JOURNAL OF PURE AND APPLIED MICROBIOLOGY, April 2013. Vol. 7(Spl. Edn.), p. 145-150

Influence of Temperature on Anaerobic Sequencing Batch Biofilm Reactor Treating Mustard Tuber Wastewater

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(Received: 03 March 2013; accepted: 14 April 2013)

Considering that mustard tuber wastewater has the characteristics of high salinity, high biodegradability, and high concentration of organic matter, a new and efficient anaerobic pretreatment unit, anaerobic sequencing batch biofilm reactor (ASBBR), was chosen to treat most organic compounds in this wastewater. By changing the temperature of the reactor, the experiment was carried out to find out the influence of temperature on effluent COD, gas production rate, and the content of the sludge dehydrogenase. Results showed that: under the condition of 50% biofilm density, draining ratio of 1/4 and 2 d hydraulic retention time, COD removal rate increased from 63.1 to 93.7% when the temperature increased from 10 to 30°C. According to the influence of temperature on effluent COD, gas production rate, and the content of the sludge dehydrogenase, ASBBR should take 30°C in mustard wastewater treatment.

Key words: ASBBR, High salinity wastewater, Mustard tuber wastewater , Temperature.

Fuling District has produced mustard tuber more than one hundred years, and became the Chinese mustard tuber production and marketing areas. In recent years, the annual average mustard wastewater of the district was 3.5 million m3. Among this wastewater, the concentrations of organic matter and suspended solids reached 0.3~20 and 0.5~7 g/L,respectively. Meanwhile, the salinity of this wastewater reached 2~15%. In general case, this wastewater without any treatment was directly discharged into the Three-Gorge Reservoir. Mustard tuber wastewater affects the efficiency of biological treatment, main conclusions are included as following: (1) high salinity inhibits the activity of many enzymes, and eventually leads to plasmolysis (Pendashteh *et al*, 2011; Aminzadeh *et al*, 2010; Rene *et al*, 2008; Zhao *et al*, 2006); (2) high salinity decreases the removal rate of BOD and COD (Peyton *et al*, 2002; Pendashteh *et al*, 2012); (3) high salt in wastewater affects anaerobic ammonium oxidation, nitrification and denitrification (Yang *et al*, 2011; Liu *et al*, 2009; Osaka *et al*, 2008); (4) high salinity can result in the inhibition of the efficiency of aerobic and anaerobic biological treatment[1] (Lefebvre *et al*, 2006; Aloui *et al*, 2009; Ozalp *et al*, 2003).

To treat this high salinity wastewater, China and abroad have investigated many aerobic and anaerobic biological treatment processes. Kubo *et al*. (Kubo *et al*., 2001) treated agro-industrial hypersaline effluent with two salt-tolerant bacteria, *Staphylococcus* sp. and *Bacillus cereus*. In a batch culture, the COD removal rate was about 90% when the pickled plum wastewater contains 15% NaCl. In another experiment, Lu *et al*. (Lu *et al*.,2009) used a hydrolysis acidification/ bio-contact oxidation system to treat oilfield-produced water with high salinity. When the

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biodegradation system was operated with 32 h HRT and 0.28 kg COD /m^3/day volumetric load, the experiment achieved 45, 63.5, 68 and 79.5% mean removal rates of NH_3-N, COD, total petroleum hydrocarbon and TSS, respectively. For anaerobic biological treatment processes, a laboratory down-flow anaerobic fixed bed reactor was used for treating low-strength saline wastewater. The COD removal rate was over 90% when HRT was 96 h. Even if HRT was decreased to 12 h, the COD removal rate could remain over 68% (Rovirosa *et al*, 2004). A lab-scale UASB reactor was used to treat synthetic wastewater; the removal efficiency of TOC could reach 88% when the wastewater was up to 1 g/L NH_4 and 2% salinity (Gomec *et al*, 2005). In this study, considering that mustard tuber wastewater has the characteristics of high salt, high nitrogen, high phosphorus and high organic concentration, an anaerobic biological treatment process is used to treat this wastewater (Chai *et al*, 2010; Wang *et al*, 2008). By changing the temperature in the reactor, the influence of the temperature on treating high salinity mustard tuber wastewater by ASBBR has been studied.

MATERIALS AND METHODS

Experiment setting

A schematic diagram of the ASBBR experimental equipment in this study is shown in Figure 1. The effective volume of the reactor was 2.4 L, with size of L×B×H = 30.0×16.0×50.0 cm. The reactor was made of plastics, which includes semi-soft fiber filler. CH4 was collected by serum bottle liquid replacement system, and the serum bottle volume was 2 L. The liquid used for replace methane was NaOH solution with 1% concentration.

High efficiency halotolerant bacteria were selected from the wastewater produced in the process of pickling the high salinity mustard tuber. Such bacteria would be inoculated into anaerobic sludge. The sludge was adapted to high-salt environment by gradually increasing the salinity and organic load.

During the experimental process, the influent salinity was controlled to be 2, 4, 6, 8, 10 g/L, respectively. The acclimatization process under a specific salinity was considered to be finished if the COD removal rate could reach 80%. Then, the salinity would be raised to the nextlevel until it reached 10 g/L. After the eight-month experiment, the whole process of anaerobic sludge acclimatization was accomplished.

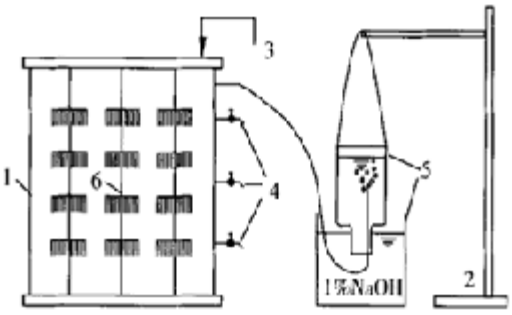
ASBBR was used as an anaerobic biological treatment unit, and its main feature was sequencing batch operation. An operation cycle was divided into four stages: influent period, reaction period, settlement period, and effluent period.The reactor was operated with 2d sequential cycles. In each cycle, the reactor was fed for 0.5 h. After 60 d, the excess sludge was discharged by pump from the bottom regularly.

Experiment water quality

The experimental wastewater used in this study was from Fuling Mustard Tuber Group Co. Ltd, Chongqing, China. During its manufacturing process, the second and third pickle liquid was of high salinity ([Cl-1] = 70~80, 140~160 g/L, respectively); thus, it could be utilized to produce mustard tuber sauce. As for the first pickle liquid and wastewater generated from the whole manufacturing process, they were of a relatively lower salinity ([Cl-1] = 10~30 g/L) and contained more impurity; therefore, they needed uniform collection and treatment before discharge. The wastewater quality is presented in Table 1.

Experimental procedure

Three group reactors were used in the experiment for parallel test. ASBBR reactor used semi-soft fiber packing, and the temperature in every reactor was 10, 20, and 30°C, respectively. Biofilm density, hydraulic retention time (the ratio between reactor volume and influent flow), and draining ratio (the ratio between the water discharged every time and the total volume of water



1 ASBBR Reactor 2 Bracket 3 Influent
4 Sampling and effluent 5 Biogas collection devices
6 Biological packing

Fig. 1. A schematic diagram of ASBBR

in the reactor) during operation were 50%, 2 d, and 1/4, respectively. Anaerobic sludge was chosen as experimental sludge which had been biofilmed and acclimatized stably. The test parameters during the experimental period consisted of the following items: effluent COD, gas production rate and dehydrogenase.

Analyses

Parameters such as effluent COD, gas production rate and dehydrogenase were tested periodically and analyzed according to the standard methods for the Examination of Water and Wastewater which was published by American public Health Association (APHA, 2005).

RESULTS AND DISCUSSION

The results of the test about the temperature on the effluent COD of ASBBR were shown in Figure 2:

The influence of temperature on effluent COD was significant; this could be concluded from Figure 2. When the temperature was 10°C, average effluent COD was 1477 mg/L and COD removal rate was 63.1%, respectively. When the temperature was 20°C, average effluent COD was 780 mg/L and COD removal rate was 80.1%, respectively. If the temperature changed into 30°C, average effluent COD was 250 mg/L and COD removal rate was 93.7%, respectively. Therefore, it was evident that the increase of temperature can improve COD removal rate of mustard tuber wastewater.

Analysis showed that, with the increase of temperature, the bacterial process rate in the reactor increased, so COD removal rate increased (Andersson *et al*, 2001). This was in accordance with the theory in which temperature was one of the important factors of certain enzyme activity. Meanwhile, the optimal temperature of certain enzyme was close to 30°C from the change trend of COD removal rate.

Table 1. The quality of influent

COD (mg/L)	3600~8200
BOD_5 (mg/L)	1400~5600
Total nitrogen (mg/L)	560~1100
Total phosphorus (mg/L)	8~19
$[Cl^{-1}]$ (g/L)	18~23
Total dissolved solids (g/L)	38~47
Total suspended solids (g/L)	2.2~4.6
Volatile suspended solids (g/L)	0.8~1.4
pH	4.5~6.2

Note: According to the preliminary experiment result that the pH value significantly affected the ASBBR treatment efficiency on the high salinity mustard tuber wastewater, the pH value of the raw water was adjusted to 7.0 ± 0.2 before the experiments.

Under different temperature, the change of gas production rate was shown in Figure 3:
Figure 3 showed that, when the temperature increased from 10 to 20°C, the gas production rate of the reactor increased from 0.105 to 0.120m^3methane/m^3/day.When the temperature increased from 20 to 30°C, the gas production rate of the reactor increased from 0.120 to 0.135 m^3methane/m^3/day. This indicated the degradation of more organic matter. Therefore, the increase of temperature could promote the increase of gas production rate.

Analysis showed that, with the increase of temperature, the gas production rate has been improved in a certain degree. This indicated more organic matter has been degraded, so COD removal rate increased (Li *et al*, 2010). Moreover, when the temperature increased from 10 to 30°C, the change trend of the gas production rate was similar to COD removal rate.

Under different temperature, the sludge dehydrogenase content was shown in Figure 4:
Figure 4 showed that, when the temperature increased from 10 to 20°C, the content of the sludge dehydrogenase increased from 4.22 to 5.10 ìg TF/g MLSS/h. When the temperature increased from 20 to 30°C, the content of the sludge dehydrogenase increased from 5.10 to 6.87 ìg TF/g MLSS/h. Hence, the increase of temperature could lead to the increase of sludge dehydrogenase content.
Analysis showed that, with the increase of temperature, the content of the sludge dehydrogenase increased, and the organic load of the unit mass sludge increased, so sludge activity also enhanced. This was in accordance with the theory which DHA concentration could indicate microorganism activity (Park *et al*, 2008). Therefore, the enhancement of sludge activity would lead to the increase of COD removal rate.

In summary, when the temperature increased from 10 to 20°C, COD removal rate increased from 63.1 to 80.1%, the gas production rate of the reactor increased from 0.105 to 0.120

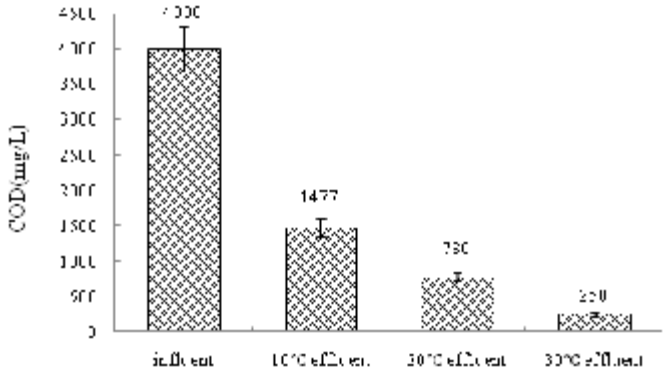


Fig. 2. Influence of temperature on effluent COD

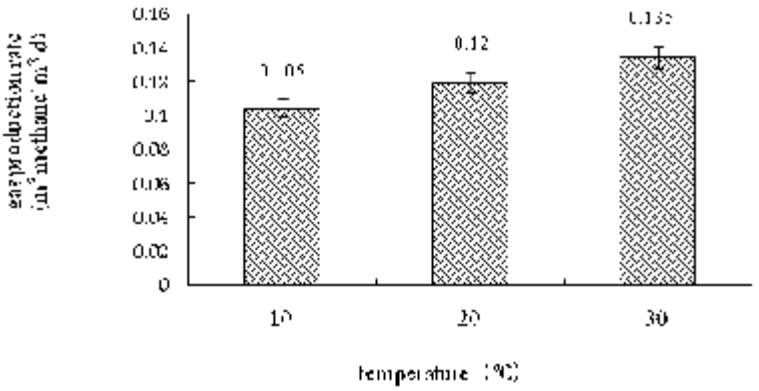


Fig. 3. Influence of temperature on gas production rate

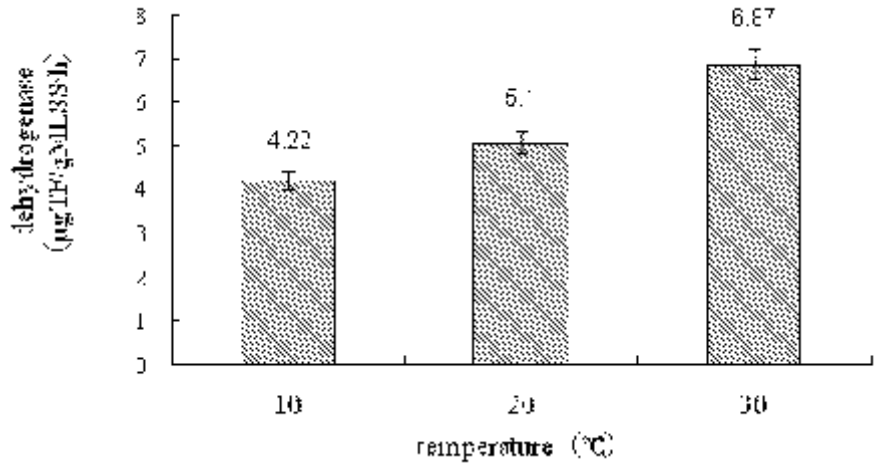


Fig. 4. Influence of temperature on dehydogenase content

m^3methane/m^3/day, and the content of the sludge dehydrogenase increased from 4.22 to 5.10 ìg TF/g MLSS/h, respectively. Meanwhile, when the temperature increased from 20 to 30°C, COD removal rate increased from 80.1 to 93.7%, the gas production rate of the reactor increased from 0.120 to 0.135 m^3methane/m^3/day, and the content of the sludge dehydrogenase increased from 5.10 to 6.87 ìg TF/g MLSS/h, respectively. At the same time, the gas production rate could indicate the degradation of organic matter; the increase of organic matter degradation would lead to the increase of COD removal rate. The dehydrogenase concentration could indicate microorganism activity; the increase of sludge activity would lead to the increase of COD removal rate.

With the increase of temperature, the reactor operation cost would increase. However, when the temperature was 30°C, COD removal rate has relatively high amount. Therefore, the test should adopt the 30°C. At the same time, as to similar salinity wastewater treatment, the optimal temperature should be determined after small experiment and pilot test.

CONCLUSIONS

The ASBBR operating at 50% biofilm density, draining ratio of 1/4 and 2 d hydraulic retention time were used to treat mustard tuber wastewater. When the temperature increased from 10 to 20°C, COD removal rate increased from 63.1 to 80.1%. Meanwhile, when the temperature increased from 20 to 30°C, COD removal rate increased from 80.1 to 93.7%. As a result, 30°C was recommended.

ACKNOWLEDGEMENTS

This study was supported by the National Natural Science Foundation of China (Grant No. 51008318).

REFERENCES

1. Aloui F, Khoufi S, Loukil S, Sayadi S., Performances of an activated sludge process for the treatment of fish processing saline wastewater. *Desalination*., 2009; **246**: 389-396.
2. Aminzadeh B, Torabian A, Azimi AA, Nabi Bidhendi GhR, Mehrdadi N., Salt inhibition effects on simultaneous heterotrophic/ autotrophic denitrification of high nitrate wastewater. *Int. J. Environ. Res*., 2010; **4**: 255-262.
3. Andersson A, Laurent P, Kihn A, Prevost M, Servais P., Impact of temperature on nitrification in biological activated carbon (BAC) filters used for drinking water treatment.*Water. Res.,* 2001; **35**: 2923-2934.
4. APHA., Standard Methods for Water and Wastewater Examination, 21st Ed, American Public Health Association, Washington, 2005.
5. Chai HX, Li XP, Zhou J, Chen Y, Long TR., Treatment of mustard tuber wastewater by anaerobic sequencing batch biofilm reactor-two-stage sequencing batch biofilm reactor-chemical-dephosphorization process. *Chinese Journal of Environmental Engineering*., 2010; **4**: 785-788 (in Chinese).
6. Gomec CY, Gonuldinc S, Eldem N, Ozturk I., Behavior of an up-flow anaerobic sludge bed (UASB) reactor at extreme salinity.*Water. Sci. Technol*., 2005; **51**: 115-120.
7. Kubo M, Hiroe J, Murakami M, Fukami H, Tachiki T., Treatment of hypersaline-containing wastewater with salt-tolerant microorganisms. *J. Biosci. Bioeng*., 2001; **91**: 222-224.
8. Lefebvre O, Moletta R., Treatment of organic pollution in industrial saline wastewater: a literature review. *Water. Res*., 2006; **40**: 3671-3682.
9. Li RP, Chen SL, Li XJ., Biogas production from anaerobic co-digestion of food waste with dairy manure in a two-phase digestion system. *Appl. Biochem. Biotech*., 2010; **160**: 643-654.
10. Liu C, Yamamoto T, Nishiyama T, Fujii T, Furukawa K., Effect of salt concentration in anammox treatment using non woven biomass carrier. *J. Biosci. Bioeng*., 2009; **107**: 519-523.
11. Lu M, Zhang Z, Yu W, Zhu W., Biological treatment of oilfield-produced water: a field pilot study. *Int. Biodeter. Biodegr.,* 2009; **63**: 316-321.
12. Osaka T, Shirotani K, Yoshie S, Tsuneda S., Effects of carbon source on denitriûcation efficiency and microbial community structure in a saline wastewater treatment process. *Water. Res*., 2008; **42**: 3709-3718.
13. Ozalp G, Gomec CY, Ozturk I, Gonuldinc S, Altinbas M., Effect of high salinity on anaerobic treatment of low strength effluents.*Water. Sci. Technol.,* 2003; **48**: 207-212.
14. Park JJ, Byun IG, Park SR, Park TJ., Nitrifying bacterial communities and its activities in aerobic

biofilm reactors under different temperature conditions.*Korean. J. Chem. Eng*., 2008; **25**: 1448-1455.

15. Pendashteh AR, Fakhru'l-Razi A, Madaeni SS, Abdullah LC, Abidin ZZ, Biak DRA., Membrane foulants characterization in a membrane bioreactor (MBR) treating hypersaline oily wastewater. *Chem. Eng. J*., 2011; **168**: 140-150.
16. Pendashteh AR, Abdullah LC, Fakhru'l-Razi A, Madaeni SS, Abidin ZZ, Biak DRA.Evaluation of membrane bioreactor for hypersaline oily wastewater treatment.*Process.Saf.Environ*., 2012; **90**: 45-55.
17. Peyton BM, Wilson T, Yonge DR., Kinetics of phenol biodegradation in high salt solutions.*Water. Res*., 2002; **36**: 4811-4820.
18. Rene ER, Kim SJ, Park HS., Effect of COD/N ratio and salinity on the performance of sequencing batch reactors.*Bioresource. Technol.,* 2008; **99**: 839-846.
19. Rovirosa N, Sanchez E, Cruz M, Veiga MC, Borja R., Coliform concentration reduction and related performance evaluation of a down-flow anaerobic fixed bed reactor treating low-strength saline wastewater. *Bioresource. Technol*., 2004; **94**: 119-127.
20. Wang XM, Zhou J, Long TR, Liu J., Efficiency of phosphorus removal from high-salinity mustard tuber wastewater by combined biological/chemical process. *China Water and Wastewater*., 2008; **24**: 29-33 (in Chinese).
21. Yang JC, Zhang L, Hira D, Fukuzaki Y, Furukawa K. Anammox treatment of high-salinity wastewater at ambient temperature.*Bioresource. Technol*., 2011; **102**: 2367-2372.
22. Zhao X, Wang Y, Ye Z, Borthwick AGL, Ni J., Oil field wastewater treatment in biological aerated filter by immobilized microorganisms. *Process.Biochem*., 2006; **41**: 1475-1483.