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Study on anaerobic treatment of wastewater containing hexavalent chromium*

XU Yan-bin (许燕滨)†, XIAO Hua-hua (肖华花), SUN Shui-yu (孙水裕)
(*Faculty of Environmental Science and Engineering, Guangdong University of Technology, Guangzhou 510090, China*)
†E-mail: hopeybxu@tom.com

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Abstract: A self-made anaerobic bio-filter bed which was inoculated with special sludge showed high efficiency in removing hexavalent chromium. When pump flow was 47 ml/min and COD_{Cr} of wastewater was about 140 mg/L, it took 4 h to decrease the Cr^{6+} concentrations from about 60 mg/L to under 0.5 mg/L, compared with 14 h without carbon source addition. Cr^{6+} concentrations ranged from 64.66 mg/L to 75.53 mg/L, the system efficiency was excellent. When Cr^{6+} concentration reached 95.47 mg/L, the treatment time was prolonged to 7.5 h. Compared with the contrast system, the system with trace metals showed clear superiority in that the Cr^{6+} removal rate increased by 21.26%. Some analyses also showed that hexavalent chromium could probably be bio-reduced to trivalent chromium, and that as a result, the chrome hydroxide sediment was formed on the surface of microorganisms.

Key words: Hexavalent chromium, Wastewater, Anaerobic, Bio-filter bed, Hexavalent chromium bio-removal
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INTRODUCTION

Industrial effluents containing chromium have toxicity as their dissolved Cr^{6+}. The Cr^{6+} salts can easily go into the circulation system through the lung, infiltrate cells, combine with big molecules in vitro after being reduced to Cr^{3+} and eventually form the "ultimate carcinogen" (O′Brien *et al*., 2003; Shakoori *et al*., 2004). If untreated wastewater containing Cr^{6+} is directly discharged to the environment, the environment and ecosystem would be polluted and people's health would be endangered seriously (Trumble and Jensen, 2004).

The treatment of heavy metal wastewater by using microorganism is one of the most active research fields in recent years (Leusch *et al*., 1995; Kaewsarn, 2002; Wu *et al*., 1996). Generally, physicochemical methods, such as ion exchange, active carbon absorption, chemical precipitation and electrochemistry, are widely used in wastewater treatment, but there are still some common problems with the methods mentioned above. When the heavy metals concentrations range from 1 to 100 mg/L, relatively, treatment and materials will cost much. Some treatments show poor selectivity for competitive absorption of alkaline-earth metals. The supernatant liquid overflowing from the chemical reactor may easily rebound to go beyond the effluent standards of Cr^{6+}. Moreover, a lot of sludge due to chemical sedimentation may cause secondary pollution (Jang *et al*., 2001). Compared with the conventional chemical treatment, biological treatment shows some advantages, such as low operation cost, steady effect, easy recovery of some valuable metals (Rehman and Shakoori, 2001; Wang *et al*., 2001). In our experiment, the wastewater containing Cr^{6+} from a hardware factory in Panyu District of Guangzhou City was used in the system domestication, while the synthetic Cr^{6+} wastewater was used in parameter screening. Because of the low redox-potential in anaerobic environment,

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Cr^{6+} can be bio-reduced to Cr^{3+} and the wastewater can be detoxified (De Lima *et al*., 2001; Deng and Xu, 2004). As the organic matter concentrations in the effluent vary with the production scale and procedure, some additional nutrients may be needed in order to meet the microorganisms' energy demand (Sağ *et al*., 2000a; 2000b). Process parameters and Cr^{6+} removal principle are discussed below.

EXPERIMENT MATERIALS

Testing strain

Strains A & B are two highly efficient Cr^{6+} removal bacteria screened in our previous research (Deng and Xu, 2004; Xu *et al*., 2005) and some of their main physical characters are listed in Table 1. The anaerobic sludge was obtained from a brewery in Guangzhou.

Source of wastewater containing Cr^{6+}

Synthetic wastewater prepared with potassium dichromate was used for screening process conditions, contents of nutrients and supplements are listed in 2.3.3.

Actual wastewater from a hardware factory in Panyu district of Guangzhou city was used in system domestication, containing 60 mg/L Cr^{6+} and 202 mg/L COD_{Cr}.

Culture medium

1. Enrichment culture medium (g/L)

KH_2PO_4 0.5, $(NH_4)_2SO_4$ 2.0, NH_4Cl 0.1, Na_2SO_4 0.5, $NaHCO_3$ 0.5, $CaCl_2$ 0.1, $MgSO_4$ 0.1, yeast extract 2.0, $C_3H_5NaO_3$ 3.0; trace metal supplement solution 6 ml, distilled water 1 L.

2. Domestication culture medium (g/L)

NH_4Cl 0.2, KH_2PO_4 0.1, $NaHCO_3$ 0.4, $MgSO_4$ 0.1; trace metal supplement solution 6 ml, actual wastewater 1 L.

3. Experimental culture medium (g/L)

NH_4Cl 0.2, KH_2PO_4 0.1, $MgSO_4$ 0.1, glucose 0.4; trace metal supplement solution 6 ml, tap water 1 L.

TESTER AND METHODS

Tester

The self-made device (Fig.1) consisted of two columns−A and B. Pan activated carbon (Type Z40 granule) and grit (about Φ 5 mm) were chosen as the carrier and filled into column B at a ratio of 2:1 in subsection, while column A just served as a conditioner. The filling-height was about 700 mm. The available volume of column B was 3 L.

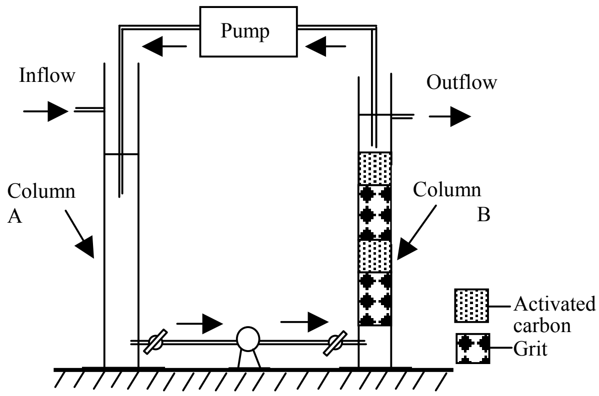

Fig.1 The self-made anaerobic bio-filter bed

Experimental methods

1. Enrichment and domestication

To get enough biomass of strain A & B, after they were enriched by several multiples of the enlarged cultures, anaerobic sludge inoculated by the enriched bacteria was pumped into the column B and stirred continuously until the carriers were covered by biofilm. Then domestication began: wastewater containing Cr^{6+} and domestication culture medium were added into the system, and the concentration of Cr^{6+} in the source increased gradually from 10 mg/L. Meanwhile, the mixed liquor was circulated by constant flow pump to avoid oxygen dissolution. During the domestication process, with the increase of Cr^{6+} concentration, some microorganisms of the biofilm might be killed by the toxicity of Cr^{6+}, and the others

Table 1 The clone configurations of two strains

Strains	Color	Shape	Edge	Diameter	Location
A	Straw yellow	Round	Smooth trim	That of needle	Surface
B	Straw yellow	Shapeless	Lacy	About 0.2 cm	Surface

that had good endurance and deoxidization capability could survive and propagate continuously. The domesticated sludge was composed of many kinds of microorganisms including bacterial strain A & B.

2. Influence of carbon source on removal effect

In general, the actual wastewater contained more or less COD_{Cr} matter, while in the synthetic wastewater, COD_{Cr} mainly came from the carbon source (e.g. glucose). Unless especially mentioned, the Cr^{6+} concentration of the tested wastewater was 60 mg/L in the following research. To determine the optimum addition amount of carbon source, experiments were carried out by comparing the removal efficiencies of two different conditions−with carbon source addition (0.4 g glucose/L wastewater) and without carbon source addition, while the flow of the constant flow pump was controlled to about 58 ml/min and the other nutrient conditions were fixed.

3. Influence of pump flow rate on removal effect

Batch treatment procedure was adopted in the tests mentioned in this paper. Seven liters of wastewater was pumped into the tester in each batch, and by adjusting the pump flow rate, the wastewater circulation cycle could be controlled. In this way, the influence of the circulation time on the anaerobic treatment system could be determined.

The optimal circulation cycle was obtained by comparing the treatment effect due to flow rate of 78 ml/min, 58 ml/min, 47 ml/min, 39 ml/min and 33 ml/min, and the corresponding circulation cycles of 1.5 h, 2.0 h, 2.5 h, 3.0 h and 3.5 h, respectively.

4. Impact of Cr^{6+} load on removal effect

The endurance of the filter bed system to Cr^{6+} was observed by increasing wastewater Cr^{6+} concentrations from 60 mg/L to 90 mg/L by increments of 10 mg/L.

5. Impact of trace metal ions on removal effect

We used some recommended addition doses of trace metal ions to prepare a mixture liquid of several metal ions. Six ml metal ions mixture per liter wastewater was considered as the general addition dose. The effect of trace metal ions could be found by comparing the Cr^{6+} removal efficiency of the system under different volumes of metal ions mixture liquid: zero, half and full.

6. Microscopic observation of biofilm

Zoogloea, protozoa, and bacteria were observed by microscope or Gram-dye.

RESULTS AND DISCUSSION

Establishment of the system

The Cr^{6+} treatment effect of the system is shown in Fig.2 after domestication. In the initial stages of domestication after finishing biofilm-forming in the reaction column, Cr^{6+} was hardly detected after a 1~2 h's operation. And then, the treatment effect became worse gradually in the later 12 d's continuous treatment, and the removal efficiency became relatively constant in the 16th day. It could be thought that the physical adsorption ability of the activated carbon in column B had reached saturation and that the system was indeed entered the domestication period. After four weeks domestication, the system completed domestication and realized the expected capability.

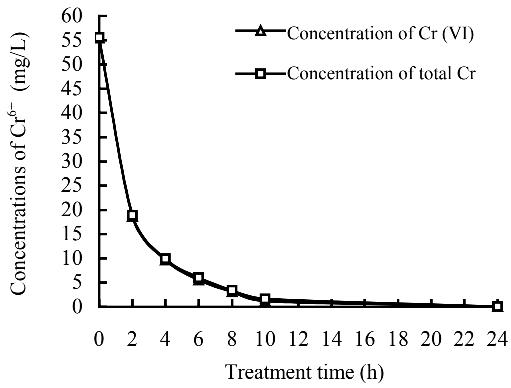

Fig.2 Cr^{6+} removal efficiency of the system after a complete domestication

Influence of carbon source on removal effect

Fig.3 shows that the treatment effect obviously improved after addition of carbon source. At the cycling flow rate of 58 ml/min, the removal rate of the first circulation in the system with carbon source addition was higher than that in the system without carbon source addition by 21.02%. Moreover, to reach the first class effluent standard of Cr^{6+} (0.5 mg/L), compared to a situation of no carbon source addition, carbon source addition could also reduce the treatment time from 14 h to 4 h. But addition quantity of 0.4 g/L glucose made the COD_{Cr} value take longer time to reach the first class effluent standard than Cr^{6+} does, which showed that the total COD_{Cr} after addition of carbon source should be controlled below 140 mg/L, so that all the parameters including COD_{Cr}, total chromium and hexavalent chromium of the outflow can meet the first class effluent standard.

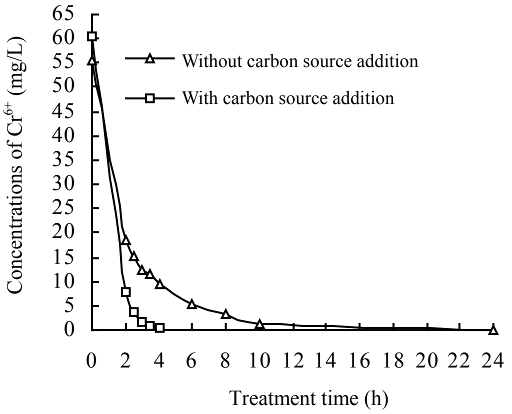

Fig.3 Influence of reaction time on Cr^{6+} residual concentration without carbon source addition

Influence of pump flow rate on removal effect

Fig.4 shows that the treatment time of the wastewater Cr^{6+} is related to the flow rate of constant flow pump. Larger flow rate does not necessarily mean better treatment effect. With the flow rate increasing from 33 ml/min to 39 ml/min, the treatment time shortens when the outflow accords with first class effluent standards, and it reaches a trough at 47 ml/min, then increases again. Forty-seven ml/min was chosen and applied in the following experiments.

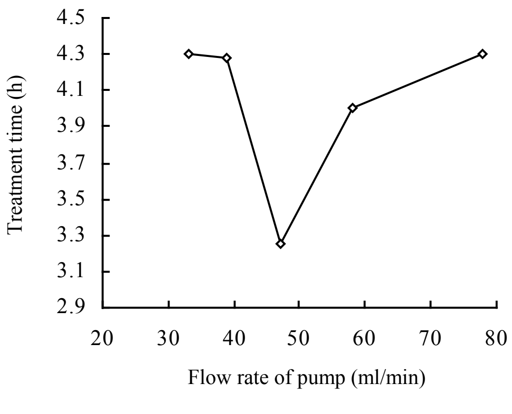

Fig.4 Relationship between pump flow rate and reaction time

Impact of Cr^{6+} load on removal effect

The Cr^{6+} concentration of the inflow was about 60 mg/L at the end of domestication. In order to observe the endurance of the system to Cr^{6+} load impact, inflows containing Cr^{6+} of 70 mg/L, 80 mg/L, 90 mg/L were circulated in the tester at 47 ml/min respectively to get the resulting treatment time, as shown in Fig.5.

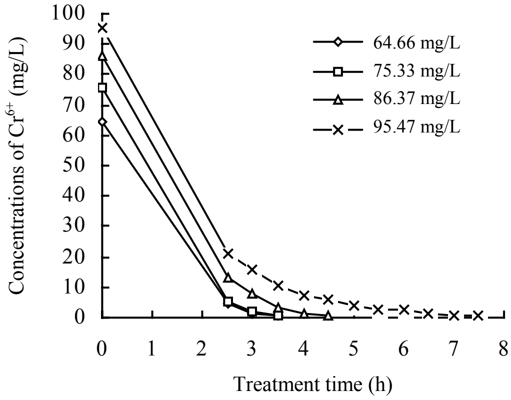

Fig.5 Influence of different Cr^{6+} concentrations of wastewater inflow on Cr^{6+} removal efficiency

When the concentration reached 75.53 mg/L, there was a little decrease in treatment efficiency, that is, this system has strong ability of impact resistance. With increase of Cr^{6+} concentration from 64.66 mg/L to 95.47 mg/L, the treatment efficiency decreased from 92.64% to 71.18%, and the treatment time up to par also increased from 3.5 h to 7.5 h. It was obvious that there is still potential to improve the Cr^{6+} load by further domestication according to actual demand.

Impact of trace metal ions on treatment effect

Fig.6 shows that the removal efficiency of Cr^{6+} is obvious improved after addition of trace metal ions into the anaerobic system.

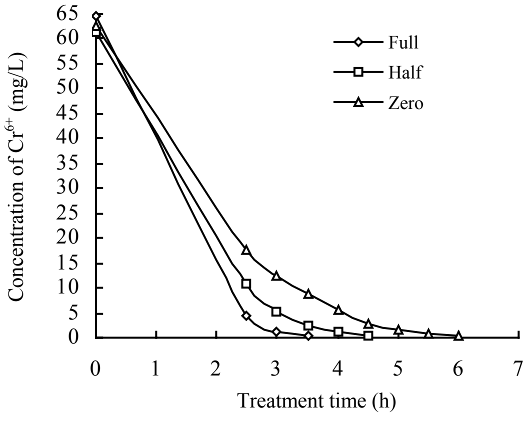

Fig.6 Effect of some trace metals on the Cr^{6+} removal efficiency of the system

At the end of the first circulation cycle, the removal rate of Cr^{6+} was 93.15% and the treatment time was about 3.5 h with 6 ml trace metal supplement solution addition. Having the quantity of trace metal supplement solution added, decreased the Cr^{6+} removal to 82.38% and prolonged treatment time by 1 h. When the control's (without addition of trace metal ion) removal rate reduced to 71.89%, the treatment time was about 6 h.

It is possible to realize the same good effect in actual wastewater treatment by adjusting the supplemental concentration of trace metal ions.

Research on the whither of Cr^{6+}

Microscopy revealed that, under perfect operating conditions obtained through the above experiments, the microfauna on padding grew well. Some ciliata such as *Metopus*, *Colpidium* were observed on surface of the upper buff biofilm, while the inside and lower part were grayish black, and neither protozoa nor micrometazoa were found, but some bacteria such as coccus (G^+), Micrococcus tetragenus (G^+), Bacillus (G^+), Microbactem (G^-) and Spirillum were identified by Gram-staining.

After the system had been running continuously for more than 80 d, the result was steady. Table 2 shows that the concentration ratio of total chromium to hexavalent chromium in the synthetic wastewater prepared with potassium dichromate and other nutrients is 100:99.9. After 2.5-h treatment, the concentration of Cr^{6+} decreased to 0.42 mg/L and the total chromium also decreased to 0.53 mg/L.

Table 2 Change of residual chromium concentration (mg/L)

Parameters	Chromium concentration (mg/L)			
	0 h	2.5 h	3.0 h	3.5 h
Cr (VI)	64.66	4.43	1.27	0.42
TCr	64.71	4.54	1.37	0.53
COD_{Cr}	136.2	108.9	96.8	87.2

Therefore, the removal mode of hexavalent chromium in wastewater may be that Cr^{6+} is reduced to Cr^{3+} by anaerobe and adsorbed on the surface of solids to form sediment. Or trivalent chromium salts formed by biological deoxidization process hydrolyze in the neutral or alkaline liquid of anaerobic system and form undissolvable chromium hydroxide.

CONCLUSION

The anaerobic biofilter system containing strain A & B showed good stability and high removal efficiency for synthetic wastewater treatment. Some main factors that affect the Cr^{6+} removal efficiency of the anaerobic system are carbon source, flow rate of pump, concentration of trace metal ions and Cr^{6+}. Microscopy revealed microfauna on the surface of the upper biofilm and some bacteria, such as coccus (G^+), Micrococcus tetragenus (G^+), Bacillus (G^+), Microbactem (G^-) and Spirillum were identified by Gram-staining on the inter and lower biofilm. From the change of the concentration proportions of total chromium to hexavalent chromium in the inflow and outflow, it was concluded that Cr^{6+} may be reduced to Cr^{3+} by anaerobe and adsorbed on the surface of solids by forming some sediment such as undissolvable chromium hydroxide for hydrolyzation of Cr^{3+} salts in the neutral or alkalinic environment of anaerobic system.

Further research will be carried out on some aspects such as the sorts of nutrients, the life or regeneration cycle of the bio-system.

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