

**Research Article** 





# Biosorption of iron by amended Aspergillus versicolor from polluted water sources

#### Abstract

Bacterial and fungal biomasses are economic natural adsorbents of metals because of negative surface charge and membrane composition. Hence, the present work aimed to investigate the use of *Aspergillus versicolor* (Ascomycota) as a biosorbent for the removal of iron from polluted water. Effects of pH, initial metal concentration, temperature and quantity of fungus on the biosorption capacity are also evaluated. The fungus is grown on Potato Dextrose Agar (PDA) plates amended with 100 ppm of Fe (II) ions and the standard spread plate method is applied. The plates are incubated at 27°C for 4 to 7 days, then the grown fungi are killed by adding 0.5 N NaOH, washed with distilled water, dried in an oven and finally grinded into fine powder. The optimum pH value for iron biosorption is 6. The rate of adsorption of iron by *A. versicolor* is very fast and reaching a maximum within 15 min. Maximum removal efficiency is observed at the biomass dosage concentration of 0.3 g *A. versicolor*:

Keywords: aspergillus versicolor, biosorption, economic, fungi, iron

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## Introduction

Environmental pollution by heavy metals has become a question of public concern considering their hard consequences.<sup>1,2</sup> Air, food, soil and water are narrated to be the media where heavy metals such as copper, cadmium, nickel, lead, zinc and iron are introduced into the environment.<sup>3</sup> Presence of these metals in waste stream ground and drinking water is a very serious concern since these metal ions are toxic to various life forms,<sup>4</sup> that heavy metals and other constituents leach into the soil and damage the flora and fauna on the earth.<sup>5</sup> Heavy metals are released from industrial operations such as electroplating, steel manufacturing, wood preservation, tanning, glass manufacturing and chemical processing which are eventually accumulating and circulating throughout the food chain causing severe problems to humans, animals and environment.

Iron is one of most spreading heavy metals making up to 5% of the earth's crust, it is one of the major impurities that are commonly present in many water sources which cause several troubles for the human health. There are different methods for removal of iron from ground water, e.g., oxidation with chlorine and potassium permanganate treatment with limestone, liquid-liquid extraction, ion exchange, chemical precipitation, bioremediation, activated carbon and some filtering materials.6-8 These methods are generally of high cost with some disadvantages such as incomplete metal removal, much reagent and time consuming, energy requirements and generation of toxic sludge or other waste products that require further disposal or treatment.9 Biosorption using biomaterials has received considerable attention for detoxification of toxic heavy metals from wastewaters as they possess various merits such as low cost, high efficiency, minimization of chemical and or toxic sludge, regeneration of biosorbent, metal recovery etc.10-12

In metal polluted environments, microbial populations adapt themselves to high concentrations of heavy metals and become resistant to toxic concentrations.<sup>13</sup> Algae, bacteria, fungi and yeasts have the ability to adsorb heavy metal ions.14,15 Bacterial and fungal biomasses are economic natural adsorbents of metals because of negative surface charge and membrane composition; the cell wall of the microorganisms essentially consists of various organic compounds such as carboxyl, acidic polysaccharides, lipids, amino acids and other components, extracellular polymeric substances present on the outer surface of some microorganisms that, also, contribute to biosorption of metal ions since those polymers contain negatively charged functional groups, such as carboxyl, phosphate and sulphate. Among microorganisms, fungi provide an effective, economic, and environmentally-friendly method of removing harmful wastes that accumulate as byproducts of industrial activities. The marine red alga Laurencia catarinensis was used in the biosynthesis of silver nanoparticles.16 Several studies have been forwarded towards the use of fungal biomass such as Rhizopus arrhizus, Saccharomyces cerevisiae and Aspergillus niger in the biosorption of heavy metals.<sup>17</sup> It has been documented that Aspergillus spp are capable of the biosorption of metal ions, out of 23 fungi isolated from tannery effluent, 4 Aspergillus species namely A. terreus, A. tamarii, A. flavus and A. niger were selected for evaluating chromium tolerance and biosorption potential. In their review, (Siddiquee et al., 2015) reported that Aspergillus spp among other filamentous fungi were the most resistant to all the metals tested as cadmium, copper and nickel. Many species of genus Aspergillus have been used for heavy metal ion adsorption such as A. niger which has been used for the removal of Cr (VI),<sup>18,19</sup> A. fumigatus to get rid of cadmium,20 A. versicolor for removal of lead ions from aqueous solutions<sup>21</sup> and cadmium,<sup>22</sup> A. terreus for biosorption of lead, mercury and cadmium ions,23 A. cristatus for cadmium biosorption24 and A. flavus has showed a higher affinity towards Fe(II).<sup>2</sup>

The main interest of the present study is to investigate the use of the highly ubiquitous *Aspergillus versicolor* resistant fungi powder as a bio sorbent for the removal of iron from aqueous solutions as a function of initial pH, initial metal concentration, temperature and quantity of the resistant fungus.

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# **Materials and methods**

#### Chemicals

Ferrous ammonium sulphate (Aldrich), Hydrochloric acid (BDH), 1, 10 phenanthroline (Aldrich) (0.2%) solution of phenanthroline hydrochloride or hydrate (phen.) in 0.1 M HCl, tri sodium citrate (Aldrich) and hydroxylamine hydrochloride (Aldrich).

## Reagents

## Standard Iron (II) solution

(1000 ppm) Fe (II) stock solution is prepared by dissolving 0.7016 g of ammonium ferrous sulphate ( $NH_4$ )<sub>2</sub>SO<sub>4</sub>·FeSO<sub>4</sub>·6H<sub>2</sub>O, (Aldrich, USA) in doubly distilled water (DDW) containing 5 mL conc. H<sub>2</sub>SO<sub>4</sub> and accurately diluted to volume in 100 mL volumetric flask.

#### I, IO Phenanthroline (0.2%)

In 100 mL vol. flask 0.2 gm of phenanthroline are dissolved in doubly distilled water (DDW) and diluted to the mark with 0.1 M HCl.

**Tri sodium Citrate (10 %) solution:** In 100 mL vol. flask, 10 g of tri sodium citrate are dissolved in DDW and diluted to the mark.

#### Hydroxylamine hydrochloride (10%)

10 g of hydroxylamine–HCl are transferred to 100 mL vol. flask and dissolved in DDW, then completed to the mark.

#### **Media preparation**

Potato dextrose broth and agar media are prepared using 20 g of potato slices boiled with 100 mL of DDW for 30 min, the filtrate is obtained by strain through several layers of cheese cloth and then press the cloth to extract the remaining liquid. 2 g of dextrose are added, then make the filtrate up to 100 mL if necessary. For agar plates, 1.5 g of agar are added to this mixture and the flask was closed with cotton plug and aluminum foil and autoclaved for 20 minutes at 121°C.

#### **Preparation of Fungal Biosorbents**

*A. versicolor* was obtained from Assuit University mycological centre under the AUMC No. 90, it was grown on Czapek yeast extract agar (CYA).

For fungal biosorption study, Potato Dextrose Agar (PDA) plates were amended with 100 ppm of Fe (II) ions and the standard spread plate method was performed. The fungus was incubated for 4 to 7 days at 27°C. ), after incubation period; the fully grown fungi (resistant metal fungi) are killed by adding 0.5 N NaOH in a conical flask containing the fungal mat and kept in a water bath for 15 minutes, the mat was washed with doubly distilled water for about 6–7 times till the pH reaches 7,the mat is then transferred to a sterile Petri dish and placed in hot air oven for 24 hours at 75°C.

The dried dead fungal mat was powdered to the smallest particle size using mortar and pestle, whereas it is known that the smaller the particle size, the larger the surface area. Biomass has been crushed to prevent particle aggregation for enhancing the biosorption capacity, then stored in sterile container for further study.

#### Instruments

UV/VIZ. Spectrophotometer (Shimadzu UV/VIZ. Perkin Elemer Lambada 3B Spectrophotometer using 1cm Quartz cell" was used for the determination of residual iron ions in the effluent of the samples after each adsorption process. Flame Atomic Absorption Spectrophotometer AA 240FS, Agilent Technologies, was used for rapid and conformational determination of iron ions; pH meter: the microprocessor pH meter BT 500 BOECO, Germany, which was calibrated against two standard buffer solutions at pH 4 and 9 was used for carrying out pH measurements and Mechanical Shaker with up to 200 rpm with speed control was used. The morphologies of the prepared samples were investigated using Scanning Electron Microscopy: TEM (JEOL–JEM– 100CX II).

## Procedure

#### Spectrophotometric determination of iron

Sorption studies were performed in batch mode. After the biosorption process, the remaining iron ions in the solution are determined spectrophotometrically as follows . Transfer to 25 mL volumetric flask 0.5 mL of the 10 % hydroxylamine solution, 2 mL 10 % tri sodium citrate solution and 5 mL of the standard Fe (II) solution. The pH is adjusted to the range 3–4. Add 2.5 mL of 0.2 % 1, 10 phenanthroline solution, dilute to the mark with DDW and mix thoroughly. After 5 min the absorbance of the solution is measured at 512 nm against a blank.

# Optimization of factors affecting the adsorption of iron

## **pH** Optimization

To investigate the effect of pH on the adsorption % of iron from aqueous media by *A. versicolor* dried powder, aliquots of 25 mL containing 100 ppm of the metal ion are transferred to a set of 100 mL conical flasks each containing 0.3 g of the fungal powder, adjust the pH of each flask to a value in the range 1–10, respectively, using 0.1M NaOH and 0.1M HCl solutions and stir for 15 minutes. Centrifuge the contents of each flask and the supernatant solutions containing the residual iron contents are filtered and the concentrations of metal ions are measured by UV/VIZ spectrophotometry at 512 nm. Data are expressed as removal percentage of the initial concentration or specific biosorption q (mg/g), which is calculated by equation (1):

$$q = \frac{c_{i-C_f}}{Wt} * V \tag{1}$$

Where  $C_i$  is the initial iron concentration (ppm),  $C_f$  is the final concentration (ppm),  $W_t$  is the dose of sorbent (g) and V is the volume of solution (mL). The equation was applied in the pH range 1–10. The optimum pH is found to be 6 (Figure 1).

#### **Biomass dose optimization**

Aliquots equal to 100 ppm iron are transferred to a set of 100 mL conical flasks. Adjust the pH of each flask to the optimum value 6. Varying amounts of *A.versicolor* powder in the range 0.05–0.5g are added to each flask, respectively. The mixtures are stirred for 15 minutes, the residual iron content in the supernatant soln, separated by centrifugation is similarly determined spectrophotometrically.

## Stirring time

A group of 100 mL conical flasks, each of which is loaded with the optimum biomass dose 0.3 g of *A. versicolor* powder and aliquots of 25 mL solution containing 100 ppm of iron at the optimum pH 6 and stirring is changed for different intervals of time (5–30 min.) for each flask in its role, respectively.

#### Metal ion concentration range

Applying the optimum conditions of the weight of *A. versicolor* powder, pH and stirring time in a group of flasks, aliquots of 25 mL solution containing varying concentrations of iron in the range 90–150 ppm are added to the flasks, respectively. The same procedure is applied and the residual iron content is determined from which the removal percent is calculated.

#### **Desorption studies**

Reusability of the adsorbent is tested by regenerating the spent adsorbent following the modified procedure of Chen et al.,.<sup>25</sup> The desorption process should yield the adsorbed metal in a concentrated form, restore the biosorbent close to the original state for effective reuse with undiminished metal uptake and physical changes or damages to it.<sup>26</sup> The adsorbed iron ions on the adsorbent surface are treated with 25 mL 0.1M HCl and stirred for 1h. The amount of iron ions remained in the solution after filtration or centrifugation is measured using the recommended spectrophotometric method and the percentage desorption was calculated according to equation (2).

Desorption % = 
$$C_r/C_i x 100$$
 (2)

Where  $C_r$  is the released metal concentration and  $C_i$  is initially sorbed metal concentration.

## **Results and discussion**

Dead fungal biomass has been used in a number of studies because it is not affected by adverse operating conditions and can solve the environmental problems of high toxicity. Dead biomass achieves, relatively, best removal of heavy metals compared to the active biomass of the strain under the same conditions due to the pretreatment of the fungal biomass with heat enhancing its the stability, settling property and heavy metal uptake capacities of the biomass as previously reported.<sup>27</sup> Under optimized conditions slight decrease in heavy metal ions noted in sorption potential from industrial effluents and wastewaters could be due to various impurities present in them in the form of anions i.e.,  $SO_4^{2-}$ ,  $NO_3^{-}$ , and  $Cl^-$  that may compete for binding sites on the fungal cell walls and then reduce the uptake of metallic ions from industrial wastewater using fungi.18 The dried powdered biomasses have been introduced as a new field of biotreatment technology, as effective and relatively simple methods for heavy metals recovery.28 Several studies have explained that inactive dried microbial biomass can passively bind metal ions via various physicochemical mechanisms.<sup>29</sup> It has been suggested that the pretreatment by iron ions stimulates the surface characteristics groups by removing or masking them or by increasing the metalbinding sites.<sup>30</sup> The use of fungal biomass is reported in literature for the removal of different heavy metals from wastewater e.g., out of 23 fungi isolated from tannery effluent, 4 Aspergillus species

namely A. terreus, A. tamarii, A. flavus and A. niger were selected for evaluating chromium tolerance and biosorption potential<sup>31</sup> A. flavus was used by Verma et al.,<sup>2</sup> for the removal of Fe(II), the uptake of chromium by Aspergillus foetidus,13 heavy metal biosorption using Aspergillus niger, 14,15,18 heavy metal removal by fungus Mucor rouxii Khambhaty et al.,19 and biosorption of hexavalent chromium by dead fungal biomass of marine A. niger.<sup>32</sup> The use of non-living biomass of A. versicolor in biosorption is more useful than bioaccumulation for water treatment, as the removal takes less time and continuous supply of nutrients is not required. A. versicolor was used previously for the removal of lead ions from aqueous solutions<sup>21</sup> and for Cr (VI), Cu (II), Ni (II) and RB reactive textile dye.<sup>28</sup> A. versicolor is a one of the most common fungi, it is frequently found in soil, plant debris, marine environments, and indoor air environments.33 It is also among the most common of indoor molds.<sup>34,35</sup> It is considered to be osmophilic as it is able to survive in solutions up to 30% NaCl or 40% sucrose.36

Different factors that affect the adsorption process have been extensively studied to improve the biosorption capacity Q (mg/g) of *A. versicolor* towards iron ions from aqueous solutions.

#### Optimum pH

Earlier studies showed that the pH value has been observed as one of the important parameters which control the heavy metal biosorption.37-39 The variation of adsorption rates at different pH values is dependent on both the metal chemistry in solution and the surface chemistry of the sorbent.<sup>40</sup> Adsorption of iron by A.versicolor powder has been found to increase with the increase in pH value until attains its maximum at pH 6. The biosorption of the dried biomass is increased with the solution pH, due to the excess amounts of OHions within the solution, the binding sites on the fungal cell wall are negatively charged. The cell wall, as explained previously, is made up of several components such as carboxyl, carbonyl, alcoholic and amino groups which determines the biosorption ability based on its protonation or unprotonation nature. This exerted an influential attraction between active sites and positively charged ions. Thus, it is most possible that at low pH values, positively charged surface will not favor the binding with iron ions due to columbic repulsion. With increase in pH values, surface becomes more and more negatively charged and thereby favoring iron ion binding (Figure 1), (Table1).

#### Stirring time

The kinetics of adsorption describing the shaking time in the removal of iron is one of the characteristics defining the efficiency of the biosorption rate. The results indicate that maximum biosorption capacity takes place after 15 minutes where the uptake was 22.2 mg/g for iron ions. After this period, the equilibrium is reached (Figure 2 & Table 2).

### **Biomass dose**

The increase in the biosorbent concentration from 0.05-0.5g results in extensive increase in the metal adsorption. The increase of the adsorption surface area and the availability of free adsorption sites help in the removal of iron. Maximum removal efficiency is observed at the biomass dosage concentration of 0.3 g *A. versicolor*, which achieves iron removal of 22.25 mg/g, after this concentration equilibrium is reached (Figure 3 &Table 3).



Figure I Effect of Initial pH on biosorption capacity of iron by A. versicolor resistant powder.



Figure 2 Effect of stirring time on biosorption capacity of iron by A. versicolor resistant powder.



Figure 3 Effect of biomass dose on biosorption capacity of iron by A. versicolor resistant powder.

| рН    | Std.<br>deviation | q (mg/g)        |  |  |  |  |
|-------|-------------------|-----------------|--|--|--|--|
| 1.00  | 0.1               | 4.1000±.05774   |  |  |  |  |
| 2.00  | 0.15275           | 8.1667±.08819   |  |  |  |  |
| 3.00  | 0.15275           | 12.1667±.08819  |  |  |  |  |
| 4.00  | 0.26458           | 16.2000±.15275  |  |  |  |  |
| 5.00  | 0.20817           | 20.1667±.12019  |  |  |  |  |
| 6.00  | 0.15275           | 22.1667±.08819  |  |  |  |  |
| 7.00  | 0.20817           | 18.1667±.12019  |  |  |  |  |
| 8.00  | 0.1               | 13.0000±.05774  |  |  |  |  |
| 9.00  | 0.11547           | 8.1333±.06667   |  |  |  |  |
| 10.00 | 0.1               | 3.1000±.05774   |  |  |  |  |
| Total | 6.39491           | 12.5367±1.16754 |  |  |  |  |

 Table 2 Effect of stirring time on biosorption capacity of iron by A. Versicolor

 resistant powder, number of determinations N=3

| Time of stirring | Std.<br>deviation | q (mg/g)        |
|------------------|-------------------|-----------------|
| 3.00             | 0.1               | 2.1000±.05774   |
| 6.00             | 0.15275           | 6.0333±.08819   |
| 9.00             | 0.15275           | 11.1667±.08819  |
| 12.00            | 0.15275           | 15.1667±.08819  |
| 15.00            | 0.15275           | 22.1333±.08819  |
| 18.00            | 0.2               | 20.2000±.11547  |
| 21.00            | 0.28868           | 12.1667±.16667  |
| 24.00            | 5.91805           | 13.8333±3.41679 |
| 27.00            | 0.1               | 4.0000±.05774   |
| 30.00            | 0.1               | 1.1000±.05774   |
| Total            | 7.25873           | 10.7900±1.32526 |

## Effect of initial metal ion concentration

The interest in processes including heavy metal removing by microorganisms has increased considerably in recent years due to the biotechnological potential of micro–organisms in metals recovery,<sup>22</sup> the maximum metal uptake was 22.2 mg/L at 90 ppm iron concentration, then iron removal decreases with the increase of iron concentration. At higher concentrations, the available sites for sorption become fewer in comparison with the molecules of solute present. Hence, the

removal of metal ions is strongly dependent upon the initial solute concentration, so heavy metal tolerant microorganisms such as *A. versicolor* instinctively use defense mechanisms on exposure to metal stress (Figure 4 & Table 4).<sup>41</sup>

Table 3 Effect of biomass dose on biosorption capacity of iron by A.Versicolor resistant powder, number of determinations N=3

| Dose  | Std.<br>deviation | q (mg/g)       |
|-------|-------------------|----------------|
| 0.05  | 0.1               | 8.1000±.05774  |
| 0.10  | 0.1               | 12.1000±.05774 |
| 0.15  | 0.15275           | 14.1667±.08819 |
| 0.20  | 0.1               | 17.1000±.05774 |
| 0.25  | 0.1               | 20.0000±.05774 |
| 0.30  | 0.07638           | 22.3167±.04410 |
| 0.35  | 0.03215           | 22.2267±.01856 |
| 0.40  | 0.03215           | 22.2367±.01856 |
| 0.45  | 0.02517           | 22.2267±.01453 |
| 0.50  | 0.07638           | 22.2333±.04410 |
| Total | 5.00218           | 18.2707±.91327 |

**Table 4** Effect of metal concentration on biosorption capacity of iron by A. *Versicolor* resistant powder, number of determinations N=3

| Metal<br>concn. | Std.<br>deviation | q (mg/g)       |
|-----------------|-------------------|----------------|
| 15.00           | 0.06715           | 10.2000±.03877 |
| 90.00           | 0.1               | 22.1000±.05774 |
| 100.00          | 0.1               | 20.4000±.05774 |
| 110.00          | 0.15275           | 18.0333±.08819 |
| 120.00          | 0.15275           | 15.0333±.08819 |
| 130.00          | 0.1               | 13.1000±.05774 |
| 140.00          | 0.2               | 11.2000±.11547 |
| 150.00          | 0.14142           | 9.9000±.10000  |
| Total           | 4.39107           | 15.6952±.95821 |

#### Effect of temperature

Temperature is another most important factor in the biosorption process of metals. The increase in temperature has improved the iron biosorption rate and decreased the contact time required for heavy metal removal. The temperature of the adsorption medium is considered to be an important parameter for energy dependent mechanisms in metal removal using biosorbent. Maximum removal of iron is found to be 22.2 mg/g at a temperature equals to 31°C. Temperature affects the cell wall stability components, its configuration and ionization of chemical moieties and energy–independent mechanisms are likely to be affected due to temperature changes since the processes responsible for removal are largely physiochemical in nature.<sup>42</sup> Similar results have been recorded in the bioaccumulation of Cu (II) and Cr (VI) by *Streptococcus Equisimilis* and *Aspergillus* sp (Figure 5 & Table 5).<sup>43</sup>



Figure 4 Effect of metal concentration on biosorption capacity of iron by A. versicolor resistant powder.



Figure 5 Effect of temperature on biosorption capacity of iron by A. versicolor resistant powder.

## The morphologies of the prepared fungi

Scanning electron microscope (SEM) provides information about the sample's surface topography and composition. SEM micrographs of dried biosorbent particles showed fairly regular spherical structures with an external surface which, although rather smooth, displayed a number of cracks likely to favor solute adsorption through enhanced diffusion to active sites (Figure 6).

#### Adsorption isotherm studies

It is more significant to study the adsorption behavior in order to provide predictions of the performance of the biosorption process under different operating conditions using the appropriate adsorption isotherm model. For solid–liquid adsorption system, the adsorption behavior can be described as adsorption isotherm model. The adsorption isotherm means the distribution of adsorbate molecules between the solid phase and the liquid one at equilibrium. Equilibrium

is said to be reached when the concentration of adsorbate in bulk solution is in dynamic balance with that on the liquid adsorbate interface.

#### Langmuir adsorption isotherm

The equilibrium adsorption data for the concentrations of iron ions is fitted into the linear form of Langmuir's isotherm equation, to determine the distribution of iron ions between the adsorbent and solution according to equation (3):

$$\frac{\mathbf{c}_e}{\mathbf{Q}_e} = \frac{1}{\mathbf{Q}_m K_L} + \frac{\mathbf{c}_e}{\mathbf{Q}_e} \tag{3}$$

Where  $C_e$  is the equilibrium concentration of the iron ions in solution (mg L<sup>-1</sup>),  $Q_e$  is the equilibrium concentration of iron ions on *A. versicolor* adsorbent (mg g<sup>-1</sup>),  $Q_m$  and  $K_L$  are Langmuir constants related to sorption capacity and the rate of adsorption respectively. Maximum adsorption capacity ( $Q_m$ ) is the monolayer capacity of the adsorbent (mg g<sup>-1</sup>) and  $K_L$  is the Langmuir adsorption constant. A plot of  $C_e/Q_e$  against  $C_e$  over the entire concentration range is a straight line with a slope of  $1/Q_m$  and intercept of  $1/Q_m K_L$ . The correlation coefficient ( $R^2$ ) values reported are very close to 1 indicating that the adsorption follows the Langmuir adsorption isotherm. The quality of Langmuir isotherm can be determined by the magnitude of a dimensionless constant  $R_L$  known as the separation factor expressed in equation (4):

$$\boldsymbol{R}_{\boldsymbol{L}} = \frac{1}{1 + \boldsymbol{C}_0 \boldsymbol{K}_{\boldsymbol{L}}} \tag{4}$$

where  $C_o$  is the initial concentration of the iron ions in mg L<sup>-1</sup> and  $K_L$  is the Langmuir constant described earlier. The adsorption process is favorable within the range  $0 < R_L < 1$ , unfavorable when  $R_L > 1$ , becomes linear when  $R_L = 1$ , and the process is irreversible when  $R_L = 0$ . The value of  $R_L$  is 0.047 for *A. versicolor*; hence the adsorption process is favorable (Figure 7), (Table 6).

#### Freundlich adsorption isotherm

The linear form of the Freundlich adsorption model equation (5):

$$logeQelogeKF \frac{1}{n} logec_e \qquad (5)$$

Where  $Q_e$  is the amount of iron ions adsorbed at equilibrium per gram of the adsorbent (mg g<sup>-1</sup>),  $C_e$  is the equilibrium concentration of iron ions in the solution (mg L<sup>-1</sup>), and  $K_f$  and n are the Freundlich adsorption model constants related to the adsorption capacity and adsorption intensity respectively. Log  $Q_e$  was plotted against log  $C_e$  and a straight line is obtained giving the intercept of log  $K_f$  and the slope of 1/n. The reported numerical value of 1/n is less than1, (Figure 8 & Table 6).

#### Interferences

On the spiking of the iron authentic samples under the optimum conditions, with different concentrations of other metal ions e.g., Mn(II), Ni(II) and Cu (II), higher values for the removed iron were obtained, which means that there is no complete selectivity for the resistant *A. versicolor* powder towards iron ions. Perhaps, these results may be attributed to the use of the single recommended pH value for the adsorbate (pH 6) where some other metal ions, which may be copresent with iron in the matrix solution, are, also, coadsorbed at this pH value to the powdered biomass. Thus, the biosorption of Cu (II) by *Aspergillus flavus* was conducted at pH 8–9 and Pb (II) by *Aspergillus niger* at pH 4–5.4. Also, it was reported by Ratnasri & Hemalatha<sup>27</sup> that the pH value must be less than that for the precipitation of respective metal ions, thus the sorption of Fe<sup>3+</sup> on some *Aspergillus* species was performed at pH 2.6 and for other species with Mn<sup>+2</sup> at pH 8.

However, the amending of the fungus with iron ions resulted in obtaining the resistant *A. versicolor* powder which proved as efficient, low cost, available, economic biosorbent for removal of heavy metals in general, and especially, for the removal of relatively large amounts of iron.

#### **Desorption studies**

## Effect of pH on desorption of iron

In strong acidic media at pH range (1.8–2.4) *A.versicolor* resistant powder showed high desorption percentages. With increasing the pH values desorption percentage decreases (Figure 9 & Table 7).

Table 5 Effect of temperature on biosorption capacity of iron by A.Versicolor resistant powder, number of determinations N=3

| Tempr. | Std.<br>deviation | q (mg/g)       |
|--------|-------------------|----------------|
| 25.00  | 0.1               | 19.1000±.05774 |
| 27.00  | 0.15275           | 20.1667±.08819 |
| 29.00  | 0.20817           | 21.2333±.12019 |
| 31.00  | 0.1               | 22.4000±.05774 |
| 33.00  | 0.15275           | 21.1667±.08819 |
| 35.00  | 0.1               | 20.1000±.05774 |
| 37.00  | 0.15275           | 19.1667±.08819 |
| 39.00  | 0.1               | 18.4000±.05774 |
| 41.00  | 0.1               | 18.0000±.05774 |
| Total  | 1.39417           | 19.9704±.26831 |



Figure 6 Scanning electron micrograph of Aspergillusversicolor biomass.



Figure 7 Langmuir isotherm plots for the adsorption of iron ions by A. versicolor.



Figure 8 Freundlich isotherm plots for the adsorption of iron ions by A. versicolor.



Figure 9 Effect of pH on desorption%.

## Stirring time

The maximum desorption percentage is found to be 80 during the first 20 minutes, then gradually decreased. (Figure 10 & Table 8).

## **Real samples**

Water samples collected from Bahr Youssef canal, ground water, Ibrahemia canal and tap water from the districts in which drinking water plants mix the River Nile water with some ground water, are subjected to the adsorption procedure as illustrated previously and the residual iron is analyzed by two methods of finish viz., colorimetry and AAS (Table 9).



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## Figure 10 Effect of on stirring time desorption%.

Table 6 Freundlich adsorption isotherm Parameters

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| Langmuir and Freundlich adsorption isotherm Parameters of Iron ions by A. Versicolor |                         |        |                                |        |  |  |
|--|-------------------------|--------|--------------------------------|--------|--|--|
| No.  | Langmuir isotherm param | neters | Freundlich isotherm parameters |        |  |  |
| 1  | Qmax(mg/g)              | 22.25  | 1/n                            | 0.8545 |  |  |
| 2  | KL                      | 0.2    | KF                             | 1.8    |  |  |
| 3  | R <sup>2</sup>          | 0.961  | $\mathbb{R}^2$                 | 0.997  |  |  |
| 4  | RL                      | 0.047  |                                |        |  |  |

| рН    | Std.      | Desorption%     | Stirring | Std       |                 |
|-------|-----------|-----------------|----------|-----------|-----------------|
|       | deviation |                 | time     | deviation | Desorption%     |
| 1.80  | 0.15275   | 80.1667±.08819  |          |           |                 |
| 2.40  | 0.1       | 79.9000±.05774  | 20.00    | 0.15275   | 80.1667±.08819  |
| 3.00  | 0.15275   | 73.1333±.08819  | 25.00    | 0.1       | 60.0000±.05774  |
| 3.60  | 0.2       | 69.2000±.11547  | 30.00    | 0.20817   | 50.2333±.12019  |
| 4.20  | 0.20817   | 64.0667±.12019  | 35.00    | 0.15275   | 41.1667±.08819  |
| 4.80  | 0.15275   | 60.1333±.08819  |          |           |                 |
| 5.40  | 0.20817   | 55.1667±.12019  | 40.00    | 0.28868   | 35.1667±.16667  |
| 6.00  | 0.32146   | 54.1333±.18559  | 45.00    | 0.15275   | 34.1333±.08819  |
| 6.60  | 0.15275   | 50.1333±.08819  | 50.00    | 0.1       | 33.1000±.05774  |
| 7.20  | 0.15275   | 47.1667±.08819  | 55.00    | 0 26458   | 32 2000+ 15275  |
| 7.80  | 0.20817   | 43.2333±.12019  | 55.00    | 0.20100   | 52.200015215    |
| 8.40  | 0.1       | 39.9000±.05774  | 60.00    | 0.1       | 32.1000±.05774  |
| Total | 13.35395  | 59.6944±2.22566 | Total    | 15.88023  | 44.2519±3.05615 |

 Table 7 Effect of pH on desorption%, number of determinations N=3

Table 8 Effect of stirring time on desorption%, number of determinations  $N{=}3$ 

 Table 9 Concentration of Fe (II) on real water samples, number of determinations N=3

| Collected samples     | Final concentration | Std.<br>deviation | Recovery%       |  |
|-----------------------|---------------------|-------------------|-----------------|--|
| Drinking water        | 0.32                | 0.10263           | 86.1133±0.05925 |  |
| Bahr Youssef<br>Canal | 1.06                | 0.07638           | 80.8833±0.04410 |  |
| Ground water          | 1.36                | 0.1               | 82.2000±0.05774 |  |
| Ibrahimia Canal       | 1.72                | 0.1               | 81.6000±0.05774 |  |

Table 10 Summery of factors affecting the extraction of iron by A.Versicolor, number of determinations N=3

|                         | Colorimetry |        |                   |               | AAS           |         |                   |               |               |
|-------------------------|-------------|--------|-------------------|---------------|---------------|---------|-------------------|---------------|---------------|
| Parameter               | Optimum     | Mean   | Std.<br>deviation | Std.<br>error | q (mg/g)      | Mean    | Std.<br>deviation | Std.<br>error | q (mg/g)      |
| pH                      | 6.00        | 22.166 | 0.15275           | 0.0881        | 22.166±0.0881 | 22.100  | 0.10000           | 0.5773        | 22.100±0.5773 |
| Time of stirring, min   | 15.00       | 22.133 | 0.15275           | 0.0881        | 22.133±0.0881 | 22.2833 | 0.15275           | 0.0881        | 22.283±0.0881 |
| Dose, g                 | 0.30        | 22.316 | 0.07638           | 0.0441        | 22.316±0.0441 | 22.050  | 0.18028           | 0.104         | 22.050±0.1040 |
| Metal Concentration, mg | 90.00       | 22.1   | 0.1               | 0.0577        | 22.100±0.0577 | 21.933  | 0.05774           | 0.3333        | 21.933±0.3333 |
| Temperature °C          | 31.00       | 22.4   | 0.1               | 0.0577        | 22.400±0.0577 | 22.400  | 0.10000           | 0.0577        | 22.400±0.0577 |

# Conclusion

Biosorption of iron ion by *A. versicolor* resistant fungus is shown to be an effective bioremoval process. It could retain relatively high quantities of metal ions with increased capacity towards the adsorption of the amending metal ion, although its selective adsorption hasn't completely achieved. The kinetics of biosorption is rapidly enhanced with temperature increase and acidic pH. Both Langmuir and Freundlich adsorption isotherm models are suitable for predicting the biosorption capacity mg/L of *A.versicolor* within the range of the studied variables.

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## **Conflict of interest**

The authors declare no conflict of interest.

## **Ethical approval**

This article does not contain any studies with human participants or animals performed by any of the authors.

## References

- Hassouna MEM, ElBably MA, Mohammed AN, et al. Assessment of carbon nanotubes and silver nanoparticles loaded clays as adsorbents for removal of bacterial contaminants from water sources. *J Water Health*. 2017;15(1):133–143.
- Verma TK, Tiwari KL, Jadhav SK. Removal of Fe(II) Using Aspergillus flavus strains From Aqueous Solution, Indian. J Sci Res. 2017;13(2):63– 67.
- Garba ZN, Ugbaga NI, Abdullahi AK. Evaluation of optimum adsorption conditions for Ni (II) and Cd (II) removal from aqueous solution by modified plantain peels (MPP). *Beni–Suef University. J Basic and Applied Sciences.* 2016;5(2):170–179.
- Wasiu MO, Ayodele OE, Ayodele TI, et al. Heavy metal contamination in stream water and sediments of gold mining areas of South Western Nigeria'. *African Journal of Environmental Science and Technology*. 2016;10(5):150–161.
- Gayatri Y, Shailaja RM, Vijayalakshmi B. Biosorption of lead by *Bacillus licheniformis* isolated from e-waste landfill, Hyderabad, Telangana, India. J of Bioassays. 2017;6(2):5240–5244.
- Ryoma BE, Kawasaki N, Ogata F, et al. Removal of lead and iron ions by vegetable bio mass in drinking water. J Oleo Science. 2006;55(8):423– 427.
- Berbenni P, Pollice A, Canziani R, et al. Removal of iron and manganese from hydrocarbon contaminated ground waters. *Bioresource Technology*. 2000;74(2):109–114.
- Regina de Moreira FPM, Madeira VS, Jose HJ, et al. Removal of iron from water using adsorbent carbon. *J Separation Science and Technology*. 2005;39:271–285.
- Owalude SO, Tella AC. Removal of hexavalent chromium from aqueous solutions by adsorption on modified groundnut hull. *Beni–Suef University J. Basic and Applied Sciences*. 2016;5:377–388.
- Sen M. Enhanced biological removal of Cr(VI) in Continuous Stirred Tank Reactor (CSTR) using Aspergillus sp. Brazilian Journal of Biological Sciences. 2018;5(9):33–45.
- El Gendy MMAA, Hassanein NM, Ibrahim HA, et al. Heavy Metals Biosorption from Aqueous Solution by *Endophytic Drechslera hawaiiensis* of Morus alba L. Derived from Heavy Metals Habitats. *Mycobiology*. 2017;45(2):73–83.
- Iskandar NL, Zainudin NAIM, Tan SG. Tolerance and biosorption of copper (Cu) and lead (Pb) by *filamentous fungi* isolated from a fresh water ecosystem. *J Environ Sci.* 2011;23(5):824–830.
- Prasenjit B, Sumathi S. Uptake of chromium by Aspergillus foetidus. J Mater Cycles Waste Manag. 2005;7:88–92.
- Kapoor A, Viraraghavan T, Cullimore DR. Removal of heavy metals using the fungus *Aspergillus niger*. *Bioresource Technology*. 1999;70(1):95–104.
- Ahluwalia SS, Goyal D. Microbial and plant derived biomass for removal of heavy metals from wastewater. J Bioresource Technology. 2007;98(12):2243–2257.

- 16. Abdel Raouf N, Alharbi RM, Al Enazi NM, et al. Rapid biosynthesis of silver nanoparticles using the marine red alga *Laurencia catarinensis* and their characterization. *Beni–Suef University Journal of Basic and Applied Sciences*. 2018;7(1):150–157.
- Javaid A, Bajwa R, Javaid A. Biosorption of heavy metals using a dead macro fungus *Schizophyllum commune fries:* evaluation of equilibrium and kinetic models. *Pak J Bot.* 2010;42:2105–2118.
- Srivastava S, Thakur IS. Biosorption Potency of Aspergillus niger for Removal of Cr(VI). Current Microbiology. 2006;53(3):232–237.
- Khambhaty Y, Mody K, Basha S, et al. Kinetics, equilibrium and thermodynamic studies on biosorption of hexavalent chromium by dead fungal biomass of marine *Aspergillus niger*. J Chem Eng. 2009;145(3):489–495.
- Al Garni SM, Ghanem KM, Bahobail AS. Biosorption Characteristics of *Aspergillus fumigatus* in Removal of Cadmium from an Aqueous Solution. *African Journal of Biotechnology*. 2009;8:4163–4172.
- Cabuk A, Ilhan S, Filik C, et al. Pb<sup>2+</sup> Biosorption by Pretreated Fungal Biomass. *Turkish Journal of Biology*. 2005;29(1):23–28.
- Soleimani N, Fazli MM, Ramazani A, et al. Application of Live, Dead, and Dried Biomasses of *Aspergillus Versicolor* for Cadmium Biotreatment. J Hum Environ Health Promot. 2016;1(2):87–98.
- Sun YM, Horng CY, Chang FL, et al. Biosorption of Lead, Mercury and Cadmium Ions by *Aspergillus terreus* Immobilized in a Natural Matrix. *Polish Journal of Microbiology*. 2010;59(1):37–44.
- Hassan SW, El Kassas HY. Biosorption of Cadmium from Aqueous Solutions Using a Local Fungus Aspergillus cristatus (Glaucus Group). African Journal of Biotechnology. 2012;11:2276–2286.
- Chen CY, Yang CY, Chen AH. Biosorption of Cu(II), Zn(II), Ni(II) and Pb(II) ions by cross–linked metal–imprinted chitosans with epichlorohydrin. *J Environ. Manag.* 2011;92(3):796–802.
- Tella AC, Owalude SO, Ojekanmi CA, et al. Synthesis of copperisonicotinate metal-organic frameworks simply by mixing solid reactants and investigation of their adsorptive properties for the removal of the fluorescein dye. J New Chem. 2014;38(9):4494–5000.
- Ratnasri PV, Hemalatha KPJ. Studies on Biosorption of Different Metals by Isolates of Aspergillus Species. IOSR Journal of Pharmacy and Biological Sciences (IOSR–JPBS). 2015;10(5):01–05.
- Taştan BE, Dönmez G. Single and binary biosorption of heavy metal ions and a reactive dye by *Aspergillus Versicolor*. *Commun. Fac Sci Univ Ank Series B*. 2010;56(1):13–25.
- Dhankhar R, Hooda A. Fungal biosorption-an alternative to meet the challenges of heavy metal pollution in aqueous solutions. *J Environmental* technology. 2011;32(5–6):467–91.
- 30. Al Asheh SMR, Jumah YR, Banat F, et al. The use of experimental factorial design for analyzing the effect of spray dryer operating variables on the production of tomato powder. *Trans IChem E*. 2003;81:81–88.
- Sugasini A, Rajagopal K, Banu N. A Study on Biosorption Potential of Aspergillus sp of Tannery Effluent. Advances in Bioscience and Biotechnology. 2014;5:853–860.
- Wang J, Chen C. ' Biosorbents for heavy metals removal and their future'. J Biotechnology advances. 2009;27(2):195–226.
- Fomicheva GM, Vasilenko OV, Marfenina OE. 'Comparative morphological, ecological, and molecular studies of *Aspergillus verseicolor* (Vuill.) Tiraboschi strains isolated from different ecotopes'. *Microbiology*. 2006;75(2): 228–234.

- 34. Yinan L, Zhao W, Xu, J, et al. 'Characterization of two related exoantigens from the biodeteriogenic fungus *Aspergillus versicolor*'. *International Biodeterioration & Biodegradation*. 2011;65(1):217–226.
- Nielsen KF. 'Mycotoxin production by indoor molds'. Fungal Genetics and Biology. 2003;39(2):103–117.
- Klaus HD. 'Compendium of soil fungi'. Eching: IHW–Verlag; 2007. 97 p.
- Blázquez G, Hernáinz F, Calero M. et al. 'Removal of cadmium ions with olive stones: the effect of some parameters'. *J Process Biochem*. 2005;40(8):649–654.
- Dogan CE, Turhan K, Akcin G. et al. 'Biosorption of Au (III) and Cu (II) from aqueous solution by a non-living *Cetrariaislandica (L.)*'. *J Ach Ann Chim.* 2006;96(3–4):229–236.
- Siddiquee S, Rovina K, Azad SA, et al. 'Heavy metal contaminants removal from wastewater using the potential *Filamentous Fungi* Biomass: A Review'. *J Microb Biochem Technol*. 2015;7:384–393.

- Iram S, Abrar S. 'Biosorption of copper and lead by heavy metal resistant fungal isolates'. *J Scientific and Research Publications*. 2015;5(1):2250– 3153.
- Fiol N, Villaescusa I, Martinez M, et al. 'Sorption of Pb(II), Ni(II), Cu(II) and Cd(II) from aqueous solution by olive stone waste'. *J Sep Purif Tech.* 2006;50(1):132–140.
- Bayramoğlu G, Bektaş S, Arica MY. 'Biosorption of heavy metal ions on immobilized white-rot fungus *Trametes versicolor*'. J Haz Mat. 2003;101(3):285–300.
- Dehghani MH, Sanaei D, Ali I, et al. 'Removal of chromium (VI) from aqueous solution using treated waste newspaper as a low-cost adsorbent: kinetic modeling and isotherm studies'. *J Mol Liquids*. 2016;215:671– 679.