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Original Article

Copper biosorption by *Rhodococcus erythropolis* isolated from the Sossego Mine – PA – Brazil

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ABSTRACT

This work evaluates the copper biosorption capacity (Cu^{2+}) by the biomass of a bacterium strain identified as *Rhodococcus erythropolis* isolated from copper mining environmental samples. The copper uptake was analyzed under different influences of pH, biomass concentration, and metal concentration. The total copper uptake was measured using ICP-OES and the metal adsorption capacity calculated by construction of Langmuir and Freundlich isotherm models. *R. erythropolis* biomass is a potential material to treat and to remove copper from low concentration of aqueous contaminated effluents reaching a total absorption of 68.03 mg of Cu^{2+} per gram of biomass according to the Langmuir model at pH = 6, the optimal pH for maximum copper uptake. Our results showed a unique absorption rate when compared to other bacteria belonging to the *Rhodococcus* genus or even when compared to other bacteria genera able to adsorb metal from aqueous environments.

1. Introduction

The increase of industrial activities from productive sectors, the huge amount of water in the industrial processes and also the quantity and complexity of the waste generated, make the development of treatment processes for cleaning wastewater urgent and necessary [1]. The presence of metallic ions in wastewater is extremely undesirable, once metals are recalcitrant and very toxic to life. Metals such as copper, nickel, cobalt and zinc are a serious concern due to their extensive use in several products within all the productive chain divisions: from the traditional heavy industries to high technology and state of the art products. This wide use

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tends to rise the concentration of metals in the environment, exposing all levels of the food chain to elevated concentration rates of metal, which can reduce biodiversity in general and cause serious diseases in human beings [2].

As persistent, exchangeable between living creatures and literally non-biodegradable, the available choices to prevent metal spreading in the environment are basically decreasing their discharge with stringent environmental regulations that compelling industries to switch to cleaner production methods or removing the metals from the effluents.

Some of important metal contaminated effluents are found in mining areas. Despite the negative environmental impact of mining, it is still a worldwide important industrial sector of economy. In Brazil, the state of Pará has the largest reserves of copper ore, accounting for 82% of the entire Brazilian copper reserve [3], second most representative state in the collection of CFEM (Financial Compensation for the Exploitation of Mineral Resources) [4,5].

The Sossego Mine was Vale's first copper operation. The plant has the capacity to process 41,000 t of ore per day, which is equivalent to 15 million tons per year, from open pit mining. The proved reserve is of the order of 255 Mt with an average content of 1.0% copper and 0.3 g/t gold [6,7].

The plant operation began in April 2004, with an installed capacity of 540,000 t of concentrate per year, with 30% copper and 8 g/t gold; [6] the Sossego mine has a dam with low concentrations of metallic copper residues, which has the potential to cause serious environmental impacts.

Biotechnology focusing on microorganism abilities to deal with metals can help recover metal from contaminated effluents. The superior capacity of microbial biomass to absorb metals has attracted the attention of researchers when compared to other inorganic and organic sorbents [8]; studies show that, in most cases, the accumulation of metals by microorganisms is due to surface phenomena [9,10], generally called biosorption.

Biosorption is defined as a process in which microbial biomass is used for retaining, removing or recovering heavy metals from water [10–13]. The most common biomass used for this purpose can be from algae, yeasts, fungi and bacteria [14,15]. Dead and living cells can adsorb metal since it is a process independent of metabolism [16]. Increasing attention has been given for metal removal by biosorption mainly due to the high selectivity and removal rates, low costs, minimization of chemical sludge volumes, possibility to treat large volumes and high efficiency for low concentration effluents [18,19]. Yet, one of the most important factors that can change the scenario of metal-contaminated effluents treatment is the possibility to recover and to reuse the adsorbed metal [8,13,20]. Recovery and reuse could significantly reduce the costs or even generate income, improving mining technology.

We here focused on the study of copper biosorption capacity of a strain identified as *Rodhococcus erythropolis* isolated from the Sossego mine area. This species, poorly documented as being related to metal contaminated sites, presented a singular capacity to adsorb copper, experimentally reaching 101.90 milligrams of copper per gram of biomass. To predict and to calculate the full adsorbance capacity, the Langmuir and Freundlich models were used. According to the Langmuir model, the best prediction model for our study, the *R. erythropolis* biomass could achieve 68.03 mg of copper per gram of biomass in aqueous environment. This biosorption rate has never been reported for this bacterium so far.

2. Material and methods

2.1. Microorganism isolation and identification

The strain studied was isolated from a high copper concentration area that has been under study since 2013, focusing on bioprospection of highly resistant copper bacteria [21]. The isolation followed the methods already described [21], with adaptations, and this resistant bacterium was isolated, identified and stored at −80 °C.

The identification was performed by mass spectrometry and confirmed by 16S rRNA gene sequencing (data not shown). The bacterium isolated from the Sossego Mine was cultivated in LB medium (Luria-Bertani) composed of 10 $\mathrm{g\,L^{-1}}$ peptone, 10 $\mathrm{g\,L^{-1}}$ NaCl and 5 $\mathrm{g\,L^{-1}}$ yeast extract. A total volume of 1 mL of culture sample in the exponential growth phase ($\mathrm{D.O.}_{600\,\mathrm{nm}} = 0.8$) was collected and centrifuged at 5000 g for 10 min. The cells were submitted to total protein extraction following the manufacturer's protocol and then analyzed by MALDI Microflex LT (Bruker Daltonics, Bremen, Germany) using the BioTyper 3.0 program to compare the acquired spectra to the reference database (total of 5625 species). Scores higher than 2.0 are reliable for genus and species identification; scores between 1.7 and 2.0 are reliable for genus identification; and scores under 1.7 are not reliable for identification and represent no similarity between the spectrum acquired and the spectra in the database. The spectra from the *R. erythropolis* were acquired in triplicate in three different identification events; in other words, they were acquired in three different protein extractions from three independent cultures.

2.2. Copper stock solution

Copper ($\mathrm{Cu^{2+}}$) ion solutions were prepared using copper chloride (Sigma–Aldrich) as reagent. The final concentrations for the experiments were prepared from dilutions of a concentrated stock solution equal to 10 $\mathrm{g\,L^{-1}}$ $\mathrm{Cu^{2+}}$. The stock solution was prepared using Milli-Q water and then filtered with a 0.22 μm membrane filter (Millipore) and stored at 4 °C.

2.3. Biomass preparation

The *Rhodococcus erythropolis* strain was grown in LB medium, under orbital shaking at 180 rpm and 28 °C. Cells were collected at the end of the exponential phase and then centrifuged in a CR22N centrifuge (Hitachi, Japan) at 10,000 × *g* for 10 min. The culture medium was totally discharged and the cell mass washed three times with saline solution (NaCl 0.9 $\mathrm{g\,L^{-1}}$). The cells were suspended in saline solution and freeze-dried in L101 freeze-drier (Liotop, Brazil). After lyophilizing, the biomass was autoclaved at 121 °C for 20 min and dried in an oven Esco Isotherm (Esco, Singapore) at 105 °C for 12 h.

2.4. Biosorbent characterization

The approximated composition of the pure pellet with the biomass was determined by energy dispersive X-ray spectroscopy (EDX) using an EDX LINK ANALYTICAL, (Isis System Series 200), SiLiPentafet detector, resolution of 133 eV to 5.9 keV, coupled to a scanning electron microscope LEO 440.

Prior to the analysis, pure pellets were made with the biomass, that is, without the presence of KBr. Each tablet was glued onto carbon double-sided adhesive tape and then the EDX analyses were performed. The analyses occurred by collecting four regions of the biomass surface.

2.5. Biosorption experiments

The biosorption experiments were carried out through the contact between the biomass and the Cu^{2+} solution. The copper ion solutions were prepared from stock solution of copper chloride in Milli-Q water in different concentrations. Experiments ($V_F = 50\,mL$) were performed in 100 mL Erlenmeyer flasks incubated under horizontal shaking at 150 rpm and 28 °C for 120 min. Samples were collected at 0; 1; 5; 20; 60; 90 and 120 min, and analyzed for copper removal content (R%) using Eq. (1):

$$R(\%) = \frac{C_i - C_f}{C_i} \times 100 \qquad (1)$$

where C_i is the initial concentration of the metal and C_f is the final concentration. In addition to R%, the biosorption capacity of the bacteria was also analyzed using Eq. (2):

$$q_e = \frac{C_i - C_e}{m} \times V \qquad (2)$$

where q_e is the adsorbed metal amount in milligrams (mg) by bacterial mass in grams (g), "m" represents the mass of biomass used in grams (g), C_e is the equilibrium concentration and V represents the volume of the experiment in liters (L).

The analyzed pH values were 4.00, 5.00 and 6.00 ± 0.20. The analyzed cell concentrations were $0.1\,g\,L^{-1}$, $0.5\,g\,L^{-1}$, $1.0\,g\,L^{-1}$ and $2.0\,g\,L^{-1}$. The metal concentrations analyzed were $0.5\,mg\,L^{-1}$, $5.0\,mg\,L^{-1}$, $10\,mg\,L^{-1}$, $50\,mg\,L^{-1}$, $100\,mg\,L^{-1}$, $200\,mg\,L^{-1}$ and $300\,mg\,L^{-1}$.

2.6. Isotherms

The copper adsorption capacity was evaluated using isotherms. The graph obtained for this evaluation correlates the values of residual metal concentration and the uptake capacity (q_e) of sorbent used. The most widely used and reported models in the literature are the Langmuir [22] and Freundlich models.

The metal uptake capacity (q_e) is given in milligrams of metal per gram of the biomass ($mg\,g^{-1}$) and the residual concentration of C_e in milligrams of metal per liter of solvent ($mg\,L^{-1}$) in the medium. The general Freundlich equation is presented in Eq. (3):

$$q_e = k_f C_e^{1/n} \qquad (3)$$

and its linearized form in Eq. (4)

$$\log_{q_e} = \log k_f + \frac{1}{n} \log C_e \qquad (4)$$

where $\log k_f$ is the measure of the capacity of the sorbent and $1/n$ is the intensity of adsorption.

The general equation of Langmuir can be presented in Eq. (5):

$$q_e = \frac{Q_m b C_e}{1 + b C_e} \qquad (5)$$

And its linearized form in Eq. (6)

$$\frac{C_e}{q_e} = \frac{1}{Q_m b} + \frac{C_e}{Q_m} \qquad (6)$$

where q_e is the metal uptake ($mg\,g^{-1}$), C_e is the concentration of the metal at equilibrium ($mg\,L^{-1}$), Q_m is the maximum adsorption capacity and b is Langmuir constant related to affinity of adsorption.

3. Results and discussion

3.1. Microorganism identification

The isolated strain from the Sossego Mine in Canaã dos Carajás was identified by MALDI-TOF mass spectrometry using the BioTyper (Bruker, Germany) program. The spectra acquired from de study strain (Fig. 1) obtained score 2.04 with *R. erythropolis* spectrum from the database. This score is suitable for species identification according to the manufacturer. The BioTyper technology compares the mass from ribossomal proteins to the spectrum database. The ribossomal protein spectrum of a bacteria is a type of fingerprint and each bacterium has its own spectrum independently of the culture media [23]. The use of mass spectra generated by MALDI-TOF technology to identify bacterial strains is fast, simple and sample preparation is cheaper when compared to classical molecular biology identification. A concern about this new method is the reliability of the identification, once its database is small when compared to ribossomal database and most of the BioTyper database is represented by medical important bacteria. Despite all the doubts about identification reliability, several studies showed that this technique can be used for general bacterial identification and also for environmental isolated strains with good rates of success [21,24–27]. With a high mass sensitivity, the mass comparison is very accurate when similar spectra in database is present. In our case, *R. erythropolis* spectrum is present in databases and as its score is above 2.0, the identification can be trusted with almost zero probability of misidentification.

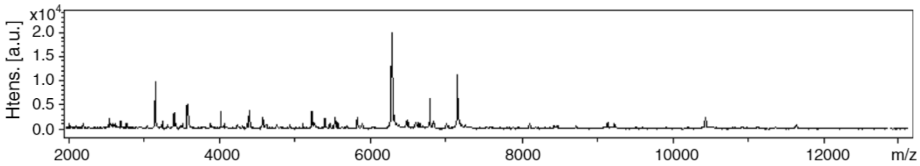

Fig. 1 – Protein spectrum provided by MALDI TOF for identifying *Rhodococcus erythropolis*.

3.2. *Biosorption experiments parameters*

The metal adsorption by biomass can be affected by some physio-chemical parameters intrinsically related to the nature of biomass. Different biomasses can be differently influenced by different parameters. To test the *R. erythropolis* biomass in copper adsorption, a variation of pH of aqueous solution was made focusing on the balancing of electrically loaded species found in the biomass and its response regarding the best rates of copper adsorption. Likewise, the quantity of biomass is another factor that can influence the copper uptake. Large amounts of biomass can interfere with biosorption forming mass agglomerates and decreasing the biomass maximum capacity of copper uptake [28]. Lastly, low concentrations can underestimate the capacity of biomass adsorption and the best relation between biomass and copper concentration must be reached.

3.3. *Hydrogen potential influence (pH)*

The biosorption experiments were carried out without and with pH control with a fixed relation between copper ($100\,\mathrm{mg\,L^{-1}}$) and *R. erythropolis* biomass ($1\,\mathrm{g\,L^{-1}}$). In the experiment with pH control, the determined pH values of 4.0, 5.0 and 6.0 were controlled all along the experiment, whereas in the experiment without pH control, the pH was adjusted to the desired values at the beginning only and no pH control was used during the experiment. These values of pH were chosen to represent the desorption point (pH = 4); the precipitation point where copper starts to precipitate only due to the pH (pH = 6) and the intermediary value (pH = 5). Fig. 2 shows the behavior of copper adsorption when pH is not controlled during the experiment. According to this graph, copper uptake occurs in the first minutes of contact for all pHs tested. For pH = 6, the percentage of copper removal by biomass was higher (22.0%) when compared to other pHs tested (17.8% for pH = 4 and 15.0% for pH = 5). During the biomass/copper contact over time, we did not observe severe changes in copper adsorption; for all the experiments, a tendency of pH stabilization between pH = 4.5 and pH = 5.5 occurred followed by copper removal stabilization between 15.0% and 22.0% of the total copper in solution. When pH is controlled (Fig. 2), the copper uptake capacity is reached in up to 20 minutes and throughout the experiment, no severe changes could be observed as occurred in the experiment with no controlled pH. The major and important difference in this experiment was the copper removal rate for pH = 6.0. In this pH, the maximum copper removal was 58.0%, which means about 2.6 times more when compared to the same pH in the experiment without pH control. The maintenance of pH = 4.0 for *R. erythropolis* biomass resulted in a decrease of metal removal from 17.8% to 11.7%, whereas at pH = 5.0, we observed an increase of only 3.4% in copper uptake as compared to pH = 5.0 in the experiment without pH control (pH = 5.0, Fig. 2).

Our results indicate the *R. erythropolis* biomass presents an elevated capacity to uptake copper in pH = 6.0 as the optimum for absorption. The pH maintenance can drastically improve the metal adsorption and it alone is responsible for differences of more than 34% in total copper removal.

3.4. *Biomass*

Once the influence of the pH in copper adsorption by *R. erythropolis* biomass is tested, experiments relating the mass of copper adsorbed versus biomass of *R. erythropolis* (q_e) were performed. As already shown, copper uptake occurred in the first 20 min of the experiment reaching the maximum capacity, which was almost constant throughout the sampling time. Without the controlled pH, the maximum absorption capacities were $24.3\,\mathrm{mg\,g^{-1}}$; $16.5\,\mathrm{mg\,g^{-1}}$ and $24.8\,\mathrm{mg\,g^{-1}}$ for pH = 4.0, pH = 5.0 and pH = 6.0, respectively. According to these data, variation in copper adsorption by the biomass was low and saturation for uncontrolled pH was around $16.0\,\mathrm{mg\,g^{-1}}$ and $25\,\mathrm{mg\,g^{-1}}$ for all the pHs tested. When controlled pH is used, the *R. erythropolis* biomass could adsorb up to 70 mg of copper per gram of biomass at pH = 6.0, which means 2.8 times more than at pH = 6.0 in the non-controlled pH experiment. For pH = 4.0, no consistent differences could be observed between the control and the experiment with controlled pH. For pH = 5.0, the controlled experiment resulted in an increase of copper uptake by 1.5 times, from $16.5\,\mathrm{mg\,g^{-1}}$ in the non-controlled pH, up to $26.0\,\mathrm{mg\,g^{-1}}$.

The capture of metallic cations by the microbial mass has been reported to be influenced by hydrogen ion concentration mainly in pH between 2.5 and 6.0 [29,30]. Higher pH values increase the negative character of the cell surface, favoring electrochemical attractions and increasing the adsorption of metallic cations. In turn, lower pH values favor anion adsorption, since they tend to make the character of the cell surface positive, protonating its functional groups [15,17,29,31]. Taking these studies into consideration, with a pH control, we may guarantee the negative character of the biomass even with the increase of hydrogen protons in solution by the displacement of this ion by metal protons. In this situation, the availability of ligand sites is maintained by the constant addition of OH^- groups, which prevents the competition between H^+ and metal cations, increasing the capacity of metal adsorption to the maximum performance related to the biomass constitution.

Not only can pH influence the metal adsorption by biomass, but another important factor is the relation between biomass

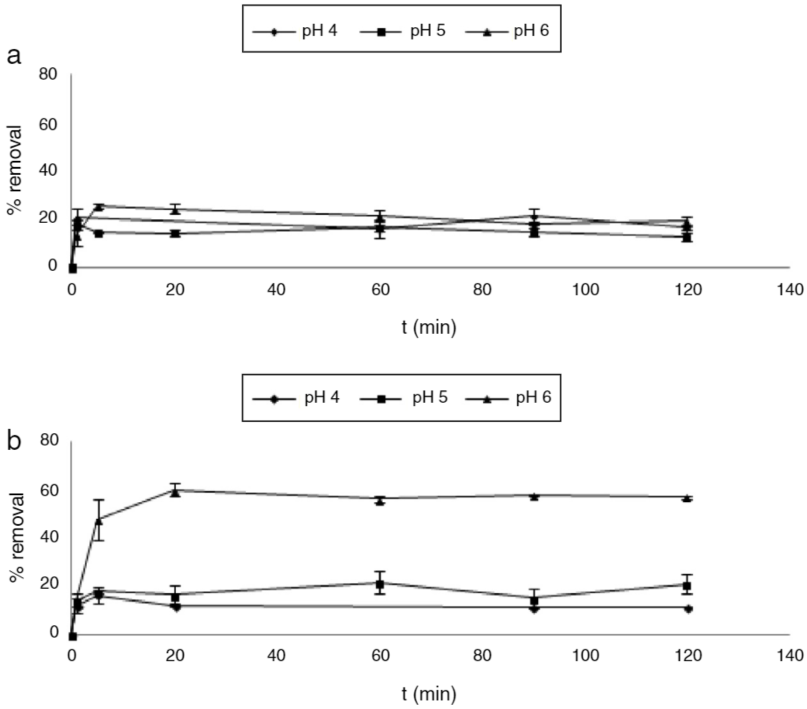

Fig. 2 – (a) *R. erythropolis* Cu^{2+} removal percentage data, in an experiment with no pH control, where $[Cu^{2+}] = 100\,mg\,L^{-1}$, [biomass] = $1.0\,g\,L^{-1}$ and pH values = 4.5 and 6; (b) *R. erythropolis* Cu^{2+} removal percentage data, in an experiment with pH control, where $[Cu^{2+}] = 100\,mg\,L^{-1}$, [biomass] = $1.0\,g\,L^{-1}$ and pH values = 4.5 and 6.

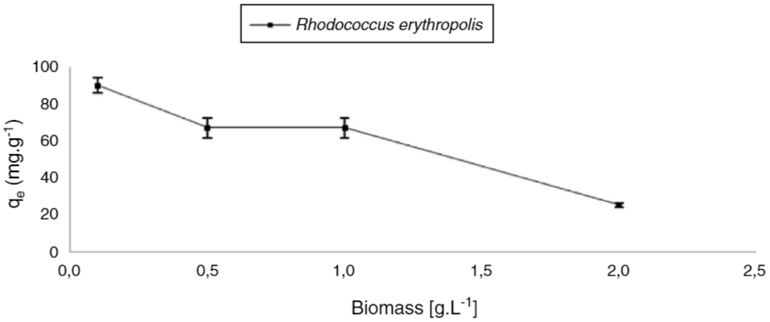

Fig. 3 – Effect of biomass concentration on copper biosorption ($[Cu^{2+}] = 100\,mg\,L^{-1}$, pH 6, $t_{contact} = 120\,min$).

concentration in copper solution [32,33]. Fig. 3 shows the copper biosorption capacity (q_e) for R. *erythropolis* in the optimal pH (pH = 6.0) for different biomass concentrations: $0.1\,g\,L^{-1}$, $0.5\,g\,L^{-1}$, $1.0\,g\,L^{-1}$ and $2.0\,g\,L^{-1}$ (Fig. 3).

As the figure shows, the optimized copper uptake was achieved in $0.1\,g\,L^{-1}$ biomass concentration with an adsorption rate of $86.00\,mg\,g^{-1}$. This rate is very high compared to other studies already published [20,28,34–36] and to our knowledge, never seen before in copper adsorption by the *Rhodococcus* genus biomass, making this strain one of the most promising biomasses for copper adsorption.

With the increase of biomass concentration, the q_e decreased and had its lower value at $2\,g\,L^{-1}$ ($q_e = 26.41\,mg\,g^{-1}$), 3.3 times lower than $0.1\,g\,L^{-1}$ ($q_e = 86.00\,mg\,g^{-1}$). The low concentration of biomass optimizes the surface contact [32,33] between copper and the sorbent, making available all the possible copper ligation sites. However, the increase of the biomass concentration disturbs the biosorption process due to the biomass aggregation effect [28], reducing its availability due to the blocking of copper ligation sites by biomass-biomass ligation, which is undesirable in adsorption processes.

3.5. *Biosorbent characterization*

To complement the results discussed so far, characterization trials of the biosorbent were carried out to determine the possible functional groups available for capturing the copper metal ions. Fig. 4 shows the spectrum obtained in the FTIR analyses.

According to the signals obtained in the FTIR analyses, functional groups such as carbonyls, carboxylic acids and amides can be stated to be present in the biosorbent structure to capture metallic ions, in this case, copper ions. The evidences are indicated in the peaks presented in the

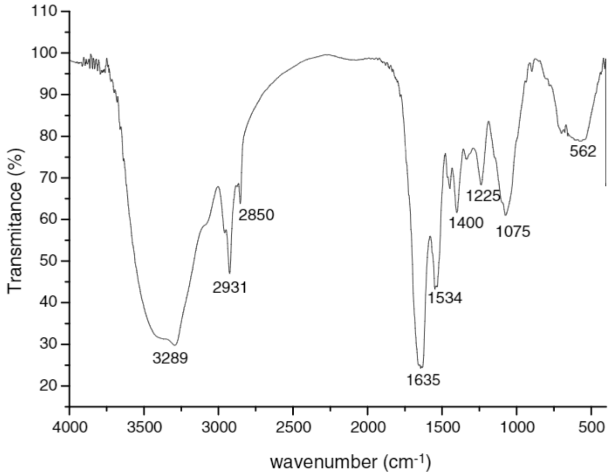

Fig. 4 – Biosorbent characterization spectrum obtained by FTIR.

regions of 1800 cm^{-1} to 1600 cm^{-1}, 3200 cm^{-1} at 2500 cm^{-1}, and 3500 cm^{-1} at 3070 cm^{-1}, respectively. The use of this technique for characterizing organic functional groups participating in biosorption phenomena is widely used and reported in the literature [37–39], corroborating the brief discussion as follows about the interaction of these groups with the metallic ions.

Biosorption phenomena occur through physical-chemical interactions between the metals and the functional groups present on the cell surface. They are based on physical adsorption and ion exchange, phenomena that can be performed with dead biomass, since they are independent of the active metabolism [40]. The functional anionic groups are known to be present in the peptidoglycans, teicuric and theicuronic acids of Gram-positive bacteria; the peptidoglycan, phospholipids and lipopolysaccharides of Gram-negative bacteria are the main components responsible for the anionic character and, consequently, for the ability of the cell wall to pick up metals. As current examples in the literature, the work by Amirnia 2015 [41] and Bueno *et al.* 2008b indicates that the carboxylic, hydroxylic and amine groups present in the microbial cell wall were responsible for the binding and capture of copper.

3.6. *Copper concentration*

Copper concentration can influence adsorption rates independently of the biomass used. In order to test the capacity of the *R. erythropolis* biomass in adsorb different copper concentration, experiments were performed with a fixed biomass concentration (0.1 $g L^{-1}$) and variation of copper in solution (0.5 $mg L^{-1}$ to 200 $mg L^{-1}$). The experiments data show the highest removal rate achieved by *R. erythropolis* was 40% at the minimum concentration studied, the optimal performance for copper uptake. The increase of copper concentration causes a drop in the percentage of copper removal [33], which indicates the saturation of the biomass reaching its maximum copper uptake capacity. This saturation capacity (q_e) was verified at optimal values of pH and biomass concentration.

Table 1 – Freundlich and Langmuir isotherm constants and regression coefficients at optimum conditions.

	Rhodococcus erythropolis					
	Langmuir			Freundlich		
ppH 6	*b* (L/mg)	q_{max} ($mg g^{-1}$)	R^2	K_f	1/*n*	R^2
	0.0939	68.03	0.9964	4.8787	0.6279	0.9639

According to the data acquired, with the raise of copper concentration up to 200 $mg L^{-1}$, the q_e reached 102 mg of copper per gram of biomass. With the increase of the metal concentration in the medium, more ions are available for adsorption process increasing the q_e value [33,42,43]. The same behavior of adsorption related to the increase of metallic concentration in the medium was already well reported in the literature [20,28,38]. Although the adsorption by microbial biomass has been well reported, copper adsorption by *R. erythropolis* biomass is poorly known as its adsorption rates, which seem to be one of the bests rates for biomass already described regarding biomass copper adsorption studies.

3.7. *Isotherms*

The experimental data obtained in the biosorption experiments were exploited using adsorption isotherms, more specifically the Langmuir and Freundlich models.

Table 1 shows the kinetics parameters discovered for calculating isotherms for the two models using experimental data. Analyzing the linear regression coefficients, both were above 0.9, providing acceptable adjustments for both models [44]. However, the model that was slightly more adequate was the Langmuir model.

Although Langmuir's isotherms do not correspond to the phenomenon of biosorption (ion exchange), it is the most utilized model once it uses only two very simple constants: q_{max}, which correspond to the higher rate of metal uptake, and coefficient *b*, which relates the affinity between the sorbent and the metal [45]. Low values of *b* indicate high affinity; therefore, good sorbents present high values of q_{max} and low values of coefficient *b* [45]. Thereby, according to the parameters obtained for the Langmuir model, *R. erythropolis* obtained a $q_{max} = 68.03\,mg\,g^{-1}$ at pH = 6.0, confirming this pH as more efficient for biosorption process. The values show that the biomass of *R. erythropolis* is an excellent biosorbent for capturing bivalent copper ions and there is no record in the literature for this species regarding copper biosorption. Some studies reported biosorption information for *Rhodococcus* genus only, as Bueno and co-workers, who reported a $q_{max} = 32.2\,mg\,g^{-1}$ for copper using *Rhodococcus opacus*, also at pH = 6.0 [28]. Another study of *R. opacus* conducted by Silvas and co-workers reported a $q_{max} = 48.2\,mg\,g^{-1}$ for a multi-elemental solution on the sum of all adsorbed metals [46]. In a study by Cayllahua & Torem [47], *R. opacus* presented a $q_{max} = 59\,mg\,g^{-1}$ for aluminum [47]. Another metal already studied focusing on the adsorption by *R. opacus* achieved a q_{max} of 94.3 milligrams per gram of biomass [28].

All these studies demonstrate the high efficiency and the novelty of copper biosortion using *R. erythropolis* biomass. This high efficiency experimentally detected is also indicated in Freundlich isotherm by the K_f value, which suggests high copper affinity and adsorption by biomass. Also, values of $1/n$ between 0 and 1 indicate a naturally favorable adsorption process [48]. In our study, the biomass of *R. erythropolis* presented values of $1/n$ between 0 and 1, which is highly favorable and considered efficient in copper adsorption. Comparing Langmuir and Freundlich models with the experimental data (Fig. 5), the Freundlich model overestimates the adsorption capacity, while the Langmuir model underestimates the adsorption capacity. Although neither of the models ideally represent the real adsorption capacity of *R. erythropolis* biomass, the Langmuir model is more adequate to predict the adsorption once all the points predicted by this model are closer to the experimental data than the Freundlich model, reflecting the real results in a more reliable way.

Although extremely dependent on a series of physiochemical factors such as pH, biomass and metal concentration, the low costs and eco-friendly alternative compared to activated charcoal, solvent extraction and chemical oxidation treatments for metal contaminated sites make biosorption one of the best alternatives. The possibility of the absorbed metal to be recovered makes biosorption even more attractive, once low concentration metal effluents can be re-mined, turning residues into potential mineral resources, reducing the extraction of non-renewable ones. Many microorganisms have already been reported to have biosorption capacities and are one of the most widely studied groups for this purpose. Contaminated environments deserve specific attention once they bear many resistant species that could adapt to, overcome and even use toxic metals in metabolism. Our study payed focused one of several existing metal contaminated areas and isolated a *R. erythropolis* strain with high biosorption capacities for copper. The biomass of the isolated strain was tested for pH, biomass concentration and copper concentration in aqueous environment. The *R. erythropolis* biomass presented an excellent copper adsorption capacity experimentally reaching 102.00 mg of copper per gram of biomass at pH = 6.0, which, according to our studies, was the best pH for copper uptake with this specific biomass. To study all the adsorption potential, the Langmuir and Freundlich models were used to measure copper adsorption, of which the Langmuir model was more suitable according to the data acquired experimentally.

As observed, the biosorption phenomena are extremely dependent on a series of physical and chemical factors discussed here, among others that may not have been explored so far. Although the results were very positive, our tests were carried out on model and monoelementary effluents, which greatly reduces the chance of possible interferences found in real effluents; these conditions could be adverse to the application of certain biosorbents.

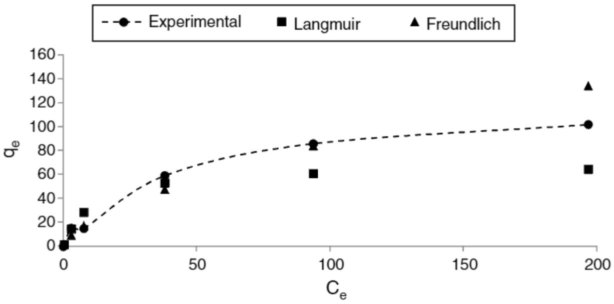

Fig. 5 – Comparative between the experimental data and the adjusted isotherms at pH 6.

4. Conclusion

In the Langmuir model, the adsorption potential of *R. erythropolis* biomass reached 68.03 mg of copper per gram of biomass, at pH 6, showing a removal difference of approximately 34%, when compared with experiments at pH 5. The characterization experiments were conclusive for determining the carbonyl and carboxyl functional groups, indicating the presence of organic compounds, such as carboxylic acids, amides and ketones, available for capturing copper ions. Besides the novelty about this bacterium not being reported on metal biosorption and the limited information about this genus, the isolated strain presented one of the highest copper uptake capacities already described. For this reason, *R. erythropolis* biomass should be exploited as an alternative to copper uptake both for environmental treatment purposes and to developed low concentration metal residues in mining.

Conflicts of interest

The authors declare no conflicts of interest.

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