Bioaugmentation treatment of PV wafer manufacturing wastewater by microbial culture

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ABSTRACT

The wastewater of silicon photovoltaic (PV) battery manufacturing contained polyethylene glycol (PEG) and detergents, which possessed the characteristics of high content of organics and low bioavailability, and then resulted in high treatment costs. To address the difficulties of existing treatment facilities in stably meeting discharge standards, eight tons of microbial culture (consisting of *Bacillus* sp. and *Rhodococcus* sp.) were added into the aerobic treatment unit. Subsequently, the effectiveness of the microbial culture in small-scale biological wastewater treatment was evaluated, and the operating conditions for engineering applications were optimized. The application study showed that the average chemical oxygen demand (COD) removal efficiency reached 95.0% when the pH value was 7, the gas–water ratio was 28:1, the reflux ratio was 50%, which indicated an increase of 51.2% contrasting with the situation without bioaugmentation. The volume load of the treatment facilities after augmentation increased by 127.9% and could tolerate the COD shock load reached 2,340 $mg \cdot L^{-1}$. At last, the effluence met the class I standard of the *Integrated Wastewater Discharge Standard* (GB8978–1996).

Key words | bioaugmentation, bio-refractory pollutants, microbial culture, PV wafer manufacturing, shock load

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INTRODUCTION

Several companies are engaged in the production of photovoltaic (PV) silicon batteries, resulting in the generation of wastewaters including wastewater from acidic washing processes, cleaning processes, and alkaline washing processes. The organic pollutants in the wastewater mainly consisted of polyethylene glycol (PEG) and detergent. PEG is produced by the polymerization of ethylene oxide and has good water solubility (Bernhard *et al.* 2008). At present, membrane filtration and various existing physical and chemical wastewater treatment methods such as active carbon adsorption, O_3 oxidation, and H_2O_2 oxidation cannot effectively treat PEG (Zhao *et al.* 1989); in particular, a larger molecular mass is associated with lower removal efficiency (Watson & Jones 1977). The existence of a high amount of surfactants in detergent also leads to difficulty in wastewater treatment. As a result, traditional biochemical techniques cannot achieve stable quality for the discharged water. Pretreatments such as high-level oxidation and internal electrolysis to reduce the concentration of organic materials requires the addition of a large amount of pharmaceutical reagents, which results in extremely high cost and does not have satisfactory effectiveness (Ji *et al.* 2013).

Bioaugmentation originated in the mid-1970s (Young 1976) to enhance the ability of wastewater treatment systems to remove pollutants by adding functional microbes to the system or polluted sites (Fantroussi & Agathos 2005). Incorporation of bioaugmentation technology into traditional bioremediation has become a developmental trend in this field (Alves *et al.* 2002) and has high feasibility (Fujita *et al.* 2000). Screening of highly competitive, adaptable, and effective bacterial strains is a decisive factor for bioaugmentation technologies (Wang *et al.* 2007). Current research in China and abroad indicates that bioaugmentation treatment of wastewater containing PEG and detergents is feasible. Kawai *et al.* (1978) proposed PEG metabolic pathways by studying the PEG degradation products and changes in enzymatic activity after the treatment of co-cultured symbiotic flavobacteria and *Pseudomonas*. Zgoła-Grześkowiak *et al.*

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(2006) used active sludge to treat PEG300 and achieved a degradation rate of 99%. Yamashita *et al.* (2005) found that *Pseudonocardia* sp. strain K1 can biodegrade PEG6000 via aerobic metabolic pathways. Huang *et al.* (2005) studied the biodegradation effect of sludge on simulated wastewater containing PEG600, PEG6000, and PEG20000 under both anaerobic and aerobic conditions and found that the aerobic degradation rates of all three wastewaters reached approximately 80% within five days. Ojo & Oso (2008, 2009) isolated and identified detergent-degrading bacteria from wastewaters and conducted laboratory biodegradation simulation studies on 76 wastewater samples collected from different industries. It is concluded that synthetic detergents are essentially biodegradable.

The afore mentioned studies have provided guidance for the bioaugmented treatment of wastewater; however, to date, no studies on engineering applications using bioaugmentation methods alone to treat the aforementioned mixed wastewaters have been reported. In this study, after the fermentation of two mixed bacterial strains, a bioaugmentation method was applied to aerobically treat wastewater containing both PEG and detergents without any pretreatments through acidification, micro-electrolysis, or advanced oxidation techniques. The presented method can solve a variety of drawbacks in previous wastewater treatment processes without bioaugmentation, such as discharge of water below quality standards, difficulty in removing chemical oxygen demand (COD), complicated operating procedures, and high operational costs.

MATERIALS AND METHODS

Feed water and microbial cultures

The characteristics of influent wastewater in the present study are shown in Table 1.

The microbial culture consisted of PEG-degradation bacteria (*Bacillus* sp.) and detergent-degradation bacteria (*Rhodococcus* sp.). Two strains were intermediate cultured in flasks, then mixed (1:1) in the fermenter. Finally, 8 tons microbial culture were achieved.

The medium of microbial culture consisted of two parts: (1) the basal medium: $2\,g \cdot L^{-1}$ Na_2HPO_4, $0.5\,g \cdot L^{-1}$ KH_2PO_4, $0.5\,g \cdot L^{-1}$ $(NH_4)_2SO_4$, $0.3\,g \cdot L^{-1}$ $MgSO_4 \cdot 7H_2O$, $5\,ml \cdot L^{-1}$ trace element solution (Kaminski *et al.* 1983), pH 7.0; (2) carbon source: $5\,ml \cdot L^{-1}$ of PEG (provided by the manufacturer) and $5\,ml \cdot L^{-1}$ of detergent (provided by the manufacturer) were added in the *Bacillus* sp. culturing medium and *Rhodococcus* sp. culturing medium, respectively.

Previous wastewater treatment process

The process of previous wastewater treatment without bioaugmentation is presented in Figure 1.

Small-scale experiment

Microbial culture was inoculated into the PEG-containing wastewater, detergent-containing wastewater, and PEG-detergent-mixed wastewater (mixed at a ratio of 1:3 according to the actual conditions) in a 1-L reactor by a ratio of 20%. (1-L reactor used in small-scale consisted of two major components, the 1-L glass volumetrical cylinder and air pump, which afforded sufficient dissolved oxygen (DO) above $2.0\,mg \cdot L^{-1}$. The mixed liquor suspended solid (MLSS) executed in this reactor was the same as the full-scale reactor.) At the same time, the sludge from the contact oxidation tank was inoculated into the mixed wastewater at a MLSS content of approximately $2{,}500\,mg \cdot L^{-1}$ in a 1-L reactor as a control (no bacterial culture added). After aeration, samples were collected at different time points to determine the COD. Through a

Table 1 | The characteristics of influent wastewater

Content	Total stream	PEG wastewater	Detergent wastewater
Quantity of wastewater, $m^3 \cdot d^{-1}$	872 ~ 1,765	270 ~ 510	492 ~ 1,397
Mean quantity of wastewater, $m^3 \cdot d^{-1}$	1,584 ($\sigma^a = 178$)	337 ($\sigma = 45$)	1,211 ($\sigma = 170$)
Range of COD, $mg \cdot L^{-1}$	520 ~ 2,408	2,171 ~ 6,103	381 ~ 1,098
Mean COD, $mg \cdot L^{-1}$	1,098 ($\sigma = 372$)	2,699 ($\sigma = 821$)	685 ($\sigma = 267$)
Range of pH	5 ~ 6	4.5 ~ 6.5	5 ~ 6
Mean pH	5.5	–	–

[a]σ refers to the standard deviation.

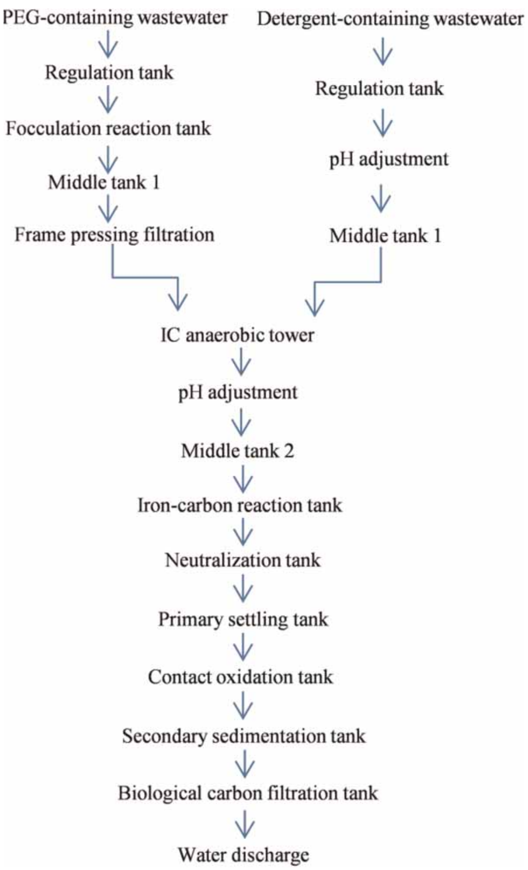

Figure 1 | Previous wastewater treatment process.

comparison with the controls, the feasibility of the microbial culture for degrading the pollutants was determined.

Full-scale experiment engineering application

The contact oxidation tank is a five-channel push-flow reactor pond. Eight tons of mixed microbial culture were added into the contact oxidation tank in three days, which the dosage on first, second and third days amounted to 4 tons, 2 tons, and 2 tons, respectively. After the augmentation treatment with the inoculated bacteria, several parameters including the system pH, gas–water ratio, reflux ratio, and shock load were investigated. The experimental design was shown in Table 2.

Based on Table 2, different factors were set up. Each experiment was conducted under the above optimal condition of several levels, except for the (A). Optimal pH experiment, the (A), was carried out in 1-L reactor, while others were in contact oxidation tank.

Table 2 | Experimental programme

Section	Factors	Variable values	Test items and frequency
(A)	Optimal pH	pH = 6,7,8	COD in 1 L reactor, every 3 h
(B)	Optimal gas–water ratio	28:1 & 56:1	COD in contact oxidation tank, at 24 h after start-up
(C)	Optimal reflux ratio[a]	100 & 50%	COD in contact oxidation tank, at 24 h after start-up
(D)	Shock load of the bioaugmentation system	About 2,400 $mg \cdot L^{-1}$	Effluent COD of system, every 24 h

[a]Reflux ratio was the ratio of reflux sludge from secondary sedimentation tank to contact oxidation tank and the influent flow of contact oxidation tank.

Analytical methods

The concentrations of COD was determined by procedures given in the standard methods (National Environmental Protection Bureau 2002), pH was determined by a pH meter (Mettler-Toledo SG2). The calculation and statistical analysis were performed using Microsoft Excel 2007.

RESULTS AND DISCUSSION

COD removal by previous wastewater treatment process without bioaugmentation

The detection data, between 2012 and 2013, showed that as well as using previous wastewater treatment process without bioaugmentation, pretreatments such as anaerobic treatment and iron–carbon micro-electrolysis had poor performance in removing COD. As shown in Figure 2, the COD in the influent water in the contact oxidation tank was in the range of 1,088–1,661 $mg \cdot L^{-1}$, with an average value of 1,379 $mg \cdot L^{-1}$; the COD in the effluent water range was in the range of 569–1,125 $mg \cdot L^{-1}$, with an average value of 673 $mg \cdot L^{-1}$; and the average removal efficiency of COD was 48.8%. It was difficult for final effluent COD in the treatment to meet the class I standard of the *Integrated Wastewater Discharge Standard* (GB8978–1996).

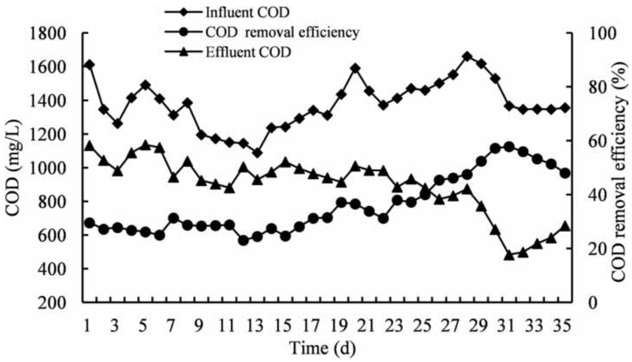

Figure 2 | COD removal performance using previous wastewater treatment process without bioaugmentation.

Mackul'ak *et al.* (2013) indicated that the antibiotic function and slow biodegradation of PEG are the main contributors to the difficulty in the removal of COD. In addition, linear alkyl benzene sulphonate, the main component of the anionic detergent, also contributes a large amount of biorefractory organisms (Ojo & Oso 2008).

Small-scale test on wastewater augmentation treatment of microbial culture

As shown in Figure 3 and Table 3, the COD levels in the three types of bacteria-treated wastewaters all showed a continuously decreasing trend. Within 15 h, the removal rate of bacteria-treated mixed wastewater was $28\ mg \cdot L^{-1} \cdot h^{-1}$, showing a superior removal outcome compared with the non-bacteria-treated sample that had a removal rate of $5\ mg \cdot L^{-1} \cdot h^{-1}$. Within 46 h, the removal efficiency of treated mixed wastewater sample reached 78.9%, which was higher than that in the non-bacteria-treated sample at 17.3%. The results from the small-scale test indicated that bioaugmentation treatment of mixed wastewater yielded the better outcome, and within 63 h, the COD removal amount was $1,114\ mg \cdot L^{-1}$ higher than the non-bacteria-treated mixed wastewater. It is speculated that the two bacterial strains' enzyme systems complement each other, resulting in loop-opening of the PEG and surfactant macromolecules to form small molecules that are easily biodegraded.

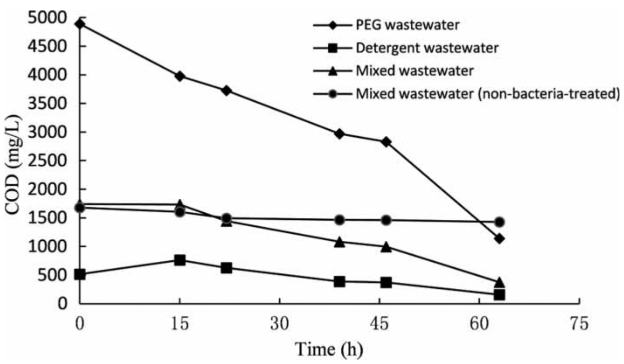

Figure 3 | Small-scale test for COD removal performance after field bioaugmentation treatment of wastewater.

Table 3 | Data of small-scale test for COD removal performance

Time, h	PEG wastewater, $mg \cdot L^{-1}$	Detergent wastewater, $mg \cdot L^{-1}$	Mixed wastewater, $mg \cdot L^{-1}$	Mixed wastewater (non-bacteria-treated), $mg \cdot L^{-1}$
0	4,889	514	1,743	1,680
15	3,976	764	1,731	1,604
22	3,726	629	1,447	1,495
39	2,971	388	1,084	1,467
46	2,829	373	998	1,462
63	1,140	160	376	1,427

In addition, the removal rate in 15 h and removal efficiency in 46 h of PEG wastewater and detergent wastewater were $60\ mg \cdot L^{-1} \cdot h^{-1}$, $13\ mg \cdot L^{-1} \cdot h^{-1}$ and 76.7%, and 79.1%, respectively, indicating that this microbial culture could tolerate about $4,889\ mg \cdot L^{-1}$ of COD, and the removal rate was correlated with the initial COD concentration.

After 46 h, the COD removal rate of all three wastewaters rapidly increased, mainly because of the gradual accommodation of microbial strains to the environment and increase in the number of bacteria. As a result, the mixed-culture bacteria grew well, and this treatment method showed satisfactory performance in treating wastewater and was thus used in engineering practice for further verification.

Engineering applications of augmentation of wastewater treatment by microbial culture

Optimization of parameters of bioaugmentation treatment systems

The effects of pH value, gas–water ratio, reflux ratio, and shock load on the bioaugmentation system were studied, and the results are shown in Figure 4. As displayed in Figure 4(a), when the pH value was 7 or 8, the COD removal efficiency of the simulated system was relatively high, as evidenced by the COD removal amounts of, respectively, $245\ mg \cdot L^{-1}$ and $302\ mg \cdot L^{-1}$ after 3 h, followed by a

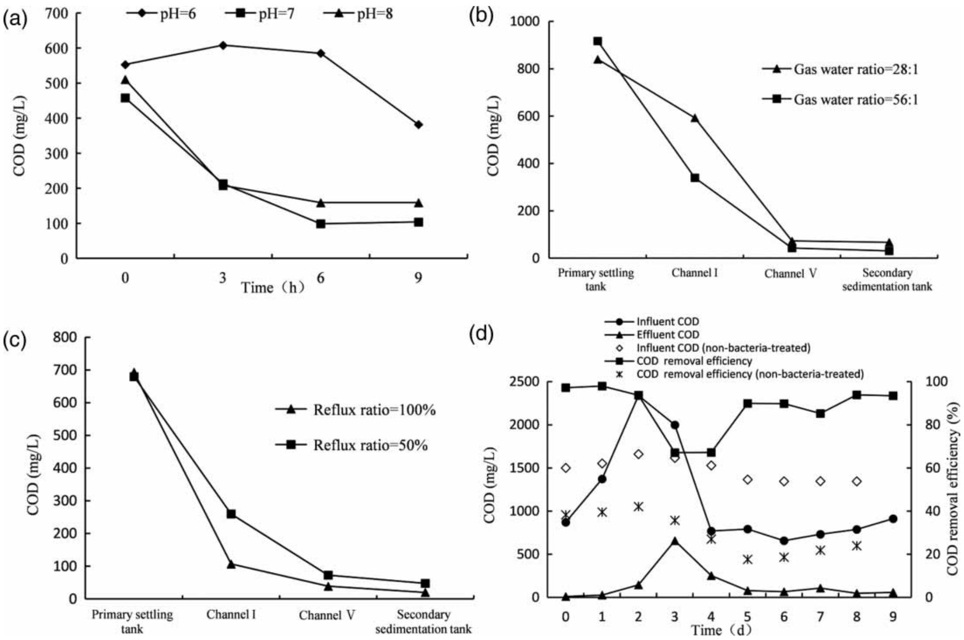

Figure 4 | Effect of pH on COD removal (a), gas–water ratio on COD removal (b), reflux ratio on COD removal (c), and shock load on COD removal (d) in the bioaugmentation system.

decrease to, respectively, 99 $mg \cdot L^{-1}$ and 159 $mg \cdot L^{-1}$ after 6 h. The removal amount was approximately 11 times higher than observed at a pH value of 6. These results indicated that neutral or mild alkaline conditions are suitable for the growth and metabolism of this microbial culture, possibly because extreme acidic or alkaline environments can change the electric charge distribution on the microbial cell membrane and the enzymatic activity during metabolism, thus hindering the growth of, or even leading to the death of, the microbes (Jiang *et al.* 2013).

The on-site wastewater pH value ranged from 5 to 6; thus, to use the microbial culture for the bioaugmentation treatment of wastewater, it was necessary to adjust the pH value of the wastewater to 7 prior before its entry into the contact oxidation tank. In the validation test for the engineering application, the on-site bioaugmentation system showed good performance when the pH was 7; however, it is worth noting that the pH level decreased during the running of the system, which is similar to the findings of Huang *et al.* (2005).

As shown in Figure 4(b), the COD removal in the contact oxidation tank when the gas–water ratio was 56:1 in channel I was 330 $mg \cdot L^{-1}$ higher than when the gas-water ratio was 28:1, indicating that a high gas–water ratio can accelerate the degradation of the pollutants in the reactor, mainly because PEG is usually decomposed through aerobic pathways (Marchal *et al.* 2008) and a high gas–water ratio helps improve microbial activity. The analysis also showed that COD was removed effectively at each step of the system under the two gas–water ratios, and the COD content in the effluent water from the secondary sedimentation tank was stable below 100 $mg \cdot L^{-1}$. Taking into account energy usage, the gas–water ratio was recommended as 28:1, which may reduce the aeration rate by 50% compared with that without bioaugmentation treatment. According to Kawai *et al.* (1978), the oxygen uptake of intact cells increases along with the addition of PEG, as the rate of absorption also increases with polymerization. Therefore, to treat a high volume of wastewater or wastewater with a high concentration of pollutants, it is need to increase the gas–water ratio to improve the activity of the microbes and thus enhance the removal efficiency of the pollutants.

The concentration of reflux sludge mainly depends on the reflux ratio (Jeppsson 1996). As indicated in Figure 4(c), the COD removal in channel I of the contact oxidation tank when the reflux ratio of the system was 100% was 166 $mg \cdot L^{-1}$ higher than when the reflux ratio was at 50%. This result indicated that the increase in the sludge reflux ratio enhanced the sludge concentration in the reactor pond to some extent, thus reducing the effluent COD concentration (Zhang *et al.* 2007). The analysis also showed that COD was removed effectively at each step of the system under the conditions with both different reflux ratios, and the effluent COD level from the secondary sedimentation tank was below 50 mg · L^{-1}. Excessively small reflux ratio may lead to long retention in the secondary sedimentation tank and thus result in floating of the sludge (Henze *et al.* 1993). In addition, taking into account energy usage, the reflux ratio was recommend to be set at 50%.

As shown in Figure 4(d), when the system encountered a high COD shock (1,373–2,340 mg · L^{-1}), the COD removal efficiency in the contact oxidation tank decreased from 98.05 to 67.1%. This result indicates that the substrate concentration suitable for normal growth of this microbial culture was higher than 1,373 mg · L^{-1}, i.e., approximately 573 mg · L^{-1} higher than that used for *A. johnsonii* by Jiang *et al.* (2013). However, despite the decrease in the COD removal efficiency of the system, the actual removal amount increased first and then decreased, reaching a maximum of 2,193 mg · L^{-1}, which was 1,333 mg · L^{-1} higher than at Day 1 and 1,253 mg · L^{-1} higher than that before the bioaugmentation treatment. On Day 4, the influent COD was below 1,000 mg · L^{-1} and the COD removal efficiency of the system was gradually restored. On Day 5, the COD removal efficiency was restored to 89.9% and the effluent COD level was 80 mg · L^{-1}. On Day 8, the removal efficiency reached 93.9% and the effluent COD level was 48 mg · L^{-1}. These results indicated that after the restoration of the system water inlet normally, the effluent COD could meet the discharge standard in one day. Considering the impact of the wastewater push speed, it was estimated that the system could be restored after three days. The volume load of the system was increased to 1.27 kg COD · m^{-3} · d^{-1} by bioaugmentation treatment, which was 127.9% higher than without bioaugmentation. The substrate concentration affected the performance of bacteria, mainly because of the hydrophilic polyoxyethylene and hydroxyl groups in the PEG. The chain length and quantity of monomer epoxy vinyl (EO) in PEG affect the biodegradability of non-ionic surfactants (Qin *et al.* 2002). The quantity of polyoxyethylene is inversely related to the degradability (Li *et al.* 2004). Birch (1984) studied the biodegradability of the linear primary alcohol, oxo alcohol (50 and 70% linearity), and linear secondary alcohol ethoxylates containing 10, 20, 30, and 40 EO. Regardless of the length of the chains, the degradation of the ethers reached 98%. With an increase in EO quantity, the primary degradation of the linear secondary alcohol of the oxo alcohols (AEO) decreases. Therefore, it is speculated that before the COD level reaches 2,340 mg · L^{-1}, the bacterial strain can break down the PEG and detergent into small molecules; as the PEG concentration increases, the EO quantity also increases, which results in toxicity which suppresses the growth of the bacteria and thus decreases their biodegradation capacity (Jiang *et al.* 2013).

Study on the long-term effect of bioaugmentation treatment

The applied study, in 2013, showed that after the augmentation treatment, the system became more stable with a stronger anti-shock capacity. Consequently, the degradation of detergent component induced the reduction of surfactant-induced foaming, and then the volume load of the system increased by 127.9% compared with the value before the augmentation treatment. As shown in Figure 5, the average removal efficiency of the COD in the system was 95.0% and the average removal amount was 780 mg · L^{-1}, which was higher, by 51.5% and 30.2%, respectively, than the pretreatment levels.

Economic and technological analysis

The iron–carbon micro-electrolysis and anaerobic treatment in the process could be canceled after bioaugmentation (Figure 1). The operating expenses of a biochemical system include electricity, reagents, labor, and others (excluding depreciation). The actual operating cost for the application with bioaugmentation treatment was 1.13 yuan (RMB)/ton, which was 0.73 yuan (RMB)/ton lower than without bioaugmentation treatment (see Table 4). In addition, compared with the previous procedure, the total wastewater hydraulic retention time (HRT) at the steps of the iron–carbon micro-electrolysis and anaerobic treatment was reduced by approximately 23.5 h; conversely, 3.6 h, approximately, less than the engineering application of Zhou *et al.* (2009).

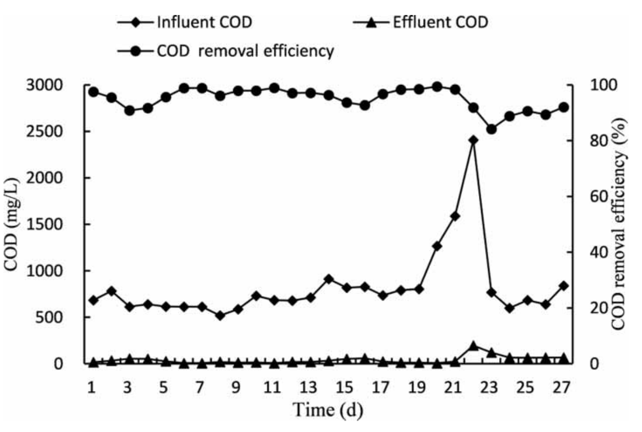

Figure 5 | Long-term COD removal performance of the bioaugmentation system.

Table 4 | Comparison of biochemical system operating costs

Project	Electricity	Reagents	Labor
Without bioaugmentation (yuan/ton)	1.17	0.26	0.43
With bioaugmentation (yuan/ton)	0.66	0.22	0.26
Total cost of water treatment without bioaugmentation (yuan/ton)	1.86		
Total cost of water treatment with bioaugmentation (yuan/ton)	1.13		

CONCLUSIONS

In this study, a microbial culture was applied for the bioaugmentation treatment of wastewater from the manufacturing of PV silicon batteries. Under optimal conditions with a pH value of 7, a gas–water ratio of 28:1, and a reflux ratio of 50%, the average COD removal efficiency reached 95.0% and the average removal quantity reached 780 $mg \cdot L^{-1}$. In addition, the bioaugmentation system could tolerate a COD shock load of 2,340 $mg \cdot L^{-1}$. The effluence met the class I standard of the *Integrated Wastewater Discharge Standard* (GB8978–1996). At last, with bioaugmentation treatment, the HRT was reduced by approximately 23.5 h and the cost reduced by 0.73 yuan (RMB)/ton. In brief, this approach simplified the process, strengthened the anti-shock capability, and reduced the operating costs without the risk of secondary contamination in the physicochemical procedure.

RECOMMENDATIONS

To ensure long-term stable performance and to optimize the properties of this microbial culture, the following recommendations are offered:

- Because of the significant impact of pH on the activity of the microbes, extra attention should be paid to adjust the pH of the water flowing in to the bioaugmentation system and the operating system.
- In the event of sudden increases in the quantity of wastewater or the concentration of pollutants in the water, or sludge floating in the secondary sedimentation tank, it is necessary to consider increasing the reflux rate while monitoring the change in the dissolved oxygen content in the contact oxidation tank.
- In the events of high shock load (COD concentration exceeding 1,373 $mg \cdot L^{-1}$) or sudden increases in wastewater inflow, it is necessary to consider increasing the gas–water ratio, enhancing the reflux rate, and the approaches for gathering and diluting wastewater by making full use of the anaerobic tower and/or iron–carbon reactor pond which has been established previously. Using these approaches, workload of the system can be reduced, the wastewater treatment efficiency can be increased, and stable operation of the system can be ensured.

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