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Aerobic decolourization of the indigo dye-containing textile wastewater using continuous combined bioreactors

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Abstract

An aerobic bioprocess was applied to Indigo dye-containing textile wastewater treatment aiming at the colour elimination and biodegradation. A combined aerobic system using continuous stirred tank reactor (CSTR) and fixed film bioreactor (FFB) was continuously operated at constant temperature and fed with the textile wastewater (pH: 7.5 and total chemical oxygen demand (COD): 1185 mg l^{-1}). The CSTR is a 1 l continuous flow stirred tank reactor with a 700 ml working volume, and operated with a variable wastewater loading rate (WLR) from 0.92 to 3.7 g l^{-1} d^{-1}. The FFB is a 1.5 l continuous flow with three compartments packed with a rippled cylindrical polyethylene support, operated with a variable WLR between 0.09 and 0.73 g l^{-1} d^{-1}. The combined two bioreactors were inoculated by an acclimated microbial consortium and continuously operated with four total WLR. This system presented high COD elimination and colour removal efficiencies of 97.5% and 97.3%, respectively, obtained with a total hydraulic retention time (HRT) of 4 days and total WLR of 0.29 g l^{-1} d^{-1}. The effects of WLR on absorption phenomena on the yield of conversion of substrate on biomass ($R_{TSS/COD}$) and on the yield of conversion of substrate on active biomass ($R_{VVS/COD}$) are discussed. The increase of WLR and the decrease of HRT diminished the performances of this system in terms of decolourization and COD removal explained by the sloughing of biofilm, and the washout phenomena.

Keywords: Textile wastewater; Indigo; Aerobic; Microbial consortia; Decolourization

1. Introduction

Textile industries consume a considerable amount of water in their manufacturing processes. Considering both the volume and the effluent composition, the textile industry wastewater is rated as the most polluting among all industrial sectors. Important pollutants in textile effluents are mainly recalcitrant organics, colours, toxicants and inhibitory compounds, surfactants, chlorinated compounds (AOX), pH and salts [1]. Alterations to their chemical structures can result in the formation of new xenobiotic compounds which may be more or less toxic than the potential compounds [2]. It has been proven that some of those dyes and/or products are carcinogens and mutagens. Apart from the aesthetic deterioration of the natural water bodies, dyes also cause harm to the flora and fauna in the natural environment [3–5]. Therefore, industrial effluents, like textile wastewater containing dyes must be treated before their discharge into the environment. The dye wastewater from the textile is one of the most difficult wastewater to treat [6,7]. Because of their commercial importance, the impact and toxicity of dyes that are released in the environment have been extensively studied [8].

Colour can be removed from wastewater by chemical and physical methods including absorption, coagulation–flocculation, oxidation and electrochemical methods. These methods are quite expensive, have operational problems [9], and generate huge quantities of sludge [10]. Among low cost, viable alternatives, available for effluent treatment and decolourization, the biological systems are recognised, by their capacity to reduce biochemical oxygen demand (BOD) and chemical oxygen demand (COD) by conventional aerobic biodegradation [4,8,11].

Work on the use of combined bacterial process to treat textile wastewater has been carried out over the years by many research groups [8,12]. Recent study has used the combina-

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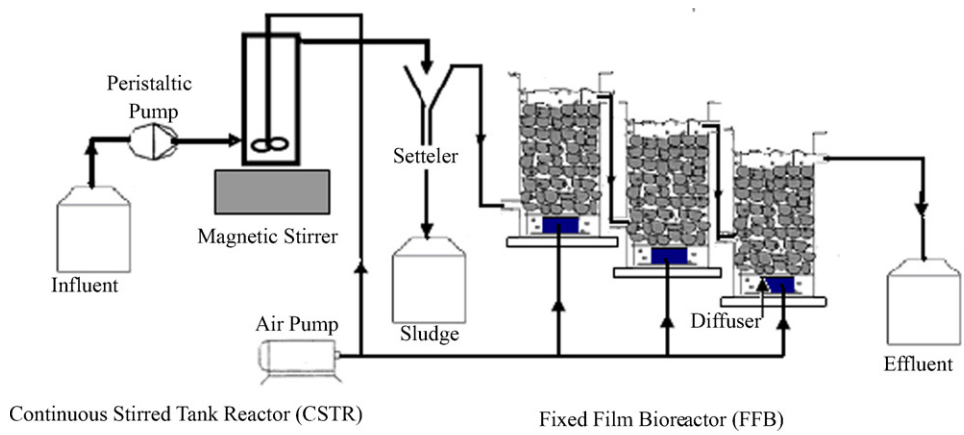

Fig. 1. Schematic representation of the experimental set-up used for textile wastewater treatment.

tion of anaerobic and aerobic steps in an attempt to achieve not only decolourization but also mineralization of azo dyes. The use of exclusively aerobic processes to decolourize azo dye-containing wastewater was not common and rarely used; because dyes are difficult to remove by using the conventional wastewater treatment systems based on aerobic processes [13]. Aerobic processes have been recently used for the treatment of textile wastewater as standalone processes and it is confirmed that they are efficient, cost-effective for smaller molecules and that the aerobic reactor is an effective technique to treat industrial wastewater [5,10,14–19]; or in combination with anaerobic processes [12,19–22].

With this aim, the research investigated the aerobic biodegradability of the textile dye (Indigo) by a combination of two aerobic bioreactors (CSTR and FFB). The effects of HRT and WLR on the treatment efficiencies in terms of COD and colour removal efficiencies were investigated.

2. Materials and methods

2.1. Operational conditions of laboratory bioreactors

A laboratory scale aerobic bioprocess as shown in Fig. 1 was used in this study. The aerobic system used was a combined CSTR and FFB bioreactor. The system was operated continuously at a constant temperature of 30 °C using an external water bath. The continuous stirred tank reactor with a 700 ml working volume was used. Mixing was assured by the continuous rotation of the magnetic stirrer. The FFB is a 1.5 l continuous flow with three compartments packed with a rippled cylindrical polyethylene support. The coupled system was first inoculated with a microbial consortia obtained from a textile wastewater treatment plant. These inocula were selected because of the large variety of microorganisms that could be found in the biomass degrading dyes in textile wastewater, and because mixed cultures offer considerable advantages over the use of pure culture. In fact, individual strains may attack the dye molecules at different position or may use decomposition products produced by another strains for further decomposition.

Each bioreactor was operated in batch-wise until biofilm formation was realised in the FFB and the microbial consortia becomes stable in the CSTR bioreactor. In fact, it is mentioned that adaptation is important for successful decolourization, and as acclimation occurred, the decolourization time becomes constant [14].

The system was fed by a peristaltic pump with the textile effluent (containing the Indigo dye) obtained from textile wastewater plant in Ksar Hellal (Tunisia), and its pH was maintained at approximately 7.5. Air was provided from the bottom of the aeration of the combined bacterial process using diffusers and an air pump. Bioreactors operating conditions are depicted in Table 1.

2.2. Analytical methods

The effluent from each bioreactor was collected daily, centrifuged at 7000 rpm for 10 min and analysed for colour, COD, pH, total suspended solids (TSS), volatile suspended solids (VSS) and colonies forming units (CFU). COD and colour measurements were carried out on the clear supernatant. Colour was measured by an UV–vis spectrophotometer (Jenway UV visible spectrophotometer) at a wavelength of 620 nm in which maximum absorbance spectra was obtained. The total and soluble COD was measured following standard methods. Measured COD and absorbance values were used for calculation of biodegradation and decolourization efficiencies.

The TSS was determined by drying samples at 105 °C for 24 h. The VSS, which is assumed as active biomass, was measured by calculating the loss of the sludge before and after

Table 1
Bioreactors operating conditions

HRT (day)	WLR ($g\,l^{-1}\,d^{-1}$)
4	0.296
3	0.394
2	0.592
1	1.185

HRT: hydraulic retention time; WLR: wastewater loading rate.

treating at 550 °C. Total plate counts were done in triplicate by spreading 0.1 ml of diluted effluent onto agar solidified selective medium supplemented with filtered influent. The pH was measured using a digital calibrated pH-meter (HANNA pH 210). Conductivity and total dissolved salts (TDS) were determined using a calibrated conductivity meter (METTLER TOLEDO MC 226).

3. Results and discussions

3.1. Characterisation of textile wastewater

Table 2 shows the characteristics of the textile influent. Its organic load was due to the presence of grease, dirt and/or sizing agents, nutrients from dye bath additives and residual dyes. These different components explain the values of total COD and BOD_5 about 1185 and 900 mg l^{-1}, respectively. Compared with another study of Buirton and Quezada Moreno [14], the influent used to feed bioreactors presented low COD and BOD_5 values due to the dilution results from different washings. Alkali or acids from the bleaching, desizing, scouring and mercerizing steps result in an extreme pH of 11–12 plus a high salt content of 2.84 g l^{-1}. The influent presented a basic pH of 9.6; this is not suitable for the biological activity of microorganisms. For those reasons, the influent pH was adjusted daily at 7.5 corresponding to the optimal pH for a maximal biological activity. This influent presented a blue colour, with maximal absorbance spectra of 0.98 obtained at the wavelength of 620 nm. This value shows that this influent is charged in Indigo compared with those used by another works such that of Sen and Demirer [1]. The values of CODs, which are very close to total COD and to the COD/BOD_5 ratio of 1.31 (Table 2), confirmed that this influent is easily biodegradable and presents approximately the same ratio (1.35) like the one used by Buirton and Quezada Moreno [14].

3.2. Effects of WLR and the HRT on the performances of the combined system

The treatment performance of the coupled system are based on the measurement of COD, OD 620 nm, conductivity, TDS, pH, TSS and VSS. The corresponding values are depicted in Figs. 2 and 3. The system was operated under four WLR and HRT.

This study shows that the influent presented a soluble COD (CODs) of 1075 mg l^{-1} and an $OD_{620\,nm}$ of 0.98, respectively (Fig. 2). The influent was biodegraded first by the communities of microorganisms present in the CSTR, and secondly by the fixed microorganisms on the biofilm of the FFB bioreactor. In fact, the CSTR effluent presented a COD of 200 mg l^{-1} for the first two WLR and it is about 350–500 mg l^{-1}, respectively for the third and the fourth WLR. It presented an OD about of 0.3, 0.42, 0.5 and 0.6, respectively, for the first, the second, the third

Table 2
Characteristics of textile wastewater containing azo dye

Parameters	Values			
Total COD (mg l^{-1})	1185	4100	1157.5	1029
Soluble COD (mg l^{-1})	1075	–	–	–
Total suspended solids TSS (mg l^{-1})	980	–	–	–
Volatile suspended solids (VSS) (mg l^{-1})	540	–	–	–
pH	9.56	8.18	9.9	9.91
TDS (g l^{-1})	2.84			
OD (620 nm)	0.98	–	0.18[a]	0.21[a]
BOD_5	900	3018	162.5	170
COD/BOD_5	1.31	1.35	–	–
References	This work	[14]	[1]	[1]

[a] OD at 669 nm.

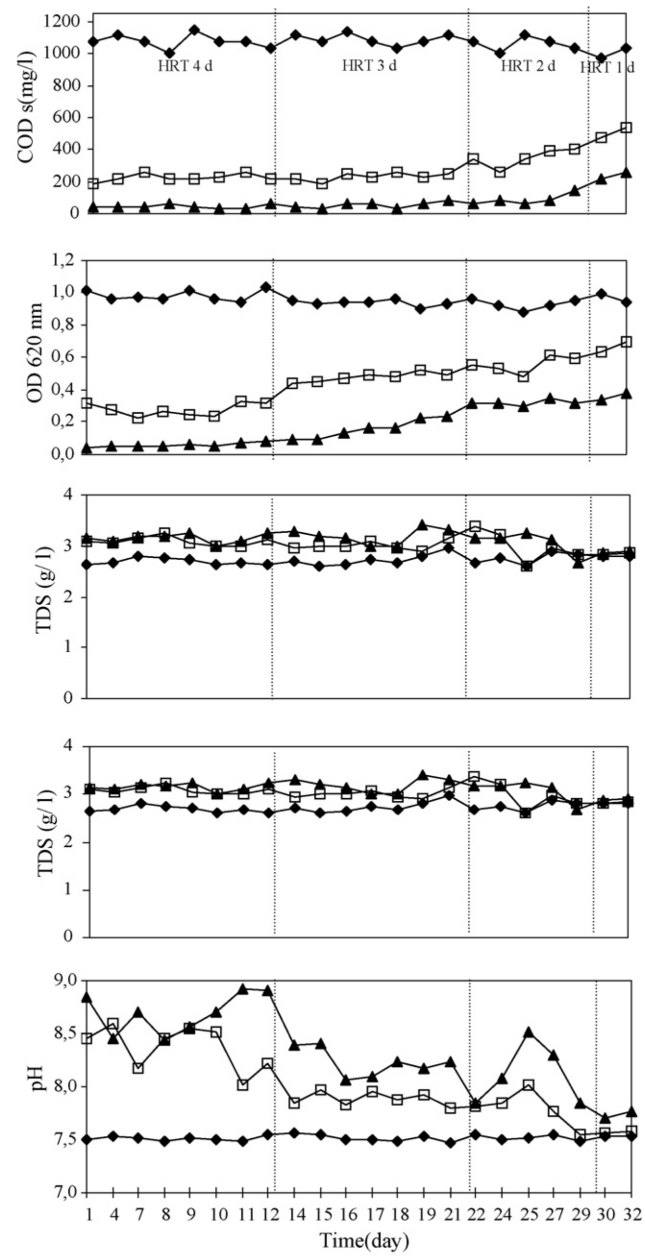

Fig. 2. Time course of COD, OD, conductivity, TDS and pH of the influent (♦), CSTR (□) and FFB (▲) under various WLR (WLR1 = 0.296 g l^{-1} d^{-1}, HRT4 = 4 days; WLR 2 = 0.394 g l^{-1} d^{-1} HRT3 = 3 days; WLR 3 = 0.592 g l^{-1} d^{-1}, HRT2 = 2 days; WLR 4 = 1.185 g l^{-1} d^{-1}, HRT1 = 1 day).

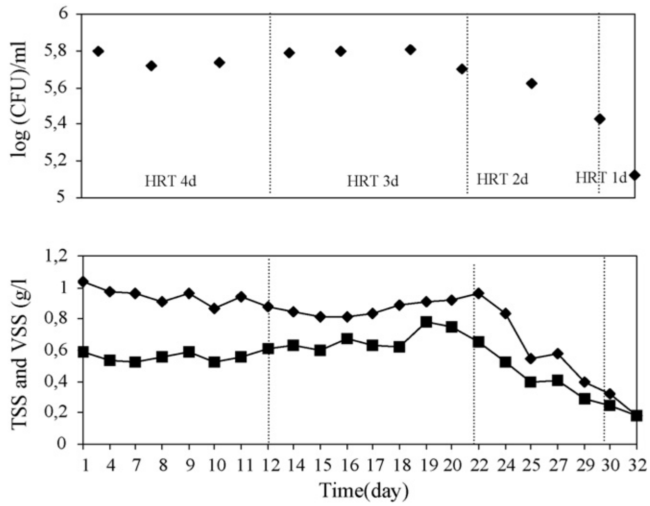

Fig. 3. Effect of wastewater loading rates on the total suspended solids (♦), volatile suspends solids (■) and cell density expressed in log (CFU)/ml in the CSTR effluent.

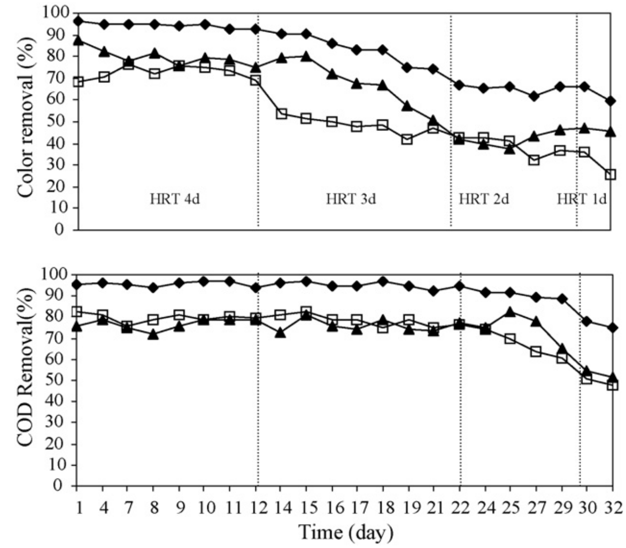

Fig. 4. Effect of the wastewater loading rates on colour and COD removal efficiencies in CSTR (□), FFB (▲) and combined system (♦).

and the fourth WLR. The CODs of FFB effluent increased from about 45 mg l^{-1} for the first three WLR to 150 mg l^{-1} for the fourth WLR, while the OD increased from 0.05 to 0.3, respectively, at the beginning and at the end of treatment. However, CODs and OD_{620} decreased with increasing both the HRT and the WLR. An increase in the conductivity and therefore the TDS was observed as shown in Fig. 2. This can be due to the presence of newly formed salty compounds.

For the first three WLR, the pH of the CSTR effluent increased from 7.5 to about 8.0 (Fig. 2). Similar trends are also observed for the FFB reactor. These pH values are, therefore, within the acceptable range for proper reactor operation. This result is explained by the biodegradation of dye by azoreductase enzymes and by the presence of new compounds like aromatic amines with alkaline characteristics [15,23]. During the final WLR the effluent pH was similar to that of the influent, indicating the low biodegradability.

Fig. 3 shows the TSS, VSS and CFU trends from the CSTR reactor. These results are useful for the study of the effect of WLR and HRT on the biomass evolution. As shown in Fig. 3, the initial biomass of 1 g l^{-1} that represented the inocula remained stable until day 24. During the last two WLR, the decrease of the biomass, VSS and CFU was essentially due to the washout phenomena of microorganisms.

3.3. Colour removal efficiency in CSTR and FFB bioreactors

The colour removal efficiency shown in Fig. 4 was above 95% in the first WLR. However, initial concentration did not inhibit the microorganisms and the degradation time was enough to allow the microorganisms to decolourize the dye [14]. These colour changes could be explained by structural modification of dye molecule due to azo bond reduction [12,24]. The decrease of colour removal efficiency by 33% observed in all the next three WLR is due to the decrease of biological activity in microbial consortia and in the biofilm. These results are explained by the increase of WLR and to the decrease of biodegradation time (HRT). The colour removal efficiencies obtained with FFB reactor are higher than those obtained with the CSTR reactor (Table 3). This is due to the advantage of the fixation of the microorganisms on the support and to the absence of washout phenomena. The aerobic biofilm reactor is an effective technique to treat industrial wastewater. The system is known to achieve high volumetric loads, no washout problems and a compact design of the reactor. The biofilm can support different populations which degrade different substrates at different points within the biofilm. For a wastewater containing a mixture of conventional pollutants and toxic organics, all organics will diffuse into

Table 3
Aerobic continuous reactor systems treating azo dye-containing wastewater

Bioreactor	Dye	Volume (l)	HRT (h)	WLR ($g\,l^{-1}\,d^{-1}$)	Microflora	COD removal (%)	Colour removal (%)	Reference
Rotating biological contractor	Blue G	1.7	48	0.25	*Coriolus versicolor*	–	80	[9]
Rotating drum bioreactor	Acid Orange 7	1	1.5	0.88	Two bacterial strains	100	–	[15]
Micro-aerophilic aerobic system	Mixture of dyes	–	7.7	2.23	Microbial consortia	85.3	94	[10]
Fixed film bioreactor	Blue G	10	50	0.96	Activated sludge + fungus	82	90	[27]
CSTR	Indigo	0.7	96	0.92	Activated sludge	80	75	This work
FFB	Indigo	1.5	96	0.09	Activated sludge	77	80	This work
Combined CSTR/FFB	Indigo	2.2	96	0.29	Activated sludge	97.5	97.3	This work

Table 4
Aerobic batch bioreactor systems treating azo dye-containing wastewater

Bioreactor	Dye	Volume (l)	Time (hour)	Concentration ($mg\,l^{-1}$)	Microflora	COD removal (%)	Colour removal (%)	Reference
Batch	Acid red	0.050	–	20	Activated sludge	–	100	[26]
Batch	Methyl red	–	18	100	Mixte culture	100	90	[2]
Batch	Reactif black, reactif red	0.15	–	100	*Geotrichum* sp.	–	100	[32]
Batch	Reactif blue, reactif green, reactif orange	–	48	200	*P. sordaria*	–	90	[33]
Batch	Indigo	0.25	96	20	C*hrysosporium ph, Philinus* gilvus	100	–	[28]
Sequenced batch biofilter	Acid red 151	8.9	12	55	Activated sludge	99	87.6	[14]

the biofilm but the conventional organics will be degraded much more quickly leaving only the toxics to penetrate deeper into the biofilm [5,25]. In fact, increasing WLR to 1.185 $g\,l^{-1}\,d^{-1}$ produces a decrease of dye degradation on day 30, possibly due to inhibitory effects of high concentration of dye or intermediate products on the microbes, or the blockage of active sites of azoreductase enzymes by dye molecules with different structures [26]. However, at high dye concentration, the FFB biofilm becomes progressively less and less capable of eliminating dye from the FFB. The most serious consequence of high influent concentration of dye is that it probably leads to accelerated biofilm growth and subsequent sloughing of biofilm [15], and to the entrainment of microorganisms with the effluent in the CSTR bioreactor.

3.4. COD removal efficiency in CSTR and FFB bioreactors

As shown in Fig. 4, the overall COD removal efficiency was higher than 90% during the application of the first three WLR. When the HRT was shortened from 4 to 3 and from 3 to 2 days, the system was able to maintain above 90% of COD removal due to the acclimatization of microorganisms to the high concentration of dye. This high removal efficiency could be attributed to both degradation and conversion of the aromatic amines and to the most difficult component which will be biodegraded into new components [7]. The decrease of COD removal efficiency by 20% observed in the fourth WLR may be explained by the phenomena of washout in the CSTR reactor and the sloughing of biofilm in the FFB [15], which are due to the increase of WLR (from 0.394 to 1.185 $g\,l^{-1}\,d^{-1}$) and the toxicity of Indigo on microorganisms. This decrease may also be explained by the reduction in the number of viable microbes and the reduction of the biological activity of the microbial consortia. These two mechanisms are demonstrated by the counting of CFU, TSS and the VSS determined from the CSTR as shown in Fig. 3.

The use of this aerobic processes to decolourize azo dye-containing wastewater was not common [13], because aerobic processes are being recently used for the treatment of textile wastewater as a standalone processes [10,14–18], or in combination with anaerobic processes [12,19–22]. However, this combination between these two aerobic bioreactors demonstrated efficacy and presented many advantages. On one hand, the COD and colour removal efficiencies are higher than those obtained with CSTR and FFB bioreactors as a standalone processes. In fact, the CSTR cannot receive high WLR and can present the phenomena of washout, and the FFB can present an accelerated biofilm growth and subsequent sloughing of that biofilm. On the other hand, this combined system was able to maintain its high capacities even at high load and performed better compared with other aerobic bioreactor operating at about the same conditions [27] or combined anaerobic–aerobic systems [20].

The enhancement of biodegradation and of the decolourization efficiencies obtained with a WLR of 0.29 $g\,l^{-1}\,d^{-1}$ and a high HRT of 4 days operating conditions explained the advantage of the combined bioreactors. This long HRT of 4 days was used because the Indigo is one of the dyes that are difficult to be biodegraded confirming therefore the results obtained by Doralice and Regina [28] as shown in (Table 4).

3.5. Variation of $R_{TSS/COD}$ and $R_{VVS/COD}$ in the CSTR system

Calculated values of yields ($R_{TSS/COD}$ and $R_{VVS/COD}$) in the CSTR system are depicted in Fig. 5. During the application of the first WLR, constant values of yields were obtained and they are, respectively, of 0.45 and 0.55 g of biomass per gram of consumed substrate. These values were correlated with high COD removal efficiencies which could be attributed to biodegradation on the first time and the conversion on the second time.

For the second WLR, both $R_{TSS/COD}$ and $R_{VVS/COD}$ increased with time. This increase would be attributed to both mechanisms; the absorption of the dye on the biomass and the biodegrada-

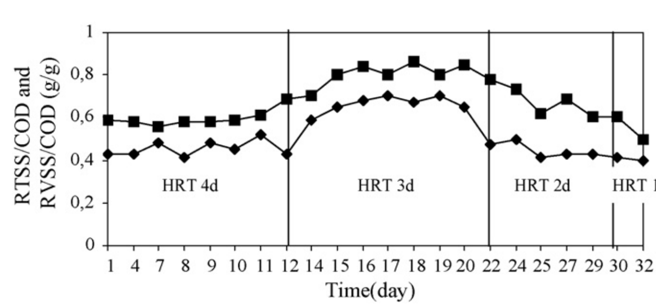

Fig. 5. Effect of the wastewater loading rates on the $R_{TSS/COD}$ (■) and $R_{VVS/COD}$ (♦) in the CSTR bioreactor.

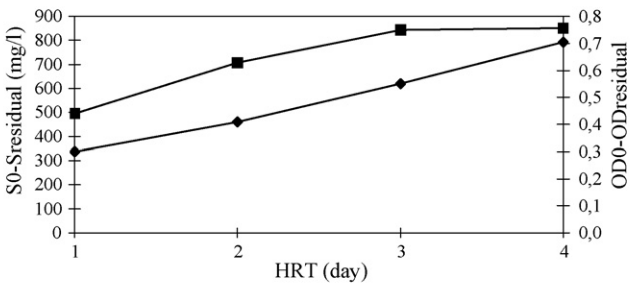

Fig. 6. Effect of HRT on the $S_0 - S_{residual}$ (■) and $OD_0 - OD_{residual}$ (♦) in the CSTR bioreactor.

tion [14]. As shown in Fig. 6, at a HRT longer than 3 days, the decolourization ($OD_0 - OD_{residual}$) still continues despite the stability of biodegradability ($S_0 - S_{residual}$); this seems to be the result of the aromatic amines conversion into new components. However, at a HRT lower than 3 days, biodegradability and decolourization are proportional. Consequently, it seems that the decolourization is the result of biodegradation enhanced by the absorption of the dye on the biomass [6,8].

For the third and the fourth WLR, the two yields decreased despite the increase of the loading rate, accompanied with a decrease of the COD removal. In addition to the reduction of the number of microbes and the reduction of biological activity in microbial consortia, as discussed previously, we can also attribute this decrease to the absorption mechanism of the dye on the biomass and to the low biodegradation time. In fact, a higher initial concentration of dye may enhance the absorption process. Bustard et al. [29] observed that the biosorptive capacity increased significantly at a higher concentration of dye. Aksu and Tezer [30] proposed that the biosorption is a result of interaction between the active groups on the cell surface such as amino acids, lipids and other cellular components of the microorganism and dye anions which are typically azo based chromophore combined with vinyl sulfone reactive groups. They suggested that activated sludge has an extensive uptake capacity for organic pollutants due to acidic polysaccharides, lipids, and other components. Chu and Chen concluded that the chemical structure, basicity and molecular weight of basic dye molecule have an influence on the absorption capacity of activated sludge biomass [31].

4. Conclusion

It has been demonstrated that an aerobic biological system can be established in a continuous flow using combined CSTR-FFB system for textile wastewater treatment. Compared with other aerobic processes, it presented a higher degradative capacity of 97.5% and a higher colour removal efficiency of 97.3%, obtained with a long HRT of 4 days and low WLR of $0.296\,g\,l^{-1}\,d^{-1}$. The performance of the system decreases with the increase of WLR. Enzymatic activities for selected strains from the bioreactor by their biodegradative capacities were investigated for the decolourization. The obtained results will be exploited for further study on the treatment of wastewater containing dyes by membrane bioreactor.

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