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

Khairia M. Al-Qahtani1, Mohamed H.H. Ali2, Mohamed S. Abdelkarim2, Afify D. G. Al-Afify2

1, Chemistry Department, Faculty of Science – Princess Nourah bint Abdulrhaman
University, Riyadh, Saudi Arabia
2, National Institute of Oceanography & Fisheries, Cairo, Egypt

*Corresponding author:
Mohamed H.H. Ali
E-mail address: mhha_ali@yahoo.com

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.
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 a 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 biomass 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:

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

\[ R\% = \left( \frac{C_i - C_f} {C_i} \right) \times 100 \]

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        | \( S_{BET} \) (m²/g⁻¹) | \( r \) (nm) | \( V_p^{total} \) (cm³/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.](image)
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...
The peak at 1630 cm$^{-1}$ is attributed to stretching vibration of C=C (Gnanasambandam and Protor 2000). The weak stretch of -COO- of pectin or C=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.

| Wave number. (Cm$^{-1}$) | Attribution               | Wave number. (Cm$^{-1}$) | Attribution               |
|-------------------------|---------------------------|---------------------------|---------------------------|
| Before adsorption       | After adsorption          | Attributions              | Before adsorption          | After adsorption          |
| 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                    | Alkyl Halide stretch C-I  | 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                    | Alkyl Halide stretch C-I  | 693.2839                  | 692.32                   | Alkyl Halide stretch C-I |
|                         |                           |                           | 512.97                    | 532.257                  |
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, Ni²⁺ and Pb²⁺ 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 qe of 49.8 and 48.8 mg/g followed by Ni(II) with removal percentage of 68.1% (qe = 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](image)

![Figure 5. Effect of contact time (min) on biosorption of Pb(II), Ni(II) and Cu(II) onto the surface of biosorbents](image)
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)](image-url)
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) | $Q_{\text{max}}$ (mg/L) | Ref.                  |
|----------------------------------|--------------|----|----------|------------|-------------------------|-----------------------|
| *Synechococcus sp.* PCC 7942    | Ni, Cu, Pb   | 5.0| 1        | 80         | 3.2, 11.3, 30.4         | Gardea-Torresdey et al. 1998 |
| *Oscillatoria sp.*              | Cr           | 5.5| 1        | 210        | 25                      | Shukla et al. 2012    |
| *Anabaena sphaerica*            | Cd, Pb       | 6.0| 2        | 60 – 90    | 111 and 121             | Abdel-Aty et al. 2013 |
| *Sargassum sp*                  | 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| 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:

\[
\frac{1}{q_e} = \frac{1}{Q_{\text{max}}} + \frac{1}{b Q_{\text{max}}} \cdot \frac{1}{c_e}
\]
where: $q_{\text{max}}$ (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 |
| $b$   | $Q_{\text{max}}$ | $R_L$ | $R^2$ | $b$ | $Q_{\text{max}}$ | $R_L$ | $R^2$ | $K_f$ | $n$ | $R^2$ | $K_f$ | $n$ | $R^2$ |
| 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 biosorbert surface and adsorbate (Bernard et al. 2013). The pseudo-second order model equation is expressed as follows

$$\frac{t}{q_t} = \frac{1}{k_2q_e^2} + \frac{1}{q_e} t$$

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, $k_2$ (mg/g.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 biosorberts, 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                   | 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  |
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) 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) ions by cyanobacterial biosorbents biomass.
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 qe 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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