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Influences of different substrates on simulated lignite biogas production

Wang Aikuan *, Qin Yong, Shao Pei

School of Resources and Geoscience, China University of Mining and Technology, Xuzhou 221116, China
Key Laboratory of CBM Resources and Pooling Process, Ministry of Education of China, Xuzhou 221116, China

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ABSTRACT

Using lignite samples, selected from Zhaotong basin, Yunnan province, China, as the parent source, simulating experiments of lignite biogas were conducted with 0.1% methanol, 5 mg/L yeast extract and 0.2 mol/L sodium acetate solutions as the exogenous substance respectively. Variation characteristics of gas production, gas composition, VFA content and activity of coenzyme F_{420} in the simulated process were analyzed to discuss the influence of different substrates on lignite biogas generation. The results show that 0.1% methanol and 5 mg/L yeast extract solutions increase VFA contents in the biogas generation system ($p < 0.05$) and inhibit coenzyme F_{420} and methanogen activities significantly, so they decrease both gas amounts ($p < 0.05$) and CH_4 contents ($p < 0.05$). 0.2 mol/L sodium acetate solution activates coenzyme F_{420} and methanogen activities and improves the efficiency of enzymatic reaction, so the gas quantity ($p < 0.05$) and the CH_4 content ($p < 0.01$) increase significantly. Therefore, sodium acetate can be one kind of good exogenous substance for the generation of lignite biogenic gas.

1. Introduction

Coalbed biogas has received great attention for its vital energy and resource significance [1–4]. Biogenic coalbed gas generated from the biodegradation of coalbed organic matter by fermentation and a combination of methanogens and other anaerobic or facultative anaerobic microorganisms. The biodegradable organic matter in coal is the material basis for biogas generation. The active maintenance of anaerobic microbial flora is a prerequisite for coalbed biogas production [5,6]. At present, many scholars have successfully simulated coalbed biogas production under laboratory conditions and achieved many significant breakthroughs, which lay the foundation for both the further understanding of the coalbed biogas production mechanism and coal micro-organism underground gasification exploration. It is also important for the theoretical and practical exploration and development of coalbed gas resource [7–10].

Under suitable conditions, there exist activated methanogens in coal seams, which could generate bio-methane using substances in coal [11–13]. But it is not clear how to activate the anaerobic microbes effectively underground. For example, a whole methanogen ecological system was found in a coalbed methane well in the west of Canada, but the difficult nutritional conditions limited their activation [14]. Furthermore, coal micro-organism underground gasification exploration requires underground injection of a nutrient solution and bacterial species. Thus, the difficulty is great and there is no relevant report in this aspect.

On the basis of previous work [15], this paper utilizes lignite-source methanogens by enrichment culture as a bacterial source to conduct simulation experiments in the laboratory. Different substrates are added to simulate the lignite biogas production process. Gas production, gas composition, VFA content and activity of co-enzyme F_{420} in the system are selected as the bacterial degradation indexes of lignite biogas. The influences of different substrates on lignite biogas production are studied and the result will be significant for the increasing production and efficiency of biogenic coalbed gas.

2. Sample collection and experimental method

2.1. Lignite samples

The lignite used in the experiment is a newly-exposed lignite sample collected in an opencast coal mine named Hongni, in Zhaotong basin, Yunnan province, China. The basic properties of the sample are shown in Table 1. The reflectivity of vitrinite (R_o) of the coal sample is 0.31%, placing it in the soft lignite phase of the immature stage. The volatile matter content of the sample is high, indicating an easier degradable potential. The sample contains rich hydrogen, rich oxygen and rich nitrogen, showing a good

* Corresponding author. Tel.: +86 13815383142.
E-mail address: wake198110@163.com (A. Wang).

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Table 1
Basic properties of the lignite sample.

Depth (m)	R_o (%)	Industrial analysis (%)			Element analysis (%)					Maceral (%)		
		M_{ad}	A_d	V_{daf}	C_{daf}	H_{daf}	O_{daf}	N_{daf}	S_{daf}	Huminite	Inertinite	Liptinite
20	0.31	6.32	6.47	49.77	69.87	5.84	21.50	1.61	1.18	82.4	14.5	3.1

biochemical activity, which is beneficial to micro-organism cultivate enrichment and simulation experiments.

After the collection, the sample is stored in an anaerobic glove box to prevent the oxidation and destruction of the ecological system in lignite. After the smash of lignite, coal dust below 60-order is screened for the experiment. The bacteria solution used in the experiment is the parent methanogens that had been enrichment cultured from a lignite sample collected in Zhaotong basin, Yunnan province [15]. After culture enrichment, the bacterial concentration was 2.0×10^{10}/mL. Enrichment medium composition and flora enrichment methods in the experiment are shown as literature [15].

2.2. Experimental methods

Simulation experiments were divided into control and experimental groups. In the control group, 1 mL of seed liquid and 19 mL of sterile de-oxygenated deionized water were added to the reactor, with 2 g of the inoculated lignite sample. Three experimental groups were prepared: 1 mL seed liquid, 2 g of 60-order lignite sample and 15 mL of deionized water are added to these 3 experiment groups. 4 mL 0.1% (volume) methanol solution was added to Group 1; 4 mL of 5 mg/L yeast leaching solution was added to Group 2 and 4 mL of 0.2 mol/L sodium acetate solution was added to Group 3. To deduct the gas production of the bacterial solution itself, a blank experiment was conducted. Namely, only 1 mL of seed liquid and 19 mL of sterile de-oxygenated deionized water were added to the reactor. All the designs are of two groups in parallel. Split charging, inoculation and other operations were carried out in an anaerobic glove box. Each reactor was sealed with an aluminium cap after sealing with an isobutyl rubber plug. After sealing, it was carefully moved from the glove box and placed in a vibration cultivating box at 35 °C. In the experiment, lignite gas production was regularly examined in terms of gas components, VFA content and activity of co-enzyme F_{420}. Intubation tubes were used in the bottle plug for drainage and gas collection.

The measurement of gas composition was carried out using the gas chromatographic method (7890GC) and implemented according to GB13610-2003 of China. A thermal conductivity detector was used, with a detection temperature of 80 °C, a chromatographic column temperature of 50 °C and a driving gas of 0.5 MPa normal oxygen.

After 6000 rpm centrifugation, a GC910 gas chromatograph (produced by Shanghai Kechuang Chromatography Instruments Co. Limited., China) was used to measure VFA content in the reaction liquid. The measured components included acetic acid, propionic acid, butyric acid and valeric acid. The chromatographic instrument uses nitrogen as a carrier gas. Also, a hydrogen flame ionization detector was utilized, with a column temperature of 190 °C and a sample size of 2 μL.

For testing of co-enzyme F_{420}, normal saline is added to 10 mL of reaction solution and then stirred. After 6000 r/min centrifugation for about 15 min, the precipitation was put into a 95 °C water bath for 30 min then cooled, followed by the addition of ethanol (ethanol: precipitation = 2.5:1 (v/v)). After 6000 r/min centrifugation for 15 min, the supernatant was taken and divided into two parts. One part was regulated to pH = 3 with 6 mol/L HCl and the HCl volume was recorded. Distilled water was added to the other part until the volume was the same as the HCl. The two solutions are colorimetrically determined at a wavelength of 420 nm, and the activity of co-enzyme F_{420} is calculated using the formula:$C = \frac{Af}{sL}$. In the formula, C is the concentration of co-enzyme F_{420}, (μmol/L), A is the extinction value of pH < 3 solution, (g/(cm mol)), f is the dilution multiple, L is the thickness of the colorimeter cell, (cm), s is the extinction value of F_{420} and s = 54.3.

2.3. Data processing and analysis

SPSS 13.0 software was used and the t-test was adopted to check the data differences of corresponding indexes between the experimental group and control groups. Test results with $p \leqslant 0.01$, refers to a highly significant difference; if $p \leqslant 0.05$, it refers to significant difference; and if $p > 0.05$, it refers to no significant difference.

3. Results and discussion

3.1. Effects of different substrates on lignite biogas production

Total biogas production of experimental group and control groups in the simulation are shown as Table 2 and Fig. 1.

In the initial stage of the experiment, the gas production in the methanol treatment group is similar to that in the control group. However, from the 24th day, gas production in the methanol group significantly decreases and, on the 48th and 60th days, there is almost no gas production, which indicates that 0.1% methanol has an obvious inhibition effect on lignite biogas production ($p < 0.05$). Studies have reported that methanogens can directly convert methanol to methane and also can directly convert methanol into acetic acid and then form methane [16,17]. However, this requires methanogens to have some acid resistance [18] because, in the interaction processes of these two approaches, CO_2 is generated, which may decrease the pH value in the system. In this simulation experiment, methanogens are mature bacteria inoculated

Table 2
Production of lignite biogas when added different types of substrate (mL/g).

Groups	Time (d)					Sig. p
	12	24	36	48	60	
Control group	1.376	2.637	5.275	11.122	11.811	
0.1% methanol	1.376	0.688	0.459	0.344	0.115	0.033
5 mg/L yeast leaching solution group	1.605	1.376	1.261	1.032	0.459	0.042
0.2 mol/L sodium acetate group	1.835	4.357	6.536	15.021	15.136	0.015