Hydrometallurgy 97 (2009) 98–104

Contents lists available at ScienceDirect

Hydrometallurgy

journal homepage: www.elsevier.com/locate/hydromet

Removal of copper(II) ions from synthetic solution and real wastewater by the combined action of dried *Trametes versicolor* cells and montmorillonite

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ARTICLE INFO

Article history:
Received 18 March 2008
Received in revised form 23 December 2008
Accepted 24 January 2009
Available online 7 February 2009

Keywords:
Biosorption
Copper(II)
Immobilization, White rot fungi
Isotherms, Wastewater

ABSTRACT

A combination of white rot fungi *Trametes versicolor* and montmorillonite type clay was studied for its ability to remove copper(II) ions from copper contaminated solutions. The effects of process parameters i.e. pH, sorbent dosage, contact time and temperature on the biosorption were examined. Reasonably rapid metal sequester rate was obtained. Maximum copper(II) biosorption capacity was found as 9.89×10^{-4} mol g^{-1} (62.80 mg g^{-1}) at pH 5.0, 1.2 g L^{-1} sorbent dosage and 20 °C. Batch biosorption process follows Langmuir, Freundlich and Dubinin-Radushkevich isotherm models. Zeta potential measurements showed that immobilized biosorbent was negatively charged in the pH range of 2.0–8.0. Changes in the texture surface of the biosorbent were screened by using scanning electron microscopy after immobilization and metal sorption processes. Interactions between sorbent and metal ion were examined by FTIR analysis. The proposed immobilized fungal biomass was successfully used to sequester of copper ions from real wastewater in continuous mode. These results indicated that montmorillonite immobilized cells of *T. versicolor* were an efficient alternative for the removal of copper(II) ions from synthetic solutions and real wastewater.

1. Introduction

Pollution from heavy metals has become a serious problem recognized in many countries of the world. The heavy metal contents in various water supplies often exceed the allowable limits. The major sources of water pollution with heavy metals are the aqueous waste streams of many industries (Lodeiro et al., 2006; Basso et al., 2002). For example, effluents from metal cleaning and plating baths, paper board mills, pulp, wood pulp production and fertilizer industries contain often high concentrations of copper ions (Özer et al., 2004).

In humans, copper is necessary for the development of connective tissue, nerve coverings and bone (Fraga, 2005) and is also incorporated into several enzymes which catalyze important reactions (Seth et al., 2004). However, it becomes toxic and carcinogenic when a large dose is ingested. There are several reports on the acute copper poisoning/toxicity in the literature. These include, abdominal pain, nausea, vomiting, headache, lethargy, diarrhea, tachycardia, respiratory difficulties, hemolytic anemia, gastrointestinal bleeding, liver and kidney failure and death (Janet et al., 2005; World Health Organization, 1998). Since heavy metal ions are not biodegradable agents, they are usually removed from the contaminated water by physical or chemical treatment processes. Conventional treatment methods (precipitation, membrane separation, ion exchange, reverse osmosis and electrolysis etc.) are not often feasible because of high treatment cost, the need for continuous input of chemicals, and the production of toxic sludge (Tunali and Akar, 2006a,b; Han et al., 2006; Abu Al-Rub et al., 2006).

Biosorption technology is based on the interaction between toxic metals and the binding functional groups on the cell wall structure of the microorganisms. These are mainly composed of polysaccharides, lipids and proteins. Biosorption has recognized as a potential alternative method over the conventional separation techniques. This process utilizes live, dead, pretreated and immobilized forms of biological cells like bacteria, fungi, yeast and algae as sorbent materials (Gadd, 1990; Volesky, 1990; Kapoor and Viraraghavan, 1997).

Immobilized forms of the biosorbent materials have more convenient in practical applications because of their mechanical strength, anti-degradation ability, physical morphology and chemical stability as well as capability of regeneration (Kapoor and Viraraghavan, 1998; Cabuk et al., 2006a). Different kinds of support materials like alginate (Prakasham et al., 1999), Ca-alginate, polyvinyl alcohol (PVA), polyacrylamide, polyisoprene, polysulfone (Bai and Abraham, 2003), silica gel (Cabuk et al., 2006a) and sepiolite (Bağ et al., 1998) have been used for the immobilization of biological cells.

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doi:10.1016/j.hydromet.2009.01.009

Trametes versicolor is a white rot fungi with commercial availability and has been used in sequester of several metal ions from aqueous solutions (Bayramoğlu et al., 2003; Wilkoazka et al., 2002; Arıca et al., 2003) and decolorization of dye bearing solutions (Chander and Arora, 2007; Blánquez et al., 2004). However to our knowledge there is no report in the literature concerning the use of *T. versicolor* immobilized on natural clay as a sorbent material.

In the present paper we report the study of using a combination of *T. versicolor* cells and montmorillonite type clay mineral for removal of copper(II) ions from aqueous solution. The sorption properties of developed sorbent were studied in batch mode as a function of initial pH, sorbent dosage, contact time and temperature. The sorption equilibrium was modeled by Freundlich, Langmuir and Dubinin-Radushkevich (D-R) isotherm models. The copper (II) removal potential of the sorbent system by using a real wastewater was also reported in the continuous mode.

2. Material and methods

2.1. Preparation of the sorbent material

The montmorillonite used in this study was provided from Eskisehir, Turkey. It was crushed, ground and sieved through a 150 µm sieve, and dried at 110 °C in an oven prior to use as a support material. The chemical analysis of the clay mineral was performed by using a 8400/60 Fison Model XRF equipment. Montmorillonite is composed of SiO_2 49.86%, MgO 12.37%, Al_2O_3 9.24%, Na_2O 6.39%, Fe_2O_3 4.51%, CaO 2.52%, K_2O 1.93% and loss on ignition 13.18% and CEC (cation exchange capacity) is 102.18 meq/100 g clay (Şölener et al., 2008). The pure culture of *T. versicolor* (ATCC 200801) was obtained from the collection of Biology Department of Eskişehir Osmangazi University. The culture was routinely maintained by sub-culturing on malt extract agar slants. The details of the cell cultivation were described earlier (Cabuk et al., 2006b). The cultured fungi were separated from the culture medium by filtration through Whatmann No 1 filter paper and the harvested mycelia were washed with several times thoroughly with deionized water. The filtration process was followed by drying in an oven at 60 °C overnight. Subsequently, dried sorbent was ground and sieved to obtain particle size of 150 µm. A procedure previously recommended by Mahan and Holcombe (1992) was followed for the preparation of the sorbent material. 150 mg of dried sorbent was mixed with 2 g of montmorillonite and the mixture was wetted with deionized water and thoroughly mixed. The resulting paste was heated in an oven at 60 °C until the constant weight. The wetting and drying steps were repeated for five times to maximize contact between sorbent and clay surface. The immobilized sorbent was then sieved to obtain original particle size. The surface characteristics of biomass were analyzed by FTIR and SEM methods and the surface charge of the biomass was measured using a Zeta potential analyzer (Malvern Zetasizer nano ZS). FTIR spectra of unloaded and copper(II) loaded sorbent were recorded in a Perkin Elmer Spectrum 100IR infrared spectrometer in the region of 400–4000 cm^{-1} with the samples prepared as KBr pellets under high pressure. The surface structure and morphology of the sorbent material before and after copper(II) sorption were characterized using scanning electron microscope (JEOL 560 LV SEM), with 20 kV and at 250 times of magnification. Prior to analyze, samples were sputter coated with a thin layer of gold under argon atmosphere to improve the electron conductivity and the image quality.

2.2. Preparation of copper (II) solutions

The stock solution of copper(II) (1.0 g L^{-1}) used in this study was prepared by dissolving an accurate quantity of $CuSO_4.5H_2O$ in deionized water. The required concentrations were prepared by diluting this stock solution. The pH of the test solutions was adjusted to the desired values by adding 0.1 N HNO_3 or 0.1 N NaOH solutions.

2.3. Real wastewater sample

The industrial wastewater sample was collected from the main drain of the casting unit of metal processing industry in Eskişehir, Turkey. Wastewater sample was placed in a sterile container and transferred to laboratory and stored at 5 °C.

2.4. Sorption experiments

50 mL of copper(II) solution (50 mg L^{-1}) was put in a beaker and 0.05 g of immobilized material was added. The mixture was stirred at 200 rpm at room temperature for 1 h and was then centrifuged at 4500 rpm for 5 min for the separation of solid phase from the liquid phase. The residual copper(II) concentrations in the supernatant were analyzed using an Atomic Absorption Spectrophotometer (Hitachi 180-70, Japan) with an air-acetylene flame. Copper hallow cathode lamp and deuterium background correction were used. The spectral slit width and specific current/wavelengths were 1.3 nm and 7.5 mA/324.8nm, respectively. The instrument response was periodically checked by standard copper(II) solutions. The sorption capacity of immobilized system (q_e) was determined by using the following equation;

$$q_e = \frac{V(C_i - C_e)}{m} \tag{1}$$

where C_i and C_e are the initial and the equilibrium copper(II) concentrations (mg L^{-1}), respectively, V is the volume of the solution (L) and m is the amount of sorbent used (g).

The effect of pH on the copper(II) biosorption capacity of sorbent system was examined by equilibrating the sorption mixture containing immobilized sorbent (0.05 g) and metal solutions (50 mL of 50 mg L^{-1}) at different pH values between 1 and 6, adding freshly prepared 0.1 M HNO_3 or 0.1 M NaOH solutions. The pH of the medium was checked during the sorption process and no significantly change was observed in the initial pH values. The effect of sorbent concentration was investigated by using sorbent amount ranging from 0.4 to 4.0 g L^{-1}. This was followed by the assessment of the effect of equilibrium time varied between 5 and 60 min.

The appropriateness of the Langmuir, Freundlich and (D-R) isotherm models was investigated by varying the initial copper(II) concentrations between 25 and 200 mg L^{-1} at the temperature range of 20 to 40 °C and the pH value of 5.0.

2.5. Applicability

The proposed sorption method was applied to real wastewater sample in a continuous system. Fixed bed experiments were

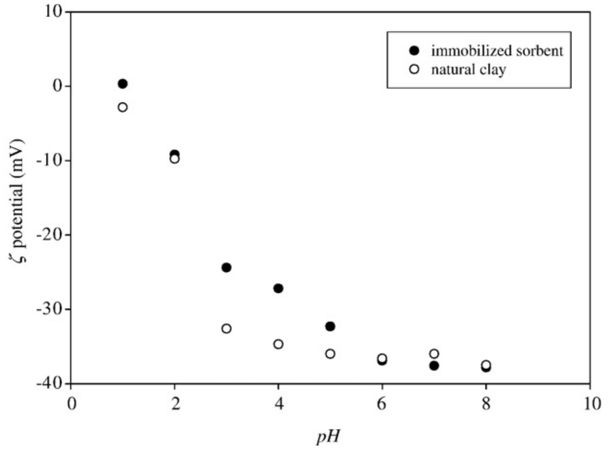

Fig. 1. ζ potentials as function of pH for natural clay and immobilized sorbent.

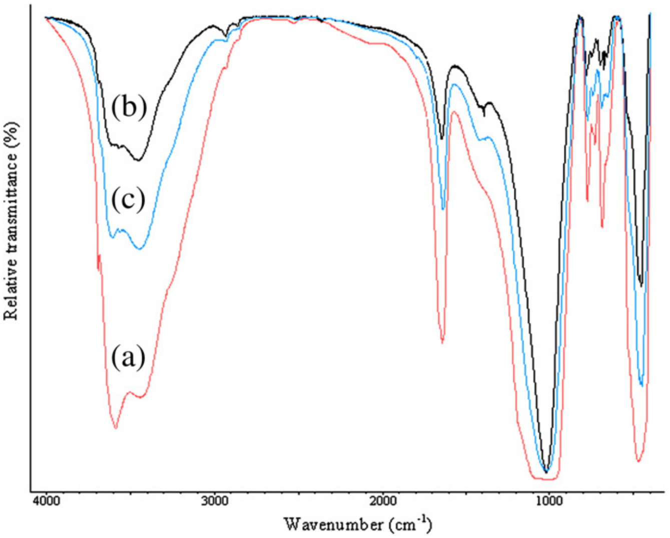

Fig. 2. FTIR spectra for (a) clay; (b) *T. versicolor*-clay and (c) copper(II) loaded *T. versicolor*-clay.

performed in a glass column (10.0 cm length and 5 mm internal diameter). 0.06 g of immobilized sorbent was packed between two layers of glass wool into the column. Wastewater sample (pH 5.0) was pumped from bottom to top through the column at a flow rate of 1.0 mL min^{-1} using a peristaltic pump (Heidolph PD5201). The effluent was analyzed for the residual copper(II) concentration by AAS as described above. The column experiments were carried out at room temperature. The sorption yield was calculated by using the following equation:

$$\textit{Sorption yield} = \frac{C_i - C_e}{C_i} \times 100. \qquad (2)$$

3. Results and discussion

3.1. Characterization of sorbent

Fig. 1 shows the zeta (ζ) potentials of natural clay and immobilized sorbent as a function of solution pH in the range of 1.0–8.0. The natural clay was negatively charged at all pH values studied and the negative charge density (zeta potential) of natural clay increased from −2.83 to −34.70 mV with an increase in the pH of the medium from 1.0 to 4.0. The zeta potential of clay did not significantly change with further increase in the solution pH. In the case of the immobilized sorbent, zeta potential was positive (0.32 mV) at pH 1.0. This could be attributed to the immobilization of *T. versicolor* which was positively charged at strongly acidic conditions due to the protonation of functional groups on the biomass surface. Above pH 1.0 immobilized system was negatively charged which was providing attractive forces to positively charged copper(II) ions.

Table 1
Band shifting after immobilization and Cu(II) sorption.

Band positions (cm^{-1})			
Suggested assignment	Montmorillonite	Immobilized sorbent	Cu(II) loaded sorbent
–OH stretching or H-bonding to Si–O–Al linkage	3586	3604	3605
–OH stretching or H-bonding to Si–O–Al linkage	3429	3436	3443
CH_2-symmetric stretching[a]		2926	2930
CH_2-asymmetric stretching[a]		2856	2857
–OH bending vibrations	1636	1634	1633
CH_2-bending vibrations[a]		1390	1413
Si–O vibrations	1090–964	1013	1017
Si–O–Al (octahedral) bending vibrations	686	688	689
Si–O–Si bending vibrations	473	449	455

[a] Denotes additional bands after immobilization.

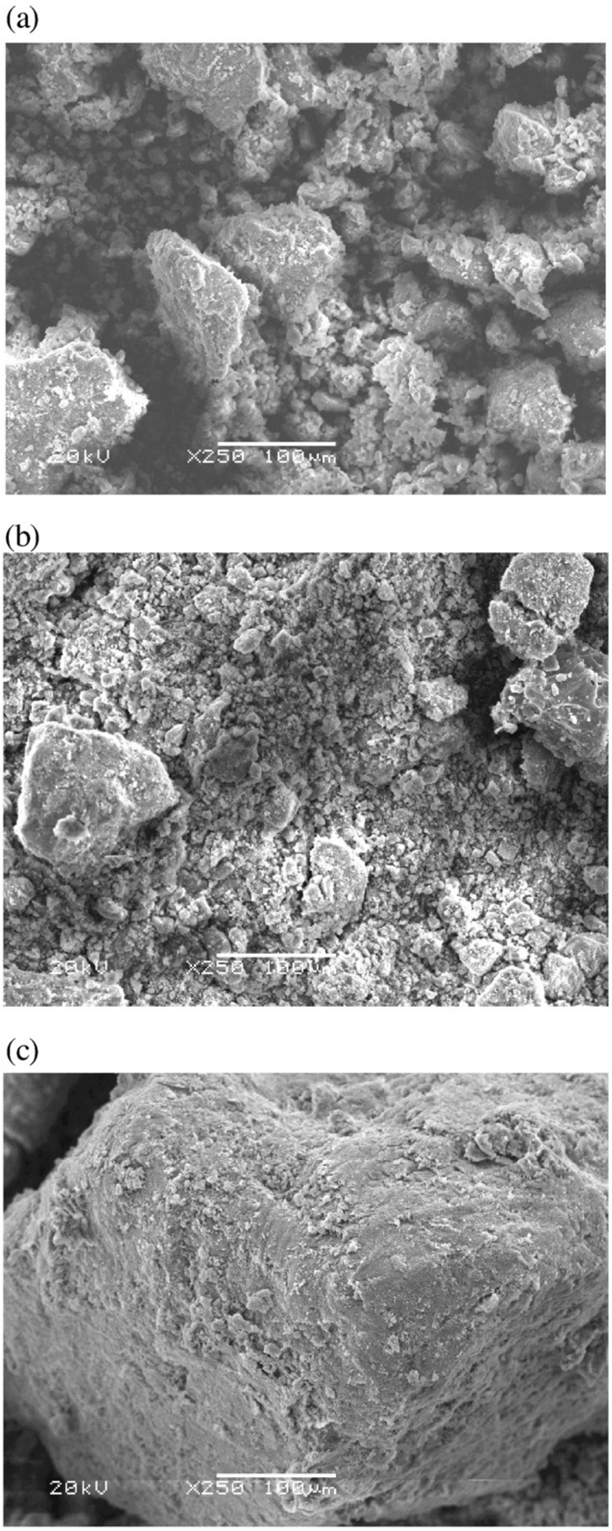

Fig. 3. SEM images for (a) montmorillonite; (b) unloaded and (c) copper(II) loaded immobilized biomass.

The FTIR spectra of clay, clay-based immobilized sorbent and copper(II)–loaded immobilized sorbent (Fig. 2) in the range of 400–

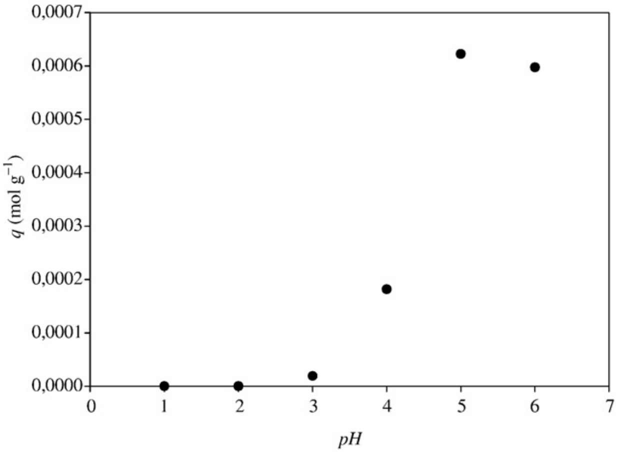

Fig. 4. Effect of initial pH on the copper (II) sorption by immobilized sorbent.

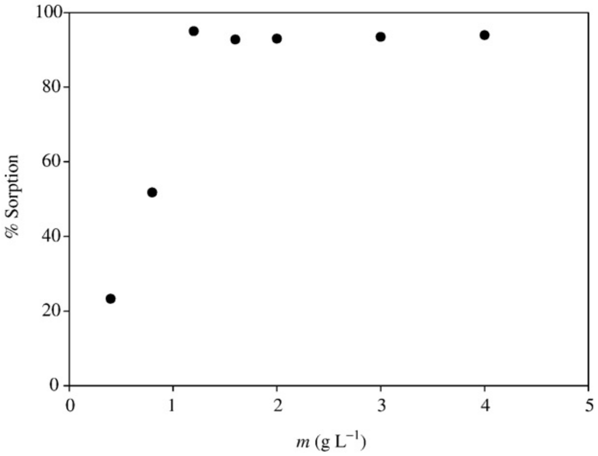

Fig. 6. Effect of sorbent dosage on the copper (II) sorption yield by immobilized sorbent.

4000 cm^{-1} were taken and compared with each other to obtain information on the immobilization of the *T. versicolor* with clay mineral and the nature of the possible sorbent–metal ions interactions. Band positions for each sample are listed in Table 1.

The bands at 3586 and 3604 cm^{-1} are indicative of H–O–H stretching vibration of water molecules in the natural clay mineral and clay-biomass sorbent, respectively (Fig. 2 (a) and (b)). The broad band at 3436 cm^{-1} in the FTIR spectra of immobilized sorbent is due to H–O–H vibrations of adsorbed water and this peak shifted to 3443 cm^{-1} after copper(II) sorption (Fig. 2 (c)) by immobilized sorbent. The bands observed at 2926 and 2856 cm^{-1} are indicative of symmetric and asymmetric stretching vibrations of –CH$_2$ groups and their bending vibrations were observed at 1390 cm^{-1} for clay-based biological sorbent but these stretching bands are not observed in the FTIR spectrum of natural clay mineral. These findings supported that the fungal biomass was deposited at the surface of clay particles. In addition, the adsorption band at 1636 cm^{-1}, assigned to –OH bending vibration of H$_2$O of the natural clay, weakened in the FTIR spectra of immobilized sorbent as a result of immobilization procedure.

The broad Si–O coordination band between 1090 and 964 cm^{-1} is observed as a result of Si–O vibrations. The deep band at around 686 cm^{-1} represents the bending of Si–O in Si–O–Al groups of the octahedral sheet. The bands at 473 and 449 cm^{-1} are due to Si–O–Si bending vibrations for the natural clay and immobilized sorbent, respectively.

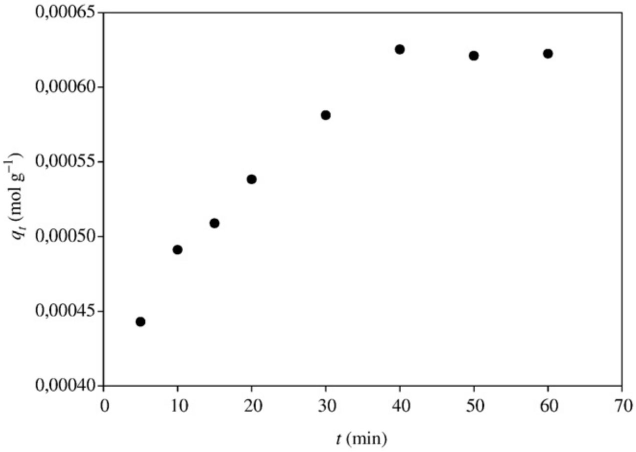

Fig. 5. Equilibrium time profile for the copper (II) sorption by immobilized sorbent.

The bands at 3436 and 1390 cm^{-1} for unloaded immobilized biomass shifted to 3443 and 1413 cm^{-1}, respectively, after copper(II) sorption. Also bending vibrations of Si–O–Si was observed at 455 cm^{-1} in the FTIR spectra of Cu(II)-loaded immobilized biomass. Consequently, the changes observed in the spectra indicated the possibly interaction between copper(II) ions and the functional groups on the surface of the biomass during the sorption process.

Fig. 3 shows the SEM images of natural clay (a) and unloaded immobilized sorbent (b). It can be seen that after the immobilization procedure, *T. versicolor* was uniformly distributed on the montmorillonite. The SEM micrograph of copper(II) loaded immobilized sorbent was also represented in Fig. 3(c). The structure of the immobilized sorbent changed after the copper(II) sorption and the pore sizes of the sorbent significantly decreased. This could be attributed to the contribution of macro-porous structure of the sorbent for the sorption of copper(II) ions.

3.2. Effect of pH

The batch studies were conducted with different initial pH values ranging from 1.0 to 6.0 in order to elucidate the effect of pH on the sorption of copper(II) by immobilized sorbent. The results presented in Fig. 4 showed that the copper(II) sorption was significantly affected by the initial pH of the solution. The maximum uptake of copper(II) ions by immobilized system was observed at pH 5.0 and it decreased below this pH value and almost no sorption took place at an initial pH between 1.0 and 3.0. Metal binding sites on the sorbent system were closely associated with hydronium ions (H$_3$O$^+$) at lower pH, which cause a decrease in the electrostatic attraction between positively charged sorbent surface and copper(II) ions. The sorption capacity of immobilized sorbent increased when the pH of the solution was increased from 3.0 to 5.0. An increase in the sorption capacity with an increase in the solution pH can be explained by the negatively charged metal binding sites on the sorbent surface as a result of deprotonation and it facilitated the approach of the copper(II) ions. The precipitation of copper(II) in hydroxide form was seen to occur at higher pH values from 6.0. Therefore the experiments were not conducted beyond pH 6.0 to avoid copper precipitation. It can be concluded from these results, copper(II) removal by sorbent system is purely sorption and precipitation does not contribute the removal process.

3.3. Effect of contact time

Fig. 5 shows the effect of time course profiles for the sorption of copper(II) by immobilized sorbent. In order to determine the effect of contact time on the sorption of copper(II) ions, the contact time was

Table 2
Isotherm model constants for the sorption of copper (II) onto immobilized system.

t (°C)	Langmuir constants				Freundlich constants			D-R constants			
	q_{max} (mol g^{-1})	K_L (L mol^{-1})	r_L^2	R_L	n	K_F (L g^{-1})	R_F^2	q_{max} (mol^{2}mol^2 kJ^{-2})	B (mol g^{-1})	$r_{D\text{-}R}^2$	E (kJ mol^{-1})
20	9.89×10^{-4}	3.19×10^{3}	0.990	0.091	3.147	6.49×10^{-3}	0.969	2.11×10^{-3}	1.93×10^{-3}	0.982	11.56
30	9.30×10^{-4}	2.03×10^{3}	0.995	0.135	2.074	1.65×10^{-2}	0.968	2.85×10^{-3}	2.26×10^{-3}	0.984	9.88
40	8.94×10^{-4}	1.43×10^{3}	0.999	0.182	1.915	1.81×10^{-2}	0.976	2.78×10^{-3}	2.30×10^{-3}	0.989	9.72

varied from 5 to 60 min. It was observed that copper(II) sorption by immobilized sorbent relatively occurs rapidly and equilibrium was reached within 40 min. The initial rapid phase is probably due to the abundant of availability of active metal binding sites on the sorbent surface and the gradual occupancy of those sites. The sorption becomes less efficient in the slower stage (Saeed et al., 2005a,b; daCosta and Leite, 1991) as a result of the competition for decreasing available active binding sites on the sorbent surface for metal ions remained in the solution. The rate of metal-sorption is of great significance for developing a microbial origin sorbent-based water-treatment technology (Saeed et al., 2005a,b) and practical application of process (Akar and Tunali, 2005).

3.4. Effect of sorbent dosage

The effect of sorbent dosage on the copper(II) sorption percentages is presented in Fig. 6. The sorption yield of copper(II) ions increased with an increase in the sorbent dosage and reached the maximum value at a sorbent dosage of 1.2 g L^{-1}. Almost no considerable changes were observed for further increase in the sorbent dosage up to 4.0 g L^{-1}. Therefore 1.2 g L^{-1} was chosen as the optimum sorbent dosage in further experiments. An increase in the sorption yield with the sorbent dosage can be explained by the increased surface area of sorbent and availability of more binding sites for metal ions (Gong et al., 2005).

3.5. Sorption isotherms

The equilibrium copper(II) sorption data were evaluated with Freundlich, Langmuir and Dubinin-Radushkevich (D-R) isotherm models at a temperature range of 20–40 °C. The Freundlich isotherm equation is an empirical equation based on the sorption onto heterogeneous surface and is given by Eq. (3) (Freundlich, 1906);

$$lnq_e = lnK_F + \frac{1}{n} lnC_e, \tag{3}$$

where K_F (L g^{-1}) and n (dimensionless) are Freundlich isotherm constants that indicate the extent of the sorption, and the degree of nonlinearity between solution concentration and sorption, respectively.

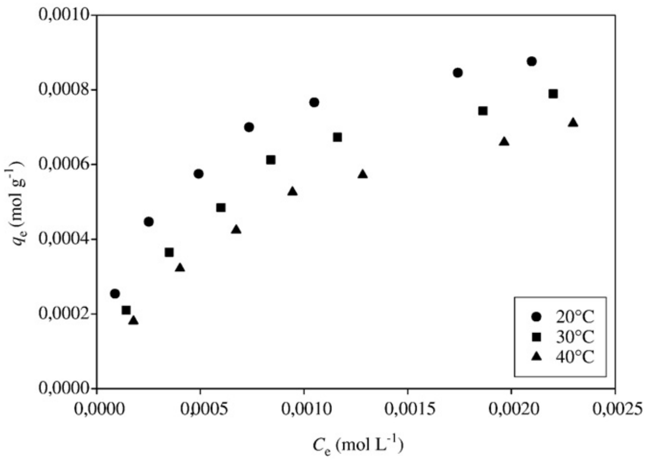

Fig. 7. General isotherm plots for the copper (II) sorption by immobilized sorbent at different temperatures.

The Langmuir isotherm model based on the sorption onto a surface a finite number of identical sites by monolayer sorption and expressed by (Langmuir, 1918);

$$\frac{1}{q_e} = \frac{1}{q_{max}} + \left(\frac{1}{q_{max}K_L}\right)\frac{1}{C_e} \tag{4}$$

where q_e and q_{max} are the equilibrium and monolayer copper(II) sorption capacities of the sorbent (mol g^{-1}), respectively, C_e is the equilibrium copper(II) concentration in the solution (mol L^{-1}) and K_L is the Langmuir adsorption constant (L mol^{-1}) and is related to the free energy of adsorption (Weber and Chakravorti, 1974).

The effect of isotherm shape has been discussed (Hall et al., 1966) with a view to predict whether a sorption system is favorable or unfavorable. The essential feature of the Langmuir isotherm can be expressed by means of 'R_L', a dimensionless constant referred to as separation factor or equilibrium parameter and R_L is calculated using the following equation,

$$R_L = \frac{1}{1 + K_L C_o}, \tag{5}$$

where K_L is the Langmuir constant (L mol^{-1}) and C_o is the highest initial copper(II) concentration (mol L^{-1}).

The D-R isotherm model is more general than the Langmuir isotherm. It was applied to distinguish the nature of sorption as physical or chemical (Benhammou et al., 2005). The D-R isotherm equation (Dubinin and Radushkevich, 1947) is:

$$lnq_e = lnq_m - \beta\varepsilon^2 \tag{6}$$

where β is a constant related to the mean free energy of sorption (mol^2 J^{-2}), q_m is the theoretical saturation capacity, and ε is the

Table 3
Sorption results of copper ions by different sorbents from the literature.

Sorbent material	Sorption capacity (mg g^{-1})	References
Cotton boll	11.40	Ozsoy and Kumbur (2006)
Activated nylon-based membrane	10.79	He et al. (2008)
Tea waste	48.00	Amarasinghe and Williams (2007)
Botrytis cinerea	9.23	Akar and Tunali (2005)
Ca-alginate immobilized algae	75.60	Bayramoglu and Arica (2009)
Expanded perlite	8.62	Sari et al. (2007)
Manganese oxide modified bentonite	105.38	Eren (2008)
2,2-dipyridyl immobilized bentonite	54.07	Erdem et al. (2009)
Newspaper pulp	30.00	Chakravarty et al. (2009)
Cankırı bentonite	44.84	Veli and Alyüz (2007)
Rubber leaf powder	8.92	Wan Ngah et al. (2008)
Bacillus sp.	16.25	Tunali et al. (2006b)
Chitosan coated PVC	87.90	Popuri et al. (2009)
Aspergillus flavus	10.82	Akar and Tunali (2006)
Activated carbon from Hazelnut husk	6.65	Imamoglu and Tekir (2008)
Ca-alginate immobilized fungi	2.76	Yahaya et al. (2009)
Trametes versicolor immobilized montmorillonite	62.80	This work

Table 4
The some chemical characteristics of wastewater sample at various pH values.

Parameters (mg L^{-1})	Effluent quality (pH 2.24)	Effluent quality (pH 5.0)
Copper	46.86	28.13
Lead	12.35	1.29
Nickel	12.05	9.43
Cadmium	14.83	10.41
Iron	214.00	110.00
Zinc	393.00	188.50
Sodium[a]	57.50	746.50
Potassium	230.50	78.50
Calcium	112.00	111.50
Magnesium	182.00	149.00

[a] Since the pH of the sample was adjusted to 5.0 with NaOH, the concentration of sodium increased.

Polanyi potential, which is equal to $RT\ln(1+1/C_e)$ R (J mol^{-1} K^{-1}) is the gas constant, and T (K) is the absolute temperature.

The constant β gives an idea about the mean free energy E (kJ mol^{-1}) of sorption and can be calculated using the relationship (Hobson, 1969; Hasany and Chaudhary, 1996; Dubey and Gupta, 2005);

$$E = \frac{1}{(2\beta)^{1/2}}. \tag{7}$$

E value gives information about sorption mechanism as chemical ion-exchange or physical sorption. The mean free energy of copper(II) sorption at the temperature of 20 and 40 °C ranged from 11.56 to 9.72 kJ mol^{-1}. This result may be attributed to a chemical ion exchange mechanism for the copper(II) sorption process.

The Langmuir, Freundlich and D-R parameters and general isotherm plots for the sorption of copper(II) onto immobilized sorbent at different temperatures are given in Table 2 and Fig. 7. It is indicated that the Freundlich, Langmuir and D-R isotherm models are suitable for describing the copper(II) sorption equilibrium by immobilized sorbent in the studied concentration and temperature range with the regression coefficient (r^2) values more than 0.975. If the R_L values lie between 0 and 1, the adsorption process is favorable (Hall et al., 1966). The R_L values for this study were between 0.091 and 0.182, therefore, the sorption of copper(II) was favorable.

The Freundlich constants K_F and n indicate the sorption capacity of the sorbent and a measure of the deviation from linearity of the sorption, respectively. The values of K_F and n at the different temperatures were between 6.49×10^{-3} - 1.81×10^{-2} and 3.147–1.915, respectively. The well description of the experimental results with all isotherm models investigated in this study implies that the sorption of copper(II) ions onto immobilized sorbent is complex and involving more than one mechanism.

The maximum monolayer sorption capacity (62.80 mg g^{-1}) of the immobilized sorbent was comparable and found to be higher than that of many sorbents reported in the literature (Table 3).

3.6. Sorption of Cu(II) from industrial waste

The proposed copper(II) sorption process was applied to real wastewater in a column at predetermined optimum conditions. When the pH of the wastewater was adjusted to 5.0, the concentration of some metal ions decreased due to the precipitation of metal ions in the form of metal hydroxide except for sodium. The various characteristics of the wastewater at original pH of 2.24 and 5.0 are given in Table 4. The sorption studies were carried out after the precipitate was removed by filtration and 66.40% sorption yield was observed for real wastewater sample. In industrial wastewater the sorption yield slightly decreased due to the presence of other metal ions in the wastewater along with the Cu(II) which interfere for the copper sorption. However in the light of these results it could be concluded that the prepared sorbent was found to be very efficient for the removal of copper(II) ions from real wastewater.

4. Conclusion

The presented study is based on the immobilization of *T. versicolor* on montmorillonite and the sorption of copper(II) ions by the immobilized system. The initial pH of the medium is significantly effected the sorption capacity of sorbent and the optimum value was found as 5.0. The sorption equilibrium was rapidly established (40 min). The equilibrium sorption data were well described by Freundlich, Langmuir and D-R isotherm models. The sorption energy in the range of 9.72 to 11.56 kJ mol^{-1} showed the one of the sorption mechanisms can be chemical ion-exchange. Complexation with the functional groups on the sorbent surface which are responsible for copper(II) sorption was also approved by FTIR analysis. The proposed sorption method was successfully applied to real wastewater including analyte ions. Consequently, the sorption process for copper(II) by prepared sorbent material was found to be simple, fast and efficient.

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