

## Research Article

# Usefulness of the Biomass of Tobacco (*Nicotiana tabacum*) for The Elimination of Chromium (VI) from Polluted Waters

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**Abstract:** The tobacco plant is capable of accumulating heavy metals in its different parts, making it a good candidate for use in bioremediation, although there are few reports in which the biomass of this plant is used for the removal of heavy metals in solution. Also, cigarette residues are an environmental problem, so the use of these residues is an opportunity to obtain biomass for the removal of heavy metals from polluted environments. The objective of this work was to determine the removal capacity of Cr (VI) by commercial tobacco biomass, finding that 1 g of biomass removal 72 mg/L of the metal at 24 h, pH 2.0, 28 °C and 100 rpm, while at higher temperatures the removal is higher, and if the concentration of the metal is increased (1 g/L), its removal capacity is reduced, since 64.72% is removed at 24 h at 28 °C, although at 60 °C, 1 g/L is removal at 8 h. If the concentration of the bioadsorbent is increased, the metal removal does not increase. Finally, 5 g of biomass eliminates 66.1% and 74% of Cr (VI) present in naturally contaminated soil (100 mg/g) and water (100 mg/L), respectively.

**Keywords:** biomass; bioremediation; chromium (VI); heavy metals; tobacco

## 1. Introduction

Currently, due to mining, industrial, urban and smelting activities, one of the biggest environmental problems is heavy metal contamination of water sources around the world, because due to its toxicity and potentially harmful effects on the different ecological systems and the environment, are considered a serious problem for the inhabitants of the towns that are supplied with contaminated water, and considering that the increase in the concentration of these metals in the different water sources, is a consequence of the various anthropogenic activities, which are the support of human life, cause serious economic problems, both locally and nationally due to the increase in the costs of medical treatments and a decrease in the productivity of the inhabitants of the contaminated areas<sup>1</sup>.

The foregoing entails a large number of problems in the life of the planet, since in plants these metals end up deposited in the soil transported to them by polluted rivers, causing different effects such as: decreased growth or yellowing of the leaves (chlorosis). In addition to being very dangerous for human life, where the effects can be skin rashes, upset stomach and ulcers, respiratory problems, weakening of the immune system, kidney and liver damage, lung cancer, heart conditions, bone, testicular, central and peripheral nervous system or death<sup>2</sup>.

Therefore, it is of the utmost importance to find more efficient methods for the retention and extraction of polluting metals from contaminated places and reduce their toxicity to guarantee the preservation of ecosystems and human life. Among the different existing methods for the control of heavy metals we can find methods such as: precipitation, oxidation-reduction, ion exchange, filtration, electrochemical treatment, membrane technologies and recovery by evaporation, adsorption and bioadsorption, and the metals that are considered as heavy are: lead, tin, iron, cadmium, mercury, chromium, vanadium, among others<sup>3</sup>.

Secondly, hexavalent chromium, also known as chromium (VI) ( $\text{Cr}^{6+}$ ), is the toxic form of the metal chromium, while some fewer toxic forms of chromium occur naturally in the environment (soil, rocks, dust, plants, and animals), it is mainly produced by industrial processes<sup>4</sup>. Inhalation of this metal can cause cancer and non-cancer health effects. Cancer effects: Breathing chromium (VI) over a long period of time increases the risk of lung and nasal cancers, while non-cancer effects, such as breathing chromium (VI) at high levels over time, can cause or worsen certain health conditions, including: Irritation of the nose, throat, and lungs (runny nose, cough), allergy symptoms (wheezing, shortness of breath), nasal sores, and very high air levels in workplaces can cause perforation of the membrane that separates the nostrils, and is increasingly being recognized as a neurotoxicant<sup>4</sup>.

Different materials have already been studied as potential biosorbents to eliminate this metal. Those materials include microorganisms, like bacteria, mushrooms, algae, waste and lignocellulosic material, and others, like shellfish, which can be removal  $\text{CO}_2$  from the atmosphere<sup>5-7</sup>, and several studies have shown that metal bonding occurs especially through a chemical functional group (carboxyl and hydroxyl groups)<sup>8</sup>. Some reports in the literature that use low-cost materials for the elimination, reduction and/or removal of this metal are: oat biomass (*Avena sativa*)<sup>9</sup>, tella residue and pea seed shell (*Pisum sativum*)<sup>10</sup>, avocado seed<sup>11</sup> inert biomasses of *Dioscorea rotundata* and *Elaeis guineensis*<sup>12</sup>, amla wood sawdust (*Embllica officinalis*)<sup>13</sup>, rice husk<sup>14</sup>, *Arachis hypogea* husk<sup>15</sup>, *Heinsia crinita* seed coat biomass<sup>16</sup>, bagasse<sup>17</sup>, onion waste<sup>18</sup>, and modified biomass of rice husk (*Oriza sativa* L.)<sup>19</sup>.

On the other hand, the ability of tobacco plants to accumulate most of the heavy metals in their roots makes them an excellent candidate for use as a Phyto-stabilizer. However, further studies should be carried out to determine the mechanism that tobacco uses to fix these metals in its roots. In previous works it was found that tobacco (*Nicotiana tabacum*) is capable of accumulating heavy metals, because this plant grows in the presence of a mixture of toxic metals, it becomes an accumulator of cadmium, magnesium and arsenic, through the Phyto-stabilization of heavy metals<sup>20</sup>, the cadmium and lead contents in cigarettes and tobacco leave<sup>21</sup>, the elimination of mercury by roots, leaves and stems of tobacco transgenic for the merApe9 gene<sup>22</sup>, the endocytic uptake of uranium in tobacco BY-2 cells<sup>23</sup>, the uptake of cadmium, chromium, and lead, by two tobacco cultivars<sup>24</sup>, the accumulation of lead by *N. tabacum*<sup>25</sup>, and the phytoextraction of cadmium and lead<sup>26</sup>, but there are few reports regarding the removal of heavy metals by the biomass of natural and/or commercial tobacco. Therefore, the objective of this work was to analyse the removal capacity of chromium (VI) in aqueous solution by the biomass of commercial tobacco.

## 2. Materials and Methods

### 2.1 Biosorbent

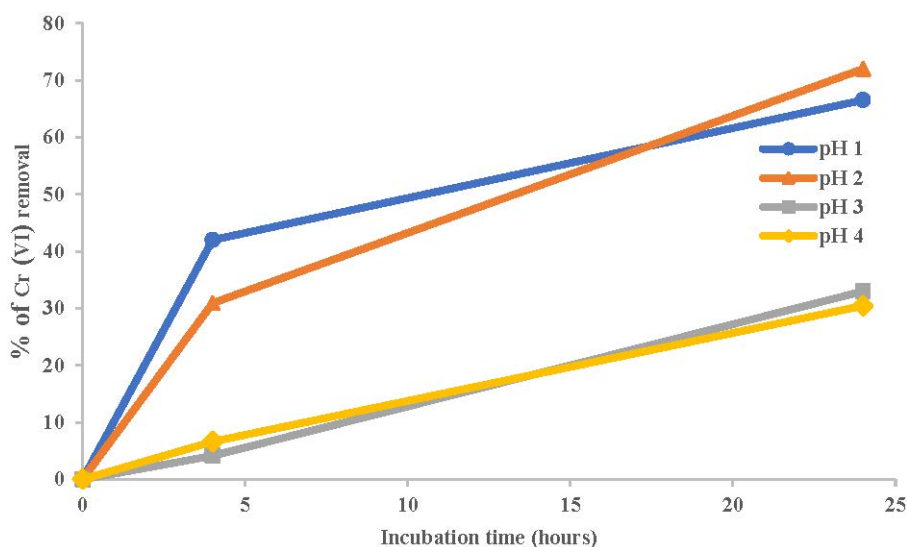
The commercial tobacco was obtained from a convenience store, located in a commercial plaza in the city of San Luis Potosí, S.L.P., México. To obtain the biomass, the tobacco was washed for 72 h at 28 °C with EDTA at 10% (w/v) in trideionized water, changing the solution every 12 h. Subsequently, it was washed with trideionized water for a week at 28 °C, with water changes every 12 h, and boiled for 60 min to remove dust, and the colour of the solution. It was dried at 80 °C for 24 h in a bacteriological oven, and the product was ground in blender, sterilized at 115 °C for 30 min, stored in amber vials until use.

## 2.2 Biosorption Studies and Determination of Hexavalent Chromium

For this, was utilized 1 g of dried biomass mixed with a solution of 100 mg/L of chromium (VI) in 100 mL of trideionized water, in an-Erlenmeyer flask at the different conditions analysed. The flasks were incubated at 100 rpm on a shaking bath Yamato BT-25 model. Samples of 5 mL were taken at different times and centrifuged at 3000 rpm for 5 min. The supernatant liquid was separated and analysed for chromium (VI) ions by a spectrophotometric method employing diphenylcarbazide<sup>27</sup>. The information shown in the results section are the mean from three experiments carried out by triplicate.

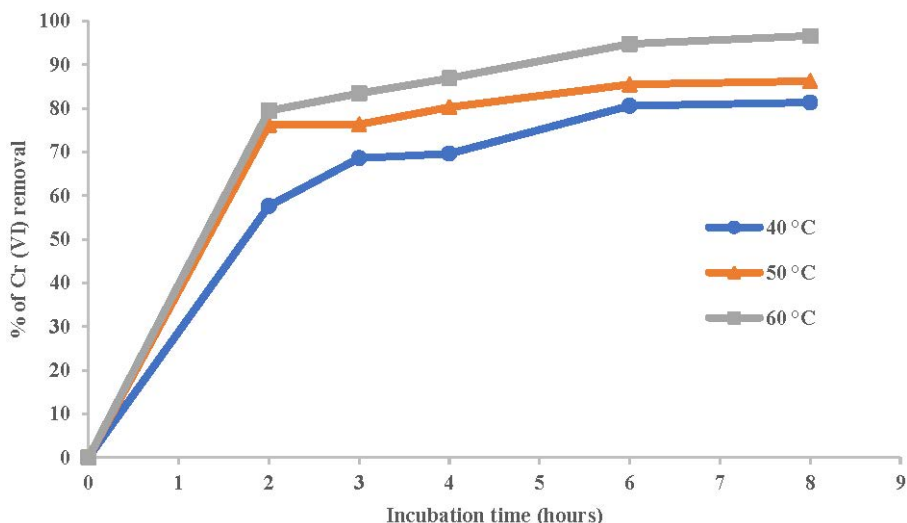
## 3. Results

The optimum time for chromium (VI) removal for *N. tabacum* biomass was 24 h, pH 2.0, with 1 g/100 mL of biosorbent, at initial metal concentration of 100 mg/L, and 28 °C (Figure 1). It was used a pH meter Corning Pinnacle 530 model and we use nitric acid 1M to maintain the pH worth. The most adsorption efficiency of the metal was observing a maximum at pH 2.0 for the biomass analysed. As the initial pH values increased from 2.0 to 4.0, the removal efficiencies of chromium (VI) decreased from 72% to 30.46%, respectively, in the same conditions (Figure 1).



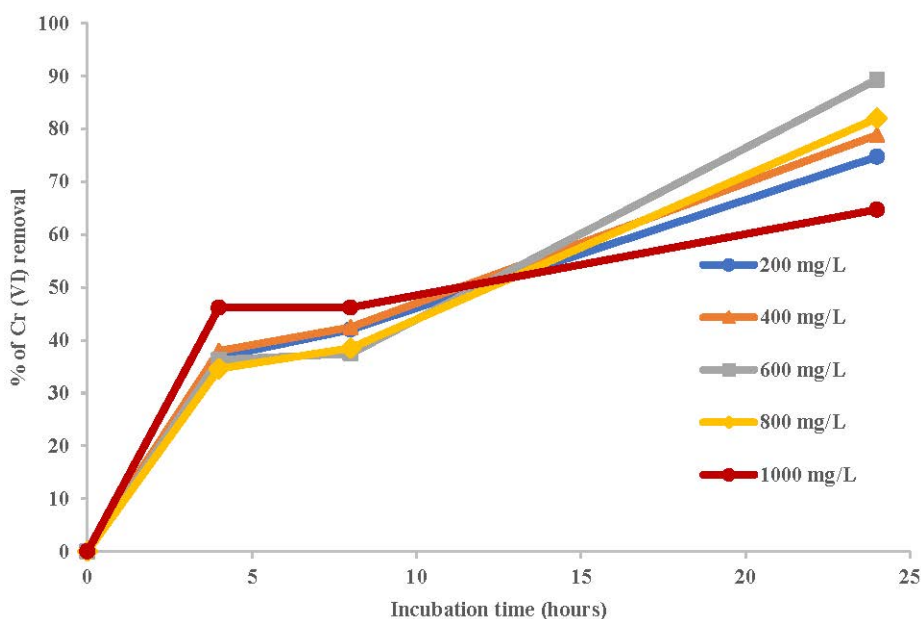
**Figure 1.** Effect of incubation time and pH on chromium (VI) removal by the biomass of *N. tabacum*. 100 mg/L Cr (VI), 100 rpm, 28 °C, 1 g of natural biomass.

In relation to the temperature, the highest removal was observed at 60 °C, since at 24 h 100% of the metal in solution is removed, while at 28 °C, 66.54% is removed at the same time (Figure 2).

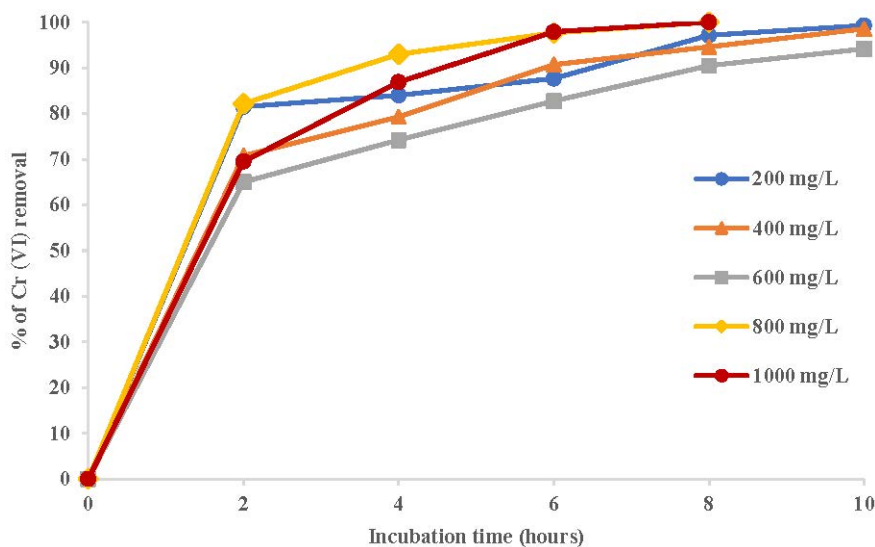


**Figure 2.** Effect of temperature on the removal of chromium (VI) by *N. tabacum*. 100 mg/L Cr (VI). pH 2.0. 100 rpm. 1 g of natural biomass. 24 h of incubation.

Regarding the effect of different concentrations of chromium (VI) in solution, on its removal, at a pH of  $2.0 \pm 0.2$ , with 1 g of *N. tabacum* biomass, at 28 °C, and 60 °C and 100 rpm. It was found that, at a higher concentration of the metal, the removal is greater, at 28 °C the maximum removal (89.36%) was at 600 mg/L, decreasing with 800 mg/L (82.02%) and 1000 mg/L (64.72%), while to 60 °C, the total removal of 800 mg/L and 1000 mg/L, was at the 8 h of incubation in the same conditions (Figures 3 and 4).

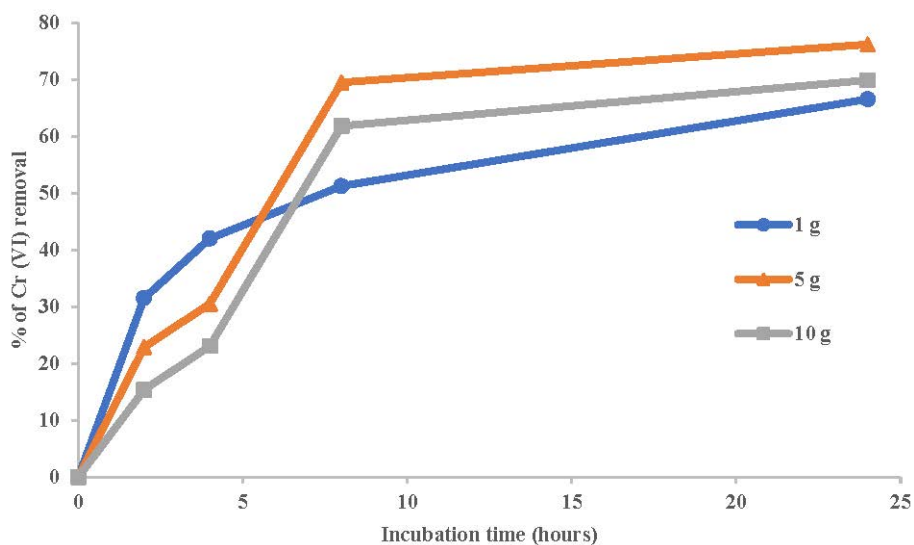


**Figure 3.** Effect of concentration of chromium (VI) (mg/L), on the removal of the metal by *N. tabacum*. pH 2.0. 28 °C. 100 rpm. 1 g of natural biomass. 24 h of incubation.



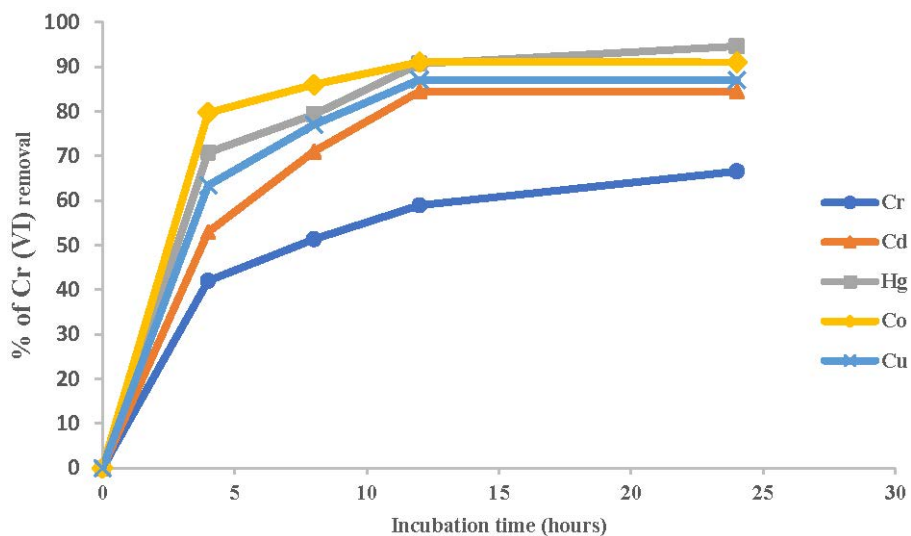
**Figure 4.** Effect of concentration of chromium (VI) (mg/L) on the removal of the metal by *N. tabacum*. pH 2.0. 60 °C. 100 rpm. 1 g of natural biomass. 24 h of incubation.

In Figure 5, the effect of biomass concentration on metal removal is observed. If the concentration of this is increased, the elimination of the metal in solution is not affected, because with the concentrations of biomass analysed, the removal is similar after 24 h of incubation.



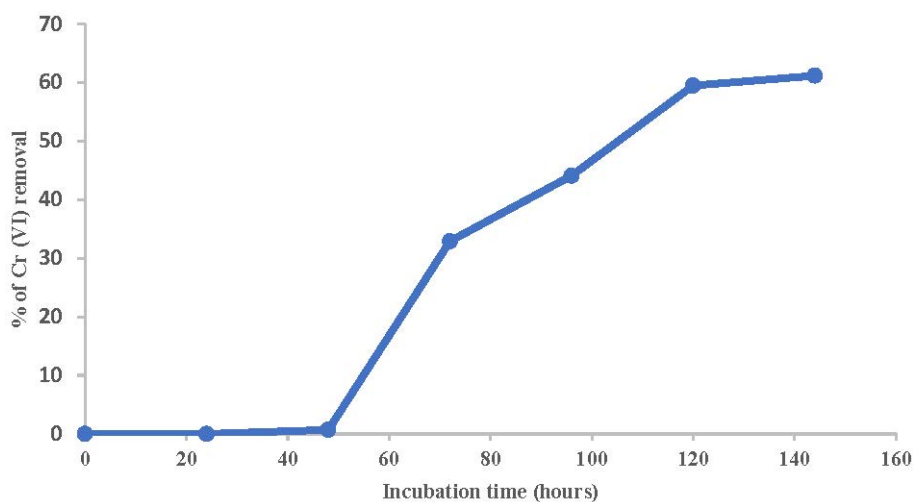
**Figure 5.** Effect of *N. tabacum* biomass (g/L) on the removal of 100 mg/L of chromium (VI). pH 2.0. 28 °C. 100 rpm.

The presence of other metals in solution such as cadmium (II), mercury (II), cobalt (II), and copper (II) (100 mg/L), do not interfere significantly with the removal of chromium (VI) in solution, because in the presence of the different heavy metals, an increase in the removal of chromium (VI) is observed (Figure 6).

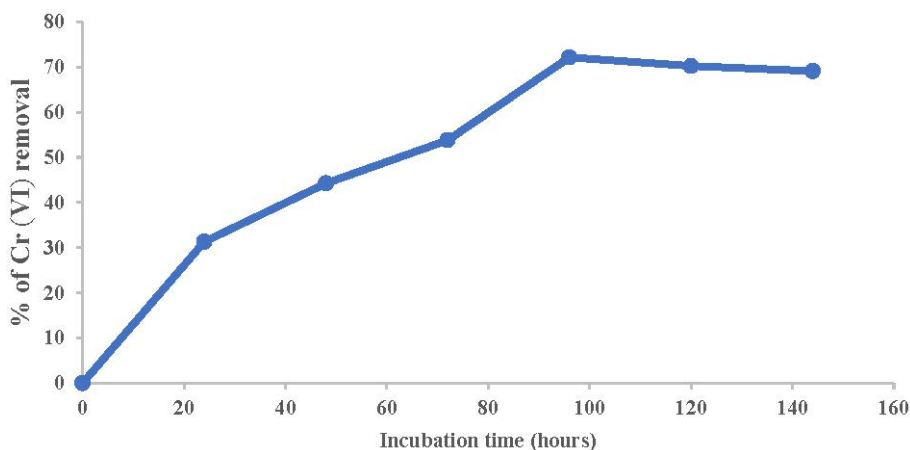


**Figure 6.** Effect of different heavy metals (100 mg/L) on bioadsorption of 100 mg/L of chromium (IV). 1 g of biomass. 24 h. 28 °C, pH 2.0, with constant agitation.

Finally, chromium (VI) bioremediation test was carried out from soil and water contaminated with 200 mg/L of the metal (adjusted), obtained from a tannery, located in Celaya, Guanajuato, México, with 5 of natural biomass, observing that this removal 66.1% and 74.2% of the metal, from the samples of soil and water contaminated, respectively, at 7 days of incubation (Figures 7 and 8).



**Figure 7.** Removal of chromium (VI) in industrial wastes incubated with 5 g of biomass. 28 °C, 100 rpm, 5 g of contaminated soil, (200 mg Cr (VI)/g soil, adjusted, pH 6.8).



**Figure 8.** Removal of chromium (VI) in industrial wastes incubated with 5 g of biomass. 28 °C, 100 rpm, 100 mL of contaminated water (200 mg Cr (VI)/L, adjusted, pH 8.2).

Finally, in the Table 1, we shown the adsorption percentages of chromium (VI) with different biosorbents.

**Table 1.** Comparison of percentages of adsorption of chromium (VI) with other biosorbents.

Adsorbent	pH	Adsorption Capacity (mg/L)	Reference
<i>Avena sativa</i>	1.0	100	9
<i>Pisum sativum</i>	2.0	5.0	10
<i>Dioscorea rotundata</i>	2.0	325.88	12
<i>Embllica Officinalis</i>	2.0–3.0	416.0	13
<i>Oriza sativa L.</i>	5.2	94.3	14
<i>Heinsia crinita</i>	2.0	49.45	16
<i>Allium cepa L.</i>	1.0	49.0	18
<i>Oriza sativa L.</i>	1.0	50.0	19
<i>Eichhornia crasipes</i>	1.5	2.5	28
<i>Nicotiana tabacum</i>	2.0	72.0	This work

## 4. Discussion

With respect to the optimum incubation time and pH for metal removal by *N. tabacum* biomass, the highest removal was to 24 h and pH 2.0. About, the *A. sativa* biomass, eliminate 100 mg/L of chromium (VI) after 8 h, pH 1.0, 1 g of biosorbent, and 28 °C<sup>9</sup>, with *D. rotundata*, the removal of hexavalent chromium, was of 325.88 mg/L, in 200 min, pH 2.0 (0.03 g de biosorbent)<sup>12</sup>, for *E. Officinalis* (2.5 g/L of biomass) was report a capacity of removal of 416 mg/L, in 100 min<sup>13</sup>, with *O. sativa L.* there was a biosorption of 94.3 mg/L, pH 5.2, 2 h at 28 °C<sup>14</sup>, and with the *A. cepa* biomass, the removal was of 49 mg/L [50 mg/L of initial concentration of chromium (VI)] pH 1.0, 28 °C, and 0.5 g/L of biomass<sup>18</sup>. The differences founded in these conditions, could partly explain, by changes in the permeability of unknown origin, providing greater or lesser exposure of the functional groups of the cell wall of the biomass analysed<sup>8,29</sup>.

On the other hand, industrial effluents often contain more than one type of metal ion, which can interfere in the elimination/recovery of the metal of interest by the biomasses to be studied<sup>8</sup>. In this work, the presence of other metals in solution such as cadmium (II), mercury (II), cobalt (II), and copper (II) (100 mg/L), does not interfere with the removal of the metal in solution in this study, but on the contrary, the removal of the metal under study increases from 72% to 84.5%–91%, in presence of the other heavy metals, and this coincides with some reports in the literature for other biomasses where it is reported like the biomass of two comercial strains of *Agaricus bisporus*<sup>30</sup>, the biomass of *Farfantepenaeus duorarum*<sup>31</sup>, for *Zhihengliuella* sp., ISTPL4 does not interfere the removal of different heavy metals in the presence of others<sup>32</sup>. Also, the presence of other metals does not significantly interfere with the adsorption of cadmium (II) by



*Pleurotus eryngii*<sup>33</sup>. On the other hand, it was found that the occurrence of  $\text{PO}_4^{-3}$ ,  $\text{NO}_3^{-}$  and  $\text{SO}_4^{-2}$  seriously affected the chromium (VI) sorption by rice husk<sup>14</sup>, and to the yeast *Saccharomyces cerevisiae*, in which the presence of heavy metals interferes with the removal of the same<sup>34</sup>.

Finally, it has been reported the ability of different microorganisms and natural biomasses, in order to analyse the possible use of biomass of *N. tabacum* for the elimination of metal from waste industrial plants, a bioremediation test was adapted in aqueous solution, incubating 5 g of biomass with 5 g of non-sterile soil, contaminated with 200 mg/g of soil, pH 6.8 and 100 mL of contaminated water with 200 mg/L, pH 8.2, resuspending the soil in trideionized water to a final volume of 100 mL, at 28 °C, and 100 rpm, observing that after 7 days of incubation the concentration of chromium (VI) of the soil and water samples, decreased between 66.1% and 74%, respectively, without significant changes in the content of total chromium (data not shown). In the control of the experiment (without biomass), the concentration of the metal of samples decreased between 7% and 14% (data not shown), which may be caused by the microflora autochthonous and/or reducing components present in the samples<sup>35</sup>. The capacity of chromium (VI) removal from wastewater by these biomasses, is equal or better than others analyzed, for example: the biomass of two commercial strains of *A. bisporus*<sup>30</sup>, in which after 7 days of incubation the concentration of this metal of the soil and water samples, decreased between 66.1% and 76.2%, too, 5 g of biomass of *F. duorarum*, removal 80% of chromium (VI) from soil contaminated with 297 mg of the metal/g of soil, after 96 h of incubation at 28 °C<sup>31</sup>, the modified biomass of rice grain (*O. sativa* L.) efficiently removes ground metal and contaminated wastewater (71 and 73%, respectively), at 10 days of incubation with 10 g of modified biomass<sup>19</sup>, the fungal biomass of *Aspergillus niger* removal 69% of the metal in situ, (7 days of incubation and 5 g of biomass (100 mL water)<sup>36</sup>. Fungal strains isolated from effluent contaminated (*Aspergillus* spp., *Rhizopus* spp., *Trichoderma* spp., and *Penicillium* spp.), showed a good adsorption capacity of chromium removal efficiency at 150 ppm of hexavalent chromium (5.49 mg/g and/or 98.75%)<sup>37</sup>, the hexavalent chromium removal (181.56 mg/g) and total chromium biosorption (110.35 mg/g) at influent solution pH of 1.0 and 2.0<sup>38</sup>, too for the bioremediation of solid waste from lemon pulp contaminated with chromium (VI) by filamentous fungi: *A. niger* (90%), *Penicillium islandicum* (5%), and *Penicillium expansum* (6%), initial pH 2.5, 3 días, 28 °C<sup>39</sup>, and the fungal strain of *Trichoderma pseudokoningii*, isolated from tannery effluent showed 97–99% chromate removal in 10 days<sup>40</sup>, and for the *Eichhornia crassipes* biomass, which removal 100 mg/L in contaminated water, at 7 days of incubation, with 10 g of natural biomass in 100 mL<sup>41</sup>.

The results obtained in this work, suggest the potential applicability of this biomass for the removal of this metal from wastewater. However, the capacity of biosorption may be affected by the high concentrations of these contaminants, decreasing the capacity adsorption by the biomasses. So that, actually the use of biomasses does not live, it is a great alternative to try to eliminate these contaminants from the different contaminated sites, by her low cost, great adsorption capacity, easy accessibility and its production in large quantities.

Finally, the *N. tabacum* biomass accumulating efficiently different heavy metals<sup>20–23</sup>, but there are few reports regarding the removal of heavy metals by the biomass of natural and/or commercial tobacco, therefore, this work is important, because commercial tobacco is used, which facilitates its obtaining in sufficient quantities for its application in bioremediation.

## 5. Conclusions

The biomass analysed, eliminate 72% of the heavy metal, at 24 h of incubation, pH 2.0, 28 °C, and 100 rpm. To higher temperature the removal is most efficient, if the heavy metal concentration is increased, the removal capacity is reduced. The biosorbent concentration not influences in the removal, and this natural biomass removal efficiently the heavy metal of contaminate soil and water.

## Author Contributions

Ismael Acosta-Rodríguez: Investigation, Writing, Review and Editing. Cintya C. Huerta-Velázquez: Work Laboratory, Writing, Review. Katia B. Cruz-García: Work Laboratory, Writing, Review.



## Conflict of Interest

The authors have no conflict of interest to declare.

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