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Bio Removal Potential of Nickel(II) by Different Bacterial Species

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***Abstract*– This study is aimed to evaluate the bio removal potential for nickel (II) as a toxic heavy metal for human and plant. So, The ability of Nickel uptake by metal-resistant five different types of bacterial species well identified and had previously work heavy metal, copper and zinc bio removal as Bacillus megaterium EMCC 1013, Rhizobium rhizogenes EMCC1743, Rhizobium leguminosarum EMCC1130, Azotobacter vinelandii and Nocardiopsis Dassenvillei were evaluated their potential activity in bio removal of nickel (II).**

Our results showed that five bacterial species have great variation potential for nickel (II) bio removal. Bacillus megaterium EMCC has the highest potential for bio removal of nickel 10 ml of 600ppm with 26.67 % removal after 24 h with inoculum size 0.1 x 10 29 Cu and incubation temperature 30 °C at PH 7 and energy source glucose and ammonium oxalate as carbon and nitrogen source. The aim of our study was to evaluate the bio removal capacity of nickel as a toxic heavy metal by five different bacterial species to use them in further study in removal of nickel (II) from electroplating waste water.

In addition Bacillus megaterium EMCC as the most potent nickel (II) resistant microorganisms will very useful in biotechnology for the remediation of metal contaminated environments with nickel and can also be used in the construction of biomarkers for the detection of nickel (II) ions.

***Key words*– Bioremoval, Bacillus megaterium, waste industry, Nickel**

I. Introduction

Heavy metals considered as main dangerous pollutants for both sewage and industrial wastewater [1]. Also one of the principle response cause pollution of water and soil[2]. Human activities, such as mining industries and most of industrial discharge wastes accumulate a lot of heavy metals in the environment and also, through the food chain, leading to dangerous environmental and health issues [3].

The traditional wastewater treatment methods, such as electrochemical treatment, ion exchange, oxidation-reduction, and membrane separation, are very expensive and have several disadvantages, such as unpredictable metal ion removal, lot of reagent preparations, and production of toxic sludge, which are often hard to dewater and need high caution during disposing processes [4].

Bio removal, which includes the utilize different microorganisms to remove or degrade environmental heavy metal contaminants, has arrived ascending awareness to clean up a metal environmental contaminant [5]. Bio removal process gives a safer and less economic ways to ordinary used physiochemical strategies [6]. By utilizing different microorganisms as the biomass of bacteria [7], fungi [5], and algae [8] for the safe removal of heavy metals from waste water and sludge is attracting concern. Microorganisms with the ability to grow in the presence of heavy metals and with a significant metal uptake have a potential use in bio removal of polluted waters[9].

Two different strains of *Cupriavidus metallidurans* utilized for Ni-resistant microorganisms were isolated from a decantation tank at a zinc factory [10] and a metal-contaminated industrial site[11].

This research aimed to Screening the bio removal capacity of Nickel by utilizing assorted species of bacteria to utilize them in next researches to removal of Nickel from industrial waste water plating industries.

II. Material and Methods:

A. Microorganisms:

Three bacterial species were purchased from the Egyptian Microbial Culture Collection, Ain shams university (*Bacillus megaterium* EMCC 1013, *Rhizobium rhizogenes* EMCC1743, *Rhizobium leguminosarum* EMCC1130). *Azotobacter vinelandii* was obtained by El -Badry et al [12] and *Nocardiopsis Dassenvillei* was obtained by Elbarbary et al., [13].

B. Chemicals and instrumentation

1. Nickel (II) stock solution:

Synthetic Nickel (II) sample was prepared by dissolving Nickel Sulfate of 4.476g in one liter of distilled water to make a 1000 ppm stock solution. The solution was diluted to get the concentrations.

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2. Nickel (II) bio removal Experiments:

LB (Luria-Bertani) liquid medium (Oxoid) was used as basal media consists of different ppm concentration of nickel (II) solution. Different pH was prepared by adjustment 0.1(N) HCl and 0.1(N) Na OH solutions. After that media was autoclaved in 250 ml conical flasks containing 100 ml medium. The media were inoculated with five different bacterial species. After incubation time samples were collected and centrifuged at 6000 RPM for 10 minutes. Supernatant was assayed for the nickel removal by Optical Emission Spectrometer Model: Optima 2000 DV Perkin Elmer (Inductive Couple Plasma). Bio removal of nickel ion in basal media inoculated with five different bacterial species separately were evaluated by following equation . All the glassware was cleaned with 5% HNO_3.

3. The relative effects of different Ni (II) concentration bios removal of microbial growth

Five different bacterial species were grown in a rotary shaker at 150 RPM and pH 7.0, while the temperature was 37 °C in LB broth medium supplemented by Different concentration (10, 15, 20, 30, and 40ml) of 300 ppm of Ni (II) for each bacterial species. After 24 h of incubation the remediation percentage of Ni (II) concentration on each bacterial growth was assessed

4. Relative effects of different inoculum size of Ni (II) bios removal

Five different bacterial species were grown in a rotary shaker at 150 RPM and pH 7.0, while the temperature was 37 °C in LB broth medium supplemented by Different inoculum size (0.1×10^{29}, 0.5×10^{29} , 1×10^{29}, 3×10^{29} and 5×10^{29}) cfu of each bacterial species. After 24 h of incubation the remediation percentage of Ni (II) concentration on each bacterial growth was assessed

5. Relative effects of different Temperature on Ni (II) bio removal

Five different bacterial species were grown in a rotary shaker at 150 RPM and pH 7.0, while the temperature was 37 °C in LB broth medium supplemented by Different incubation temperature 20, 25, 30, 35 and 40 °C. After 24 h of incubation the remediation percentage of Ni (II) concentration on each bacterial growth was assessed

6. Relative effects of different pH of Ni (II) bios removal

Five different bacterial species were grown in a rotary shaker at 150 rpm and pH 7.0, while the temperature was 37 °C in LB broth medium supplemented by Different PH (4, 5 , 6 , 7 and 8). After 24 h of incubation the remediation percentage of Ni (II) concentration on each bacterial growth was assessed

7. Relative effects of different Carbon sources on Ni (II) bio removal

Five different bacterial species were grown in a rotary shaker at 150 rpm and pH 7.0, while the temperature was 37 °C in LB broth medium supplemented by Different carbon sources (glucose, starch, sucrose and dextrose). After 24 h of incubation the remediation percentage of Ni (II) concentration on each bacterial growth was assessed

8. Relative effects of different Nitrogen sources on Ni (II) bio removal

Five different bacterial species were grown in a rotary shaker at 150 RPM and pH 7.0, while the temperature was 37 °C in LB broth medium supplemented by Different nitrogen sources (ammonium chloride, ammonium sulfate, ammonium oxalate, glycine and asparagine). After 24 h of incubation the remediation percentage of Ni (II) concentration on each bacterial growth was assessed**.**

III. Results and discussion

Pollution produced by the accumulation of heavy metals considers serious issue that can make passive effects on the hydrosphere. One of the best methods in elimination the toxicity of heavy metals from the environment is using Bacterial bio removals.

The environmental pollution caused by heavy metal toxicity is ascending all the world along technological development. Nickel one the most heavy metals used and the more wide expansion contaminants of the environment [14]. Wastewater has a high concentration of different heavy metals not degraded by the ordinary methods of wastewater recycling. The main source of heavy metals is the industrial activities such as metal processing, mining and electroplating, tanning, carpet washing and dying. Presence of high concentration of toxic heavy metals in waste water can cause severe problems to human health [15]. Bio removal can be used to effectively reduce contaminant toxicity, mobility or volume to levels that are innocuous to human health and ecosystem[16].

III.1. The relative effects of different Ni (II) concentration bios removal of microbial growth

Five different bacterial species *Nocardiopsis Dassenvillei*, *Azotobacter vinelandii*, *Bacillus megaterium* EMCC 1013, *Rhizobium rhizogenes* EMCC1743 and *Rhizobium leguminosarum* EMCC1130 were screened for their removal activity percentage of Ni (II) using different concentrations of Ni (II) with 13.41, 20.84, 21.52, 14.13 and 20.67 % respectively for 10 ml of 300 ppm Ni (II). Their removal activity decreased by an increase in Ni (II) concentration as shown in (figure No. 1)

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The effect of different Ni (II) dose on the bio removal potency showed that decrees in growth rate as compared with control. This has been explained that the exposed microorganism to metal stress devote its energy from growth to maintenance of other functions as a greater demand of energy to resist metal toxicity [17]. As described by Alboghobeish et al[18]. Bio removal efficiency increased with time, and maximum efficiency was observed at 72 h of growth (11.78% reduction in Ni^{2+} concentration). Bio removal was negligible after this time. Specific surface properties and the physiological state of the microorganisms might have a role in metal uptake. High biomass production is also important for better bio removal.

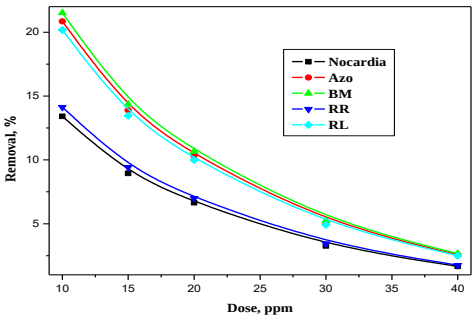

Fig. 1 Relative effect of different Ni (II) (ppm) concentration bio removal of different bacterial species

III.2. Relative effects of different inoculum size of Ni (II) bios removal

Different inoculum sizes of five bacterial used for Ni (II) bio removal were evaluated as shown in figure no 2 which appear that Nickel at concentration 10 ppm appeared as an increase in bacterial cell count decrease percentage of Ni (II) bios removal. The highest bio removal was by using inoculum size 0.1 x 10 ^{29} cfu of five different bacterial species as *Bacillus megaterium* EMCC 1013 was 22 %. From the above results *Bacillus megaterium* EMCC 1013 showed the most resist potency organisms for Ni (II) bio removal organism as shown in figure No 2

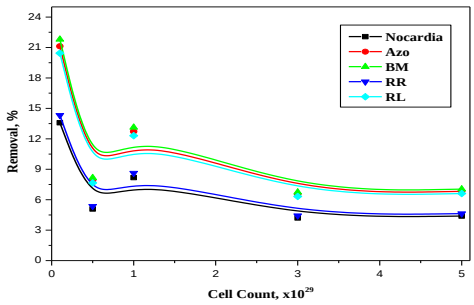

Figure No 2: Relative effects of different inoculum size on Ni (II) bio removal by different bacterial species

III.3. Relative effects of different Temperature of Ni (II) bio removal

Effect of different incubation temperature for Ni (II) bio removal using *Bacillus megaterium* EMCC 1013, *Rhizobium rhizogenes* EMCC1743, *Rhizobium leguminosarum* EMCC1130 Azotobacter *vinelandii* and *Nocardiopsis Dassenvillei*. *Bacillus megaterium* EMCC 1013 was the most resist potency organisms for Ni (II) bio removal percentage with 23.73 % at 30 ^{o}C as shown in figure No 3. As described by Alboghobeish et al. 2014[17] they isolates high resisitance bacterial starins for nickel as heavy metal under 30 ^{o}C.

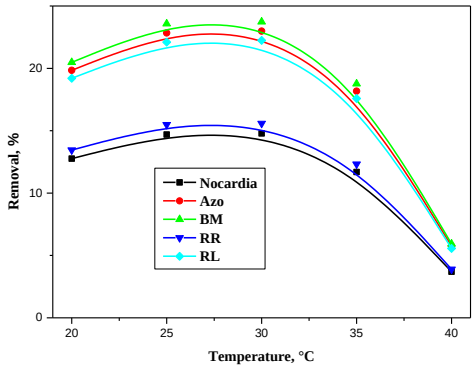

Figure No 3: Relative effects of different Temperature on Ni (II) bio removal by different bacterial species

III.4. Relative effects of different pH on Ni (II) bio removal

Effect of different pH for Ni (II) bio removal using Bacillus *megaterium* EMCC 1013, Rhizobium rhizogenes EMCC1743, Rhizobium leguminosarum EMCC1130 *Azotobacter vinelandii* and Nocardiopsis Dassenvillei. Bacillus megaterium EMCC 1013 was the most resist potency organisms for Ni (II) bio removal percentage with 17.59 % at PH 7. This result similar to Stanley et al.[19] in Bioremoval of nickel using *Pluerotus ostreatus*

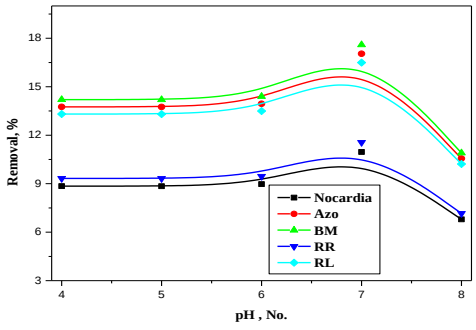

Figure No 4: Relative effects of different PH on Ni (II) bio removal by different bacterial species

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III.5. Relative effects of different Carbon sources on Ni (II) bio removal

Effect of different carbon sources for Ni (II) bio removal using five different bacterial strains was evaluated. *Bacillus megaterium* EMCC 1013 was the most potent Ni (II) bio removal percentage with 25.8 % Ni (II) bio removal with glucose utilization as carbon source figure No 5. Our results was agree with results As reported by El badry et al.[11], *Azotobacter vinelandii* isolate grows well on presence of glucose as carbon source during bio dissolution of phosphate. The bacterial growth exhibited remarkable variation according to the utilized carbon source, the best bacterial growth to produce enzyme and organic acids reached when glucose is utilized as a carbon source

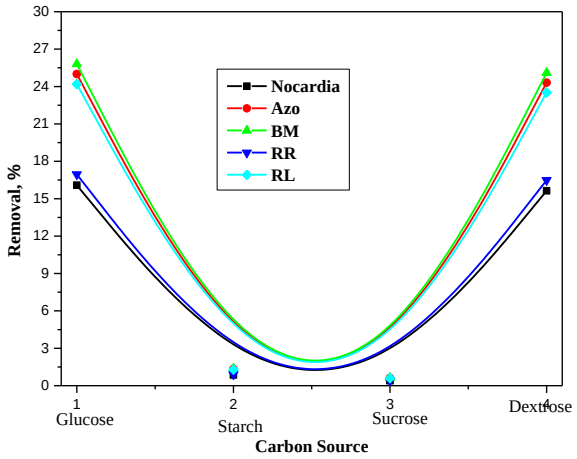

Figure No 5: Relative effects of different Carbon sources on Ni (II) bio removal by different bacterial species

III.6. Relative effects of different Nitrogen sources on Ni (II) bio removal

Effect of different nitrogen sources for Ni (II) bio removal using five different bacterial strains. *Bacillus megaterium* EMCC 1013 was the most efficacy Ni (II) bio removal percentage with 26.67 % figure No 6. As a nitrogen source, ammonium oxalate was found to give maximum soluble Phosphate. As reported by Elbarbary et al., 2015 Nocardiopsis dassenvillei had high efficacy for dissolution phosphorus using ammonium oxalate.

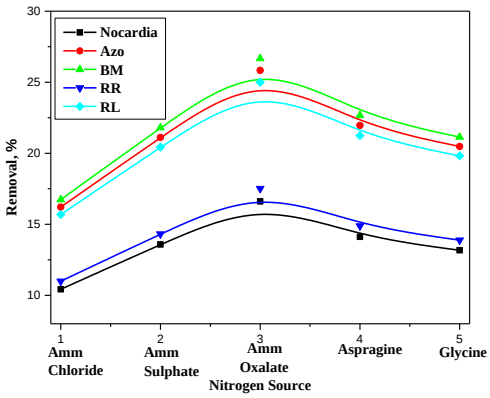

Figure No 6: Relative effects of different nitrogen sources on Ni (II) bio removal by different bacterial species

IV. Conclusion

Nickel as heavy metal elements was a removal evaluation of different bacterial species. *Bacillus megaterium* EMCC 1013 was the most potent of nickel removal by 26.67 %. The results of this work is important to be well understood the bio removal mechanism of *Bacillus megaterium* EMCC, and is significant for its pilot test and future practical application

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