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Mineralization of diazo dye (Reactive Black 5) in wastewater using recirculated up-flow constructed wetland reactor

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ABSTRACT

Application of a sequential anaerobic and aerobic process in a constructed wetland can enhance the treatment performance of textile wastewater. In this study, two laboratory-scale recirculated up-flow constructed wetland (UFCW) reactors planted with *Phragmites australis* were constructed to investigate the treatment performance between aerated and non-aerated reactors for treating 50 mg/L Reactive Black 5 (RB5)-containing wastewater. Results showed that the non-aerated reactor demonstrated an almost entire anaerobic environment yielded a better RB5 removal efficiency (89%) compared to the aerated reactor (81%). However, the aerated reactor performed higher removal on chemical oxygen demand (COD) and aromatic amines than the non-aerated reactor. Proper design ratio of anaerobic/aerobic region in UFCW is a key to accomplish not only color reduction but also COD and aromatic amines in textile wastewater treatment.

Keywords: Up-flow constructed wetland; Supplementary aeration; Reactive Black 5; Aromatic amines; Artificial aeration

1. Introduction

Azo dyes are extensively used for the dyeing of cotton and constitute about 60–70% of total dyes produced. Azo dyes are characterized by one or more nitrogen–nitrogen double bond (–N=N–) called azo group. Their removal from wastewater can be accomplished by physical [1], chemical [2], or biological processes [3,4], but biological processes are usually preferred because they are cheaper and environmentally friendly [5]. Microbial azo dye decolorization is resulted by the reduction of the azo bond under anaerobic conditions, which leads to the accumulation of toxic aromatic amines [6]. Due to mutagenic and carcinogenic nature, aromatic amine residues from anaerobic decolorization resist further anaerobic degradation [7,8] and subsequently the chemical oxygen demand (COD) concentration persists in the effluent. These aromatic amine residues can be mineralized aerobically [9]. A wastewater treatment process with the incorporation of anaerobic and aerobic conditions is, therefore, an attempt to achieve not only decolorization but also degradation of the aromatic amines [10–12].

Constructed wetland (CW) is verified as a promising environmental-friendly technology to be widely applied for wastewater treatment. Emergent plants play important roles in CW for wastewater treatment such as settlement of suspended solids, providing

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surface area for micro-organisms and providing oxygen release [13,14]. CWs were initially utilized for nutrients removal in residential and municipal sewage but increasing attention is now also being paid to using CWs to treat leachate, contaminated groundwater, and industrial effluents [15]. However, study on the application of CWs for textile wastewater treatment is still limited. Bulc and Ojstršek [16] reported that their CW model and the pilot CW demonstrated well capability to treat dye-rich textile wastewater. The CW used in their study had significant effects on color reduction, i.e. up to 70% in the CW model and up to 90% in the pilot CW. The role of the emergent plant, Phragmites australis, in the degradation of azo dye Acid Orange 7 (AO7) under vertical-flow constructed wetland (VFCW) operation has been thoroughly investigated by Carias et al. [17] and Davies et al. [18,19].

Combined anaerobic-aerobic treatment holds promise method to completely remove azo dyes from wastewater. Therefore, the anaerobic and aerobic processes should be properly incorporated in CWs in order to achieve simultaneous color and aromatic amines removals in azo dye-containing wastewater. The design concept of UFCW by flowing of wastewater through anaerobic region followed by aerobic region could efficiently treat azo dyes-containing wastewater, but very limited literatures focus on this design and application [20]. The UFCW was used for removing the azo dye AO7, and the results were reported in our previous study [20]. It was observed that the performance of the reactors depends on the artificial aeration, hydraulic retention time, and types of emergent plants. The aim of this study is to investigate the treatment performance of recirculated UFCW reactors on the diazo dye RB5-containing wastewater. The recirculated UFCW reactor was selected in the current study instead of continuous flow of azo dyecontaining wastewater into UFCW reactor due to easier handling and operation. The discharge of treated wastewater can be done as the concentrations of pollutants were removed till to desired level.

2. Materials and methods

2.1. Chemicals

RB5 ($C_{26}H_{25}N_{25}O_{19}S_6$; CAS 17095-24-8) was supplied by Sigma-Aldrich. The UV–vis spectra of RB5 were recorded from 200 to 800 nm using a UV–vis spectrophotometer (Hitachi U-2800, Japan). The maximum absorbance wavelength (λ_{max}) of RB5 was found at 597 nm. The synthetic wastewater consisted of organic carbon, nutrients, and buffer solution of the

following composition (concentration in mg/L; manufacturer; CAS): C_6H_5 COONa (107.1; Riedel de Haen; 18106), CH₃COONa (204.9; HmbG Chemicals; C0729-21416332), NH₄NO₃ (176.1; Bendosen; C0082-2270791), NaCl (7.0; HmbG Chemicals; C0753-21416,592), MgCl₂.6H₂O (3.4; HmbG Chemicals; C0472-21413962), CaCl₂.2H₂O (4.0; HmbG Chemicals; 21412322), and K₂HPO₄.3H₂O (36.7; HmbG Chemicals; C0939-21415122) giving COD Cr 326, T-N 62, and T-P 5.0 mg/L. All other chemicals were of analytical grade.

2.2. Adsorption study

This study was conducted to determine the adsorption capacity of RB5 onto gravel. The gravel with an average size of 6 mm was washed with distilled water and dried in oven at 105 °C for 24 h before used for adsorption study. RB5 of 100 mL with concentrations of 50 and 100 mg/L were put into plastic bottles. Then, 10 and 20 g of gravels were added into the bottles. The bottles were placed on a shaker for shaking 24 and 72 h continuously at 350 rpm. The solutions were filtered with 0.45 µm membrane filter and the filtrates were analyzed for RB5 concentration using UV-vis spectrophotometer (Hitachi U-2800, Japan).

2.3. Reactor setup and operation

Two laboratory-scale UFCW reactors, namely, aerated and non-aerated, were constructed at indoor and the RB5-containing wastewater was circulated between the UFCW reactor and the wastewater collection tank. The schematic diagram of a recirculated UFCW reactor is illustrated in Fig. 1. The diameter of reactor was 23.6 cm and the height was 33.8 cm. Each reactor was filled with gravels of an average diameter of 6 mm up to a height of 30 cm. With these



Fig. 1. Schematic diagram of a recirculated UFCW reactor.

dimensions of the reactor and gravel, it provided about 2.2 L of void volume which was enough for the laboratory-scale evaluation on the performance of recirculated UFCW for treating RB5-containing wastewater. The reactor also could develop sequential anaerobic and aerobic conditions at the lower bed to upper bed, respectively, along the reactor. The results obtained in the laboratory-scale reactor can be used as a fundamental design for pilot-scale study in future. Biological clogging of wetland media is often problematic and may increase the water retention time in the wetland reactor and cause the reduction of effective area available for water infiltration and subsequently affect the flow direction and hydraulic retention time. The gravels with an average size of 6 mm may avoid the clogging problem and at the same time provide adequate surface area for the growth of micro-organisms on the surface of the media. The characteristic of the recirculated UFCW reactor is shown in Table 1.

The wastewater was pumped into the reactors through the bottom point of S5, and the treated wastewater was discharged from the top point of S1. The wastewater was recycled back into reactors continuously for 48 h. Then, treated wastewater was disposed and new synthetic wastewater was prepared and the process was repeated. *Phragmites australis* (also known as common reed) was used as an emergent plant in both reactors, and the roots of the plant were located at 16 cm below the bed surface. After transplantation, the reactors were fed with tap water and then seeded with activated sludge for biofilm establishment. In the aerated reactor, two porous air spargers were installed at 16 cm below the bed surface to provide artificial aeration.

2.4. Analysis

Treatment performance of the recirculated UFCW in treating RB5-containing wastewater was evaluated

Table 1	
Characteristics of UFCW reactor	

Total column height	33.8 cm
Column diameter	23.6 cm
Height of gravel bed	30 cm
Total volume	15 L
Volume of gravel bed	13 L
Average gravel size	6 mm
Average gravel bed porosity	17%
Average void volume of gravel bed	2.2 L
Hydraulic retention time	2 days
Average flow rate	30 mL/min

based on the removal of organic matters, color, and intermediate products being generated. Wastewater samples were analyzed for COD, RB5, and aromatic amines. Concentration of COD was determined by using HACH DR2800 spectrophotometer. UV–vis spectrophotometer (Hitachi U-2800) was used to determine the concentration of RB5 as well as the absorbance of aromatic amines at 254 nm. Oxidationreduction potential (ORP) was monitored along the sampling points in the wetland reactors with HANNA ORP meter.

3. Results and discussion

3.1. Plant monitoring

Plants differ widely in their capacity to adapt to oxygen deficiency [21]. Most plant species that are not adapted to waterlogging exhibit injury symptoms, wilting, leaf senescence, and epinasty are likely to be the first. What follows is a rapid decline in or even its termination [22]. In such cases, the respiratory consumption by plant roots, soil fauna, and micro-organisms can totally deplete the oxygen. While many plants can survive under these anaerobic/anoxic conditions for a short time, only the plant species found in wetlands can persist under these conditions. In the present study, the selected emergent plant, Phragmites australis, could transfer atmospheric oxygen to the rhizome and out through the roots ultimately to the gravel and rhizosphere. This feature permits roots to respire aerobically and to grow in soil conditions that are often anoxic. Oxygen transport into the root zone through lenticles and aerenchymous tissue has been measured between $2.08 \text{ g} \text{ O}_2/\text{m}^2/\text{d}$ by Brix and Schierup [23] and 5–12 g $O_2/m^2/d$ by Armstrong et al. [24] in *Phragmites australis* grown in gravel beds.

Visually, inspection of plant's growth can be done for the sign of toxicity such as chlorosis, necrosis, and malformation. The density or height of the shoots of emergent plant and the number of leaves also were used to evaluate the stress or sign of toxicity of the plants due to the presence of pollutant [25-28]. The photos presented in Fig. 2 show the growth of transplanted emergent plants in recirculated UFCW reactors before and after the addition of RB5-containing wastewater. The emergent plants showed a healthy growth condition at the first day of transplantation. After addition of RB5-containing wastewater into the recirculated UFCW reactors, toxic signs of emergent plants could be observed such as chlorosis, where stems and leaves turned to yellow, and plants were wilted. This suggested that the emergent plants were in stress condition with RB5-containing wastewater.



Fig. 2. Observation of emergent plants: (a) first day of transplantation, (b) one week, and (c) one month after the addition of RB5-containing wastewater.

After three weeks of acclimatization, the emergent plants were found to adapt well with the environment and stems turned to green and new shoots started to grow in both the aerated and non-aerated reactors. New shoots were produced first in the aerated reactor, which indicated that artificial aeration provided greater positive effect on emergent plants' growth compared to non-aerated reactor. Fig. 2c shows that emergent plants in the aerated reactor grew more abundant compared to those in the non-aerated reactor. The increase in the number of shoots and leaves and the height of plants in both reactors were observed as the operation proceeded.

3.2. ORP profile

As a general rule of thumb, ORP readings greater than 100 mV are indicative of aerobic condition, whereas ORP readings less than -100 mV indicate anaerobic condition [10]. Anoxic condition may present between 100 and -100 mV. The distributions of ORP in the aerated and non-aerated reactors were different (Fig. 3). In the aerated reactor, almost all of the beds showed low positive ORP readings which ranged from 23 to 42 mV. This suggested that the aerobic or anoxic environment might present in the bed region between 9 and 30 cm from the bottom of the reactor. The micro-organisms that grew under anoxic condition (called facultative microbes) used molecules other than oxygen such as nitrate, as their energy source for metabolism. The slight increase in ORP values at 16 cm of bed region could be explained as the existing of roots zone and air spargers provided additional oxygen into the wetland reactor. The top layer of the non-aerated reactor showed a relatively low positive ORP value of 15 mV. A sharp decrease in



Fig. 3. Trend of ORP along UFCW reactors.

ORP value was observed at 23 cm from the bottom of the reactor. The bed regions below 23 cm demonstrated negative ORP readings in the range of –118 to 126 mV. This indicated that only the top layer of the reactor was in aerobic or anoxic condition, whereas the bed regions below 23 cm were in anaerobic condition. The aerobic, anoxic, and anaerobic regions developed in the recirculated UFCW reactor will influence the microbial activity in the reduction of color, degradation of organic compounds and aromatic amines, nitrification, and denitrification.

3.3. Adsorption study

The results of adsorption study showed a very low adsorption capacity of RB5 onto gravels (Table 2). An increase in the amount of gravels and contact time did not give significant difference on dye removal. The adsorption capacity for RB5 ranged from 0.02 to 0.06 mg/g. The values of dye removal for RB5 per

russipation cupacity of RDs onto gravers							
ntration (mg/L) Adsorption capacity (mg/	Adsorption capacity (mg/g)						
10 g gravels	20 g gravels						
24 h 72	24 h 72 h						
0.06 0.0	0.02 0.03						
0.04 0.0	0.02 0.04						
24 h 72 0.06 0.0 0.04 0.0	24 h 0.02 0.02						

Table 2 Adsorption capacity of RB5 onto gravels

gram of gravel were very low and can be neglected. As a result, the role of gravels as an adsorbent in dye removal was considered not of importance.

3.4. Treatment performance

Table 3 shows the treatment performance of recirculated UFCW reactors for treating 50 mg/L of RB5containing wastewater. The adsorption study showed a very low adsorption capacity of RB5 onto gravels which can be ignored. This indicated that color was removed through microbial degradation in the recirculated UFCW reactors. Various populations of microorganisms grew in the recirculated UFCW reactors to break down pollutants by means of anaerobic and aerobic activity. Overall, the results showed high removal efficiencies on color and COD in both reactors. The average COD concentration of 50 mg/L RB5-containing wastewater was 392 mg/L. According to Vymazal et al. [29], treatment efficiency of CW for the removal of organic compounds highly depends on the oxygen concentration in wetland bed, design of CW, treatment condition, and filled medium characteristics. This idea is comparable with our results in which COD removal in the aerated reactor (92%) was higher than that in the non-aerated reactor (83%) as shown in Table 3. Artificial aeration used in the aerated reactor successfully developed an almost wholly aerobic/ anoxic conditions and could efficiently mineralize organic compounds in wastewater. The artificial aeration boosted the COD removal rate of wetland system by increasing the aerobic region [30]. Aromatic amines that are formed as a result of azo dye reduction have been reported to be degraded more easily under aerobic condition [31,32]. The absorbance at 254 nm represents the presence of aromatic amines in the wastewater. It was found that higher amounts of aromatic amines were accumulated in the non-aerated reactor as shown in Table 3 and Fig. 4. It could be ascribed to the complete color reduction under anaerobic condition. Moreover, the aromatic amines are hard to be degraded anaerobically, which led to a lower COD removal efficiency.

The RB5 is an intensely colored dye having one naphthalene ring and two aromatic amine rings with sulfonate functional groups joined by two azo bonds and is recalcitrant to biological degradation. In the recirculated UFCW reactors, the microbial populations developed on the support materials gravels and root systems of the emergent plant. Generally, aerobic, anoxic, and anaerobic microbes grew at different depth along the recirculated UFCW reactors as discussed in the ORP profile. Besides, anaerobic and aerobic microniches may develop at the inner and outer biofilm of gravel surfaces. The anaerobic and anoxic microbes were responsible for the reduction of azo bond that led to decolorization. In this study, anoxic microenvironment prevailing in the recirculated UFCW especially in

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Parameter	Influent (mg/L)	Reactor	Effluent (mg/L)	Removal (%)	
COD	394 ± 53	Aerated	34 ± 22	92 ± 4	
		Non-aerated	66 ± 40	83 ± 10	
RB5	48 ± 5	Aerated	9 ± 4	81 ± 8	
		Non-aerated	5 ± 1	89 ± 3	
Aromatic amines at 254 nm (abs)	1.12 ± 0.02	Aerated	0.49 ± 0.04		
		Non-aerated	0.72 ± 0.09		

Table 3 Treatment performance of UFCW reactor in RB5-containing wastewater



Fig. 4. UV–vis spectrum analysis of the water samples collected from UFCW reactors.

the aerated reactor facilitates the reduction of dye molecules by mimicking anaerobic microenvironment. Reduction of azo bond by anaerobic and anoxic microbes has been reported by researchers. The sequential anoxic-aerobic bioreactor achieved complete decolorization and up to 98% removal of COD load of synthetic dye wastewater containing 100 mg/L of C.I. Acid Red 88 was reported by Manjinder et al. [33]. The anoxic conditions in the column provided suitable environment for the reduction of azo bond as the latter is reported to be readily reduce under anaerobic conditions [33]. Alessandro et al. [5] observed that the color removal mainly took place under anaerobic conditions, although a small increase in color removal was still observed under anoxic conditions. Under anoxic conditions, the presence of small anaerobic microzones (e.g. inside the activated sludge flocks) may cause the reduction of azo bond through reduction activity by microorganisms [5].

Azo dye reduction by bacteria under anaerobic conditions is often a co-metabolic reaction, in which the reducing equivalents produced during the anaerobic oxidation of a co-substrate are used to break the azo bonds. Azo dyes act as terminal electron acceptors in the respiratory electron transfer chain [34]. The aromatic amines formed as a result of reduction process are generally not degraded under anaerobic conditions, but can be further mineralized under aerobic conditions. In this study, the removal of RB5 was due to co-metabolism process. The co-metabolism of RB5 is defined as the removal of non-growth substrate (RB5) by growing micro-organisms in the presence of growth co-substrates. A co-substrate is defined as the carbon and energy source for microbial growth and maintenance and release of the electrons for the cleavage of azo bond under reducing environments [35,36]. In the present study, sodium benzoate and sodium

acetate were used as co-substrates. The anaerobic microbes utilized the energy generated from the degradation of these co-substrates for the reductive cleavage of RB5. The mass quantities of sodium benzoate and sodium acetate used as co-substrates were 0.24 and $0.45 \,\mathrm{g}$, respectively, and giving $326 \,\mathrm{mg/L}$ of COD. With this amount of co-substrates, the RB5 removal efficiency at aerated and non-aerated reactors was 81 and 89%, respectively. As reported by Isik and Sponza [37], glucose as a co-substrate providing the reduced environment appears to be sufficient for the cleavage of azo bond in Congo Red at COD concentration as low as 100 mg/L for complete decolorization. Chinwekitvanich et al. [38] reported that the addition of tapioca as a co-substrate increased the decolorization efficiency by up to 70% in textile wastewater treatment by UASB systems. Mendez-Paz et al. [39] had reported that the low glucose concentration in the influent seems to be responsible for the lower efficiency of AO7. However, theoretically, a loading rate of 0.20 g of glucose $L^{-1}d^{-1}$ is necessary to obtain the reducing equivalents needed to achieve the complete reduction of AO7.

Decolorization of the dye solution by microbes could be due to the adsorption onto microbial cell or biochemical reaction leading to biodegradation. In adsorption, examination of the absorption spectrum would reveal that all peaks decreased approximately in proportion to each other. If dye removal is attributed to latter phenomenon, either the major visible light absorbance peak would completely disappear or a new peak would appear [40]. Fig. 4 shows the UVvis spectrum of RB5. There are three absorption peaks, one in visible region (597 nm) and two in UV region (254 and 312 nm). This can be ascribed to the presence of chromophoric azo bonds and aromatic compounds, respectively, in RB5. As shown in Table 3, RB5 removal efficiency in the aerated and non-aerated reactors was 81% and 89%, respectively. In the nonaerated reactor, almost the entire bed was in anaerobic condition, demonstrated a higher color reduction when compared to the aerated reactor. This results proved that microbial decolorization of azo dyes is more effective under anaerobic conditions, which were also reported by other researchers [10,29]. As shown in Fig. 4, the peak at 597 nm that contributed to the color was almost completely eliminated in the non-aerated reactor. However, the appearance of the absorbance peak at 254 nm shows the formation of aromatic amines after the reduction of azo bonds by anaerobic microbes. For aerated reactor, the peak of 597 nm was significantly reduced, but it still can be observed from the UV-vis spectrum. This clearly indicated that the azo dye reduction was performed by biodegradation as the adsorption of RB5 onto gravels was negligible. Since anaerobic condition only developed in the bottom layer of the aerated reactor and the anoxic microenvironment prevailing in almost the entire bed media suggested the insufficiency of anaerobes population to carry out azo bonds reduction compared to the non-aerated reactor. The effects of emergent plant on direct degradation of azo dve RB5 were not investigated in present study. However, Carias et al. [17] and Davies et al. [18,41] have reported that the emergent plant, Phragmites australis, plays an important role in the degradation of dyes. Antioxidant and detoxification enzymes of the emergent plant, Phragmites australis, in the degradation of the azo dye AO7 were studied by Carias et al. [17] and Davies et al. [18,41]. Phragmites australis produces enzymes in particular POD, which degrade AO7 [18]. Carias et al. [17] have studied the activities of antioxidative enzymes such as SOD, CAT, ascorbate peroxidase, and dehydroascorbate reductase, when Phragmites australis was exposed to AO7, indicating that the enzymes of Phragmites australis react to the presence of AO7, protecting plant cells against oxidative damage.

Libra et al. [42] reported the expected amines from hydrolyzed RB5 reduction are in the expected stoichiometric ratio of 2 mol p-aminobenzene-2-hydroxylethylsulfonic acid (p-ABHES) and 1 mol 1,2,7-triamino-8-hydroxynaphthalene-3,6-sulfonic acid (TAHNDS) per mol hydrolyzed RB5, which could be demonstrated by LC-MS analysis. The absorbance peaks of 312 nm and 254 nm in UV-vis spectrum (Fig. 4) represent naphthalene and benzene groups, respectively. There was no peak observed in wavelengths range from 300 to 400 nm in UV-vis spectrum in both effluent samples collected from the aerated and non-aerated reactors. This may be due to the instability of the TAHNDS which can auto-oxidize immediately upon contact with oxygen to further oxidize into other forms with a half life of 40-80 min as revealed by Libra et al. [42]. The intermediate aromatic amines generated from the cleavage of azo bond during anaerobic biodegradation were determined using UV-vis spectrophotometer at the wavelength of 254 nm. Table 3 shows that higher amount of aromatic amines was accumulated in the non-aerated reactor compared to the aerated reactor. This result is comparable with the UV-vis spectrum in Fig. 4, which shows a peak in the UV region in the non-aerated reactor. On the other hand, the artificial aeration in the aerated reactor boosted the mineralization of aromatic amines until no peaks were detected in the UV region of UV-vis spectrum.

The profiles of decolorization and accumulation of aromatic amines in the UFCW reactors were observed

for 48 h. As shown in Fig. 5, the decolorization rate in the non-aerated reactor was higher than that in the aerated reactor especially at the beginning stage. This was ascribed to the anaerobic region in the nonaerated reactor which was larger compared to that in the aerated reactor as discussed in ORP profile. The larger population of anaerobic microbes in the nonaerated reactor contributed to higher RB5 removal efficiency. The cleavage of azo bond in RB5 leads to decolorization and formed intermediate aromatic amines. The peaks at 597 and 254 nm represent the azo bond and aromatic amines, respectively, and the ratio of 254/597 nm shows the accumulation of aromatic amines in the effluent sample. In the non-aerated reactor, it was observed that the absorbance ratio increased tremendously as the operation time proceeded. This shows the accumulation of intermediate aromatic amines in the non-aerated reactor. As the



Fig. 5. Color (a) and aromatic amines (b) monitoring of the water samples collected from UFCW reactors.

azo bond reduced by the anaerobic microbes, the aromatic amines generated could not be further mineralized by the dominant anaerobic microbes. Partial amount of aromatic amines might be mineralized by aerobic microbes that grew near to the root systems of the emergent plant or at the top layer of the reactor. On the other hand, the absorbance ratio only increased slightly in the aerated reactor which showed that most of the aromatic amines generated were mineralized by the aerobic microbes that grew in the reactor. The results showed that biodegradation of azo dye RB5 was initiated with the cleavage of azo bond by anaerobic microbes. The intermediate aromatic amines produced could contribute to higher COD in effluent. As a result, these intermediate aromatic amines must be further degraded by aerobic microbes for the complete mineralization of RB5. The incorporation of anaerobic followed by aerobic conditions was important for the simultaneous removal of color and aromatic amines. The on and off of aeration in the UFCW reactor can be used to control the ratio of the aerobic and anaerobic regions in the wetland bed for the complete removal of dyeing wastewater.

4. Conclusion

The results presented in this study demonstrated that it is possible to obtain high decolorization as well as high biodegradation of organic compounds using recirculated UFCW reactor. Artificial aeration accelerated the development of aerobic environment in the UFCW reactor resulting in a higher COD removal efficiency (92%) than the non-aerated reactor (83%). No significant peaks were detected in the UV region of the UV-vis spectrum of aerated reactor that evidences the aromatic amines were efficiently being removed. The absorbance ratio profile showed tremendous decrease in aromatic amines in the aerated reactor. Anaerobic environment promoted a better performance of azo bond reduction by anaerobes reported at 89% RB5 removal efficiency in the non-aerated reactor. The RB5 concentrations of treated wastewater were significantly reduced from 50 to 9 and 5 mg/L in the aerated and non-aerated reactor, respectively. Further study will be conducted to control the aerobic and anaerobic region in the recirculated UFCW in order to remove the color and aromatic amines completely, and original textile wastewater will also be tested.

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