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# Optimization, equilibrium, kinetic modeling, and thermodynamic studies of biosorption of aniline blue by the dead biomass of *Aspergillus fumigatus*

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ABSTRACT

The effect of biosorbent concentration, initial pH, temperature, agitation rate, adsorption time, and initial dye concentration was studied for the biosorption of aniline blue dye using dead fungal biomass of *Aspergillus fumigatus*. The maximum biosorption of aniline blue was observed at the sorbent concentration of 1 g/L, initial pH 10, temperature 30°C, agitation rate 160 rpm, and initial dye concentration of 50 mg/L. The experimental data were analyzed using Freundlich, Temkin, and Scatchard equilibrium isotherm models out of which the Freundlich isotherm ($R^2 = 0.98$) was found to best fit the experimental data. Thermodynamic parameters such as enthalpy change (−3.08 kJ/mol) and entropy change (8.32 J/mol K) were also calculated. The biosorption kinetics of Aniline blue was found to obey pseudo-second-order kinetic model ($R^2 = 0.967$). The results showed that biosorption was favorable and spontaneous, thus, indicating the positive affinity of the dye towards the adsorbent. The characteristics of the fungal biosorbent were studied using Fourier transform-infrared spectroscopy (FTIR) and scanning electron microscopic (SEM).

*Keywords:* Aniline blue; *Aspergillus fumigatus*; Thermodynamic analysis; Biosorption isotherm; Kinetic model

## 1. Introduction

Pollution control is one of the prime concerns of today's society. Untreated or partially treated wastewater and industrial effluents discharged into the natural ecosystems pose a serious problem. Synthetic dyes are extensively used in many industries, such as textile, leather tanning, paper production, food technology, and hair coloring [1]. Wastewater discharged from these industries is usually polluted by dyes that react with metal ions present in the wastewater to form toxic substances which damage the esthetic quality of water bodies, reduce light penetration, and photosynthesis. Some of the dyes are carcinogenic, allergenic, and mutagenic causing many waterborne diseases that are threatening to both the mankind and other living beings. Dyes are biologically nondegradable and are, therefore, difficult to decolorize once released into the aquatic environment [2,3].

Chemical methods, such as coagulation, flocculation, electro-floatation; conventional oxidation methods

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using oxidizing agents and irradiation; and physical methods like nanofiltration, reverse osmosis, electrodialysis are available for the decolorization of industrial effluents but are not widely used in large scales because of their higher costs and the disposal problems involved [4–6]. Adsorption has been found to be superior to other techniques for water decolorization in terms of initial cost, flexibility and simplicity of design, ease of operation, insensitive to toxic pollutants, recovery/reuse of adsorbent, and high efficiency. Various adsorbent materials have been used to remove dyes from wastewater. The use of activated carbon [2], clay minerals [6,7], fly ash [8], agricultural wastes [9], micro-organisms [10], and sand [11] as adsorbents were reported for wastewater decolorization. Activated carbon is quite expensive and regeneration produces secondary effluents resulting in considerable loss of the adsorbent [12]. This has led many researchers to search for low-cost and locally available adsorbents.

Biological treatment is the most economical alternative when compared with physical and chemical processes. Biological methods, such as fungal decolorization, microbial degradation, adsorption by (living or dead) microbial biomass, and bioremediation systems are commonly used in the treatment of industrial effluents [4]. Biosorption is a well-known equilibrium separation process for the uptake of dyes using dead biomass, such as plants, fungi, or algae [10,13,14]. It does not result in the formation of harmful substances and is a feasible method for industrial scale [10,15]. Therefore, the dead biomass of *Aspergillus fumigatu*s was used as a biosorbent for the removal of basic dye aniline blue in the present study.

## 2. Materials and methods

### 2.1. Preparation of biosorbent

The filamentous fungus *A. fumigatus* MTCC 870 was obtained from Microbial Type Culture Collection and Gene Bank, Institute of Microbial Technology, Chandigarh, India. *A. fumigatus* was cultivated by submerged fermentation; 1,000 mL of the growth media consisted of 10 mL Czapek concentrate, 1 g $K_2HPO_4$, 5 g yeast extract and 30 g sucrose. The flasks were incubated on a rotary shaker with 160 rpm at 30˚C for three days. The conical flasks were then autoclaved to kill the micro-organisms. The biomass was recovered from the broth and was washed using distilled water for three times followed by 0.1% NaCl treatment and was then dried overnight. The obtained dry flakes of biomass were crushed and sieved. Particles of size 300–500 μm were selected for biosorption studies.

### 2.2. Preparation of dye solution

Aniline blue solution is used as dye solution in this study. Aniline blue (CI 42755) is also known as Acid blue 22, Soluble Blue 3M, and Marine Blue V. It was purchased from SRL India Ltd., Mumbai, India. 1% (w/v) stock solution was prepared by completely dissolving 1 gm of dye in 100 mL of distilled water. The stock solution was diluted to 100 mg/L and used as working stock solution for the biosorption studies.

### 2.3. Batch adsorption studies

Weighed amount of dried biomass was introduced into the experimental dye solution. Biosorption experiments were carried out in a rotary shaker using 250 mL Erlenmeyer flasks with 100 mL experimental dye solution. The effect of biosorbent dosage with the concentrations of 0.1, 0.2, 0.3, 0.4, and 0.5 g in 100 mL of 50 mg/L experimental dye solution (pH 10) was studied at 30˚C at 160 rpm for 60 min. The effect of initial pH on biosorption was studied for different pH ranging from 8 to 12 in 100 mL of 50 mg/L experimental dye solution using the optimum biosorbent dosage of 1 g/L at 30˚C at 160 rpm for 60 min. Then, the effect of agitation on biosorption was studied at different agitation rates namely 100, 120, 140, 160, and 180 rpm using 50 mg/L experimental dye solution, optimum biosorbent dosage 1 g/L, optimum initial pH 10 at 30˚C for 60 min. Similarly, the effects of initial dye concentration and time were studied using the dye solution with five different concentrations ranging from 25 to 125 mg/L prepared in different conical flasks. The initial pH was adjusted to the optimum value of 10. The optimum biosorbent dosage level of 1 g/L was added and kept in an orbital shaker with 160 rpm at 30˚C for 60 min. Samples were withdrawn at different time intervals, centrifuged at 6,000 rpm for 10 min and the absorbance of the supernatant was measured at maximum wavelength of $\lambda_{max}$ at 550 nm using UV spectrophotometer (2201 Systronics). The absorbance was reported in terms of the dye uptake capacity.

### 2.4. FTIR and SEM analysis of biosorbent

The characteristic functional groups present in the fungal biosorbent before and after were analyzed using BRUKER α-T Fourier transform-infrared spectroscopy (FTIR) and the surface characteristics were studied using QUANTA 200 scanning electron microscopic (SEM).

## 3. Results and discussion

### 3.1. Effect of biosorbent dosage and pH

The effect of biosorbent concentration on dye uptake capacity of *A. fumigatus* biomass was studied by varying the biosorbent concentration from 1 to 5 g/L with 100 mL dye solution with fixed initial dye concentration of 50 mg/L. The biosorption capacity of the sorbents decreased from 11.05 to 2.89 mg/g for aniline blue due to increase in biosorbent dosage (Fig. 1). The maximum dye uptake capacity of 11.05 mg/g was obtained for 1 g/L of biosorbent dosage. The decrease in dye uptake capacity may be due to the decreased number of possible binding sites, decreased availability of solute, less electrostatic interactions, and interference between binding sites. The maximum dye uptake was observed for 1 g/L biosorbent dosage, thus the optimum biosorbent concentration was found to be 1 g/L. The effect of pH on biosorption of aniline blue was studied by varying the pH from 8 to 12 with an interval of 1 for fixed initial dye concentration of 50 mg/L and biosorbent dosage of 1 g/L. The biosorption capacity of the biomass was increased when the pH of the solution was increased from 8.0 to 10.0 (Fig. 2). Under low alkaline conditions (pH 10 to 12), biosorption capacity was significantly decreased. The maximum dye uptake capacity of 11.7 mg/g was obtained at pH 10. The increase in the biosorption capacity with increase in pH can be explained by the dye binding sites on the biosorbent surface becoming positively charged thereby enhancing the biosorption of the dye molecules due to the attractive forces [16].

### 3.2. Effect of agitation rate

The effect of agitation rate on dye uptake capacity of *A. fumigatus* biomass was studied by conducting experiments at 100, 120, 140, 160, and 180 rpm using 1 g/L of biosorbent and 100 mL of 50 mg/L of dye solution at pH 10. The dye uptake capacity was increased with increase in agitation rate up to 160 rpm (Fig. 3). The maximum dye uptake capacity of 12.6 mg/g was obtained at an agitation rate of 160 rpm. The decrease in dye uptake capacity was observed when the agitation rate increased from 160 to 180 rpm.

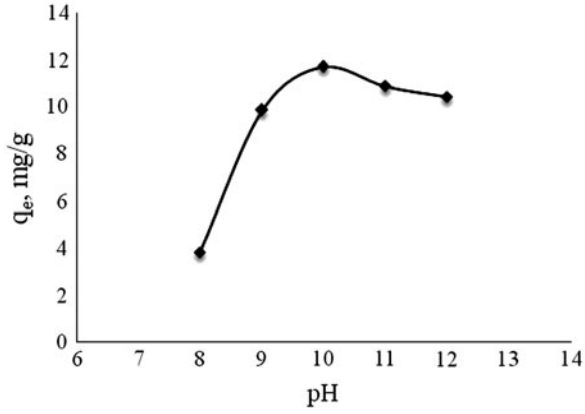


Fig. 2. Effect of pH on equilibrium uptake of aniline blue by *A. fumigatus* biomass at 30°C, 50 mg/L of dye solution, 1 g/L of sorbent, 160 rpm and 60 min.

### 3.3. Effect of time and initial dye concentration

The effect of initial dye concentration on dye uptake capacity of *A. fumigatus* biomass was studied by conducting experiments using varied initial dye solution concentration from 25 to 125 mg/L, pH 10, 1 g/L of biosorbent and at 160 rpm. Samples were collected from 20 to 100 min with an interval of 20 min. The effect of contact time and initial dye concentration

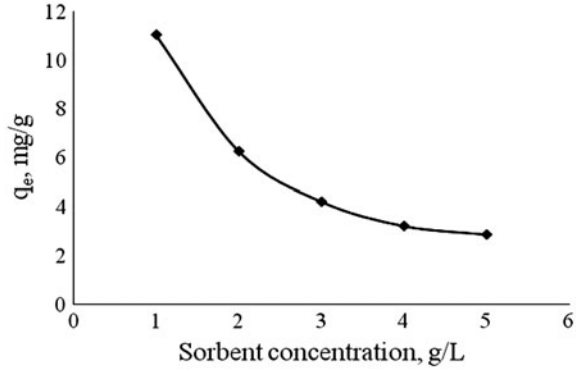


Fig. 1. Effect of sorbent concentration on equilibrium uptake of aniline blue by *A. fumigatus* biomass at 30°C, 50 mg/L of dye solution, 60 min, 160 rpm and pH 10.

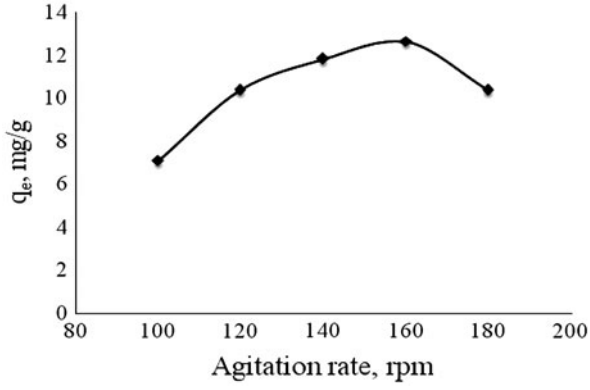


Fig. 3. Effect of agitation rate on absorption of aniline blue by *A. fumigatus* biomass at 30°C, 50 mg/L of dye solution, 1 g/L of sorbent, pH 10 and 60 min.

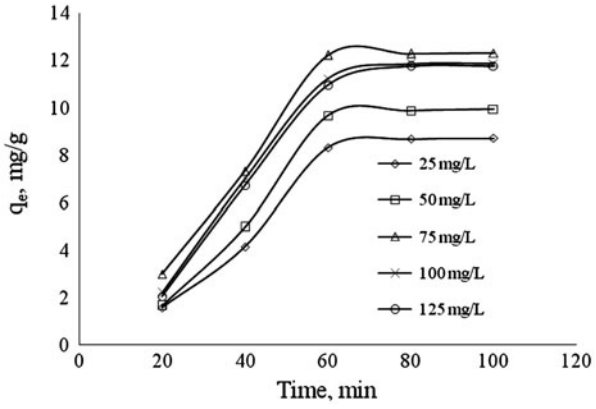


Fig. 4. Effect of initial dye concentration on equilibrium uptake of aniline blue by *A. fumigatus* biomass at 30°C, 1 g/L of sorbent, 160 rpm and pH 10.

on biosorption of aniline blue is shown in Fig. 4. It is observed from Fig. 4 that the dye uptake capacity was increased with increase in contact time and finally attained saturation at 60 min, called equilibrium time for all dye concentration studied. Similar biosorption pattern was previously reported for the removal of Remazol Black B by biomass of *Rhizopus arrhizus* [17]. The dye uptake capacity was also influenced remarkably by the initial dye concentration. The equilibrium dye uptake capacity was found to be more for 75 mg/L of initial dye concentration, when compared with all other initial dye concentrations studied. The maximum dye uptake capacity obtained was 12.23 mg/g. An increase in the initial dye concentration increases the number of collisions between dye ions and the sorbent, which enhances the sorption process. In addition, a higher initial dye concentration provides an important driving force to overcome all mass transfer resistances between the aqueous and solid phases, thus increasing the dye uptake capacity [18].

### 3.4. Biosorption isotherms of aniline blue by A. fumigatus

Analysis of the experimental data is important to develop an equation which accurately represents the results, so that they can be used for the design of efficient adsorption process. Langmuir theory assumes homogeneous type of biosorption within the biosorbent, meaning that once a dye molecule occupies a site, no further biosorption can take place at that site [19]. Langmuir isotherm is given by Eq. (1). The essential features of Langmuir isotherm can be expressed in terms of dimensionless constant in Eq. (2).

$$\frac{C_e}{q_e} = \frac{1}{bQ_0} + \frac{1}{Q_0}C_e \tag{1}$$

$$R_L = \frac{1}{1 + bC_0} \tag{2}$$

If the $R_L$ value lies between 0 and 1, the biosorption process is considered to be favorable. The constants $Q_0$ and $b$ can be determined from the linear plots of $C_e/q_e$ vs. $C_e$ at different temperature.

Freundlich isotherm is an empirical model that can be used for nonideal heterogeneous sorption expressed in Eq. (3) [20]. Plots of $q_e$ against log $C_e$ at different temperatures is used to determine Freundlich's constants $n$ and $k_f$.

$$\ln q_e = \ln k_f + \frac{1}{n}\ln C_e \tag{3}$$

Temkin isotherm emphasizes the effect of indirect adsorbent–adsorbate interactions on adsorption isotherm [21]. Experimental equilibrium data have been analyzed by Temkin isotherm in Eq. (4) and the constants were determined from the plots of $q_e$ vs. ln ($C_e$) at different temperatures. The constant $B$, related to the heat of adsorption increases with the increase in temperature proving endothermic adsorption.

$$q_e = B\ln A + B\ln C_e \tag{4}$$

The shape of the Scatchard isotherm plot from Eq. (5) relates the type of interaction between the biosorbate and the biosorbent. The negative slope of the linear Scatchard plot indicates the independent interactions between the biosorbate and the binding sites [22].

$$\frac{q_e}{C_e} = q_m K_L - q_e K_L \tag{5}$$

The $R_L$ of Langmuir isotherm was obtained from 0.0063 to 0.0193 for biosorption of aniline blue dye (Table 1), indicating that the biosorption process is favorable. The constants $Q_0$ and $b$ can be determined from the linear plots of $C_e/q_e$ vs $C_e$ at different temperatures. The numerical values of Freundlich's constant $n$ were between 1.05 and 1.50 for aniline blue dye, which is a measure of the deviation from linearity of the biosorption. The values are greater than unity, indicating that reactive dyes are favorably adsorbed by *A. fumigatus* at all the temperatures studied. The constant $B$ (Temkin isotherm) is related to the heat of adsorption, it increases in accordance to the temperature proving that the biosorption is endothermic in nature. The low value of constant $n$ and $k_f$ for Freundlich at 30°C shows that the biosorption isotherm of aniline blue dye on *A. fumigatus* biomass is best described by Freundlich isotherm, therefore

Table 1
Isotherm constants for the biosorption of aniline blue dye onto *A. fumigatus* biomass at various temperature

| Isotherms | 20℃ | 30℃ | 40℃ | 50℃ |
|---|---|---|---|---|
| *Langmuir* | | | | |
| $Q_0$ (mg/g) | 0.583 | 0.687 | 0.769 | 0.896 |
| $b$ (L/mg) | 1.02 | 1.89 | 3.13 | 1.67 |
| $R^2$ | 0.75 | 0.91 | 0.83 | 0.83 |
| $R_L$ | 0.0191 | 0.0104 | 0.0063 | 0.0117 |
| *Freundlich* | | | | |
| $n$ | 1.20 | 1.07 | 1.38 | 1.11 |
| $k_f$ (L/mg) | 2.48 | 1.44 | 4.09 | 1.77 |
| $R^2$ | 0.97 | 0.98 | 0.98 | 0.95 |
| *Temkin* | | | | |
| $A$ (L/mg) | 1.11 | 1.28 | 1.11 | 1.03 |
| $B$ | 8.06 | 3.40 | 7.01 | 19.93 |
| $R^2$ | 0.95 | 0.93 | 0.97 | 0.80 |
| *Scatchard* | | | | |
| $q_m$ (mg/g) | 3.381 | 15.7 | 25.24 | 11.41 |
| $K_L$ (l/mg) | 0.91 | 0.86 | 0.78 | 0.60 |
| $R^2$ | 0.95 | 0.92 | 0.97 | 0.78 |

indicating the nonideal heterogeneous biosorption of aniline blue dye by *A. fumigatus* biomass.

### 3.5. Kinetic modeling of biosorption of aniline blue by A. fumigatus

It is important to predict the time at which dye is removed from aqueous solution in order to design appropriate sorption treatment process. The kinetics of adsorption describes the solute uptake rate, which in turn governs the residence time of the adsorption. The biosorption mechanism and rate controlling steps have been studied using the pseudo-first-order, pseudo-second-order kinetic and external diffusion models. Pseudo-first-order kinetic law is given by Lagergren and it is given as Eq. (6) [23].

$$\ln(q_e - q_t) = \ln q_e - K_1' t \tag{6}$$

The slope and intercept of $\ln(q_e - q_t)$ vs. time yield the values of $K_1'$, and the predicted $q_e$ and experimental $q_e$ values are obtained for the biosorption of aniline blue dye solution on *A. fumigatus* biomass. In pseudo-second-order model, it is assumed that the sorption capacity is proportional to the number of active sites occupied on the sorbent, hence the kinetic rate law can be written as Eq. (7) [24].

$$\frac{t}{q_t} = \frac{1}{h} + t\left(\frac{1}{q_e}\right) \tag{7}$$

where $h = K_2'/q_e^2$. The values $(1/h)$ and $q_e$ can be determined from the intercept and slope, respectively, from a linear plot of $t/q_t$ and time.

The external diffusion model is given by Eq. (8) [25].

$$\ln\frac{C_t}{C_0} = -k_{\text{ext}} t \tag{8}$$

The plot of $\ln(C_t)$ vs. time gives a linear relationship for external diffusion model. The $R^2$ values of the models are given in Table 2. It was found that the pseudo-second-order kinetic model fits the experimental data very well for the biosorption of aniline blue dye onto *A. fumigatus* biomass when compared to other models studied.

### 3.6. Thermodynamics of biosorption of aniline blue by A. fumigatus

The thermodynamic parameters are considered to be the actual indicators for the industrial application of a biosorption process. Thermodynamic properties are used to determine the feasibility of the biosorption mechanism and the heat of adsorption [26]. The change in Gibbs free energy ($\Delta G$), enthalpy ($\Delta H$), and entropy ($\Delta S$) of adsorption were calculated from the variation of the thermodynamic equilibrium constant $K$ with temperature using Eqs. (9)–(11) [27].

$$\Delta G = -RT\ln K \tag{9}$$

$$\ln\frac{K_2}{K_1} = -\frac{\Delta H}{R}\left(\frac{1}{T_2} - \frac{1}{T_1}\right) \tag{10}$$

$$\Delta G = \Delta H - T\Delta S \tag{11}$$

Table 2
Kinetic constants for the biosorption equilibrium of aniline blue dye by *A. fumigates*

| Kinetic model | Aniline Blue |
|---|---|
| *Pseudo-first-order* | |
| $K_1'$ ($\text{min}^{-1}$) | 0.018 |
| $R^2$ | 0.276 |
| *Pseudo-second-order* | |
| $K_2'$ (g/(mg min)) | 0.306 |
| $R^2$ | 0.967 |
| $h$ (mg/(g min) | 6.89 |
| $q_{\text{cal}}$ (mg/g) | 8.62 |
| *External diffusion model* | |
| $k_{\text{ext}}$ (1/min) | 0.001 |
| $R^2$ | 0.067 |

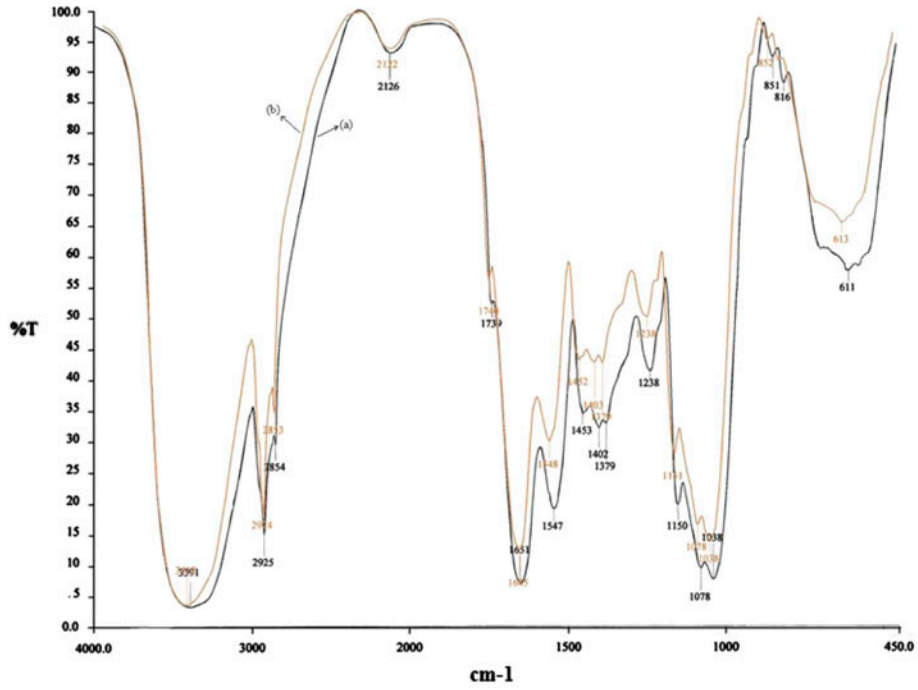


Fig. 5. FT-IR spectra of *A. fumigatus* biosorbent (a) before adsorption (b) after adsorption.

Isotherm data on biosorption process of aniline blue dye onto *A. fumigatus* biomass at different temperatures were used to find the thermodynamic parameters. The $\Delta G$ was decreased from −252.68 to −335.90 kJ/mol when the temperature of biosorption increased from 40 to 50°C. The negative value of $\Delta G$ indicates that the biosorption process was favorable and spontaneous in nature. The adsorption capacity was found to increase with increase in temperature from 40 to 50°C. The $\Delta H$ and $\Delta S$ of the biosorption of aniline blue were found to be 3.08 kJ/mol and 8.32 J/mol K, respectively. The positive value of $\Delta H$ confirmed the endothermic nature of the biosorption process and the positive value of $\Delta S$ showed the positive affinity of the dye towards the adsorbent. Thermodynamic analysis clearly demonstrated that biosorption of reactive dye onto biosorbent prepared from biomass of *A. fumigatus* biomass was more favorable.

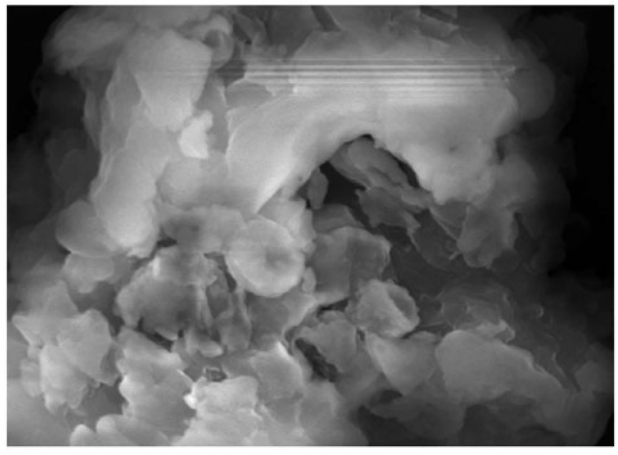

Fig. 6. SEM image of *A. fumigatus* biosorbent before adsorption.

### 3.7. FTIR and SEM analysis of biosorbent

The FTIR analysis of biosorbent was carried out to confirm the existence of functional groups in biomass of *A. fumigatus* before and after adsorption. The slight reduction of stretching vibration of the adsorption bands without change in peak positions is observed in both spectra, taken before and after adsorption, are shown in Fig. 5. This clearly indicates that the adsorption of aniline blue on the biomass of *A. fumigatus* may be due to the physical forces and not by chemical interactions. SEM analysis is one of the most widely used methods to investigate the surface morphology and physical properties of the adsorbent. It is used to understand the biosorption mechanism [6]. The SEM micrographs of biosorbent before and after adsorption are shown in Figs. 6 and 7, respectively. The surface

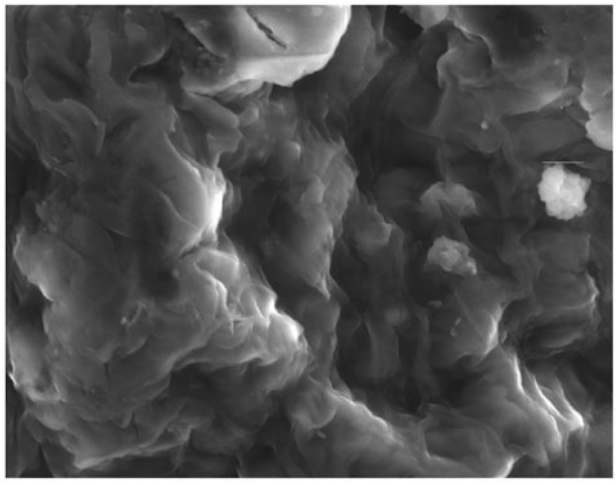

Fig. 7. SEM image of *A. fumigatus* biosorbent after adsorption.

was found to be more rough and porous in the SEM micrograph of biosorbent before adsorption than after adsorption. These micropores could be considered as a great factor in increasing the adsorption capacity of the biosorbent.

## 4. Conclusions

The removal of aniline blue by *A. fumigatus* dead biomass was systematically investigated and optimized under different conditions. Adsorption equilibrium was correlated well with Freundlich isotherm in comparison with other isotherms. The pseudo-second-order kinetic model was found to fit very well with the dynamic behavior for the adsorption of aniline blue on *A. fumigatus*. The thermodynamic quantities such as $\Delta H$, $\Delta S$, and $\Delta G$ value show that *A. fumigatus* has considerable potential as an adsorbent for the removal of aniline blue. The results also indicated that the adsorption was spontaneous and physical in nature. Thermodynamic analysis clearly demonstrates that biosorption of reactive dye aniline blue onto *A. fumigatus* biomass was more favorable.

### Symbols

$Q_{0,b}$ — Langmuir isotherm constant (L/mg)
$A$ — Temkin constant (L/g)
$B$ — Temkin constant (RT/b)
$C_0$ — initial liquid-phase concentration (mg/L)
$C_e$ — equilibrium liquid-phase concentration (mg/L)
$\Delta H$ — heat of adsorption (J/mol)
$K_{fo}$ — Freundlich isotherm constant (L/mg)
$K_L$ — Scatchard isotherm constant (L/mg)
$N$ — Freundlich isotherm constant
$q_e$ — equilibrium uptake capacity (mg/g)
$K_1'$ — pseudo-first-order rate constant ($min^{-1}$)
$K_2'$ — pseudo-second-order rate constant (g/(mg min))
$k_{ext}$ — external diffusion constant ($min^{-1}$)
$\Delta G$ — Gibbs free energy change (J/mol)
$b$ — equilibrium constant of biosorption
$\Delta S$ — entropy change (J/mol)
$R_L$ — dimensionless constant separation factor
$T$ — absolute temperature (K)

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