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Isolation and Characteristics of a Bacterial Strain for Deodorization of Dimethyl Sulfide

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Abstract

The removal characteristics of dimethyl sulfide (DMS) with a peat packed tower were studied. The peat itself did not remove DMS. The peat inoculated with activated sludge as a source of microorganisms showed an efficient removal of DMS. Dominant microorganisms for degradation of DMS in the peat packed tower were some chemolithotrophic and non-acidophilic sulfur-oxidizing microorganisms originating from sludge. A dominant DMS-oxidizing strain Au7 was isolated and identified as chemolithotrophic Thiobacilli. Product of DMS oxidation by strain Au7 was sulfate. The optimum pH of DMS removal by strain Au7 was 7-5.45.

Keywords: Biological deodorization; Peat packed tower; Activated sludge; Dimethyl sulfide; Chemolithotrophic Thiobacilli

1. Introduction

With the development of national economy and improvement of people's living standard, odor problem becomes more and more important, as well as odor pollution treatment. In every treatment method, biological deodorization has been the research emphasis in recent 10 years, because of its many advantages, such as high deodorization efficiency, simplified device and low treatment cost. From 1980s, many microbial deodorization device and equipment have been used in metallurgy industry, petroleum industry, chemical industry and slaughter and wastewater treatment, and some effects have been achieved [1, 2]. But in the early 1990s, we just began lab research. At present, there are few reports about biological deodorization [3, 4, 5].

Dimethyl sulfide (DMS) is chalcogenide odorous substance with extremely low oder threshold, which is main induced smelly component of many life and production odor source. But there is little research on degradation microorganisms of DMS. This paper researched on DMS biological deodorization by biofiltration. Peat has many advantages, such cheap price, easily obtained, and relative low pressure loss which is ideal deodorization packed, so peat was chose as packed of deodorization device in this research. Use original peat and peat inoculated with activated sludge as packed, research removal effect of DMS by biofilter and variation of microbial community on the packed pre-and post ventilation. On this basis, screen dominant DMS degrading bacteria and research their deodorization characteristics preliminarily.

2. Matertials and Methods

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2.1 Experimental Device

Fig 1 is DMS gas generating device schematic diagram. 50 mL bottle with 25 mL DMS liquid was laid in 500 mL jar, then air stripping generated DMS gas in the jar and the concentration of DMS gas was controlled by changing gas flow. Biological deodorization device schematic diagram showed in Fig2. Diameter and height of aeration column is 35mm and 400mm, respectively; peat packed tower main parameter are as follows: inner diameter 50mm; height 500mm; packed height 40cm; packed volume: 0.8L.

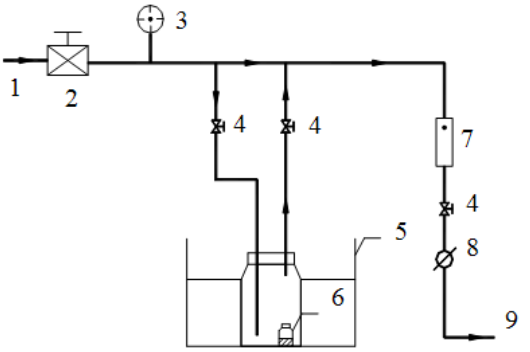

Figure 1 DMS gas generating device

1 air 2control valve 3 pressure gauge 4 interprets valve 5water bath case 6 DMS liquid 7 flow meter 8 sampling place 9 air outlet

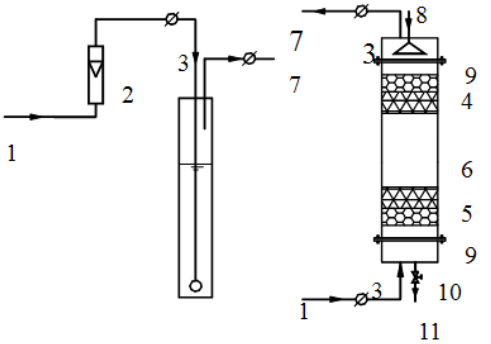

Figure 2 Biological deodorization device schematic diagram

a）aeration column b）peat packed tower 1 air inlet 2 flow meter 3 sampling place 4 rasching rings 5 glass wool 6 peat 7 air outlet 8 spraying liquid inlet 9 flanged valve 10 gate valve 11 deodorized liquid outlet

2.2 Measurement of DMS Concentration

DMS was analyzed by injecting samples into a gas chromatography equipped with FPD detector, in which supporter was chromosorb with 60-80 mesh, β, β-oxygen O, O nitriles was used as stationary phase, and column temperature was 80℃.

2.3 Isolation and Enumeration of Strains

To determine microbe quantity and screen dominant bacteria, microorganism counts were needed to be done. Bacterial count adopted plate counting method which measure viable bacteria number [7,8]. Nutrient medium, chiari medium, Waksman medium and thiosulfate medium were isolation and enumeration of heterotrophic bacteria, fungi, acidphilic sulfide bacteria and non-acidphilic sulfide bacteria, respectively.

2.4 Identification of Strains

After gaining pure strains by isolation, use traditional microorganism classification method to identify strains [7, 8, 9, 10, 11].

2.5 Detection of Concentration of S2O32-、SO42- and S2-

Methods reported as in literature [12].

2.6 Removal of Dimethyl Sulfide by Peat Packed Tower

Ventilate and operate two biological packing towers which used the peat inoculated with activated sludge (30g-MLSS activated sludge/kg-dry peat) and the original peat that not be inoculated with activated sludge as packed, then research DMS removal effect by them. During operation, airspeed was SV36/h; the concentration of DMS gas was from 5mg/m^3 to 40mg/m^3 gradually during intake process; 50mL /2d kept packed moisture at about 60% and operation temperature was 18℃.

2.7 Screening and Characteristic Research on Dimethyl Sulfide Dominant Degradation Strains

1) Screening and Identification of Strains: After the peat packed tower was domesticated by DMS gas, on the basis of former microbiology analysis, screen DMS dominant degradation strains-non-acidphilic chemoautotroph bacteria from $Na_2S_2O_3$ medium plates, and identify them by traditional bacterial classification method.

2) The Growth of Strains in the $S_2O_3^{2-}$ liquid medium: Add strain suspended liquid to culture liquid C containing $Na_2S_2O_3$, and shaking culture at 30℃, then measure growth condition of strains.

3) Removal of DMS by Aeration Column: The experiment that aeration columns remove DMS divided into two groups: one group, only add 250mL aseptic culture liquid C to aeration columns; the other group, add 240mL aseptic culture liquid C and 10mL strain suspended liquid to aeration columns. During intake process, the concentration of DMS was controlled at about 150mg/m^3, and gas flow was controlled at 200ml/min.

3. Resrults and Discussions

3.1 DMS Removal Effect by Original Peat and Peat Inoculated with Activated Sludge

Fig3 showed DMS removal effect by original peat packed tower and peat packed tower inoculated with activated sludge. From Fig3, original peat biological packed tower without being inoculated with activated sludge didn't have the ability in removing DMS, but peat biological packed tower inoculated with activated sludge removed DMS to some extent. Activated sludge was source of abundant microorganisms, which

preliminarily explained that DMS removal was bio-oxidation, not peaty physical action, moreover microorganisms of degrading DMS existed in activated sludge, but there weren't indigenous microorganisms that can degrade and use DMS on the peat.

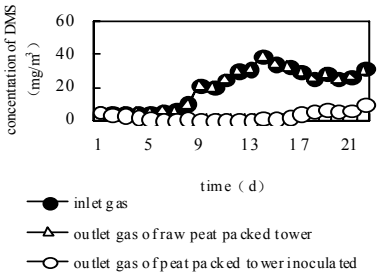

Figure 3 Removal of DMS by peat packed tower

3.2 Effects of removal rate to a change of the pH during removing DMS by peat inoculated with activated sludge

Fig4 showed changes of removal rate and the pH during removing DMS by peat packed tower inoculated with activated sludge. During the operation of peat biological packed tower inoculated with activated sludge, when inlet 6 to 7d with 5mg/m3 DMS gas, there was no DMS gas in outgas. With enhanced DMS load during inlet, pH of the peat decreased gradually, which showed microorganisms oxidized DMS into sulfate resulting in pH decrease. When pH was down to 5.45, removal rate of DMS began to decrease, and in outgas DMS gas was detected, which indicated pH had a great impact on DMS removing, and also microorganisms of degrading DMS had the best degrading ability when pH was no lower than 5.45.

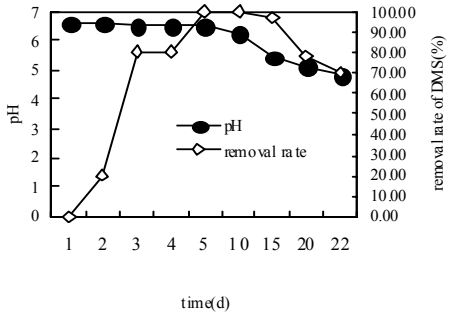

Figure 4 Changes of pH and removal efficiency of DMS by peat packed tower inoculated with activated sluge

3.3 Microbiology Analysis

Table 1 showed microorganism number in the peat inoculated with activated sludge at pre-and post ventilation. Compared with pre-ventilation, after ventilating DMS gas, number of heterotrophic microorganisms in the peat inoculated with activated sludge decreased significantly, almost 100 times;

there was not obvious change in fungi number; the number of acidphilic autotroph bacteria decreased, on the contrary, the number of non- acidphilic autotroph bacteria increased by 100 times.

Table 1 Microorganism Number in the Peat inoculated with Activated Sludge at pre-and post Ventilation(ind./g dry peat)

Gas	nutrient medium	chiari medium	Waksman medium	$Na_2S_2O_3$ medium
	heterotrophic bacteria	*fungi*	*acidphilic sulfide bacteria*	*non-acidphilic sulfide bacteria*
no	1.52×10^{10}	4.83×10^{6}	1.05×10^{6}	1.90×10^{6}
DMS	2.80×10^{8}	2.10×10^{6}	1.01×10^{5}	7.6×10^{8}

In $Na_2S_2O_3$ medium whose pH was 7, $Na_2S_2O_3$ and CO_2 were sole energy and carbon source, so only can grow chemoautotrophy sulfur bacteria belonging to non- acidphilic bacteria. After ventilation, their number increased by 100 times, which indicated that during biofilter removing DMS, non-acidphilic chemoautotrophy sulfur bacteria had a great impact on DMS oxidation. When pH decreased to some extent, it was not suitable for these microorganisms to grow, so removal rate of DMS decreased when pH decreased to 5.45.

Although there was no nutrient being added during operation, the peat itself contained abundant organic compounds. pH decrease had a little influence on the growth of heterotrophic bacteria, but the number of heterotrophic bacteria decreased significantly after ventilation,. So it is considered that DMS had inhibition on the growth of heterotrophic bacteria, resulting in significant decrease of the number of heterotrophic bacteria.

3.4 Screening and Characteristic Research on Dimethyl Sulfide Dominant Degradation Strains

Obtain a well-growth typical strain from screening in the $Na_2S_2O_3$ medium plates, names Au7. Table 2 showed main characteristics of Au7, and Fig 5 was optical and electron microscope photograph of Au7.

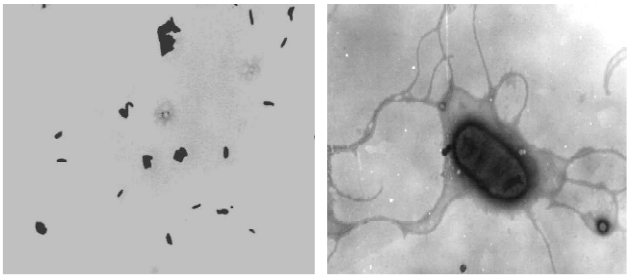

Figure 5 Micrograph of Strain Au7

The cells of Au7 are in the sharp of short rod, gram negative, capable motility, with color of yellow-white and round, and its diameter is 0.5-1mm on the thiosulfate medium plates. In the medium, sole source of energy and carbon source on the growth of strains is $CO_2$and $S_2O_3^{2-}$. In both the medium of 2g/l yeast extract that replaced $Na_2S_2O_3$ and the medium being added 2g/l yeast extract, strains couldn't grow, which indicated this strain is strict chemoautotrophy sulfur oxidation microorganism, which can reduce NO_3 to NO^{2-} but not reduce NO^{2-} to generate N_2 , and its the optimum growth temperature and pH is 7 and 30℃, respectively . According to Berger's Manual of Systematic Bacteriology, it can judge that Au7 is a member of thiobacillus （*Thiobacillus sp.*）in sulphur bacteria.

Table 2 Main Characteristics of Strain Au7

Determination items	Results
shape	Short-bar
size	0.3～0.5×1.5～2.0μm
motility	＋
gram stain	negative
colony characteristics on the $Na_2S_2O_3$ plates	diameter 0.5 ～ 1mm ， with yellow-white sulfur deposition
trophic type	obligate autotrophy
nitrate reduction	－
the optimum growth temperature	30℃
the optimum growth pH	7

3.5 The Growth of Strain Au7 in S2O32- Liquid Medium

Fig 6 showed that the growth effect of Au7 in $S_2O_3^{2-}$ liquid medium. As Fig6 showed, with the decrease of the concentration of $S_2O_3^{2-}$, both cell number and the concentration of SO42 increased gradually; it can be seen the accumulation of sulfur by naked eye in the culture medium; because of the accumulation of SO42-, pH decreased gradually in the liquid. Those data indicated, Au7 oxidized thiosulfate to sulfate, and its intermediate is sulfur.

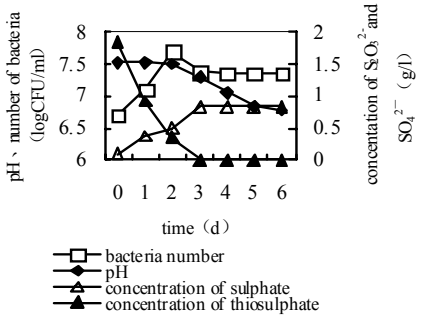

Figure 6 Growth and thiosulphate oxidation by strain Au7

3.6 Removal of DMS by Aeration Columns

After ventilating DMS gas in the aseptic culture liquid aeration columns, at the first 5h, the concentration of DMS in outlet was the same as that of inlet; within 24h, the concentration of SO_4^{2-} didn't change in the aseptic culture liquid aeration columns, either, which showed DMS can't be removed by aseptic culture liquid. Fig 7 showed the removal effect of DMS by aeration columns inoculated by Au7. During intake process, the concentration of DMS was 150 mg/m3; after 2h, the concentration of DMS was 64.25 mg/m3 in outgas; but after 24h, when OD(A660)=0.032, DMS wasn't be detected in outgas and bacterial liquid was still even in the aeration columns, with no bacterial mass appearing. When OD (A660) was up to 1, no longer increase, strains stopped growing, maybe because of lack of nitrogen source. DMS gas was generated by air stripping. During operation period without any sterilization measurement,

external microorganisms could enter aeration columns inevitably, so in the aeration columns except Au7, there were some other heterotrophic bacteria that didn't predominate in quantitatively, compared with DMS dominant degrading autotrophic strain—Au7. After 10 days, the amount ratio of autotrophic bacteria and heterotrophic bacteria was 10:1, and it has been this ratio all long.

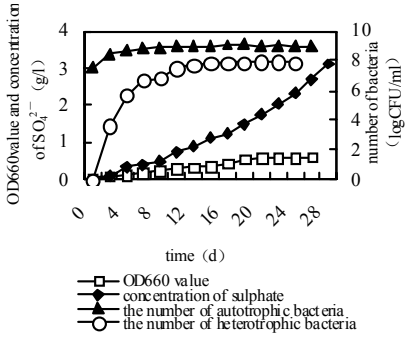

Figure 7 Removal of DMS by peat packed tower

4. Conclusions

Biofilter that used original peat as packed almost didn't remove DMS, but biofilter that used peat inoculated with activated sludge as packed removed DMS effectively, which showed that in the activated sludge there exist microorganisms that can degrade organic sulfide.When pH was no lower than 5.45, peat biofilter inoculated with activated sludge had the best DMS removal effect.

In the biofilter, dominant microorganisms that removed DMS were non-acidphilic chemoautotroph sulfur bacteria; heterotrophic microorganism lost their dominant position in the original peat; fungi number, which changed little relatively, were second-dominant strains degrading DMS.

Obtain one DMS dominant degradation strain-Au7 from screening. From identification, it belonged to thiobacillus. End products were SO_4^{2-} by strain Au7 oxidizing DMS. The optimum pH of degrading DMS was 7.

5. Acknowledgment

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