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MICROBIOME OF *HYOPHILA INVOLUTA* (HOOK.) A. JAEGER IN QUANTIFYING CHROMIUM IMMERSION AND BIOACCRETION

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Abstract

Heavy metal adulteration in aquatic source is difficult to identify and their presence can result in bioaccretion and biomagnification. The present work objectives to assess chromium immersion and bioaccretion by Hyophila involuta (Hook.) A. Jaeger. H. involuta was collected from Pachamalai Hills. The plant was eroded and air dried. The dried thallus was immersed in 50 ml potassium dichromate solution with a chromium concentration of 170µg/ml. Biomass concentration and treatment duration was optimized with biomass ranging from 0.5 g to 3 g and days of immersion from 1 to 3. The thallus showed high levels of chromium immersion. With 24 hours of immersion, 0.5 g biomass removed 17.8 % chromium from water. With increase in biomass concentration there was increase in chromium removal with maximum removal of 82 % when 3 g biomass was used. In day two, 3 g biomass removed 94 % chromium from water and in day 3, biomass concentration from 2 g to 3 g could bring down the detectable range of chromium to zero. Cell wall of bryophytes is thought to have net negative charges that can bio absorb cations. No major studies are there utilizing this ability of bryophytes. Hyophila is common taxa in Tamil Nadu and hence the ability of H. involuta in bioimmersion of chromium can be exploited in water treatment plants.

Key words: Bryophyte, Hyophila involuta, Chromium, Bioimmersion

Introduction

Heavy metals are metal ions having high density and atomic mass. Their lopsided presence can have deleterious effects on the ecosystem. Heavy metal pollution is an area of concern, especially in regions enduring economic development. Rapid urbanization pooled with escalating industrial activities, has led to enlarged heavy metal pollution in water sources. Physio-chemical methods of heavy metal removal, including membrane filtration, reverse osmosis, adsorption on to activated carbon, coagulation etc are not viable due to their high costs. As a result, scientists have been exploring the prospective of low-cost adsorbents for heavy metal amputation. Plants play a key role in combating heavy metal pollution over their ability to pile up and remove pollutants via bioaccretion and phytoremediation (1,2). Among plants, bryophytes are anticipated to be an ecofriendly alternative for heavy metal amputation. Conferring to Nickrent *et al.* (3), bryophytes were the primary green plants to abide in terrestrial ecosystems, and as a result, they had to craft shielding systems counter to the ample sophisticated concentrations of heavy metals extant on land when associated to those in the sea. The presence of these mechanisms empowered many bryophytes to inhabit environments tainted with heavy metals (Shaw *et al.,* 1989) and to accrue momentous quantities of these metals in highly polluted areas, all without suffering any discernible negative influences on their environment (4).

Bryophytes have proven to be effective experimental organisms for probing phytoremediation, owing to their distinguishing morphological and physiological characteristics (5-7). The absence of a root system in these plants implies that they can absorb heavy metals not only from the soil through surface contact but also from atmospheric sources (5,8). The absence of a cuticle layer in bryophytes exposures their cell walls to ionized metals, facilitating their immersion (9,10). This capacity is further enriched by their capability for intense ionic interactions (11) and augmented surface area per unit mass (12). As a result, bryophytes exhibit a rapid response to fluctuations in heavy metal concentrations, surpassing the receptiveness of most vascular plants (7). Conversely, the lack of specialized vascular tissues (1) and slow growth rate (2) of mosses countenance their growth segments to assimilate heavy metal acquaintance over time, providing a more inclusive picture of environmental conditions than would be possible with only current measurements. This is exclusively imperative in areas with swiftly shifting heavy metal levels.

Chromium, a naturally occurring heavy metal, has been extensively exploited in various industrial applications, resulting in momentous environmental pollution and detrimental health consequences. In view of this, the present work aims to assess chromium immersion and bioaccretion by *Hyophila involuta* (Hook.) A. Jaeger.

Materials and methods

Collection and identification of gametophyte

The thallus of *Hyophila involuta* was collected from Pachamalai Hills located within Tiruchirappalli district, where it was found attached to soil and stone substrates. The plant was carefully collected, washed and air dried. Identification of the gametophytic plant was accomplished by combining morphological observations with information gathered from standard literature resources.

Bioaccretion of Chromium

Studies were conducted to determine bioaccretion of hexavalent chromium by the bryophyte. Potassium dichromate ($K_2Cr_2O_7$) was used as the source of hexavalent chromium. Preparation of standard graph and assay of chromium was done as per Eaton *et al*. (13).

Preparation of Primary Stock solution

Stock solution of Potassium dichromate ($K_2Cr_2O_7$) was prepared by dissolving 141.4 mg of $K_2Cr_2O_7$ in 50 ml distilled water and making the volume up to 100 ml in a standard flask. This solution contains 500µg / ml of Chromium.

Preparation of Working Stock solution

The primary stock solution was diluted 1:100 with distilled water, yielding a solution with a chromium concentration of 5 µg/mL.

Preparation of diphenylcarbazide solution

250 mg of 1,5-diphenylcarbazide was dissolved in 50 ml of acetone. The solution was stockpiled in an amber coloured bottle.

Colorimetric method for quantifying chromium

2 mL of the sample was transferred to a beaker, and 120 mL of distilled water was added. The pH of the solution was attuned to 2 ± 0.5 using 0.2 N H_2SO_4. The solution was diluted to a final volume of 100 mL using distilled water. 2 mL of diphenylcarbazide solution was auxiliary to the sample solution. The mixture was meticulously assorted and endorsed to stand for 10 minutes. The absorbance of the solution was restrained at 540 nm. A blank solution was prepared by swapping the 2 mL sample with 2 mL of distilled water, and the same procedure was followed.

Bioaccretion studies

The gametophyte was washed well to eradicate soil particles and dried. It was used as the plant material. The thallus was immersed in 50 ml potassium dichromate solution with a chromium concentration of 170µg/ml. Biomass concentration and treatment duration was optimized with biomass ranging from 0.5 g to 3 g and days of immersion from 1 to 3. Chromium immersion was studied using 1,5-diphenylcarbazide reagent.

Results and discussion

The vegetative gametophyte was scrutinized under a stereo zoom microscope and identified with the aid of standard literature and online resources revealing a characteristic leafy, erect morphology. The leaves exhibited contortion upon desiccation and wide spreading when moist, with characteristic oblong-lingulate shape, concave form, and clasping base. The costa was strong, tapering, and smooth, with a percurrent pattern. Based on these features, the specimen was identified as *Hyophila involuta* (Figure 1).

Figure 1- *Hyophila involuta*

Certain bryophyte species exhibit metal tolerance, enabling them to withstand elevated levels of heavy metals that are toxic to other species. In the present study, *H. involuta* showed high levels of bioimmersion of chromium. With 24 hours of immersion, various concentration of biomass showed considerable immersion of chromium (Figure 2). 0.5 g of biomass could absorb 1.120 mg of chromium that account for 17.8 % removal from the solution. With increase in biomass concentration, the percentage removal also increased, with 3 g biomass, 6.87 mg chromium removal was noted. Calero *et al.* (14) investigated the bioimmersion kinetics of heavy metals utilizing olive by-products. Analysis of the results revealed that the pseudo-second-order model is the most suitable for modelling the biosorption of the three metal ions. For the three metal ions, an increase in initial metal concentration leads to an enhancement in maximum biosorption capacity and initial biosorption rate. Temperature exhibits a negligible effect on the biosorption capacity of Cd(II) and Cr(III). However, for Pb(II), temperature plays a crucial role, with a pronounced increase in biosorption capacity observed between 25°C and 40°C.

On day two, 0.5 g biomass absorbed 62.5 % total chromium from solution and the percentage of removal increases with higher biomass concentration (Figure 3). At 3.0 g biomass concentration, 94.3 % chromium removal was noted. Research on the biosorption capabilities of bryophytes is limited. Nevertheless, a study by Sari *et al*. (15) examined the biosorption of Cd(II) and Cr(III) ions from aqueous solutions using *Hylocomium splendens* moss. The saturation capacity (qm) of *H. splendens* biomass was determined to be 32.5 mg/g for Cd(II) ions and 42.1 mg/g for Cr(III) ions.

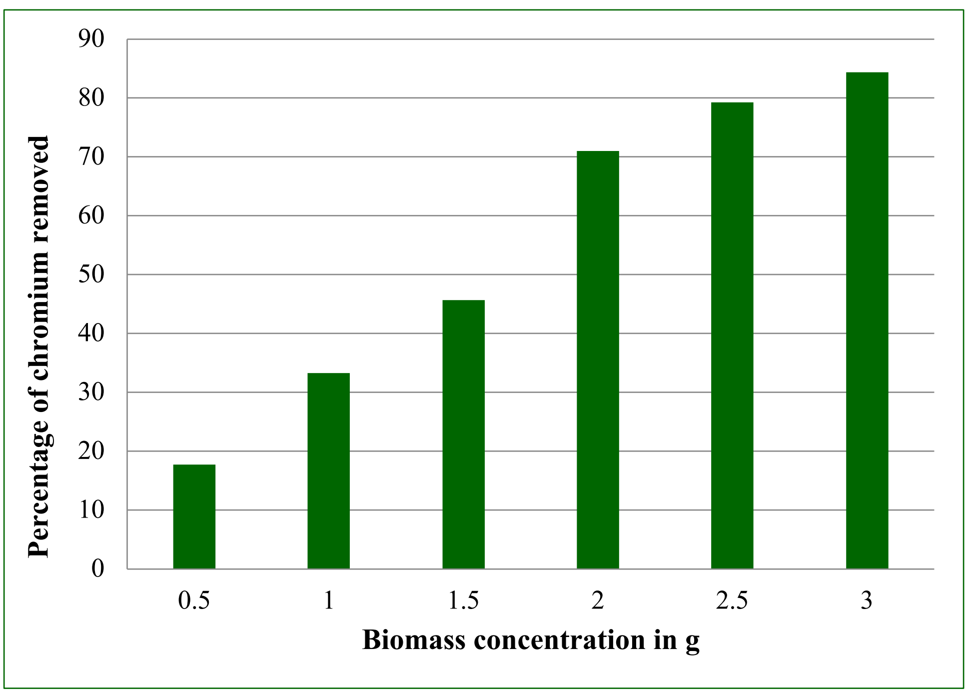

Figure 2- Chromium immersion after 24 hours of treatment

After 144 hours of treatment, biomass concentration from 2 g to 3 g could bring down the detectable range of chromium to zero. This indicates the potential of dried thallus of *Hyophila involuta* in removing chromium from water. The plant can be utilized for field studies towards removal of chromium from spent water.

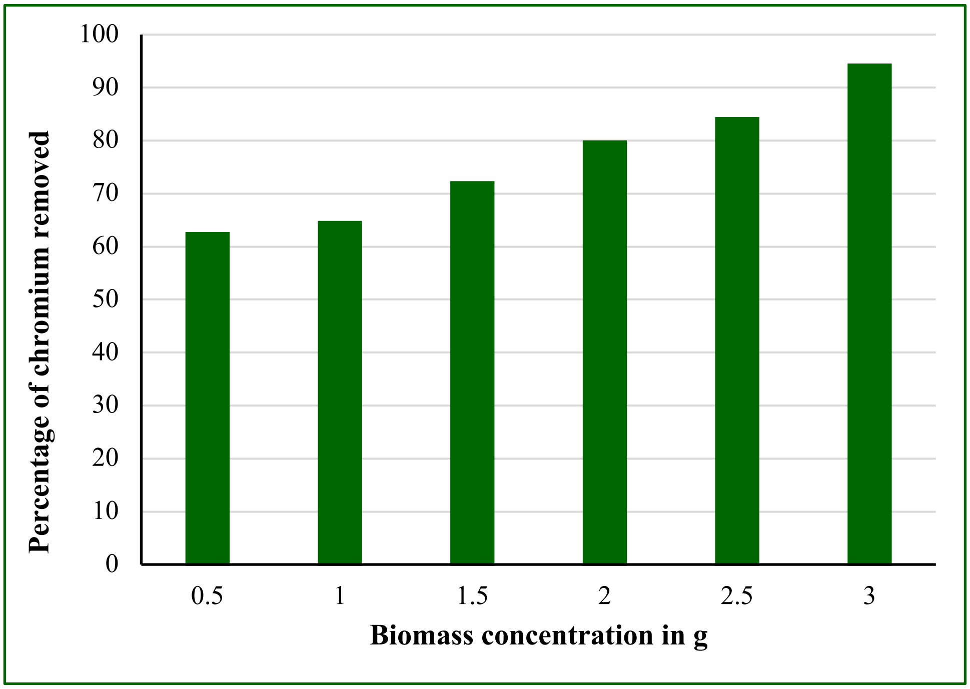

Figure 3- Chromium immersion after 120 hours of treatment

Conclusion

From the above discussions on *Hyophila involute* showed significant results in Chromium immersion. The thallus showed high levels of chromium immersion. With 24 hours of immersion, 0.5 g biomass removed 17.8 % chromium from water. With increase in biomass concentration there was increase in chromium removal with maximum removal of 82 % when 3 g biomass was used. In day two, 3 g biomass removed 94 % chromium from water and in day 3, biomass concentration from 2 g to 3 g could bring down the detectable range of chromium to zero. Cell wall of bryophytes is thought to have net negative charges that can bio absorb cations. No major studies are there utilizing this ability of bryophytes. *Hyophila* is common taxa in Tamil Nadu and hence the ability of *H. involuta* in bioimmersion of chromium can be exploited in water treatment plants.

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  Selva Ganapathy Muthusamy: Conceptualization, Methodology, Resources, Data curation, Investigation, Formal analysis, Writing - original draft, Validation.

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