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Assessing the Suitability of Green Algae (Chlorella Sorokiana) as Biosorbent for Removal of Cu (II) and Zn (II) Ions from Synthetic Wastewater in a Batch System

***[1]OLIOMOGBE, TI; [2]NKOLLO, MI; [1]EMEGHA, JO**

*[1]Department of Energy and Petroleum Studies, College of Natural and Applied Sciences, Novena University Ogume, P.M.B. 02, Kwale, Delta State, Nigeria
[2]Department of Optometry, College of Medical and Health Sciences, Novena University Ogume, P.M.B. 02, Kwale, Delta State, Nigeria

**Corresponding Author Email:* Imanobetim@gmail.com
**ORCID:* https://orcid.org/0000-0002-6833-4336
**Tel:* +2348138771527

Co-Authors Email: nkolloinnocent@gmail.com; jjjemegha@yahoo.com

ABSTRACT: Heavy metal contamination in wastewater poses a significant threat to environmental and human health. This study investigated the potential of Chlorella sorokiana, a green alga, as a biosorbent for Cu (II) and Zn (II) removal from synthetic wastewater. Batch experiments were conducted at 28°C, pH 6, and a contact time of 60 minutes. Chlorella sorokiana displayed promising biosorption capacity, removing 85% and 75% of Cu(II) and Zn(II), respectively. Further optimization revealed enhanced removal efficiencies at pH 5 (88.5% Cu(II) and 82% Zn(II)) and a biosorbent dosage of 0.5 g/L (90.7% Cu(II) and 87.1% Zn(II)). These findings demonstrate the efficacy of Chlorella sorokiana as a biosorbent for heavy metal remediation. This research highlights the potential of biosorption as a sustainable and environmentally friendly approach for wastewater treatment, offering a promising solution for mitigating heavy metal pollution.

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The surge in global economic development, across both developing and developed nations, has given rise to environmental pollution (Oliomogbe *et al.*, 2023; Ukhurebor *et al.*, 2023a; Brack *et al.*, 2022). It has been recorded that an alarming 90% of untreated wastewater are released to freshwater bodies, rendering them unfit for both human consumption and sustaining aquatic life (Emegha *et al.*, 2023; UN-Water, 2011). Heavy metals have higher atomic weight and density, which is at least five times greater than that of water (Tchounwou *et al.*, 2012). This increase in heavy metals in our environment and water bodies stems predominantly from the pace of global industrialization (Ivanova *et al.*, 2018). Resultantly, the pollution caused by heavy metals emerges as an increasingly critical environmental challenge. Industries engaged in mining, energy production, chemical processing, electroplating, and more, contribute substantially to the discharge of heavy metal-laden wastes, triggering serious consequences for human health and the ecological balance (Ukhurebor *et al.*, 2023b; Wang and Chen, 2006). Existing methodologies aimed at heavy metal removal, encompassing precipitation, filtration, ion exchange, and carbon adsorption, have exhibited notable constraints pertaining to selectivity, cost implications, and the generation of secondary waste (Taha *et al.*, 2023). In response to these limitations,

**Corresponding Author Email:* Imanobetim@gmail.com
**ORCID:* https://orcid.org/0000-0002-6833-4336
**Tel:* +2348138771527

biosorption, leveraging biomaterials as adsorbents, emerges as a compelling approach. Its efficiency, cost-effectiveness, minimal resource requirements, eases of handling and negligible environmental impact position it as a promising solution (Egboduku *et al.*, 2023; Jiménez-Cedillo *et al*., 2013). The paradigm of biosorption harnesses the capability of select biomasses to selectively sequester ions in aqueous solutions (Priya *et al.,* 2022). Microorganisms, known for their expansive surface-volume ratio and effectiveness at absorbing toxic substances, have garnered significant interest. Microalgae species, such as Chlorella vulgaris and Scenedesmus Obliquus, have undergone intensive investigation for their ability in eliminating pollutants (Wilde and Benemann, 1993). These microalgae, with their extensive surface areas and potent binding affinities, prove to be effective biosorbents. Their cell walls contain functional groups like carboxyl, hydroxyl, amines, and sulfates, which play a pivotal role in attracting and sequestering heavy metal ions (Barakat, 2011). In this study, the focus is directed towards employing Chlorella sorokiniana as the biosorbent for the removal of copper and zinc ions from aqueous solutions. Through batch biosorption experiments and the optimization of influencing factors. Hence, the objective of this paper was to assess the suitability of the green algae (Chlorella sorokiana) as biosorbent for the removal of Cu (II) and Zn (II) ions from synthetic wastewater in a batch system.

MATERIALS AND METHODS

Preparation of Synthetic Wastewater containing Zinc and Copper: Industrial wastewater was not collected for this experiment; instead, synthetic wastewater was generated in the lab and used. Synthetic wastewater was chosen for a number of reasons. Such as a controlled composition of heavy metals, isolation of contaminants and lastly, synthetic wastewater enables comparison of result with earlier research which frequently uses standardized synthetic wastewater to maintain uniformity. Cu (II) and Zn (II) stock solutions comprising 1 g/L of the salts $CuSO_4$ and $ZnSO_4$ were dissolved in distilled water and stirred with a magnetic stirrer until the salts were completely dissolved in order to produce synthetic wastewater with known concentrations. 0.1 M HCl and 0.1 M NaOH solutions were applied to the mixture to alter its pH. The appropriate concentrations were produced by diluting the stock solution to specified values between 50 and 200 mg/L. The solution was then placed in a refrigerator until it was needed for the experiment after being filtered to remove any leftover particles.

Nutrient Medium: Algae culture media is made with broth. Bold Basal Media (BBM), a widely used and generally upgraded medium for algal growth, was chosen for this investigation. They are easy to use, and they have all the minerals and trace elements needed for the selected algal strains to flourish. To create the media, 0.0950g of broth is dissolved in 50 ml of distilled water. After that, the medium was sterilized for 20 minutes at 121 °C and 15 psi in an autoclave.

Algae samples selection and collection: There are numerous micro algae species available but for this research work, the green algae Chlorella sorokiana (Figure 1) was selected. The species is well-known for producing a lot of biomasses and being simple to grow. Due to its capacity to thrive in a variety of environmental settings, it is frequently employed in research and commercial applications.

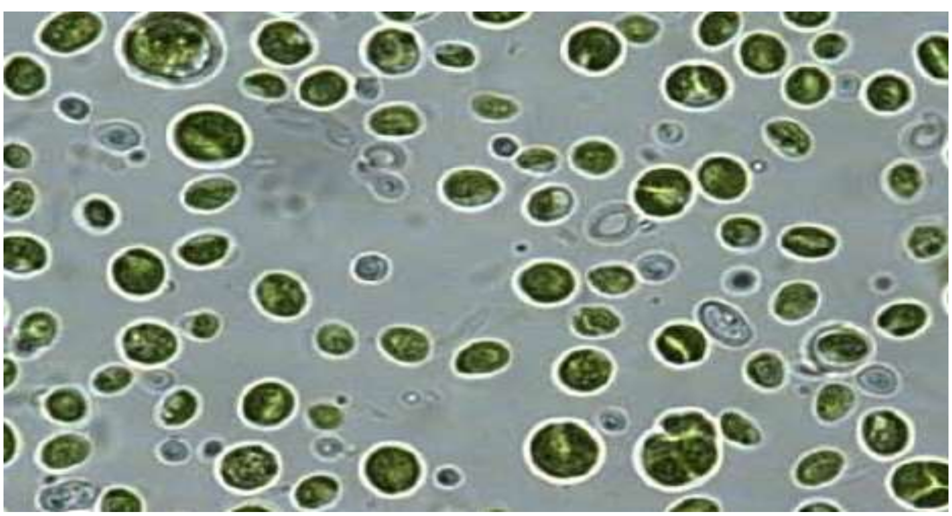

Fig 1: Chlorella Sorokiana strain under the microscope

Microalgae Cultivation: Microalgae from Chlorella spp. was raised in Bold Basal Medium (BBM). This medium was chosen because a variety of different green algal cultures had been grown on it. The stock culture contains 50.6 106 cells per milliliter. The cultivation process started by inoculating the Microalgae cells at a density of 50.6 million (10^6) cells per milliliter (mL) of the cell suspension into each 250 ml Erlenmeyer culture flask in order to achieve a 10% (v/v) inoculum density. The Microalgae cells were cultivated for a day under neutral pH, illuminated with white light from a Phillips source at a specific intensity, to provide them with the light energy they need for growth. The light intensity was set at 90 micromoles (µmol) of photons per square meter per second (m^2s^{-1}). Culture flasks were created on an orbital shaker that rotated at 70 rpm and 28 °C. Algae cultures were routinely inspected and nutrient concentrations were adjusted as required. All contaminants that were present were removed during the cultivation process. The required biomass concentration was reached after 14 days in order to prepare for harvesting. Centrifugation was chosen as the harvesting method. Before being collected, the culture was centrifuged at 4000 rpm for five minutes. The harvested microalgae were rinsed in distilled water to remove any remaining growing material or residue.

Biosorbents preparation: To get rid of extraneous objects, the microalgae were gathered and given four thorough washes with distilled water. After that, they were put in a sterile container to calculate the moist weight. The biomass was then put into an oven that was set to a low temperature of 60^0C. This was left in place until the biomass had dried entirely. Finally, the biomass was crushed and sieved through a 2 mm mesh size sieve before being kept in sterile, clean bottles and having the concentration determined.

Batch Biosorption Experiment. The biosorption study was initiated using an experimental batch process. This method is used to evaluate the ability of biological material to remove pollutants from wastewater or solutions. 1g of Chlorella spp. microalgae biomass was added to each conical flask that contained a stock solution of 10 mg/L of Cu (II) or Zn(II). The pH of the metal solutions was adjusted to the appropriate level (6) using 0.1 N NaOH. Each flask was shaken for 60 minutes at 28°C and 120 rpm using a rotary shaker. After the sorption process, the mixture was filtered through filter paper to separate the metal-adsorbed metal from the flocculated biomass. The number of heavy metals that had been absorbed was measured and assessed. The percentage removal efficiency (% R) of the heavy metal ions by the biosorbents was calculated using.

$$\%R = \frac{C_i - C_e}{C_i} * 100 \quad (1)$$

Where C_e is the metal ion concentration in the aqueous phase after contact and C_i is the initial metal concentration before contact.

Concentration Optimization of Biosorption Conditions: For biosorption to remove heavy metals from a solution, biomass concentration is required. Batch-based experimentation was used. Cu (II) and Zn (II) solutions were kept at a constant concentration of 7 mg/L in order to examine the impact of biomass content on biosorption capability. The biomass concentration was adjusted to 0.25, 0.5, 0.75, and 1.0 (g/l), while the pH was kept constant at the same level as the preceding experiment. The mixes were allowed to come to room temperature and then shaken for three hours at 120 rpm on a rotary shaker. After mixtures were filtered via filter paper, the amount of heavy metal absorbed at different biosorbent dosages was measured with an atomic adsorption spectrometer. Three runs of the experiment were carried out to ensure accurate results.

The pH Optimization of Biosorption Conditions: A batch experiment was used to determine the optimal pH for biosorption. Solutions containing 10 mg/l of Cu (II) and Zn (II) were prepared. The initial pH of the samples was adjusted to be between 3 and 7, respectively, usingH_2SO_4. After that, 0.5g of the biosorbents were weighed and added to each flask. On a rotary shaker set at 120 rpm, the mixtures were shaken for three hours at room temperature. An atomic adsorption spectrometer (AAS) was then used to determine the amount of heavy metal adsorbed at different pH levels after the mixtures had been filtered via filter paper.

RESULTS AND DISCUSSION

The application of Chlorella sorokiana biosorbent for heavy metal biosorption in wastewater: When present in large concentrations in water bodies, heavy metals can result in a variety of ecological and health problems. However, copper and zinc are two common heavy metals that are prevalent in industrial effluents and pose significant environmental risks. Following the biosorption procedure using Chlorella sorokiana, the final concentration of copper in the wastewater was reduced from its original measurement of 10 mg/L to 1.5 mg/L (Figure 2). This translates to a remarkable 85% copper reduction from the effluent. Such a significant decrease in copper heavy metal is evidence of Chlorella sorokiana's viability as a biosorbent for copper removal. The elimination of zinc was also investigated. After a biosorption experiment was conducted by Chlorella sorokiana, the starting concentration of 10 mg/L was decreased to 2.5 mg/L (Figure 2). As a result, 75% of the zinc in the effluent was removed. The higher percentage of removal of copper for Chlorella sorokiana compared to zinc indicates that the algae has a greater affinity for copper ions, making it an excellent choice for the removal of this heavy metal from wastewater.

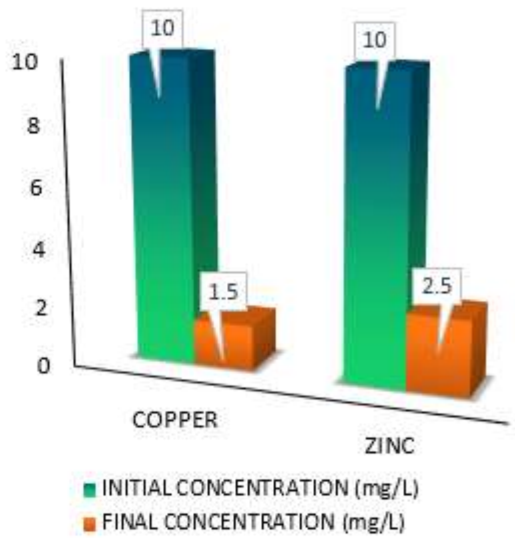

Fig 2 Initial and final concentration of zinc and copper using Chlorella sorokiana

pH optimization of Chlorella sorokiana Biosorbent for zinc and copper extraction: Table 1 demonstrates that the pH optimization by biosorption experiment yielded the greatest results for zinc removal at pH 5, with 82% removal effectiveness. The removal efficiency decreases at pH 4 and 6, while pH 7 had the lowest removal efficiencies. This is due to the fact that zinc hydroxide's solubility rises as pH falls. Since zinc hydroxide is less soluble at pH 5, the biosorbent is more likely to absorb it. With a removal effectiveness of 88.5% (Table 2), pH 5 is also the ideal pH for the elimination of copper. At pH 4 and 6, the removal efficiency drops, whereas pH 3 has the lowest removal efficiencies. Similarly, this is due to the fact that copper hydroxide becomes more soluble as pH drops. Since copper hydroxide is less soluble at pH 5, the biosorbent is more likely to absorb it. Consequently, these results align with the studies conducted by Wanta *et al.* (2020) that employed Chlorella spp. as biosorbents. Generally speaking, the pH range of 4 to 6 was ideal for the biosorption of heavy metals. This resulted from the fact that heavy metal hydroxides' solubility decreases in this pH range, making them more likely to be absorbed by the biosorbent. The results of this study shows that Chlorella sorokiana is a promising biosorbent for removing copper and zinc from wastewater. The removal efficiencies were highest (88.5% for copper and 82% for zinc) at pH 5, likely due to the optimal electrostatic interaction between the positively charged metal ions and the negatively charged functional groups on the algal cell wall. Lower pH values might decrease the solubility of metal ions, potentially hindering their interaction with the biosorbent. Conversely, at higher pH (pH 7), the surface charge of the algae may become more negative, leading to repulsion of the metal ions and consequently lower removal efficiencies.

Table 1: Optimization of pH using Chlorella sorokiana spp for Zinc removal

pH	Initial Concentration	Final Concentration	% Of Heavy Metal Removal
3	10mg/L	3.96mg/L	60.4%
4	10mg/L	3.3mg/L	67%
5	10mg/L	1.8 mg/L	82%
6	10mg/L	2.1 mg/L	79%
7	10mg/L	4.12mg/L	58.8%

Table 2 Optimization of pH using Chlorella sorokiana spp for copper removal.

pH	Initial Concentration	Final Concentration	% Of Heavy Metal Removal
3	10mg/L	3.8 mg/L	62%
4	10mg/L	2.9 mg/L	71%
5	10mg/L	1.15mg/L	88.5%
6	10mg/L	1.9mg/L	81%
7	10mg/L	2.2mg/L	78%

Optimization of Biosorbent dosage for zinc and copper removal: The percentage of heavy metal removal for copper and zinc increased with biosorbent dosage but plateaued at higher dosages due to the biosorbent's saturation with heavy metals. Biosorption site saturation, equilibrium dynamics, and mass transfer restrictions may contribute to this plateau. To remove copper, an optimal dosage of 0.5 g/L of biosorbent is recommended (Table 4). At this dosage, the removal percentage reaches 90.7%. Comparatively, the clearance rate for zinc at the same dosage is slightly lower at 87.1% (Table 3). Nonetheless, the greater affinity copper possesses for the biosorbents accounted for the variation in their clearance rates. Also, at 1.0g/L of biosorbent dosage, there was a decrease in removal efficiency. This could be attributed to limitations in mass transfer or aggregation of algal particles, hindering their interaction with metal ions. The results of the study suggest that Chlorella Sorokiana spp. is effective in removing copper and zinc from wastewater. A dosage of 0.5 g/L of biosorbents is recommended for both metals, making this method economical and efficient for eliminating heavy metals from wastewater.

Table 3: Zinc removal using biosorbent concentrations.

Biosorbent Dosage	Initial Concentration	Final Concentration	% Of Heavy Metal Removal
0.25 g/L	7mg/L	1.92	72.57%
0.5 g/L	7mg/L	0.9	87.1%
0.75 g/L	7mg/L	1.8	74.2%
1.0 g/L	7mg/L	2.43	74.2%

Table 4: Copper removal using biosorbent concentrations.

Biosorbent Dosage	Initial Concentration	Final Concentration	% Of Heavy Metal Removal
0.25 g/L	7mg/L	1.1	84%
0.5 g/L	7mg/L	0.65	90.7%
0.75 g/L	7mg/L	1.5	78%
1.0 g/L	7mg/L	1.7	75.7%

Conclusion: This study significantly contributes to the field of heavy metal bioremediation by demonstrating Chlorella sorokiana's potential as a cost-effective biosorbent for copper and zinc removal. The optimal conditions identified provide valuable insights for optimizing future biosorption processes. Further research on real wastewater application and removal mechanisms will solidify Chlorella sorokiana's role in sustainable wastewater treatment.

REFERENCES

Barakat, MA (2011). New trends in removing heavy metals from industrial wastewater. *Arab. J. Chem*. 4(4): 361–377. https://doi.org/10.1016/j.arabjc.2010.07.019

Brack, W; Barcelo Culleres; D; Boxall, A (2022). One planet: one health. A call to support the initiative on a global science–policy body on chemicals and waste. *Environ. Sci. Eur*.34(1): 1–10. https://doi.org/10.1186/s12302-022-00602-6

Egboduku WO; Egboduku T; Emegha JO; Imarhiagbe O (2023). Biosorbents Derived from Invasive Plants for Environmental Remediation. In Adsorption Applications for Environmental Sustainability, IOP Publsh, 2023. Pp 9.1-9.15 https://doi.org/10.1088/978-0-7503-5598-8ch9

Emegha JO; Oliomogbe TI; Okpoghono J; Babalola AV; Ejelonu CA; Elete DE; Ukhurebor KE (2023). Green Derived Biosorbents for the Degradation of Petroleum Contaminants. In Adsorption Applications for Environmental Sustainability.IOP Publsh. Pp. 12.1-12.20

Ivanova, N; Gugleva, V; Dobreva, M; Pehlivanov, I; Stefanov, S; Andonova, V (2018). Silver Nanoparticles as Multi-Functional Drug Delivery Systems. In: Farrukh, M.A., Ed., Nanomedicines, IntechOpen, London, 71-91.

Jiménez-Cedillo, MJ; Olguín, MT; Fall, C; Colin-Cruz, A (2013). As(III) and As(V) Sorption on Iron-Modified Non-Pyrolyzed and Pyrolyzed Biomass from Petroselinum Crispum (Parsley). *J. Environ. Manag*. 117: 242–252. . https://doi.org/10.1016/j.jenvman.2012.12.023

Oliomogbe, T.I; Emegha, J.O; Ukhurebor, K.E (2023). Microorganism Derived Biosorbent in the Sequestration of Contaminants from the Soil, In Adsorption Applications for Environmental Sustainability. IOP Publsh,. Pp 10.1-10.20.

Priya, AK; Gnanasekaran, L; Dutta, K; Rajendran, S; Balakrishnan, D; Soto-Moscoso, M (2022). Biosorption of Heavy Metals by Microorganisms: Evaluation of Different Underlying Mechanisms. *Chm J.Sci* , 307: 135957.

Taha, A; Hussien, W; Gouda, SA (2023). Bioremediation of Heavy Metals in Wastewaters: A *Concise Review. EJABF*. 27(1): 143–166.

Tchounwou, PB; Yedjou, CG; Patlolla, AK; Sutton, DJ (2012). Heavy Metal Toxicity and the Environment. Experientia. Supplementum, 101: 133–164.

Ukhurebor, K.E; Aigbe U.O; Onyanche RB (2023a). Introduction to the state of the art and relevant aspects of the applications of adsorption for environmental safety and sustainability. In Adsorption Applications for Environmental Sustainability.IOP Publishing Pp 1-1.17.

Ukhurebor KE; Aigbe UO; Onyanche RB. (2023b). The challenges of, and perspectives on adsorption applications for environmental sustainability. In Adsorption Applications for Environmental Sustainability .IOP publish. 2023b

UN-Water (2011). Water Quality and Sanitation. *Water Res.* Pp45: 2985–2995

Wang, J; Chen, C (2006). Biosorption of Heavy Metals by Saccharomyces Cerevisiae: A Review.*J Biotechnol. Adv*.24: 427–451.

Wanta, KC; Hidayat, C; Catherine Miryanti, A; Kristijarti, AP (2020). Biosorption of Copper (II) Ions using Living Chlorella sp. from Aqueous Solution. IOP Conference Series: MSE, 1: 012026. https://doi.org/10.14716/ijtech.v14i1.4995

Wilde, EW; Benemann, JR (1993). Bioremoval of heavy metals by the use of microalgae. *Biotechnol. Adv*. 11(4): 781-812.