Environ Chem Lett (2005) 2:203–207
DOI 10.1007/s10311-004-0098-z

ORIGINAL PAPER

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Treatment of azo dye Orange II in a sequential anaerobic and aerobic-sequencing batch ractor system

Received: 24 August 2004 / Accepted: 8 December 2004 / Published online: 2 March 2005

Abstract We studied the biodegradation of Orange II in a sequential anaerobic and aerobic-sequencing batch reactor system. Granular activated carbon was used either packed into a column or added directly into the anaerobic reactor to investigate the treatment performance between the two operation conditions. We found that the circulation of mixed liquor between the anaerobic reactor and the carbon-packed column enhanced the chemical oxygen demand from 28 to 52% and Orange II removal efficiencies from 88 to 96%, under simultaneous adsorption and biodegradation process. The morphology of microbes was observed under an electron-scanning microscope.

Keywords Activated sludge · Orange II · Sequencing batch reactor · Color removal · Azo dye

Introduction

The removal of textile dyes from wastewater is one of the most important environmental issues to be solved today. Many dyes used in textile industry are particularly difficult to remove by conventional waste treatment methods since they are designed to be resistant to degradation or fading by oxidizing agents and light (Osman et al. 2004). As the characteristics of dye wastewater are very variable, many different physical, chemical and biological treatment methods have been employed for its treatment.

Reactive azo dyes are recalcitrant to microbial degradation because they have complex aromatic molecular structures; and the strong electron-withdrawing property of the azo groups is though to protect again attack by oxygenases so that the conventional aerobic wastewater treatment processes usually cannot efficiently decolorize azo dye contaminated effluents (Nigam et al. 1983). However, under anaerobic conditions, they undergo reductive fission, yielding colorless aromatic amines, compounds that in turn generally require aerobic conditions for their biodegradation (Brown and Laburer 1983). The most logical treatment strategy for complete degradation of azo dyes is therefore a sequential anaerobic–aerobic system.

One of the possibilities of using activated carbon in wastewater treatment is the direct addition of powdered activated carbon to an activated sludge system, thus combining adsorption and biodegradation. The potential superiority of powdered activated carbon-enhanced activated sludge system over a conventional activated sludge system has been demonstrated in many pilot and full-scale test treatments (Sublette et al. 1982; Martin et al. 2004). The Biological Activated Carbon-process is more effective than the consecutive adsorption and biodegradaion treatment. This is provided by the biological regeneration (bioregeneration) of activated carbon loaded with the contaminants (Alexander et al. 2001). The Biological Activated Carbon-process represents a kind of a depot of the substrate and oxygen needed for the microorganisms present on the adsorbent, protect them from too high concentration of the substrate or toxic substances. Microorganisms regenerate surface of activated carbon using organic substrate as a source of carbon and energy (Ivana et al. 1998).

The objective of this study was to investigate the decolorization of azo dye Orange II in a sequential anaerobic and aerobic sequencing batch reactor system. A granular activated carbon column was installed and the circulation of mixed liquor between the anaerobic reactor and carbon column was carried out to enhance the treatment performance under simultaneous adsorption and biodegradation process. Besides, the granular activated carbon was added into the anaerobic reactor to compare the treatment performance with granular activated carbon column, in terms of chemical oxygen demand and Orange II removals. The morphology of activated sludge in anaerobic and aerobic reactors; and granular activated carbon from column and anaerobic reactor were observed under an electron-scanning microscope and compared.

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Table 1 Treatment conditios in sequential anaerobic and aerobic-sequencing batch reactor system

Term	Time (day)	Orange II concentration (mg/l)	OLR (g COD/l-'day)	Adsorbent
1	0–24	100	5.62	–
2	25–46	100	11.24	–
3	47–69	100	11.24	GAC-column
4	70–90	100	11.24	–
5	91–114	100	11.24	GAC-SBR

OLR: organic loading rate; COD: chemical oxygen demand; SBR: sequencing batch reactor; GAC: granular activated carbon

Experimental

Two identical laboratory-scale plexiglass reactors, each with a total volume of 5l were used to simulate the activated sludge process. The reactors were operated with Fill, React, Settle, Draw and Idle periods in the ratio of 0.5:21.5:1.0:0.75:0.25 for a cycle time of 24h. In the aerobic reactor, aeration was provided during Fill and React periods by using porous air diffusers while a mixer was used in an anaerobic reactor to provide efficient mixing. In each cycle, 3l of new synthetic wastewater was added in the anaerobic reactor during Fill period and the same amount of treated effluent was removed during Draw period after settling for 1h. In the case of aerobic reactor, the influent was from the discharged of anaerobic reactor.

Activated sludge from municipal wastewater treatment plant was used as inoculums. That inoculum was selected because of a large variety of microorganisms that could be found in the biomass degrading municipal wastewaters. The activated sludge was acclimatized in the laboratory for 1 month by feeding it with synthetic wastewater. The synthetic wastewater consisted of bacto-peptone (188), sucrose (563), NH_4Cl (344), $MgSO_4$ (49), $FeCl_3$ (11.3) and KH_2PO_4 (318) giving chemical oxygen demand of 800–850mg/l. The variation of treatment conditions studied for the biological systems are shown in Table 1. In the simultaneous adsorption and biodegradation study (Term 3), a granular activated carbon column (4cm Dia./ 30cm H) was installed and the mixed liquor was circulated between the anaerobic reactor and the column. During Fill period in anaerobic reactor, the carbon-packed column was aerated with air for about 20min. The amount of granular activated carbon used was about 160g. In Term 5, the carbon was put directly into the anaerobic reactor.

The effluent of each reactor was collected at the end of the Draw period and analyzed for chemical oxygen demand according to the procedure outlined in the Standard Methods (APHA 1989). In the chemical oxygen demand and Orange II measurement, samples were prepared by filtering through a membrane filter 0.45m. The Orange II concentrations were measured in the UV-Vis spectrophotometer (UV-1200, Shimadzu) at λ_{max} 480nm. The mixed liquor suspended solids, mixed liquor volatile suspended solids, suspended solids concentrations and sludge volume index in the reactors were monitored three times per week following the Standard Methods (APHA 1989). The percentage of Orange II or chemical oxygen demand removal was calculated according to the following equation:

$$\text{Removal}\,(\%) = 100 \times \frac{C_0 - C}{C_0}$$

where C_0 is the initial concentration (mg/l), and C is the concentration after treatment (mg/l).

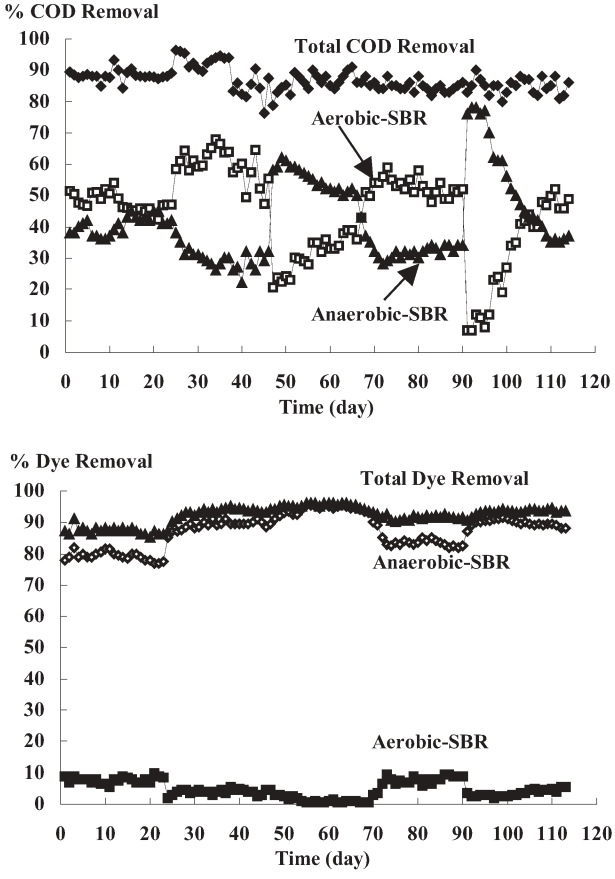

Fig. 1 (**a**) Chemical oxygen demand (COD) and (**b**) Orange II removal efficiencies in anaerobic and aerobic sequencing batch reactor (SBR) system

Results and discussion

Chemical oxygen demand and Orange II removals

Figure 1 shows the chemical oxygen demand and Orange II removal efficiency in the sequential anaerobic and

aerobic sequencing batch reactor system under various operating conditions such as organic loading rate, installation of carbon column and the addition of granular activated carbon into the sequencing batch reactor. Generally, most of the Orange II was removed in the anaerobic reactor and the balance was further reduced in the aerobic reactor. The total Orange II removal efficiency was above 85% throughout the experiment. However, it was the reverse in the case of chemical oxygen demand removal where most of the chemical oxygen demand was removed by the aerobic reactor especially in the system without the application of granular activated carbon.

The increase of organic loading rate from 5.62 to 11.24g chemical oxygen demand/lday had enhanced Orange II removal efficiency from 80 to 88% in the anaerobic reactor but decreased chemical oxygen demand removal efficiency from 42 to 28%. The lower chemical oxygen demand removal efficiency could be due to the methanogenesis process inhibited by low pH in the anaerobic reactor (about 4.5) or intermediate products produced from the biodegradation of Orange II.

During Term 3, a granular activated carbon column (4cm Dia./ 30cm H) was installed and the mixed liquor was circulated between the anaerobic reactor and the column. This study was conducted for about 23 days to investigate the feasibility of the granular activated carbon column to enhance the treatment performance by simultaneous adsorption and biodegradation process. As can be seen from Term 3 of Fig. 1, the chemical oxygen demand and Orange II removal efficiencies was improved by 28–52 and 88–96%, respectively, at the steady state of the system. During the first 3 days of carbon column installation, the chemical oxygen demand removal increased up to 62%, which was ascribed to the adsorption of organic substrates on the carbon, followed by a gradual decrease of removal efficiency as the adsorption capacity of granular activated carbon was exhausted. If bioregeneration does not occur in the carbon column, Orange II and organic materials will be adsorbed on the carbon gradually until the equilibrium concentration was reached, then the color and chemical oxygen demand removal rates will decreased along with the decreased of the adsorption capacity. The occurrence of bioregeneration will lead to a renewal of adsorptive potential, a higher stability of the system and a prolong duration of service by the granular activated carbon. In Term 4, the circulation of mixed liquor was stopped and it was found that the chemical oxygen demand and Orange II removal efficiencies dropped significantly. This indicated that the installation of carbon column had enhanced the treatment performance by simultaneous adsorption and biodegradation process.

In Term 5, the granular activated carbon was added directly into the anaerobic reactor instead of using carbon column. As similar to granular activated carbon column, the addition of carbon increased the chemical oxygen demand and Orange II removal efficiencies in the first few days due to adsorption process. However, the chemical oxygen demand removal efficiency decreased drastically after day 6 of the experiment. At steady state, the chemical oxygen demand removal efficiency was about 35%, almost closed to the system without carbon addition. However, the Orange II removal efficiency was able to maintain at 89%, about 7% higher than the system without carbon addition. As compared to the Term 3, where carbon column was used to improve the treatment efficiency, the addition of carbon into anaerobic reactor is only able to enhance Orange II but not in chemical oxygen demand removal. This could be due to the microbes growth on the carbon in column which was different from the anaerobic reactor. In addition, the aeration of carbon column with air for about 20 min during Fill period in anaerobic reactor provided enough oxygen for microbes attached on carbon to degrade the organic substrates. The results showed that the installation of granular activated carbon column and circulation of mixed liquor between the anaerobic reactor and carbon column had advantages in chemical oxygen demand and Orange II removals compared to the addition of granular activated carbon directly into sequencing batch reactor.

Changes of mixed liquor suspended solid and sludge volume index

Figure 2 depicts the changes of mixed liquor suspended solids and sludge volume index in the biological system studied. The data shown in Fig. 2 was the average of mixed liquor suspended solids, mixed liquor volatile suspended solids concentrations and sludge volume index which collected three times per week. The activity of activated sludge was represented by the ratio of mixed liquor volatile suspended solids to mixed liquor suspended solids. In the anaerobic reactor, the mixed liquor suspended solids increased gradually from Term 1 to Term 5, whereas there was a small fluctuation of mixed liquor suspended solids in aerobic reactor throughout the experiment. The increase of mixed liquor suspended solids in the sequencing batch reactors showed that the activated sludge microbes could adapt to the Orange II-containing wastewater. Meanwhile, the ratio of mixed liquor volatile suspended solids to mixed liquor suspended solids in aerobic reactor increased from 0.79 to 0.86, while in the case of anaerobic reactor, it was increased from 0.88 to average 0.91 in Terms 2 and 3, but decrease to 0.87 and 0.82 in Terms 4 and 5, respectively. The decrease of the ratio in anaerobic reactor during Terms 4 and 5 could be due to the ash contain from the granular activated carbon. The lower ratio increase in anaerobic reactor than in aerobic reactor was ascribed to the lower growth rate in anaerobic conditions because of lower energy profits in anaerobic respiration for microbes.

As shown in Fig. 2b, the average of sludge volume index in anaerobic reactor was about 37.5ml/g. On the other hand, the sludge volume index in the Term 1 of aerobic reactor was 110ml/g but decreased gradually until 45ml/g at the last two terms of experiment. These results

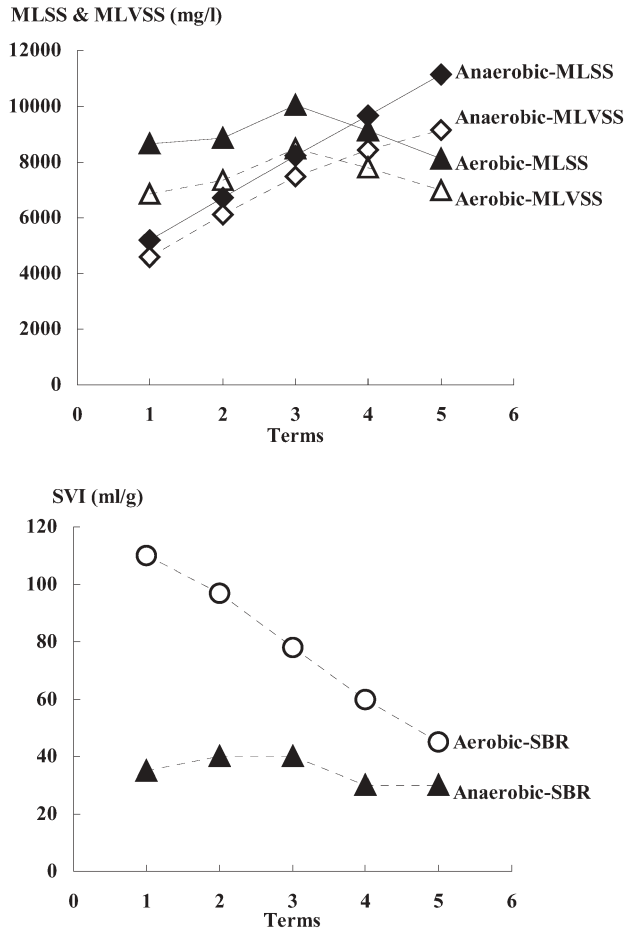

Fig. 2 The changes of mixed liquor suspended solids (MLSS) and mixed liquor volatile suspended solids (MLVSS) concentrations and sludge volume index (SVI) in anaerobic and aerobic sequencing batch reactor (SBR) system

showed that the sludge settle ability in both sequencing batch reactors were good indicated the suitability of operation periods and also the presence of Orange II in wastewater inhibited the excessive growth of filamentous bacteria.

Microscopic images of activated sludge microbes

The microbial observation of suspended and attached microorganisms in anaerobic and aerobic reactors; and granular activated carbon from column and anaerobic reactor was carried out. The electron-scanning microscope photographs for Fig. 3a–b,c and d was taken at Terms 2, 3 and 5, respectively. The different patterns of biomass could be observed. It was found that the microorganism's morphology was different among the anaerobic and aerobic reactors; and granular activated carbon from column and anaerobic reactor. As shown in Fig. 3a, we observed that the cocci-shaped bacteria were the dominant microbes in the aerobic reactor with diameter about 2μm. In the case of anaerobic reactor (Fig. 3b), the

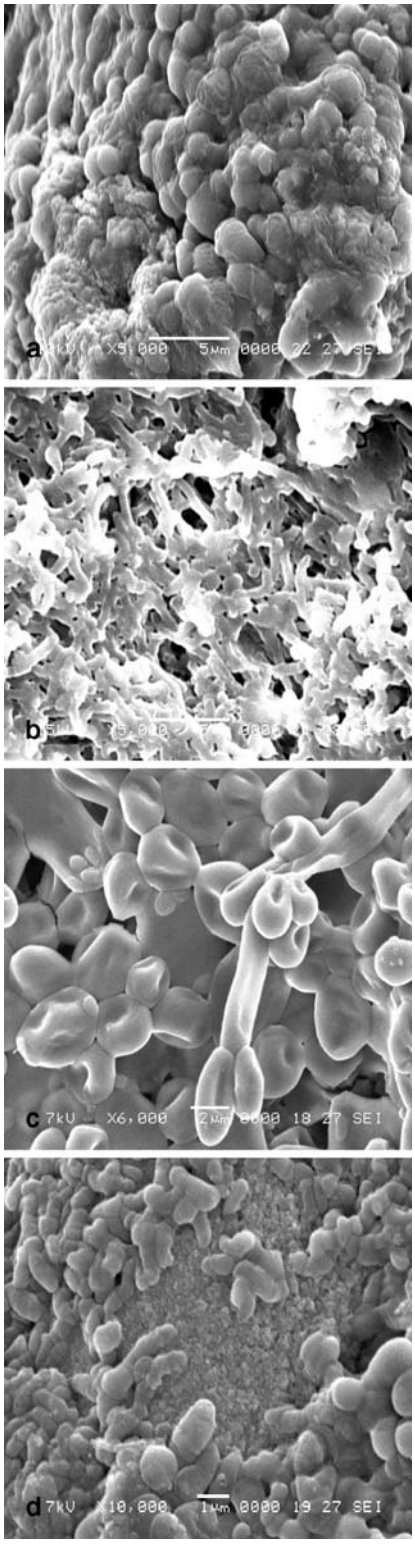

Fig. 3 Microscopic images for activated sludge in (**a**) aerobic sequencing batch reactor (**b**) anaerobic sequencing batch reactor (**c**) granular activated carbon column and (**d**) granular activated carbon from anaerobic sequencing batch reactor

microscopic structure was in the rod-shaped or filament form. The high removal efficiency of Orange II could be due to this microbe in anaerobic reactor. Figure 3c and d showed that there were a lot of microbes attached on the surface and macro-pore of granular activated carbon. The microscopic images between the carbon from column and anaerobic reactor have some differences. The dominant microbes attached on the carbon from column were spherical-shaped with average 2μm in diameter. Besides, the filament-shaped microbes also observed in the system. On the other hand, the dominant microbes on the carbon from anaerobic reactor were in a small rod-shaped with diameter and length of 1 and 2μm, respectively. The discrepancies in the microorganism's composition of the granular activated carbon, could be the reason for different treatment efficiencies in chemical oxygen demand of the two operation conditions employed.

Conclusion

The increase of organic loading rate from 5.62 to 11.24g chemical oxygen demand/ld had enhanced Orange II removal efficiency from 80 to 88% in anaerobic sequencing batch reactor but decreased chemical oxygen demand removal efficiency from 42 to 28%. The circulation of mixed liquor between the anaerobic sequencing batch reactor and granular activated carbon column had enhanced the chemical oxygen demand and Orange II removals under simultaneous adsorption and biodegradation process, whereas the addition of granular activated carbon into anaerobic sequencing batch reactor just improved the Orange II removal efficiency. The different morphology of microbes observed under electron-scanning microscope in anaerobic and aerobic sequencing batch reactors; and granular activated carbon from column and anaerobic sequencing batch reactor could be the reason for different removal efficiency in chemical oxygen demand and Orange II between the systems and operation conditions employed.

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