



# Application of the Biomass from the Shell of *Pachyrhizus erosus* in the Removal of Hexavalent Chromium from Contaminated Waters

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## Authors' contributions

*This work was carried out in collaboration among all authors. Authors LLGF and VEMM wrote the protocol and wrote the first draft of the manuscript. Authors LLGF and VEMM and IAR managed the analyses of the study. Author IAR designed the study, managed the literature searches, formal analysis, writing-review, editing and supervision. All authors read and approved the final manuscript.*

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## ABSTRACT

**Aims:** Recently, the removal capacity of different heavy metals from sites contaminated by low-cost materials has been studied, with promising results. These adsorbents include dead microorganisms, clay minerals, agricultural waste, industrial waste, and other materials. The objective of this work was studying the removal capacity of Chromium (VI) by the biomass of the shell of *Pachyrhizus erosus*, by the Diphenylcarbazide colorimetric method.

**Place and Duration of Study:** Sample: Faculty of Chemical Sciences. Autonomous University of San Luis Potosí, S.L.P., between July and November 2021.

**Methodology:** The biomass was obtained from the shell of *P. erosus*, acquired in the Republic market of the city of San Luis Potosí, during the month of July 2021. For obtain the biomass, the shell was washed during 24 hours in EDTA at 10% (p/v), and 1 week with tridesionized water under constant stirring, with water changes every 12 hours, and we work with 100 mL of a 100 mg/L solution of Chromium (VI) obtained by diluting a 1 g/L standard solution prepared in tridesionized water from K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub>. The pH of the solution was adjusted with 1 M H<sub>2</sub>SO<sub>4</sub> and/or 1 M NaOH, before adding it to the biomass.

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**Results:** The highest bioadsorption was at 7 hours, at pH 1.0 and 28°C. Regarding temperature, the highest removal was at 60°C and 1.5 hours, with a removal of 90.69%. At the metal concentrations analyzed, the natural biomass showed a good removal capacity, in addition too efficiently removal the metal *in situ* (93.6% of contaminated water), at 3 days of incubation, with 5 g of biomass, and 100% of removal from contaminated earth, at 52 hours of incubation, with 5 g of biomass).

**Conclusion:** Therefore, this biomass can be used to remove it from industrial wastewater and contaminated earth.

*Keywords:* Removal; biomass; chromium (VI); *Pachyrhizus erosus*.

## 1. INTRODUCTION

Industrial and mining activity represents an important Public Health problem for the toxicity that causes the development of these and by the wide variety of sources of exposure, by releasing metals into the environment toxic such as lead, mercury, cadmium, arsenic, chromium, among others, harmful to the human health and for most forms of life. It has been reported that more of 90% of small and medium-sized industries tanneries in the country, make dumping of their chromium-contaminated effluents, without any type of treatment; polluting water sources throughout the territory national; causing damage to health and environment [1]. High concentrations of some of these elements in the environment or in certain processes can trigger a series of problems ranging from degradation and loss of soil fertility, until affecting processes wastewater treatment, since they inhibit the metabolism of the microorganisms used in the decontamination of resources natural (water, soil, and air) degraded by anthropogenic activities [2].

Common sources of wastewater that contain high concentrations of Chromium hexavalent; chemical compound considered as a carcinogen; come mainly of the metal cleaning industry, coating, curing, phosphate refining and bauxite, chlorine generation, manufacture of batteries and tanneries [3]. Heavy metals in general exert biological effects that are detrimental for most organisms; reason why have been cataloged as a class emerging cancer in humans. Exposure to Chromium causes several toxic effects, namely dermatitis, allergies, cancer, mutations, and teratogenic effects, which have been attributed to hexavalent Chromium ions [Cr (VI)] [4]. Chromium contamination and the impact on public health, has led man to seek alternatives to solve this problem, using

traditional methods such as: reverse osmosis, electro dialysis, ultrafiltration, ion exchange and chemical precipitation [5]; but the high cost of conventional methods led to the development of technological alternatives, which, in addition to take advantage of and apply the natural processes that occur in an ecosystem to purify a polluting residue, offer the possibility of recover the resources present in it for later use, also generating an economic value that contributes to sustainability of the system, about the Chromium (VI) removal process there are many investigations in this regard [6], like the use of the biomass of *Agaricus bisporus* for the bioremediation of this metal in solution [7], the application of *Coriandrum sativum* biomass in the removal of Chromium (VI) from polluted waters [8], the biosorption of the same metal in aqueous solution by *Ananas comosus* biomass shell [9], their removal of their by *Macadamia* nutshell powder [10], their adsorption by modified *Oyster* shell types [11], the adsorption of Chromium (VI) from the synthetic aqueous solution using chemically modified dried water hyacinth roots [12], and for the biomass from the coffee industry [13]. For the above, the objective of this work was studying the removal capacity of Chromium (VI) by the biomass of the shell of *Pachyrhizus erosus*.

## 2. MATERIALS AND METHODS

### 2.1 Bioadsorbent Used

The biomass was obtained from *P. erosus* shell acquired in the Republic Market of the capital city of San Luis Potosí, S.L.P., México, between the months July-August 2021 (Fig. 1). It was washed 24 hours with EDTA at 10% (w/v), one week with trideionized water with constant agitation, and water changes every 24 hours. Subsequently, it was boiled for 60 minutes to removal the dust and adhering organic

components, and it was washed again under the same conditions for 24 hours. It was dried at 80°C for 24 hours in a bacteriological oven, ground in a blender and stored in an amber bottle until use [9].

## 2.2 Chromium (VI) Removal Studies

A series of Chromium (VI) solutions ( $K_2Cr_2O_7$ ) (100 mg/L), the pH of the solution to be analyzed was adjusted with  $H_2SO_4$  1 M and/or 1 M NaOH, before adding it to biomass. The amount of biomass added to each flask is 1.0 g/100 mL

and 100 mg/L of metal solution, and was take samples at certain times, the biomass is removed by centrifugation (3000 rpm/5 min) and the supernatant was analyzed to determine the concentration of the metal ion by the Diphenylcarbazide colorimetric method, with which a complex of purple color is formed, and read at a 540 nm absorbance [14]. The removal of the metal was carried out at different pH's, temperatures, initial concentration of the metal and biomass, as well as the possible use of biomass to remove the metal from industrial waste (Figs. 2 and 3).

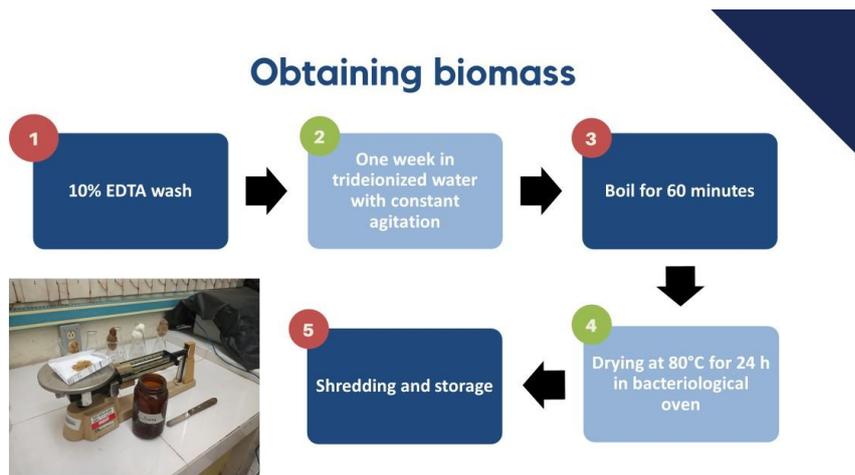


Fig. 1. Obtaining the cellular biomass from *P. erosus*

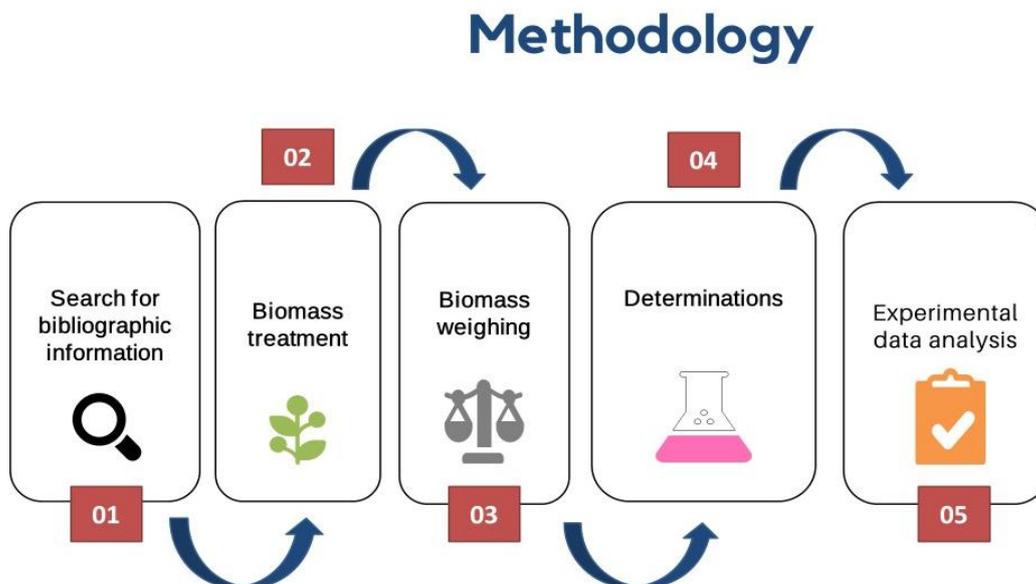
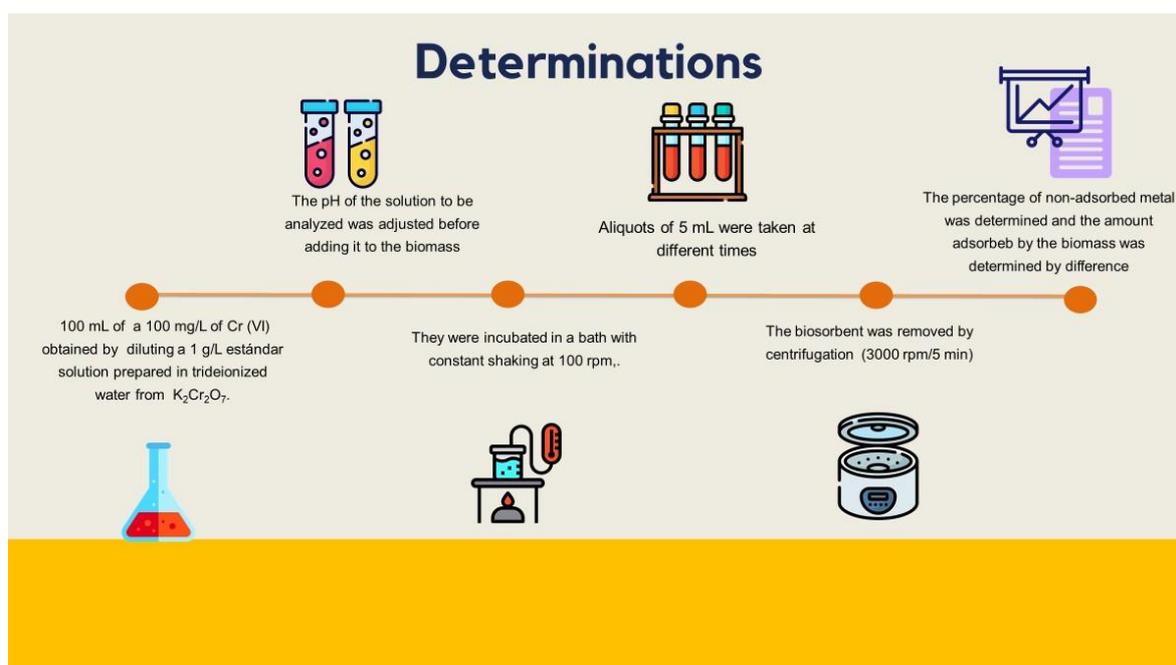


Fig. 2. Methodology used for the determination of Chromium (VI)



**Fig. 3. Determination of chromium (VI) concentration in solution**

### 2.3 Determination of the Optimum Time and pH for the Chromium (VI) Removal

For this determination, were prepared 4 solutions of Chromium (VI) standard (100 mg/L) in water trideionized; adjusted to different values of pH (1.0, 2.0, 3.0 and 4.0). Later it was added to each of the 4 Erlenmeyer flasks of 250 mL which contain 1 g of sterile biomass of *P. erosus*, and 100 mL of Chromium standard solution (VI), incubating them at 28°C in a bath with constant stirring at 100 rpm, then taken 5 mL aliquots at different incubation times, the bioadsorbent was removed by centrifugation (3000 rpm/5 min) and in the supernatant was determined the percentage of Chromium (VI) not adsorbed and by difference determines the amount adsorbed by the biomass. The optimal pH and incubation time determined by comparing the percentage metal removal rate measured every given time and pH analyzed.

### 2.4 Determination of the Optimum Temperature for the Chromium (VI) Removal

A standard solution of Chromium (VI) was prepared (100 mg/L) in trideionized water and adjusted to pH optimum obtained from the previous test. Subsequently, 100 mL of this

solution, were added to each of the 250 mL Erlenmeyer flasks, containing 1 g of *P. erosus* biomass, and incubated at 28°C, 40°C, 50°C and 60°C, in a bath with constant stirring at 100 rpm. Aliquots were taken 5 mL at different incubation times, and the cellular bioadsorbent was removed by centrifugation (3000 rpm/5 min) and the supernatant was determined the percentage of Chromium (VI) not adsorbed. The optimal incubation temperature was determined with the comparison of the percentages of Chromium (VI) removal of at the different temperatures at which were analyzed.

### 2.5 Effect of Initial Concentration of the Metal on the Bioadsorption of Chromium (VI)

Five standard solutions of Chromium (VI) were prepared (200, 400, 600, 800 and 1000 mg/L) in water trideionized, which were adjusted to optimal pH of removal obtained in the tests before made. Next, 100 mL of each Chromium (VI) standard solution were added to each of the 250 mL Erlenmeyer flasks containing 1 g of *P. erosus* biomass and incubated at 28°C and 60°C in a bath with constant agitation at 100 rpm, were taken aliquots of 5 mL at different times, removing the bioadsorbent by centrifugation (3000 rpm/5 min) and the supernatant was determined the concentration of Chromium (VI) not adsorbed.

## 2.6 Effect of Initial Concentration of Biomass on the Bioadsorption of Chromium (VI)

Some standard Chromium (VI) solutions (100 mg/L), in 100 mL of tri-deionized water and adjusted to pH optimum of removal were placed in 250 mL Erlenmeyer flasks, with different concentrations of the biomass under study, and were incubated at 28°C, in a bath with constant agitation at 100 rpm, taking 5 mL aliquots at different times, removing the cellular bioadsorbent by centrifugation (3000 rpm/5 min), and in the supernatant the concentration of Chromium (VI) not adsorbed was determined.

## 2.7 Bioremediation Assays

To two 250 mL Erlenmeyer flasks containing each 5 g of biomass, 95 mL of water and/or 5 g of earth contaminated with 100 mg/L of Cr (VI) (adjusted), from effluent from a Tannery of the city of Celaya, Guanajuato, Mexico. The mixture was incubated at 28°C with constant stirring (100 rpm), taking 5 mL aliquots at different times, removing the cellular bioadsorbent by centrifugation (3000 rpm/5 min), and in the supernatant the concentration of Chromium (VI) not adsorbed was determined.

## 2.8 Determination of the Concentration of Chromium (VI)

The concentration of the metal ion was determined by the Diphenylcarbazide colorimetric method, with which a complex of purple color is formed, which is read at a 540 nm absorbance [14].

## 3. RESULTS AND DISCUSSION

### 3.1 Effect of Incubation Time and pH

The optimum time and pH for the removal of Chromium (VI) (100 mg/mL) by the biomass of *P. erosus*, was 100% at 7 hours, pH 1.0, 28°C, 100 rpm, and 1.0 g/100 mL of bioadsorbent (Fig. 4), using a Corning Pinnacle 530 model pH meter and 1 M HNO<sub>3</sub> to keep the pH value constant, since the capture speed is controlled by the time at which the adsorbate it is transported from the outside to the inside of

the bioadsorbent particles [15]. These results are least efficient for those reported with the biomass of macromycete *A. bisporus*, which the removal was 100% at 21 minutes [7], for the *C. sativum* biomass, when the highest removal was observed within 3 hours, with 1.0 g of natural biomass, and 28°C [8], for the *A. comosus* biomass shell, the highest biosorption of the metal (100 mg/L) occurs within 10 hours, at pH of 1.0, 28°C with 5 g of natural biomass [9], too an optimum time of 2 and 4 hours for the removal of Chromium (VI) by *Macadamia* nutshell powder [10], 2 hours for the modified Oyster shell types, and chemically modified dried water hyacinth roots [11,12], an optimum time of 10 minutes using orange peel and wheat bran, for the removal of Chromium (VI) from wastewater [16], 6 hours for Chromium (VI) removal and total Chromium biosorption from aqueous solution by *Quercus crassipes* acorn shell [17], 30 minutes for the decontamination of the same metal on modified chicken feather [18], and 80 and 50 minutes with orange peels and *Fomitopsis pinicola* [19].

On the other hand, the highest metal adsorption was observed at a pH of 1.0 with the analyzed biomass (Fig. 4), and this is probably what the dominant species (CrO<sub>4</sub><sup>2-</sup> and Cr<sub>2</sub>O<sub>7</sub><sup>2-</sup> of Cr ions in solution, interact more strongly with the ligands carrying positive charges [20,21], and this is like that reported for the fungal *A. bisporus*, *C. sativum*, and *A. comosus* biomass, which the optimum pH of removal was 1.0 [7,8 and 9], too the same pH valor using orange peel and wheat bran, *Q. crassipes* acorn shell, and modified chicken feather [16,17 and 18], a pH between 1.1-2.0 using different types of biosorbents (*F. pinicola*, a mixture of cones, peach stones, apricot stones, *Juglans regia* shells, orange peels, and *Merino* sheep wool to removal Chromium (VI) from aqueous solution [19]. But these results are different for the adsorption ability for toxic chromium (VI) ions in aqueous solution of some modified Oyster shell types, in which is reported an optimum pH of 6.0 [11], a pH of 3.0 for the chemically modified dried water hyacinth roots [12], a pH of 9.0 for calcite based biocomposites [22], and an optimum pH of 2.0 for the bioremediation with *Arequipena* papaya seed (*Vasconcellea pubescens*) for total chromium removal [23].

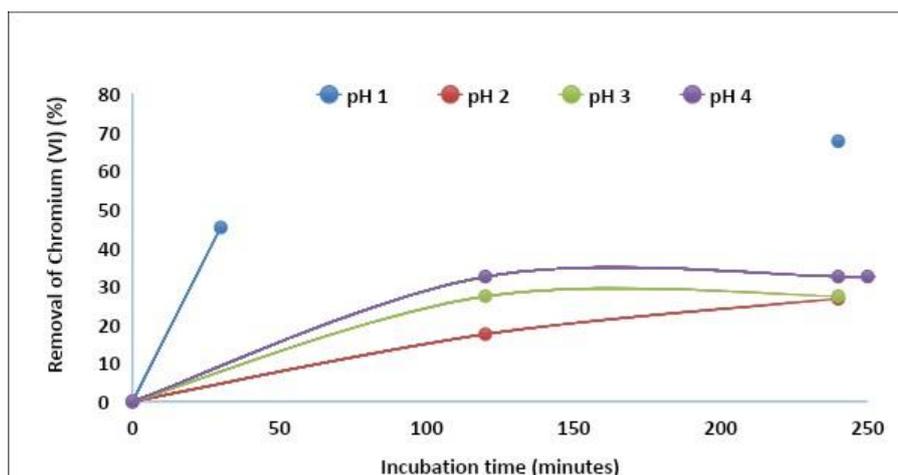


Fig. 4. Effect of pH and incubation time on the bioadsorption of Chromium (VI) in aqueous solution by the biomass of *P. erosus* 100 mg/L. 1 g of biomass, 28°C 100 rpm

### 3.2 Effect of Incubation Temperature

Also, it was found that at a higher temperature, the metal is removed in a greater proportion, since the temperature increases the active surface charge activity and the kinetic energy of the bioadsorbent, thus improving the removal of metal ions [6], therefore, at 60°C and 28°C, 90.69% and 90.7% of the metal in solution are removed in a time of 1.5 and 7 hours respectively (Fig. 5). These results are consistent with those reported for *C. sativum* biomass, when the highest removal was observed at 60°C, in 12 minutes, when the metal is completely adsorbed [8], for the *A. comosus* biomass, which the highest removal was observing at 60°C, in 3 hours, when the metal is completely adsorbed [9], 50°C for the decontamination of hexavalent chromium on modified chicken feather [18], if increase the temperature from 20°C to 45°C, too increase the Chromium (VI) removal with the modified biomass from wheat residues (*Triticum aestivum*) in wastewater [24], for Hibiscus sabdariffa flower biomass, the highest removal was observed at 50°C and 60°C in 40 minutes, when the metal is completely adsorbed [25], and for the removal of the same metal by the use of the residue of *Beta vulgaris* var. *Cyca* biomass [26]. But these results are different for the chemically modified dried water hyacinth roots, with an optimum temperature of 25°C [12], and for calcite based biocomposites, in which is increase the temperature decrease the removal of the metal [22], and an optimum temperature between 25°C to 30°C for the removal of Chromium (VI) using activated carbon from coffee lees [27].

### 3.3 Effect of Initial Concentration of the Metal on the Bioadsorption of Chromium (VI)

Regarding the effect of different concentrations of the metal in solution, at pH 1.0, with 1 g of biomass, at 28°C and 100 rpm, it was found that at a concentration of 200 mg/L, the greatest removal of the metal is evidenced, well 200 and 1 000 mg/L are removal at 1.5 and 4 hours respectively (Fig. 6A). On the other hand, it was also found that at 60°C, the metal concentration does not influence significantly in the removal of Chromium (VI), as at the temperatures analyzed the removal is similar between 10 and 25 minutes (Fig. 6B). This is like that reported for *A. bisporus* biomass, where at low concentrations of the metal (200 mg/L and 28°C), this biomass showed the best removal responses, adsorbing 100% at 60 minutes while with 1.0 g/L of Chromium (VI), removal was 90.3%, while, at 60°C, the biomass studied removal 100% at 90 minutes with 1 000 mg/L of the metal [7], for the removal of the same metal for *C. sativum* biomass, with a elimination of 100% at 4 and 24 hours, at 28°C, for 200 and 1000 mg/L, respectively [8], for the *A. comosus* biomass, which the highest removal was observing at 60°C, in 3 hours, when the metal is completely adsorbed, and at low metal concentrations (200 mg/L) a 28°C, was observe the best results for the elimination because the removal of the metal was 100% and 48%, at 24 hours for 200 and 1000 mg/L, respectively [9], for the chemically modified dried water hyacinth roots, with an optimum concentration reported of the metal was 10 mg/L [12], for calcite based biocomposites, in which is

increase the metal concentration of 250 to 2000 mg/L decrease the removal of the metal [22], if increase the initial concentration of the metal, from 1 to 3 g, decrease the Chromium (VI) removal with modified biomass from wheat residues (*T. aestivum*) in wastewater [24], and it has shown that as the initial concentration of hexavalent chromium increases, decrease the biosorption capacity of the activated carbon from the coffee grounds and therefore the efficiency [27], and these results are different for the modified Oyster shell types, and *Q. crassipes* acorn shell, which if increase the metal concentration too increase the removal of the same metal [11,17], for the decontamination of hexavalent Chromium on modified chicken feather, the percentage removal gradually increases with increasing initial concentration of the metal from 5 to 25 mg/L [18], and using orange peel and wheat bran, the concentration of the metal does not affect the removal of Chromium (VI) under the conditions selected for the wastewater [16].

### 3.4 Effect of initial concentration of *P. erosus* biomass on the bioadsorption of Chromium (VI)

The influence of the biomass concentration on the Chromium (VI) elimination capacity is shown in Fig. 7, in which it is observed that if the biomass concentration increases, the elimination of the metal in solution increases, since with 5 grams of biomass 93.3% of the metal is removed at 2.7 hours, while with 1.0 g, 52% is removed at the same time of incubation,

because there are more bioadsorption sites of it, since the amount of bioadsorbent added determines the number of binding sites available for the biosorption of heavy metals [20,21], and these results are like those reported for *A. bisporus* biomass, when If the amount of biomass is increased from 1 to 5 g, the removal of the metal in solution also increases, with 90.3%, with 1.0 g of biomass at 160 minutes, while with 5 g of biomass, the removal is 100 % at 100 minutes, at pH 1.0, 28oC and 100 rpm [7], for the *A. comosus* biomass, in which with 5 g of biomass at 10 hours, the metal is fully removed, while with 20 g of biomass, the removal is 100 % at 5 minutes, at pH 1.0, 28oC and 100 rpm [9], for the modified Oyster shell types, if increased the concentration of biomass of 1.65 mg/g to 2.92 mg/g, the removal capacity increase from 29.3% to 58.3% [11], for the chemically modified dried water hyacinth roots, with an optimum adsorbent concentration reported was 14 g/L [12], too, it was observed that quantitative removal of the hexavalent Chromium ion increases with increasing biosorbent dose and maximum removal was achieved by using 0.15 g/50 mL of chicken feather [18], if increase the initial concentration of the biomass, from 1 to 3 g, increase the Chromium (VI) removal with modified biomass from wheat residues (*T. aestivum*) in wastewater [24], and are different for the *C. sativum* biomass, if was increasing the amount of biomass, the removal of the metal in solution decreased slightly, well the removal obtained was observed between 100%-80%, with 1-5 g of natural biomass [8].

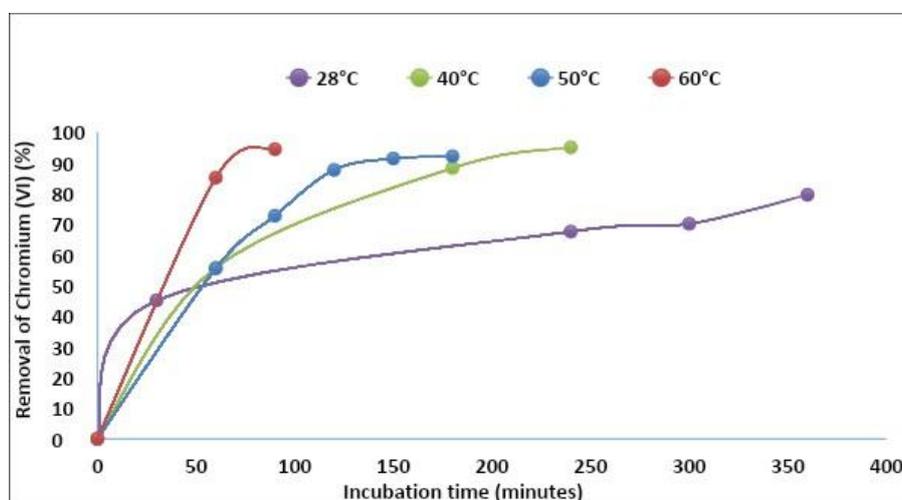
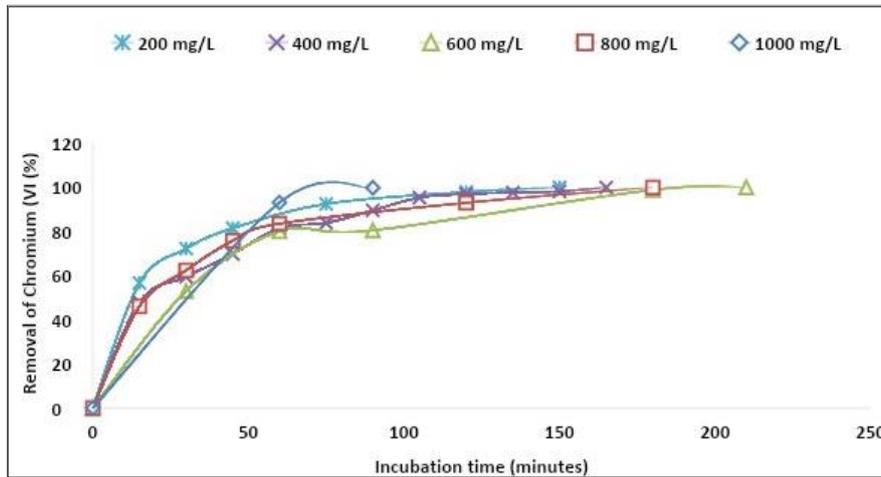
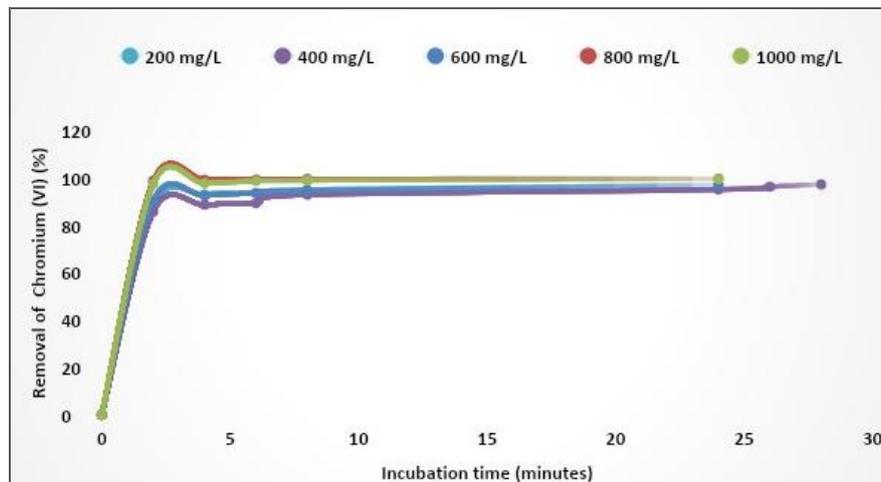


Fig. 5. Effect of incubation temperature on the bioadsorption of Chromium (VI) in aqueous solution by the biomass of *P. erosus* 100 mg/L. 1 g of biomass. 100 rpm



A



B

Fig. 6. Effect of initial concentration of Cr (VI) on the bioadsorption of Chromium (VI) in aqueous solution by the biomass of *P. erosus*. 1 g of biomass. 100 rpm. pH 1.0. 100 rpm

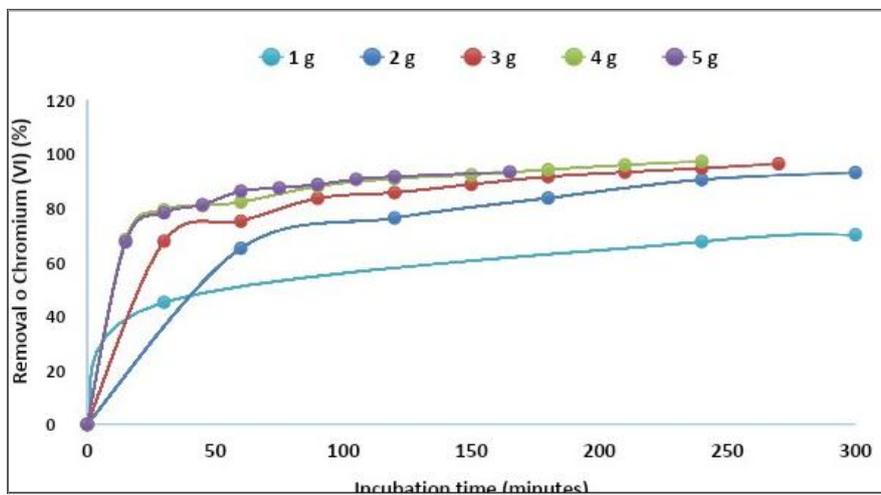
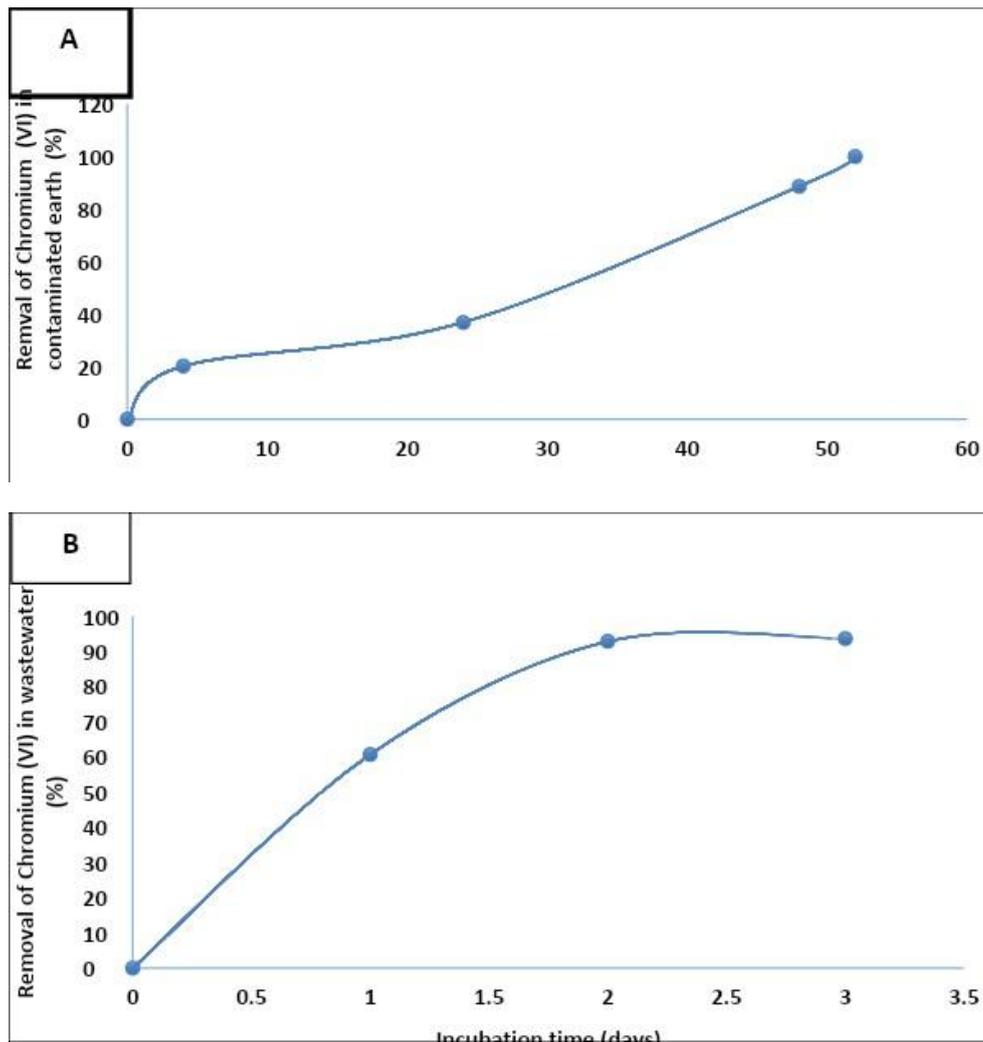


Fig. 7. Effect of initial concentration of the natural biomass on the bioadsorption of Chromium (VI) in aqueous solution. 100 mg/L of Cr (VI). 100 rpm. pH 1.0



**Fig. 8. Bioremediation of Cr (VI) from earth and water contaminated with 100 mg/g soil (pH 6.8), and 100 mg/L Chromium (VI) (pH 8.2) (5 g of *P. erosus* biomass, 28°C, 100 rpm. A: Earth, B: Water**

### 3.5 Bioremediation of Chromium (VI) from Earth and Water Contaminated

A bioremediation test was adapted in aqueous solution and earth to analyze the possible use of this biomass in the removal of the metal from industrial waste, therefore, 5 g of biomass were incubated with earth and wastewater contaminated [100 mg/L and 100 mg/g earth, of Chromium (VI) adjusted], at 28°C and 100 rpm (samples obtained from a tannery, from the state of Celaya, Guanajuato, Mexico), observing that after 52 hours 100% of the metal is removed from wastewater (Fig. 8A) and 93.6% of it at 52 hours, contaminated earth, respectively (Fig. 8B). The metal removal capacity from wastewater by the biomass is equal or better than others analyzed, for

example: The removal of the same metal by *A. bisporus* [7], *C. sativum* biomass [8], and *A. comosus* biomass [9], for the chemically modified dried water hyacinth roots [12], using orange peel and wheat bran [16], for catalytic removal of heavy metals from electroplating industrial effluent [22], for the bioremediation with Arequipeña papaya seed (*V. pubescens*) for total chromium removal [23], for the Chromium (VI) removal with modified biomass from wheat residues (*T. aestivum*) in wastewater from the Rio Seco and Uchumayo industrial park areas [24], for the removal of Chromium (VI) in aqueous solution by *H. sabdariffa* flowers biomass, and for the use of residue of *B. vulgaris* var. *Cycla* biomass [26], for the bioremediation of different hazardous metals from contaminated soil by *Nicotiana tabacum* plant

[28], and for the removal of heavy metals during primary treatment of municipal wastewater [29].

#### 4. CONCLUSION

Different methods have been exploited for water purification. Some processes are costly, complicated, time and energy consuming, skilled personnel, and especially create amine residues in the sludge. In the last decade, adsorption is evaluated as a promising alternative because it is the most efficient and thus most applied in water treatment, and thousands of adsorbents from available sources have been recommended in the literature to removal contaminants, for instance, clay minerals, biomass, materials from agricultural wastes, and by-products, and the biomass-based adsorbents have gained considerable attention thanks to their availability in huge quantities in most places worldwide, in particular, the use of waste biomass from invasive species as adsorbent materials for the removal of pollutants from aqueous streams could decrease their threat [19,30 and 31]. In this work, the biomass of a commercial legume plant of *P. erosus* was analyzed for the removal of Chromium (VI) in aqueous solution, with the following conclusions:

1. The biomass of *P. erosus* eliminates 100 mg/L of Chromium (VI) at 7 hours of incubation, with 1 g of biomass, 28°C, pH 1.0 and 100 rpm.
2. If the temperature is increased, the removal efficiency is increased.
3. To lower metal concentration, is higher the removal efficiency.
4. To higher biomass concentration, the removal efficiency increases.
5. In bioremediation tests, it was found that biomass efficiently removes metal from contaminated earth and waters with Chromium (VI), therefore, their application is viable for its treatment, in addition, the biomass used is natural, of easy obtained, handling, and affordable cost.

#### DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded

by the producing company rather it was funded by personal efforts of the authors.

#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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