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MUTAGENICITY ASSESSMENT OF PRODUCED WATER DURING PHOTOELECTROCATALYTIC DEGRADATION

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Abstract—Oilfield produced water was treated by photocatalysis, electro-oxidation, and photoelectrocatalysis, respectively. The chemical composition and toxicity of the raw effluent and treated products were assessed by chemical and mutagenicity analysis. The raw effluent exhibited mutagenic activity in both strains of *Salmonella typhimurium*. The lowest concentration of the dichloromethane extract capable of inducing a positive response in strains TA98 and TA100 were as low as 4 and 5 μg/plate, respectively. All three technologies could detoxify direct-acting mutagenic organic pollutants efficiently, although they could not completely eliminate mutagenicity in the water after 60 min of treatment. At equivalent doses, photoelectrocatalysis exhibited the greatest capability to reduce genotoxicity, whereas photocatalysis was the least effective and did not cause appreciable change in mutagenicity. The gas chromatography–mass spectrometry (GC-MS) analysis revealed that *n*-alkanes (259.4 ng/L) and phenolic compounds (2,501.4 ng/L) were the main organic constituents in the oilfield produced water. Thus, the results of both biological and chemical analysis indicate that photoelectocatalysis was the most effective technology for degradation of oilfield wastewater.

Keywords—Toxicity assay Photoelectrocatalysis Chlorination disinfection Ames test Oilfield wastewater

INTRODUCTION

Large amounts of wastewater are generated annually during the exploitation and production of crude oil and natural gas around the world. In most oilfields, the volume of produced water can be more than 10 times the volume of hydrocarbons depending on the age of a specific oilfield [1]. For example, most of China's large oilfields, such as Daqing, Shengli, and Liaohe, are all in the middle or final stage of development and production, and as a result, the water content of the formation producing the oil can be as high as 90% [2]. Most produced water is reused and reinjected into the underground to enhance oil recovery. However, large amounts of produced water are also discharged directly into the environment after only simple treatment. Previous studies indicated that this type of produced water contains chemicals that can cause acute and chronic toxicity in laboratory testing [3–6]. Furthermore, the increasing number of ecological accidents stemming from crude oil residues has been linked to the complex composition of produced water [7,8]. Thus, produced water without adequate treatment has become a source of environmental concern.

The heterogeneous photocatalytic process employing semiconductor catalysts is an advanced oxidation technology for degrading organic pollutants and has received increasing attention over the last three decades [9]. However, only a few reports have been published regarding the photocatalytic decomposition of organic pollutants in wastewater associated with crude oil production [10–12]. One of the reasons for the infrequent application of the heterogeneous photocatalytic process is its inefficiency of treating produced water containing abundant chloride ions. Chloride ions would compete for surface active sites and form weak Cl radicals [13,14], imposing a strong deleterious effect on slurry photocatalytic degradation of organic pollutants because of the coagulation role of electrolytes for TiO_2 slurry solution [15,16].

On the other hand, electrolytic production of active chlorines, such as free chlorines and subsequently hypochlorous acid (HOCl) and hypochlorite (OCl^-), is widely used for disinfection of industrial watstewater [17,18] and drinking water [19,20]. Our previous work on the photoelectrocatalytic degradation of soluble organic pollutants in high-saline produced water indicated that this hybrid photoelectrocatalysis was efficient and cost effective [21]. One main concern is whether active chlorines produced in the photoelectrocatalytic process would lead to the formation of more toxic intermediates, because numerous studies have reported that mutagenic, carcinogenic, or both disinfection byproducts might be produced from the use of chlorination [22–24]. Another question is whether photoelectrocatalysis is effective enough to degrade any toxic intermediates instantly.

Clearly, it is necessary to clarify the mutagenic potentials and mechananisms of photoelectrocatalytic degradation and the effect of degradation on the mutagenic potential of oilfield produced water. In the past decades, most studies have focused mainly on the toxicity of raw oil produced water to the marine environments [25,26], whereas few data are available on photoelectrocatalytic degradation of mixed organic pollutants. The goals of this investigation were to examine the potential mutagenicity of produced water during photoelectrocatalytic degradation and to evaluate the feasibility of the photoelectrocatalytic process as an alternative for treatment of oilfield produced water.

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MATERIALS AND METHODS

Reagents

Dimethyl sulfoxide and Dexon were purchased from Sigma Chemical (Saint Louis, MO, USA). D-Biotin, L-histidine, agar, nutrient broth, and other chemicals of analytical grade were all purchased from Huankai (Guangzhou, China). *Salmonella typhimurium* strains TA98 and TA100 were obtained from the Guangzhou Sanitation Prevention Station, China. All reagents were used without further purification.

Photoelectrocatalysis procedures

Titanium dioxide nanometer particles were prepared by a modified sol-gel technique reported previously [21]. The experimental setup was a custom-made slurry photoelectrochemical reactor described elsewhere [27]. A 350-ml sample of produced water containing a certain amount of nano-TiO_2 photocatalysts was fed into the photoelectrochemical reactor. The reactor was timed to start when the direct current power, illumination, and compressed air supply were turned on. Except where indicated, the experimental conditions were chosen as follows: cell voltage 30.0 V, airflow 0.05 MPa, catalyst 2 g/L, pH 7.0, initial chemical oxygen demand (COD) 645.0 mg/L, and reaction interval 60 min. Procedures of photocatalysis (PC) were the same as those of photoelectrocatalysis (PEC), except that no cell voltage was employed. Also, the electro-oxidation (EO) procedures were the same as those of PEC without illumination. The sample solution was collected and allowed to precipitate. The supernatant was separated and used for COD measurement, GC-MS analysis, and bioassay.

Preparation of sample extracts

Each raw effluent or degradation sample of 250 ml was extracted three times with 10 ml of dichloromethane by a liquid–liquid extraction method. The combined extract was rotary-evaporated at 35°C or less to near dryness. The residue was transferred to a screw-capped vial and was subject to a gentle stream of nitrogen to remove remaining solvent. The dry extract was redissolved in 50 μl of dichloromethane, and hexamethylbenzene was added as the internal standard for GC-MS quantification of individual phenolic compounds and *n*-alkanes. To test for mutagenicity, 1.7 L of raw effluent or degradation samples were extracted, and the dried extracts were redissolved in different volumes of dimethylsulfoxide to create a series of solutions with various concentrations of organic pollutants. The solutions were filtered through a Millipore (0.45 μm) membrane (Sangon, Shanghai, China) to remove bacteria.

GC-MS analyses

All samples were analyzed with a Hewlett-Packard (HP) 6890 gas chromatograph with an HP 5972 mass selective detector (GC-MS; Hewlett-Packard, Little Falls, DE, USA) operated in the electron impact mode (70 eV). An HP-5 capillary column (50 m × 0.32 mm internal diameter, film thickness 0.25 μm) was used for chromatographic separation. The column temperature was programmed from 45 to 200°C at 3°C/min, followed by an increase to 290°C at 5°C/min (held for 30 min). The injector and ion source temperatures were set at 280 and 180°C, respectively. The carrier gas was helium of ultrahigh purity at a constant flow rate of 2.2 ml/min. A 1-μl sample was injected in the splitless mode. Quantification was done with an internal calibration method.

Mutagencity assay

Salmonella mutagenicity tests were performed according to the standard plate incorporation method [28]. The testing strains were the *Salmonella typhimurium* classical strains TA98 and TA100, which are capable of detecting base frameshift–type and base pair substitution–type mutagenicity, respectively. Five to six different concentrations for each sample were assayed in triplicates. A known mutagen, Dexon, was used as the positive control. Solvent and blank controls were also employed in each assay. The revertant colonies on each plate were counted after 48 h of incubation at 37°C in a sealed container, and the number of *his*$^+$ revertants in each sample was recorded as the mean value from three plates. The results were expressed as revertant colonies per plate. The mutagenic index (MI) was also calculated for each dose as the ratio of average numbers of revertants per plate in a sample and in the negative control (solvent). A sample was considered mutagenic when the MI value was equal to or greater than 2 for at least one of the tested doses and if it had a reproducible dose–response curve [29].

Quality control

For each of eight to 10 samples, a procedure blank and at least one replicate were included. To minimize procedural blanks, the entire analytical procedure was conducted carefully: All glassware was washed with water and soap, rinsed with deionized water, and kilned at 420°C for more than 4 h before use to remove organic contamination. The analytical results indicated that the procedure blanks contained no detectable amounts of target substances, and so no blank correction was necessary. The instrument was calibrated daily with calibration standards, and the relative percent difference between the five-point calibration and daily calibration was less than 20%.

RESULTS AND DISCUSSION

Mutagenicity of organic pollutants in raw and degraded water

Mutagenicity testing for two strains of *Salmonella typhimurium* was conducted on four samples (i.e., raw effluent and effluent samples upon photocatalysis at 60 min [PC60], electro-oxidation at 60 min [EO60], and photoelectrocatalysis at 60 min [PEC60]). All four samples showed strong mutagenic activities in both TA98 and TA100, with the raw effluent sample being the most effective in inducing mutations in *Salmonella* Typhimurium (Table 1). The mutagenic activity of raw effluent in TA98 and TA100 was detectable at concentrations as low as 4 (MI = 3.0) and 5 μg/plate (MI = 2.2), respectively. This could suggest that both the frameshift-type and base pair substitution–type chemicals could be present in the raw effluent extracts, and their contributions to the toxicity were almost equivalent. An increase of the raw effluent concentration up to 10 μg/plate amplified the reversion frequency, and the number of revertant colonies was increased up to 7.3- and 3.0-fold for TA98 and TA100, respectively (Table 1). Mutagenic activities were also detected in the other three samples subject to degradation with single or combined photoelectrocatalytic technologies, but at much higher concentrations for both TA98 and TA100 than those detected in raw effluent. The detectable concentrations of PC60, EO60, and PEC60 were 5 (MI = 2.4), 10 (MI = 2.6), and 30 (MI = 2.6) μg/plate,

Table 1. Mutagenicity in bacterial strains TA100 and TA98 exposed to dichloromethane extracts in different degradation processes[a]

Dose		No. of revertants *his*$^+$/plate in *Salmonella typhimurium* (MI)[b]	
μg/plate	ml/plate	TA98	TA100
Raw effluent			
0[c]	0	31 ± 1.2	103 ± 8.7
2	0.41	33 ± 4.5 (1.1)	121 ± 9.7 (1.1)
3	0.61	46 ± 3.6 (1.5)	125 ± 13.2 (1.2)
4	0.82	93 ± 8.4 (3.0)	167 ± 5.6 (1.7)
5	1.03	130 ± 7.9 (4.1)	231 ± 20.4 (2.2)
10	2.05	228 ± 5.2 (7.3)	304 ± 16.4 (3.0)
PC60			
0	0	31 ± 1.2	103 ± 8.7
3	1.13	423 ± 3.2 (1.3)	116 ± 12.3 (1.1)
4	1.51	57 ± 6.3 (1.8)	146 ± 20.1 (1.4)
5	1.89	76 ± 10.2 (2.4)	203 ± 15.2 (1.9)
10	3.78	104 ± 8.9 (3.3)	241 ± 10.4 (2.3)
20	7.56	171 ± 13.4 (5.5)	299 ± 18.3 (2.9)
30	11.3	194 ± 17.5 (6.2)	376 ± 30.1 (3.7)
EO60			
0	0	29 ± 4.7	103 ± 8.7
5	1.81	23 ± 3.5 (0.8)	120 ± 19.1 (1.1)
10	3.62	77 ± 9.5 (2.6)	128 ± 5.3 (1.2)
20	7.24	101 ± 1.0 (3.5)	165 ± 14.0 (1.6)
30	10.86	153 ± 15.8 (5.2)	222 ± 4.9 (2.2)
40	14.48	169 ± 5.7 (5.8)	284 ± 35.1 (2.8)
PEC60			
0	0	29 ± 4.7	103 ± 8.7
2.5	0.58	25 ± 3.2 (0.8)	101 ± 11.5 (0.9)
5	1.16	27 ± 2.1 (0.9)	135 ± 15.8 (1.3)
10	2.32	42 ± 6.2 (1.4)	201 ± 3.6 (1.9)
20	4.64	53 ± 2.1 (1.8)	343 ± 23.5 (3.3)
30	6.96	75 ± 15.1 (2.6)	562 ± 22.8 (5.5)
40	9.28	103 ± 3.5 (3.5)	697 ± 35.1 (6.8)
Control+[d]		606 ± 54.4 (21)	831 ± 82.5 (8.1)

[a] *his* = hisdine prototrophy; MI = mutagenic index; PC60 = photocatalysis at 60 min; EO60 = electro-oxidation at 60 min; PEC60 = photoelectrocatalysis at 60 min.
[b] Mean ± standard deviation.
[c] Negative control.
[d] Positive control (50 μg/plate of Dexon [Sigma Chemical, St. Louis, MO, USA]).

respectively, in TA98 and were 10 (MI = 2.3), 30 (MI = 2.2), and 20 (MI = 3.3) μg/plate, respectively, in TA100 (Table 1).

It is apparent that all three advanced oxidation technologies were able to detoxify direct-acting mutagenic organic pollutants efficiently, but both the detoxification efficiency and degradation mechanism varied with each technology. The detoxification efficiency of the photocatalytic process was the least effective among the three for both the base frameshift and base pair substitution chemicals, whereas the electro-oxidation process was the most effective for the base pair substitution chemicals. The detoxification efficiency of the photoelectrocatalytic process was the best for base frameshift substances, although none of the strains eliminated mutagenicity from the wastewater. Overall, at equivalent doses, PEC60 exhibited the greatest capability to reduce genotoxicity, whereas PC60 showed almost no such ability.

The high COD removal efficiency was consistent with the toxicity decrease in oilfield produced water [21]. In the photoelectrocatalytic process, the COD removal efficiency (47.4%) was consistent with the detoxifying efficiency. However, in both the photocatalytic and electro-oxidation processes, the detoxifying efficiency did not conform to the COD removal efficiency. The COD removal efficiency with photocatalysis (38.0%) was much higher than that with electro-oxidation (13.5%), but the detoxifying efficiency with photocatalysis was much lower than that with electro-oxidation. Apparently, further tests on toxicity with samples treated by different technologies were desirable. To this end, the mutagenicity was also evaluated per liter of wastewater. The lowest dose of raw effluent and the capability of inducing a doubling of revertants for PC60, EO60, and PEC60 were 0.82 (MI = 3.0), 1.89 (MI = 2.4), 3.62 (MI = 2.6), and 6.96 (MI = 2.6) ml/plate, respectively, in TA98 and 1.03 (MI = 2.2), 3.78 (MI = 2.3), 10.86 (MI = 2.2), and 4.64 (MI = 3.3) ml/plate, respectively, in TA100 (Table 1). Once again, the trend of mutagenic efficiency per liter of wastewater was similar to that per unit of organic extracted mass. Therefore, both the combined photoelectrocatalytic and single photocatalytic or electro-oxidation processes were considered effective, despite the difference between the COD removal and detoxifying efficiencies associated with these technologies.

For the purpose of comparison, the mutagenicity of each sample was standardized with the MI10.0 value [30]. The MI10.0 is defined as the mutagenicity intensity at the dose of a sample solution that initially contains 10.0 μg of organic extract or 10.0 ml of wastewater and that is calculated from the dose–response curve of the Ames assay on each sample. Each measurement confirmed a linear relationship between the dose of the sample to a plate and the mutagenicity intensity. For both per microgram extract and per liter wastewater, the

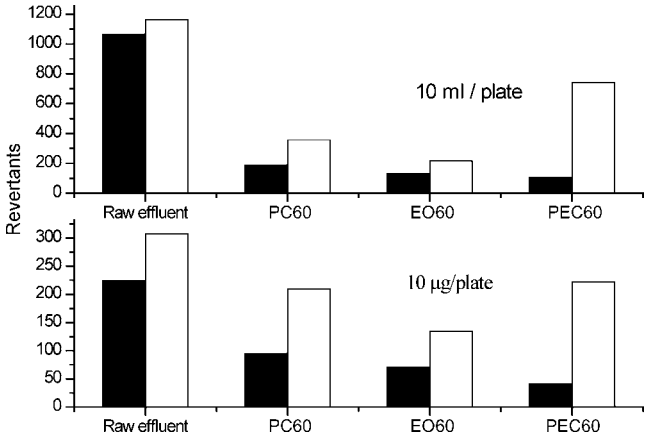

Fig. 1. The mutagenicity intensity at 10.0 μg organic extract or 10.0 ml of raw effluent and wastewater degraded by photocatalysis (PC), electro-oxidation (EO), and photoelectrocatalysis (PEC) processes. ■ TA98; □ TA100.

detoxification order for base pair substitution chemicals was photoelectrocatalysis > electro-oxidation > photocatalysis, whereas the detoxification order for base frameshift substances was electro-oxidation > photoelectrocatalysis > photocatalysis (Fig. 1). Electro-oxidation is thus more efficient than photocatalysis degradation in reducing the bacterial mutagenicity of produced water.

Levels of organic pollutants in raw and degraded water

To further elucidate the relationship between the detoxification efficiency and reaction mechanism, raw effluent and three treated samples were all subject to GC-MS analysis. The efficiency of the different treatment technologies was examined qualitatively by comparing the chromatographic peaks. The raw produced water sample contained a complex mixture of organic pollutants. Upon treatment by the three technologies, the intensities of most peaks decreased significantly. Both the intensity and the number of chromatographic peaks were reduced more substantially with the photoelectrocatalytic treatment (almost all constituents were removed) than with the photocatalysis or electro-oxidation process alone (Table 2 and Fig. 2).

The GC-MS analysis revealed that *n*-alkanes and phenolic compounds were the main organic constituents in the raw produced water. This finding conformed to a previous conclusion that the most prevalent (~90%) groups detected in produced waters were C_{10}–C_{30} straight-chain alkanes [31]. Moreover, the distribution of *n*-alkanes increased slightly from C_{14} to C_{18}, reached the maxima at C_{18}, and gradually decreased with increasing number of carbons to C_{34} in this produced water [32]. The same pattern for the distribution of alkanes was also observed in our raw effluent sample (Fig. 2A). By comparison, *n*-alkanes were highly depleted in the samples treated by the photoelectocatalytic, photocatalytic, and eletro-oxidation technologies (Fig. 2B–D), with the degradation mode and efficiency varying with different treatments. For example, the photocatalytic process preferably degraded the high–molecular weight *n*-alkanes, such as C_{30} and C_{33} (Fig. 2B), whereas the low–molecular weight *n*-alkanes (from C_{14} to C_{21}) were removed more efficiently by electro-oxidation (Fig. 2C). On the other hand, both high– and low–molecular weight *n*-alkanes (from C_{14} to C_{16} and C_{30} to C_{33}) were completely eliminated by the photoelectrocatalytic treatment (Fig. 2D). Quantitatively, the concentrations of individual *n*-alkanes varied in the

Table 2. Concentrations of phenolic compounds identified at different degradation processes[a]

Phenolic compound	Retentation time (min)	Raw effluent (ng/L)	PC60 (ng/L)	EO60 (ng/L)	PEC60 (ng/L)
Phenol	8.49	215.2	178.8	ND	ND
2-Methylphenol	11.7	363.0	359.9	ND	ND
3-Methylphenol	12.82	385.6	451.3	ND	ND
2,4-Methylphenol	13.92	71.2	15.6	ND	ND
2-Ethylphenol	15.57	36.5	55.0	ND	ND
2,5-Dimethylphenol	16.12	375.3	135.3	ND	ND
3,4-Dimethylphenol	17.11	188.4	89.7	ND	ND
3,5-Dimethylphenol	17.41	105.9	76.6	ND	ND
2,4-Dimethylphenol	18.16	112.1	60.9	ND	ND
3-Ethyl-5-methylphenol	18.46	60.4	ND	ND	ND
2,4,5-Trimethylphenol	19.69	44.7	15.0	ND	ND
2-Ethyl-5-methylphenol	20.34	74.8	48.9	ND	ND
2-Ethyl-6-methylphenol	21.21	58.4	46.8	ND	ND
3,4,5-Trimethylphenol	21.42	41.4	ND	ND	ND
2,3,5-Trimethylphenol	21.64	51.8	24.7	ND	ND
2,3,6-Trimethylphenol	22.01	12.1	4.2	ND	ND
2,3,5,6-Tetramethylphenol	22.32	5.8	10.4	ND	ND
2-(1,1-Dimethylethyl)phenol	22.72	18.9	23.3	ND	ND
2-Methyl-5-(1-methylethyl)phenol	22.97	28.5	26.0	ND	ND
4-(2-Propenyl)phenol	23.61	57.8	28.7	ND	ND
4-(1-Methylpropyl)phenol	24.37	14.9	3.4	ND	ND
4-(2-Propenyl)phenol	24.61	29.4	5.6	ND	ND
3,5-Diethylphenol	25.04	26.7	17.9	ND	ND
2,5-Diethylphenol	25.68	52.1	52.3	ND	ND
2-(2-Methyl-2-propenyl)phenol	26.44	14.5	4.5	ND	ND
2-(2-Methyl-2-propenyl)phenol	26.71	29.0	ND	ND	ND
2-Methyl-6-(2-propenyl)phenol	28.21	27.2	12.2	ND	ND
Σ Phenols		2,501.4	1,747.0	0	0
Degradation efficiency			30.2%	100%	100%

[a] PC60 = photocatalysis at 60 min; EO60 = electro-oxidation at 60 min; PEC60 = photoelectrocatalysis at 60 min; ND = not detected.

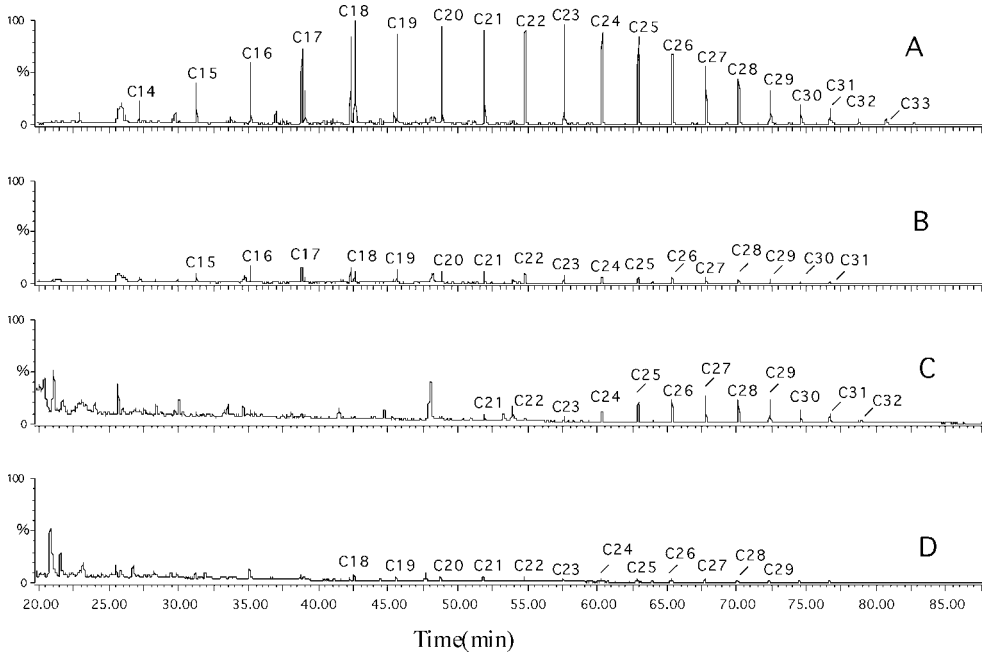

Fig. 2. Mass fragmentograms of *n*-alkanes (mass to charge ratio [*m/z*] = 85). (**A**) Raw effluent; (**B**) photocatalysis 60 min (PC60); (**C**) electro-oxidation 60 min (EO60); (**D**) photoelectrocatalysis 60 min (PEC60).

range of 2.2 to 30.5 ng/L, with a total *n*-alkane concentration of 259.4 ng/L in raw produced water. By comparison, the concentrations of individual *n*-alkanes varied between 0 and 2.2 ng/L, with a total *n*-alkane concentration of 10.4 ng/L in the sample subject to 60 min of photoeletrocatalytic degradation. The total contents of *n*-alkanes were 47.3 and 45.4 ng/L, respectively, in the samples upon photocatalytic and electro-oxidation treatments. Overall, the degradation efficiencies of total *n*-alkanes by photocatalysis, electro-oxidation, and photoelectrocatalysis processes were 81.8, 82.5, and 96.0%, respectively.

The concentration of total phenolics was 2,501.4 ng/L, with concentrations of individual compounds in the range of 5.8 to 385.6 ng/L (Table 2). After 60 min of photocatalytic degradation, the total phenolic concentration decreased to 1,747.0 ng/L, with the concentrations of individual components in the range of 0 to 451.3 ng/L. These results indicate that the concentrations of most phenolic compounds were considerably depleted (~30%) on photocatalytic treatment, with the exception of five single phenolic components. Surprisingly, most phenolic compounds were completely degraded with the electro-oxidation or photoelectrocatalytic treatment (Table 2). This difference in degradation efficiency was attributed to the different reaction mechanisms involved [33]. In the photocatalytic process, the intermediate products are free to diffuse away from the catalyst surface and into the bulk solution before colliding with another •OH radical, resulting in low reaction efficiency. On the other hand, the rate of electron–hole recombination is greatly reduced because of a potential bias applied with the other two treatments, allowing photogenerated holes to effectively oxidize the intermediate species present in the solution. Another explanation is that all phenolic compounds can be electrochemically degraded at the anode because they are present as anions at medium and high pH values.

The results from the toxicity tests and GC-MS analysis allowed us to assess the relationship between the mutagenicity of oilfield produced water and its organic pollutant contents. Most of the *n*-alkanes are expected to have low toxicity according to a previous study [34]. In contrast, the phenolic compounds, which are potential or known human carcinogens, are of considerable health concern even at low concentrations [35,36]. As a result, the mutagenic toxicity of produced water should depend on the number and the abundances of phenolic compounds present. As discussed earlier, the sample degraded by photocatalysis was more toxic than that by electro-oxidation or photoelectrocatalysis (Table 1). Therefore, photoelectrocatalysis was more efficient than photocatalysis for detoxifying oilfield produced water. These results were consistent with those of our previous investigation that the COD concentrations were lower in samples subject to the photoelectrocatalytic treatment and higher in those subject to the photocatalytic treatment [21].

The mutagenicity of produced wastewater during photoelectrocatalysis

The mutagenicity of the organic extracts from produced water subject to photoelectrocatalysis for various time lengths was examined in two strains of *Salmonella typhimurium*. The profiles of the mutagenic toxicity for both TA98 and TA100 decreased rapidly with degradation time, except for a small increase for TA100 at 60 min (Fig. 3). Furthermore, the samples of raw effluent, PEC30, and PEC60 showed significant mutagenic activities in both strains (Tables 1 and 3). In strain TA98, the detectable concentrations were 4 (MI = 3.0), 10 (MI = 2.2), and 30 (MI = 2.6) μg/plate, respectively. The disproportional correlation between the amount of base frameshift substances and the reaction time indicates that base frameshift substances could be efficiently degraded by the photoelectrocatalytic technology. When the reaction times were 120 and 240 min, no mutagenic activity was detected, even at a concentration of up to 200 μg/plate. On the other hand, the detectable concentrations of raw effluent, PEC30, and PEC60 were 5 (MI = 2.2), 20 (MI = 2.2), and 20 (MI = 3.3) μg/plate, respectively, in strain TA100 (Tables 1 and 3). Within

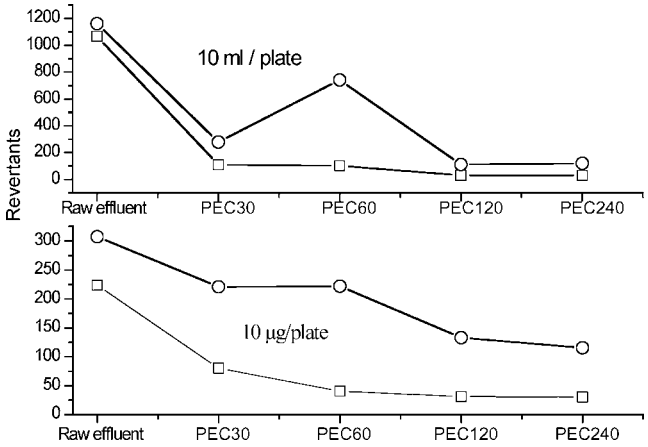

Fig. 3. The mutagenicity intensity at 10.0 μg organic extract or 10.0 ml of raw effluent and wastewater treated with different photoelectrocatalytic (PEC) intervals. □ TA98; ○ TA100.

the first 30 min of degradation, a significant decrease of base pair substitution chemicals was noted. The amount of base pair substitution chemicals was not reduced anymore in the next 30 min. Although the dose at PEC30 and PEC60 was equal to 20 μg/plate, the mutagenic index (MI) of PEC30 and PEC60 were 2.2 and 3.3, respectively. The number of revertants was also positively correlated with toxicity [28]. Therefore, the mutagenicity of those base pair substitution substances increased initially with reaction time. This could indicate that some of the base pair substitution intermediates appeared instantaneously during the first 60 min, and thus contributed to a high level of mutagenicity. As the reaction time reached 120 min, the lowest mutagenic dose approached 100 (MI = 2.6) μg/plate. At 240 min, mutagenicity became extremely low (>100 μg/plate), and the contents of extracted organic pollutants were also considerably low. Some instantly generated base pair substitution intermediates appeared to be produced during the first 60 min of reaction and were gradually transformed with increasing reaction time. Taken together, the photoelectrocatalysis appears to be a promising oxidation technology for decontaminating oilfield produced water.

During the course of photoelectrocatalytic degradation, mutagenicity was also evaluated per liter of wastewater. In strain TA98, the detectable concentrations of lowest mutation dose of raw effluent for PEC30, PEC60, PEC120, and PEC240 were 0.82 (MI = 3.0), 6.54 (MI = 2.2), 6.96 (MI = 2.6), >261.6 (MI = 1.7), and >425.2 (MI = 1.6) ml/plate, respectively. By comparison, the detectable concentrations of lowest mutation dose were 1.03 (MI = 2.2), 13.08 (MI = 2.2), 4.64 (MI = 3.3), 130.8 (MI = 2.6), and >212.6 (MI = 1.7) ml/plate, respectively, in strain TA100 (Tables 1 and 3). These results further confirmed that toxic intermediates were generated during the initial 60 min of degradation, and these toxic intermediates could be detoxified as the degradation progressed. Clearly, photoelectrocatalysis was able to simultaneously lower the amount of organic pollutants and reduce mutagenicity in existing substances per liter.

Furthermore, the mutagenicity of each sample at various degradation timepoints was standardized with the MI10.0 value (Fig. 3). Apparently, the mutagenicities of the oilfield produced water samples subject to the photoelectrocatalytic process for various time lengths showed a similar decreasing trend, with the exception of TA100 as mentioned before. However, the mutagenicity values fluctuated more if the toxicity was calculated as sample volume per plate. These results also confirmed that the COD concentration decreased continually with increasing photoelectrocatalytic reaction time [21].

For comparison, produced water samples treated by photoelectrocatalysis for different time lengths were also subject to GC-MS analysis. Both the number and intensity of chromatographic peaks decreased substantially with increasing re-

Table 3. Mutagenicity in bacterial strains TA100, TA98 exposed to dichloromethane extracts during photoelectrocatalytic degradation[a]

Dose		No. of revertants *his*$^+$/plate in *Salmonella typhimurium* (MI)[b]	
μg/plate	ml/plate	TA98	TA100
PEC30			
0[c]	0	31 ± 1.2	129 ± 20.4
2.5	1.63	32 ± 0.7 (1.0)	147 ± 1.4 (1.1)
5	3.27	46 ± 2.1 (1.5)	179 ± 35.3 (1.4)
10	6.45	70 ± 17.6 (2.2)	196 ± 4.9 (1.5)
20	13.08	104 ± 7.8 (3.3)	279 ± 26.1 (2.2)
30	19.62	247 ± 36.0 (7.9)	472 ± 2.8 (3.7)
40	26.16	280 ± 10.6 (8.9)	560 ± 44.5 (4.4)
PEC120			
0	0	31 ± 1.2	103 ± 8.7
50	65.4	33 ± 6.5 (1.1)	185 ± 21.7 (1.8)
100	130.8	44 ± 4.6 (1.4)	268 ± 17.9 (2.6)
200	261.6	54 ± 8.2 (1.7)	381 ± 23.4 (3.7)
PEC240			
0	0	31 ± 1.2	103 ± 8.7
50	106.3	35 ± 4.5 (1.1)	174 ± 19.2 (1.7)
100	212.6	33 ± 8.4 (1.1)	173 ± 27.8 (1.7)
200	425.2	50 ± 6.2 (1.6)	
Control+[d]		606 ± 54.4 (21)	831 ± 82.5 (8.1)

[a] *his* = hisdine prototrophy; MI = mutagenic index; PEC30 = photoelectrocatalysis at 30 min; PEC120 = photoelectrocatalysis at 120 min; PEC240 = photoelectrocatalysis at 240 min.
[b] Mean ± standard deviation.
[c] Negative control.
[d] Positive control (50 μg/plate of Dexon [Sigma Chemical, St. Louis, MO, USA]).

Table 4. Concentrations of *n*-alkanes identified at different photoelectrocatalytic intervals[a]

n-Alkanes	Retention time (min)	Raw effluent (ng/L)	PEC30 (ng/L)	PEC60 (ng/L)	PEC120 (ng/L)	PEC240 (ng/L)
C14	27.19	5.1	ND	ND	ND	ND
C15	31.24	8.8	ND	ND	ND	ND
C16	35.14	12.5	ND	ND	ND	ND
C17	38.83	13.2	1.8	1.6	0.9	ND
C18	42.32	30.5	2.3	2.2	1.0	0.5
C19	45.66	19.0	1.4	0.8	0.3	0.3
C20	48.84	19.0	ND	ND	ND	ND
C21	51.88	15.6	1.5	0.8	0.7	0.7
C22	54.82	16.4	0.9	1.1	0.7	0.6
C23	57.62	18.4	0.7	0.8	0.7	0.6
C24	60.31	17.4	ND	ND	ND	ND
C25	62.91	17.7	1.2	0.9	0.9	0.9
C26	65.42	14.2	0.4	0.6	0.6	0.6
C27	67.82	13.1	0.5	0.7	0.6	0.5
C28	70.16	11.0	0.3	0.4	0.5	0.2
C29	72.41	10.6	0.6	0.6	0.6	0.4
C30	74.6	6.4	ND	ND	ND	ND
C31	76.72	5.3	3.2	ND	ND	ND
C32	78.77	2.2	ND	ND	ND	ND
C33	80.74	3.1	ND	ND	ND	ND
Σ Alkanes		259.4	14.8	10.4	6.9	5.1
Degradation efficiency			94.3%	96.0%	97.3%	98.1%

[a] PEC30 = photoelectrocatalysis at 30 min; PEC60 = photoelectrocatalysis at 60 min; PEC120 = photoelectrocatalysis at 120 min; PEC240 = photoelectrocatalysis at 240 min; ND = not detected.

action time. A detailed examination unveiled that all phenolic compounds were completely degraded within 30 min, whereas the concentrations of *n*-alkanes were gradually depleted. The profile of *n*-alkanes before and after treatment showed a sharp reduction in the number of detectable *n*-alkanes with increasing degradation time (Table 4). At 30 min, the removal efficiency varied between 38.7 and 100%, and an average of 94.3% of *n*-alkanes was removed by photoelectrocatalysis. With increasing degradation time, the removal efficiency of total *n*-alkanes maintained a moderate rise. The average efficiency for removal of total *n*-alkanes reached 98.1% at 240 min. Apparently, most of the organic pollutants including 100% of phenolic compounds and 94.3% of *n*-alkanes could be removed within 30 min. Also, chlorophenols, usually present in drinking water because of chlorine disinfection [37], were not detected in any of our treatment processes. On the other hand, several phthalate esters, probably originating from the polyvinyl chloride composition in the base of the reactor, were detected during photoelectrocatalysis. Thus, overall toxicity was reduced significantly [38].

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