\mathrm{e}}^2 kt}{1+q_{\mathrm{e}}kt} \qquad (2)$$

where $q_{\mathrm{e}}$ is an adjustable parameter that represents the amount of aluminium removed from solution at equilibrium, $k$ is the pseudo-second order constant and $t$ the time expressed in minutes. The obtained parameters are shown in Table 3.

In addition, Fig. 5 also shows the fit of the experimental data taken from Fig. 1, shown by Zhang and Taylor [45], to a pseudo-second order kinetic equation (Eq. (2)).

The fit is acceptably good, with relatively low errors in the parameters calculated, particularly in the pseudo-second order rate constant (Table 3). This indicates that this kind of equation, extensively used in passive metal uptake processes like biosorption [39], can be useful to quantitatively reproduce the entire curve associated to the two phase process described in different papers [45--48]. This proposal is not necessarily in contradiction with a dual description of the metabolic uptake process; the first step of the dual process would correspond to a diffusion control (Eq. (3)), taking place at short times. This implies proportionality with the squared root of time, as observed in Figure 6, which is frequently found in biosorption (passive uptake) of metals kinetic experiments [38, 49--52]

$$Q_{\mathrm{t}}=k_{\mathrm{d}}t^{1/2} \qquad (3)$$

where $k_{\mathrm{d}}$ is the intraparticle diffusion constant.

Figure 7 shows the pseudo-second order kinetic constants as a function of $Al^{3+}$ concentration. This figure contains data from either biosorption or bioaccumulation studies taken from different sources [45, 53-55]. A decreasing trend, commonly obtained when a pseudo-second order adsorption kinetic behaviour is involved [56], can be observed. The data in Fig. 7 fit with accuracy to the linearized equation:

$$k = \frac{a}{b + [\mathrm{Al}]_i} \quad (4)$$

where $a$ and $b$ are fitting parameters. This empirical equation has been described previously by Ho and Mckay [57]. However, as has been discussed by Guiso et al. [58], until now there is no theoretical justification for that observed trend with the concentration, although these last authors suggest the possibility that conformational changes in the biomass, may occur.

The analysis of data carried out in this work suggest that pseudo-second order equation can be a useful descriptive tool in $Al^{3+}$ or other metal kinetic studies of toxicity. According to Gregory Taylor et al. [48], $Al^{3+}$ toxicity is a major factor limiting growth of plants on a worldwide basis, and exerts a toxic effect within minutes of exposure. Although several resistance mechanisms have been proposed, the mechanistic basis of $Al^{3+}$ transport and the overall subcellular distribution remain speculative.

### 3.3 Adsorption isotherms and comparison with bioaccumulation results

Figure 2 shows fitted data corresponding to the aluminium adsorption by *R. ulmifolius* leaves. As can be observed in the Fig. 2, *R. ulmifolius* leaves show no pH effect in the pH range of 2.5--4. The simple Langmuir equation (Eq. 5) is able to accurately describe the effect of aluminium concentration on *R. ulmifolius* uptake capacity at constant pH:

$$Q_e = \frac{Q_{max} K [Al]_e}{1 + K [Al]_e} \quad (5)$$

where $Q_{max}$ is the maximum $Al^{3+}$ biosorption capacity and $K$ represents an affinity constant. A high value of parameter $K$ indicates a high affinity of the biosorbent for the sorbate.

The values of $Q_{max}$ and the affinity constant, $K$, obtained in this work are compared with those obtained for the interaction of $Al^{3+}$ with other biomaterials [53--55, 59] in Table 2. It can be apparently observed three groups of data; those with a low affinity constant, but high $Q_{max}$ values, which correspond essentially to brown algae. The other group is comprised by *R. ulmifolius* data, which show a high affinity constant but low $Q_{max}$ values. Finally, there is an intermediate group between them. These data suggest a more energetic binding between aluminium and *R. ulmifolius* biomaterial than with brown algae, although the monolayer coverage was reached at lower concentrations. Therefore, a higher value of the affinity constant for $Al^{3+}$--*R. ulmifolius* interactions would facilitate natural attenuation by a stronger fixation of aluminium in leaching processes, for example.

Figure 8 shows an empirical semi-logarithmic relationship between the Langmuir affinity constants shown in Table 2 for the $Al^{3+}$--biomass interaction, and the logarithm of the stability constant ($\beta$) for the complex of $Al^{3+}$ with different low molecular-mass organic ligands: formic acid (1.36), acetic acid (1.51), propionic acid (1.69), phthalic acid (3.18), citric acid (8.32), salicylic acid (12.9) and catechol (16.3). Values were taken from Vance et al. [60] for a ligand/metal relation of 1:1. The good linearity of the fit seems to confirm the fact that aluminium complexes are primarily produced on sites containing oxygen as ligand [60]. It is also worth mention that an increase in the value of the Langmuir adsorption constants from algal biomass, characterized by the predominance of carboxylic acids as complexing sites, to *R. ulmifolius* with abundance of hydroxyl groups (phenolic) (7.8) is observed. However this point should be confirmed with more work related to $Al^{3+}$--biomass interactions.

From Fig. 2 it can also be seen that the levels of Al adsorbed on dead leaves of *R. ulmifolius* are comprised between c.a. 4000 and 10 000 mg/kg of dry material. We have used these units to build the

isotherms in order to compare directly with bioaccumulation results found in the literature.
Brooks et al. [61] were the first to define the term “hyperaccumulator” to describe plants that can accumulate more than 1000 mg of dry weight of nickel per kg of biomass in aerial parts. Steve-Jansen et al. [36] indicate in 2002 that “hyperaccumulation” has not been used referred to aluminium, and suggest that 1000 mg/L aluminium in dried leaf tissue is an appropriate standard for the aluminium hyperaccumulation definition. However, Masunaga et al. [62] on the basis of nutritional characteristics of plants containing aluminium suggested a criterion based on aluminium concentration (>3000 mg/L) is more suitable to define aluminium hyperaccumulators.
The results obtained in this work show that strictly according to the definition, dead leaves of *R. ulmifolius* cannot be considered a hyperaccumulator, a concept initially associated to metal accumulation with the participation of metabolism. However, it can be considered that *R. ulmifolius* dead leaves hyperaccumulate $Al^{3+}$ according to the high level of adsorbed $Al^{3+}$ present in the leaves, and the criteria commonly used to classify a plant as hyperaccumulator. In addition, the presence of levels up to 10 000 mg/kg manifests that passive adsorption (biosorption) allows to reach much higher levels than those obtained by a metabolic way (bioaccumulation/hyperaccumulation): about 75 mg/kg initially present in the leaves before contacting $Al^{3+}$ solutions. These data is also coincident with the observation that the average content of $Al^{3+}$ in herbaceous tissues of plants is around 200 mg/L of the dry mass (0.02%).
The high capacity of *R. ulmifolius* dead leaves to retain $Al^{3+}$ suggests a possible application of this biomaterial as amendment in aluminium rich soils, contributing to natural attenuation processes. Their presence in soils would help to decrease the energy cost of plants around and to metabolically reduce aluminium levels through different mechanisms of resistance (hyperaccumulation or exclusion). Plants must consume some energy to synthesize complexing agents and to control possible toxic effects of aluminium [63]. The mechanisms of plants resistance to heavy metals are commonly described in terms of the location where metal interaction occur, as internal (fixation in the symplasm) or external (the attachment of the metal is produced in the external parts or apoplasm). This last process is considered to avoid the penetration of the metal in the cytoplasm [29, 36], although there are more works about fixation of aluminium in the symplasm of the plants. Therefore, the data present in this work could be useful in modelling at least the external mechanism of aluminium (or other metals) fixation in the apoplasm of plants. Adsorption studies on dead biomaterials, as those carried out in this work, can also help to obtain useful kinetic and thermodynamic data, such as isotherms, rate equations and constants.

## 4. CONCLUDING REMARKS

The relatively fast aluminium removal kinetics, less than one hour to achieve equilibrium, and the relatively high Langmuir affinity constant and maximum aluminium uptake (up to 10 000 mg/kg), by a non-metabolic process, indicate that *R. ulmifolius* leaves can act as good potential adsorbent for aluminium removal in relatively acidic environments (pH 2.5--4). Therefore, an application as phytostabilizer in amendments or in natural attenuation cycles, in wastewater containing aluminium treatments or as a detoxifying agent for aluminium in a simulated acidic gastrointestinal fluid is suggested.
A pseudo-second order kinetic equation is able to acceptably describe the dependence of $Al^{3+}$ uptake with

time. In addition, a comparative analysis with aluminium bioaccumulation published data allows concluding the utility of the pseudo-second order equation in the interpretation of metabolic uptake of this metal, not previously suggested and with some interest in the interpretation of $Al^{3+}$ tolerance by plants. However, the potential physiological use of the equations presented in the paper has to be taken with caution because this study has been carried out with dead biomaterial.

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**Figure 1.** Potentiometric mass titration curves for suspensions of *R. ulmifolius* leaves (0.75 g/L □ and 1.5 g/L △). The solid line corresponds to the titrations of blank solution (0.03 M $NaNO_3$ and a small amount of 0.1 M NaOH, pH 11.6)

**Figure 2**. Aluminium sorption isotherms for suspensions of 2.5 g/L of *R. ulmifolius*. The symbols correspond to the experimental points at pH 4 (○) and pH 2.5 (△). The lines represent the fits to Langmuir equation (Eq. (5))

**Figure 3**. Fourier-transform infrared spectrum for *R. ulmifolius* leaves

**Figure 4.** Aluminium uptake from solution as a function of contact time for *R. ulmifolius* leaves. Initial aluminium concentration: 10 mg/L (right *y*-axis) and 100 mg/L (left *y*-axis), $T$ = 21°C, biomass dose of 2.5 g/L and not fixed pH. The lines represent the fit to Eq. (2)

**Figure 5.** Kinetic data of aluminium removal from solution obtained from Zhang et al. [45] the lines represent the fit to Eq. (2)

**Figure 6.** Intraparticle diffusion model for kinetic data of Al removal by *R. ulmifolius*. Initial aluminium concentration: 10 mg/L (right *y*-axis) and 100 mg/L (left *y*-axis), $T$ = 21°C and not fixed pH. The lines represent the fit to Eq. (3)

**Figure 7.** Pseudo-second order kinetic constants, $k$, as a function of Al concentration. This figure contains data from biosorption and bioaccumulation studies taken from different sources: this work (△); [54] (○); [53] (□) and [45] (▽). The inset represents the linear relationship between the inverse of k and the initial Al concentration for the same data.

**Figure 8**. Empirical semi-logarithmic plot of affinity Langmuir constants vs. stability constants of complexes $Al^{3+}$--L for several ligands (L). From L = formic acid (lowest value) to L = catechol (highest value). Data taken from [60]

**TABLE 1** Metal concentrations in native *R. ulmifolius*

| mg/kg | This work | [33] | [34] | [35] |
|---|---|---|---|---|
| Cd | 0.015 | | 0.006--0.01 | 0--0.1 |
| Pb | 0.34 | 149 (leaves) to 1178 (roots) | 0.86--7.6 | 0.5--1 |
| Mg | 3501 | | | |
| Al | 72.3 | | | |
| Ca | 11776 | | | |
| Fe | 82.9 | | | 363.8--422.8 |
| Ni | 1.13 | 151 (roots) | | 5.5--6.4 |
| Cu | 5.2 | | 12.1--17.6 | 11.4--13.8 |
| Zn | 20.5 | 91 (leaves) to 957 (soil) | 25--57 | 37.2--63.7 |
| Hg | 0.12 | | | 0.1--1.5 |

**TABLE 2** Langmuir affinity constants and $Q_{max}$ values for Al removal using different biomaterials.

| Sorbent | $Q_{max}$ mmol/g | $K$ L/mmol | $K_D = 1/K$ µmol/L | Reference |
|---|---|---|---|---|
| *Padina pavonica* (brown alga) | 2.86 | 0.540 | $1.85 \times 10^3$ | [54] |
| Cattle manure vermicompost | 0.309 | 9.47 | $1.06 \times 10^2$ | [59] |
| Cattle manure vermicompost (Kaolin wastewater) | 0.0407 | 34.40 | 29.1 | [59] |
| *Streptomyces rimosus* | 0.436 | 1.62 | $6.18 \times 10^2$ | [55] |
| *Turbinaria conoides* (brown alga) pH 3.5 | 2.18 | 0.149 | $6.71 \times 10^3$ | [53] |
| *Turbinaria conoides* (brown alga) pH 4 | 2.37 | 0.168 | $5.95 \times 10^3$ | [53] |
| *R. ulmifolius* leaves pH 2.5 | 0.370 ± 0.020 | 30 ± 11 | 33.5 | This work |
| *R. ulmifolius* leaves pH 4.0 | 0.353 ± 0.015 | 46 ± 11 | 21.8 | This work |

**TABLE 3** Kinetic parameters obtained from the pseudo-second order model (Eq. 2) for the binding of aluminium to *R. ulmifolius* (this work) and to *Triticum aestivum L.* [45]

| | **Pseudo-second order (Eq. 2)** | | | | |
|---|---|---|---|---|---|
| | $[Al]_i$ (mg/L) | $Q_e$ (mg/kg) | $k$ (kg/mg·min) | $r^2$ | Reference |
| | 10 | 3508 ± 159.7 | $(1.1 \pm 0.27) \times 10^{-4}$ | 0.8669 | This work |
| | 100 | 7831 ± 290.0 | $(5 \pm 1) \times 10^{-5}$ | 0.9175 | This work |
| Atlas 66 | 2.02 | 525 ± 50.3 | $(4.5 \pm 1.6) \times 10^{-5}$ | 0.9299 | [45] |
| Neepawa | 2.02 | 531 ± 33.0 | $(6.1 \pm 1.5) \times 10^{-5}$ | 0.9597 | [45] |
| PT-741 | 2.02 | 375 ± 25.2 | $(8.9 \pm 2.4) \times 10^{-5}$ | 0.9507 | [45] |
| Scout-86 | 2.02 | 649 ± 28.3 | $(3.2 \pm 0.5) \times 10^{-5}$ | 0.9888 | [45] |