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Use of *Cyperus alternifolius* and *Eichhornia crassipes* for removing heavy metal from shrimp farm effluent in wetlands.

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Abstract. Water quality deterioration by nutrient enrichment from shrimp farming has been a concern in recent years, especially heavy metal in water resource. Phytoremediation technology using aquatic weed in wetlands increasingly applied to remediate the eutrophic waters. Objectives of this research were to evaluate the effectiveness and potential of both *Cyperus alternifolius* and *Eichhornia crassipes* in lab scale constructed wetland in reducing heavy metal as iron (Fe) and copper (Cu) using phytoremediation technologies in 1, 3, 7, 10, and 14 days. The results showed that the survival rates (SVR) of both aquatic plants in the experiment was the highest (100%) after 14 days of treatment. As a result of Zn, Fe, Cu concentration in control and experimental plot tended to decrease from start time to 14 days, concentration of iron were 0.0 mg/l in both the experiment and control plot at 14 days, the experiment plot showed effectively used in reducing a concentration of zinc and copper between the day 1 and day 14. To conclude, the results indicated that both plants could be used in wetlands engineered for iron and copper remediation.

1. Introduction

Vietnam is the leading producer of shrimp in the world with a high production per year, there are many kinds of shrimp as black tiger shrimp (*Penaeus monodon*), whiteleg shrimp (*Litopenaeus vannamei*), and giant freshwater shrimp (*Macrobrachium rosenbergii*). In Vietnam, shrimp farming has rapidly expanded in recent years. Consequentially, the uneaten feed and excreted waste caused a number of water quality problems. The wastewater from shrimp farming is responsible for nutrient enrichment in receiving waters surrounding shrimp farm. Conventional wastewater treatment systems are costly to install and operate for treating the the wastewater from shrimp farming. Therefore, the use of bioremediation is an interesting alternative to solve this issue.

The principles of phytoremediation systems for cleaning up stormwater include: (a) identification and implementation of efficient aquatic plant systems; (b) uptake of dissolved nutrients including nitrogen, phosphorous, and metals by the growing plant; and (c) harvest and beneficial use of the plant biomass produced from the remediation system[1]. Macrophytes are commonly used in artificial wetland constructed for treatment of wastewater domestic sewage treatment in many countries. This plant is capable of removing organic matter, suspended solids and nutrients such as nitrogen and phosphorus from water. There are many species of macrophytes such as umbrella sedge or umbrella palm (*Cyperus alternifolius*) and water hyacinth (*Eichhornia crassipes*). First of all, umbrella sedge is



a plant growing in wetlands. It is a multi-year-old and grows in humid soil and marshy areas. The plant has strong underground root, and can be easily multiplied using seeds or pieces of the plant. The advantage for the bioremediation using *Cyperus alternifolius* is that it eliminates excess nutrients of the wastewater [2].

In a literature, *Cyperus alternifolius* was employed to treat municipal wastewater from Yazd city (center of Iran) by constructing vegetated wetlands. Also *Cyperus alternifolius* can grow well on any form of nitrogen and as they can develop a deep and dense root system [3]. This plant is selected as part of the wetland flora to reduce metal pollution in water resource, affected industrial activities[4]. Water hyacinth can grow well in water with concentration phosphate (P) levels above 20 ppm [5]. Water hyacinth (*Eichhornia crassipes*) have the optimal temperature for the plant growth from 20 to 30 °C, Water hyacinth produces new seedlings at higher temperatures and the reproductive mechanism of water hyacinth is not affected by plant size [6]. Water hyacinth can change its living state based on water characteristics or with low salinity [7]. Especially at low salinity of 0.2‰ will limit the ability of hyacinth development [8]. Water hyacinth can remove toxic heavy metals such as cadmium (Cd), lead (Pb), mercury (Hg) in a metal-containing solution without nutrients in the winter [9]. The values of the concentration factors in the roots tended to decrease, and the root concentration was higher at the leaf tips [10]. Results of [11] show that water hyacinth can reduce chemical oxygen demand (COD) and biological oxygen demand-5 days (BOD₅) by 79% and 86%, respectively. A research [12] illustrates that water hyacinth is a good absorption of Cd and Zn. In Malaysia, water hyacinth is used to treat copper (Cu), cadmium (Cd), lead (Pb) and zinc (Zn) for four days. Water hyacinth can be considered as a treatment for heavy metal content in livestock wastewater, rather than pollutants in the water environment [13].

The present study seems part of management options in the near future for enhancing the water quality from wastewater of shrimp farming by natural and artificial wetland technology. The objectives of this study were to evaluate Zn, Fe and Cu removal from this wastewater, compared to the control and QCVN08 BTNMT: 2015. Second one is that check the growth performance of *Cyperus alternifolius* and *Eichhornia crassipes* live in a lab scale constructed wetland.

2. Materials and Methods

2.1. Experimental set-up

The lab-scale artificial wetland for treatment the aquaculture wastewater was constructed by glass tank sizes as 1000 × 450 × 600 mm (length × width × height, respectively), equipped with a circulation system, with the height of water immersing the plants is of 500 mm. The volume of the submerged part in glass fish tank is around 98 L and the volume of the plastic tank with circulating pump around 80 L. This tank was divided into two compartments for growing *Cyperus alternifolius* and *Eichhornia crassipes* separately. Each compartment comprised of three layers of (a) bottom layer, 10 kg lava stone, (b) middle layer, 10 kg large gravel, and (c) top layer, 15 kg small gravel.

The two lab-scale wetlands was set up at the laboratory of Nguyen Tat Thanh University, each with a control and a treatment plot (Figure 1). Sample polluted water was selected for this study at the freshwater shrimp farmers, cultivating giant freshwater shrimp along the Saigon River, Ho Chi Minh. *Cyperus alternifolius* had fresh weight 163.66±20.74 g, length of plant 57.5±0.71 cm, length of root 28.00±2.83 cm and *Eichhornia crassipes* had fresh weight 98.81±15.29 g, length of root 20.25±1.06 cm, grown in the lab-scale set up at day 0, while no plant was experiment at start time, while no plant was maintained in the control plot. During experimental time, water samples were collected at day 1, 3, 7, 10, and 14. The collected samples were analyzed for the nitrogen and phosphorus content as NO₃-N, NH₄-N, PO₄-P. Specially, the control and the treatment plot were daily analyzed for water quality parameters, including temperature (°C), pH, electrical conductivity (EC), and total dissolved solids (TDS).

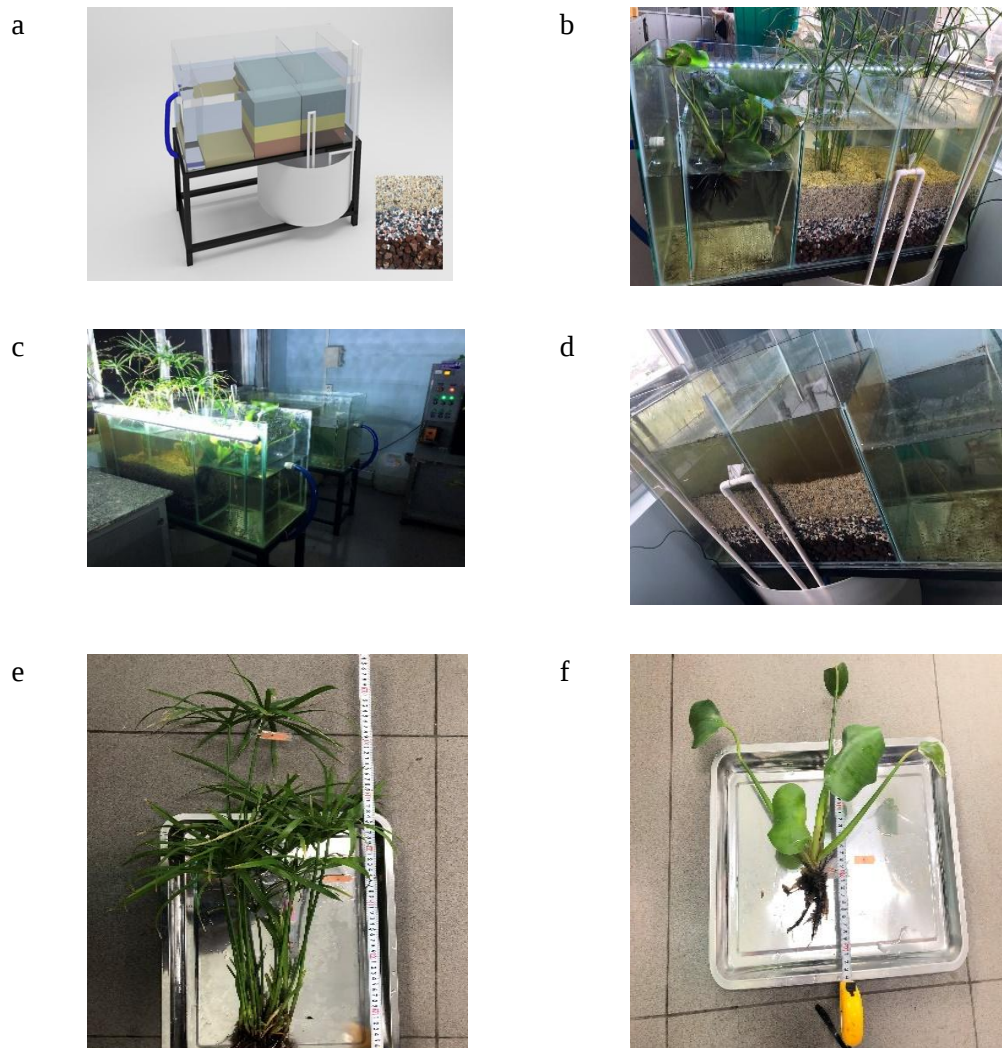


Fig. 1. Design of the lab-scale wetlands in this study (a-d), for treating shrimp farming effluent by using *Cyperus alternifolius* (e) and *Eichhornia crassipes* (f).

2.2. Samples collection and analysis

Samples were collected and stored with 2 L plastic bottles placed in a refrigerator at 4 °C with preservation as appropriate at the laboratory of Nguyen Tat Thanh University. Activities of analysis were conducted at this laboratories and prior to filtration. The pH, conductivity, total dissolved solids, and temperature of water samples were determined by an integrated measurement instrument (Mi805 Milwaukee Instruments, Colorado, USA). In addition, the dissolved oxygen (DO) were determined by using Milwaukee MW 600 Dissolved Oxygen Meter, Colorado, USA. Hanna Instruments HI83399-02 is a multiparameter photometer for measuring key water quality parameters as zinc (Zn), iron (Fe) and copper (Cu) in this study. Moreover, copper was further analyzed following the EPA method and iron by TPTZ method.

2.3. Analysis of results

Mean values and standard errors of water quality parameters were calculated from replicates, to be done within each treatment on one sampling time to understand differences between the control and

the experiment. This analysis of the results is following several standards, namely National Standards of Vietnam: (QCVN 08-MT: 2015), (QCVN 01 - BYT: 2009), and WHO Guidelines (World Health Organization 2006).

3. Results and Discussions

3.1. The growth performance of *Cyperus alternifolius* and *Eichhornia crassipes*

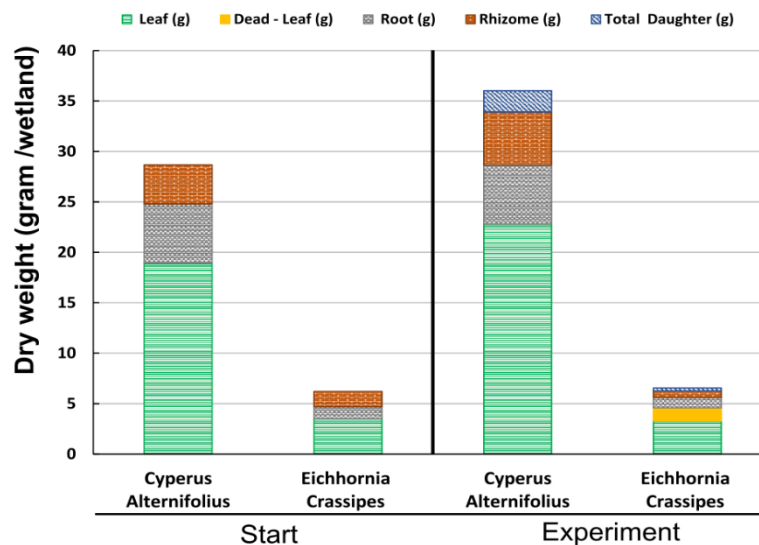


Fig. 2. The growth performance of *Cyperus alternifolius* and *Eichhornia crassipes*

Dry weight of *Cyperus alternifolius* and *Eichhornia crassipes* in wetland were measured and shown in Figure 2. After 14 days of experiment, we can see that the fresh weight and dry weight of the *Cyperus alternifolius* increased from 163.66 g to 185.28 g and in the dry weight from 28.68 g to 36.02 g when compared to the control group we could a marked growth of plants in the experimental environment. The same was true for *Eichhornia crassipes* after the end of the experiment where the fresh weight increased from 80.92 g to 101.12 g as well as the dry weight from 4.76 g to 5.36 g and increased compared to the control group and achieved a 100% survival rate.

3.2. Zn, Fe, Cu concentration reduction

This study inherits the research content [17]. Summary tables (Table 1) are presented both the primary publication [17] and data of the reduction of Fe and Cu are described below. According to the publication [17], pH average value recorded for the control was 7.92 and for the experiment was 8.08.

- The average dissolved oxygen concentration (DO) recorded for the studied water samples of this study was 6.51 mg/L at the control plot and 5.58mg/L at the experiment compartments.

- At start time of this study was 7.15 ± 0.21 mg/L of $\text{NO}_3\text{-N}$, and the water quality was higher than the limit allowed by A2 in (QCVN 08: 2015), regulation $\text{NO}_3\text{-N} \leq 5$ mg/L for water supply and conservation of aquatic ecology. After 14 days, the concentration of $\text{NO}_3\text{-N}$ of control was higher than experiment plot, and if surface waters often contains $\text{NO}_3\text{-N}$ less than 1 mg/L of nitrate $\text{NO}_3\text{-N}$ [18], then it will be not beneficial for aquatic life.

Results presented that Fe, and Cu concentration in control and experimental plots tended to decrease from day 0 until day 14 because of phytoremediation potential of some heavy metals by aquatic plants [19], [20]. The growth of plant weight of *Cyperus alternifolius* and *Eichhornia crassipes*

in during the experiments of the wastewater treatment system is technically important by achieving a 100% survival rate. So, the faster of both plant adapt and grow was very important, the more nutrients removed from the wastewater, showed the effectiveness of a lab scale constructed wetland with lava filter.

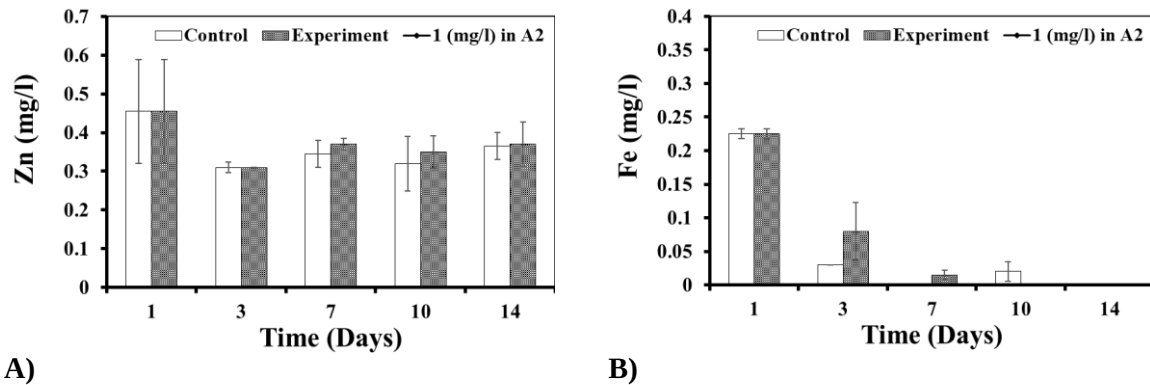


Fig. 3a-b. Concentration of Zn and Fe (mg/L) in water samples in this study.

The Zn levels for all 14 days of treatment are shown in Figure 3 with reducing from day 0 to day 14. The Fe concentration of the first day of both control and experimental plots was 0.225 ± 0.007 mg/L iron is an essential element and plays an important role in aquatic weed and aquatic animals in water resources. Firstly, iron is an integral part of the structure of the haemoglobin molecule, which transports oxygen in the blood in many vertebrates and invertebrates. Secondly, iron plays an important role in the photosynthesis of plants. Fe concentration is listed in the water quality in QCVN, Vietnam[15] and in the TCVN of Industrial Waste water and Discharge Standard[16]. The Fe levels for all 14 days of treatment are shown in Figure 3b. Fe concentration of the first day of both control and experimental plots was 0.225 ± 0.007 mg/l. Concentration of Fe in both plots tended to decrease at 3 days and continuing to the value of 0.0 mg/l at the end of 14 days. Especially at 3 days, the concentration of Fe in the experiment was higher than the control plot. Change of Fe concentration was affected by growing *Cyperus alternifolius* and *Eichhornia crassipes*.

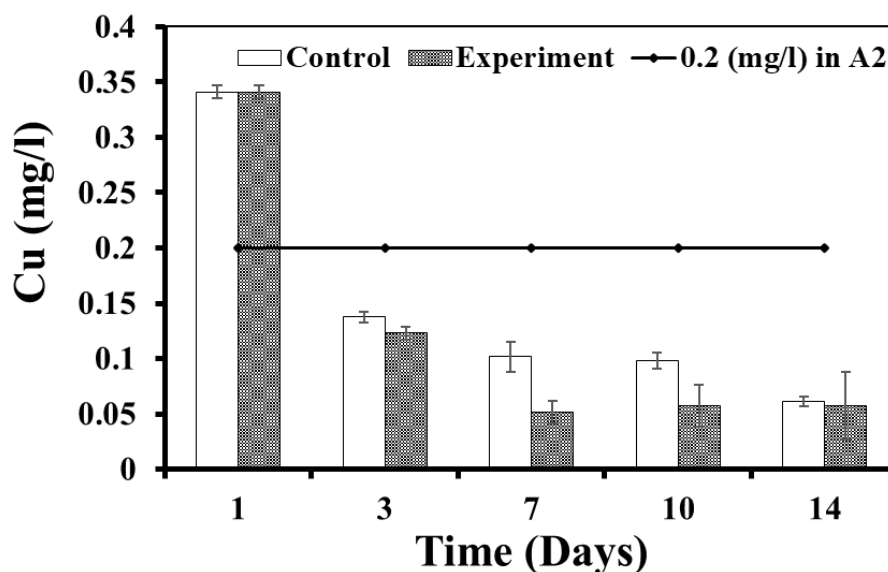


Fig. 4. Concentration of Cu (mg/L) in water samples in this study.

The Cu concentration of the first day of both control and experimental plots was 0.341 ± 0.005 mg/l. Cu concentration in control and experimental plots tended to decrease from 3 days until the end of 14 days, Cu concentrations in experimental plot decreased to 0.0515 ± 0.1 mg/l and 0.1015 ± 0.1 mg/l in the control plot at 7 days but there was a slight increase at 10 days of the experimental plot from 0.0515 ± 0.1 mg/l to 0.0575 ± 0.2 mg/l.

4. Conclusion

This study showed that two aquatic macrophytes as *Cyperus alternifolius* and *Eichhornia crassipes* had a high survival rate and an increase of their dry weight. So, this lab scale constructed wetland with a lava filter could be effectively used in reducing heavy metal of zinc, copper and iron levels of wastewater from shrimp farms. If we want an eco-friendly future without water pollution, we need to reduce heavy metal in water resource by developing new green technology and eco-friendly, for example, building eco-gardening and protecting natural wetland is based on a global approach to the biocontrol solutions. Particularly on large wetland, we need biological treatment of wastewater, focusing on the phytoremediation potential of selected aquatic plants of *Cyperus alternifolius* and *Eichhornia crassipes*. In this study, the concentration of several metals can be reduced to safe levels, such as to <0.6 mg/L (zinc), to <0.25 mg/L (iron), and to <0.15 mg/L (copper), where the limit is 1, 1, and 0.2 mg/L, respectively.

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