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Effect of pentachlorophenol and chemical oxygen demand mass concentrations in influent on operational behaviors of upflow anaerobic sludge blanket (UASB) reactor

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Abstract

Upflow anaerobic sludge blanket (UASB) reactor that was seeded with anaerobic sludge acclimated to chlorophenols was used to investigate the feasibility of anaerobic biotreatment of synthetic wastewater containing pentachlorophenol (PCP) with additional sucrose as carbon source. Two sets of UASB reactors were operated at one time. But the seeded sludge for the two reactors was different and Reactor I was seeded with the sludge that was acclimated to PCP completely for half a year, and Reactor II was seeded with the mixed sludge that was acclimated for half a year to PCP, 4-CP, 3-CP or 2-CP, respectively. The degradation of PCP and the operation fee treating the wastewater are affected by the concentration of MEDS (microorganism easily degradable substrate). So the confirmation of the suitable ratio of [COD] and [PCP] was the key factor of treating the wastewater containing PCP economically and efficiently. During the experiment, the synthetic wastewater with 180.0 mg L^{-1} PCP and 1250–10000 mg L^{-1} COD could be treated steadily in the experimental Reactor I. The removal efficiency of PCP was more than 99.5% and the removal efficiency of COD was up to 90%. [PCP] (concentration of PCP) in effluent was less than 0.5 mg L^{-1}. [PCP] in influent could affect proper [COD] (concentration of COD) range in influent that was required for maintenance of steady running of the experimental reactor with a hydraulic retention time (HRT) from 20 to 22 h. [PCP] in influent would directly affect the necessary [COD] in influent when the UASB reactor ran normally and treated the wastewater containing PCP. When [PCP] was 100.4, 151.6 and 180.8 mg L^{-1} in influent, respectively, [COD] in influent had to be controlled about 1250–7500, 2500–5000 and 5000 mg L^{-1} to maintain the UASB reactor steady running normally and contemporarily ensure that [COD] and [PCP] in effluent were less than 300 and 0.5 mg L^{-1}, respectively. With the increase of [PCP] in influent, the range of variation of [COD] in influent endured by the UASB reactor was decreasing. The ratios of [COD] and [PCP] in influent could affect removal efficiency of PCP and COD, the concentration of total volatile fatty acids (VFA) in effluent, biogas quantity and methane content in biogas. [PCP] in influent was linearly or semi-logarithmically correlated to [COD] in effluent when [COD] in influent was 5750 ± 250 mg L^{-1}, and so was the relationship between [COD] in influent and [PCP] in effluent when [PCP] in influent was 100.4 or 151.6 mg L^{-1}, less than the maximum permissible [PCP]. The sources of seeded sludge, the way of sludge acclimation and the characteristics of anaerobic sludge could all affect the UASB reactor capacity treating PCP. When [PCP] were less than 180.8 mg L^{-1} for Reactor I and 151.6 mg L^{-1} for Reactor II, the variation of [PCP] in influent had little effect on the UASB reactor volume gas production rate and substrate gas production rate. And [VFA] and pH value in effluent were affected a little. Volume biogas production rate and substrate biogas production rate of the UASB reactor were only affected by [COD] and loading rate in influent. But when [PCP] was more than 151.6 mg L^{-1} for Reactor II, the biogas production fell quickly and was over 3 days later. [VFA] in effluent from Reactor II increased up to 2198.1 mg L^{-1} quickly and the pH value fell to less than 7. Reactor II could not run normally. The component of VFA accumulated quickly was mainly acetate (above 50%). With [PCP] increased from 7.9 to 180.8 mg L^{-1} gradually in influent, the methane content in biogas from Reactor II decreased from 70% to 60%, but the reactor could still run normally. Then as for Reactor II, the content of methane have fallen from 75% to 45% or so quickly. And Reactor II could not run steadily. So the conclusion could be drown that too high [PCP] in influent for UASB reactor mainly inhibited the activity of methane-producing bacteria cultures utilizing the acetate.

Keywords: Pentachlorophenol; Anaerobic biotreatment; Chemical oxygen demand

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1. Introduction

Pentachlorophenol (PCP) is a biocide widely used worldwide for wood and wood product preservation. And large quantities of PCP are also produced from the bleaching process with chlorine gas in pulp and paper industry. Due to its toxic properties, PCP is an environmentally significant chemical that acts on a variety of organisms as a potent inhibitor of oxidative phosphorylation. In cells, it disrupts the proton gradient across membranes [1]. Along with other chlorophenols (CPs), it has been designated as a priority pollutant by the Environmental Protection Agencies in many countries [2].

Now PCP has been the target of a number of investigations focused on its possible biotreatment [3–13]. Both anaerobic and aerobic biodegradation pathways for PCP and other CPs have been widely reported over the past decades years [14,15]. Mono- and, to a less extent, dichlorophenols can be metabolized by aerobic microorganisms, but aerobic attack becomes less effective to more highly chlorinated compounds. Additional researches have also indicated that biodegradation of highly chlorinated compounds (such as PCP) by aerobic bacteria is hindered. In contrast, reductive dechlorination of chlorophenols has been demonstrated to occur under anaerobic conditions [8,16,17]. The rate of dechlorination under such conditions is actually greater for more heavily chlorinated compounds. Even though PCP has been shown to resist biodegradation, several pathways for the microbial degradation of PCP have been identified. These pathways are PCP methylation, reductive or oxidative dehalogenation and ring cleavage.

For the above reasons and the environmental significance of PCP, the anaerobic degradation of PCP was investigated extensively [18–22]. In anaerobic biodegradation of PCP, chlorines are removed from the aromatic ring via reductive dechlorination [23–25]. The position of chlorine atom on the aromatic ring of PCP and other chlorophenols is an important factor that affects reductive dechlorination. In anaerobic cultures fed higher chlorophenols, PCP and 2,4,6-trichlorophenol (TCP) inhibited the culture, while 3,4,5-TCP was degraded [26]. Acclimation of cultures can mitigate this regiospecificity of dechlorination. Following acclimation to 3-CP, cultures degraded both 3,4-DCP and 3,5-DCP [27]. Similarly, cultures acclimated to 4-CP degraded 2,4-DCP and 3,4-DCP [27]. When cultures were acclimated to 3,4-DCP and 2,4-DCP, PCP was biodegraded to 3-CP [25]. In another case, acclimation to a mixture of 2-PCP, 3-CP and 4-CP resulted in sludge able to mineralize PCP to carbon dioxide (CO_2) and methane (CH_4) [24]. While study on anaerobic treatment has focused on conventional anaerobic systems or on methanogenic systems alone, anaerobic acidogenesis has been studied mainly in connection with the concept of two-phase digestion [28–29]. Two-phase digestion physically separates acidogens from methanogens, allowing for higher loading rate and shorter retention time as well as enhanced effluent quality [28]. In these investigations, the toxicity of PCP was exhibited by the glucose-utilizing in these systems and the distinct inhibition of glucose assimilation at the $10\,mg\,L^{-1}$ PCP and higher was found. Biodegradation of PCP may be expected since an accumulation of excess reducing equivalents in the form of hydrogen that is characteristic of acidogenic systems and could cause reductive dechlorination as electron acceptors. Otherwise, anaerobic/aerobic biodegradation of PCP has also been demonstrated to be an effective treatment process when used as part of an integrated system [19]. Both anaerobic and aerobic biodegradation pathways for PCP and CPs have been reported over the past decades. Chlorinated phenols are anaerobically biodegraded through the reductive chlorination. Researchers have degraded PCP and other chlorinated compounds with both acclimated and unacclimated cultures obtained from the sediments and sewage in batch tests. Therefore, they have also exhibited that biodegradation pathways are influenced by the culture's sources and acclimation processes [19]. Possibility of anaerobic biodegradation of PCP was demonstrated by the observation that an accumulation of less chlorinated phenols occurred with PCP disappearance in the anaerobic sewage sludge. Reductive dechlorination firstly is a significant process for the biotreatment of PCP, because dechlorinated products are usually less poison and more easily biodegraded either anaerobically or aerobically.

With thorough investigations on the metabolism and variation of anaerobic microbes, some of their potentials that haven't been found under aerobic conditions like detoxification and degradation of the poisonous and noxious organic pollution were found [17,30]. The anaerobic microbes can degrade the most halogenated organic compounds by co-metabolism. Now the investigations also showed that although the dechlorinated microbes could acquire energy from the reduction reaction, it was difficult for them to obtain the necessary carbon sources maintaining their growth from the reduction dechlorinated reaction. The carbon sources and nutrient essential for the dechlorinated microbes growth were usually obtained from the MEDS or other microbial co-metabolism of benzene rings. Therefore, highly attention to utilizing anaerobic microbial co-metabolism to treat the aerobic difficultly degradable organic compound was paid by the adding of the microbial easily degradable substrate (MEDS) [30].

The microbial easily degradable substrate (MEDS) in wastewater is usually expressed by chemical oxygen demand (COD). The degradation of PCP and the operation fee treating the wastewater are affected by the concentration of MEDS. If the concentration of MEDS was too low, the microbes in the system could not grow normally and the degradation rate of PCP was affected. If it was too high, COD in the effluent could not reach the standards of wastewater discharge and the operation fee treating the wastewater increased correspondingly and the bio-degradation of PCP was inhibited due to glucose-utilizing effect of microbe. So the confirmation of the suitable ratio of [COD] and [PCP] was the key factor of treating the wastewater containing PCP economically and efficiently. In this paper, the effect of PCP and COD mass concentrations in influent on operational behaviors of upflow anaerobic sludge blanket (UASB) reactor was investigated.

2. Materials and methods

2.1. Source of anaerobic activated sludge

Anaerobic activated sludge was obtained from the wastewater treatment station of Hangzhou citric acid plant in Zhejiang Province. The seeded activated sludge for the reactors was acclimated for half a year to PCP, 4-CP, 3-CP or 2-CP, respectively. During acclimation, chlorophenols were added at the selected time, and at the same time, certain quantity of sucrose was added to promote acclimation and heighten the activity of acclimating sludge.

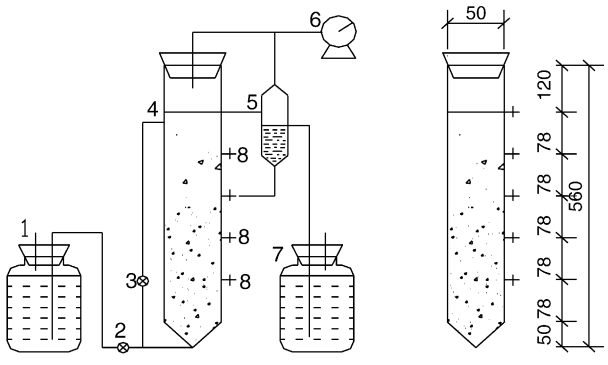
2.2. Instruments, equipments and components of wastewater

2.2.1. Instruments and equipments

Waters high performance liquid chromatography instruments were employed to measure chlorphenols, 721 model spectrophotometer was employed to analyze VFA in effluent, 102-G gas chromatography instruments were employed to analyze the components of biogas. Reactors and process in this experiment are illustrated in the Fig. 1. The experiments were performed in a temperature controlled room at $28 \pm 1\,^{\circ}C$. The total volume of each reactor was 1100 ml and the efficient volume was 866 ml. Reactor I was seeded with the anaerobic activated sludge that was acclimated to PCP for half a year, and Reactor II was seeded with the equal amount of anaerobic activated sludge that was the mixture of equal scale sludge acclimated to PCP, 4-CP, 3-CP and 2-CP for half a year, separately. The amount of seeded sludge was 20,100 mg-VSS.

2.2.2. Components of wastewater

Synthetic wastewater was used in this experiment in order to keep the influent stable. Table 1 showed the detailed components of the synthetic wastewater. [COD] of stock solution was 30,000 mg L^{-1}, but was diluted to a desired concentration before use. Stock solution for PCP was prepared at concentration



unit: mm

1.feeding tank 2.feeding pump 3.recirculating pump 4.UASB reactor 5.Gas-liquid-sludge separator 6.gas meter 7.effluent tank 8.sampling ports

Fig. 1. Schematic diagram and dimension of experimental reactor.

Table 1
Compositions of the synthetic wastewater

Constituent	mg L^{-1}
Yeast cream	300
Ammonium chloride (NH_4Cl)	7300
Potassium dihydrogen ortho-phosphate (KH_2PO_4)	2500
Sodium bicarbonate ($NaHCO_3$)	33000
Dipotassium hydrogen ortho-phosphate ($K_2HPO_4{\cdot}3H_2O$)	1300
sucrose	27500

of 10,000 mg L^{-1} in methanol, but it was added to the synthetic wastewater at the specified concentration.

2.3. Start-up and operation of UASB reactors

Initially, the sequencing batch influent and closed recycling was adopted in this experiment because the anaerobic flocculation sludge acclimated for half a year was in suspended and its sedimentation was very poor. When the removal efficiency of substrate was above 80% and sedimentation of sludge was improved partly, the influent flow was slightly increased and hydraulic load and substrate load (organic compound and PCP load) were increased with time. Then shortening hydraulic retention time (HRT) was implemented to accelerate start-up of reactors and formation of PCP-degradation anaerobic granular sludge. The stage of start-up was about 3 months and during this course the PCP-degradation anaerobic granular sludge was formed gradually. After start-up, HRT of the two reactors was 20–22 h and the highest [PCP] in influent was 181 mg L^{-1} for Reactor I and 151.6 mg L^{-1} for Reactor II. Apparently, this result was above others reported in the past [20,31,32].

2.4. Analytical methods

To determine the residual PCP concentration, effluent was extracted with acetonitrile, centrifuged for 10 min at 1000 rpm, and filtered through 0.45 um filters. Extracts were analyzed using an Agilent 1100 serial HPLC system. The HPLC system contained a vacuum degasser, quaternary pump, autosampler, column compartment, diode array and multiple wavelength detectors (DAD). The column was hypersil reversed-phase ODS-C-18 supplied by Agilent, USA. PCP measurement conditions were mobile phase of acetic acid (2%) 10% and methanol 90% at a flow rate of 1.00 ml min^{-1}, signal wavelength at 220 nm, with 20 nm bind width, reference wavelength at 300 nm, with 50 nm bind width. [PCP] was quantified using an HPLC method. Detection limit for PCP was set at 0.01 mg L^{-1} [33]. The acidity ethylene glycol colorimetry was employed for the measurement of [VFA] in effluent [34], and hydrogen and methane in biogas was quantified with a gas chromatograph (Shimadzu 14B, Japan) equipped with a packed column and flame ionization detector. Column, injector and detector temperature were maintained at 55, 90 and 90 °C, respectively. Nitrogen was employed as carrier at a flow rate of 20 ml min^{-1} [35]. COD was analyzed using the acidity potassium dichromate method [35]. Biogas was quantified with a wet gas flow meter and pH value was

determined with a pH meter (PHS-9V) [35]. In this experiment, the sludge was taken from all the sampling ports and then the sludge from the different heights of the UASB reactor was mixed. The total volume of sample of sludge was 100 ml. And all the mixed sludge was put into the 100 ml measuring cylinder and settling for 30 min. The volume of sedimentation was SV. The relationship of SVI and SV could be expressed by the equation, SVI = SV/MLSS. MLSS was measured by the gravimetric method [35]. Each test was repeated for two or three times during 3–7 days, and the measurement errors for COD and PCP in wastewater should be less than 5%.

3. Results and discussions

3.1. The combined effect of [PCP] and [COD] in influent on removal efficiency of substance in UASB reactor

In this experiment, results indicated that the lowest [COD] (or COD load rate) maintaining reactor normal operation (HRT 20–22 h) was decided directly by [PCP] in influent for UASB reactor treating the toxic organic wastewater containing PCP after start-up of reactor and sludge granulation. For example, when [PCP] was 100.4 mg L^{-1} (PCP load rate was 120.0 mg L^{-1} d^{-1}) in influent, PCP removal efficiency of 99.8% was attained when [COD] was 1250 mg L^{-1} (COD load rate was 1.5 g L^{-1} d^{-1}), and COD removal efficiency was 90.3%. If [COD]and [PCP] in effluent were not more than 300 and 0.5 mg L^{-1}, respectively, and with [PCP] as above, [COD] in influent could be controlled between 7500 and 1250 mg L^{-1}. When [PCP] was increased to 151.6 mg L^{-1} (PCP load rate was 180 mg L^{-1} d^{-1}) in influent, PCP removal efficiency could reach only 98.8% with the COD concentration of 1250 mg L^{-1} in influent and COD removal efficiency could only reach 85.8%. And therefore, [PCP] was up to 1.730 mg L^{-1} in effluent. In the case, if the above effluent demand was satisfied ([COD] ≤ 300 mg L^{-1}, [PCP] ≤ 0.5 mg L^{-1}), the highest [COD] in influent was allowed not more than 5000 mg L^{-1}, and the lowest [COD] in influent should be increased up to 2500 mg L^{-1} (COD load rate was about 3.0 g L^{-1} d^{-1}). When [PCP] in influent was up to 180.8 mg L^{-1} (PCP load rate was 216.7 mg L^{-1} d^{-1}), [COD] in influent should be maintained about 5000 mg L^{-1} to satisfy the above demand in effluent ([COD] ≤ 300 mg L^{-1}, [PCP] ≤ 0.5 mg L^{-1}) (Table 2). [COD] values of 1250, 2500 and 5000 mg L^{-1} were the essential lowest [COD] to maintain the UASB reactor running normally when [PCP] was 100.4, 151.6 and 180.8 mg L^{-1}, respectively, in influent. In this investigation, the results also demonstrated that [PCP] in influent affected not only [COD] which maintained the UASB reactor running normally in influent, and [PCP] and [COD] in effluent, but also the relative relationship between [PCP] in effluent and [COD] in influent. It was demonstrated by regression analysis for the data in Table 2 that when [PCP] in influent was 100.4 mg L^{-1}, [PCP] in effluent was markedly linearly related to [COD] in influent. The relationship can be expressed by the regression equations, $[PCP]_{effluent} = 0.172–1.518 \times 10^{-5}$ $[COD]_{influent}$ ($r = -0.9629^{**}$). While [PCP] in influent was increased to 151.6 mg L^{-1}, [PCP] in effluent was markedly semi-logarithmically related to [COD] in influent, the relationship can be expressed by the regression equations, $\ln[PCP]_{effluent} = 0.561–4.331 \times 10^{-4}$ $[COD]_{influent}$ ($r = -0.9712^{**}$).

Additionally, when $[COD]_{influent}$ was approximately 5750 ± 250 mg L^{-1}, and $[PCP]_{influent}$ was not above the highest permissible concentration for UASB reactor, [COD] in effluent was markedly positive correlation to [PCP] in influent under condition that all $[PCP]_{effluent}$ was less than 0.5 mg L^{-1} (Table 3). But certain differences between $[PCP]_{influent}$ and

Table 2
Effect of [COD] and [PCP] in influent on removal efficiencies of PCP and COD

Influent				Effluent		Removal efficiency	
[COD] (mg L^{-1})	[COD] t^{-1} (mg L^{-1} d^{-1})	[PCP] (mg L^{-1})	[PCP] t^{-1}(mg L^{-1} d^{-1})	[COD] (mg L^{-1})	[PCP] (mg L^{-1})	$\omega(COD_{cr})$ (%)	ω(PCP) (%)
1000	1200	100.4	120	261.9	0.854	73.81	99.15
1250	1500	100.4	120	120.7	0.16	90.34	99.84
		151.6	181	176.8	0.73	85.86	98.86
		180.8	216.7	368.9	14.96	70.53	91.73
2500	3000	100.4	120	156.2	0.144	93.75	99.86
		151.6	181	220.8	0.288	91.17	99.81
		180.8	216.7	671.9	1.6	73.12	99.11
5000	6000	100.4	120	184.2	0.066	96.32	99.93
		151.6	181	291.6	0.196	94.17	99.87
		180.8	216.7	295.6	ND	94.06	100
7500	9000	100.4	120	379.5	0.052	95.26	99.95
		151.6	181	458.4	0.061	94.27	99.96
		180.8	216.7	2889	5.51	63.88	96.7
10000	12000	100.4	120	532.6	0.027	94.67	99.97
		151.6	181	659.4	0.024	93.41	99.98
		180.8	216.7	–[a]	–[a]	–	–

Note: The results were the mean values in 3–7 days.

[a] The reactor performances were deteriorated because of high concentration of PCP.

Table 3
Effect of [PCP] in influent on [PCP] in effluent ([COD] in influent was 5750 ± 250 mg L^{-1})

	Influent, [PCP] (mg L^{-1})												
	7.88	11.59	27.26	33.67	42.89	55.13	65.76	90.08	110.1	124.5	136.6	170.8	180.8
Effluent, [PCP] (mg L^{-1})													
Reactor I	ND	ND	ND	0.22	0.04	0.16	0.25	0.20	/	0.49	0.18	ND	ND
Reactor II	ND	0.37	ND	ND	ND	ND	0.44	ND	0.10	ND	0.36	0.43	1.47[a]

[a] The [PCP] in influent was obtained after the maximum permissible [PCP] in the bioreactor was exceeded for 48 h. ND, not detected. The others were the means of continual experiments for 3–7 days.

$[COD]_{effluent}$ existed in both UASB reactors, the relationship can be expressed by the regression equations (Fig. 2):

$$\text{Reactor I}: [COD]_{effluent} = 106 + 1.15\,[PCP]_{influent},$$

$$r = 0.9739^{**}$$

$$\text{Reactor II}: \text{In}[COD]_{effluent} = 4.717 + 0.0127\,[PCP]_{influent},$$

$$r = 0.9362^{**}$$

in these equations, [PCP] means the pentachlorophenol mass concentration in influent or effluent and the unit is mg L^{-1}; [COD] means the concentration of COD in the influent or effluent and the unit is mg L^{-1}, and ** means marks difference (1% marked level).

According to the above regression equations, [PCP] and [COD] in effluent can be directly predicted by [PCP] and [COD] in influent after anaerobic biotreatment. Therefore, the feasibility of process and whether effluent can satisfy discharge criteria or not could be estimated. And the anaerobic treatment efficiency could be increased by adjusting the scale of [PCP] and [COD].

Comparing Reactors I and II (Fig. 2), when [PCP] in influent was below 100.4 mg L^{-1}, both reactors could run normally and the COD removal efficiency in Reactor I was a bit superior to Reactor II. But when [PCP] in influent was more than 151.6 mg L^{-1}, [COD] in effluent from Reactor II increased rapidly. When [PCP] was up to 180.1 mg L^{-1}, the running of Reactor II defeated. This demonstrated that the inhibition of PCP appeared in Reactor II, but Reactor I could still run steadily. It was assumed that the anaerobic sludge seeded for Reactor I was acclimated to PCP completely, but the anaerobic sludge seeded for Reactor II was the equal scale mixture of four types of sludge that were acclimated to 2-CP, 3-CP, 4-CP and PCP for half an year (the anaerobic sludge that was acclimated to PCP was accounted for about 25%). During the acclimation to PCP for half a year, the microbes existing in the acclimated anaerobic sludge have been adapted to the various kinds of mid products from the PCP anaerobic degradation and could degrade them fast. Then the anaerobic sludge acclimated to 2-CP, 3-CP and 4-CP, respectively could only degrade the corresponding acclimating substrate and their homologues and could not degrade all mid products from the PCP degradation. Other studies have also showed that different substrate for the acclimations of anaerobic sludge could affect the degrading characteristics of acclimated sludge obviously. The above results also demonstrated that, with the increase of [PCP] in influent, the UASB reactor's capacity of adaptation to the change of [COD] in influent became poor and the range of enduring to the change of [COD] in influent became small, especially for Reactor II, and too low or high [COD] all resulted in dropping of the removal efficiency of PCP and COD, and evenly worse, the reactor could become acidified. It was assumed to be the interaction effect between easily degradable substrate and PCP-dechlorination in wastewater. Suitable quantity of microbial easily degradable substrate could promote the PCP-dechlorination and degradable metabolism. With the increase of [PCP] in influent, the desired amount of easily degradable substrate increased correspondingly. On the other side, PCP as a type of strong electron acceptor had stronger inhibition effect on the microbe degrading easily degradable substrate with [PCP] increasing. It resulted in microbial activity degrading COD and PCP worse, especially for methane-producing bacteria. Then it made the UASB reactor capacity of enduring the change of COD load rate in influent dropping.

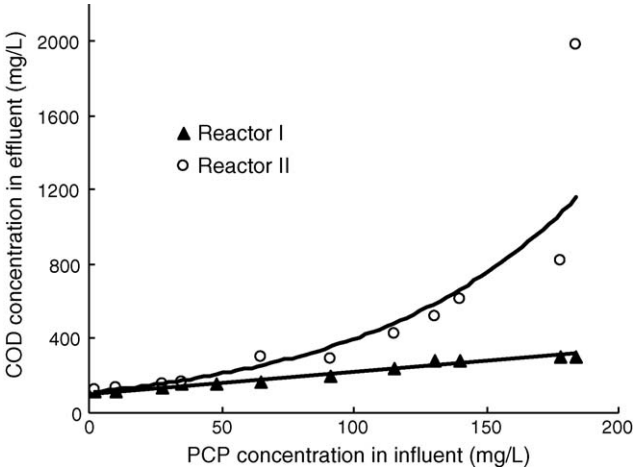


Fig. 2. The relationship between PCP in influent and COD in effluent.

3.2. The effect of [PCP] in influent on [VFA] and pH in effluent

From the experimental results (Table 4), when [COD] in influent was approximately 5750 ± 250 mg L^{-1}, the change of [PCP] in influent within the range of the highest permissible [PCP] had little effect on [VFA] and pH in effluent; but when [PCP] in influent was more than the highest permission, [VFA] in effluent was increased sharply up to 2198.1 mg L^{-1} (for Reactor II) and pH value fell to 7 fast. The result by GC analysis demonstrated that the major component of VFA accumulated

Table 4
Effect of [PCP] in influent on [VFA] and pH in effluent ([COD] in influent was 5750 ± 250 mg L^{-1})

	Influent, [PCP] (mg L^{-1})											
	7.88	11.59	27.26	33.67	42.89	55.13	65.76	90.08	110.1	136.6	170.8	180.8
Effluent												
[VFA] (mg L^{-1})												
Reactor I	107.2	85.6	114.5	85.6	ND[a]	78.4	71.2	ND	107.2	49.6	92.8	71.2
Reactor II	64.0	49.6	78.4	100.0	ND	49.6	114.5	ND	121.7	285.6	316.3	2198.1
pH												
Reactor I	7.86	7.89	7.61	7.57	7.70	7.67	7.61	7.68	7.56	7.33	7.55	7.46
Reactor II	7.61	7.49	7.56	7.54	7.50	7.63	7.61	7.81	7.43	7.41	7.37	7.00

[a] *Notes*: ND, not detected. The others were the means of continual experiments for 3–7 days.

when [VFA] was increased sharply in effluent was acetic acid (above 50%) and the second was propionic acid. From the above results, high [PCP] mostly inhibited the microbial metabolism for these two organic acids. In the anaerobic digester, the acetate metabolism microbial cultures were mainly methane-producing bacteria. That is to say, the microbial activities degrading these two organic acids especially for methane -producing microbial cultures were inhibited.

Comparing Reactors I and II (Table 4), When [PCP] in influent was below 110.1 mg L^{-1}, both reactors could run normally and [VFA] in effluent was less than 107.2 mg L^{-1} for Reactor I and 121.7 mg L^{-1} for Reactor II. And at this time, the pH value was 7.56 and 7.43 for Reactors I and II, respectively. When [PCP] increased gradually, [VFA] in effluent from Reactor I maintained in the lower level steadily, but [VFA] in effluent from Reactor II increased rapidly because of the different seeded acclimation sludge. When [PCP] was increased to 180.8 mg L^{-1}, [VFA] in effluent from Reactor I was 71.2 mg L^{-1} and the pH value was 7.46, Reactor I could still run steadily. But [VFA] in effluent from Reactor II was increased to 2198.1 mg L^{-1} and the pH value fell to 7.00 quickly. This demonstrated that PCP and its mid products from degradation inhibited the activity of the methane-producing bacteria quickly. Reactor II happened to acidity evidently. Under the same operational conditions, the reasons of vast differences of two parallel anaerobic reactors were different seeded sludge. The anaerobic sludge and its biomass that could degrade PCP and its mid products seeded into Reactor II was about 25% of Reactor I. It resulted that the methane-producing bacteria were inhibited and the normal operation of Reactor II was influenced when [PCP] in influent was higher.

3.3. The combined effect of [PCP] and [COD] in influent on the biogas output and the content of methane in UASB reactor

The experimental results illustrated that when [PCP] was not more than the highest permissible concentration, the change of [PCP] in influent had no effect on the biogas producing rate that only varied with the change of COD load rate in influent. when [PCP] in influent of Reactor II was above the highest permission, gas output dropped distinctly. Usually, it could be observed after 2 h, and much less biogas was produced and evenly no biogas was detected 3 days later. Then Reactor II would happen to acidity rapidly and normal running defeated when [PCP] reached up to 151.6 mg L^{-1} (Figs. 3 and 4) .But Reactor I was evidently superior to Reactor II due to the different acclimation sludge seeded. The anaerobic sludge in Reactor I could resist and degrade the higher [PCP] and striking loading.

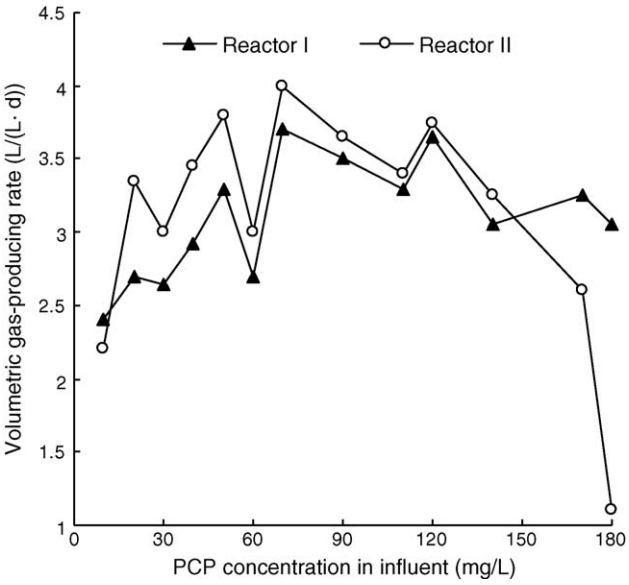


Fig. 3. Effect of PCP in influent on volumetric gas-producing rate.

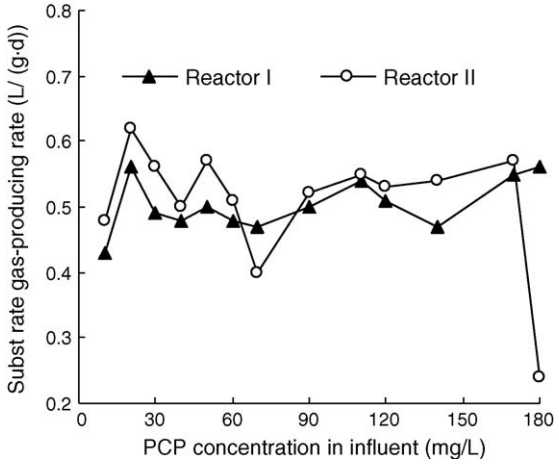


Fig. 4. Effect of PCP in influent on substrate gas-producing rate.

Table 5
Effects of [PCP] and [COD] in influent on CH_4 and H_2 contents in biogas

	Influent ([PCP] = 100.4 mg L^{-1}), [COD] (mg L^{-1})					Influent ([PCP] = 151.6 mg L^{-1}), [COD] (mg L^{-1})					Influent ([PCP] = 180.8 mg L^{-1}), [COD] (mg L^{-1})				
	1250	2500	5000	7500	10000	1250	2500	5000	7500	10000	1250	2500	5000	7500	10000
Components of biogas															
$\Phi(CH_4)$ (%)	74.1–78.0	67.1	57.7–60.8	57.0–57.6	53.4–56.2	72.1–79.9	64.7–66.3	57.9–61.2	–	55.4	70.1–71.4	57.7–63.2	57.8–59.0	34.4–40.9	27.7–35.5
$\Phi(H_2)$ (%)	–	–	–	0.08–0.22	0.04–0.15	–	–	–	–	0.08	–	–	–	0.48–0.72	0.53–0.81

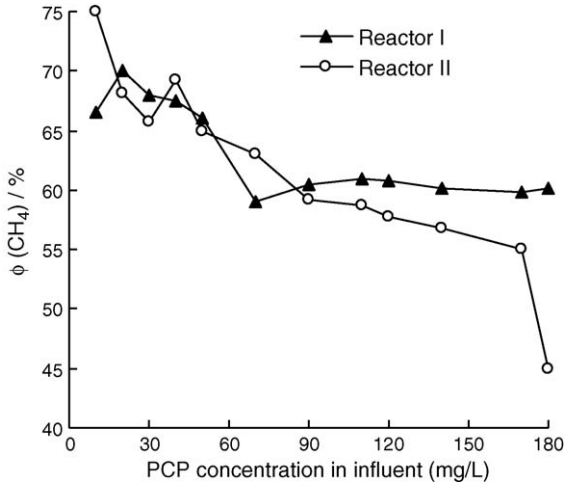


Fig. 5. Effect of PCP in influent on CH_4 content in biogas.

The combined effect of [PCP] and [COD] in influent on the content of methane was showed from two respects, one was that when [COD] in influent was approximately 5750 ± 250 mg L^{-1} and [PCP] in influent was increased from 7.9 to 180.8 mg L^{-1}, the content of methane correspondingly fell 60% from initially about 70%, and Reactor II was more distinctly than Reactor I (Fig. 5). The other was that the increase of [COD] resulted in the lower content of methane under the same [PCP] condition. However, the content of hydrogen was positive correlation to [PCP] and [COD] in influent (Table 5). It illustrated from the above results that PCP as a type of strong electron acceptor, had stronger effect on the methane-producing bacteria than the other two kinds of microorganism cultures (ferment bacteria and hydrogen and acetic acid producing bacteria) during the anaerobic digestion. This was because part of methane-producing bacteria having the carbon dioxide as the electron acceptor and transforming the carbon dioxide into methane needed fairly low redox potential and the ability of PCP acquiring electrons was stronger than carbon dioxide. The changes of [PCP] and [COD] could markedly influence the flow direction of electron and the contribution of reduction capacity of NADH in anaerobic digestion. With the increase of [PCP] in influent, the competition between PCP and the methane-producing bacteria for electrons and reduction capacity was increased correspondingly. As a result, that carbon dioxide was reduced into methane was influenced and the content of methane fell and the content of carbon dioxide increased in biogas. When [COD] was 1250 mg L^{-1}, the reason of higher methane content in biogas was probably that the concentration of microbial easily degradable substrate was too low.

4. Conclusions

(1) [PCP] in influent would directly affect the necessary [COD] in influent when the UASB reactor ran normally and treated the wastewater containing PCP. When [PCP] was 100.4, 151.6 and 180.8 mg L^{-1} in influent, respectively, [COD] in influent had to be controlled about 1250–7500 mg L^{-1}, 2500–5000 mg L^{-1} and 5000 mg L^{-1} to maintain the UASB

reactor running normally and ensure that [COD] and [PCP] were less than 300 and 0.5 mg L^{-1}, respectively. With the increase of [PCP] in influent, the range of variation of [COD] in influent endured by the UASB reactor was decreasing. [COD] values of 1250, 2500 and 5000 mg L^{-1} were the essential lowest [COD] to maintain the UASB reactor running normally when [PCP] were100.4, 151.6 and 180.8 mg L^{-1}, respectively, in influent. Ratio of [COD] and [PCP] in influent could affect removal efficiency of PCP and COD. But too low or too high COD affect the removal efficiency of PCP and even the PCP biodegradation was inhibited.

(2) [PCP] and [COD] in effluent can be directly predicted by [PCP] and [COD] in influent after anaerobic biotreatment. [PCP] in influent was linearly or semi-logarithmically correlated to [COD] in influent when [COD] in influent was 5750 ± 250 mg L^{-1}, the regression equations were as following:

$$\text{Reactor [I]} : [\text{COD}]_{\text{effluent}} = 106.16 + 1.15\,[\text{PCP}]_{\text{influent}},$$

$$r = 0.9736^{**}$$

$$\text{Reactor II} : \text{In}[\text{COD}]_{\text{effluent}} = 4.717 + 0.0127\,[\text{PCP}]_{\text{influent}},$$

$$r = 0.9362^{**}$$

and so was the relationship between [COD] in influent and [PCP] in effluent .when [PCP] in influent was 100.41 or 151.62 mg L^{-1}, The relationship can be expressed by the regression equations, $[\text{PCP}]_{\text{effluent}} = 0.172\text{–}1.518 \times 10^{-5}\,[\text{COD}]_{\text{influent}}$ ($r = -0.9629^{**}$).While [PCP] in influent was increased to 151.6 mg L^{-1}, [PCP] in effluent was markedly semi-logarithmically related to [COD] in influent, the relationship can be expressed by the regression equations, $\ln[\text{PCP}]_{\text{effluent}} = 0.561\text{–}4.331 \times 10^{-4}\,[\text{COD}]_{\text{influent}}$ ($r = -0.9712^{**}$).

(3) The sources of seeded sludge, the way of sludge acclimation and the characteristics of anaerobic sludge could all affect the UASB reactor capacity treating PCP. When HRT was about 20–22 h, Reactor I seeded with the anaerobic sludge acclimated to PCP completely could treat the wastewater containing [PCP] less than 180.8 mg L^{-1}. It was obviously better than Reactor II that was seeded with the equal scale mixed anaerobic sludge acclimated to 2-CP, 3-CP, 4-CP and PCP, respectively (the anaerobic sludge acclimated to PCP was only 25% of total biomass), Reactor II could only treat the wastewater containing PCP with [PCP] less than 151.6 mg L^{-1}.

(4) When [PCP] were less than 180.8 mg L^{-1} for Reactor I and 151.6 mg L^{-1} for Reactor II, the variation of [PCP] in influent had little effect on volume gas production rate and substrate gas production rate of the UASB reactors. And [VFA] and pH value in effluent were affected a little. The volume gas production rate and substrate gas production rate of the UASB reactors were only affected by [COD] and load rate in influent. But when [PCP] was more than 151.6 mg L^{-1} for Reactor II, the biogas production fell quickly and the biogas production was over 3 days later. [VFA] in effluent from Reactor II raised up to 2198.1 mg L^{-1} quickly and the pH value fell to less than 7. And at this time, the UASB reactor could not run normally. The analysis results by GC showed that the component of VFA accumulated quickly was acetate (above 50%). With [PCP] increased from 7.9 to 180.8 mg L^{-1} gradually in influent, the methane content in biogas from Reactor I decreased from 70% to 60%, but the reactor I could still run normally. Then at this time, as for Reactor II, the content of methane have fallen from 75% to 45% or so quickly. And the reactor could not run steadily. So the conclusion could be drown that too high [PCP] in influent for UASB reactor mainly inhibited the activity of methane-producing bacteria cultures utilizing the acetate.

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