

Adsorption of Ni(II) by *Exiguobacterium* sp. 27 and Polyaniline Nanoparticles

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ABSTRACT: The present study was carried out to investigate three different adsorbents; dry cells of the bacterium *Exiguobacterium* sp. 27, polyaniline and a mixture of both in different ratios for removal of Ni(II) from aqueous solution. The results showed that the adsorption of Ni(II) using dry cells of *Exiguobacterium* sp. 27 was very fast and the equilibrium time was 15 min, while the equilibrium time for polyaniline was 120 min. Finally, for a mixture of dry cells of *Exiguobacterium* sp. 27 and polyaniline, the equilibrium time was 40 min. Adsorption isotherm and kinetics were studied. The reaction of *Exiguobacterium* sp. 27 when exposed to Ni(II) solution was found to be pseudo-second order reaction and the reaction obeyed the Langmuir equation. Whereas, the reaction of polyaniline with Ni(II) solution was pseudo-second order reaction and the reaction obeyed Freundlich equation. The experimental studies showed that mixture of *Exiguobacterium* sp. 27 and PANI had better metal uptake than both *Exiguobacterium* sp. 27 and PANI individually. This work provided an evidence for the possible application of nanoparticles of bacterial cells and/or polyaniline for Ni(II) biosorption from solution for a safe and clean environment.

Key words: Biosorption, Polyaniline, *Exiguobacterium* sp. 27, Nanoparticles, Freundlich and Langmuir isotherm

INTRODUCTION

Water is one of the great important environmental resources affecting human being and other living things. Water systems are frequently polluted owing to direct and indirect human activities. Water pollution by heavy metals remains as an important environmental issue because of its negative impact on health and ecosystem (Nriagu *et al.*, 1988). Nickel ions are non-biodegradable toxic and may cause dermatitis and allergic sensitization (Al-Rub *et al.*, 2002, Al-Rub *et al.*, 2003). Adsorption process is the most frequently applied method in industries for heavy metal removal. A lot of studies on this process have been carried out (Ossman *et al.*, 2013). Many studies have recently devoted the usage of different adsorbent materials in processes involving the removal of nickel ions from aqueous environments such as seaweeds (Vijayaraghavan *et al.*, 2005), crab shell (Vijayaraghavan *et al.*, 2004), dried aerobic activated sludge (Aksu *et al.*, 2000), loofa sponge-immobilized biomass of *Chlorella sorokiniana*

(Akhtar *et al.*, 2004), activated carbon prepared from almond husk (Hasar, 2003), spent animal bones (Al-Asheh *et al.*, 1999) and waste of factory tea (Malkoc *et al.*, 2006) but low adsorption capacities or efficiencies limit their applications. Therefore, investigating new adsorbents with higher adsorption capacities and efficiencies has been the aim of many researchers. Conductive electro active polymers such as polypyrrole and polyaniline can be used for the removal of heavy metals from water and waste waters (Eisazadeh, 2007a; Eisazadeh, 2007b; Eisazadeh *et al.*, 2008). Polyaniline (PANI) is a poly aromatic amine that can be easily synthesized chemically from Brønsted acidic aqueous solutions (Li *et al.* 2008). It is one of the most potentially useful conducting polymers and has received considerable attention in recent years because of its environmental stability, low cost of raw materials, and easy synthesis (Ansari *et al.*, 2006; Hung *et al.*, 1986; Kang *et al.*, 1998; Bhadra *et al.*, 2009). Recently, applications of PANI conducting

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polymers for strong adsorption of heavy metal ions from aqueous solutions have been reported (Ansari et al., 2006; Ansari, 2006; Ansari et al., 2008; Huang et al., 2006; Li et al., 2009a; Li et al., 2009b; Stejskal et al., 2009). Another attracting method for removing heavy metals is biosorption. Biosorption has recently attracted growing interest. It has the advantage of achieving high purity of the treated wastewater involving inexpensive sorbents. Studies on various types of non-living biomass such as algae, fungi, bacteria, yeast, nut hulls, and wood sawdust in addition to some other lignocellulosic wastes, have shown that such biomaterials may be used for removal of toxic metal ions from wastewater (Figueira et al., 2000; Schiewer et al., 2000; Nuhoglua et al., 2002; Lodeiro et al., 2005; Wang et al., 2005; Allaboun et al., 2008; Aksu et al., 2006; Grimm et al., 2008). In this paper, three different nanosorbents namely; dry cells of *Exiguobacterium* sp. 27 (representing the biological sorbent), polyaniline (representing the chemical sorbents) and finally a mixture of both in different ratios were investigated for the removal of nickel ions from aqueous solution.

MATERIALS & METHODS

The bacterial strain used in this study was isolated from Lake Mariout (Egypt) and identified as *Exiguobacterium* sp. 27 using 16S rDNA. It grew well in media containing 1 mM Ni. All bacteriological media components, chemicals, solvents and reagents, were of analytical grade and obtained from commercial suppliers. Metal salts used were Nickel chloride hexa hydrate ($\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$). Aniline hydrochloride $\text{C}_6\text{H}_8\text{ClN}$ (CDH Company) and Ammonium peroxydisulfate ($(\text{NH}_4)_2\text{S}_2\text{O}_8$

Cells mass were prepared in a batch culture. Seed culture was first prepared in a 100 ml Erlenmeyer flask containing 50 ml of LB medium, pH 9 and kept on a rotary shaker at 300 rpm and 37°C till $\text{OD}_{(600)} = 0.8$.

A batch of dry cells were prepared by inoculating 2 l stirred tank bioreactor (Biostat, B. B-3.2 Braun, biotechnologia, Germany) with a working volume 0.7 - 1.2 liters. The bioreactor is equipped with temperature, pH and agitation controllers (Fig.1). The fermenter was filled with 1 l LB – broth medium, pH adjusted to 9.0 using pH electrode, and then autoclaved for 20 min at 121°C. Under aseptic conditions, the fermenter was inoculated with 50 ml of the seed culture previously prepared, and then incubated for 12 h at 37°C with agitation at 300 rpm. One run from the fermenter produced about 4 g fresh cells. Cells were washed 2-3 times by distilled water after centrifugation, dried in an oven for 24 h at 60°C till constant weight and stored in a desiccator to remove moisture. The dried cells were blended for 3 min to become soft powder.

Aniline hydrochloride was dissolved in distilled water in a volumetric flask to 50 mL of solution. Ammonium peroxydisulfate was dissolved in water also to 50 mL of solution. Both solutions were kept for 1 h at room temperature (~18–24 °C), then mixed in a beaker, stirred, and left at rest to polymerize. Next day, the PANI precipitate was collected on a filter, washed thoroughly with 0.2 M HCl, and acetone. Polyaniline hydrochloride powder was dried in air and then in vacuum at 60 °C.

The removal of Ni(II) was carried out into three stages, the first dealt with the removal of nickel using nanoparticles of dry bacterial cells. The second dealt



Fig. 1. Bacterial growth in the stirred tank bioreactor (fermenter)

with the removal of nickel using nanoparticles of polyaniline powder, while the third one dealt with the removal of nickel using a mixture of polyaniline and bacterial cells in different ratios. In all stages, the effect of operating conditions namely; contact time, pH, nickel concentration and dose of adsorbent on the nickel uptake have been studied. Residual nickel concentration was measured using Inductively Coupled Plasma (ICP)-Atomic adsorption (Prody model, Leemans Company, USA).

Data were expressed as removal percentage of the initial concentration or specific biosorption q (mg/g). Ni(II) biosorption removal (%) was determined by equation (1).

$$\% \text{ Removal} = \frac{(C_i - C_f)}{C_i} * 100 \quad (1)$$

Where C_i is the initial Ni(II) concentration (ppm), C_f is the final Ni(II) concentration (ppm), while specific biosorption q (mg/g) was calculated by equation (2):

$$q = \frac{(C_i - C_f)}{Wt} * V \quad (2)$$

Where C_i is the initial Ni(II) concentration (ppm), C_f is the final Ni(II) concentration (ppm), Wt is the dose of sorbents (g) and V is the volume of solution (ml)

Measurement of nanoparticle size depended on measuring the rate of fluctuations in laser light intensity scattered by particles which diffused through a fluid (Michael *et al.*, 2008). The powder sample was put in water and followed by the ultrasonic for 15 min until a good dispersion, then the sample was measured by submicron particle size analyzer (Beckman Coulter, Miami, U.S.A.).

In order to know the structure of the adsorbents, scanning electron microscopy (SEM) was employed to visualize sample morphology using a JEOL JSM-5300 Scanning Electron Microscope operated between 15 and 20 KeV.

RESULTS & DISCUSSION

In order to understand the morphology of the biosorbent, SEM analyses of *Exiguobacterium sp. 27* dry cells, polyaniline and the mixture of both were carried out before and after adsorption. Nickel ions found to play a major role on the surface morphology of adsorbents. The comparison of SEM micrographs showed that the particle had undergone remarkable physical disintegration after adsorption in all the three adsorbents (Pandiyani *et al.*, 2011). Fig.2 shows cells

of *Exiguobacterium sp. 27* before (a) and after (b) exposure to Ni ions solution. The unexposed cells were smooth and had certain dimensions but after exposure to Ni solution, they became distorted and swollen with meanders on the surface. This may be due to precipitation of Nickel ions around the cells and formation of links with bacterial cell wall functional groups.

Fig.3 shows Polyaniline before (a) and after (b) exposure to Nickel ions solution. Particles of polyaniline were organized, fine and well shaped before being exposed to Nickel ion solution, but after exposure, they became like anemones and corals reefs. A mixture of polyaniline and *Exiguobacterium sp. 27* before (a) and after (b) exposure to nickel ions solution was studied (Fig.4). The mixture before exposure had wide pores appearing in the first picture as dark regions (Fig.4a), while in the second picture this dark region diminished (Fig.4b), suggesting that pores of mixture were filled with Ni(II). Moreover, *Exiguobacterium sp. 27* dry cells in mixture became more distorted.

In order to establish equilibration time for maximum uptake and to know the kinetics of adsorption process, the adsorption of Ni(II) using two different nano-adsorbents (polyaniline (75.7 ± 1 nm) and *Exiguobacterium sp. 27* bacterial dry mass (69.9 ± 1 nm) were studied as a function of contact time and results are shown in Fig.5. It is clear that the removal was rapid in the initial stage and significant removal of Ni (II) ions occurred after 15 min for *Exiguobacterium sp. 27* and 120 min for polyaniline. The time observed in this study was shorter than that reported by other biosorbents. For example, the equilibrium time required for Ni removal by *B. subtilis*, *P. aeruginosa* and *E. cloacae* was 3.5 h (Pandiyani *et al.*, 2011). Which was considered sufficient for the removal of ions. This might be attributed to the large surface area of the adsorbents; ($435.39 \text{ m}^2/\text{g}$ for polyaniline and $455.338 \text{ m}^2/\text{g}$ for bacterial dry mass). As the surface accumulation site became exhausted, the rate of uptake is controlled by the rate of transport from the exterior to the interior sites of the biomass particles.

The solution pH usually plays a major role in adsorption, and seems to affect the solution chemistry and the activity of the functional groups of the sorbents. For metals, the pH strongly influences the speciation and biosorption availability of the metal ions (Gabr *et al.*, 2008). In Fig.6, the effect of pH on the adsorption of Ni(II) was tested in the range of 3 – 7 because at higher pHs, the solubility of metal complexes decreases sufficiently allowing precipitation, which may complicate the sorption process. The activity of binding sites can also be altered by adjustment of the pH. For instance, during the adsorption of metal ions,

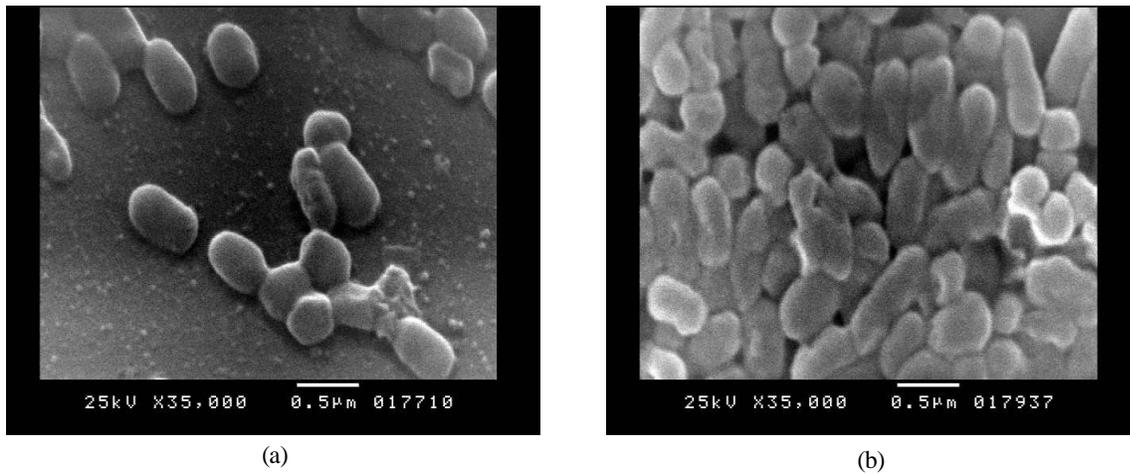


Fig.2.SEM micrograph of *Exiguobacterium* sp. 27 cells before (a) and after (b) exposure to Nickel ions

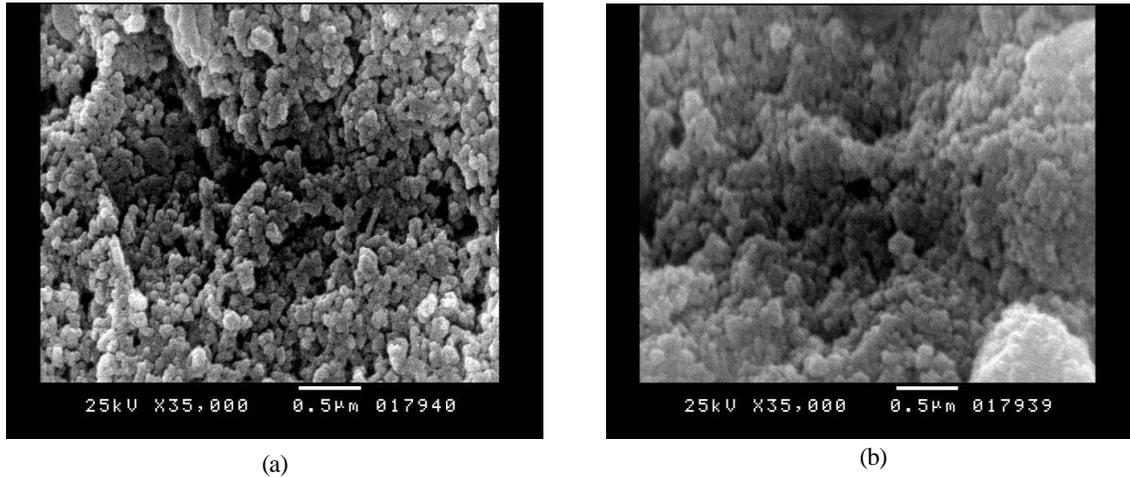


Fig. 3.SEM micrograph of Polyaniline before (a) and after (b) exposure to Nickel ions

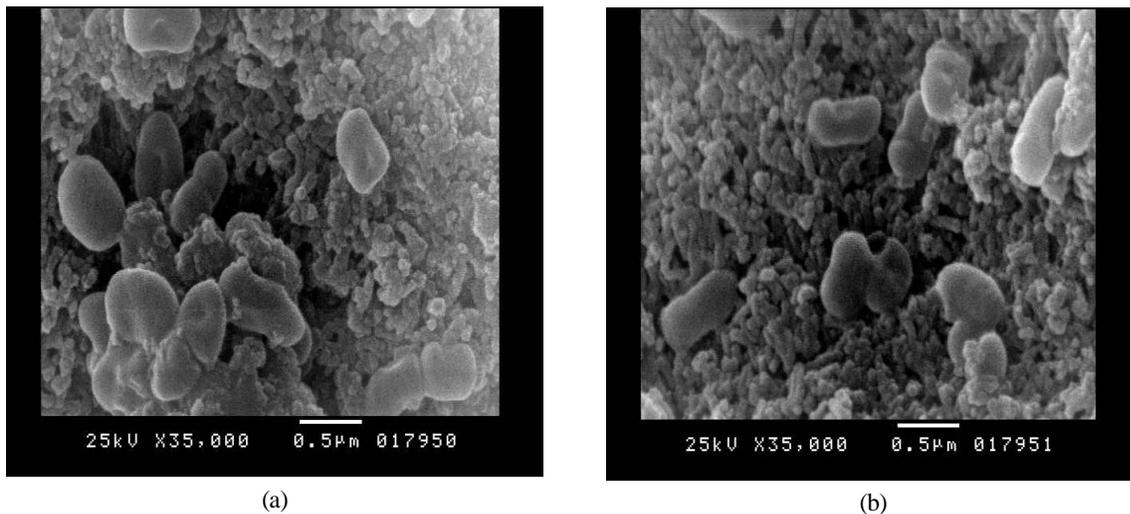


Fig.4.SEM micrograph of mixture of Polyaniline and *Exiguobacterium* sp. 27 before (a) and after (b) exposure to Nickel ions

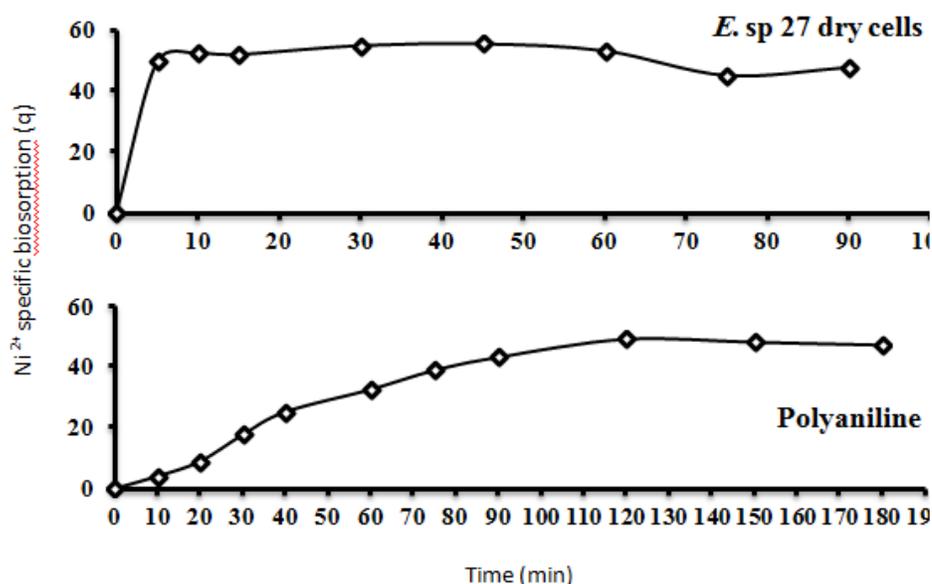


Fig. 5. Effect of contact time on the uptake efficiency, q (mg/g) of Ni(II) ions.

Initial concentration of Ni(II): 100 ppm, Agitation speed: 100 rpm, weight of adsorbent: 1 g/l and initial pH 7.0 for *E. sp 27* dry cells while pH 3.0 for Polyaniline

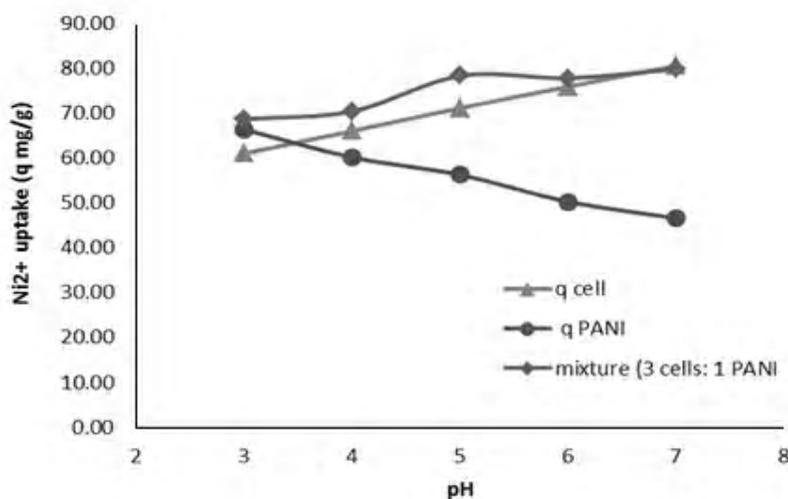


Fig. 6. Effect of initial pH on the uptake efficiency (q mg/g) of Ni(II) ions. Initial concentration of Ni(II) 100 ppm, Agitation speed: 100 rpm, weight of adsorbent 1 g/l for 40 min

pHs 3 to 7 have been found favourable, due to the negatively charged carboxyl groups ($pK_a=3-5$), which are responsible for binding metal cations via ion exchange mechanism (Vijayaraghavan *et al.*, 2008). In general, metal uptake by bacterial biomass increases with increasing pH until it reaches a maximum after which the metal uptake decreases. The optimal pH value for Ni(II) adsorption by *Exiguobacterium sp. 27* was 7.0. These results suggest that the adsorption of metals on the biomass surface is controlled by ionic attraction. At low pH values, the inactivated cell surface becomes more positively charged, leading to reducing the

attraction between metal ions and functional groups at the cell wall. In contrast, when the pH increased, the cell surface became more negatively charged and the process of retention was favoured (Gabr *et al.*, 2008). In case of polyaniline, the maximum Ni(II) sorption achieved at pH 3 and this is due to hydrogen ion activity. Polyaniline has a reactive NH group in the polymer chain flanked on either side by a phenylene ring, imparting a very high chemical flexibility. It undergoes protonation and deprotonation in addition to adsorption through nitrogen, which, having alone a pair of electrons, is responsible for the technologically

interesting chemistry and physics. Protonation of polyaniline not only involves the ingress of protons, but is also accompanied by ingress of anions, to maintain charge neutrality. This suggests that the behavior of polyaniline depends on the pH (Eisazadeh, 2007). At very low pHs, H⁺ ions compete with Ni cation for the exchange sites in the system. The heavy metal cations are completely released under circumstances of extreme acidic conditions. At pHs 3 – 7, there are three species present in solution, Ni(II), NiOH⁺ and Ni(OH)₂. These species are adsorbed and electrostatically interaction at the surface of polyaniline. As the pH decreases, the surface of the polyaniline exhibits an increasing positive characteristic. H⁺ ions present at a higher concentration in the reaction mixture and compete with Ni(II) ions for the biosorption sites resulting in the reduced uptake of Ni ions. At higher pH values, precipitation of Ni(OH)₂ occurred and both sorption and precipitation would be the effective mechanisms to remove the nickel ions in aqueous solution. At around pH 3, nickel cations, mainly Ni(II), would be expected to interact more strongly with the negatively charged binding site in the sorbent. So it is concluded that the decrease concentration of H⁺ ions in solution develops a negatively charged polyaniline surface, which can interact with the positively charged metal ions. The polyaniline surface became less positive due to the decrease of proton competitive adsorption and the deprotonation of PANI and therefore ionic exchange and electrostatic attraction between the metal ions and polyaniline was likely to be increased (Kong *et al.*, 2011).

To investigate the effect of initial metal ion concentration, the initial Ni(II) concentration was changed between 50 – 150 mg/l values in each experiment set. The maximum percentage of Ni(II) removal (48.5 % occurred upon using biomass, and 60 % upon using polyaniline) in the given time duration of 15 and 120 min, respectively (Fig.7). Data clearly show that by increasing the concentration, there is an increase in the specific uptake for both types of biosorbents. This might be attributed to the metal ion accumulation due to increasing electrostatic interactions, involving on site gradually moving downward affinity for metal ions (Pandiyani *et al.*, 2011). The initial solute concentration seems to have impact on adsorption, with a higher concentration resulting in a high solute uptake (Das, 2012). This is because at lower initial solute concentrations, the ratio of the initial moles of solute to the available surface area is low; subsequently, the fractional sorption becomes independent of the initial concentration. However, at higher concentrations, the sites available for sorption became fewer compared to the moles of solute present

and; hence, the removal of solute is strongly dependent upon the initial solute concentration. It is always necessary to identify the maximum saturation potential of an adsorbent, for which experiments should be conducted at the highest possible initial solute concentration.

The dosage of a biosorbent strongly influences the extent of biosorption. In the current work biosorbent dosages ranged from 0.67 to 1.6 g/l. Data in Fig. 8 indicate that lower biosorbent dosages yielded higher uptakes. Similar observation was reported (Aksu *et al.*, 2006; Vijayaraghavan *et al.*, 2006). The quantity of biosorbed solute per unit weight of biosorbent decreased with increasing biosorbent dosage, which may be due to the complex interaction of several factors. An important factor at high sorbent dosages is that the available solute is insufficient to completely cover the available exchangeable sites on the biosorbent, usually resulting in low solute uptake (Tangaromsuk *et al.*, 2002). Moreover, the interference onto binding sites due to increased biomass dosages cannot be overruled & thus results in the low specific uptake rate (Pandiyani *et al.*, 2011). In many instances, lower biosorbent dosages yield higher uptakes and lower percentage removal efficiencies (Vijayaraghavan *et al.*, 2008).

Models have an important role in technology transfer from a laboratory- to industrial-scale. Appropriate models can help in understanding process mechanisms, analyze experimental data, predict answers to operational conditions and optimize processes. In order to investigate the adsorption isotherm, the experimental data of equilibrium isotherms were interpreted using two equilibrium models; the Freundlich and the Langmuir. This modelling permits us to determine the maximal capacity of removal.

Langmuir's isotherm model suggests that the uptake occurs on homogeneous surface by monolayer sorption without interaction between adsorbed ions. The linear form of Langmuir isotherm equation is represented by the following equation

$$\frac{C_e}{q_e} = \frac{1}{Q^0 b} + \frac{1}{Q^0} C_e \quad (3)$$

Where Q^0 is the maximum metal ions uptake per unit mass of adsorbent (mg/g) related to adsorption capacity and b is Langmuir constant (L/mol) related to energy of sorption. Therefore, a plot of C_e/q_e versus C_e , gives a straight line of slope $1/Q^0$ and intercept $1/(Q^0 b)$.

Freundlich isotherm describes the adsorption equation for non ideal adsorption that involves

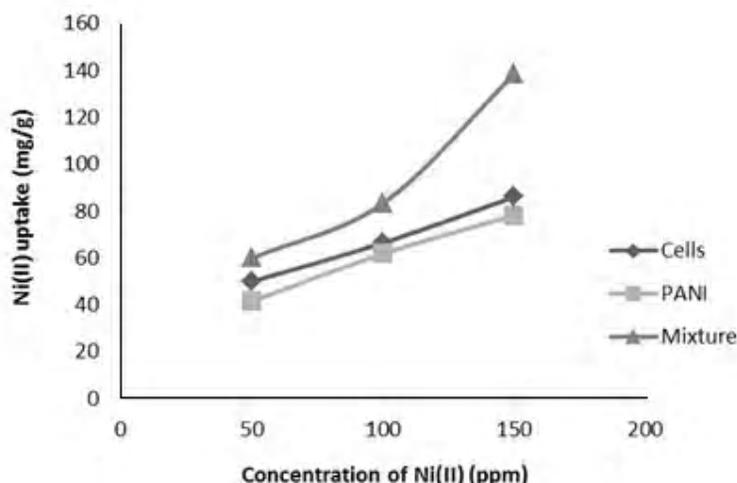


Fig.7. Effect of initial Ni(II) concentration on the uptake efficiency (q mg/g) of Ni(II) ions. pH 7.0 in case of cells, pH 3.0 in case of polyaniline, and pH 5.0 for mixture (3Cells:1 PANI) Agitation speed: 100 rpm, weight of adsorbent: 1 g/l for 40 min

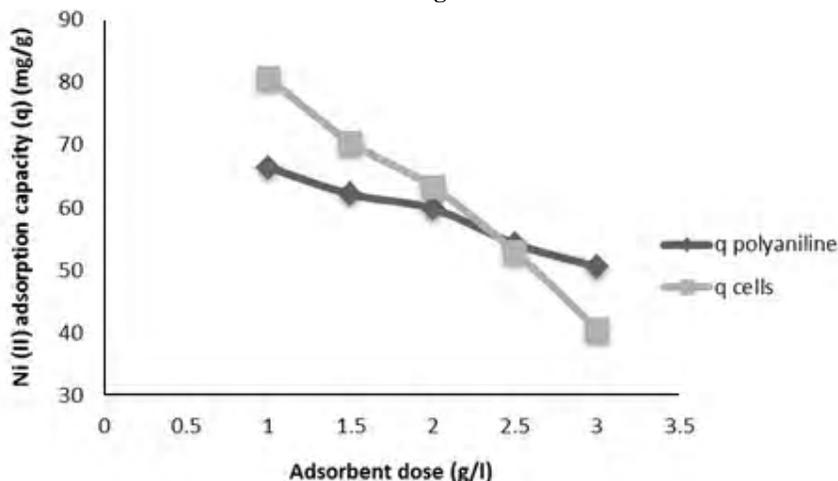


Fig.8. Effect of adsorbent dose on the uptake efficiency (q mg/g) of Ni(II) ions. pH 7.0 in case of cells and pH 3.0 in case of polyaniline, Agitation speed: 100 rpm, Ni(II) concentration 100 ppm for 40 min

heterogeneous adsorption. This empirical isotherm is expressed by the following equation:

$$q_e = K_F C_e^{1/n} \quad (4)$$

The equation is conveniently used in the linear form by taking the logarithm of both sides as

$$\log q_e = \log K_F + \frac{1}{n} \log C_e \quad (5)$$

Freundlich constants, K_F and $1/n$, are related to adsorption capacity and intensity of adsorption, respectively. The values of n and K_F can be calculated from the slope and intercept of the plot of $\log q_e$ versus $\log C_e$ derived from Eq. (5). The magnitude of the exponent $1/n$ gives an indication of the favorability of adsorption.

The applicability of the Freundlich sorption isotherm was also analyzed, using the same set of experimental data, by plotting $\log(q_e)$ versus $\log(C_e)$. The data obtained from linear Freundlich isotherm plot for the adsorption of the Ni(II) is presented in Table 1.

Table 1 reveals that the data were fitted better by Langmuir equation than by Freundlich equation in case of *Exiguobacterium* sp. 27. While in case of polyaniline the data were fitted better by Freundlich equation than by Langmuir equation.

For any practical applications, the process design, operation control and sorption kinetics are very important (Alomáa *et al.*, 2012). The sorption kinetics in a wastewater treatment is significant, as it provides

Table 1. Langmuir and Freundlich isotherms for Exiguobacterium Sp. 27 and Polyaniline (150 ppm, 100rpm, 1g of adsorbent and pH7 for cells and pH 3.0 for PANI)

<u>Isotherm model</u>	<u>Exiguobacterium sp. 27</u>	<u>PANI</u>
<u>Langmuir</u>		
Q ₀ (mg g ⁻¹)	125	112
b (L mg ⁻¹)	0.07272	0.0135
No. of parameter estimate	2	2
	4	4
R ²	0.934	0.959
<u>Freundlich</u>		
1/n	0.245	0.296
KF (mg g ⁻¹)	0.1033	1.015
No. of parameter estimate	2	2
	4	4
R ²	0.882	0.975

valuable insights into the reaction pathways and the mechanism of a sorption reaction (Vijayaraghavan *et al.*, 2008). Also, the kinetics describes the solute uptake, which in turn controls the residence time of a sorbate at the solid-solution interface (Vijayaraghavan *et al.*, 2008). The overall adsorption can occur through one or more steps. In order to investigate the mechanism of process and potential rate controlling steps, the kinetics of Ni (II) adsorption were analyzed using pseudo-first-order (Lagergren, 1898) and pseudo-second-order (Ho *et al.*, 2000) kinetic models. The conformity between experimental data and the model predicted values was expressed by the correlation coefficients (R², values close or equal to 1).

The adsorption kinetic data were described by the Lagergren pseudo-first-order model (Lagergren, 1898), which is the earliest known equation describing the adsorption rate based on the adsorption capacity. The linear form equation is generally expressed as follows:

$$\log(q_e - q_t) = \log q_e - \frac{K_1}{2.303} t$$

In order to obtain the rate constants, the values of $\log(q_e - q_t)$ were linearly correlated with t by plot of $\log(q_e - q_t)$ versus t to give a linear relationship from which K_1 and predicted q_e can be determined from the slope and intercept of the plot, respectively.

The variation in rate should be proportional to the first power of concentration for strict surface adsorption. However, the relationship between initial solute concentration and rate of adsorption will not be linear when pore diffusion limits the adsorption process.

The adsorption kinetic may be described by the pseudo-second order model. The linear equation is generally given as follows:

$$\frac{t}{q_t} = \frac{1}{K_2} \frac{1}{q_e^2} + \frac{1}{q_e} t \quad (7)$$

If the second-order kinetics is applicable, then the plot of t/q_t versus t should show a linear relationship. Values of K_2 and equilibrium adsorption capacity q_e were calculated from the intercept and slope of the plots of t/q_t versus t . The linear plots of t/q_t versus t show good agreement between experimental and calculated q_e values (Table 2). The correlation coefficients for the pseudo second-order kinetic model are greater than 0.97, which led to believe that the pseudo second order kinetic model provided good correlation for the adsorption of Ni(II) onto both of *Exiguobacterium* sp. 27 and Polyaniline nanopowder.

Table 2. Kinetic parameters for the adsorption of Ni(II) onto both of Exiguobacterium sp. 27 and Polyaniline nanopowder

Kinetics models	Exiguobacterium sp. 27	PANI
<u>Pseudo first order kinetics</u>		
K ₁ (min)	0.0483	0.0105
q ₁ (mg/g)	19.890	10.461
R ₁ ²	0.8304	0.8168
<u>Pseudo second order kinetics</u>		
K ₂ (g/mg min)	0.0026	0.0009
q ₂ (mg/g)	55.619	42.523
R ₂ ²	0.989	0.9789

An experimental design (Table 3) was established to study the effect of combining both nanoparticles of *E.sp.27* and polyaniline on nickel specific uptake. The two sorbents were mixed in different ratios 2:1, 1: 1 and 1: 2 to reach the optimal biosorbent dose (0.67 g/l). Experiment was performed in 50, 100 and 150 ppm metal ion concentration for 40, 80 and 120 min., pH was kept at 3, 5 and 7. The results reveal that the maximum Ni(II) specific biosorption(138.34 mg/g) was detected using 0.67g/l adsorbent in the ratio of 2: 1 polyaniline: *Exiguobacterium sp.27* with pH 5, Ni(II) concentration 150 mg/l and retention time 40 min. Here the equilibrium was reached quickly for Ni(II) specific biosorption indicating that the active sites of the mixture were well exposed. The polyaniline chains contained amine and secondary amino groups, which acted as linkages between the benzene rings and the Ni(II). When polyaniline was used as an adsorbent, only chelation existed between Ni(II) and polyaniline. However, three-removal mechanisms chelation, ion exchange and electrostatic attraction existed simultaneously when polyaniline/ *Exiguobacterium sp. 27* mixture was used as an adsorbent. As a result, the adsorption capacity of polyaniline/*Exiguobacterium sp. 27* mixture was

larger than that of polyaniline. Hence, the adsorption capacity for Ni(II) could be improved a lot when the polyaniline/*Exiguobacterium sp. 27* was used as the adsorbent (Kong *et al.*, 2011).

An analysis of variance (ANOVA) was used to perform a systematic analysis of the relative importance of each factor. All statistical analyses, including regression analysis and ANOVA, were carried out using STATISTICA (Statsoft). The statistical analysis of experimental design included a discussion of the statistical weight of each factor in the mixture process of adsorbents and a variance analysis. Calculations were performed according with the basic concepts of the Taguchi method (Guijarro *et al.*, 2011).

From previous statistical analysis, the model equation can be concluded as the following:

$$q = 0.658 * \text{Concentration} - 2.0285 * \text{pH} - 0.107 * \text{Time} + 6.616 * \text{Ratio} \quad (8)$$

According to the model equation, the most effective factor in Ni (II) ion uptake is the mixture adsorbents ratio while the lowest effect factor is the time(Table 3).

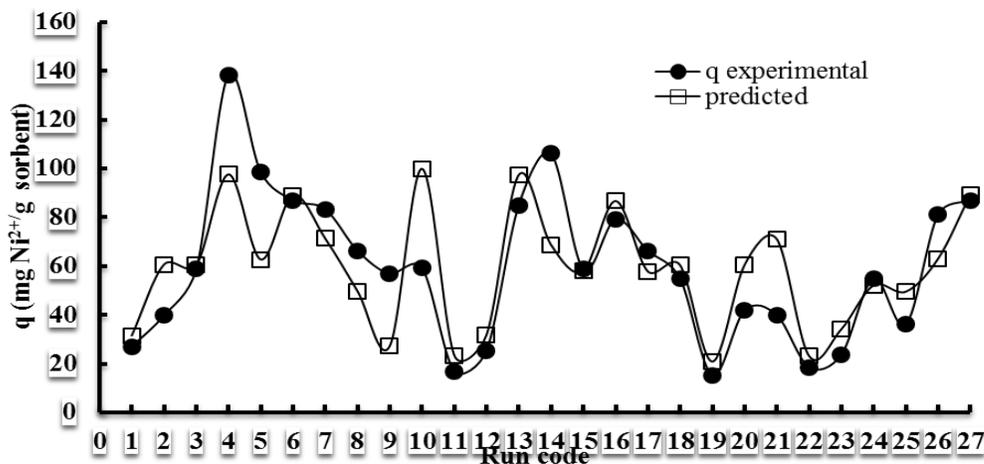


Fig. 9. Ni(II) specific sorption (q) resulted from Experiment and another predicted by model

CONCLUSION

The present study was carried out to evaluate the efficiency of a local bacterial isolate to biosorb nickel ions from aqueous solution. On three stages, to reach optimum Ni(II) biosorption, three nanosorbents were investigated; dry cells of *Exiguobacterium sp. 27* (representing the biological sorbent), polyaniline (representing the chemical sorbents) and finally a mixture of both in different ratios. For dry cells of *Exiguobacterium sp. 27*, the equilibrium time was 15 min, whereas it was 120 min for polyaniline. Finally, in the mixture of dry cells of *Exiguobacterium sp. 27* and

polyaniline, the equilibrium time was 40 min. Adsorption isotherm and kinetic studies were investigated for Ni(II) specific sorption. The reaction of *Exiguobacterium sp. 27* when exposed to Ni(II) solution was pseudo-second order reaction and the reaction obeyed the Langmuir equation. While the reaction of polyaniline with Ni(II) solution was pseudo-second order reaction and the reaction obeyed Freundlich equation. The experimental studies showed that mixture of *Exiguobacterium sp. 27* and PANI had better metal uptake than both *Exiguobacterium sp. 27* and PANI individually. Another experimental design

Table 3. Experimental design of Ni(II) uptake by a mixture of dry cells *Exiguobacterium* sp. 27 and polyaniline

Run code	pH	Ni(II) Conc. (ppm)	Time (min)	Ratio	q (mg/g)
1	3	50	80	2	26.94
2	7	100	40	2	39.78
3	5	100	80	2	58.71
4	5	150	40	2	138.34
5	5	100	120	3	98.48
6	5	150	120	2	86.96
7	5	100	40	3	83.33
8	7	100	80	1	66.3
9	3	50	120	2	56.82
10	5	150	80	3	59.29
11	7	50	80	2	16.84
12	5	50	40	2	25.3
13	3	150	80	2	84.98
14	3	100	40	2	106.1
15	5	100	40	1	58.71
16	5	150	80	1	79.1
17	3	100	80	1	66.3
18	5	100	80	2	54.92
19	5	50	80	1	15.2
20	5	100	80	2	41.67
21	3	100	80	3	39.8
22	5	50	120	2	18.5
23	5	50	80	3	23.57
24	7	100	120	2	54.92
25	5	100	120	1	35.98
26	7	100	80	3	81.02
27	7	150	80	2	86.96

Note: In ratio column1 = Nanoparticles of Polyaniline (1): *Exiguobacterium* sp. 27 dry cells (1)2 = Nanoparticles of Polyaniline (2): *Exiguobacterium* sp. 27 dry cells (1)3 = Nanoparticles of Polyaniline (1): *Exiguobacterium* sp. 27 dry cells (2)

was established to study the effect of combining both nanoparticles (*E.sp.27* and polyaniline) on nickel specific uptake. The most effective factor in Ni(II) ion uptake was the mixture adsorbents ratio, while the lowest effect factor was the time. This work provides an evidence for the possible application of nanoparticles of bacterial cells and/or polyaniline for biosorption of Ni(II) from solution for a safe and clean environment. Also provides the basic scientific data to design and apply for strategies to remediate metal-contaminated effluents in industrial plants.

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