

Study of Factors Influencing the Content of Total Nitrogen in Anaerobic Fermentation Slurry

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Abstract: The content of total nitrogen in anaerobic fermentation slurry produced from pig dejecta was measured to determine how the changes of fermentation conditions affect it. The experimental results indicated that the content of total nitrogen in anaerobic fermentation slurry changed low when fermentation temperature was 15°C and changed obviously at 30°C and 55°C. The effect of material concentration on the content of total nitrogen in anaerobic fermentation slurry was more obvious. The change of the content of total nitrogen was most notable when material concentration was 3.7%. The content of total nitrogen changed low when material concentration was 0.925% and 1.85%. Without agitation, the content of total nitrogen changed low. With agitation, the change of the content of total nitrogen was higher. Increase of inoculation volume might increase the change of the content of total nitrogen. The change of the content of total nitrogen was higher when pH value was 7 and the content of total nitrogen changed slowly at pH5 and pH 8.5.

Keywords: anaerobic fermentation slurry; total nitrogen; anaerobic fermentation process

1. INTRODUCTION

Nitrogen (N) is one of three important elements which are necessary for the growth of farm crops. A large number of experiments indicate that anaerobic fermentation slurry is a high quality organic fertilizer^[1-11]. When anaerobic microbes synthesize self-material to satisfy the self-growth, they need less nitrogen and phosphorus for consuming. There are much nitrogen and phosphorus in anaerobic fermentation slurry. This is one of the reasons why the anaerobic fermentation slurry is better than other organic fertilizers as a fertilizer.

Soil and Fertilizer Institute, Hubei Academy of Agricultural Sciences analyzed statistically several hundreds of samples of anaerobic fermentation slurry and cinder in different methane tank. The results showed that the content of nitrogen in different samples differs greatly, the average content was 0.39%^[12]. The other important reason is the effect of different fermentation technological parameters on the content of total nitrogen in anaerobic fermentation slurry except the difference of raw materials. However, researches on the factors influencing the content of ammonia nitrogen in anaerobic fermentation slurry are little. It is not in favor of the development and usage of anaerobic fermentation slurry. The aim of this paper is to study the change rules of the content of total nitrogen in anaerobic fermentation slurry produced under different technological conditions with fermentation time and probe technological conditions for the production of superior anaerobic fermentation slurry.

2. MATERIALS AND METHODS

2.1 Raw Material

Raw materials used in this experiment were fresh pig dejecta collected in Xinmuda piggery in the east suburb of Zhengzhou, China. The feed for the swine is mainly chaff, corn, and vegetable leaves. Test samples were prepared from the fresh pig dejecta removed the blocks of impurities through rough filter. Main physicochemical properties of the material include 81.5% water, 15% organic material, 0.91% nitrogen, and 0.33% P with pH value of the feed liquid from 7.4 to 8.0. The inocula were anaerobic fermentation slurry from the methane tank that was running normally in Typical Garden of Future Agriculture in Qi country, Henan Province and waste liquid after anaerobic fermentation in IC reactor in Key Laboratory of Renewable Energy of the Ministry of Agriculture, Henan Agricultural University.

2.2 Experimental Principle

Experiments that measured technological parameters of fermentation were operated in BIOF-2010 Biological Fermentation Tank. Its micro-computer control system can control technological parameters such as fermentation temperature, rate of agitation, pH value automatically. Its control scope of temperature is 5~65°C, scope of rate of agitation is 50~800r/m, scope of pH value is 2~12 and working pressure is 0 ~ 0.05MPa. Through controlling important technological parameters influencing anaerobic fermentation such as fermentation temperature, material concentration, rate of agitation, inoculation volume and pH value, we studied the change rules of the content of total nitrogen under different technological conditions.

Material concentration (%) is the percent of the quantity of dry pig dejecta (g) to the quantity of anaerobic fermentation slurry (g). Inoculation volume (%) is the percent of volume of inocula (ml) to volume of anaerobic fermentation slurry (ml).

Materials were fed in batches. A process of fermentation was completed with feeding once. A sample was taken once a day and the content of total nitrogen of the sample was measured once a day. Five technological parameters of fermentation were assorted and operated in experiments.

1) Study on the effects of fermentation temperature on the content of total nitrogen in anaerobic fermentation slurry. Temperature was 15°C, 30°C and 55°C respectively, the other conditions were constant: material concentration 3.7%, rate of agitation 25 r/m, inoculation volume 30% and pH value 7.

2) Study on the effects of material concentration on the content of total nitrogen in anaerobic fermentation slurry. Material concentration was 0.925%, 1.85% and 3.7% respectively, the other conditions were constant: fermentation temperature 30°C, rate of agitation 25 r/m, inoculation volume 30% and pH value 7.

3) Study on the effects of rate of agitation on the content of total nitrogen in anaerobic fermentation slurry. Rate of agitation was 0 r/m, 25 r/m and 50 r/m respectively, the other conditions were constant: fermentation temperature 30°C, material concentration 3.7%, inoculation volume 30% and pH value 7.

4) Study on the effects of inoculation volume on the content of total nitrogen in anaerobic fermentation slurry. Inoculation volume was 10% and 30% respectively, the other conditions are constant: fermentation temperature 30°C, material concentration 3.7%, rate of agitation 25 r/m and pH value 7.

5) Study on the effects of pH value on the content of total nitrogen in anaerobic fermentation slurry. pH value was 5, 7 and 8.5 respectively, the other conditions are constant: fermentation temperature 30°C, material concentration 3.7%, rate of agitation 25 r/m and inoculation volume 30%.

2.3 Experiment Methods

Total nitrogen was measured by digestion^[13]. The supernatant fluid after treating original anaerobic fermentation slurry with LXF—II B type centrifugal machine of multiple pipes with 5000r/m speed was test sample. Use the method of $H_2SO_4-H_2O_2-HClO_4$ digesting sample, take a share of solution digested and measure the nitrogen with distillation titration method. The content of total nitrogen can be measured with equation (1)^[14].

$$V_N = (V_f - V_0) \times N \times 14 \times 1000 / (W \times V) \quad (1)$$

Where, V_N —the content of total nitrogen in anaerobic fermentation slurry, mg/L;

V_f —volume of H_2SO_4 standard liquid consumed with titrating test sample, ml;

V_0 —volume of H_2SO_4 standard liquid consumed with titrating blank, ml;

N —normality of H_2SO_4 standard liquid, ml;

14—weight per gram molecule N, g;

1000—scale factor;

W —volume of test sample of anaerobic fermentation slurry, ml; and

V —volume of solution digested using to distil, ml.

3. RESULTS AND DISCUSSION

The source of substrate nitrogen that can be inverted and used by microbe is wide. According to the difference of their existence, it can be assorted into organic nitrogen, ammonia nitrogen and nitronitrogen. In anaerobic fermentation slurry, the main existence is organic nitrogen and ammonia nitrogen, and nitronitrogen is little. The total nitrogen measured with kjeldahl method includes ammonia nitrogen and organic nitrogen compound that can be inverted to ammonia salt on this condition. This kind of organic nitrogen compound mainly is protein, peptone, amino acid, nucleic acid, urea and other organic nitrogen compound with negative 3 valence nitrogen. It does not include trinitride, azine, azo, hydrazone, nitrate, nitroso, nitro, nitrite, nitrile, oxime and semicarbazone nitrogenous compound^[15].

3.1 Effect of Fermentation Temperatures

Changes of the content of total nitrogen in anaerobic fermentation slurry with fermentation time at different fermentation temperatures could be seen from Table 1 and Figure 1. The other experimental conditions were controlled as follows: material concentration 3.7%, rate of agitation 25 r/m, inoculation volume 30% and pH value 7.

According to the experiment design of anaerobic fermentation, three experiments of 15°C, 30°C and 55°C were settled and belonged to fermentation of low temperature, medium temperature and high temperature respectively. The curves of Figure 1 showed the experimental results of three experimental operating modes, their difference showed the effect of temperature factor on the content of total nitrogen in anaerobic fermentation slurry. As to the curve of 15°C, the content of total nitrogen in anaerobic fermentation slurry increased on the 1st day of fermentation, then keep about 800mg/L, the whole curve was straight and does not wave. It illuminated that the content of total nitrogen changed low in low temperature. When temperature is low, the speed of growth and reproduction of microbe is slow, and is difficult to start, in addition, the efficiency of using substrate is not high and nitrogenous derivative produced is little. So the change of the content of total nitrogen is little. The curves of 30°C and 55°C wave largely, the content of total nitrogen change obviously. It illuminated that microbe in fermentation tank dissolved substrate constantly, grew and reproduced largely. The biochemical reaction in system is acute. In the beginning of fermentation, the content of total nitrogen increased, it could be conjectured that this was the accumulation of proportion nitrogenous derivative produced by microbe. However the content of total nitrogen began to decrease after the 3rd day, probably because the lack of other nutrient element caused the slow-up of bacteria metabolism. The trend of two curves of 30°C and 55°C are same, the apex

appeared on the third day, but the apex of 55°C curve is obviously higher than that of 30°C curve. It could be conjectured that the higher temperature, the higher the apex of the content of total nitrogen.

Table 1: Changes of the content of total nitrogen in anaerobic fermentation slurry with fermentation time at different fermentation conditions(mg·L⁻¹)

		Fermentation time(d)				
		1	2	3	4	5
Fermentation Temperatures (°C)	15	285.60	770.02	786.80	795.21	898.84
	30	1187.76	899.61	1635.22	1125.61	952.03
	55	746.77	1174.04	2788.81	1713.63	1310.42
Material Concentrations (%)	0.925	105.00	226.80	327.61	375.20	400.41
	1.850	344.96	509.60	705.60	652.41	681.82
	3.700	1187.76	899.64	1653.20	1125.62	952.01
Rates of agitation (r·m ⁻¹)	0	736.40	770.00	817.61	1175.42	1008.03
	25	1187.76	899.64	1635.20	1125.61	952.04
	50	1125.04	944.72	962.36	1864.80	1405.61
Inoculation volumes (%)	10	968.24	944.72	1293.61	814.62	824.54
	30	1187.76	899.64	1635.22	1125.60	952.02
pH values	5	1310.41	1041.63	1209.64	1232.01	1338.43
	7	1187.76	899.64	1635.22	1125.61	952.04
	8.5	940.82	1047.20	1014.63	996.82	1019.22

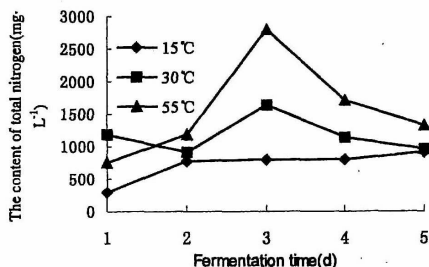


Figure 1: Changes of the content of total nitrogen with different fermentation temperature

3.2 Effect of Material Concentrations

Changes of the content of total nitrogen in anaerobic fermentation slurry with fermentation time at different material concentrations could be seen from Table 1 and Figure 2. The other experimental conditions were controlled as follows: fermentation temperature 30°C, rate of agitation 25 r/m, inoculation volume 30% and pH value 7.

As to Figure 2, the three curves showed the results measured of three experimental operating

modes that material concentration was 0.925%, 1.85% and 3.7% respectively. Because of the difference of initial substrate concentration, the gradient of initial nitrogen content showed by curves existed and was 105mg/L, 345mg/L and 1187.8mg/L. The trend of three curves differed highly, it could be conjectured that material concentration had an obvious effect on the content of total nitrogen. Figure 2 showed that the change of nitrogen content was most notable on the condition of higher material concentration. Substrate concentration was high, nutrient was sufficient, the growth of bacteria bloomed relatively, biochemical reaction was acute; Substrate concentration was lower, the change of nitrogen content was not high, such as when material concentration was 1.85%, the content of total nitrogen increased slowly to 705.6mg/L from 344.96mg/L at the beginning, then kept in 650mg/L. When material concentration was 0.925%, nitrogen content always kept between 100mg/L and 200mg/L. By estimate, it was because that C:N in pig dejecta was low (about 13:1), when substrate concentration was lower, element C was used rapidly by microbe, element C became the limiting factor for microbe growth, it caused that anaerobic fermentation was poor and microbe metabolism was inhibited.

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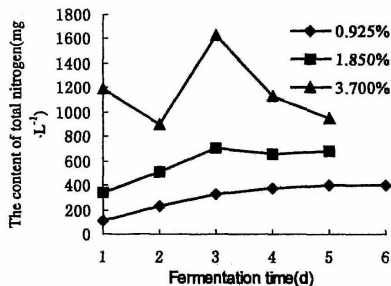


Figure 2: Changes of the content of total nitrogen with different concentration

3.3 Effect of Rates of Agitation

Changes of the content of total nitrogen in anaerobic fermentation slurry with fermentation time at different rates of agitation could be seen from Table 1 and Figure 3. The other experimental conditions were controlled as follows: fermentation temperature 30°C, material concentration 3.7%, inoculation volume 30% and pH value 7.

In this paper, the rates of agitation were controlled at 0 r/m, 25 r/m, and 50 r/m respectively and the other control conditions were constant. The advantage of agitation in anaerobic fermentation had three aspects: the 1st was prompting substrate temperature to distribute evenly, the 2nd was having bacteria and substrate had the sufficient chance of contacting, the third was increasing speed of anaerobic fermentation

with agitation. As to the bio fermentation tank used in experiments, heating layer was in the underlayer of tank and was 1/2 total tank volume. Without agitation, there was high temperature difference in the superjacent bed and underlayer of fermentation material. Furthermore, from the experiments we could see, without agitation, obvious stratification appeared, substrate deposited in the bottom of tank and the superjacent liquid only had a small quantity of flocculi. Because of the incompleteness of substrate reaction, the effect of anaerobic fermentation was poor. The experimental results (as Figure 3) showed that without agitation, the content of total nitrogen changed low and the average growth rate in the first

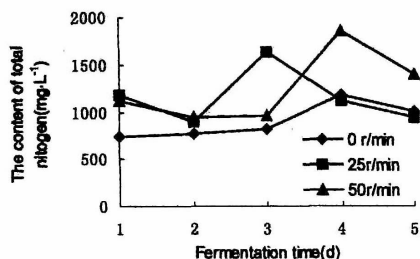


Figure 3: Changes of the content of total nitrogen with different rate of agitation

four days of fermentation was only 22.3mg/L. With agitation, the change of content of total nitrogen was higher. However, the increase of the content of total nitrogen with 50r/m rotation of agitation was not more dominant than that of 25r/m. It illustrated that it was not that the higher the rotation, the more rapid the increase of the content of total nitrogen.

3.4 Effect of Inoculation Volumes

Changes of the content of total nitrogen in anaerobic fermentation slurry with fermentation time at different inoculation volumes could be seen from Table 1 and Figure 4. The other experimental conditions were controlled as follows: fermentation temperature 30°C, material concentration 3.7%, rate of agitation 25 r/m and pH value 7.

When the factor of inoculation volume was studied, the two experimental operating modes that inoculation volume was 10% and 30% were chosen respectively. The difference of value was double in order to see the difference of the content of total nitrogen in anaerobic fermentation slurry with different inoculation volume easily. On the 2nd day of fermentation, the content of total nitrogen with 30% inoculation volume decreased 288.12mg/L and the content of total nitrogen with 10% inoculation only decreased 23.52mg/L; On the 3rd day, the content of total nitrogen with 30% inoculation volume increased quickly, the rate of change was obviously higher than that of 10%. The rate of change in the following several days approached, and the change trend of two curves was approximately same. But the change rate of the content of total nitrogen measured in low inoculation volume is lower than that of high inoculation volume, it could be conjectured that

increasing inoculation volume could increase the change gradient of the content of total nitrogen in anaerobic fermentation slurry.

Inocula have plentiful superior bacteria that were in favor of fermentation, these bacteria grow and reproduce rapidly in fitting circumstance. The more inocula, the more the quantity of superior bacteria fed, the more rapid their enrichment in fermentation tank. All kinds of bacteria participate in complicated biochemical reaction of nitrogenous matter in anaerobic circumstance, decompose organic nitrogen, consume nitrogen in a measure and produce some nitrogenous derivative of complicated structure. The more the kinds of bacteria and the larger the quantity of bacteria, the more frequent and complicated the reaction. So, inoculation volume could bring obvious change of the content of total nitrogen in anaerobic fermentation.

3.5 Effect of pH Values

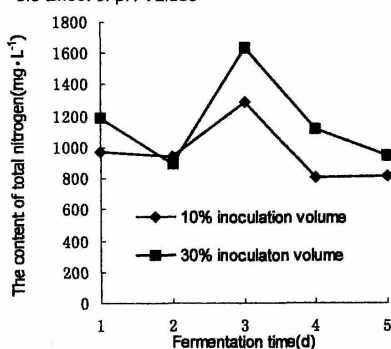


Figure4: Changes of the content of total nitrogen with different inoculation volume

Changes of the content of total nitrogen in anaerobic fermentation slurry with fermentation time at different pH values could be seen from Table 1 and Figure 5. The other experimental conditions were controlled as follows: fermentation temperature 30°C, material concentration 3.7%, rate of agitation 25r/m and inoculation volume 30%.

The three curves of figure 5 showed the experimental results of pH=5, pH=7 and pH=8.5 respectively. At pH5, the content of total nitrogen in anaerobic fermentation slurry increased slowly, the change curve approached baseline and the average change gradient was about 100 mg/L a day, but the content in the 1st day decreased. The curve of pH8.5 showed that the change gradient of the content of total nitrogen in anaerobic fermentation slurry was not high and kept about 1000mg/L. As to the experimental operating mode of pH7, the content of total nitrogen in anaerobic slurry changed obviously and the difference of change rate was large. pH=5 and pH=8.5 did not accord with the general anaerobic fermentation conditions of producing biogas, methane bacteria could not exist normally on this condition, it

was all kinds of hydrolytic bacteria that could grow and metabolize normally in digester, anaerobic fermentation could not run normally because of being restrained and from the experimental results we could see that the content of total nitrogen changed slowly here. As to pH=7, fermentation slurry showed neutrality, it could be observed from experiments that anaerobic fermentation ran normally and the change gradient of the content of total nitrogen measured was high.

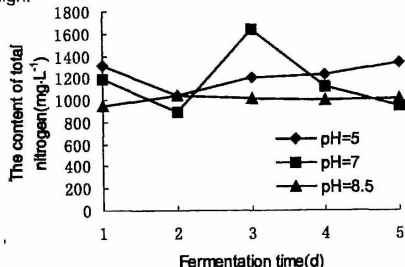


Figure 5: Changes of the content of total nitrogen with different value

4. CONCLUSIONS

The experimental results showed that:

- 1) The content of total nitrogen in anaerobic fermentation slurry produced from pig dejecta was affected by fermentation conditions such as temperature, concentration, rate of agitation, inoculation volume and pH value in different degree.
- 2) In fermentation of low temperature, the content of total nitrogen in anaerobic fermentation slurry changed slowly. In fermentation of medium and high temperature, the content of ammonia nitrogen in anaerobic fermentation slurry changed obviously, and the apex of 55°C curve was higher than that of 30°C.
- 3) The effect of material concentration on the content of total nitrogen was more obvious. When material concentration was higher, the change of the content of total nitrogen was most notable. The content of total nitrogen changed low when material concentration was low.
- 4) Without agitation, the content of total nitrogen changed low. With agitation, the change gradient of the content of total nitrogen was higher.
- 5) The change rate of total nitrogen measured in low inoculation volume always lower than that of high inoculation volume. Increase of inoculation volume might increase the change gradient of the content of total nitrogen.
- 6) The change gradient of the content of total nitrogen was higher when pH value was 7 and the content of total nitrogen changed slowly at pH5 and pH 8.5.

7) The mechanism of metabolic process of nitrogen in anaerobic fermentation needs further study.

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