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Potential of *Staphylococcus Xylosus* Strain for Recovering Nickel Ions from Aqueous Solutions

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Adel Ali Saeed Abduh Algheethi[1,*], Efaq Noman[2], Radin Maya Saphira Radin Mohamed[1], Mohamed Osman Abdel-Monem[3], Amir Hashim Mohd Kassim[1]

[1] Micro-pollution Research Centre (MPRC), Department of Water and Environmental Engineering, Faculty of Civil and Environmental Engineering, Universiti Tun Hussein Onn Malaysia (UTHM), 86400 Parit Raja, Batu Pahat, Johor, Malaysia
[2] Environmental Technology Division, School of Industrial technology, Universiti Sains Malaysia (USM), 1800, Penang, Malaysia
[3] Botany Department, Faculty of Science, Benha University, Egypt

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ABSTRACT

The potential of bacterial biomass for the biosorption of heavy metals has investigated extensively. However, the bacterial species exhibited different affinities toward the heavy metals ions based on their differences in cell wall characteristics, structure and physiological status (living or dead cells). In this study, the potential of living and dead cells of *Staphylococcus xylosus* 222W for removal nickel ions from aqueous solution as a function for physiological status, nickel and biomass concentrations, time, pH and temperature was investigated. The pre-treatment of bacterial cells was performed by the heating at 100 ºC for 15 min. The removal experiments were conducted in the lab scale. The results revealed that the dead cells exhibited more efficiency in removing nickel ions than living cells at all investigated concentrations (2 to 10 mM). The biosorption efficiency (E %) increased with increasing in biomass cells to limit concentrations (0.1 to 1 g dry wt L^{-1}). The maximum removal of nickel was 81.41 vs. 77.10 % by living and dead cells, respectively achieved after 9 and 10 hrs of the incubation period, respectively. The acidic conditions decrease the efficiency of metal removal, while the optimal removal was recorded at pH 8 for both biomass (living and dead cells). The maximum uptake capacity of *S. xylosus* 222W (living and dead cells) was recorded at 37oC, the percentage removed being 75.90 vs. 84.92 %, respectively. It can be concluded that *S. xylosus* 222W exhibited high potential and affinity to remove of nickel ions from aqueous solution.

Keywords:
Biosorption, efficiency, factors, heavy metals, *S. xylosus*

1. Introduction

The potential of bacterial cells for removing heavy metals ions from contaminated water and wastewater have been reported by many authors in literature [1,2]. However, bacterial strains have a different efficiency, capacity and affinity towards the metal ions. These differences related the

* *Corresponding author.*
E-mail address: alisaeed@uthm.edu.my (Adel Ali Saeed Abduh Algheethi)

cell wall composition and presence functional groups on their surface. Therefore, the examination for each bacterial species might lead to find a high potential strain which is more applicable for treatment of wastewater. The bacterial strains isolated from heavy metal contaminated wastewater might occur more potential to resist and remove the metal ions compared to that obtained from non-contaminated area even if both strains are classified within the same species [3,4]. Sundar *et al*. [5] revealed that the potential of B. subtilis to remove Cr^{3+} ions improved effectively after the acclimatization to high concentrations of Cr^{3+}. The removal percentage increased from 64 to 85 %. This might be due to the over production of exo-polysaccharide (EPS) by the bacterial cells which is one of the mechanism used to resist the metal ions toxicity. Naik *et al*. [6] indicated that B. cereus which has high ability to tolerate Cr^{3+} exhibited more than 75 % of Cr^{3+} removal. Moreover, understand the response of bacterial cells to the environmental factors represent the critical step to find a more suitable bacterial strain with high efficiency to remove heavy metals. These factors play an important role in the removal process by the direct affecting the bacterial cells or the chemical status of the metal ions. Nickel ions are among different heavy metal ions which have less affinity to adsorb on the bacterial cells. The morphological characteristics of the bacterial cells are the main factor in the metal removal by the biosorption process. *Staphylococcus xylosus* with a Gram positive reaction and cocci shape might provide high exposure area to adsorb of nickel ions. The cell wall of this bacteria has high content of peptides which are the active site of the biosorption process.

The present paper is a continuous work for the previous work in which *Bacillus subtilis*, and *Burkholderia cepacia* was used for biosorption of nickel ions [1]. Moreover, *S. xylosus* has exhibited more tolerant for nickel ions compared to those bacteria. Therefore, *S. xylosus* 222W might occur different reposes for the biosorption factors and this is the aim of the present work. The factors affecting biosorption process which included concentration of metal ions and biomass, living and dead cells, time, pH and temperature affecting biosorption process were investigated.

2. Materials and Methods

A. Experimental Set-up

The experimental-setup used in this work included isolation, purification, identification of *S. xylosus* 222W as well as tolerance for Ni^{2+} ions. The potential of living and dead *S. xylosus* cells for recovering Ni^{2+} ions from aqueous solution were conducted. The biosorption experiments were performed in batch solution contain 100 mL of Ni^{2+} ions. Factorial Complete Randomized Design (CRD) (5×3×***x***) in triplicate was used to investigate the factors affecting the recovery of Ni^{2+} process. Two (2) cell biomass (living and dead) and one (1) control for the recovering process (Ni^{2+} ions solution without cells) making a total of three (3) groups. Five (5) factors included concentration of Ni^{2+} ions and biomass, time, pH and temperature, while x the values used for each factor.

B. Preparation of the Bacterial Cell Biomass and Ni^{2+} Ions Solution

The preparation of *S. xylosus* 222W cell biomass was prepared as described in previous work [1]. The bacterial tolerance for Ni^{2+} ions was conducted at 15 mM Ni^{2+} ion concentrations on Brain Heart Infusion medium [4]. The bacterial biomass were prepared as living cells (untreated cells) and dead cells (pre-treated at 100 ºC for 15 mins). Nickel ions solution was prepared in a concentration of 15 mM and then diluted with distilled water to obtain the required concentrations.

C. Factors Affecting the Recovery of Ni^{2+} Ions Process

The effect of nickel ion concentrations (1, 2, 3, 4, 6, 8 and 10 mM), biomass concentrations (0.2, 0.4, 0.8, 1.6, 2, 2.4 and 3 g dry wt L^{-1} living and dead cells), time (1, 2, 3, 4, 5, 6, 7, 8, 9 and 10 hrs), pH (2, 3, 4, 5, 6, 7 and 8) and temperatures (20, 30, 37, 45 and 55ºC) was investigated. The bacterial cells were separated from the aqueous solution by centrifuging at 4020 crf for 20 min. The concentration of Ni^{2+} ions in the supernatant was determined by using UV-160 A spectrophotometer (Win. Aspect T20, 031-2004, Germany) according to the procedure described by Snell and Snell [7]. Bacterial cells efficiency in removing Ni^{2+} ions was calculated according Eq. 1 [8].

$$E = \frac{C_i - C_f}{C_i} \times 100 \quad (1)$$

where; C_i represent Ni^{2+} ions (initial concentration, mM); C_f is Ni^{2+} ion concentrations (mM) in the supernatant determined at the end of the experiment.

The differences in the removal efficiency of nickel ions were analysed using ANOVA (S-N-K test) and considered significant at $p \leq 0.05$.

3. Results and Discussion

A. Effect of Nickel Ions and Cells Concentration

The recovering different concentrations of Ni^{2+} ions by *S. xylosus* 222W (living and dead cells) is depicted in Figure 1. The results revealed that the living bacterial cells show their highest efficient for recovering low concentrations of Ni^{2+} ions with the highest recovery percentage (89.97%) recorded at 3 mM. In contrast, dead bacterial cells exhibited more efficient than living cells at low (1 and 2 mM) and high concentrations (4 mM), where the removal percentage was 91.89 vs. 85.66 % for living cells. The removal of Ni^{2+} at 3 mM by dead cells were less than that recorded at 2 and mM, but these differences are not significant and might be related the distribution of Ni2+ ions or bacterial biomass in the solution.

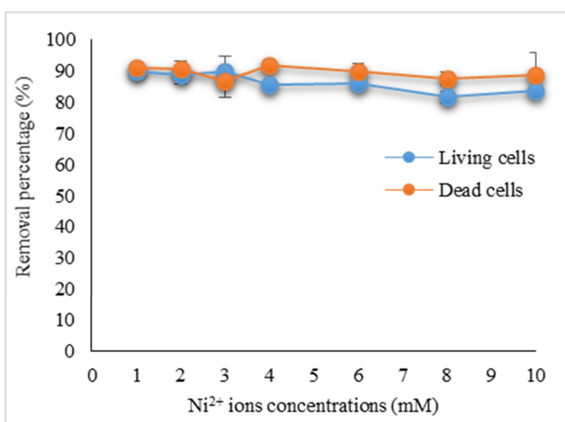

Fig. 1. Recovery of Ni^{2+} ions by *S. xylosus* 222W at different metal concentrations (living and dead cells)

The removal of Ni^{2+} ions from the aqueous solution as a function of cell biomass concentrations is presented in Figure 2. The increasing of cell concentrations associated with the removal efficiency. At 2 mM the maximum efficiency of dead cells was recoded with 3 g dry wt. L-1 (91.12 %). The highest removal efficiency by living cells was noted with 1 g dry wt. L-1 (85.03%). In contrast, no significant differences ($p<0.05$) between living and dead cells in their efficiency for recovery Ni2+ ions from the aqueous solution with 4 mM. The maximum removal was recorded with 0.8 g dry wt. L-1, and no increasing in the removal was showed with increasing cell biomass concentrations for 3 g dry wt. L-1.

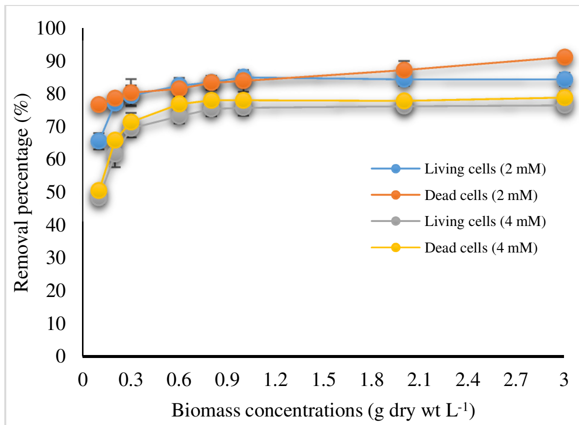

Fig. 2. Recovery of Ni2+ ions by S. xylosus 222W at different cell biomass concentrations (living and dead cells)

The recovering Ni^{2+} ions from aqueous solution by the bacterial cells as a response for different concentrations of metal ions and cell biomass depend on the equilibrium between the metal ions and active sites available on the bacterial cell surface [2,9]. At low concentrations of Ni^{2+} ions more active site are available for biosorption of metal ions, while at high concentrations of metal ions the free active sites become saturated with the metal ions with high concentrations of the metals still available in the solution. In contrast, increasing of cell biomass associate with the removal efficiency. Similar findings were also reported with different bacterial species and metal ions [1,4]. The high concentration of the biomass lead to the agglomeration of these cells which effect negatively on the removal process due to the decreasing in the surface area exposed to the metal ions even in the presence high concentrations of these ions [10]. The high efficiency of dead cells in comparison with the living cells might be explained based on the absence of competing protons produced during metabolism [11].

B. Time Dependency Studies for Nickel Biosorption

The removal percentage of Ni^{2+} ions during the period from 1 to 10 hrs is shown in Figure 3. It can be noted that the recovery of Ni^{2+} ions increasing with the time. The living bacterial cells achieved their maximum removal after 10 hrs (77.10%), while the dead cells recorded the highest removal after 9 hrs (81.41%). In comparison with previous studies, the authors mentioned that the time required for the biosorption of metal ions depend on the metal ions type and concentrations. For instance, Goksungur *et al*., [12] indicated that the removal of Cu^{2+} with low concentrations taken only 5 mins, while at high concentrations the time required for the biosorption process took between 30 and 60 min. In terms of type of the metal ions, Jang *et al*., [13] reported that the Ni^{2+}

removal reached to the steady-state level after 24 hrs. In the present study, the maximum removal was achieved on 9th and 10th hrs. This might be related to the affinity of the Ni^{2+} ions to the biosorption site located on the bacterial cell surface, where Ni^{2+} ions have less affinity to biosorp on bacterial cells in comparison with others metals [4].

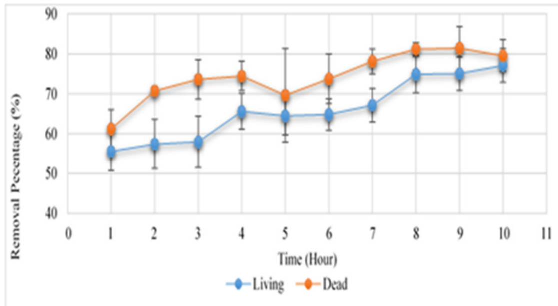

Fig. 3. Recovery of Ni2+ ions by S. xylosus 222W at different time (living and dead cells)

C. Recovering of Ni^{2+} ions in a response for pH and Temperature

Both pH and temperature exhibited an effect on the removal of nNi^{2+} from the aqueous solutions by the bacterial cells (Figure 4 and 5). pH occurred more influence on the dead bacterial cells than living cells. In contrast, temperature effected more on the living cells than dead cells. The extreme pH values (acidic condition) exhibited more influences on the removal process in comparison to the extreme temperature (high temperature). The maximum recovering efficiency was noted at pH 8 (81.6 vs. 76.58% for living and dead cells respectively). The highest removal percentage of Ni^{2+} ions by living and dead cells was observed at 37ºC (75.9 vs. 84.9 %, respectively).

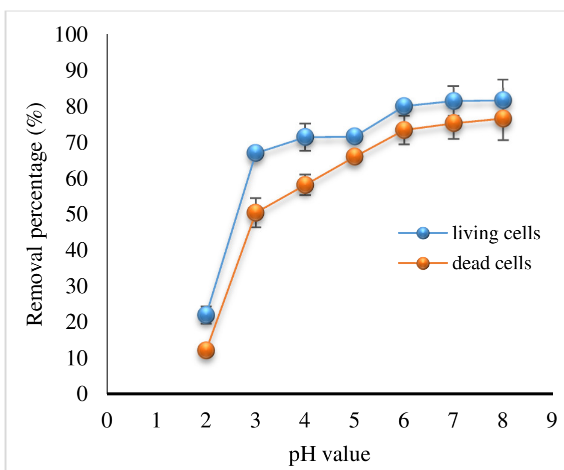

Fig. 4. Biosorption of Ni^{2+} ions by *S. xylosus* 222W at different pH values concentrations (living and dead cells)

The role pH values in the biosorption process of metal ions by the bacterial cells belongs to the effect on the chemical status of the metal ions such as solubility in the solution and releasing H^+ proton from the bacterial cells. Both effects might enhance or reduce the removal percentage.

However, many of the authors have mentioned that the optimum pH for the biosorption process is located between pH 4 and 8 [14,15].

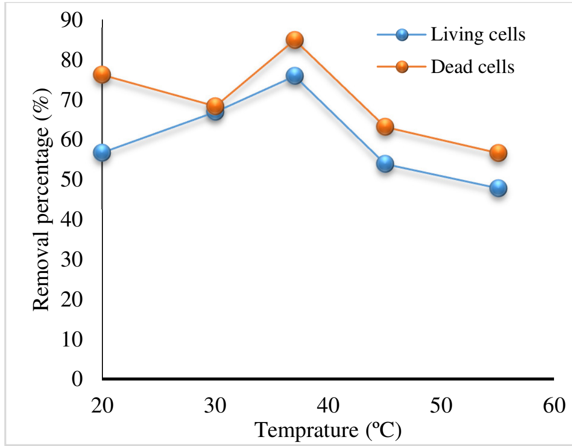

Fig. 5. Biosorption of Ni^{2+} ions by *S. xylosus* 222W at different temperature (living and dead cells)

The explanation for the effect of temperature on the biosorption process is differ from one author to another. These difference related to the nature of the biosorption process as endo-thermic or exothermic reactions. Habib-ur-Ruhman *et al*., [16] claimed that the biosorption capacity of Ni^{2+} by *Dalbergia sossoo* is an endothermic process. Therefore, the increasing of temperature associated with the increasing of removal efficiency. Selatnia *et al*., [10] reported that the adsorption generally increase with decrease in temperature and the physical adsorption reactions are normally exothermic. In the present study, the recovery efficiency correlated with the increasing of temperature from 20 to 37ºC and then reduced significantly when the temperature raised to 45 and then 55ºC. Therefore, it can be indicated that the temperature effect on the biosorption process by provided the metal ions with the energy emission and the biosorption by *S. xylosus* is an endothermic reaction. At low temperature the metal ions biosorped on the functional group of the bacteria cell surface has no energy to release into the solution. In contrast, the high temperature provide the metal ions with the energy emission and contribute in the broken the bond between metal ions and functional group and then release to be free in the solution.

4. Conclusions

It can be concluded that *S. xylosus* 222W has the potential to recovering the Ni^{2+} ions from the aqueous solution. The factors investigated here exhibited an influence on the recovering process. However, *S. xylosus* 222W has a wide range of the factors values and exhibited an efficiency for recovering metal ions even at low or high values, this properties make this bacterium more applicable for removing Ni^{2+} ions from different contaminated water or wastewater without the need to adjust the characteristics of these media.

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