Bioremediation of heavy metals from pharmaceutical industry sewages using bimetallic nanoparticles of *Eichorniacrassipes*

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**Abstract.** Biosorption remains one of the most productive and safe technologies that could be received particularly with green algae as biosorbent in the ejection of heavy metals from the effluent. Bimetallic nanoparticles were synthesized from *Eichorniacrassipes* utilizing the mix of both gold chloride and silver nitrate (3:1). Ultraviolet-visible spectroscopy (UV–vis), Scanning Electron Microscopy (SEM), Infrared Spectroscopy (FTIR) and Infrared Spectroscopy (FTIR) were used in the evaluation of the Nanoparticles. The bimetallic nanoparticles were utilized as a biosorbent for particular heavy metals (lead, zinc, copper and manganese) present in a pharmaceutical effluent. The nanoparticles were brought into the effluent at various concentrations (1mg, 5mg and 10mg) and time intervals of 24h, 48h and 72h. The decrease in concentration of each metal was determined utilizing Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-OES). The synthesis of NPs was affirmed by colour change in the blend from light green to lilac after 5min of incubation. The UV-visible spectroscopy adsorption spectrum for bimetallic nanoparticles of *E.craspides* (EC-AuAgNPs) was at 398nm while SEM micrograph shown spherical and cube shaped particles with size ranging from 0.31nm- 1.077nm. The FTIR absorbance peaks were available at 3263.93 (O–H), 2107.11 (-C≡C-), 1631.16 (C═C), 1338 (C–N), and 1034.96 cm⁻¹ (C–O). The most extreme rate decrease acquired for each metal was; Copper (67.25%, 10 mg at 72 h); Zinc (61.13%, 1mg at 72 h); Lead (93.37%, 1 mg at 72 h) and Manganese (69.26, 10 mg at 72 h). The biosorption of heavy metals by EC-AuAgNPs was contact time dependent for all the metals and biosorbent concentration dependent for only copper and manganese.

**Keywords:** Biosorbent, Nanoparticles, Heavy metals, Effluents, Concentration.

1. **INTRODUCTION**

Water pollution is a significant environmental disorder threatening contemporary societies which could prompt to the uptake and buildup of pollutants by edible plants and fish posing a threat to man and animal wellbeing [1]. Consistently, 2 million tons of industrial, agricultural waste and sewage is disposed into the water bodies [2]. The applications of insecticide, nematicides, herbicides and chemical fertilizers have been ensnared in the augmented level of organic pollutants in the environment. The increased human activities and some alteration in the geophysical condition of the
Earth have contributed to release of some inorganic pollutants such as: radioactive materials, metalloids, trace elements and heavy metals [3].

The occurrence of heavy metals in the soil can result in its bioaccumulation in the tissues of plants, when this happens; the developmental growth of the plant is directly affected. Heavy metals adversely disrupts the physiology and metabolic procedure in plants; this can cause biochemical lesion, inhibition of enzyme, retardation in growth, untimely dropping of leaves, disruption of photosynthetic activity and respiration, plant death and chlorosis [4, 5]. However, seaweeds which are macroalgae have been accounted to have probably absorbed heavy metals from the contaminated soil and water, thus rendering them less poisonous [6]. Seaweeds are predominant in the ocean at a global level and they are classified into three based on their colour as green (Chlorophyta), red (Rhodophyta) and brown (Phaeophyta). Massive populates of green seaweeds are terrestrial of fresh water based while both red and brown are mainly aquatic [7].

Biosorbents, the unreceptive uptake of pollutants by biological constituents, particularly got from green macroalgae, alginate derivatives and sea weeds have high adsorptive potential for various metallic ions. Schiewer and Patil 2008 opined that the elimination and/or recovery of metal ions from aqueous solutions are emerging potential option to the present conventional methods. Removal of heavy metals through biosorption has a number of benefits over the conventional techniques, these are cost effective, ability to recover biosorbents, reduction in biological slurry and chemical, and possibility of metal recovery [8, 9]. Biosorbents are generally copious, generally biodegradable, widely abundant, usually biodegradable and inexpensive compared to adsorbents that are industrially synthesized; they also have high affinity to get rid of toxic heavy metals present in industrial effluents [10].

Eichhorniacrassipes (water hyacinth) is a perennial, floating water weeds that are aquatic macrophyte known as nuisance weeds which overruns polluted rivers, lakes and dams [4, 11]. It has a quick propagation rate framing a thick "carpet" on the surface of water bodies. This utilization came to fruition for a couple of reasons, the first being that water hyacinth is so copious. Attempts have been made towards the removal of E. crassipes from water bodies but the resiliency of the weed has either made it difficult or the expense of removal has been excessively costly been excessively. Since the plant has a great demand for nutrient, this has become an advantage in using the metals to remove organic materials from polluted waters thereby making it appropriate [12]. While in most literature it has been referred for its negative effects, it has likewise been deductively appeared to have positive effects in remediating polluted water [13-15]. Water hyacinth has been reported to have amassed and concentrated zinc, nickel and copper in their roots to levels 2000, 1200 and 1300 more than its concentration in a water body (growth media) respectively [14].

Nanoparticles are being used for various purposes, clinical medicines, industry creation, for example, sun powered and oxide fuel batteries for vitality stockpiling to wide fuse into various materials of ordinary utilize, for example, beautifiers or garments [16]. Biomolecules that occur naturally were documented to play an active role in the establishment of nanoparticles with distinct sizes and shapes thus acting as a motivating force for the structuring of greener, safe and ecological conventions for the synthesis of nanoparticles. Bimetallic (gold and silver) nanoparticles of Eichhorniacrassipes were employed as biosorbent for the elimination of some selected metals from pharmaceutical effluent.

2. MATERIALS AND METHODS
2.1. Collection of Sample
2.1.1. Collection and Preparation of Eichhorniacrassipes

Samples of E. crassipes were collected from Ogudu tidal pond in Lagos State, Nigeria. The plants were washed few times with distilled to dispose of contaminations and different epiphytes.
They were then sun-dried for 168 hours and crushed into powder using a blender and later sieved with a fine mesh (laboratory test sieve of 100mm pore size). One gram (1g) of the macroalga was then drenched for 24 h in 100 ml of distilled water and afterward sieved to get the concentrate using filter paper. The extract of *E.crassipes* was later used for synthesis of bimetallic nanoparticles.

### 2.1.2 Collection of Pharmaceutical Effluent

Pharmaceutical effluents were collected at the point of discharge of the effluent into a neighboring water course at Ilorin, Kwara State, Nigeria, which is usually discharged into a neighboring river at Ilorin, Kwara State, Nigeria.

### 2.2 Synthesis of Bimetallic Nanoparticles

Bimetallic oxidewas set up by blending 1mM silver nitrate and 1mM gold chloride in apportion 3:1. The bimetallic nanoparticles of *Eichorniacrassipes* (EC-AuAgNPs) were synthesized by adding 10 ml of *E.crassipes* extract to 90 ml of the bimetallic solution. The mixture was incubated under the Sun and it was observed for change in colour.

### 2.3 Characterization of Nanoparticles

#### 2.3.1 UV–vis Spectroscopy

The biosynthesis of EC-AuNP was first noticed by colour change in the reaction mixture. The mixture was subjected to optical measurements using UV-visible spectrophotometer in way to determine the absorbance. The absorbance peak of EC-AuNP was determined by scanning the solution between 300 and 800 nm.

#### 2.3.2 Energy Dispersive X-ray (EDX) Spectroscopy and Scanning Electron Microscopy (SEM)

The scanning electron microscopy of the synthesized nanoparticles was achieved using Concised FEGSEM 6100Zeiss Ultra Plus Germany at an enhanced voltage of 20.0 kV with secondary electrons in low vacuum mode. The different elemental composition of the EC-AuAgNPs was determined using the same equipment.

#### 2.3.3 Fourier transform infrared (FTIR) Spectroscopy

Fourier transform infrared (FTIR) spectroscopy was carried out to evaluate the functional groups in the bioagent which was used to synthesize EC-AuAgNPs.

### 2.4 Biosorption of Heavy Metals

The EC-AuAgNPs was utilized in the adsorption of four heavy metals (lead, copper, manganese and zinc) from pharmaceutical effluent. The nanoparticles were dried at 70 °C and the dried samples were used as adsorbent various concentrations (1 mg, 5 mg and 10 mg) and time interval (24, 48, and 72 h). The mixture of EC-AuAgNPs and pharmaceutical effluent was put in a shaker for each time span. At the end of the set time, the mixture was filtered to remove the adsorbent from the mixture and the concentration of each metal in the solution was determined using inductively coupled plasma mass spectrometer [17].
2.5 IOP-OES Analysis

The certified standard materials (heavy metal standards) were obtained from DLD Scientific South Africa. Stock solutions of 100 ppm for each element were prepared, stored in the refrigerator at 4 °C. The working solutions of multi-elements standard for the calibration were prepared from the stock solution by appropriate dilutions. The sample solutions were filtered prior to ICP analysis using a 0.45 µm filter disc into ICP vials. The concentrations of heavy metals (lead, copper, manganese, and zinc) were analysed using Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-OES) Perking Elmer Precisely Optima 5300 DV. The sample concentrations were obtained by extrapolating the intensity of the samples obtained from the ICP-OES with the calibration equation \( y = mx + c \) where \( y \) is the sample intensity, \( m \) is the average intercept, \( c \) is the slope, and \( x \) is the actual concentration of the heavy metals in the sample. In this case, the dilution factor of the digested samples was taken into consideration to obtain the actual concentrations in the sample solution. The concentration range for the heavy metals were: Manganese (0.1 to 1.0 µg/mL), Zinc (1-10 µg/mL), Copper (0.05-0.5 µg/mL) and Lead (0.005-0.1 µg/mL) as seen in Table 1 while the calibration curve for each metal is shown in Figure 1.

| Heavy metals | LOD (ng/mL) | LOQ (ng/mL) | R² | Concentration range for the calibration |
|--------------|-------------|-------------|----|-----------------------------------------|
| Mn           | 0.000963107 | 0.003210358 | 0.9989 | 0.1 to 1.0 µg/mL                        |
| Zn           | 0.010628923 | 0.035429744 | 0.9969 | 1 – 10 µg/mL                           |
| Cu           | 0.00033443  | 0.001114765 | 0.9933 | 0.05 – 0.5 µg/mL                       |
| Pd           | 1.0958E-05  | 3.65268E-05 | 0.9972 | 0.005 – 0.1 µg/mL                      |

Figure 1. Calibration curves for the selected heavy metals (Mn, Zn, Cu and Pb).
The instrument’s response for all the heavy metals analyzed were all more than 0.99. However, the blank solution (deionized water) used for the sample preparation was analyzed intermittently with the samples. It was found that the heavy metals concentration was not significant and could not be accounted for as compared to the real samples.

The percentage reduction of zinc, copper, manganese and lead were calculated using the Equation (1):

\[ \text{Percentage Reduction} = \left( \frac{C_0 - C_1}{C_1} \right) \times 100 \]

Where \( C_0 \) = initial concentration of each metal and \( C_1 \) = final concentration of each

3. RESULTS AND DISCUSSION

3.1. UV–visible Analysis of Nanoparticles

A colour change from light green to lilac was noted in the mixture after 5 minutes of reaction, the colour stabilized after 10 minutes (Figure 2). The UV–visible spectrum of the nanoparticles taken revealed absorbance at 398 nm for the EC-AuAgNPs (Figure 3). The colour change of the EC-AuAgNPs observed after 5 min from green to lilac is consonance to the colour change observed by Mostafa et al. (2018) when they synthesized from Gmelina and a colour change was observed after 10 min. [18] likewise documented a change in colour from grey to violet using Persimmon leaf extract.

![Figure 2: Synthesis of Bimetallic Nanoparticles from Eichorniacrassipes: (A) Extract of E. crassipes (B) AuAg (C) Mixture of free Extract and AuAg (D) Colour change at 5 minutes of Incubation.](image)

![Figure 3: Uv-vis Absorption Spectra of EC-AuAgNPs](image)
3.2. Morpohological and Elemental Characteristics of the Nanoparticles

The SEM revealed that nanoparticles were round and cube shaped with size ranging from 0.31 to 1.07 nm (Figure 4). The energy dispersive X-ray of EC-AuAgNPs affirmed the presence of both gold and silver with weight composition of 8.18% and 19.55% respectively (Figure 5). The nanoparticles obtained in this work were spherical and cube shaped with agglomeration, the agglomeration of the nanoparticles was as a result of drying the nanoparticles before characterization. These agglomerated particles were responsible for the large size (0.31nm to 1.07nm) of particles obtained. Veerasamy et al. [19], obtained dispersed nanospherical nanoparticles using gmelina extract with size ranging from 4.07nm to 8nm.

![Figure 4: SEM Image of Bimetallic Nanoparticles of E. crassipies.](image)

![Figure 5: EDX Analysis of Bimetallic Nanoparticles of E.crassippies](image)
3.3 Analysis of Functional Groups

Five different absorbance spectra were available in the bimetallic nanoparticles of *E. crassipies*. The functional groups present are alcohols, alkyne, amide, alkenes, aromatic amines and carboxylase having different spectra of 3263.93 cm\(^{-1}\), 2107.11 cm\(^{-1}\), 1631.16 cm\(^{-1}\), 1338 cm\(^{-1}\) and 1034.96 cm\(^{-1}\) respectively (Figure 6). The absorbance spectrum of EC-AuAgNPs was 398 nm. Shankar *et al.* [20] obtained an adsorption spectrum of 430 nm using neem leaf. The different functional groups present on the bioactive molecules are involved in the synthesis of nanoparticles and its stabilization. The functional groups recorded in this work are consonance with the submission of [21], where aromatic carbonyl group were responsible for the synthesis of nanoparticles from *E. crassipies*. Likewise, [22] reported the role of alcohol, hydroxyl and carboxylic acid groups in the reduction and capping of silver and gold nanoparticles from *Turbinaria conoides*. In this work, these functional groups are not only involved in the synthesis of nanoparticles but as well play a重要 in the absorption of heavy metals. Different functional groups like carbonyl, ether, nitro group, hydroxyl, amino have been reported to participate in the biosorption of metal ions from solution [20]. As discussed by [22], the functional groups that were involved in adsorbing heavy metals by materials of biological origin are: phosphate, ester, hydroxyl, phenolic, carboxyl, sulfhydryl and amino group.

![Figure 6: FTIR Spectra of EC-AuAgNPs.](image)

3.4 Heavy Metal Adsorption By EC-AuAgNPs

The highest overall percentage reduction in concentration of copper was 67.25% at 72 h when 10 mg of the nanoparticles was utilized as biosorbent. The rate of copper uptake by the nanoparticles was both concentration and contact time dependent (Figure 7). The minimum enactment of the nanoparticles as absorbent for copper was with 1 mg at 24 h (12.72%). The highest percentage decrease in zinc by the nanoparticles at 1, 5 and 10 mg was 61.13, 34.39 and 58.12 % respectively all at 72 h. (Figure 8). The biosorption of zinc by the nanoparticles was contact time dependent but not concentration dependent because the highest reduction in concentration of zinc was accomplished with 1 mg of the nanoparticles. The bimetallic nanoparticles from *E. crassipies* at the three concentrations examined had a high affinity for the metallic ions of lead. The percentage reduction in the concentration of lead at 1 mg, 5 mg and 10 mg was 93.37 , 57.06 and 69.16 % respectively at 72 h (Figure 9). The percentage reduction in the concentration of manganese from the effluent, at 1 mg the percentage recorded are 24.44, 51. 71 and 55.28% at 24, 48h and 72h respectively (Figure 10); while at 5 mg the recorded values are 5.23 (24h), 9.60 (48 h), 51.67 % (72 h) and at 10 mg the values are 3.36 (24 h), 38.05 (48 h) and 69.26 (72 h).
Figure 7: Biosorption of Copper from Pharmaceutical Effluent using Bimetallic Nanoparticles of *E. crassipies* at Different Concentrations (1 mg, 5 mg and 10 mg) and Contact Time (24h, 48h and 72h).

Figure 8: Percentage Reduction in the Concentration of Zinc at Different Contact Time and Concentration of Nanoparticles.
4. CONCLUSION

Environmental pollution is the consequence of rapid industrialization and technological advancement and unprecedented increase in populace increase. In this work, the adsorption potential of bimetallic nanoparticles (silver and gold) of *Eichornia crassipies* was investigated for the adsorption of heavy metals (lead, zinc, copper and manganese) at different concentrations (1mg, 5mg and 10mg) and contact time (24h, 48h and 72h). The maximum percentage reduction obtained for each metal was; Copper (67.25%, 10 mg at 72 h); Zinc (61.13%, 1 mg at 72 h); Lead (93.37%, 1 mg at 72 h) and Manganese (69.26, 10 mg at 72 h). This use of *E. crassipies* has proffered solution to the various
problems caused by the sea weed. E crassipies can be grown, harvested and embrace as absorbent to remove heavy metals from different industrial effluent.

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