Comparison and analysis of organic components of biogas slurry from eichhornia crassipes solms and corn straw biogas slurry

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Abstract. Biogas slurry is one of anaerobic fermentations, and biomass fermentation biogas slurries with different compositions are different. This paper mainly presents through the anaerobic fermentation of Eichhornia crassipes solms biogas slurry and biogas slurry of corn straw, the organic components of two kinds of biogas slurry after extraction were compared by TLC, HPLC and spectrophotometric determination of nucleic acid and protein of two kinds of biogas slurry organic components, and analyzes the result of comparison.

1. Introduction

Biogas slurry is a product of anaerobic fermentation, the available nutrient ability, nutrients utilization rate is high, the crop can be quickly absorbed, not only can improve the yield and quality of crops, but also has the functions of preventing and anti-adversity effect, is a kind of high-quality organic liquid fertilizer [1]. Biogas slurry not only contains rich soluble inorganic salts, but also contains biochemical products of anaerobic fermentation, which has the functions of nutrition, bacteriostasis, stimulation and anti-adversity [2]. At the same time, biogas slurry can kill and inhibit crop diseases and insect pests, and has repellent effect on some insects. At present, the biogas slurry has the dual effect of biological fertilizer and biological pesticide and will not bring resistance and environmental pollution, but also reduces nitrate accumulation in crops [3], broad-spectrum known as anaerobic microorganism processing agent in the world [4].

Water hyacinth (Eichhornia crassipes Solms) is pontederiaceae water hyacinth is Chinese, name of Eichhornia crassipes. It has a erect oval gourd stem because of its dark green leaves, commonly known as water hyacinth. Water hyacinth is an aquatic herb, which can grow at a suitable temperature and absorb large amounts of N, P and K nutrients. It has been recognized as a water plant with strong N and P capacity in the world. At present, the research on water hyacinth mainly focuses on the research of water hyacinth nutrition, water hyacinth control and comprehensive utilization of research, now studies focus on water hyacinth removal in eutrophic water capacity of nitrogen and phosphorus and its application in water treatment, but its use in fertilizer has not been reported in details [5]. In 1901, it was introduced into China as a flower. In 30s, it was introduced into China's provinces as livestock and poultry feed, and promoted as a plant for ornamental and purifying water quality. Because of its asexual propagation speed, it has been widely distributed in 19 provinces of North China, East China, central China, Southern China and the southwest, especially in Yunnan (Kunming), Zhejiang, Fujian, the invasion of Jiangsu, Sichuan, Hunan, Hubei, Henan province is serious, and has
spread to temperate regions. It caused great environmental problems and caused serious harm to the ecosystem [6]. Corn straw with C, H, O elements as the main content of the three part, accounting for more than 90% of the total elements, straw and carbohydrate rich material, the main ingredients of corn straw is wood fiber, wood fiber consists mainly of cellulose, hemicellulose and lignin [7]. At present, China's straw comprehensive utilization technologies include: organic fertilizer utilization technology, energy utilization technology, cultivation edible mushroom technology, industrial raw materials utilization technology and feed utilization technology. Although organic fertilizer utilization technology has made great progress, but the research is still not deep enough, and the research results cannot be effectively industrialized [8].

Study on water hyacinth liquid focused on the removal of N, P, and research on the application of water hyacinth also weakened, moreover, studies especially on the nutrients in the study of farmland utilization is still not seen [9]. This paper mainly presented through the anaerobic fermentation of water hyacinth biogas slurry and biogas slurry of corn straw, how two kinds of slurry the organic components extracted, using the analysis method of the analysis of the differences of organic components in biogas slurry.

2. Experiment

Experimental flow chart:

![Figure 1. Chart of experiment.](image)

2.1. Fermentation experiment of water hyacinth marsh

After the water hyacinth plant was removed, the roots were treated and cleaned to reach the required cleanliness at the experimental stage, and the shredder was used for processing. Take 100 g water hyacinth crushed in the three cervical fermentation flask, add 300 ml distilled water, adding 30 ml after experimental treatment of liquid manure, at room temperature (25 DEG C) mixed anaerobic fermentation, and the use of drainage gas gathering method of gas production record.

2.2. Fermentation experiment of corn stalk

After the corn stalks were removed, the roots and leaves were removed and cleaned, and the required cleanliness in the experimental stage was achieved, and the mills were treated. Remove the 100 g of
corn straw in the three cervical fermentation flask, add 300 ml distilled water, adding 30 ml after experimental treatment of liquid manure, at room temperature (25 DEG C) mixed anaerobic fermentation, and the use of drainage gas gathering method of gas production record.

2.3. Extraction of organic components in biogas slurry
Take 100 ml water hyacinth biogas slurry in a separatory funnel, adding 100 ml petroleum ether first-order extraction, then separated with a separatory funnel, then add 50 ml of petroleum ether for two order extraction, the organic layer was poured into the flask preservation, water hyacinth biogas slurry of petroleum ether extract.

100 ml corn straw biogas slurry in a separatory funnel, adding 100 ml petroleum ether first-order extraction, then separated with a separatory funnel, and add 50 ml of petroleum ether for second order extraction, the organic layer is poured into the flask was saved, corn straw biogas slurry of petroleum ether extract.

Take 100 ml water hyacinth biogas slurry in a separatory funnel, with ethyl acetate 100 ml one-step extraction, and then use the separatory funnel water leakage, ethyl acetate were added to the two order 50 ml extraction, the organic layer is poured into the flask was saved, the ethyl acetate extract of water hyacinth biogas slurry.

Take 100 ml corn straw biogas slurry in a separatory funnel, with ethyl acetate 100 ml one-step extraction, and then use the separatory funnel water leakage, ethyl acetate were added to the two order 50 ml extraction, the organic layer is poured into the flask was preserved, ethyl acetate extract of corn straw biogas slurry.

Take 100 ml water hyacinth biogas slurry, pour it into a rotary evaporator for evaporation, with 5 ml methanol organic matter and dissolved, the liquid poured into the flask by water hyacinth biogas raw preservation, dry methanol extract.

Corn straw biogas slurry 100 ml, pour it into a rotary evaporator for evaporation, with 5 ml methanol organic matter and dissolved, the liquid poured into the flask was preserved, corn straw raw dry methanol extract of biogas slurry.

2.4. Comparative analysis of organic ingredients
The comparative analysis of the organic extracts of two kinds of biogas slurry extract mainly adopts the following analysis methods:

Thin layer chromatography (TLC) is to apply suitable fixed phases to glass plates, plastic or aluminium substrates to form a uniform layer. After sampling and unfolding, the method compares the specific shift value (Rf) of the chromatogram obtained by the same method according to the specific value of the shift value (Rf) and the appropriate contrast, and is used for the identification of the drug, the impurity inspection or the content determination method. Thin layer chromatography (TLC) is an important experimental technique for rapid separation and qualitative analysis of small amounts of substances. It is also used to track the reaction process. In the course of this experiment, two kinds of samples were analyzed by fluorescence detection, iodine vapor color detection and sulfuric acid color detection.

Two kinds of biogas slurry pretreatment

Extraction of organic components from two biogas slurry

Fluorescence color test, Iodine vapor color test, Sulfuric acid color test

Figure 2. flow chart of experiment.
High Performance Liquid Chromatography (HPLC) is an important branch of chromatography, the mobile phase is liquid, using high pressure infusion system, mixed solvent, buffer flow will have a single solvent with different polarity or different proportion of the pump into the fixed phase column, the column the components are separated into. To detect, so as to realize the analysis of the sample.

Spectrophotometry is used to determine the absorbance of a light at a specific wavelength or a certain wavelength range. In this paper, the content of nucleic acid and protein in biogas slurry was analyzed by spectrophotometer.

2.5. Results and analysis

2.5.1. Experimental results and analysis of fermentation. Analysis of the fermentation of water hyacinth biogas slurry: in the process of experiment, it was found that the color of liquid in the fermentation device changed from light color to dark color, and the amount of drainage in this process also increased gradually. We further studied the fermentation broth of 120 h. The record of test is shown in Table 1.

![Table 1. Fermentation of eichhornia crassipes solms.](image)

Analysis of the fermentation of corn straw biogas slurry: during the experiment, it was found that the color of the liquid in the fermentation plant changed from light color to dark color, and the amount of drainage in this process also increased gradually. We further studied the fermentation broth of 120 h. The record of test is shown in Table 2.

![Table 2. Fermentation of corn straw slurry.](image)

2.5.2. Results and analysis of thin layer chromatography. After TLC analysis of chromogenic results can be obtained in fluorescence color experiment, water hyacinth biogas slurry of petroleum ether extract, ethyl acetate extract of water hyacinth biogas slurry, water hyacinth Biogas Dry methanol extraction system of the original color liquid three subjects. The result is obvious in the color experiment in iodine vapor. Water hyacinth biogas slurry of the petroleum ether extract, ethyl acetate extract of water hyacinth biogas slurry, biogas slurry of the petroleum ether extract of corn straw color liquid three subjects. The result is obvious; the color experiment in concentrated sulfuric acid, alcohol, ethyl acetate extract of water hyacinth biogas slurry, biogas slurry of the petroleum ether extract of corn straw color liquid three subjects. The result is obvious. Through the results of color rendering experiments, we can preliminarily judge that there may be many organic compounds with high polarity. In view of this series of phenomena, the experimental subjects were analyzed by high performance
liquid chromatography (HPLC).

2.5.3. Results and analysis of high performance liquid chromatography. Sample HPLC test results (methanol: water, 6:4, retention time, 40 min).

According to figure 3 and figure 4. Through the comparative analysis of water hyacinth and biogas slurry of corn straw and found that the two test objects in second minutes to peak area obviously fourth minutes in water hyacinth biogas slurry, first to second minutes is obvious in the peak district; corn straw biogas slurry, fifteenth to sixteenth minutes and seventeenth to nineteenth two minutes has obvious peak area. It can be explained that water hyacinth contains organic compounds with higher polarity, which can peak before corn stalk. From the map, it also reflects the obvious difference between the two kinds of stock solution. This series of differences makes the water hyacinth and corn stalk biogas slurry have different characteristics.
According to figure 5 and figure 6. Through the comparative analysis of the water hyacinth and corn straw biogas slurry of petroleum ether and methanol extract showed that two test objects in second minutes to the peak area, was fourth minutes and tenth minutes in the corn straw, to eleventh minutes, Twelfth minutes to fourteenth minutes, sixteenth minutes to eighteenth minutes, three hours there will be clear that the peak oil organic ingredients of corn straw biogas slurry in soluble components of larger organic species rich.

According to figure 7 and figure 8. Through the comparative analysis of the water hyacinth and corn straw biogas ethyl acetate methanol extract showed that the two test objects in second minutes to peak area obviously in twelfth minutes to fourth minutes, fourteenth minutes have obvious peak area. In second minutes to fourth minutes, the difference between the peak value of the two subjects was large, and the peak time was also different. Among them, the extraction time of water hyacinth biogas
slurry is earlier, the peak value is higher, and it also shows that the organic matter in water hyacinth slurry is good in ester solubility, organic matter composition is more, and polarity is higher.

2.5.4. Results and analysis of nucleic acids and proteins detected by spectrophotometer. By spectrophotometer in nucleic acid and protein and pH test analysis of water hyacinth and corn straw biogas slurry.

Table 3. Analysis of maize biogas slurry.

| Test Name          | Parameters | Wavelength(s) | Result | Displayed Units |
|--------------------|------------|---------------|--------|----------------|
| DNA concentration  | f 1=62.9   | A1=A\textsubscript{260nm} | A\textsubscript{1}=0.009 | DNA mg/ml       |
|                    | f 2=36.0   | A2=A\textsubscript{280nm} | A\textsubscript{2}=0.015 | Protein mg/ml   |
|                    | f 3=1552   | A\textsubscript{ref}=A\textsubscript{320nm} | A\textsubscript{ref}=0.001 |              |
|                    | f 4=757.3  |              | C-DNA: 0.097 |              |
| Protein concentration | f 1=49.1   | A1=A\textsubscript{260nm} | A\textsubscript{1}=0.002 | DNA mg/ml       |
|                    | f 2=3.48   | A2=A\textsubscript{230nm} | A\textsubscript{2}=0.006 | Protein mg/ml   |
|                    | f 3=183    | A\textsubscript{ref}=A\textsubscript{320nm} | A\textsubscript{ref}=0.004 |              |
|                    | f 4=75.8   |              | C-DNA: 0.081 |              |
|                    |            |              | C-Pro: 0.4224 |              |
|                    |            |              | Ratio: 0.912  |              |

Table 4. Analysis of Eichhornia crassipes solms biogas slurry.

| Test Name          | Parameters | Wavelength(s) | Result | Displayed Units |
|--------------------|------------|---------------|--------|----------------|
| DNA concentration  | f 1=62.9   | A1=A\textsubscript{260nm} | A\textsubscript{1}=0.007 | DNA mg/ml       |
|                    | f 2=36.0   | A2=A\textsubscript{280nm} | A\textsubscript{2}=0.014 | Protein mg/ml   |
|                    | f 3=1552   | A\textsubscript{ref}=A\textsubscript{320nm} | A\textsubscript{ref}=0.013 |              |
|                    | f 4=757.3  |              | C-DNA: 0.408 |              |
|                    |            |              | C-Pro: 5.643 |              |
|                    |            |              | Ratio: 9.361 |              |
| Protein concentration | f 1=49.1   | A1=A\textsubscript{260nm} | A\textsubscript{1}=0.010 | DNA mg/ml       |
|                    | f 2=3.48   | A2=A\textsubscript{230nm} | A\textsubscript{2}=0.014 | Protein mg/ml   |
|                    | f 3=183    | A\textsubscript{ref}=A\textsubscript{320nm} | A\textsubscript{ref}=0.007 |              |
|                    | f 4=75.8   |              | C-DNA: 0.770 |              |
|                    |            |              | C-Pro: 2.428 |              |
|                    |            |              | Ratio: 0.841 |              |

In table, F1, F2, F3 and F4 are related calculation factors A1, A2, A\textsubscript{ref}, and the wavelength used in calculation.

From the two kinds of biogas slurries, composition contrast can be seen in the table that two kinds of slurries have obvious differences in the content of DNA and protein. Moreover, differences in the content of protein and nucleic acid are also relatively obvious. It also shows that the two kinds of functional differences in biogas slurry as fertilizer. After repeated measurements of pH meter, water hyacinth biogas slurry pH was 5.68. The pH value of corn straw biogas slurry was 5.08. Different acidity and alkalinity have different effects on fertilizer.

3. Conclusion

Eichhornia crassipes solms biogas slurry and biogas slurry of corn straw by anaerobic fermentation experiment, the two kinds of slurries after TLC analysis method can find that there are obvious differences in two kinds of organic components of biogas slurry, the analysis results in high performance liquid chromatography in corn straw biogas slurry which is rich in organic ingredients,
after the spectrophotometric determination of nucleic acid and protein, the two kinds of slurry organic components are obviously different, which was caused by the characteristics of two kinds of slurries with certain differences. According to the difference of two kinds of biogas slurries, the influence of application should be further studied.

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