# The influence of pollutants on plant growth and treatment efficiency of horizontally-constructed wetlands

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Received 9 August 2022; accepted 26 October 2022

#### Abstract:

Constructed wetlands (CWs) have been widely used for wastewater treatment due to advantages such as low cost and easy operation. However, pollutant concentrations directly affect treatment efficiency by inhibiting plant and microbial growth. This study aims to evaluate the relationship between pollutant concentrations, tolerance of plants, and treatment efficiency of horizontal subsurface flow (HSF) CWs receiving synthetic wastewater. The results showed that *Cyperus alternifolius* L. grew well in conditions with concentrations of chemical oxygen demand (COD) and NH<sub>4</sub><sup>+</sup> up to 75 and 125 mg/l, respectively, and pH 5-8. The lowest treatment efficiency was 56.7% at an initial COD concentration of 1000 mg/l. There were several dead plants in the CWs with a NH<sub>4</sub><sup>+</sup> concentration of 200 mg/l. When the initial COD and NH<sub>4</sub><sup>+</sup> concentrations were in the range of 500-750 and 75-125 mg/l, respectively, the treatment efficiencies of organic matter (COD) and NH<sub>4</sub><sup>+</sup> were 82.5-85.1 and 31-69.7%, respectively. The pH value was stable at a neutral level during the treatment process. The concentration ranges of COD (500-750 mg/l) and NH<sub>4</sub><sup>+</sup> (75-125 mg/l) were the optimal thresholds for pollution control while also ensuring normal plant growth. This study serves as a basis for considering wastewater characteristics in the design of HSF CW systems in practice.

Keywords: constructed wetlands, Cyperus alternifolius L., pollutant factors, wastewater treatment.

Classification numbers: 3.1, 5.1, 5.3

### 1. Introduction

Together with fast socio-economic development, the problem of water pollution has raised concerns. Therefore, the efficient removal of nutrients and organic matter in wastewater before disposal is necessary. CWs utilise plants, substrates (e.g., soil, sand, and gravel), and microbial communities in the rhizosphere to transform and remove organic and/or inorganic pollutants [1-3]. With several advantages such as low maintenance cost and lack of chemical treatment, wetland technology has the proven ability to be an eco-friendly and effective wastewater treatment method [4, 5]. There are two main parts of an artificial wetland: substrates (or filter materials) and vegetation. Substrates can improve treatment capacity and stabilise the performance of CWs [5]. Substrates also provide reaction interfaces for various physical, chemical, and biological processes, attachment surfaces for microorganisms, support plant growth, and create favourable hydraulic conditions for sewage flow [6]. Plants play significant roles in wetland treatment processes [2, 3].

With the ability to release oxygen from their roots and root exudates, plants can enhance both aerobic and anaerobic degradation to reduce organic matter, ammonium, and other contaminants in wastewaters [1, 7]. Common wetland plants such as reed (Phragmites australis), umbrella plant (Cyperus alternifolius, Cyperus papyrus), and water spinach (Ipomoea aquatica) have been widely used in CWtreated wastewater due to their tolerance to pollutants and adaptability [2, 8-10]. Vegetation is an important factor affecting the effectiveness of CWs in eliminating COD, BOD, TSS, and NH<sup>+</sup> in all kinds of wastewater [1, 2, 8, 11]. However, excessive nutrient concentration could suppress plant growth and transformation processes, thus affecting the wastewater treatment process in CWs. There is a little information about the correlation between pollutant concentration and the treatment time and efficiency of HSF CW. This study aims to evaluate the effects of pollutant factors (varied pollutant concentrations) on plant growth and treatment efficiency of HSF CWs, thus providing the basic for proper application of CWs in wastewater treatment.





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## 2. Materials and methods

## 2.1. Materials

*Cyperus alternifolius* was collected from the pig farming wastewater treatment system in Chuong My, Hanoi and was grown in HSF CWs filled with sand, limestone, and gravel.

The wastewater samples were synthetic wastewater in which pH values, COD, and  $NH_4^+$  concentrations were varied as presented in Table 1. The basis of the concentration range was to emulate the characteristics of wastewater types and plant tolerance thresholds reported in previous studies [8].

Table 1. Characteristics of the input synthetic wastewater.

pН	COD (mg/l)	NH <sub>4</sub> <sup>+</sup> (mg/l)
3	250	25
4	500	50
5	750	75
6	1000	100
7	•	125
8		150
9		200

#### 2.2. Experiment design

The HSF CWs had a length x width x height of 50x30x30 cm. The HSF CWs were constructed by using limestone (1-2 cm grain size, 10 cm height), gravel (0.5-1 cm grain size, 10 cm height), and sand (5 cm height) as substrates. *Cyperus alternifolius* was grown on filter media with an initial height of 15 cm and density of 18 shoots/CW.

The wetland was constructed and placed outdoors under a glass roof. The temperature ranged from 25 to 32°C, the average humidity was about 82%, and the average period of sunshine was 11.5 hours per day. The experiment was conducted in 7 days with a wastewater volume of 15 l for each system. 50 ml of sample was taken after 24, 48, 72, 96, 120, and 144 hours. The determined factors were pH and pollutant concentrations (COD,  $NH_4^+$ ). The growth performance of *Cyperus alternifolius* was determined by counting shoots and measuring plant height by the end of the experiment. All the experiments were conducted in triplicate.

#### 2.3. Sample analysis

The removal efficiency, H (%), by the CWs was calculated by using the difference in concentration (mg/l) of the inlet and outlet divided by concentration (mg/l) in the inlet. The hydraulic loading rate was calculated by the inlet flow rate divided by the area of the CW unit. The loading rate (g/m<sup>2</sup>/d) was calculated by multiplying the hydraulic loading rate by the inlet concentration (mg/l). The removal rate, R (g/m<sup>2</sup>/d), was defined as the hydraulic loading rate (m/d) multiplied by the difference in concentration (mg/l) between the inlet and outlet.

#### 2.4. Statistical analysis

Statistical analyses of the experimental data were performed using the SPSS 20.0 package for Windows. All data were tested for goodness of fit to a normal distribution, using the Kolmogorov-Smirnov one-sample test.

#### 3. Results and discussion

#### 3.1. Effect of pH on plant growth

pH is an important value in wastewater treatment as it strongly affects the growth of plants and microorganisms. Thereby, it affects the ability to metabolize nutrients, or the pollutant removal capacity of the wastewater treatment method [5].

The changes in pH values during the experiment are presented in Fig. 1. The results showed that the pH value was almost unchanged during the treatment process for wastewater with alkaline pH (pH=8-9), but for wastewater with low pH, it tends to rise to neutral (pH=7.2-7.5).



Fig. 1. The change in pH in of the HSF CWs during the experimental period.

Microbial transformation processes and wetland plants could attribute to the neutral pH results. In the range of neutral pH, the aquatic plants grew well in wastewater. After 7 days, the heights of Cyperus alternifolius were 16.8-21.5 cm (Fig. 2). The acidic wastewater (pH=3-4) or alkaline wastewater (pH=9) slightly affected plant growth. This result is similar to previous studies' results for some aquatic plants used for wastewater treatment [4, 7]. The height of the plants in those wastewaters only increased by 1.8 cm (pH=3) and 3.8 cm (pH=9), while they increased by 5.5-6.5 cm when grown in wastewater with pH=5-8. The plants also produced new shoots. After 7 days, the numbers of new shoots ranged from 2-11 in which the number of new shoots was highest when the wastewater was at neutral conditions (pH=6-8) (Fig. 3). The growth of Cyperus alternifolius increased with the increase of pH, reaching its highest value at pH=7, then the growth slightly decreased from pH 8 to 9.



Fig. 2. Effect of pH on the height of *Cyperus alternifolius* in the HSF CWs after 7 days.



Fig. 3. Effect of pH on the shoot generation of *Cyperus alternifolius* in HSF CWs after 7 days.

## 3.2. Effect of COD on wastewater treatment removal and efficiency

COD is a significant parameter in wastewater treatment; it is also a value used to evaluate the efficiency of wastewater treatment methods. Previous studies have proven that the presence of vegetation in CWs could remove an average of 57.8% of COD [4, 11]. The potential COD removal of vegetation was different for plant species and wastewater types. As shown in Fig. 4, the COD removal efficiency from synthetic wastewater in the HSF CWs planted with by Cyperus alternifolius was highest (H=90%) with an initial COD concentration of 250, and lowest (H=56,7%) with an initial COD concentration of 1000 mg/l. The result (H=90%) was higher than previous studies (H=85%) and even in the study that used same vegetation in treatment (H=69.87%). After 3 days, COD concentrations reduced from 750 mg/l to less than 300 mg/l (meeting the national standard for aquacultural wastewater QCVN 62:2016/BTNMT, column B).

The removal rate of COD by *Cyperus alternifolius* in the HSF CWs increased with the increase of COD concentration from 250 up to 750 mg/l, and the COD removal reduced when the COD concentration was 1000 mg/l. The results shown in Fig. 5 also indicated that HSF CWs planted with *Cyperus alternifolius* could remove 19.16 g/m<sup>2</sup>.d after 24 h when COD the concentration was 750 g/m<sup>3</sup>. The high potential of COD removal could be due to metabolism activities of microorganisms and the role of vegetation [1, 4].

The effects of COD concentration on the growth of *Cyperus alternifolius* in the HSF CWs during the experimental period are shown in Figs. 6 and 7. The height and the number of shoots of *Cyperus alternifolius* was almost unchanged with COD concentrations of 250-750 mg/l, but they strongly decreased when the COD concentration reached 1000 mg/l. Therefore, *Cyperus alternifolius* could tolerate high COD concentrations up to 1000 mg/l. This result was similar to results of [8] with the plant *Phragmites australis*.



Fig. 4. The treatment efficiency of COD by the HSF CWs planted with *Cyperus alternifolius* during the experiment.



Fig. 5. The removal rate of COD by the HSF CWs planted with *Cyperus alternifolius* during the experiment.



Fig. 6. Effects of COD concentrations on the height of *Cyperus alternifolius* in HSF CW after 7 days.



Fig. 7. Effects of COD concentrations on the shoot generation of *Cyperus alternifolius* in HSF CW after 7 days.

## 3.3. Effect of $NH_4^+$ on removal rate and wastewater treatment efficiency

In CW-treated wastewaters, vegetation can directly absorb and metabolize nutrients like nitrogen and phosphorus and enhance nutrient transformation processes such as nitrification, denitrification, adsorption, and desorption [3]. The NH<sub>4</sub><sup>+</sup> removal efficiency (%), and NH<sub>4</sub><sup>+</sup> removal rate (g/m<sup>2</sup>.d) by HSF CWs planted with *Cyperus alternifolius* are shown in Figs. 8 and 9, respectively. In this study, NH<sub>4</sub><sup>+</sup> removal efficiency (%) and NH<sub>4</sub><sup>+</sup> removal rate (g/m<sup>2</sup>.d) increased with an increase of treatment time. When the NH<sub>4</sub><sup>+</sup> concentrations at the inlet were high, about 75-125 mg/l, and after 7 days of treatment, the NH<sub>4</sub><sup>+</sup> removal efficiencies (%) were 31-69.7%. These results were similar to other studies that reported the removal of NH<sub>4</sub><sup>+</sup>-N by CWs planted with *Cyperus papyrus* - about

69.69% [2, 9]. With initial  $NH_4^+$  concentrations of 75-125 mg/l, the  $NH_4^+$  removal rates in this study were 1.22-1.66 g/m<sup>2</sup>d after 24 hours and decreased as  $NH_4^+$  concentrations in wastewater decreased over time.



Fig. 8. The NH<sub>4</sub><sup>+</sup> removal efficiency by HSF CWs planted with *Cyperus alternifolius* during the experiment.



Fig. 9. The NH<sub>4</sub><sup>+</sup> removal rate by HSF CWs planted with *Cyperus alternifolius* during the experiment.



Fig. 10. Effect of  $NH_4^*$  concentration on the height of *Cyperus* alternifolius in HSF CWs after 7 days.





Plants can directly use ammonium nitrogen (NH<sub>4</sub><sup>+</sup>-N) as a nitrogen source in metabolism processes [5]. Nitrogen plays an important role in plant construction and growth. Indeed, nitrogen combines with C, H, and O to make amino acids - blocks of protein - which are elements of protoplasm, enzymes, chlorophyll, and many vitamins [12]. As shown in Figs. 10 and 11, the growth of Cyperus alternifolius was affected by NH<sup>+</sup> concentration. With an initial NH<sup>+</sup> concentration of 25-75 mg/l, plant growth was good with plant heights of 22.4-20.7 cm, and the number of shoots were in the range of 11-14. However, the plant height and capacity of shoot generation decreased with an increase in  $\mathrm{NH}_{\scriptscriptstyle\!\!A}^{\,\scriptscriptstyle +}$  concentration. With an initial  $\mathrm{NH}_{\scriptscriptstyle\!\!A}^{\,\scriptscriptstyle +}$  concentration of 150 mg/l, several yellow leaves were observed. When NH<sup>+</sup> concentration was 200 mg/l, there were some dead plants. High concentrations of  $NH_4^+$  were potentially toxic to Cyperus alternifolius's growth, and therefore could result in their death [13]. The results of this study demonstrated that pH, the concentration of organic matter (COD), and  $NH_{A}^{+}$ strongly affected plant growth and the removal efficiency of HSF CW, therefore, considerations for setting up wastewater treatment plants using CWs should carefully take this data into account in practice to ensure treatment performance.

#### 4. Conclusions

This study evaluated the effects of pollutant factors including COD,  $NH_4^+$ , and pH on the growth of *Cyperus alternifolius* and the treatment performance of the HSF CW-treated synthetic wastewater. The following concentrations of organic matter (COD: 500-750 mg/l) and  $NH_4^+$ (75-125 mg/l) ensure normal growth of *Cyperus alternifolius* and good treatment performance of the HSF CWs. *Cyperus alternifolius* grew well at pH 6-8, and this plant species could be used as a vegetation element in the HSF CWs due to their ability to endure high concentrations of organic matter (COD: 500-750 mg/l) and  $NH_4^+$ (75-125 mg/l). This study serves as a basis for considering wastewater characteristics in the application of HSF CW systems in practice.

#### **CRediT** author statement

Nguyen Thanh Binh: Formal analysis, Data curation, Writing - Original draft; Bui Thi Kim Anh: Conceptualisation, Methodology; Nguyen Van Thanh: Visualisation, Investigation, Software; Dang Dinh Kim: Supervision, Validation; Nguyen Minh Phuong: Writing -Reviewing and Editing.

### **COMPETING INTERESTS**

The authors declare that there is no conflict of interest regarding the publication of this article.

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