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Screening of Fermentation Starters for Organic Fertilizer Composted from Chicken Manure

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Abstract In order to find fermentation starters that can promote rapid and effective composting of chicken manure, shorten the fermentation period, and improve the compost quality, fermentation starters with different microbial combination ratios (C1, C2, C3, C4 and C5) were designed and fermentation experiment was carried out on these five fermentation starters, commercially available organic fertilizer starter (SS), and chicken manure without inoculating fermentation starter. In the process of the fermentation, the changes in the fermentation temperature, water content and pH value were monitored; the effects of organic manure on the germination rate and growth of rapeseed seeds were measured; viable count, water content, and pH value of prepared fermentation starters were measured; main indicators (organic matter, nitrogen, phosphorus, and potassium) of organic fertilizer generated from fermentation were measured. From the indicators, the formula C3 and C4 were better, in the beginning of fermentation, the temperature rose rapidly, and it reached 40 ℃ in the second day; the water content dropped to below 35% in the 18th day; the pH value underwent the process of declining-rising-stable, and the overall value showed slight rise, but the change was not great (0 – 0.5). C3 and C4 fermentation products had better effects on the germination and growth of rapeseeds. The germination rate of the rapeseed seeds was 100% and 97.5% and the bud length was 15.94 mm and 14.57 mm, respectively, and the root length was 45.97 mm and 39.44 mm, respectively. The content of organic matter in fermented organic manure was 86.62% and 85.17% respectively, and the total content of nitrogen, phosphorus and potassium was 8.85% and 8.22% respectively, and the pH was 7.5 and 8.0 respectively. All of these complied with the industry standard NY525 – 2012 (organic matter ⩾ 45%, and nitrogen, phosphorus, and potassium ⩾ 5%, pH 5.5 – 8.5). The fermentation of fresh chicken manure was completed within 20 days, conforming to the requirements of large-scale organic fertilizer production factory for composting chicken manure into organic fertilizer.

Key words Chicken manure, Compost, Organic fertilizer, Fermentation starter, Screening

1 Introduction

At the same time of providing much meat, eggs and products for people, large-scale poultry farming also brings the bad smell of waste, chicken manure. It is possible to use poultry manure to produce biogas as biological energy. This has been studied extensively[1,2]. Poultry manure also can be used as feed for pigs[3], cattle and sheep[4]. However, modern agriculture, especially green and organic agriculture, needs much non-polluted and composted organic fertilizer, to improve the soil and provide safe and effective fertilizer for crops. Therefore, composting the chicken manure is of great significance for turning it into fertilizer conducive to agricultural production, particularly for production of directly edible cash crops. According to extensive studies, the composted organic fertilizer can significantly change the form of lead (Pb) in polluted soil and reduce the bioactivity of Pb. The function of reducing the toxic effects in Pb heavy metal polluted soil is chicken manure > cow dung > pig manure[5]. Therefore, chicken manure organic fertilizer is an effective modifier for heavy metal contaminated soil[5]. Although there have been extensive studies on fermentation of chicken manure[6,7]. In large-scale production, the fermentation starter composted from chicken manure can increase the temperature rise of fermentation at the early stage of composting. In the process of fermentation, it is favorable for rapid decomposing and killing pathogens and parasitic ovum in manure, and removing residual toxic substances harmful to growth of plants. At present, many small and medium-sized livestock and poultry farms directly dry manure and sell dried manure. On the one hand, the stench in the drying process exerts a great impact on the environment; on the other hand, the loss of nutrients in chicken manure can not turn them into crops can provide nutrients for the crops. Large farms need organic fertilizer fermentation factories to solve livestock and poultry manure problems. However, from the production line, we know that most organic fertilizer fermentation starters have a long fermentation cycle, the fermentation takes about 40 days[7-9]. Factors, such as site, production cycle, and season, bring certain difficulty for organic fertilizer production factors to achieve the desired production targets. Most organic fertilizer producers hope to shorten the fermentation cycle to about 20 days, to reduce the area, shorten the production cycle and increase the production. In fact, manure of different livestock contains different ingredients, and different organic waste residues contain different ingredients, it is necessary to properly design

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types and compatibility of microorganisms in the fermentation starter. For this, we carried out the screening experiment for the organic fermentation starter composted from chicken manure.

2 Materials and methods

2.1 Materials We used the chicken manure composted fermentation starter developed jointly by the Microorganism Application Research Office of School of Life Sciences in Zhengzhou University and Zhengzhou Huantong Biotechnology Co., Ltd. We prepared the fermentation starter with different bacteria (multiple of mould, yeast, bacteria and actinomyces) in accordance with characteristics of chicken manure, the number was C1, C2, C3, C4, and C5, and detected the viable count, pH value, and water content as per the national standard GB 20287 – 2006. The results conformed to requirements of solid microbial agents (as listed in Table 1). Besides, we took the commercially available fermentation starter (marked as SS) as the control group.

Table 1 Quality indicators of fermentation starter composted from different bacteria compatibility of chicken manure

Name	C1	C2	C3	C4	C5	National standard[a]
Mould // cfu/g	4.03×10^{10}	1.51×10^{10}	1.01×10^{10}	6.03×10^{10}	1.05×10^{10}	$\geq 1\times10^{7}$
Bacteria // cfu/g	2.95×10^{10}	1.01×10^{10}	2.43×10^{10}	1.10×10^{9}	1.92×10^{10}	$\geq 1\times 107$
Actinomyces // cfu/g	4.48×10^{8}	1.60×10^{10}	1.90×10^{8}	1.00×10^{8}	1.01×10^{8}	$\geq 1\times10^{7}$
Total // cfu/g	7.02×10^{10}	4.12×10^{10}	3.46×10^{10}	6.15×10^{10}	2.98×10^{10}	$\geq 2.0\times10^{9}$
pH	7.0	6.5	7.5	7.5	6.5	5.5 ~ 8.5
Water content // %	16.40	12.50	18.30	13.65	17.70	≤20

Note: [a] refers to *Microbial Inoculants in Agriculture* (GB 20287 – 2006).

The raw material for fermentation was fresh chicken manure, collected from a large chicken factory in Kaiyang City of Guizhou. It had water content of 78% and pH of 7.5. It was fermented in an organic fertilizer factory near the chicken farm. Auxiliary materials were barley bran, purchased from the local market.

2.2 Methods

2.2.1 Treatment of fermentation compost. In this experiment, we designed 7 treatments: C1, C2, C3, C4 and C5 were fermentation starters composted from different microbial strains, SS was commercially available fermentation starter, and CK was the control group without fermentation starter. We carried out the chicken manure fermentation experiment for these treatments. We repeated each treatment three times, the experiment lasted 18 days. We mixed 5000 kg chicken manure for each treatment, added 800 kg fresh rice bran, and inoculated 0.5 kg microbial inoculum, composted and fermented. The compost was 1.2 m high and 1.5 m wide in the base.

2.2.2 Measurement of indicators. We measured the temperature value of fermentation compost of each treatment at about 9:00 every day, measured the pH value every other day using precision test paper pH 5.5 – 9.0 (Shanghai Xinsheng Chemical Science Co., Ltd.), measured the water content every 3 days, measured the effects of fermented products on the germination rate and growth of rapeseed on the 19th day, measured the organic matter, nitrogen, phosphorus and potassium in the products, by the method specified in NY525 – 2012.

2.2.3 Effects on the germination rate of rapeseeds. We weighed 5 g of fermented chicken manure organic fertilizer, extracted for 1 h with deionized water, then filtered with 3 layers of gauze, took 5 mL of the filtrate, add to the culture dish, put 2 filter papers into the culture dish, and placed 20 rapeseed seeds in the middle of two layers of filter paper, and repeated 3 times per treatment. At the room temperature (32 ℃), we cultured 48 h, counted the germination rate and the length of buds and roots. According to these, we analyzed the effects of the fermented organic manure on the germination rate and growth of rapeseed, and the clear water treatment group was the blank control. *Germination rate* (%) = (*number of germinated seeds / total number of tested seeds*) × 100.

2.2.4 Measurement of main indicators of fermentation starters generated from different compatibility. Calculation of viable count: we thinned the fermentation starters generated from different compatibility by times and spread evenly onto the plate PDA (for fungi viable count), LB (for bacterial viable count) and Gaoshi's number 1 (for viable count of actinomyces). After certain time of growth at corresponding temperature, we separately counted strains of the fungi, bacteria, and actinomyces. We measured the pH value of the fermentation starter using the precision test paper pH5.5 – 9.0, and measured the water content using the drying method.

2.3 Data processing We processed data and plotted charts by manual calculation and computer software Excel.

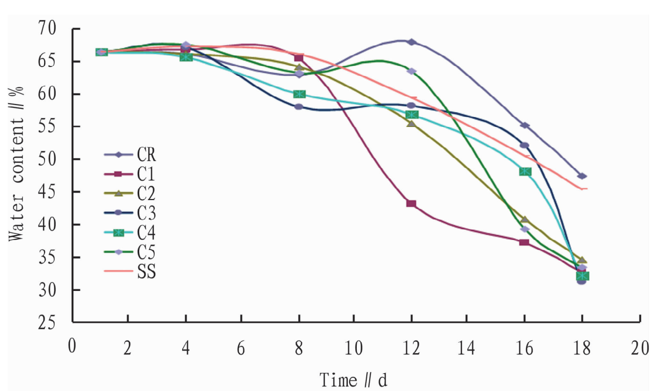


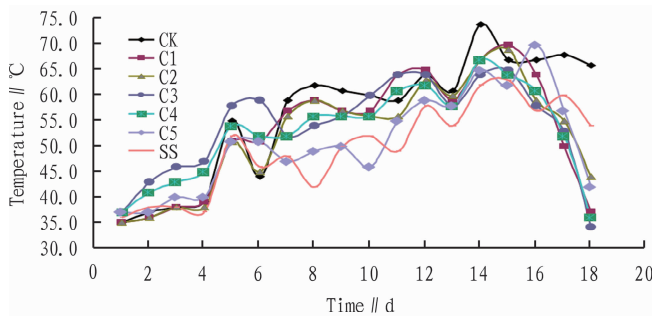
Fig. 1 Changes in water content in the fermentation process of chicken manure

3 Results and analyses

3.1 Changes in water content After inoculating different microbial fermentation starters in the chicken manure, the changes in water content in the fermentation process are shown in Fig. 1. From the fermentation process, the water content of each treatment was declining. After 18 days of fermentation, the water content of SS and CK was 44.98% and 46.96%, respectively, and the water content of other treatments was below 35%. After the fermentation, the water content of C3, C4, C1, C5 and C2 was 31.19%, 32.09%, 32.58%, 33.38% and 34.43% respectively, conforming to the requirements of chicken manure organic fertilizer producers. And after a period of composting, the water content could be dropped below 30%, meeting the requirements of industrial standard.

3.2 Changes in temperature In the fermentation process of chicken manure, the fermentation starters C1, C2, C3, C4, C5, SS and CK, on the second day of inoculation, the treatments C3 and C4 rose to 40 ℃ above, C5 rose to 40 ℃ on the third day; other treatments reached the 50 ℃ on the fifth day and consistent with C5, but still lower than C3 and C4. On the 18th day of fermentation, the temperature of the treatments was 35 ℃ – 45 ℃, and the fermentation was basically completed (Fig. 2). According to the speed of temperature rise after the inoculation and the temperature rise on the 18th day of fermentation, high temperature retention, and temperature drop, treatments C3 and C4 were better.

3.3 Changes in the pH In the fermentation process, the pH of each treatment underwent the process of falling-rising-stable, which was slightly higher than that of the initial pH, with a variation range of 0 to 0.5. After completion of fermentation, the pH of each treatment was 7.5 – 8.0, which complied with the national standard NY525 – 2012 of the People's Republic of China. However, the pH of C3 fluctuated in the fermentation, but it returned to the initial state at the end of the fermentation (Table 2).



Note: The temperature fluctuation curve is due to the temperature drop after turning the compost.

Fig. 2 Changes in temperature in the fermentation process of chicken manure

Table 2 Changes in the pH in the fermentation process of chicken manure

Time // d	CK	C1	C2	C3	C4	C5	C6	SS
1	7.5	7.5	7.5	7.5	7.5	7.5	7.5	7.5
3	7.0	6.5	6.5	6.5	6.5	6.5	7.0	7.0
5	6.5	7.0	6.5	6.5	6.5	6.5	7.0	7.0
7	6.5	7.5	7.5	7.0	7.5	7.5	7.5	7.5
9	7.0	7.0	7.0	7.0	7.0	6.5	7.0	6.5
11	7.0	7.0	7.5	7.5	7.5	7.0	7.5	6.5
13	7.0	7.5	7.5	7.0	7.0	7.5	7.5	7.0
15	7.5	8.0	7.5	8.0	8.0	7.5	8.0	8.0
17	7.5	8.0	7.5	7.5	7.5	7.5	8.0	8.0
19	8.0	8.0	8.0	7.5	8.0	8.0	8.0	8.0

3.4 Effects of fermented compost on germination rate and growth of rapeseeds The germination test of rapeseeds showed that the fermentation starters with different compatibility had little effect on the germination rate of rapeseeds ($P < 0.05$), and even for the fermented compost without fermentation starter (*CK*), although the fermentation was not completely finished, with fermentation of 18 days, the germination rate of rapeseeds reached 92.50%. The buds and roots with addition of organic fertilizer filtrate were significantly better than that of the clear water control (CK_W), indicating that a period of fermentation of chicken manure had little effect on the germination rate of rapeseeds, but promoted growth of roots and buds. According to the germination rate, average bud length and root length of rapeseeds, treatments C2, C3, C4, and C5 were better, the germination rate was 100%, 100%, 97.50% and 95%, respectively, and average bud length and average root length remained a high level, as listed in Table 3.

Table 3 Effect of fermentation starters with compatibility of different strains on the germination rate and growth of rapeseeds

Treatment	CK_W^*	*CK*	C1	C2	C3	C4	C5	SS
Germination rate // %	100.00	92.50	95.00	100.00	100.00	97.50	92.50	95.00
Average bud length // mm	5.34	10.48	9.74	12.11	15.94	14.57	10.62	14.18
Average root length // mm	16.21	31.33	26.23	34.65	45.97	39.44	30.35	39.03

Note: * CK_W is clear water control group.

3.5 Measurement of main indicators of organic fertilizer after fermentation In the fermentation process of composting chicken manure into organic fertilizer, we visually observed the temperature and water content, measured the organic matter content and nitrogen, phosphorus, potassium and pH of organic fertilizer fermented by C3 and C4 treatments, by the method specified in national standard NY525 – 2012 (industry standard for short). The results are listed in Table 5. From Table 5, we can see that the organic matter content of each treatment was above 80%, complying with the requirements of being equal to or greater than 45% in the industry standard; nitrogen, phosphorus, and potassium were greater than 8% (8.85% and 8.22, respectively), also meeting the provisions of the industry standard (equal to or greater than 5%); the pH of two kinds of fermentation starters after fermentation was 7.5 and 8.0, which was in the range of 5.5 – 8.5, also conforming to the provision of the industry standard.

Table 4 Measurement of indicators of organic fertilizer fermented with different compatibility of strains

Treatment	C3	C4	Industry standard[a]
Organic matter//%	86.62	85.17	≥45
Nitrogen//%	4.01	3.73	
Phosphorus//%	1.95	1.63	
Potassium//%	2.99	2.86	
Nitrogen, phosphorus, and potassium//%	8.85	8.22	≥5
pH	7.50	8.00	5.5 ~ 8.5

Note: the industry standard refers to NY525 – 2012.

4 Conclusions and discussions

According to the experiment results, the treatments C3 and C4 have better fermentation, and their composite indicators are better than commercially available fermentation starter (SS). Therefore, C3 and C4 can be used in turning chicken manure into organic fertilizer. Modern chicken farms use water to rinse the poultry manure, dehydrate and ferment the organic fertilizer, firstly dry for a period of time, then use it for fermentation of organic fertilizer. In the fermentation process, the water content is higher, generally 78% – 81%. Although when using some commercially available fermentation starters, we controlled the water content of chicken manure in strict accordance with the instructions, it is rarely to complete the fermentation in a short period. This study is based on the requirements of chicken manure organic fertilizer producers, that is, the fermentation cycle is shortened to 20 days, and the water content at the end of fermentation is 45% or less. And then after a period of composting, it is able to produce organic fertilizer complying with the industry standard. Focusing on these targets, combining with raw materials provided by chicken manure organic fertilizer production factories, we designed the strain compatibility. After 18 days of fermentation, we screened C3 and C4 formula, the characteristics are rapid temperature rise, long temperature holding, and rapid decomposition. At the early stage of composting, the temperature rise is rapid, it can hold 45 ~ 67 ℃ for 15 d, favorable for killing pathogens and parasitic ovum in manure, and removing residual toxic substances harmful to growth of plant, to realize harmless, composting, and balanced nutrients. The content of nitrogen, phosphorus and potassium was 8.85% and 8.22%, respectively; the content of organic matter was 86.62% and 85.17%, respectively; the content of nitrogen, phosphorus and potassium was 8.85% and 8.22% respectively, and the germination rate was 100% and 97.50% separately. Besides, the two kinds of fermentation starters complied with national standard (viable count: 3.46×10^{10} cfu/g, 6.15×10^{10} cfu/g, water content 18.30% and 13.65%, and the pH was 7.5). In the fermentation process of chicken manure, the inoculation amount was only 0.01%, far lower than other similar products[6–10], which reduce the fermentation cost. In conclusion, the comprehensive evaluation of indicators of C3 and C4 fermentation starter are better and can be used for organic fertilizer fermentation of fresh chicken manure with high water content. Besides, the pH value of chicken manure after adding barley bran was 7.5, while the pH of fermented C3 and C4 was 7.5 and 8.0 respectively. In the process of fermentation, the pH fluctuated in the range of 7.5 – 8.0, first falling then rising. The trend of change is consistent with results of other researchers[7], but there is also opposite case[8,9]. Some scholars obtained the pH value greater than 9, 8.3 – 9.4[12], 8.0 – 9.0[8], and 7.5 – 9.0[5], indicating that the type of fermentation starter is related to the pH of the fermentation products. To control the pH of the compost after fermentation, attention should be paid to the selection of fermentation starters. In this experiment, the pH of the compost after fermentation of chicken manure first fell then rose or remained stable and the range of variation was 0 – 0.5, and the pH of fermented bean dung was declining within the amplitude of 0.5 – 1.0[7]. Therefore, different fermentation starters and different raw materials of compost, the pH value of the compost after fermentation is different, in practical application, it is required to carefully select fermentation starters to facilitate the fermentation of organic fertilizer to improve the growth and quality of selected crops.

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6 Agricultural sacrificial activities of Zhuang People

Zhuang people mainly live in Guangxi. Spirits they believe are various. In agricultural production, they worship many gods. Land god is the master of an area and controls the flood, drought, and livestock pestilence of the area. On festivals or in the event of serious disaster, the villagers must go to the land temple to pray. Before praying, they should not eat dog meat. Every year, the whole village will hold a ceremony of small sacrifice offering; every three years, they will hold a ceremony of grand sacrifice offering. In the beginning of every spring, Zhuang people will pray to ask the land god to protect good weather, bumper harvest, and peace and safety of men and livecstock. In autumn, they will redeem their vow, to express gratitude to the land god. The land god is the patron saint of villages. In the event of the pest disaster, the villages will hammer at the gong to gather villagers, kill pigs, sheep, and chicken as sacrifice to make public offering, and use animal blood to stain paper, insert in the field, to signify that the land god has arrived there, then the disasters can be avoided. On June 6 of the lunar calendar, every family kills chicken to worship the land god and drink wine and sing songs, and insert small paper flags, to pray for bumper harvest. In the event of pest disaster, they play dragon game to dispel pests. At the time of sowing, they sprinkle few rice seeds or peanut shells along village roads, to pray for even growth of seedlings. In July, some villages offer sacrifices to seedlings, offer pork and wine as sacrifice, and both the young and old villagers drink wine together. In autumn harvest, Zhuang people hold a ceremony of eating new rice. They should not harvest rice until they offer sacrifices. According to legend, the cattle are dispatched by the heaven to help farmers. April 5 of the lunar calendar is called the birthday of cattle king. On this day, it is forbidden to use cattle. Instead, they offer colored glutinous rice to feed cattle. After busy spring plough, they hold a ceremony of collecting the soul of cattle, called the festival of cattle soul. On this day, they put chicken, fish, meat, and glutinous rice into the cattle pen, and use such rice and meat to feed cattle.

7 Agricultural sacrificial activities of Yi People and Tujia People

The Yi people are distributed in Sichuan, Yunnan, Guizhou and Guangxi. They believe that the gods make all things and dominate everything; the land is the source of food and clothing, so every year they will choose auspicious day to worship the gods; and the water gods dominate the fields and protect the fields from floods and droughts. They offer sacrifices to the fire god to pray for bumper harvest. Qiang people live in the upper reaches of Min River. They worship various gods. Offering sacrifice to the heaven is the grandest activity, it is generally held in March or May of the lunar calendar. In case of drought, they hold a ceremony for praying rain. Before praying, everyone is not allowed to cut firewood, dig herbs or hunt, called " forbidden mountain" , if violated, patrol villagers will beat them to bleed to please the gods. After the forbidding the mountain, the wizard wields the magical power to pray rain, if not effective, the wizard leads villagers to the top of the " holy mountain" to shout the rain. Tujia people mainly live in boundary area of Hunan, Hubei, Sichuan, and Guizhou. They believe that all things in the world are dominated by the gods, and thus they worship a large number of gods. In agriculture, they worship the land god. In the Spring Festival, they slaughter pigs and take pig hoof as sacrifice of land god. On February 2 of lunar calendar, they kill chicken to offer sacrifice to the land god. Li people live in Hainan Island and believe that harvest of crops is the charity of land ghost. They believe all things have spirits and in some places, there are still relics of totem adoration; they all respect seniors and ancestors, and during festivals, they will go to them to pray for peace and harvest. Owing to the spread of Christianity, some of Li have taken to this religion. Before transplanting rice seedlings, they firstly transplant several rice seedlings to play the rice soul; when the rice becomes ripe, they should offer several rice dumplings to express gratitude to the land ghost. And the wife should go to the field to greet the land ghost.

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