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Simultaneous removal of color, organic compounds and nutrients in azo dye-containing wastewater using up-flow constructed wetland

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ABSTRACT

Combination of aerobic and anaerobic processes in constructed wetlands can enhance the treatment performance in textile wastewater. This study assessed the treatment of azo dye Acid Orange 7 (AO7) and nutrients using five laboratory-scale up-flow constructed wetlands (UFCW) with and without supplementary aeration, and with different emergent plants. Supplementary aeration controlled the size of aerobic and anaerobic zones in the UFCW reactors as evidenced by the oxidation–reduction potential (ORP) and dissolved oxygen (DO) profile of the UFCW. The AO7 removal efficiency was above 95% in all UFCW reactors and most of the color was extensively removed in the anaerobic region of the UFCW beds. The intermediates produced through the breakage of azo bond were significantly reduced in the UFCW reactors with supplementary aeration. The results indicated the applicability of the UFCW reactors to the treatment of azo dye-containing wastewater. The removals of T-N and T-P were in the range of 60–67% and 26–37%, respectively, among the UFCW reactors. The COD and NH_4-N removals in the aerated reactors were about 86 and 96%, respectively. On the other hand, the COD and NH_4-N removals were in the range of 78–82% and 41–48%, respectively, in the non-aerated reactors. The supplementary aeration enhanced the removal efficiencies in organic matter, NH_4-N and aromatic amines in the UFCW reactors.

1. Introduction

Constructed wetlands are recognized as eco-friendly, low maintenance and low-cost alternative technology for wastewater treatment. They were mainly used for treatment of domestic and municipal sewage, storm water and agricultural runoff [1–4]. Nowadays, the application of constructed wetlands is gaining in importance as one of the most promising alternatives for the treatment of industrial effluents in developing countries resulted from the transfer of the knowledge, technical collaboration and co-operation by the developed countries [5]. The accelerating industrialization in developing countries with an enormous consumption of vast kind of chemicals in various industries causes a growing environmental contamination hazard. It is well known that some industrial wastewater contain high concentration of less biodegradable, persistent and toxic contaminants such as dyes, aromatic and polyaromatic hydrocarbons, halogenated organic pollutants and so on. There are few studies on the application of constructed wetlands to industrial wastewater treatment. The performance of this eco-technology as industrial wastewater treatment measure should be evaluated from engineering point of view.

Azo dyes are the most widely used dyes in textile industry. The degradation of azo dye is difficult due to their complex structure and synthetic nature. Azo dyes are hence one of the most problematic contaminants in textile industry. A variety of physical, chemical and biological processes have been used to treat textile effluents [6]. Biodegradation of azo dye generally requires sequential anaerobic and aerobic processes [7–10]. The azo bonds can be broken biologically by some anaerobes. The aromatic amine residues from anaerobic decolorization resisted further anaerobic degradation [11], and they were also reported as mutagens [12]. The aromatic amine residues can be mineralized aerobically [13]. The aerobic processes could remove organic matter but exhibit a relatively low in color and nutrients removals [14,15], whereas anaerobic processes show a great removal in color, still, it cannot remove the organic matter and nutrient effectively [16].

The anaerobic and aerobic processes should be properly incorporated in constructed wetland in order to achieve simultaneous color, nutrients and carbon removals in azo dye-containing wastewater. The design of constructed wetland to control the aerobic and anaerobic regions is very important in order to achieve the whole process such as breakage of azo bond, mineralization of intermediate aromatic amines, nitrification and denitrification.

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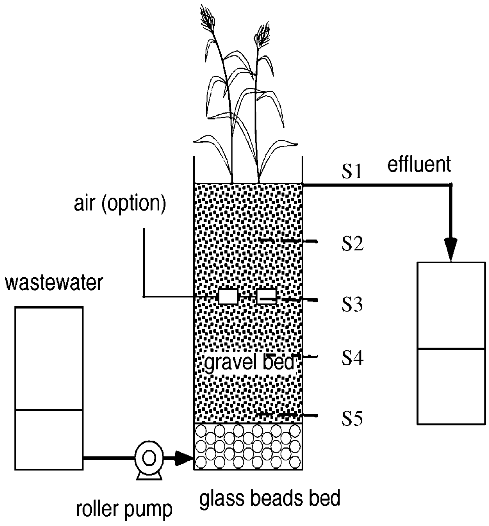

Fig. 1. Experimental setup of up-flow-constructed wetland.

Up-flow constructed wetland (UFCW) is proposed in the present study for the treatment of azo dye-containing wastewater. In the UFCW, the anaerobic condition is followed by aerobic condition because the upper surface region of constructed wetland is maintained in aerobic condition. Supplementary aeration is also applied to control the aerobic region of the UFCW reactor and promote the activity of aerobes.

The objective of present study is to examine the efficacy of UFCW reactor for the treatment of azo dye-containing wastewater. Acid Orange 7 (AO7) was used as a model less biodegradable organic contaminant in industrial wastewater. The effects of emergent plants and supplementary aeration on the simultaneous removal of color, organic matter and nutrients were investigated. Five UFCW reactors were used to examine the COD, nutrients, AO7, ORP and DO profiles along the reactors. Besides, the effects of azo dye AO7 on the growth of emergent plants were also discussed.

2. Materials and methods

Five parallel laboratory-scale up-flow constructed wetland reactors (namely reactor R-A, R-B, R-C, R-D and R-E) were prepared and located indoors. The temperature was controlled at 23 ± 3 °C. Artificial light was used and controlled by timer which ran on a cycle of 12 h on and 12 h off. Fig. 1 shows the schematic diagram of an UFCW reactor. Each UFCW reactor consisted of a wetland reactor, a wastewater reservoir, a roller pump and an effluent reservoir. The wetland reactor and operation characteristics are summarized in Table 1. The diameter of each wetland reactor was 18 cm and the height was 70 cm. Each wetland reactor was filled with 5 mm glass bead to a depth of about 6 cm to ensure the uniform distribution of inflow, and round washed gravel, 5.7 mm in average diameter, was filled to a height of 60 cm above the glass bead bed. The bed void height and water level in the reactor were almost the same. These wetland reactors were designed with five water sampling points by using a small pipe (7 (S5), 21 (S4), 36 (S3), 51 (S2) and 66 cm (S1) from bottom) along the reactors as shown in Fig. 1.

Table 1
Reactor characteristics.

Total column height	70 cm
Column diameter	18 cm
Total volume	17.8 L
Height of gravel bed	60 cm
Volume of gravel bed	15.3 L
Average gravel size	5.68 mm
Average gravel bed density	1.68 g/mL
Average gravel bed porosity	29.5%
Average void volume of gravel bed	4.52 L
HRT (based on average void volume of bed)	3 d
Average flow rate	1.04 mL/min

Table 2
Wetland conditions.

Reactor	Emergent plant	Aeration
R-A	Manchurian wild rice	None
R-B	*Phragmites australis*	None
R-C (control)	None	None
R-D	*Phragmites australis*	Aerated
R-E	None	Aerated

Constructed wetlands were planted with the shoots of *Phragmites australis* and Manchurian wild rice, which were transferred from Sakata Waterfowl Wetland, Niigata, Japan. After transplantation, the wetland reactors were loaded with tap water to establish the emergent plants and also seeded with a small amount of activated sludge. Five wetland reactors were operated with different conditions as summarized in Table 2. Out of five reactors, the reactor R-A was planted with Manchurian wild rice and the reactor R-B with *P. australis*. R-C was a control. The other two reactors, R-D (planted with *P. australis*) and R-E (unplanted), were installed with four porous air spargers at 30 cm below the bed surface for supplementary aeration.

The composition of wastewater was as follows: C_6H_5COONa 107.1, CH_3COONa 204.9, NH_4NO_3 176.1, NaCl 7.0, $MgCl_2{\cdot}6H_2O$ 3.4, $CaCl_2{\cdot}2H_2O$ 4.0 and $K_2HPO_4{\cdot}3H_2O$ 36.7, in mg/L. The characteristics of the wastewater were as follows: COD_{Cr} 326 mg/L, T-N 62 mg/L, and T-P 5.0 mg/L [17]. After the acclimatization, the wetland reactors were loaded with synthetic wastewater from low until the desired concentration. The concentrations of synthetic wastewater used were the quarter strength at the beginning, followed by the half till the full concentration. The average void fraction of gravel was about 29.5%. The wastewater was fed to the reactors to give hydraulic retention time (HRT) based on the void bed volume of 3 d. After the wetland reactors were operated for about two and half months, AO7 was added into the synthetic wastewater to give the concentration of 50 mg/L.

The influent and effluent water samples of UFCW reactors were collected periodically to evaluate treatment performance of the reactors. After the reactors reached steady state, water samples were collected from the five sampling points along the reactor to examine the pollutants' profiles. Wastewater samples were analyzed for COD, ammonium-N (NH_4-N), nitrate (NO_3^-), nitrite (NO_2^-), total nitrogen (T-N), total phosphorus (T-P) and AO7. COD was determined using a HACH DR/890 portable colorimeter. NH_4-N, T-N, and T-P were determined by the standard method (JSWA, 1994). NO_3^- and NO_2^- were measured using an ion chromatography (Shimadzu, Japan). The concentrations of the AO7 were determined by the absorbance at wavelength 480 nm using a UV/Vis spectrophotometer (Hitachi U-1100, Japan). ORP and DO were measured with an ORP meter (RM-20P, TOA-DKK, Japan) and DO meter (DOL-10, DKK, Japan), respectively.

3. Results and discussion

3.1. Dissolved oxygen and oxidation–reduction potential along UFCW system

The dissolved oxygen (DO) and oxidation-reduction potential (ORP) are the measures of the oxidizing (aerobic) and reducing

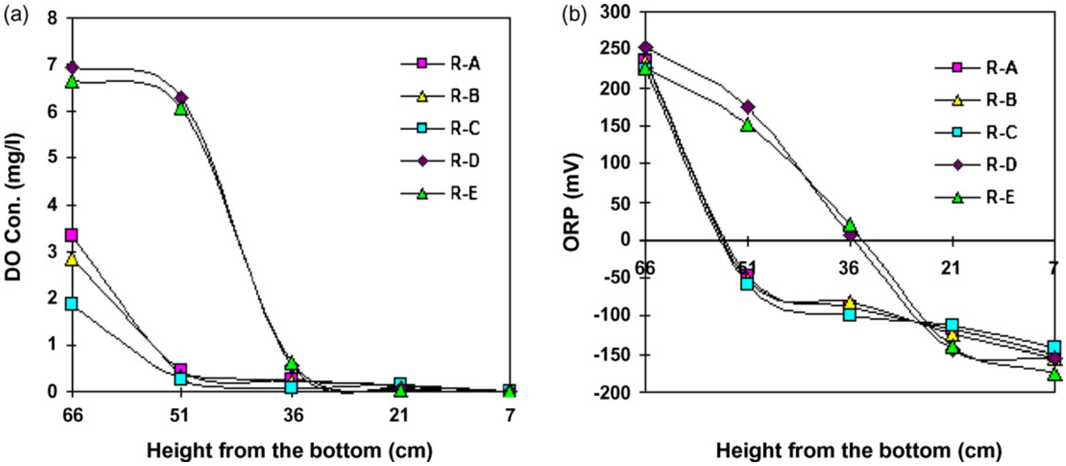

Fig. 2. Trend of DO (a) and ORP (b) along the up-flow constructed wetland.

(anaerobic) condition of aquatic environment. Redox potentials greater than 100 mV are commonly interpreted to indicate an aerobic environment, whereas ones less than −100 mV are to indicate an anaerobic environment [18]. The distribution of ORP and DO along UFCW reactors are shown in Fig. 2(a) and (b), respectively. The DO and ORP distributions were different between the UFCW reactors with supplementary aeration (R-D and R-E) and without supplementary aeration (R-A, R-B and R-C).

In the non-aerated UFCW reactors, the ORP and DO at the top of reactors were in the range of 213–235 mV and 1.9–3.3 mg/L, respectively, and sharply decreased to (−68)–(−88) mV and 0.2–0.4 mg/L, respectively, below 15 cm from the surface media. This indicated that only the top layer of these three reactors were aerobic. The ORP and DO at the bottom of these reactors were about −150 mV and 0 mg/L, respectively, indicating anaerobic condition. There was little difference in the ORP and DO distributions of reactors R-A, R-B and R-C. This suggests little contribution of the emergent plants on oxygen supply through their root system into the gravel bed in macro-scale. Strong redox potential at the micro-scale within the constructed wetland have been linked to the presence of macrophytes. The measurements of Bezbaruah and Zhang [19] using microelectrodes in experimental wetlands showed that the redox potential at the surface of lateral roots of *Scirpus validus* was higher than observed in the bulk water. The increased redox potential near the surface of roots was related to the presence of oxygen released by plants [19,20]. Therefore, aerobic condition may exist in the vicinity of the root in the reactors R-A and R-B. On the other hand, the ORP and DO distributions in the aerated UFCW reactors showed different trend from those in the non-aerated ones. The ORP and DO at the aeration point, about 30 cm from the surface bed, were in the range of 6–20 mV and 0.5–0.6 mg/L, respectively, and gradually increased to 225–252 mV and 6.6–6.9 mg/L, respectively, at the top of reactors. This shows that almost half of the gravel bed was aerobic by the supplementary aeration. The ORP and DO below the aeration point were less than −100 mV and 0.2 mg/L, respectively, indicating anaerobic condition. The sequence of anaerobic and aerobic conditions in an UFCW reactor is expected to enhance the simultaneous removal in color, organic matter and nutrients in textile wastewater. The depth of aeration point can be adjusted according to the characteristics of wastewater in practical application to give optimal aerobic and anaerobic ratio [21].

3.2. Plant monitoring

During the initial period of study, the transplanted plants acclimatized themselves well in UFCW reactors as shown in the increased number of stem, leaf and height. This showed the plants could adapt well to the synthetic wastewater. However, after the

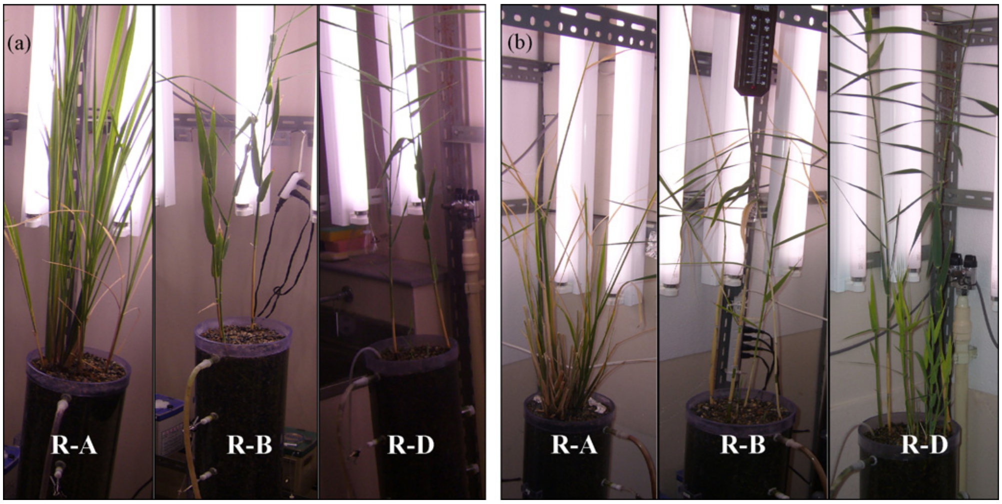

Fig. 3. Emergent plants (a) before and (b) after the addition of AO7.

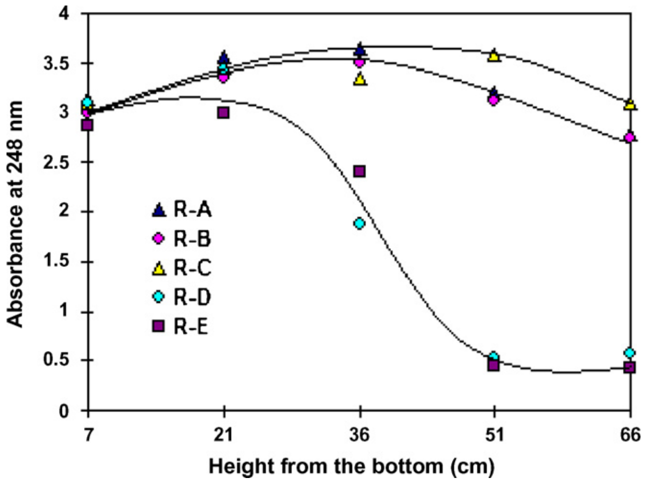

Fig. 4. Absorbance at 248 nm along UFCW reactors.

addition of AO7, toxic signs like chlorosis symptoms, leaf anatomical changes and plant death were observed in the reactor R-A (Manchurian wild rice) and the reactor R-B (*P. australis*). The growth of young stems *P. australis* was significantly inhibited in the reactor R-B. This toxic signs did not show in the reactor R-D (*P. australis*) with supplementary aeration. Fig. 3 shows the photos of transplanted plants in the reactors R-A, R-B and R-D before and after the addition of AO7. From the observation, almost all of the leaves in the emergent plants in R-A and R-B turned to yellowish and no new stems were found after the addition of 50 mg/L AO7. On the other hand, the emergent plant in R-D still in good condition and the number of stem was increased as the operation proceeded.

The emergent plant *P. australis* has been shown to survive and reproduce well in the aerated reactor than the non-aerated reactor after the addition of AO7. The high concentration in COD and nutrients, will be shown later, were not the reason for the anatomical changes, growth inhibition or death on the selected emergent plants in the reactors R-A and R-B. The toxic signs of plants in the non-aerated UFCW reactors could due to the effects of intermediate products generated through the reduction of AO7. When AO7 degradation occurs, aromatic amines such as sulfanilic acid and 1-amino-2-naphthol will be released. The wavelength of maximum absorption (λ_{max}) for sulfanilic acid was determined experimentally to be 248 nm. Fig. 4 shows the absorbance at 248 nm for the samples collected along the UFCW reactors. The figure clearly shows that most of the aromatic amines were mineralized in the aerated reactors as shown by the drastic drop in the absorbance at 248 nm above the aeration point. However, the aromatic amines were accumulated in the non-aerated reactors and slightly decreased at the top layer of reactors R-A and R-B. The amount of aromatic amines mineralized in the planted non-aerated-reactors (R-A and R-B) was higher than that in the unplanted one (R-C). This may suggest the activity of aerobic microbes in rhizosphere. These results showed the deterioration on the growth of emergent plants by AO7-containing wastewater in UFCW reactors. Further investigation will be carried out on the effects of azo dyes and aromatic amines on the emergent plants.

Table 3
Treatment performance of up-flow constructed wetland reactors in synthetic wastewater containing 50 mg/L AO7.

Parameters	Influent (mg/L)	Reactor	Effluent (mg/L)	Removal (%)
COD	383.8 ± 13.7	R-A	78.8 ± 15.0	79
		R-B	68.3 ± 18.6	82
		R-C	84.1 ± 19.1	78
		R-D	53.8 ± 15.6	86
		R-E	54.8 ± 15.6	86
T-N	55.4 ± 3.4	R-A	19.9 ± 3.4	64
		R-B	20.4 ± 3.1	63
		R-C	21.9 ± 2.9	60
		R-D	18.0 ± 3.8	67
		R-E	18.0 ± 6.0	67
T-P	6.7 ± 0.3	R-A	4.2 ± 0.4	37
		R-B	4.8 ± 0.5	31
		R-C	4.7 ± 0.3	30
		R-D	5.0 ± 0.3	26
		R-E	4.8 ± 0.2	29
NH_4-N	34.8 ± 2.8	R-A	18.0 ± 3.0	48
		R-B	19.6 ± 4.2	44
		R-C	20.4 ± 4.0	41
		R-D	1.5 ± 1.1	96
		R-E	1.7 ± 1.7	95
NO_3-N	29.5 ± 2.2	R-A	0	100
		R-B	0	100
		R-C	0	100
		R-D	4.1 ± 3.1	86
		R-E	1.9 ± 2.9	93
NO_2-N	0	R-A	0	–
		R-B	0	–
		R-C	0	–
		R-D	10.6 ± 4.7	–
		R-E	14.0 ± 4.8	–
AO7	50.5 ± 2.0	R-A	2.2 ± 0.3	96
		R-B	2.0 ± 0.6	96
		R-C	2.0 ± 0.5	96
		R-D	1.1 ± 0.6	98
		R-E	1.0 ± 0.4	98

3.3. *Treatment performance of UFCW systems*

After the UFCW reactors were operated with the synthetic wastewater for about two and half months, 50 mg/L of AO7-containing wastewater were introduced to the wetland reactors. The average COD, T-N, T-P, NH_4-N, NO_3-N and NO_2-N in the synthetic wastewater containing 50 mg/L AO7 were 384, 55, 6.7, 35, 30 and 0 mg/L, respectively. The average T-N concentration tested from experiment (55 mg/L) was lower than the theoretical value (62 mg/L). However, the average T-N concentration tested from experiment before the addition of 50 mg/L AO7 was about 61.1 mg/L which is close to the theoretical value. The addition of 50 mg/L AO7 in the synthetic wastewater might cause some effects on the T-N measurement. Table 3 shows the treatment performance of UFCW reactors for the wastewater containing 50 mg/L AO7. In general, all of the UFCW reactors show a high removal in color and organic matter but the nutrients removals were different among the reactors. The adsorption study revealed that the adsorption of pollutants on media was negligible (data not shown) except for T-P. This implies that most of the pollutants were removed biologically in the UFCW reactors. The organic matter in wastewater was removed in the wetland beds mainly through aerobic biological decomposition by microbes growing in the matrix. The removals of NH_4-N and NO_3^- were largely attributed to nitrification and denitrification, respectively, whereas the T-P removal was dominated by physical mechanisms. The details for each pollutant removal are discussed in the following section.

3.3.1. *COD removal*

The average COD concentration in synthetic wastewater was 320 mg/L and the addition of 50 mg/L AO7 increased the COD concentration to 384 mg/L. The organic matter in wastewater are degraded both anaerobically and aerobically by the heterotrophic

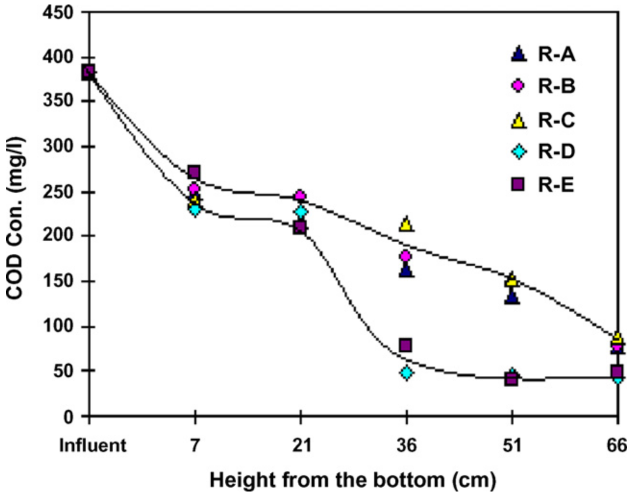

Fig. 5. COD profile along UFCW reactors.

microorganisms in the wetland reactors depending on oxygen concentration in the bed [4]. As shown in Table 3, the COD removal in the aerated UFCW reactors was higher than that in the non-aerated ones. Supplementary aeration boosted the mineralization of organic matter and enhanced the constructed wetland performance in treating high-strength wastewater. The aeration system could adjust to balance the aerobic treatment performance according to the wastewater characteristics and loading rate. The COD removal efficiencies were 79, 82 and 78% for the non-aerated reactors R-A, R-B and R-C, respectively. The uptake of organic matter by macrophytes is negligible compared to the biodegradation. The slight difference in COD removal efficiency among the non-aerated reactors might be due to the difference in oxygen diffusion rate and oxygen leakage from the macrophytes roots into the adjacent microenvironment where the degradation of organic matter by aerobic microbes may occur.

The excellent COD removal was mainly contributed by the microbial activities both aerobic biological decomposition and denitrification processes. The gravel media placed inside the UFCW reactors allowed the accumulation of immerse amounts of attached microbes, which were very helpful in rapidly catalyzing biochemical reactions. The organic biodegradation under anaerobic pathway in the wetland reactors also provides advantage of low-sludge production, which can largely prevent the wetland reactor being clogged by biomass [22]. Our results show that the difference between the planted and unplanted wetland reactors in COD removal was very little. Calheiros et al. [23] had also found no significant differences in COD removal between planted (*I. pseudocarus*, *S. secundatum*, *T. latifolia* and *P. australis*) and unplanted units in the first 17 months of operation, when the systems were subject to the same wastewater at HLRs of 3 and 6 cm d^{-1}. However, emergent plants can contribute to wastewater treatment processes in a number of ways, such as settlement of suspended solids, providing surface area for microorganisms and providing oxygen release [4,24,25].

Fig. 5 shows the COD concentration profiles in the gravel beds. In general, the COD concentration decreased by 33%, at 7 cm height from reactor bottom, which might be due to anaerobic degradation and dilution effect. As shown in the ORP and DO profiles, the anaerobic environment was developed at the bottom region of reactors either in the aerated or the non-aerated UFCW reactors. The anaerobic condition at the bottom region promoted denitrification which requires the utilization of external carbon source. The COD concentration in the non-aerated reactors (R-A, R-B and R-C) slowly decreased along the gravel bed while in the aerated reactors (R-D and R-E) the COD sharply decreased around the aeration point. These difference in COD concentration profiles well corresponded with DO and ORP profiles as shown in Fig. 2.

3.3.2. *Nitrogen removal*

As shown in Table 3, the T-N removal efficiencies were in the range of 60–67% among the wetland reactors. It was observed that the T-N removal efficiencies in the aerated reactors were higher than the non-aerated ones. Nitrification and denitrification processes play an important role in the nitrogen removal performance of wetland reactors. The removal efficiency of NH_4-N in the aerated reactors was about 95% while it was in the range of 41–48% in the non-aerated reactors. This showed the supplementary aeration extremely enhanced the removal of NH_4-N in the UFCW reactors. Besides, it was observed that the planted reactors (R-A and R-B) showed better removal in NH_4-N than the unplanted one (R-C). This could due to the uptake by emergent plants and also the release of oxygen from the root of emergent plants producing a positive effect on nitrifying bacteria in the rhizosphere [26]. Several experimental studies on N removal in treatment wetlands also showed the unplanted treatment had a lower N removal compared with planted treatment [27–30]. However, other researchers found no difference in the removal of NH_4-N and NO_3^- from wastewater as a result of the presence of plants in a constructed wetland [23,31]. The beneficial role of plants in constructed wetlands is not always evident, and that seems to depend on several parameters, such as the time length of operation, type of vegetation and characteristic of the wastewater.

It was observed that the NH_4-N removal efficiency in reactor R-A (planted with Manchurian wild rice) was slightly higher than that in reactor R-B (planted with *P. australis*). Janjit et al. [28] reported that Manchurian wild rice preserved the high ranking of nutrient removal rates among 21 kinds of plants for both area-based and weight-based calculation. The NH_4-N removal dropped significantly in the non-aerated reactors after the addition of 50 mg/L AO7 in the influent. The NH_4-N removal efficiencies were about 85, 62 and 59% in the reactors R-A, R-B and R-C, respectively, before the addition of AO7. The azo dye AO7 and the aromatic amine compounds generated may cause partial inhibitory effect on the activity of nitrifying microbes. Some researchers reported that the azo dye-bearing wastewater inhibited activated sludge nitrification resulting in the effluent with higher NH_4-N concentrations [32,33].

Anaerobic environment was developed in almost of the whole bed in the non-aerated reactors. Nitrate and nitrite were hardly detected in these reactors, indicating high-denitrification rate in the bottom region. On the contrary, the concentration of these compounds increased in the aerated reactors R-D and R-E, corresponding to the decrease in NH_4-N concentration. The higher nitrate concentration in the reactor R-D (planted with *P. australis*) may be resulted from the increase in number of nitrifying bacteria in the vicinity of the rhizome ascribed to the addition of oxygen supply through roots system. The total NO_3-N and NO_2-N concentrations in the effluent of aerated reactors was about 15 mg/L. Partial denitrification was observed in the aerobic regions of reactors R-D and R-E. However, the supplementary aeration would inhibit the denitrification process. As a result, only approximately half of the nitrate and nitrite generated from nitrification was denitrified in the aerated reactors. Thus, aeration conditions should be adjusted to provide suitable anaerobic and aerobic microbial reactions in the vertical wetland.

In order to understand the fate of nitrogen in UFCW reactors, the distributions of NH_4-N and NO_3-N along the reactors were analyzed and the results are shown in Fig. 6(a) and (b), respectively. The NH_4-N concentration profiles of aerated reactors were qualitatively the

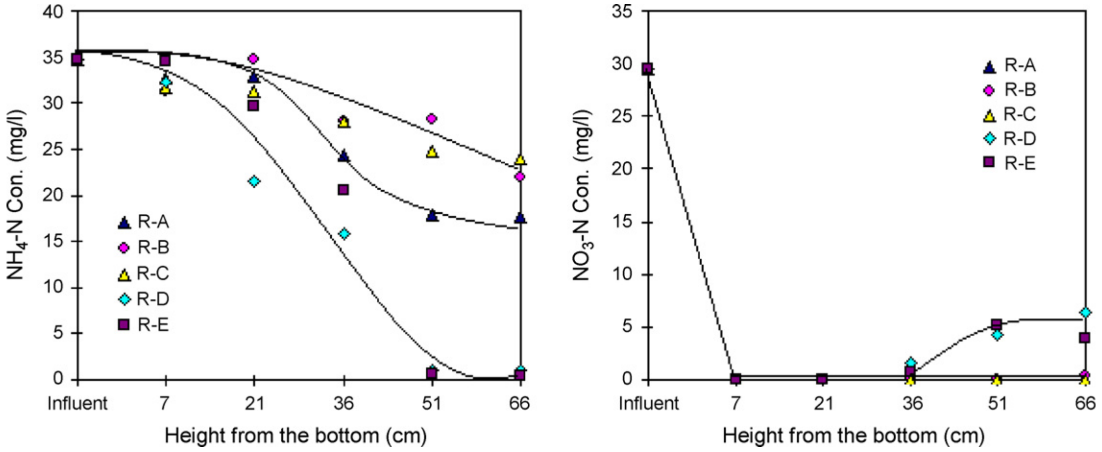

Fig. 6. NH_4-N (a) and NO_3-N (b) profiles along UFCW reactors.

same to the COD profiles, where NH_4-N concentration drastically decreased above the aeration point. This indicated that the nitrification proceeded in the upper aerobic beds. Slight decrease in NH_4-N concentration was observed from the middle part of reactors R-A, R-B, and R-C as similar to the COD profiles in the non-aerated reactors. As mentioned above, this could due to the diffusion, convection and leakage of oxygen from macrophytes roots into the adjacent microenvironment. The average NO_3-N concentration in influent was about 30 mg/L and it was fully removed at the bottom of every wetland reactor. The pH of influent was about 7.5 and was found that it increased to 8.5 at a height of 7 cm from the bottom of reactor. The anaerobic environment and abundance in external carbon source at the bottom wetland reactors encouraged the denitrification. The increased nitrate in the aerated reactors was corresponded to the nitrification that occurred in aerobic region of media beds.

3.3.3. *Total phosphorus removal*

Several studies have reported on the potential use of constructed wetlands in the removal of phosphorus from wastewaters [34,35]. T-P removal in wetlands is a manifold process. There are physical, chemical and biological functions effective for the removal of different T-P components [4,36]. As shown in Table 3, the average removal efficiencies of T-P in the reactors R-A, R-B, R-C, R-D and R-E were 37, 31, 30, 26 and 29%, respectively. The T-P removal of reactor planted with Manchurian wild rice was slightly higher than that of the reactor planted with *P. australis*. In the case without AO7 addition, the T-P removal in the reactor planted with Manchurian wild rice (R-A) was 52% and significantly differed from that in the other reactors. This again showed the high-removal efficiency of nutrients by Manchurian wild rice. T-P removal in constructed wetlands may take place due to plant uptake [37], accretions of wetland soils [38], microbial immobilization [39], retention by the substrate and precipitation in the water column [40]. Among these factors, the substrate may play the greatest role and could be the factor most amenable to control. Thus, it is important to select those substrates presenting the highest phosphate adsorption capacity [41]. In our study, most phosphorus is believed to be stored in the media bed rather than in the emergent plants. The adsorption played an important role for T-P removal as shown by the adsorption study where the adsorption capacity of T-P by gravel was about 37.7 mg/g. The T-P removal efficiency dropped gradually as the operation proceeded due to the saturation of adsorption capacity (data not shown). Basically, the T-P concentration slowly decreased along the media beds (Fig. 7). The adsorption, precipitation and uptake by plant and biomass were the cause for the T-P removal. Further study is needed to investigate the T-P distributions or mass balance in the wetland reactor.

3.3.4. *Azo dye AO7 removal*

Preliminary adsorption study was carried out by shaking 100 g of gravel with 100.0 mL AO7 solution (20 mg/L) for a contact time

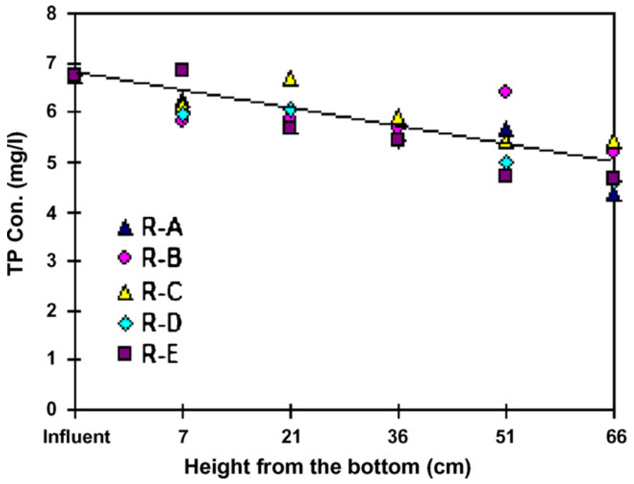

Fig. 7. T-P profiles along UFCW reactors.

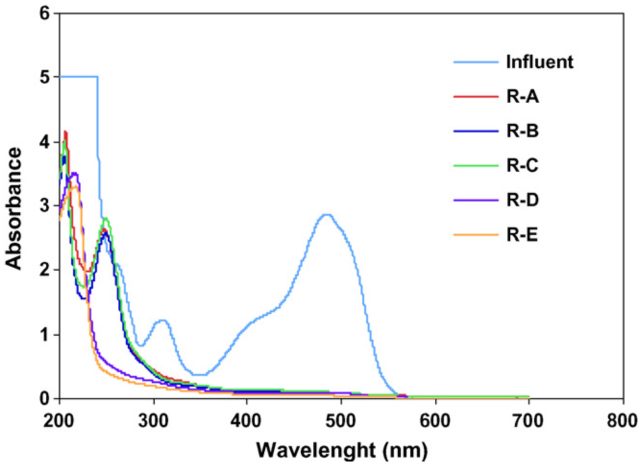

Fig. 8. UV–vis spectrum of effluent UFCW reactors.

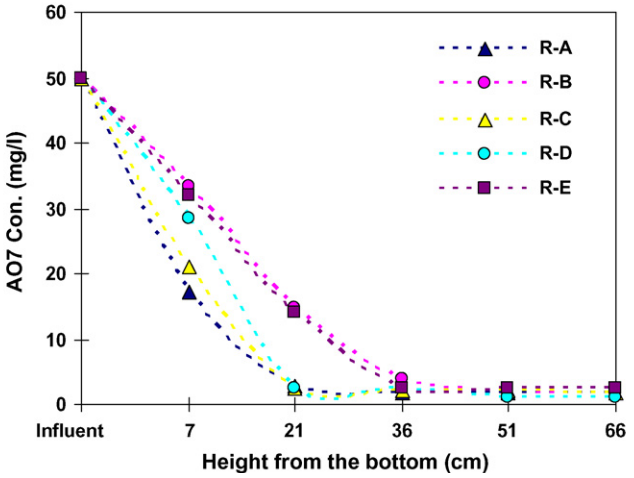

Fig. 9. AO7 profiles along UFCW reactors.

of 48 h. It was found that only about 0.5% of AO7 was removed by the gravel and this showed in the contribution of gravel bed to AO7 removal through adsorption was negligible. Fig. 8 shows the UV–vis spectrum for the effluent samples collected from UFCW reactors. The UV–vis spectra of the influent and effluent clearly show that the AO7 has been almost completely eliminated as shown with the absorbance at 480 nm. Since the adsorption of AO7 on gravel is negligible, the removal of color must be due to the biodegradation of azo bond.

Fig. 9 shows the AO7 concentration profiles along the beds. Most of the AO7 was removed in the anaerobic region of wetland reactors. In the anaerobic region, AO7 was reduced by microbes and the intermediate aromatic amines resisted further degradation. Although the cleavage of the azo bond removes the visible problem, there remains the problem of the aromatic amines. The absorbance at wavelength 248 nm for the non-aerated reactors (R-A, R-B and R-C) shows the presence of sulfanilic acid as intermediate produced through AO7 reduction. Anaerobic degradation of sulfanilic acid is difficult, as the sulfonyl group is a xenobiotic structural element, and the strongly charged anionic moiety prevents penetration of the compound through bacterial membranes [42]. As a result, the aromatic amines were accumulated in the anaerobic region of wetland beds, and started to be further mineralized above the aeration point in aerated reactors (Fig. 4). The slight drop in aromatic amines concentration in the planted non-aerated reactors (R-A and R-B) indicated that the emergent plants also play an important role in the removal of dye. A significant portion of oxygen needed to support the aerobic mineralization of aromatic amines in wetland reactors could be obtained from the roots. As shown in Table 3, the AO7 removal was above 95% in all UFCW reactors. Although the color removals among the wetland reactors were not significantly different, the data obtained shows the aerated reactors (R-D and R-E) removed aromatic amines more effectively compared to the non-aerated reactors (R-A, R-B and R-C). Moreover, the planted non-aerated rectors (R-A and R-B) showed slightly higher removal efficiency of aromatic amines than the control reactor (Fig. 4).

4. Conclusions

Supplementary aeration controlled aerobic and anaerobic conditions in the UFCW reactors as shown by the ORP and DO profiles of beds. In general, the anaerobic followed by aerobic conditions in UFCW reactors with supplementary aeration showed the high-removal efficiency in AO7, organic matter, aromatic amines and nutrients. The aerated reactors give aerobic and anaerobic regions respectively to the upper and lower media beds of wetland reactors. The non-aerated reactors give anaerobic condition in almost the whole bed of reactors.

The UFCW reactors with supplementary aeration outperformed the non-aerated reactors in terms of the removal of organic matter, NH_4-N and the aromatic amines which generate from AO7 reduction. The color and nitrate were extensively removed in the anaerobic region of wetland reactors. The sequence in anaerobic followed by aerobic processes in aerated UFCW reactors demonstrated the high efficiency in simultaneous removal of color, nutrients and organic matter in azo dye-containing wastewater. Moreover, the mineralization of aromatic amines in the aerobic region might reduce the inhibitory/toxic effects on the emergent plants. The accumulation of aromatic amines in the non-aerated reactors could be one of the reasons to cause chlorosis, anatomical changes or death on the emergent plants. There was also a slight improvement in the removal of aromatic amines by the emergent plants compared to the unplanted reactor which might be due to the mineralization by aerobic biomass that growth near to the macrophyte roots or uptake by the plants.

Up-flow constructed wetland reactors with supplementary aeration could control the aerobic and anaerobic regions in the bed and treat high-strength wastewater. As the results obtained by using the laboratory scale UFCW reactors, with and without supplementary aeration, are promising, our systems will be evaluated in pilot- and full-scale experiments in a long-term study to ensure the applicable of the systems for industrial wastewater. The present system will be one of the most promising wastewater treatment technologies to treat less-biodegradable industrial wastewater discharged in industrializing Asian regions.

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