

Accumulation of chromium (VI) from aqueous solutions using water lilies (*Nymphaea spontanea*)

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Abstract

This study describes an investigation using tropical water lilies (*Nymphaea spontanea*) to remove hexavalent chromium from aqueous solutions and electroplating waste. The results show that water lilies are capable of accumulating substantial amount of Cr(VI), up to 2.119 mg g⁻¹ from a 10 mg l⁻¹ solution. The roots of the plant accumulated the highest amount of Cr(VI) followed by leaves and petioles, indicating that roots play an important role in the bioremediation process. The maturity of the plant exerts a great effect on the removal and accumulation of Cr(VI). Plants of 9 weeks old accumulated the most Cr(VI) followed by those of 6 and 3 weeks old. The results also show that removal of Cr(VI) by water lilies is more efficient when the metal is present singly than in the presence of Cu(II) or in waste solution. This may be largely associated with more pronounced phytotoxicity effect on the biochemical changes in the plants and saturation of binding sites. Significant toxicity effect on the plant was evident as shown in the reduction of chlorophyll, protein and sugar contents in plants exposed to Cr(VI) in this investigation.

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1. Introduction

The application of bioremediation in treating heavy metals in wastewater has gained much attention lately. The discovery of metal hyper-accumulating properties in certain plants suggests the potential of using plant-based system for treatment of wastes containing metals (Ensley, 2000). Various reports have demonstrated the ability of aquatic macrophytes especially wetland plants such as *Typha* sp., *Phragmites* sp., *Scirpus* sp., *Leersia*

sp., *Juncus* sp. and *Spartina* sp. in reducing the levels of heavy metals in polluted waters (Vajpayee et al., 1995; Neralla et al., 1999; Lim et al., 2001; Deng et al., 2004; Weis and Weis, 2004; Shankers et al., 2005).

The use of aquatic plants such as water hyacinths (*Eichhornia* sp.), duckweeds (*Lemna* sp.) for wastewater treatment has been well documented (Perry and Robinson, 1997; Reed et al., 1988). These aquatic plants are, however, known to be rampant colonizers that often interfere with crops on irrigated land and fish production. Water lilies were undertaken for this study as they are aesthetically appealing to provide landscape-pleasing environment and they are potentially hyper-accumulators of nutrients and metals as they have extensive roots

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and provide large surface area for the biofilms formation and thus enhance the microbial activities. To date there have been limited studies on the use of water lilies for wastewater treatment. This paper reports a study undertaken to assess the effectiveness of water lilies to remove Cr(VI) from aqueous solutions and electroplating waste and to assess the effect of Cr(VI) on some of the plant biochemical processes. Cr(VI) is studied as this is one of the most commonly occurring toxic pollutants present in wastewaters discharged from electroplating, dye and pigment manufacturing, wood preserving and leather tanning industries. In addition to being highly toxic, Cr(VI) is mobile, and has a long residence time in surface water and groundwater; it poses health risk to humans and animals and impairs the development and growth of plants (Kleiman and Cogliatti, 1998; Chandra and Kulshreshtha, 2004; Shankers et al., 2005). Cu(II) commonly occurs together with Cr(VI), especially in electroplating wastewaters. Thus its effect on Cr(VI) accumulation by water lilies is investigated.

2. Materials and methods

2.1. Materials

The night bloomer tropical water lilies (*Nymphaea spontanea*) were grown in the glass house of Universiti Putra Malaysia, Selangor, Malaysia. Mature plants with approximately the same size and weight, aged between 7 and 8 weeks old were used in this investigation unless otherwise stated. Prior to experimentation, the plants were washed thoroughly with tap water; damaged and decayed parts were removed.

Cr(VI) and Cu(II) solutions were prepared from A.R. grade $K_2Cr_2O_7 \cdot 5H_2O$ and $Cu(NO_3)_2 \cdot 3H_2O$, respectively. Electroplating waste was collected from an electroplating factory in Seremban, Malaysia. Analysis of the waste showed that it contained 7000 mg l^{-1} Cr, 6050 mg l^{-1} K, 7500 mg l^{-1} Na and 12850 mg l^{-1} sulphate.

2.2. Methods

2.2.1. Study on metal uptake

Cr(VI) solutions at concentrations of 1, 2.5, 5 and 10 mg l^{-1} were used in the study to assess the metal accumulation of water lilies. A binary metal solution with Cr(VI) to Cu(II) ratio of 5:1 was prepared for the study on the effect of Cu(II) on Cr(VI) uptake by the plants as these metals commonly occur together in electroplating wastewaters. In the treatment of electroplating waste, the waste solution was diluted and spiked with Cu(II) to yield a solution containing 2.5 mg l^{-1} Cr(VI) and 0.5 mg l^{-1} of Cu(II). The pH of the waste solution which measured 2.0 initially was adjusted to 6.0 by adding 0.5 M NaOH prior to experimentation.

All the experiments were conducted for a period of seven days as preliminary tests showed that water lilies were able to grow well and stay healthy without nutrient supply in water for ten days. Addition of nutrients would result in algae growth that would interfere with metal uptake by water lilies. Each test plant was placed in a 15-l plastic container containing 10 l of metal solution. The pH of the solution was adjusted to 6.0–6.5 which was considered suitable for water lilies; previous studies have shown that most of the aquatic plants grew in environment where pH ranged between 5.5 and 7.5 (Sen et al., 1987; Outridge and Noller, 1991; Kleiman and Cogliatti, 1998; Lim et al., 2003). There was no change in solution pH at the end of the experiments. The experiments were carried out at room temperature ($30 \pm 2^\circ\text{C}$) in the glass house of Institute Bioscience of Universiti Putra Malaysia. Plant control free of heavy metals and metal control free of plants were established for each set of experiment. All the experiments were performed in triplicates.

The concentration of metal ions in solution was determined before the commencement of the experiment. Solution samples were collected periodically from day zero to day seven from each plastic container for the determination of metal concentrations using an inductively coupled plasma-atomic emission spectrometer (ICP-AES, Perkin Elmer P1000). Correction for evaporation was made daily by adding tap water to the mark in the plastic container.

The test plants were harvested periodically at 1, 3, 5 and 7 days after exposure to Cr(VI). They were separated into leaves, petioles and roots, washed with tap water followed by deionized water. The leaves were used for the determination of total chlorophyll using Knudson et al.'s method (1977) which is proven to remove more than 99% of the total chlorophyll obtained from healthy leaves. The materials were then oven dried at 80°C until constant weight was obtained. The dried samples were then ground and kept in desiccators before being weighed for the analyses of metal, protein and sugar concentrations. Duplicate portions of ground plant material (0.25 g) were weighed for analysis of metal concentrations while 0.1 g plant materials were used for protein and sugar analyses.

Pickford's wet ashing method (1989) was adopted for the analysis of metal contents in plants where the samples were digested in a mixture of HNO_3 and $HClO_4$ in the ratio of 4:1. Analysis of metals using NBS standard reference material No. 1571 (orchard leaves) has been carried out in this laboratory to check the accuracy of the method and good agreement between certified and experimental values were obtained (Lee et al., 1983). The method of Lowry et al. (1951) and the phenol-sulphuric acid method (Dubois et al., 1956) were adopted for protein estimation and sugar assay, respectively.

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