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Experimental Investigation of Gas Degradation by Methanotrophs at Different Air Flow-rates

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Abstract

A technical approach based on microbial technology is proposed to help resolve the problems caused by explosive gases in coal mines. The proposed technique uses methanotrophic bacteria to oxidize methane. In laboratory experiments, the oxidation effect of hanging nets impregnated with liquid containing methanotrophic bacteria was investigated at different air flow-rates. The experimental results showed that the volume of gas degraded and the gradient of degradation both increased as the gas concentration increased at constant air flow-rates. At fixed gas concentrations, the volume of degraded gas increased with increasing flow-rates of air at low flow-rates. However, the volume of degraded gas slightly decreased with increasing flow-rates of air at high flow-rates. These experimental results provide a theoretical basis for the treatment of explosive gases during exploration for natural gas and to treat potentially dangerous concentrations of gas in gobs, caves and upper corners of mineshafts. They will also be of great practical significance in coal mining.

Keywords: Coal Mines; Explosive Gases; Gas Treatment; Methanotrophs; Microbes; Oxidation

1. Introduction

Gas disasters in mines can cause serious loss of life and property and have significant social impacts [1, 2]. Extensive investigations have been conducted in China on the treatment of gas in coal mines. The national technological projects in the 7th to the 11th Five Year Plans proposed gas treatment technologies in the coal mining industry, including improvements in ventilation, gas drainage and equipment for the treatment of gases. As a result of these technologies, the numbers of accidents and casualties caused by mine gases were greatly reduced. However, the situation remains challenging. Based on data from national gas treatment work in coal mines in 2013, the numbers of accidents and deaths were 59 and 348, respectively. In 2013, six major accidents with a total death toll of 101 occurred in coal mines. Of these six major accidents, three, with a total death toll of 63, were caused by gas explosions. It can be concluded from these data that although the total number of accidents caused by gas disasters has decreased rapidly, major gas disasters have not yet been effectively controlled. The prevention of gas explosions in coal mines therefore remains a priority.

The main methods of preventing gas explosions in coal mines involve preventing the concentrations of gases reaching specified values in the upper corners of mine structures, in caving zones and in gobs. Many technologies, such as ventilation and gas drainage, have been used

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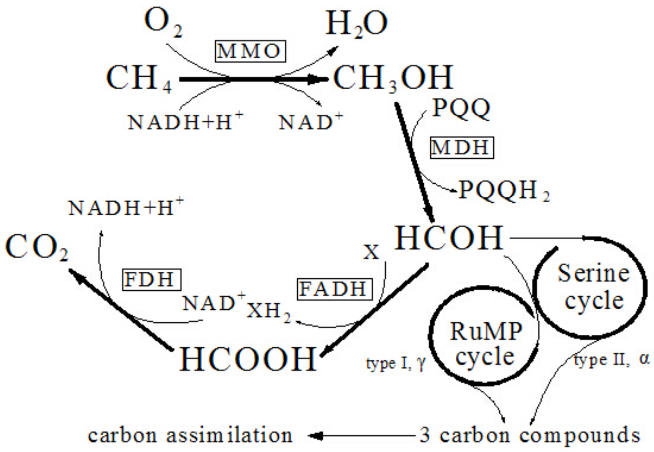

Figure 1: Oxidation pathway of methane by methanotrophs

to prevent gas concentrations reaching these critical values. Microbial technology is now at an advanced stage of development and some researchers have tried to degrade mine gases using microbes. Methanotrophs are widely distributed in the natural world and are found in extreme environments (e.g. acidic, alkaline, high-salt, high-temperature, low-temperature and infertile environments). Methanotrophs metabolize methane, and some cultured methanotrophs have been used to degrade methane into H_2O and CO_2 [3]. During the degradation process, energy for the growth of the methanotrophs is provided by the oxidation of methane. In the degradation process (Fig. 1), methane is first activated to methanol by methane monooxygenase (MMO), and the methanol is then oxidized to formaldehyde [4]. Finally, formaldehyde is assimilated by the microbes or oxidized to CO_2 by methanoic acid.

In the context of the above characteristics of methanotrophs, theoretical and practical investigation of microbial gas treatments has great potential in terms of reducing the number and severity of gas accidents in coal mines.

2. Application of methanotrophs in coal mines

The main research on methanotrophs has been concentrated in environmental fields, but some success has also been achieved in the field of coal mine research [5]. The application of microbes to the degradation of gas in coal mines began in the 1970s. Microbiologists and mining experts from the Russian Academy of Sciences and the Moscow Institute of Mining first proposed technologies to effectively control methane concentrations in mines by using microbes. Based on the oxidation of methane to CO_2 by methanotrophs, a filter layer of bacteria capable of degrading methane was formed by spraying live bacteria into mining gobs or injecting live bacteria into layers of coal via drilling [6]. Experimental data from coal mines in Donetsk and Kuznetsk showed that the methane content decreased by 30–60%.

At the 4th Conference on International Mining Ventilation, the Canadian researchers Chuck et al. presented a new technique for eliminating gas based on field investigations and laboratory tests. Bacteria that could degrade methane were separated from water samples collected from coal mines in western Canada. These bacteria were then studied in the laboratory in simulations with open, closed and continuous ventilation pipes. The best results showed that the methane content of air without bacteria reached up to 5%, whereas the air samples containing bacteria had a methane content of only 0.05%.

Other successes have also been obtained by Australian researchers. Bacteria that could degrade methane were sprayed onto the surface of coal. These bacteria used methane as their sole source of energy for growth and multiplication. Twenty days after spraying with these bacteria, the methane content in air samples had decreased by 66%. Some researchers have reported that the methane content in air samples decreased by 99% with the use of certain bacteria; in some instances, the methane content decreased by 97% after 7 days [7]. Beijerinckiasp. bacteria, which were found in acidic marshlands by US researchers, have been reported to be highly effective in degrading methane in addition to fixing atmospheric nitrogen; in one report, 90% of the methane in air samples was degraded by these bacteria.

The application of microbes to the treatment of gas has recently been proposed in China, although there has been little reported work in the field of coal mine safety. Since the beginning of the 7th Five Year Plan, a group of workers led by Shuben Li from the Lanzhou Chemistry and Physics Research Institute have conducted investigations on methanotrophs, and a number of interesting bacteria have been identified. Further work has been carried out to separate and purify Methylomonassp. GYJ-3. Investigations have been conducted on the catalytic reaction mechanism based on the separated MMO, which is composed of three groups of active components [8, 9].

Mixed bacteria composed of M3011 and GYJ3 strains have been separated and cultured by Wang and Chen and used in gas degradation tests in the Ping Ding Shan mine [10]. Twelve coal samples containing gas from different mining areas and depths were used. The test results showed that the methane contents of the samples into which the methanotrophs had been added were lower than those of the reference sample and that there was an obvious decrease in the methane concentrations in the four samples. The maximum gas concentration feasible for the desorption of coal samples decreased from 10% to 4.8%, and the maximum and average rates of degradation were 52% and 44%, respectively. These results show that an obvious biotransformation has been exerted by the methanotrophs on the methane desorbed from coal samples.

Min and Zhe from Zhe Jiang University carried out extensive investigations on the identification and separation of methanotrophs, and have reported a number of techniques for their culture and purification. New techniques for gas treatment based on bioreactors have been developed by Yu; it was found that bioreactors filled with bioceramics from the hanging membrane containing the methanotrophic bacteria improved methane degradation and were characterized by high shock resistance. Bioceramics from these hanging membranes are therefore suitable fillings for the treatment of gases in coal mines [11]. An investigation of the selection of fillings has provided basic data on the treatment of gas in bioreactors and their application in coal mines.

A strain of QJ16 bacteria that uses methane as the sole source of carbon has been obtained by Wang et al. from the National Key Laboratory and the Chemical Engineering Laboratory of Hua Dong University of Technology. Based on DNA techniques, the species and optimum growth environment were determined; it was found that the oxidative ability of methanotrophs was influenced by the Cu2+ content. This result suggests further investigations on the effective use of methanotrophs.

Investigations on the selection, separation, purification, identification and gene analysis of methanotrophs and suitable growth environments for large-scale culture have been conducted by Mao and Tan. Purified M02-019 bacteria were obtained, and tests on their degradation ability verified. The feasibility of gas treatment by microbes was also verified by contrast experiments in which water and the culture medium were injected. At the two sites in the coal mine where methanotrophs were injected, the number and intensity of jet orifices decreased rapidly. The gas concentrations in the return air at these two sites were 22.5% and 77.2%, the gas concentrations in the coals decreased by 39.7% and 13.5%, the gas pressures decreased by 76% and 18% and the average decreases in the desorption index (K1) were 62.8% and 26.9%.

A high-pressure mixing apparatus designed and manufactured by Zhang from Henan University of Engineering was applied in a study of gas degradation [12]. In this series of experiments, both the pressure and the content of oxygen were varied. A medium containing varying amounts of methanotrophs was applied to the coal samples and the volume of methane desorbed and the volume of CO_2 generated were then measured when the sample was pressurized with air containing a low concentration of oxygen.

Table 1: Components of the NMS Culture Medium

NO.	Components	Ratio
1	KNO_3, g	1.000
2	NH_4Cl, g	0.250
3	KH_2PO_4, g	0.272
4	$CaCl_2{\cdot}6H_2O$, g	0.200
5	$MgSO_4{\cdot}7H_2O$, g	1.000
6	$Na_2HPO_4{\cdot}12H_2O$, g	0.717
7	Trace element solution, mL	1
8	Distilled water, mL	1000

It can be concluded from these studies that it is feasible to solve the problems of gas in mines through the use of methanotrophs. Previous investigations have concentrated on field tests in which methane was degraded under static conditions [13]. However, in practical mining situations, the air flows as a result of ventilation and the flow-rate and gas concentration will vary with location. To systematize the treatment of mine gases by microbes, the effect of the air flow-rate on the variation in methane concentrations should be investigated. We therefore designed an experimental system to investigate the ability of methanotrophs to degrade methane based on the air flow-rate in ventilation roadways, working faces, upper corners and gobs.

3. Experimental equipment

3.1. The bacteria liquid and medium

The bacteria liquid is a kind of mixed bacteria medium containing methanotrophs with high-oxidation ability on methane. The bacteria sample is separated, purified and cultured based on soil enrichments obtained from the roots of rice at FENG Lou in Henan Province. The culture medium adopted named NMS is a kind of medium containing no carbon source and the formula of the medium is listed in Tables 1 and 2.

3.2. Experimental system

Fig. 2 shows that our bio-oxidation reaction system for mixed gases consists of a mixing system, a monitoring

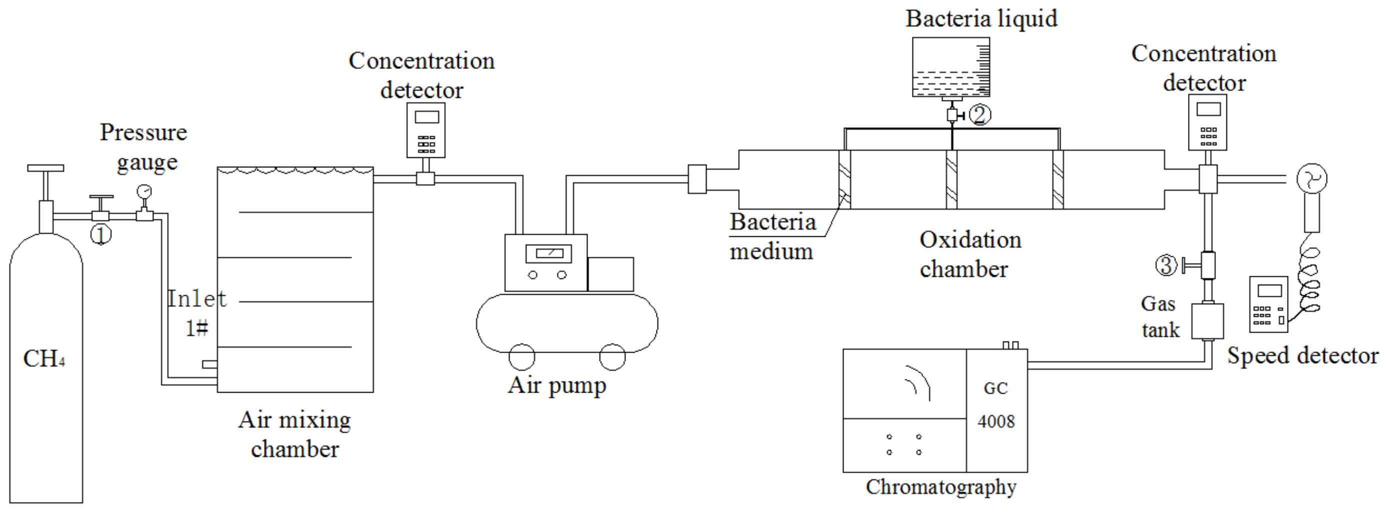

Figure 2: Experimental System

Table 2: Components of the Trace Elements

NO	Components	Ratio
1	H_3BO_3, g	0.030
2	Na_2EDTA, g	0.500
3	$CoCl\cdot6H_2O$, g	0.020
4	$NiCl_2\cdot6H_2O$, g	0.002
5	$FeSO_4\cdot7H_2O$, g	0.200
6	$CuSO_4\cdot5H_2O$, g	0.030
7	$ZnSO_4\cdot7H_2O$, g	0.003
8	$MnSO_4\cdot4H_2O$, g	0.003
9	$NaMnO_4\cdot2H_2O$, g	0.003
10	Distilled water, mL	1000

system, an oxidation reaction system and a system to measure the air flow-rate.

3.3. Experimental procedure

To simulate the effects of the hanging nets containing the methanotrophic bacteria on the degradation of methane and to investigate the effects of varying the gas concentration and flow-rate at a fixed temperature of 23°C, the experimental procedure was divided into the following steps:

1. To ensure that the experiments were conducted in a closed system, the junctions were sealed before the tests.
2. Tests were conducted on the air tightness of the system and the accuracy of the apparatus. Valve No. 1 was shut, and the air pump that controlled the flow volume was turned on so that the volume of flow through the air pump could be adjusted. The computed and monitored flow-rates were calibrated based on the size of the oxidation chamber and the distribution pipe.
3. The air pump was turned on and valve No. 1 was unseated slightly. Calibration without the addition of the bacterial medium was carried out based on the observed values from two content monitors.
4. The air pump was adjusted to ensure a fixed flow-rate and the gas concentration in the mixing chamber was changed by adjusting valve No. 1. Valve No. 2 was unseated, and the liquid containing the bacteria was injected. The gas concentrations from the two monitors were recorded and further analysis was conducted on the obtained results. Where required to analyze the air component, valve No. 3 was unseated to inflate the restoring tank and the air could then be analyzed by gas chromatography.
5. The air pump and valve No. 1 were adjusted at the same time to ensure that the gas concentration in the system was uniform. The gas concentrations and variations of the air components were analyzed at different flow-rates through the oxidation chamber.
6. Step 5 was repeated with different gas concentrations in each step.
7. Measurements were performed to prevent the accumulation of gas in the laboratory.

4. Laboratory experiments and the degradation of gas at different air flow-rates

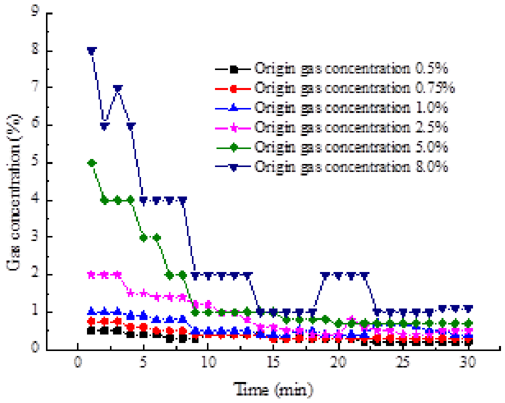

Figure 3: Gas concentration curve when the flow speed is 0.25 m/s

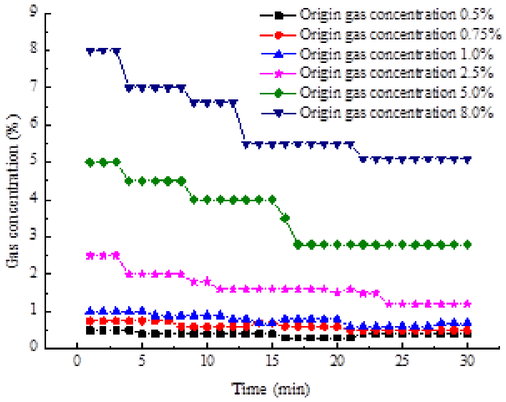

Figure 5: Gas concentration curve when the flow speed is 1 m/s

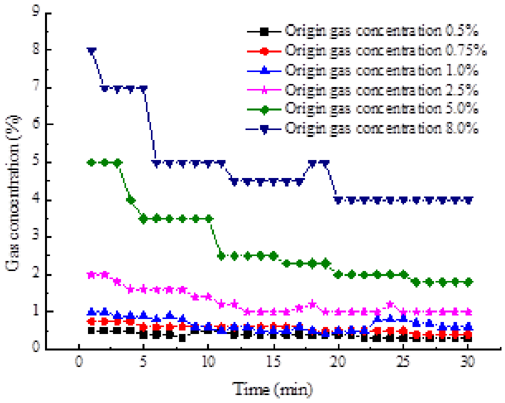

Figure 4: Gas concentration curve when the flow speed is 0.5 m/s

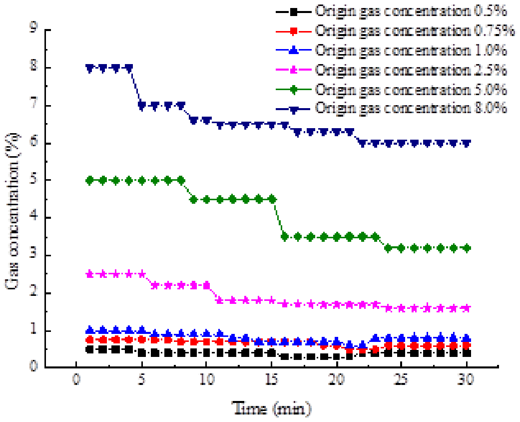

Figure 6: Gas concentration curve when the flow speed is 1.5 m/s

Safety regulations for return airways in coal mines specify that the gas concentrations in the main return airway, working faces and tail roadways should be <0.75%, <1% and <2.5%, respectively. The theoretical lower limit for gas explosions is 5%; a common gas concentration of 0.5% and a maximum concentration of 8% are also adopted. Based on the requirements fro air speed, flow-rates of 0.25 m/s, 0.5 m/s, 1 m/s, 1.5 m/s and 2 m/s were adopted. Five different gas concentrations (0.5%, 0.75%, 1%, 2.5%, 5% and 8%) were adopted at each flow-rate. The concentrations of methane and CO_2were recorded in the air after it had flowed through the oxidation chamber in which hanging nets containing methanotrophic bacteria were fixed (Fig. 3–7).

The following conclusions can be drawn from Figures 3–7 and Table 3:

1. Within the range of air flow-rates studied, both the volume and the rate of degradation of the gas increased as the gas concentration in the air flow increased at a constant flow-rate. It can be concluded that the methanotrophs rapidly oxidized methane when oxygen was abundant and that the volume of gas degraded increased as the gas concentration in the air flow increased. These results confirm the feasibility of treating mine gases using microbes.
2. Within the gas concentration range studied here, at a constant gas concentration, the volume of gas degraded increased as the flow-rate of air increased at flow-rates<1.5 m/s. However, the volume of gas degraded decreased slightly as the flow-rate of air increased to speeds> 2 m/s. The total volume of gas in the oxidation chamber per unit time increased as the

Table 3: Methane Desorption Volume in Different Flow Speeds after 30 minutes, m^3/30 min

Speed, m/s	0.5%	0.75%	1%	2.5%	5%	8%
0.25	0.006476	0.012187	0.01413	0.056638	0.123755	0.195112
0.5	0.007771	0.013424	0.023079	0.088313	0.158492	0.219015
1	0.013344	0.017723	0.026271	0.10425	0.174306	0.246864
1.5	0.022608	0.018722	0.038858	0.125051	0.198527	0.29885
2	0.015072	0.017898	0.032028	0.091374	0.17427	0.20724

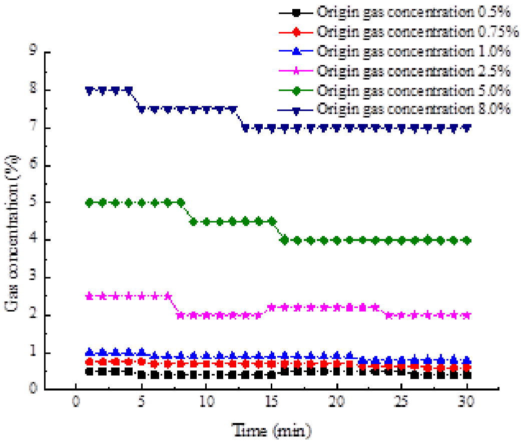

Figure 7: Gas concentration curve when the flow speed is 2.5 m/s

flow-rate increased. At low flow-rates, the length of time that there was contact between the gas and the methanotrophs increased, resulting in more complete oxidation of the methane and higher volumes of degraded gas. However, as the flow-rate increased, the contact time between the gas and the methanotrophs was insufficient for a complete reaction to take place, resulting in a lower volume of degraded gas.

3. The experiments showed that the degradation efficiency was low or even zero during the first few minutes in the oxidation chamber. This may be explained as follows. As valve No. 2 was unseated, a water film formed on the medium as a result of gravity and imbibition. The pores of the medium were therefore blocked from the air flow, and few contacts were formed between the methane gas and the methanotrophs. After a few minutes, this water film was blown away by the flow of air and the amount of contact between the methane and the methanotrophs increased rapidly, resulting in an increased volume of degraded gas. The use of a solid medium with a high porosity and large specific surface area is therefore to be preferred.
4. It was observed during the experiments that the gas concentrations fluctuated, but that the concentration was in an equilibrium state 23 min after the concentration decreased. This phenomenon may have resulted from a number of factors, such as changes in the growth environment of the microbes, or changes in the concentration of the methanotrophs during the addition process or during the cooling of the apparatus after 40 min of operation. It was observed that the gas concentration decreased monotonically after the flow of air through the oxidation chamber and then remained at an equilibrium value.
5. In the experiments, the recorded volumes of CO_2 indicated that the volume of CO_2 increased as the volume of degraded gas increased. However, these increases in volume of CO_2 were limited, and it can therefore be concluded that not all the methane was converted into CO_2. Analysis showed that some of the increase in the volume of CO_2 resulted from intermediates such as methanol and formaldehyde in the oxidation chamber.

5. Conclusions

The results obtained in this study show that the volume of degraded gas increased as the gas concentration increased at low air flow-rates. The feasibility of using methanotrophs to treat gases in mines was confirmed. The experimental results have great practical significance for mining and show that methanotrophs could be used to treat mine gases during exploration, including in the treatment of gases in gobs and caving zones and in problem areas such as upper corners.

The gas concentration and air flow-rates used in the experiments were restrained by limits on the automation of the system and the pump capacity. Despite these limitations, the feasibility of treating mine gases by applying methanotrophic bacteria was verified by our experiments, which also provide a theoretical basis for future investigations of safety issues in coal mining.

Acknowledgments

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