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Efficiency of extremophilic microbial mats for removing of Pb(II), Cu(II) and Ni(II) ions from aqueous solutions

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Abstract

Two different extremophilic films were used as natural biosorbents to remove Cu(II), Ni(II) and Pb(II) from aqueous solution. Surface area, scanning electron microscope imaging and Fourier transformation infrared were used to characterize the surface of biosorbents. The results indicated high affinity of biosorbents to remove Pb(II) Cu(II) and Ni(II) with adsorption ratio ranged between 73.6% to 100% for both two biosorbent. The two biosorbents success to remove the metal ions from the aqueous mixture in the order of Pb(II) > Cu(II) > Ni(II). The maximum removal ratios of metal ions were achieved at pH = 6, 150 min contact time, 2.5 g/L biosorbent dose and 50 mg/L metal ions. The isothermal studies showed that both Langmuir and Freundlich models well expressed the adsorption process. Kinetically, the pseudo-second order reaction well express the type of reaction than pseud-first order reaction.

Keywords: Biosorption, Cyanobacterial mats, Extreme habitat, Nickel, Lead, Copper.

1. Introduction

In recent decades, the humanity has realized the need to conserve the surrounding environment and different habitants. Pollution, especially with heavy metals, affects the life of the planet either directly or indirectly. The toxic pollutants are generating during different stages in industrial processes (Landa-Acuña et al. 2020) and mining (Beltrán- Pineda and Gómez-Rodríguez, 2016). These pollutants tend to accumulate or transport into the environment, providing an alarm issues of toxic products (Rai and Tripathi 2007). Some heavy metals e.g. Cd, Pb, Hg, As, Ni and Cr etc. are found in the industrial wastes causing serious problems to aquatic environment due to their accumulation, non-degradation nature and their long persistence in the tissues (Gupta et al. 2016). Owing to exacerbation toxic metals problems in the environment, the researchers stepped up their efforts to develop an eco-friendly, low cost and clean techniques to overcome this serious situation. Bioremediation

is considered one of the most promising, important and useful bio-techniques to overcome the contaminated environments (Ali et al. 2019; Kumar 2017)

Biological treatment is carried out through two different pathways; bioaccumulation through living cells (Ali et al. 2016; Churchill et al. 1995) or the biosorption using dry died cells (Davis et al. 2003; Singh 2020). Biosorption adsorption process involves dry died cell (solid phase) and metal ions solution (liquid phase) (Farooq et al. 2010). The high affinity between the biosorbent (dry cell biomass) and the sorbate (metal ions) lead to high attraction and bound of sorbate onto the surface of biosorbent (Das et al. 2008).

Bioremediation is a biological treatment through series steps to recover the polluted environments using living organisms, e.g. plants and/or microorganisms (fungi, bacteria and algae) to remove, adsorb, and degrade different dangerous pollutants (Maeir et al. 2009). Cyanobacteria and bacteria are the most adsorbents used to bio-remediate and degrade dyes, hydrocarbons and heavy metals (Kumar and Kundu 2020) due to their high growth rate that giving high surface area which permits high adsorptive capacity for heavy metals binding (Ahad et al. 2017).

Cyanobacterial-bacterial film is a consortium of cyanobacteria and bacterial strains embedded in polysaccharide secretions. Cyanobacterial-bacterial films are widely distributed in different normal habitats (terrestrial, freshwater and marine) and found in extreme habitats (hot springs, Antarctic ponds, and hypersaline waters (Madigan et al. 2000; Nakagawa and Fukui 2002; Singh 2014). Cyanobacterial-bacterial film have several advantages in comparison with other living organisms; they have unique composition of cell wall, high binding capacity, large surface area, low nutrients need for growth, greater polysaccharides volume (Micheletti et al. 2008a; Pathak et al. 2018).

Many scientists reported excellent biosorption removal efficiency for several cyanobacterial strains. *Oscillatoria angustissima* had high removal efficiency for Zn biosorption (641 mg/g of dry weight) (Ahuja et al. 1999). A high biosorption capacity of Cu had been reported (240 mg/g, 143 mg/g and 67.93 mg/g) for *Cyanospira capsulata*, *Cyanothece capsulata* and *S. platensis*, respectively (Celekli et al. 2010; Micheletti et al. 2008b; Paperi et al. 2006). Abdel-Aty et al. (2013) reported maximum biosorption efficiency of *Anabaena sphaerica* live cells were 121.95 mg/g and 111.1 mg/g for Cd and Pb respectively. Ahad et al. (2017) reported that *Nostoc muscorum* removed 71.4 mg/g of Cd. A high removal capacity of 1.47 mmol and 0.65 mmol for Pb and Ni was achieved using *Lyngbya taylorii* (Klimmek and Stan 2001).

Both living and dead bacterial biomass effectively can be used as biosorbents for toxic metal removal from aqueous media even at lower concentrations (Mohapatra, et al.

2019; Todorova et al. 2019). Bacterial removal of heavy metals has been reported. *Arthrobacter viscosus* has been utilized for Cr (VI) removal from aqueous solution (Lameiras et al. 1982). The removal efficiency of *A. viscosus* was optimized by the application of *E. coli* biofilm supported on granulated activated carbon (Quintelas et al. 2009). Khanafari, et al. (2008) used *Bacillus circulans* to remediate aqueous solution from Pb(II) and Cr(VI). *Pseudomonas aeruginosa* biofilm over a flexible polyvinyl conduit efficiently was able to eliminate 85 % of the Cu(II) from the solution (Qureshi et al. 2001). *Escherichia coli* biofilm supported on kaolin had metals' removal ability in the order of Fe(III) > Cd(II) > Ni(II) > Cr(VI) (Quintelas et al. 2009).

Very few studies used microbial consortium of cyanobacteria and bacteria in bioremediation treatments. This work aims to determine the adsorption efficiency of two different cyanobacterial-bacterial consortia for removing Cu, Ni and Pb ions from mixture aqueous solution. Furthermore, describe the different factors affecting biosorption process e.g. pH, contact duration time, and adsorbent dosage.

2. Materials and Methods

2.1 Cyanobacterial samples

Microbial mats were collected from hot spring (biosorbent 1) and cold spring (biosorbent 2) located in Siwa Oasis in Western Desert, Egypt. The collected samples were packed in dry clean polyethylene bags. After reaching the laboratory, the samples were washed several times with deionized water. Microbial biomass was dried at 105°C in circulated-air oven (Binder ED 115) for 48 h, then ground in an electric mill (model Kz II, Wuhan, China) and passed through a 63 µm sieve.

2.2. Characterizations

Surface area of both biosorbents was determined according to Brunauer–Emmett–Teller (BET) method using a Coulter SA3100 with outgas of 15 min at 150 °C (Brunauer et al. 1938). Pore diameter, pore volume and micropore surface area were determined by the Barrett–Joyner–Halenda (BJH) method (Barrett et al. 1951). Scanning electron microscope (JEM-2100, JEOL, Tokyo Japan) at an acceleration voltage of 200 kV was used for scanning electron microscope imaging (SEM) and energy dispersive X-ray spectroscopy (EDX). The Fourier transform infrared spectra (FT-IR) were measured before and after metal adsorption using spectrum spectrometer (6700 FTIR, Nicolet, America). The samples were ground with KBr (1:100) the measurements were taken within the range of 400–4000 cm⁻¹.

2.3. Preparation of metal ions standard solutions

Stock standard solutions of copper, nickel and lead (1000 mg/L) of analar grade (Sigma Company) were used for preparation of serial dilutions as required.

2.4. Batch biosorption studies

Biosorption of Cu(II), Ni(II) and Pb(II) from mixture aqueous solution is affected by several factors such as solution pH, dose of biomass, contact time and initial metal ions concentration. pH effect was studied using different pH values (2 - 9) under 150 min contact time and 50 mg/L metal ions concentrations. Biosorbent dose effect was studied using different dry biosorbent weights (0.5 - 4 g/L) under 150 min contact time, pH 6 and 50 mg/L metal ions concentrations. Contact time effect was investigated with different time range of 15 - 180 min under pH 6, biosorbent dosage of 2.5 g/L, initial metal ions concentrations of 50 mg/L. Finally, initial metal ions concentrations effect was carried out using different metal ions concentrations (5 – 50 mg/L) with constant pH 6, biosorbent dose of 2.5 g and contact time of 150 min. Experiments were carried out in 250 ml glass flasks in triplicate; each flask contained 100 ml of metal mixture solutions with continuous stirring at 300 rpm for 150 min. Metal mixtures were centrifuged at 5000 rpm for 7 min to separate the biosorbents. The supernatants were analyzed by Perkin Elmer inductively coupled Plasma Model Optima 3000XL ICP-OES. Results are expressed as µg/L. The adsorption capacity of heavy metal ions by different microbial mat could be expressed as follows:

$$q_e = \frac{(C_i - C_f) \times V}{M}$$

$$R\% = \frac{(C_i - C_f) \times 100}{C_i}$$

where; C_i is the initial metal concentration, C_f is the final metal concentration (mg/L), M is the mass of used biosorbent (g) and V is the volume of initial metals solution (L)

3. Results and discussion

3.1. Surface area

Surface areas of biosorbents were measured to illustrate their capacity to adsorb metals (Table 1). Biosorbent 2 has higher surface area than microbial film from biosorbent 1.

Table (1): Specific surface area, radius and pore volume of biosorbents.

Samples	S_{BET} (m ² g ⁻¹)	r (nm)	V_p^{total} (cm ³ g ⁻¹)
Biosorbent 1	3.56	156	0.114
Biosorbent 2	4.54	118	0.105

3.2. Scanning electron microscope (SEM)

The morphological differences between surfaces of biosorbents before and after adsorption of Cu(II), Ni(II) and Pb(II) ions were detected using scanning electron microscope (SEM). Figure (1a & 1c) represents SEM graph of biosorbent 1 and biosorbent 2 biomass before the adsorption of metal ions, while Figure (1b-d) represents SEM graph after adsorption of metals ions. A relative differences of surface morphology of biosorbents before and after adsorption of

metals ions were clearly observed. The surfaces of biosorbents consists of several very fine particles with irregular sizes and shapes and coarse nature, which in turn made feasible to visualize small pores. After complete adsorption of metal ions, biosorbents' surfaces exhibited irregular structure, fragile walls and appearance of bright dots onto the surface which declared absorption of metals ions (Figure 1 b-d). These results are supported by findings of El-Naggar et al. (2018) for removing Pb^{+2} using *Gelidium amansii* biomass.

3.3. Electron dispersive spectroscopy (EDX)

Elemental analysis of biosorbents was carried out using EDX spectra which is useful for the detection of chemical characterization and elemental construction of biosorbents (Dmytryk et al. 2014). EDX results before and after adsorption of metals ions are shown in Figure (2 a-d). Before adsorption; three peaks belong to O, Ca and C were observed in biosorbent 1 biomass with abundance ratios of 50.55 %, 33.8 % and 12.92%, respectively. While biosorbent 2 biomass showed slightly elemental composition difference where, O, Fe and Si were observed with abundance ratios of 43.98%, 23.0, 17.45 %, respectively (Figure 2 a & c). After biosorption of metal ions, peaks of Cu, Pb and Ni appeared onto the surface of biosorbent 1 with ratios of 6.61%; 3.3% and 2.95%, respectively (Figure 2 b). While biosorbent 2 had the ratios of 2.28, 1.83 % and 0.65 % for Cu, Pb and Ni, respectively (Figure 2 d). Kim et al. (1995) and Shukla et al. (2012) found that appearance of Pb and Cr peaks in EDX spectra after adsorption process of Pb and Cr from their aqueous solution.

Figure 1. SEM micrograph of biosorbent 1: a) before and b) after, and biosorbent 2: c) before and d) after adsorption of Cu, Ni and Pb ions from aqueous solution.

Figure 2 Energy-dispersion spectra (EDX) of biosorbent 1: a) before, b) after and biosorbent 2: c) before, d) after adsorption of Cu, Ni and Pb ions.

3.4. FT-IR analysis.

FT-IR spectrograms of biosorbents were measured before and after metals ions biosorption (Table 2 and Fig. 3). FT-IR detect the changes occurred onto the surface of biosorbents as a result of the interaction between the different functional groups found on the biosorbent surfaces and metals ions after adsorption. FT-IR results showed several peaks for biosorbents' functional groups before metal adsorption at 3430.7, 2922.6, 2519.5, 1800.2, 1630.5, 1422.2, 1032.7 and 874.5 cm^{-1} . After adsorption, these peaks were shifted to 3415.3, 2926.5, 2510.5, 1810.2, 1620.5, 1411.3, 1020.77 and 863.6 cm^{-1} , respectively (Table 2 & Figure 3). Peaks' shifting and changing of their intensity depend on the interaction between metals ions and the persist functional groups (Ali et al. 2019).

The appearance of broad peak at 3430 cm^{-1} is attributed to the stretching of O-H group revealed the presence of free hydroxyl groups of carboxylic acids (Iqbala et al. 2009). The peak appeared at 2922 cm^{-1} is assigned to the stretching vibration of -C-H and =C-H of lipids (Li et al. 2007). A minor peak appeared at 2510 cm^{-1} is corresponding to stretching acid O-H. Stretching vibration of the carbonyl group C=O was appeared at 1800 cm^{-1} (Axson

2012). The peak at 1630 cm^{-1} is attributed to stretching vibration of $\text{C}=\text{C}$ (Gnanasambandam and Protor 2000). The weak stretch of $-\text{COO}-$ of pectin or $\text{C}=\text{O}$ of amide was observed at 1032 cm^{-1} (Figure 3). In general, FT-IR analysis indicated that carbonyl, carboxyl, free hydroxyl, methylene and carbonate groups are responsible for metals biosorption.

Table 2. FT-IR spectrum verified the main function groups of biosorbent 1 and biosorbent 2 biomass before and after metals ions adsorption.

Biosorbent 1 biomass			Biosorbent 2 biomass		
Before adsorption	After adsorption	Attribution	Before adsorption	After adsorption	Attribution
Wave number. (Cm ⁻¹)			Wave number. (Cm ⁻¹)		
3430.74	3415.31	Stretching O-H	3694.943	3690.91	O-H stretching of Hydroxyl group
2922.59	2926.45	-C-H and =C-H stretch vibration	3619.733	3609.73	
2519.54	2510.54	stretching Acid O-H	2922.592	2925.48	-C-H and =C-H stretch vibration of methyl and methylene groups
1800.22	1810.19	C=O carbonyl group	2853.167	2856.06	
1630.52	1620.55	C=C stretched	1874.47	1880.26	C=O carbonyl group
1422.24	1411.28	epoxy C-O groups	1629.55	1637.27	C=C stretched
1032.69	1020.77	Weak stretch of -COO- of pectin, C=O amide	1379.82	1383.68	epoxy C-O groups
874.56	863.60	Alkyl Halide stretch C-Cl	1034.62	1031.73	Weak stretch of -COO- of pectin, C=O amide
712.57	712.57		916.0218	914.093	Alkyl Halide stretch C-Cl
609.40	664.36	Alkyl Halide stretch C-I	795.4926	792.6	
461.87	430.05		693.2839	692.32	Alkyl Halide stretch C-I
			512.97	532.257

Figure 3. FT-IR spectra of; a) biosorbent 1 biomass and b) biosorbent 2 biomass before (black line) and after (red line) biosorption of metals ions from aqueous solution

3.5. Batch biosorption experiments

3.5.1. Effect of pH

Biosorption results of Cu(II), Ni(II) and Pb(II) ions under effect of different pH values of the solution are shown in Fig. (4a&b). The results show a decrease of removal efficiency of both biosorbents at low pH values, especially < 4.5 , because the biosorbents surfaces is protonated by excess H^+ ions which compete with the positive Cu^{+2} , Ni^{2+} and Pb^{2+} ions for the active binding sites onto the biosorbents' surfaces. Whereas, H^+ ions are low abundant at higher pH values and active binding sites became negatively charged, thus the functional groups are free and the biosorption of positively charges metals ions is increased (Singh 2020). The maximum removal percentages of Pb(II), Cu(II) and Ni(II) reached to 100%, 93.2%, and 73.6% were achieved at pH 6 for biosorbent 1. Biosorbent 2 shows slight increase of removal percentages reached to 100%, 94.2% and 74.6 % for Pb(II), Cu(II) and Ni(II) respectively (Fig. 4b). Slightly acidic pH (5-6.5) was reported as optimum for metal removal (Table 3), pH 6.5 was optimum value for removal of both Cu(II) and Ni(II) using *Sargassum* sp as biosorbent (Barquilha et al. 2019), likewise pH 6 was optimum value for biosorption of Cd(II) and Ni(II) using *Cystoseria indica* (Khajavian et al. 2019).

3.5.2. Effect of Contact Time

Contact time plays an important role in the biosorption of metals ions onto the surface of different biosorbents, the rate of biosorption is very rapid in the beginning of the reaction as a result of availability and vacancy of all active binding sites. By increasing time, the biosorption rate decreases because decrease of active sites availability (Michalak et al. 2013).

The biosorption of Cu(II), Ni(II) and Pb(II) onto the surface of biosorbent 1 and biosorbent 2 are shown in Fig (5 A&B). The results showed that the uptake of Cu and Pb by two biosorbents was rapidly took placed reached to about 70% during first 30 min while the rate of Ni is lower (45%), the rate became slower by increasing the time till reached the maximum removal efficiency after 150 min then the rate tend to be steady (Figure 5 A&B). Furthermore, the results indicated completely remove of Pb(II) and Cu(II) onto the surface of biosorbent 1 after 150 min with q_e of 49.8 and 48.8 mg/g followed by Ni(II) with removal percentage of 68.1% ($q_e = 33.9$ mg/g). Biosorbent 2 showed lesser efficiency for removal of metals ions with ratios of 93.2% & 91.2% and 65% for Pb(II), Cu(II) and Ni(II) respectively. *Spirulina platensis* showed maximum biosorption efficiency for removing Cd after 90 min (Al-Homaidan et al. 2015), while *Synechococcus* sp. reached maximum removal percentage for Ni, Cu, Pb after 80 min (Gardea-Torresdey et al. 1998) (Table 3)

Figure 4. Effect of pH values on biosorption of Pb(II), Ni(II) and Cu(II) onto the surface of A) biosorbent 1 and b) biosorbent 2

Figure 5. Effect of contact time (min) on biosorption of Pb(II), Ni(II) and Cu(II) onto the surface of biosorbents

3.5.3. Effect of biosorbent dose

In general, the increased in microbial biomass is accompanied with increase of adsorption process of metals ions due to availability of high surface area provides more and more active binding sites (Michalak et al. 2013). Thereby, studying of biosorbents dose variation regarded as an important factor affects the biosorption process (Dixit and Singh 2013). Studying the various biosorbent doses effects (0.5 to 5 g/L) to remove Cu(II), Ni(II) and Pb(II) was carried out (Figure 6 A&B). A significant rapid increase of biosorption rates was observed with increase of biosorbents dose from 0.5 to 2.5 g/L further increase of dose have no significant increase of biosorption rate. The results exhibit maximum removal ratios of biosorbent 1 reached to 98.7 %, 82.6% and 72.9% for Pb, Cu and Ni respectively (Figure 6A). Slight decrease of biosorption efficiency of biosorbent 2 than biosorbent 1 was observed. The maximum removal ratios of biosorbent 2 reached to 95.9 %, 80.3% and 72.4% for Pb, Cu and Ni were obtained (Figure 6B).

3.5.4. Effect of Initial Metal Concentration

Biosorption capacity increased with increasing metals concentrations due to the available free active binding sites for interaction, further increasing of metals concentrations leads to occupying of active binding sites thus the rate of biosorption is halted (Michalak et al. 2013). The biosorption of Cu, Ni and Pb increased with the increase of their initial concentrations from 5 to 50 mg/L (Figure 7 A&B) further increase of their concentrations had insignificant increase of biosorption rate which means that the reaction reach to saturation limit (Ahad et al. 2017). The maximum removal ratios of biosorbents reached to 100 %, 91.0% and 74.3% for Pb, Cu and Ni, respectively, at 50 mg/L metals ions concentrations with corresponding adsorption capacity q_e of 49.8, 44.5 and 38.5 mg/g (Figure 6 A&B)

Figure 6. Effect of biosorbent dosage (g/L) on biosorption of Pb(II), Ni(II) and Cu(II)

Figure 7. Effect of initial metals concentration (mg/L) on biosorption of Pb(II), Ni(II) and Cu(II) onto the surface of A) biosorbent 1 and b) biosorbent 2

Table (3) Comparison between the parameters affecting maximum adsorption efficiency of different metal ions using different cyanobacterial organisms as biosorbents

Adsorbent	Elements	pH	Dose g/L	Time (min)	Q_{max} (mg/L)	Ref.
<i>Synechococcus sp. PCC 7942</i>	Ni, Cu, Pb	5.0	1	80	3.2, 11.3, 30.4	Gardea-Torresdey et al. 1998
<i>Oscillatoria sp.</i>	Cr	5.5	1	210	25	Shukla et al. 2012
<i>Anabaena sphaerica</i>	Cd, Pb	6.0	2	60 – 90	111 and 121	Abdel-Aty et al. 2013
<i>Sargassum sp</i>	Ni, Cu	6.5	1	90 – 120	0.863 – 1.109 mmol	Barquilha et al. 2019
Cyanobacterial mats	Zn, Fe, Cd	6	2.5 g/L	150	24.8, 24.4 and 15.8	Ali et al. 2019b
Cyanobacterial mats	Pb, Cu, Ni	5.5 – 6	0.5 - 1 g/L	150	49.2, 48.2 and 28.5	Recent data

3.6. Adsorption equilibrium isotherms

3.6.1. Langmuir isotherm model

Langmuir models postulate that the adsorbent has finite numbers of energetically uniform sites and the adsorbate form a homogenous monolayer onto the adsorbent surface. Langmuir model is expressed by the equation:

$$\frac{1}{q_e} = \frac{1}{q_{max}} + \frac{1}{bq_{max}} \cdot \frac{1}{c_e}$$

where: $q_{\max}$ (mg/g) the maximum adsorption capacity, q_e is metals ions concentrations adsorbed at equilibrium (mg/g), b (L/mg) Langmuir constant. Langmuir separation constant (R_L) was calculated as follow:

$$R_L = \frac{1}{1 + bC_i}$$

Where C_i is initial metal ions concentration (mg/L); b is the Langmuir constant. Separation factor (R_L) value determine Langmuir isotherm type; if $R_L = 0$ (irreversible), $R_L = 1$ (linear), $R_L > 1$ (unfavorable), or $0 < R_L < 1$ (favorable) (McKay et al. 1982).

3.6.2. Freundlich isotherm model

Freundlich isotherm model mainly used to describe the heterogeneous adsorption on the adsorbent surface by the adsorbate molecules. Freundlich isotherm model is described by the following equation

$$q_e = K_f C_e^{1/n}$$

$$\log q_e = \log K_f + 1/n \log C_e.$$

Where: q_e is the adsorbed metal amount (mg/g), C_e is the equilibrium adsorbate concentration in mg/L, K_f is the adsorbent capacity and n is the adsorption intensity.

Calculated constants of Langmuir and Freundlich for the biosorption of Pb(II), Cu(II) and Ni(II) onto the surface of used biosorbents are found in Table (4) and graphically represented in Figures (8 & 9). The results showed that both Langmuir and Freundlich models are well-fitting to describe the isothermal biosorption type with $R^2 > 0.95$. A narrow difference between the adsorption capacities of both biosorbents was observed. R_L range of $0 < R_L < 1$ indicated that favorable adsorption of Pb(II), Cu(II) and Ni(II) was achieved (Table 4). The lower values of n (< 1) for studied metals ions revealed that the adsorption is a chemical adsorption process (Desta 2013).

Table 4: Langmuir and Freundlich isotherm constants for Pb, Cu and Ni biosorption onto the surface of biosorbent 1 and biosorbent 2.

Langmuir									Freundlich					
Biosorbent 1				Biosorbent 2					Biosorbent 1			Biosorbent 2		
b	$Q_{\max}$	R_L	R^2	b	$Q_{\max}$	R_L	R^2		K_f	n	R^2	K_f	n	R^2
Pb	0.46	11.29	0.04	0.98	0.35	17.51	0.05	0.97	11.21	0.52	1.00	10.51	0.60	0.97
Cu	0.20	12.09	0.09	0.97	0.16	11.74	0.11	0.98	2.73	0.59	0.94	1.70	0.55	0.94
Ni	0.04	1.63	0.33	1.00	0.22	2.78	0.08	0.98	1.29	0.66	0.99	7.52	0.62	0.99

Fig. 8. Langmuir isotherm plot for biosorption of Pb(II), Cu(II) and (Ni(II) ions by cyanobacterial biosorbents biomass.

Fig. 9. Freundlich isotherm plot for biosorption of Pb(II), Cu(II) and (Ni(II) ions by cyanobacterial biosorbents biomass.

3.7. Kinetics studies

Studies of biosorption kinetic is an important factor to evaluate the biosorption efficiency (Moghadam et al. 2013). Thus, pseudo-first order and pseudo-second order models were used to illustrate the kinetics biosorption of Pb(II), Cu(II) and Ni(II) onto surface of the two cyanobacterial biosorbents.

3.7.1. The pseudo-first order model

This model supposes that the uptake of metals ions is directly proportional to the difference between saturation levels, and expressed as;

$$\log(q_e - q_t) = \log(q_e) - \frac{k_1}{0.203} t$$

Where: q_t is the concentration of adsorbed metal ions (mg/g) at time t , q_e is the amount of adsorbed metal ions (mg/g) at equilibrium, k_1 (min^{-1}) is the constant of pseudo-first order reaction.

3.7.2. The pseudo-second order model

This model assumes occurring of a chemical adsorption. Thus, the chemical bonds were formed between biosorbent surface and adsorbate (Bernard et al. 2013). The pseudo-second order model equation is expressed as follows

$$\frac{t}{q_t} = \frac{1}{k_2 q_e^2} + \frac{1}{q_e} t$$

Where: q_t is adsorbed metal ions (mg/g) at time t , q_e is the amount of adsorbed metal ions (mg/g) at equilibrium, k_2 ($\text{mg/g} \cdot \text{min}^{-1}$) is the constant of pseudo-second order reaction

The kinetic constants and the linear plots of the pseudo-first and the pseudo-second order reactions are given in Table 5 and presented graphically in Figs 10 & 11. In accordance of calculated constants for both biosorbents, we can conclude that biosorption of studied metals followed the pseudo-second order reaction. The recorded q_e ranged between 19.72 – 26.3 and 18.97 – 24.1 mg/g for the two biosorbents with R^2 exceed than 0.99.

Table 5: Constants of Pseudo-first order and pseudo-second order reactions for biosorption of Pb(II) Cu(II) and Ni(II) onto the surface of cyanobacterial biosorbents.

	Pseudo first order reaction						Pseudo second order reaction					
	Biosorbent 1			Biosorbent 2			Biosorbent 1			Biosorbent 2		
	q_e	K_1	R^2	q_e	K_1	R^2	q_e	K_2	R^2	q_e	K_2	R^2
Pb	5.35	0.011	0.845	4.44	0.011	0.876	26.11	0.005	0.994	24.10	0.006	0.999
Cu	6.22	0.012	0.855	10.74	0.032	0.948	26.32	0.004	0.999	23.64	0.006	0.999
Ni	8.80	0.020	0.890	6.25	0.014	0.860	19.72	0.004	0.995	18.97	0.006	0.993

Figure 10. Pseudo-first order reaction plot for biosorption of Pb(II), Cu(II) and (Ni(II) ions by cyanobacterial biosorbents biomass.

Figure 11. Pseudo-second order reaction plot for biosorption of Pb(II), Cu(II) and (Ni(II) ions by cyanobacterial biosorbents biomass.

4. Conclusion

Two different microbial mats inhabit extreme habitats collected from hot spring (biosorbent 1) and from cold spring (biosorbent 2) and used these mats to remove Pb(II),

Cu(II) and Ni(II) from aqueous solutions. Microbial mats showed an excellent affinity to adsorb and remove metal ions from mixture aqueous solutions. Maximum biosorption efficiency for Pb(II), Cu(II) and Ni(II) for two used biosorbents was achieved at pH = 6, 150 min contact time, 2.5 g/L biosorbents dose and 50 mg/L as initial metal ions concentrations. Biosorbent 1 achieved maximum adsorption capacity of 46.26, 44.5 and 38.5 mg/g whereas, biosorbent 2 showed slight increase reached to 47.26, 45.1 and 38.3 mg/g for Pb(II), Cu(II) and Ni(II), respectively. Isothermal data revealed well representation with both Langmuir and Freundlich models, and the results prove higher biosorption capacity for Pb > Cu > Ni ions. Kinetically, the biosorption reaction is obeyed pseudo-second order reaction model with q_e ranged between of 19.72 – 26.32 mg/g with $R^2 > 0.99$

Declarations

Ethics approval and consent to participate

Not applicable

Consent for publication

Not applicable.

Authors' contributions

Khairia M. Al-Qahtani final revision and editing the manuscript, supporting the work through the allowed fund, Mohamed H.H. Ali conceived, designed the experiments and performed the experiments and wrote the manuscript, Afify D. G. Al-Afify and Mohamed S. Abdelkarim sharing in the experimental parts and analyzed the data. All authors read and approved the final manuscript

Declaration of interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Availability of data and materials

All data generated or analyzed during this study are included in this published article.

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References

- Abdel-Aty AM, Ammar NS, Ghafar HHA, Ali RK (2013) Biosorption of cadmium and lead from aqueous solution by fresh water alga *Anabaena sphaerica* biomass. J Adv Res 4: 367–374.
- Ahad RIA, Goswami S, Syiem MB (2017) Biosorption and equilibrium isotherms study of cadmium removal by *Nostoc muscorum* Meg 1: morphological, physiological and biochemical alterations. 3 Biotech 7(2): 1-12.
- Ahuja P, Gupta R, Saxena RK (1999) Zn²⁺ biosorption by *Oscillatoria angustissima*. Process Biochem 34: 77–85
- Al-Homaidan AA, Alabdullatif JA, Al-Hazzani AA, Al-Ghanayem AA, Alabbad AF (2015) Adsorptive removal of cadmium ions by *Spirulina platensis* dry biomass. Saudi Journal of Biological Sciences 22(6): 795-800.
- Ali MHH, Abd Elkarim MS, Haroun S, Attwa MK, (2019) Bioremediation of Fe, Zn and Cd ions from aqueous solution using died cells of cyanobacterial mats from extreme habitat, Siwa Oasis, Egypt. Egyptian Journal of Aquatic Biology and Fisheries 22(5): 511-522.
- Ali MHH, Hussian AEM, Abdel-Satar AM, Goher ME, Napiórkowska-Krzebietke A, El-Monem AMA (2016) The isotherm and kinetic studies of the biosorption of heavy metals by non-living cells of *Chlorella vulgaris*. Journal of Elementology 21(4): 1263-1276.
- Axson JL (2012) Hydrating aldehydes in the gas phase, atmospheric consequences. Thesis, University of Colorado, Boulder, Colorado, USA.
- Barquilha CER, Cossich ES, Tavares CRG, Silva EA (2019) Biosorption of nickel (II) and copper (II) ions by *Sargassum* sp. in nature and alginate extraction products. Bioresource Technology Reports 5: 43-50.
- Barrett EP, Joyner LG, Halenda PP (1951) The determination of pore volume and area distributions in porous substances. I. Computations from nitrogen isotherms. Journal of the American Chemical society 73(1): 373-380.
- Beltrán-Pineda ME, Gómez-Rodríguez AM (2016) Biorremediación de Metales Pesados Cadmio (Cd), Cromo (Cr) y Mercurio (Hg), Mecanismos Bioquímicos e Ingeniería Genética, Una Revisión. Revista Facultad De Ciencias Básicas 12(2): 172–197. <https://doi.org/10.18359/rfcb.2027>
- Bernard E, Jimoh A, Odigure J (2013) Heavy metals removal from industrial wastewater by activated carbon prepared from coconut shell. Res J Chem Sci 3: 3-9.

491 Brunauer S, Emmett PH, Teller E (1938) Adsorption of gases in multimolecular layers. Journal
492 of the American chemical society 60(2): 309-319.

493 Celekli A, Bozkurt H (2011) Bio-sorption of cadmium and nickel ions using *Spirulina platensis*,
494 kinetic and equilibrium studies. Desalination 275:141–147

495 Churchill SA, Walters JV, Churchill PF (1995) Sorption of heavy metal by prepared bacterial
496 cell surfaces. J. Environ. Eng 121: 706–711.

497 Das N, Vimala R, Karthika P (2008) Biosorption of heavy metals, an overview. IJBT 7: 159–
498 169.

499 Davis T, Volesky B, Muccib A (2003) A review of the biochemistry of heavy metal biosorption
500 by brown algae. Water Res 37: 4311– 4330.

501 Desta MB (2013) Batch sorption experiments, Langmuir and Freundlich isotherm studies for the
502 adsorption of textile metal ions onto teff straw (*Eragrostis tef*) agricultural waste. Journal
503 of thermodynamics 47 - 53.

504 Dixit S, Singh DP (2013) Phycoremediation of lead and cadmium by employing *Nostoc*
505 *muscorum* as biosorbent and optimization of its biosorption potential. Int. J.
506 Phytoremediation 15: 801–813

507 Dmytryk A, Saeid A, Chojnacka K (2014) Biosorption of microelements by *Spirulina*, towards
508 technology of mineral feed supplements. Sci World J Article ID 356328.

509 El-Naggar NEA, Hamouda RA, Mousa IE, Abdel-Hamid MS, Rabei NH (2018) Biosorption
510 optimization, characterization, immobilization and application of *Gelidium amansii*
511 biomass for complete Pb²⁺ removal from aqueous solutions. Scientific reports 8(1): 1-19.

512 Farooq U, Kozinski JA, Khan M, Athar M (2010) Biosorption of heavy metal ions using wheat
513 based biosorbents – a review of the recent literature. Bioresour. Technol 101: 5043–5053.

514 Gardea-Torresdey JL, Arenas JL, Francisco NMC, Tiemann KJ, Webb R (1998) Ability of
515 immobilized cyanobacteria to remove metal ions from solution and demonstration of the
516 presence of metallothionein genes in various strains. Journal of Hazardous substance
517 research 1(1): 2.

518 Gnanasambandam R, Protor A (2000) Determination of pectin degree of esterification by
519 diffuse reflectance fourier transform infrared spectroscopy. Food Chem 68: 327–332.

520 Goher ME, Abdel-Satar AM, Ali MH, Hussian AE, Napiórkowska-Krzebietke A (2016)
521 Biosorption of some toxic metals from aqueous solution using non-living algal cells of
522 *Chlorella vulgaris*. Journal of Elementology 21(3): 703-714.

523 Gupta A, Joia J, Sood A, Sood R, Sidhu C, Kaur G (2016) Microbes as potential tool for
524 remediation of heavy metals, A review. J Microb Biochem Technol 8(4): 364-72.

525 Iqbala M, Saeeda A, Zafarb SI (2009) FTIR spectrophotometry, kinetics and adsorption
526 isotherms modeling, ion exchange, and EDX analysis for understanding the mechanism
527 of Cd²⁺ and Pb²⁺ removal by mango peel waste. J Hazard Mater 164: 161–171.

528 Khajavian M, Wood DA, Hallajani A, Majidian N (2019) Simultaneous biosorption of nickel
529 and cadmium by the brown algae *Cytoseria indica* characterized by isotherm and kinetic
530 models. Applied Biological Chemistry 62(1): 1-12.

531 Khanafari A, Eshgh DS, Mashinchian A (2008) Removal of lead and chromium from aqueous
532 solution by *Bacillus circulans* biofilm, Iran. J. Environ. Healt 5: 3195–200.

533 Kim YH, Yoo YJ, Lee HY (1995) Characteristics of lead adsorption by *Undaria pinnatifida*.
534 Biotechnol Lett 17: 345–350.

535 Klimmek S, Stan HJ (2001) Comparative analysis of the biosorption of cadmium, lead, nickel,
536 and zinc by algae. Environ Sci Technol 35: 4283–4288.

537 Kumar MG (2017) Microbes in heavy metal remediation, a review on current trends and
538 patents. Recent patents on biotechnology 11(3): 188-196.

539 Kumar R, Kundu S (2020) Microbial Bioremediation and Biodegradation of Hydrocarbons,
540 Heavy Metals, and Radioactive Wastes in Solids and Wastewaters. In Microbial
541 Bioremediation & Biodegradation (pp. 95-112). Springer, Singapore.

542 Lameiras S, Quintelas C, Tavares T (2008) Biosorption of Cr (VI) using a bacterial biofilm
543 supported on granular activated carbon and on zeolite. Bioresource technology 99(4):
544 801-806.

545 Landa-Acuña D, Acosta RAS, Cutipa EH, de la Cruz CV, Alaya BL (2020) Bioremediation, A
546 Low-Cost and Clean-Green Technology for Environmental Management. In Microbial
547 Bioremediation & Biodegradation (pp. 153-171). Springer, Singapore.

548 Li FT, Yang H, Zhao Y, Xu R (2007) Novel modification pectin for heavy metal adsorption.
549 Chin Chem Lett 18: 325–328.

550 Madigan MT, Jung DO, Woses CR, Achenbach LA (2000) *Rhodoferrax antarcticus* sp. nov., a
551 moderately psychrophilic purple nonsulfur bacterium isolated from an Antarctic
552 microbial mat. Arch. Microbiol 173 (4): 269–277.

553 Maeir RM, Pepper IL, Gerba CP (2009) Environmental microbiology, 2nd edn. Academic Press.
554 Elsevier, San Diego California, p 585.

McKay G, Blair HS, Gardner JR (1982) Adsorption of dyes on chitin,. Journal of Applied Polymer Science 27 (8): 3043–3057.

Michalak I, Chojnacka K, Witek-Krowiak A (2013) State of the art for the biosorption process—a review. Appl. Biochem. Biotechnol 170: 1389–1416.

Micheletti E, Colica G, Viti C, Tamagnini P, De Philippis R (2008a). Selectivity in the heavy metal removal by exopolysaccharide-producing cyanobacteria. J Appl. Microbiol 105: 88–94.

Micheletti E, Pereira S, Mannelli F, Moradas-Ferreira P, Tamagnini P, De Philippis R (2008b) Sheathless mutant of cyanobacterium *Gloeotheca* sp. strain PCC 6909 with increased capacity to remove copper ions from aqueous solutions. Appl. Environ Microbiol 74: 2797–2804.

Moghadam MR, Nasirizadeh N, Dashti Z, Babanezhad E (2013) Removal of Fe(II) from aqueous solution using pomegranate peel carbon, equilibrium and kinetic studies. International Journal of Industrial Chemistry 4(1): 19.

Mohapatra RK, Parhi PK, Pandey S, Bindhani BK, Thatoi H, Panda CR (2019) Active and passive biosorption of Pb (II) using live and dead biomass of marine bacterium *Bacillus xiamenensis* PbRPSD202, Kinetics and isotherm studies. Journal of environmental management 247: 121-134.

Nakagawa T, Fukui M (2002) Phylogenetic characterization of microbial mats and streamers from a Japanese alkaline hot spring with a thermal gradient. J. Gen. Appl. Microbiol 48 (4): 211–222.

Paperi R, Micheletti E, De Philippis R (2006) Optimization of copper sorbing–desorbing cycles with confined cultures of the exopolysaccharide-producing cyanobacterium *Cyanospira capsulata*. J Appl Microbiol 101: 1351–1356.

Pathak J, Rajneesh MPK, Singh SP, Häder DP, Sinha RP (2018) Cyanobacterial farming for environment friendly sustainable agriculture practices, innovations and perspectives. Front Environ. Sci 6: 1–13.

Quintelas C, Rocha Z, Silva B, Fonseca B, Figueiredo H, Tavares T (2009) Removal of Cd (II), Cr (VI), Fe (III) and Ni (II) from aqueous solutions by an *E. coli* biofilm supported on kaolin. Chemical engineering journal 149(1-3): 319-324. <https://doi.org/10.1016/j.cej.2008.11.025>.

Qureshi FM, Badar U, Ahmed N (2001) Biosorption of copper by a bacterial biofilm on a flexible polyvinyl chloride conduit. Applied and environmental microbiology 67(9): 4349-4352.<https://doi.org/10.1128/AEM.67.9.4349-4352.2001>.

- 589 Rai PK, Tripathi BD (2007) Removal of heavy metals by the nuisance cyanobacteria
590 *Microcystis* in continuous cultures, an eco-sustainable technology. Environmental
591 Sciences 4(1): 53-59.
- 592 Shukla D, Vankar PS, Srivastava SK (2012) Bioremediation of hexavalent chromium by a
593 cyanobacterial mat. Applied Water Science 2(4): 245-251.
- 594 Singh S (2014) A review on possible elicitor molecules of cyanobacteria, their role in improving
595 plant growth and providing tolerance against biotic or abiotic stress. J Appl Microbiol
596 117: 1364–5072
- 597 Singh S (2020) Biosorption of Heavy Metals by Cyanobacteria, Potential of Live and Dead
598 Cells in Bioremediation. In Microbial Bioremediation & Biodegradation (pp. 409-423).
599 Springer, Singapore.
- 600 Todorova K, Velkova Z, Stoytcheva M, Kirova G, Kostadinova S, Gochev V (2019) Novel
601 composite biosorbent from *Bacillus cereus* for heavy metals removal from aqueous
602 solutions. Biotechnology & Biotechnological Equipment 33(1): 730-738.
603 <https://doi.org/10.1080/13102818.2019.1610066>.

Figures

Figure 1

SEM micrograph of biosorbent 1: a) before and b) after, and biosorbent 2: c) before and d) after adsorption of Cu, Ni and Pb ions from aqueous solution.

Figure 2

Energy-dispersion spectra (EDX) of biosorbent 1: a) before, b) after and biosorbent 2: c) before, d) after adsorption of Cu, Ni and Pb ions.

Figure 3

FT-IR spectra of; a) biosorbent 1 biomass and b) biosorbent 2 biomass before (black line) and after (red line) biosorption of metal ions from aqueous solution

Figure 4

Effect of pH values on biosorption of Pb(II), Ni(II) and Cu(II) onto the surface of A) biosorbent 1 and b) biosorbent 2

Figure 5

Effect of contact time (min) on biosorption of Pb(II), Ni(II) and Cu(II) onto the surface of biosorbents

Figure 6

Effect of biosorbent dosage (g/L) on biosorption of Pb(II), Ni(II) and Cu(II)

Figure 7

Effect of initial metals concentration (mg/L) on biosorption of Pb(II), Ni(II) and Cu(II) onto the surface of A) biosorbent 1 and b) biosorbent 2

Figure 8

Langmuir isotherm plot for biosorption of Pb(II), Cu(II) and (Ni(II) ions by cyanobacterial biosorbents biomass.

Figure 9

Freundlich isotherm plot for biosorption of Pb(II), Cu(II) and (Ni(II) ions by cyanobacterial biosorbents biomass.

Figure 10

Pseudo-first order reaction plot for biosorption of Pb(II), Cu(II) and (Ni(II) ions by cyanobacterial biosorbents biomass.

Figure 11

Pseudo-second order reaction plot for biosorption of Pb(II), Cu(II) and (Ni(II) ions by cyanobacterial biosorbents biomass.