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A Horizontal Plug-Flow Baffled Bioelectrocatalyzed Reactor for the Reductive

Decolorization of Alizarin Yellow R

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Abstract:

An application-oriented membrane-free, continuous plug-flow baffled bioelectrocatalyzed reactor (PFB-BER), was designed and testified for the decolorization of Alizarin Yellow R. Decolorization efficiency (DE) with an external power source of 0.5V was higher than without electrolysis, i.e. 93.4% versus 73.6% (HRT of 24h). Product formation efficiencies of p-phenylenediamine and 5-aminosalicylic acid were above 95% and 50%, respectively. When HRT decreased to 8h and 4h, DE reduced to 69.9% and 44.9%, respectively and the corresponding electron recovery efficiency increased from 151.2% (HRT of 24h) to 340.1% (HRT of 8h). An additional electrode assembly improved DE to 96.4% (HRT of 8h) and 80% (HRT of 4h), while energy consumption (HRT of 4 h) was lower than that of HRT of 12 h with single electrode assembly under comparable DE. The PFB-BER with higher removal capacity, lower internal resistance and energy consumption provides a new solution to treat the high loading azo dye-containing wastewaters.

Key words: Plug-flow baffled bioelectrocatalyzed reactor (PFB-BER); Alizarin Yellow R (AYR); Decolorization efficiency (DE); Electrode assembly number

1. Introduction

Azo dye, characterized by one or more azo groups ($-N=N-$) in its chemical structure, is the largest and most versatile class of synthesized dyes widely applied in textile, leather, and cosmetics (Mu et al., 2009). It is estimated that between 2% and as much as 50% of different kinds of dyes would be discharged along with wastewater eventually (Tan et al., 1999), resulted in a series of environmental, ecological and aesthetic problems. Most of azo dyes are toxic, mutagenic and persistent to biodegradation, therefore, the efficient and applicable [technology](#) is in urgent need for the effective control of azo dyes contamination (Ayed et al., 2011; Solanki et al., 2013). Most of azo dyes are resistant to aerobic microbial degradation, while the azo bond could be [reduced](#) to aromatic amines under the anaerobic environment. The produced amine metabolites are less toxic and can be easily biodegraded aerobically (Tan et al., 1999).

Recently, bioelectrochemical systems (BESs) have emerged great advantages in dealing with desalination, decolorization and heavy metals with the [higher](#) efficiency (Solanki et al., 2013). Several studies have reported the reductive decolorization of azo dyes by the quick cleavage of azo bonds in either traditional dual-chamber BESs (Ding et al., 2010; Kong et al., 2014; Kong et al., 2013; Liu et al., 2009; Mu et al., 2009; Sun et al., 2011) or BESs with the improved configurations (Cui et al., 2012; Hou et al., 2011; Sun et al., 2013; Wang et al., 2013). These studies suggested a promising potential employment of BES for the enhanced degradation of azo dye-laden wastewater.

Considering the application-oriented system, most of the reported lab-scale BES modules would be restricted during enlargement in costs, operability and feasibility. For example, the dual-chamber systems possessed the higher internal resistance, leading to the lower electron transfer efficiency and the higher running cost. In comparison, the

single-chamber BESs, such as air-cathode single-chamber BES (Sun et al., 2012) or up-flow single-chamber BES (Cui et al., 2012), possessed with a low internal resistance or reduced construction and operation costs. However, in term of scale enlargement, the scalability of up-flow BESs is restricted based on its size, weight and excess energy requirement to overcome the gravitational effects.

Therefore, aiming at the large-scale treatment of azo dye-containing wastewaters, a lab-scale reactor were designed and operated, namely plug-flow baffled bioelectrocatalyzed reactor (PFB-BER), to testify the decolorization activity and efficiency of azo dye Alizarin Yellow R (AYR), one of the most popular azo dyes applied in industry, was used as the targeted pollutant. Electrochemical impedance spectroscopy (EIS) and cyclic voltammetry (CV) were employed to evaluate the electrochemical characteristic of the PFB-BER during the decolorization process. The decolorization efficiency, metabolites formation, current density and energy consumption under the key operational parameters including different loading rates and numbers of electrode assembly were also analyzed and evaluated.

2. Materials and methods

2.1. PFB-BER configuration

The PFB-BER was made of plexiglas and constructed of three channels (length of 20 cm, depth of 5 cm, and width of 5 cm for each channel) sealed tightly to ensure the anaerobic environment. Total volume and effective volume of the PFB-BER was 1.50 and 1.25 L, respectively. Graphite granular (ID from 3.0 to 5.0 mm) filling in titanium baskets (length of 45 mm, height of 30 mm, and width of 45 mm) were adopted as cathode and anode, respectively, with the volume ratio of 2:1. The schematic diagram of the PFB-BER configuration was shown in Fig. S1.

2.2. *The PFB-BER operation and inoculation conditions*

Four identical reactors were constructed at the same time, two reactors in the closed circuit with 0.5 V power supplied by a DC power (IT6921, Itech Co. Ltd., USA) and external resistance of 20 Ω . The other two were in the opened circuit without power supply as control. A saturated calomel electrode (SCE, +247 mV vs. Standard hydrogen electrode, SHE) (Shanghai Precision Scientific Instruments Co. Ltd., China) was placed between the anode and cathode as reference electrode. Data of anode and cathode potentials as well as reaction currents were recorded using data acquisition equipment (Keithley 2700, Keithley Co. Ltd., USA). All potentials reported were against SCE.

Continuous flow tests were conducted with the nutrient medium described in detail by Cui et al (2014a) and a gradual increased AYR concentration from 20 to 100 mg/L. Acetate (NaAc) worked as the potential carbon source and additional electrons donor and the targeted pollutant AYR was as electrons acceptor for cathode. A peristaltic pump (BT100-1L, Longer Pump Co. Ltd., China) was allied for the continuous feed. All reactors were inoculated with 125 mL of activated sludge (total suspended solids of 15695 ± 260 g/L) collected from Taiping wastewater treatment plant (Harbin, China).

Reactors were started when anode potential reduced to below -300 mV and the corresponding cathode potential was below -800 mV. The lowest cathode potential was determined as -897.1 ± 12.6 mV. After AYR increased to 100 mg/L, system was maintained with the hydraulic retention time (HRT) of 24 h. Then the running conditions were altered, such as the HRT was shortened to 12, 8 or 4 h, respectively, and the number of electrode assembly was doubled, to testify the AYR decolorization and products formation efficiency, as well as energy consumption. Each condition were kept running for at least one month to guarantee the steady running state.

2.3. Chemical and electrochemical analysis

The determination methods of AYR and two main decolorization products of AYR, *p*-phenylenediamine (PPD) and 5-aminosalicylic acid (5-ASA), the calculation methods including the decolorization efficiency (DE, %), the decolorization rate (DR, mol m⁻³ TV d⁻¹), the products formation efficiency (FE, %), the electron recovery efficiency (ERE_{AYR}) and energy consumption (EC, Kwh mol⁻¹ AYR), as well as cyclic voltammetry (CV) and electrochemical impedance spectroscopy (EIS) were described in detail in supplementary information (SI).

3. Results and discussion

3.1. AYR decolorization and electrochemical characteristics in the PFB-BER

AYR removal efficiency approached to 93.4 ± 1.4% in the closed circuit mode, while only 73.6 ± 2.9% of AYR was removed in the opened circuit mode (Fig. S2). The measured chroma is less than 80 (data not shown), which approached the textile wastewater discharge standard II in China. These results indicated the PFB-BER mode improves the AYR removal efficiency and decolorization activity.

The bioelectrochemical reduction behavior of AYR was analyzed by CV (Fig. 1A). When AYR was initially supplied, two obvious irreversible reductive peaks at -0.92 and -0.59 V (vs. SCE) were observed on the biocathode. The two reduction peaks had been proved as nitro (-NO₂⁻) and azo bond (-N=N-) reduction, respectively (Cui et al., 2014a). In contrast, no obvious reduction peak was found by CV analysis after 24h, attributed to the lack of reductive compounds in cathode. The results confirmed the catalytic behavior of biocathode during AYR decolorization.

Impedance response of the whole cell, anode and cathode in the PFR-BER was analysed. As shown in the Nyquist plots (Fig. 1B), R_{ohm} of the whole cell, anode and

cathode was 46.7, 23.8 and 21.9 Ω , respectively. Correspondingly, R_{ct} was 63.6, 19.3 and 48.1 Ω , respectively. The lower R_{ct} of the anode suggests that biofilm formation on the anode was responsible for the increase in anodic reaction rate and decrease in its inner resistance. The total resistance of the reactor was 121.1 Ω , which was much lower than that of the traditional dual chamber BES (774 Ω) (Cui et al., 2011). The results indicated the membrane-less design of the PFB-BER could be a better BES configuration for the treatment of pollutants.

3.2. The AYR decolorization efficiency under different HRTs

The AYR decolorization efficiency (DE) decreased with the increased loading rate of the influent AYR via shortening HRT (Fig. S3). When the influent AYR loading rate increased from 106.2 to 318.7 $\text{g m}^{-3} \text{TV d}^{-1}$ (corresponding to the shortened HRT from 24 to 8 h), the AYR DE decreased from 93.5 ± 4.7 to $69.9 \pm 3.5\%$. The lower efficiency with HRT of 8 h indicated the exceeding of cathode treatment capability. In comparison, HRT of 12 h with the loading rate of $318.7 \pm 10.6 \text{ g m}^{-3} \text{TV d}^{-1}$ was considered as **more suitable** with a shorter time than 24 h and the higher DE of 85.4%, more than 1.23 times compared with that of HRT 8h.

In the BES, the AYR reductive decolorization went through two processes, the initial reduction of AYR to 5-ASA and *p*-nitroaniline and then the further reduction of *p*-nitroaniline to PPD happened (Cui et al., 2014a). The generation of the decolorization products 5-ASA and PPD at different HRTs was shown in Fig.2A. *P*-nitroaniline was undetected in effluents under all of the tested conditions in this study. The formation efficiency of 5-ASA was only about 50% to 70% and gradually increased with the shortened HRT from 24 to 8 h. When the HRT was shortened, the accumulated AYR concentration and the formation efficiency of 5-ASA both increased. In comparison, the

formation efficiency of PPD approached to 95-100% under all of the tested conditions, indicating the complete reductive decolorization of AYR. No hydrogen was detected, indicated a much lower hydrogen generation potential than the theoretical (-414mV vs SHE, pH=7) caused by the system over potential. Meanwhile, no methane was able to be detected, so the side effect brought by methane generation can be neglected.

As shown in Fig. 2B, when the HRT was shortened from 24 to 8 h, the anode potential increased from -475 to -405 mV, and correspondingly, the cathode potential increased from -937 to -820 mV. The current density gradually grew from 15 to 20 Am^{-3} , attributed to an elevated loading rate of AYR. The calculated electron recovery efficiency (ERE) for AYR decolorization approached to 151.2, 239.2, and 340.1% under the HRT of 24, 12 and 8 h, respectively. The increase of ERE via reducing HRT indicated that the ratio of the pure biodegradation to the electrolysis participated in degradation likely gradually increased.

3.3. Enhanced AYR decolorization by an additional electrode assembly

To improve AYR DE under the shortened HRT (8 and 4 h), another group of electrode assembly was inserted, and the running results were shown in Fig. 3. By an additional electrode assembly, AYR DEs improved about 26.4 and 35.1% at HRT of 8 and 4 h, respectively. AYR DEs at HRT of 8 and 4 h with two electrode assemblies were comparable with the conditions with HRT of 24 and 12 h applying the single electrode respectively. The enhanced AYR removal efficiency was attributed to the improvement of electrochemical reaction surface. However, the anode and cathode potentials were not obviously changed.

The energy consumption (EC) decreased gradually as HRT shortened after applying two groups of electrode assembly (Fig. S4). EC at HRT of 8h (about 0.272×10^{-3}

kWh·mol⁻¹) was much higher than the condition with HRT of 24h applying single electrode assembly (0.104×10^{-3} kWh·mol⁻¹), although their DE was comparable to each other. However, under the similar DE, the one with HRT of 4h consumed lower energy (about 0.05×10^{-3} kWh·mol⁻¹) compared to the condition with HRT of 12h applying single electrode assembly (0.059×10^{-3} kWh·mol⁻¹). The results indicated the more groups of electrode assembly inclined to treat the higher loading rate AYR with advantage of both removal efficiency enhancement and energy conservation.

3.4. Outlook

Based on the great interest in exploring alternative technologies to face the huge discharge of the high toxic compound-laden wastewater, development of cost-effective BES configuration with the high applicable potential (Solanki et al., 2013) and energy reservation characteristic is in urgent need (Chen et al., 2010; Sun et al., 2009). In this study, a novel system, application-oriented horizontal continuous-flow PFB-BER was designed as the simulated modified biological treatment unit in sewage treatment plant. Through the running parameter tests (loading ratios and cathode assembly numbers) and feasibility analysis (effects and energy consumption), the PFB-BER designed in this study showed superior decolorization and energy saving characteristics.

PFB-BER was design without the membrane, which largely decreased cathode R_{ct} to 63.6 Ω and the total R_{ct} to 121.1 Ω (Fig. 2B), compared with the other modified BESs ever reported (148.9 Ω , single-chamber BES, Wang et al., 2013; 617 Ω , double-chamber MFC, Sun et al., 2011). The lower resistance gave rise to a higher current, would eventually facilitate the electron transfer and reduce the running cost. Importantly, one group electrodes took up 15.0% of the reactor volume, much lower compared with the other reported application-oriented BESs, including the up-flow biocatalyzed

electrolysis reactor (UFBER) (28%) (Cui et al., 2012) or the modified integrated BES system (55.7%) (Kong et al., 2015). Additionally, the horizontal type configuration avoided the extra costs brought by the global gravity, like some up-flow type BES (Cui et al., 2012), is more suitable to deal with large amount of wastewater in the sewage treatment plant or as the simulation for *in situ* bioelectrochemical remediation of river water body.

The removal capacity of AYR approached 622.2 ± 40.5 , 1005.1 ± 132.8 , 1919.3 ± 253.7 and 2616.2 ± 237.0 g m⁻³ TCV d⁻¹, under HRT of 24, 12, 8 and 4 h, respectively, calculated by multiplying the influent flow rate (m³ d⁻¹) with subtraction of the removed concentration of AYR (g L⁻¹) (Fig. S3) and dividing the total cathode volume (m⁻³). The AYR removal capacity obtained in this continuous flow system was relatively higher, compared with the one achieved in UFBER (about 1000 g m⁻³ TCV d⁻¹ with HRT of 8 h; about 1500 g m⁻³ TCV d⁻¹ with HRT of 4 h) and a pilot-scale anaerobic baffled reactor coupled with biocatalyzed electrolysis reactors (about 1200-1700 g m⁻³ TCV d⁻¹ with HRT of 8 h and 1400-2300 g m⁻³ TCV d⁻¹ with HRT of 4 h), respectively (Cui et al., 2014ab), as calculated in the same way. AYR decolorization resulted in two main metabolites, namely 5-aminosalicylic acid (5-ASA) and *p*-phenylenediamine (PPD) (Fig. 2A) was proved by CV and HPLC analysis, which largely reduces the toxicity of AYR and would facilitate the aerobic biodegradation process. Besides, setting up more groups of electrodes effectively enhanced the decolorization process with the higher loading rate and the lower energy consumption. Further study is needed focusing on the electrode improvement and cathode biofilm community and function analysis to give better solutions on improving the system removal efficiency and reducing the energy consumption.

It is noticed under all of the conditions, the recovery rate of PPD approached to 95%-100% and while, 5-ASA was only about 50%-70%, consistent with one of the studies on AYR reduction in up flow type BES (biocathode followed with bioanode) (Cui et al., 2012). This indicated that almost all *p*-nitroaniline was converted into PPD and while, PPD was resisted to be further degraded. The low recovery rate of 5-ASA was probably attributed to its further degradation to propionate or acetate by anaerobic microorganisms (e.g., acetogenic fermenters and methanogens) (Cui et al., 2014 a; 2014b; Pandey et al., 2007) or by bioanode compartment after the flow pass through. The possibility of electron generation by degradation of either 5-ASA or its metabolites in bioanode is not excluded and the potential contribution to biocathode requires the further investigation.

4. Conclusion

In this study, an application-oriented PFB-BER was designed and constructed, with an outstanding bioelectrochemical property, the superior AYR decolorization performances and lower energy consumption. The current study provides a new concept for BES scaling-up to energy-efficient treatment of azo dye-laden wastewaters.

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Figure Legend

319 **Fig.1** The decolorization of 100 mg L⁻¹ of AYR in the closed and opened circuit of
320 PFB-BER with HRT of 24 h. **A:** Cyclic voltammetry (CV, scan rate of 10mV/s) in the
321 closed circuit with AYR of 100 mg L⁻¹ at 0 and 24 h, respectively; **B:** The Nyquist plots
322 of anode, cathode and the whole cell by EIS analysis.

323 **Fig.2** The decolorization of 100 mg L⁻¹ of AYR under different HRTs of 24, 12 and 8 h.
324 **A:** The concentration distribution of AYR and the metabolites (PPD and 5-ASA) and
325 formation efficiency of PPD and 5-ASA in effluents. **B:** Potentials of anode and cathode
326 and the current density.

327 **Fig.3** The anode/cathode potential and the decolorization efficiency of AYR with one
328 and two electrode assembly under HRTs of 8 and 4 h;

329

Figure. 1

Figure. 2

Figure. 3