Optimum Dosage of Hyper-Thermophilic Aerobic Compost (HTAC) Produced from Sewage Sludge for Rice Yield

Yanfei Cheng¹, Ryuhei Inamori², Kakeru Ruike^{1,2}, Yuhei Inamori² & Zhenya Zhang¹

¹ Graduate School of Life and Environmental Sciences, University of Tsukuba, 1-1-1 Tennodai, Tsukuba, Ibaraki 305-8572, Japan

² Foundation for Advancement of International Science, 3 Chome-24-16 Kasuga, Tsukuba, Ibaraki 305-0821, Japan

Correspondence: Zhenya Zhang, Graduate School of Life and Environmental Sciences, University of Tsukuba, 1-1-1 Tennodai, Tsukuba, Ibaraki 305-8572, Japan. E-mail: zhang.zhenya.fu@u.tsukuba.ac.jp

Yuhei Inamori, Foundation for Advancement of International Science, 3 Chome-24-16 Kasuga, Tsukuba, Ibaraki 305-0821, Japan. E-mail: y_inamori@fais.or.jp

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Abstract

Hyper-thermophilic bacteria can shorten the duration of composting process. This study aimed to elucidate the applicability and the optimum fertilization amount of hyper-thermophilic aerobic compost (HTAC) for rice growth and yield. The stem height, tillers number and rice yields in the treatments with HTAC significantly increased with increasing fertilization amount. But the ripening rate of grain was found to be lower at higher fertilization amount (250 and 500 kg N/ha). The rice yield in 180 kg N/ha HTAC treatment was 5.38 t/ha, which is similar with that using recommended amount of chemical fertilizer. The N and P concentrations in surface water of 180 kg N/ha HTAC treatment before midsummer drainage were lower than 1 mg/L and 0.1 mg/L, reaching the environmental quality standard. Thus, the optimal fertilization amount of HTAC was deemed as 180 kg N/ha when considering the plant quality and environmental friendliness.

Keywords: Hyper-thermophilic aerobic compost, Rice growth, Yield components, Environmental quality standard

1. Introduction

Cereal accounts for about 80% of the world's food supply (Pimental and Wilson, 2004), in which the rice provides 20% of world dietary energy supply (FAO, 2004). With the rapid increase of world population, the quantity of food produced per capita has been declining since 1984 based on available cereal grains (Pimental & Wilson, 2004). Though the expansion of arable land can remit the eager increasing food demand, fertilizer still is the predominant factor with regard to the increased grain yield. It was reported that cereals account for around 60% of global fertilizer use (FAO, 2012). However, the increased application of chemical fertilizer resulted in serious soil deterioration. The structure, exchangeable cations capacity (especially calcium and magnesium) and pH of soil can be significantly affected by application of chemical fertilizers for months and years (Bernal et al., 2009). Cai et al. (2015) reported that based on the 18-year fertilization treatments, urea as a chemical N fertilizer in an intensive farming system has significantly reduced soil pH and is confirmed as the major cause of intensified acidification of the red soil in southern China. To solve these problems, the use of organic fertilizer has been paid more attention since it can improve the structure of the soil and increase its ability to hold water and nutrients (Tester et al., 1990; Creccio et al., 2001). In addition, organic fertilizer is generally made from plant residue or solid wastes by composting, which makes it renewable and environmental friendly.

Composting is a spontaneous biological decomposition process involving mineralization and humification of organic materials in a predominantly aerobic environment. During the process, bacteria, fungi and other microorganisms, including microarthropods, can break down the macromolecular organic materials into usable nutrients and stable organic substances called as compost (Nasini et al., 2016), which is also free of pathogens and seeds of weeds (Eghball et al., 2000). However, the production of complete compost generally need at least 3 months by conventional composting process (Ros et al., 2003). The long processing duration results in low

treatment efficiency of organic wastes and high cost for fertilizer production. Thus, the development of new technologies to accelerate the composting process is indispensable to meet the treatment demand for increased amount of organic wastes. Hyper-thermophilic aerobic composting is one of the innovative methods that has been applied for small local communities in Japan. The bacteria which can survive at high temperature of 80-100° C, called as *hyperthermophile*, are used during hyper-thermophilic aerobic composting. *Hyperthermophile* usually can be separated from extreme environment, such as hot spring, hot-water deposit in ocean, or oil deposit from underground. The metabolic heat release that occurs during bacterial fermentation raises the processing temperature sufficiently high at 80–100°C (Kanazawa et al., 2008). Subsequently, the composting period can be shortened to 45 days due to the high organic decomposition rate of *hyperthermophile* at high temperature. Also, a hygienic compost with better quality can be produced due to the high temperature can sterilize the pathogens. The odorous components other than ammonia also disappear in the early stage of fermentation process. However, compared with chemical fertilizer, compost contains not only mineralized nutrients, but also organic nutrients that can be slowly decomposed to release nutrients for plant growth. Thus, it is difficult to determine the fertilization amount of compost. Overdosing would induce environmental pollution and long-lived phenomena in rice crops, and infect the rice yield.

In this study, a hyper-thermophilic compost produced in Kagoshima, Japan was applied to evaluate the effects on rice growth and environmental burden derived from rice paddy drainage. The hyper-thermophilic aerobic compost was named as HTAC. The rice growth experiment was carried out in 2016. A rice cultivar called as *Koshihikari* was chose for plant growth experiment. This study aimed to elucidate the optimum application amount of HTAC for rice growth.

2. Materials and Methods

2.1. Fertilizer

HTAC used in this research was provided by Kyowa Kako Co., Ltd. It was produced by composting sewage sludge inoculated with *C. yamamurae*, which was isolated from Kirishima hot spring in Kagoshima prefecture, Japan. The sewage sludge was collected from Kagoshima wastewater treatment plant (Moriya et al., 2011). The main characteristics of HTAC are shown in Table 1. The chemical fertilizer used in this study was manufactured by Showa Sangyo Co., Ltd.

	TS	TN	NH4 ⁺ -N	TP	C/N	TK	Cu	Zn	CaO
	(%)	(mg/kg)	(mg/kg)	(mg/kg)		(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)
HTAC	73.5	28000	20000	38000	6	8100	250	470	70000

Table 1. Characteristics of HTAC

*TS, total solids content; TN, total nitrogen content; TP, total phosphorus content; TK, total potassium.

**The data were measured by Syonan analysis center Co., Ltd.

2.2. Study Site and Rice Growth Experiment

The rice growth experiment was performed on a farmland in 2016 in Tsukuba, Ibaraki, Japan (36°05'N,140°04'). This region has a northern temperate marine climate with an average annual temperature of 13.8°C and a mean annual precipitation of 1283 mm.

Square containers (0.8 m in length \times 0.5 m in width \times 0.5 m in height) equipped with water inlet and outlet were used as rice growth device to evaluate the different fertilization conditions. In each container, commercial pumice and river sand were firstly filled for 5 cm orderly from the bottom. Then, the well mixed paddy soil was filled on top for 30 cm. A specific amount of HTAC was applied in each container and mixed well with the paddy soil. The experiment without fertilizer addition was used as control. After irrigating, 8 rice plants (*Oryza sativa cultivar Koshihikari*, seeds were purchased from Japan Agricultural Cooperatives) were transplanted in each device with the interval of 20 cm. The devices were installed outdoors. The rice growth period was from May 5th to September 12th in 2016. The fertilization conditions are shown in Table 2. Mid-summer drainage was conducted between day 44 and 63 during rice growth experiment as it was proven that summer drainage could increase rice growth and yield (Hwang & Chung, 2013).

Treatment	Conditions
R1 NONE	Without fertilizer
R2 Che.60	With 60 kg N ha ⁻¹ of chemical fertilizer
R3 HTAC 60	With 60 kg N ha ⁻¹ of HTAC
R4 HTAC 180	With 180 kg N ha ⁻¹ of HTAC
R5 HTAC 250	With 250 kg N ha ⁻¹ of HTAC
R6 HTAC 500	With 500 kg N ha ⁻¹ of HTAC

Table 2. The fertilization conditions for rice growth experiment in 2016.

2.3 Calculation of Rice Yield and Yield Components

The plant height was measured by the length of rice plant from ground to the longest leaf. Tillers per plant was recorded by the total number of stem that grow after the initial parent stems.

After rice harvest, the rice plants were air-dried for two weeks. The numbers of panicle and spikelet were counted. The ripened grains were obtained by salt water selection method using NaCl solution with specific gravity of 1.06. The ripened grains were then cleaned by dilute water and air dried for 24 h. The outer hull of ripened grains was removed by hulling machine (TR-130, KETT electric laboratory). The weight of rice per plant after removing the hull was called as brown rice weight (g plant⁻¹). The ripening rate of grain, 1000-grain weight and rice yield were calculated by the following equations.

Ripening rate of grain (%)=
$$\frac{\text{number of ripened grains}}{\text{spikelets per plant}} \times 100$$
 (1)

1000-grain weight (g)=
$$\frac{\text{brown rice weight}}{\text{spikelets per plant × ripening rate of grain}} \times 1000$$
 (2)

Rice yield (t ha⁻¹) =
$$\frac{\text{brown rice weight } \times 8 \text{ (plant number per device)}}{0.4 \text{ m}^2 \text{ (cross sectional area of the device)}} \times 10000$$
 (3)

2.4 Determination of N and P in Surface Water and Soil

Surface water samples were collected from each device every week. The water samples were filtered through fiber filter with pore size of 1.2 μ m. Then, the concentration of total nitrogen (TN), NH₄⁺-N, NO₃⁻-N, and total phosphorus (TP) in all the water samples were determined. Total nitrogen (TN) was determined by alkaline potassium persulfate digestion and UV spectrophotometric method (APHA, 2012). NH₄⁺-N was measured by Indophenol-blue method (Ivancic and Degobbis, 1984). NO₃⁻-N was measured by UV spectrophotometer method at 220 and 275 nm (APHA, 2012). TP was determined by a molybdenum blue method mentioned by Murphy and Riley (1962).

In each device, soil was randomly sampled from five spots in the depth of 0-20 cm. Wet soil samples were air dried, grounded and passed through 2 mm sieve. NH_4^+ -N in soil was measured by the indophenol blue method by Hall (1993). PO_4^{3-} -P in soil was determined by the molybdenum blue colorimetric method after extraction by 0.5 M NaHCO₃ (Olsen et al., 1954).

2.5 Statistics

Analysis of variance was conducted to detect differences in plant height, rice yield, and yield components. Data were analyzed in randomized block design. The least significant difference (Duncan method) at p < 0.05 was applied to compare the means for significant differences between variety and cropping season.

3. Results

3.1 Characteristics of Rice Growth

3.1.1 Plant Height

The variation of plant height over time are shown in Figure 1a. Compared with R2 (Che. 60), R4 (HTAC 180) showed a similar level of plant height, while the higher plant height was achieved in R5 (HTAC 250) and R6 (HTAC 500). The highest plant height reached to 105.3 cm in R6 (HTAC 500) after 91 days' cultivation and began to flatten after that. As shown in Figure 1b, on day 115, plant height was found to be positively correlated with the fertilization amount of HTAC. The relationship between plant height and HTAC fertilization amount fitted the following quadratic equations: $y = 0.0343x+90.604(R^2=0.979; x, HTAC amount; y, plant height)$.

The average height in panicle formation stage and maturity stage are shown in Table 3. The plant height increased with increasing N fertilization amount in both stage in R3-R6. And R4 (HTAC 180) showed similar plant height with R2 (Che. 60).

	average height (cm)				
	Panicle formation stage	Maturity stage			
R1 NONE	59.1±3.2 ª	89.7±3.5 ª			
R2 Che.60	66.9±2.4 ^b	96.8±2.7 ^b			
R3 HTAC 60	58.9±1.6 ª	92.3±2.0 ª			
R4 HTAC 180	68.1±2.6 ^b	98.6±2.9 ^b			
R5 HTAC 250	69.8±3.3 ^b	99.1±3.1 ^b			
R6 HTAC 500	79.6±4.2°	107.1±4.5 °			

Table 3. The average plant height in panicle formation stage and maturity stage of koshihikari

Note: all the data is the average value of 8 plants.

*Data were statistically analyzed by one-way ANOVA with repeated measures and different small letters indicate significant differences (P<0.05) among four treatments of each season by Duncan's method for multiple comparisons. All the data are the means of two replicates±standard error.

3.1.2 Rice Tillers per Plant

The variation of tillers per plant are shown in Figure 2a. After transplanting, the number of stem increased with the occurrence of tiller and reached the maximum value. Then the ineffective tillers died during summer drainage, and panicle grew from the remained tillers. The tillers number in all the treatments reached the highest on day 49, which were 10, 27, 14, 24, 26, 35 in R1 to R6, respectively. Among all the treatments, R6 (HTAC 500) achieved the highest tillers number.

As seen in Figure 2b, on day 49 and day 115, tillers number were found to be positively correlated with the fertilization amount of HTAC. The relationship between tillers number and HTAC amount fitted the following quadratic equations: y = 0.0521x+11.385 (R²= 0.955; day 49; x, HTAC amount; y, tillers number), y=0.0462x+9.0525 (R²= 0.994; day 115; x, HTAC amount; y, tillers number).

3.2 Yield and Yield Components

As shown in Table 4, the rice yields in R3-R6 significantly increased with increasing fertilization amount of HTAC. The highest rice yield was 6.56 t/ha in R6 (HTAC 500). In R3-R6, the rice tillers number and spikelets number per plant also increased with the increasing fertilization amount, while the ripening rate of grain and 1000-grain weight decreased when HTAC amount higher than 250 kg N/ha.

Under these experimental conditions, the relationship between rice yield and HTAC fertilization amount fitted the following quadratic equations: $y = -1.858*10^{-5}x^2 + 0.017x + 2.667$ (R²= 0.968), and the optimal fertilization amount calculated is 457.5 kg N/ha (Figure 3a). As the rice yield of R2 (Che.60) was 5.02 t ha⁻¹, the amount of HTAC which can achieve same rice yield with R2 calculated by this modified quadratic equations is 170.0 kg N ha⁻¹.

a.



Figure 1. Variation of plant height in R1-R6 with different treatment conditions (a) and the relationship between HTAC amount and plant height on day 115 (b)



Figure 2 Variation of tillers per plant in R1-R6 with different treatment conditions (a) and the relationship between HTAC amount and tillers number on day 49 and day 115 (b)

Conditions	Tillers	Spikelets plant ⁻¹	Ripening rate of grain (%)	Brown rice Weight	1000	Rice yield
	plant ⁻¹			$(g plant^{-1})$	1000-grain weight (g)	$(t ha^{-1})$
R1 NONE	11±0.6ª	909±50 ª	77.9 ± 1.7 ^a	14.64±2.2 ª	20.7±0.2 ª	2.93±0.4 ª
R2 Che.60	$20{\pm}1.0^{b}$	1463±63 ^b	80.2±1.9 ª	25.09±3.1 ^b	21.4±0.3 ^b	$5.02{\pm}0.6^{b}$
R3 HTAC 60	11±1.1 ª	931±49ª	82.3±1.6 ^b	15.87±2.5 ª	20.7±0.1 ª	$3.17{\pm}~0.5^{\text{ a}}$
R4 HTAC 180	17±1.5 ^b	1457±74 ^b	84.7±1.4 °	26.92±2.8 ^b	21.8±0.2 ^b	5.38 ± 0.6 ^b
R5 HTAC 250	20 ± 1.2^{b}	1618±82 ^b	84.2±1.3 °	28.81±3.2 ^b	21.1±0.2 ª	5.76 ± 0.6^{b}
R6 HTAC 500	29±1.5 °	1894±96°	83.0±1.6 ^b	32.81±3.5 °	$20.9\pm\!0.3^{\rm \ a}$	6.56±0.7°

Table 4. Rice yield and yield components

Note: all the data is the average value of 8 plants.

*Data were statistically analyzed by one-way ANOVA with repeated measures and different small letters indicate significant differences (P < 0.05) among four treatments of each season by Duncan's method for multiple comparisons. All the data are the means of two replicates±standard error.



b.



Figure 3. The relationship between rice yield and HTAC amount (a) and tillers number (b)

3.3 Variation of N and P Nutrients in Water and Soil

3.3.1 Variation of N and P in Water

TN, NH_4^+ -N, NO_3^- -N, and TP concentrations in surface water in each conditions are shown in Figure 4. The TN concentration increased after transplanting and reached to the top on day 7. The highest TN happened in R2 (Che. 60, 3.89 mg/L), and followed by R6 (HTAC 500, 3.34 mg/L) and R5 (HTAC 250, 3.03 mg/L). After day 7, the TN concentration of all treatments rapidly decreased. Before midsummer drainage (day 44 to day 63), they were all lower than 2 mg/L. After midsummer drainage, the TN concentration in surface water was lower than 1 mg/L in all the treatments.

The NH₄⁺-N concentration in surface water showed the similar trends with the TN concentration. The NH₄⁺-N concentration in R6 (HTAC 500) was the highest on day 14, increasing to 0.27 mg/L. After that, the NH₄⁺-N concentration in all the treatments were maintained at concentrations that lower than 0.2 mg/L. The NO₃⁻-N only appeared in R2 (Che. 60) in the first 7 days. It was 0.68 mg/L after transplanting and then increased to 1.83 mg/L on day 7. The NO₃⁻-N concentration in other treatments were all lower than 0.05 mg/L.

The variation of TP concentration was similar with the variation of TN. The TP concentration in all treatments increased during the first 7 days after transplanting. On day 7, the highest TP concentration was 0.53 mg/L in R6 (HTAC 500). Then the TP concentrations in all treatments decreased. Before midsummer drainage, they were all lower than 0.2 mg/L.



Figure 4. Variation of TN (a), NH₄⁺-N (b), NO₃⁻-N (c), TP (d) concentrations in surface water in different treatment conditions

3.3.2 Variation of N and P in Soil

The variation of NH_4^+ -N and PO_4^{3-} -P concentrations in soil are shown in Figure 5. The NH_4^+ -N concentrations of R1 (NONE), R2 (Che. 60), R3 (HTAC 60) were lower than 50 mg/kg from initial to final. While in R4 (HTAC 180), R5 (HTAC 250), R6 (HTAC 500), the initial NH_4^+ -N concentrations were 69.7, 73.1 and 108.9 mg/kg, respectively. The NH_4^+ -N concentrations in R5 and R6 firstly decreased, and then increased to the highest (97.5 mg/g for R5, 125.9 mg/g for R6) on day 28. The PO_4^{3-} -P concentration increased with increasing fertilization amount in R3-R6. In R1-R3, the PO_4^{3-} -P concentration varied from 100 mg/kg to 200 mg/kg and almost being stable from initial to final, nevertheless, the PO_4^{3-} -P concentration in R6 was the highest (434.6 mg/kg) at initial and then varied around 400 mg/kg. It decreased to 300 mg/kg at harvest. In R5, it was varied around 300 mg/kg and decreased to 200 mg/kg at harvest.



Figure 5. Variation of NH₄⁺-N (a) and PO₄³⁻-P (b) concentrations in soil in different treatment conditions

4. Discussion

4.1 Effect on Rice Growth

In this research, the rice growth was normal after transplanting in all the treatment conditions. No death or growth defect happened in all treatments. Compost has been recognized as slow-releasing fertilizer, but the maximum tillers number happened at the same time in both treatments with chemical fertilizer and HTAC. This might due to the high ammonium concentration in HTAC that can provide sufficient nutrients for rice growth.

Rice lodging occurred on rice plant in R6 (HTAC 500) with the highest amount of HTAC. It can easily happen when the stem over grew. The normal height of *Koshihikari* is said to be less than 80 cm in panicle formation stage, and less than 100 cm in maturity stage. The height of plant is an important indicator that related with rice lodging. From Table 3, we could found that the plant in R4-R6 showed higher plant height than normal value. Basak et al. (1962) reported that high nitrogen fertilization amount might lead to rice lodging and pointed out that 100 pounds per acre (112 kgN/ha) gave optimum yield of rice and avoided rice lodging before heading. The higher fertilization amount of HTAC increased the risk of rice lodging, which might also influence the rice yield. Figure 3b shows the relationship between rice yield and tillers number. When the tillers number was lower than 22, the relationship between tillers number and rice yield fitted the following quadratic equations: y = 0.2658x+0.3913 (R²= 0.994; x, tillers number; y, rice yield). However, in R6, although the tillers number reached to 35, the rice yield did not increase significantly.

The rice yield in R6 was the highest (6.56 t/ha), which is 2.24 times that in R1 (NONE). However, the ripening rate of grain was found to be lower in R5 and R6. The higher nutrients level applied can promote plant growth, thus increasing the tillers number per plant. The more tillers can guarantee the sufficient photosynthesis for plant growth, and in turn, can potentially increase the rice yield. However, when the tillers number excessed a specific value, malnutrition would happen. The spikelets number per panicle would decrease due to the malnutrition. However, in this study, spikelets number per panicle in all treatments had no significant difference. The rice yield increased with the increase of tiller number. The NH₄⁺-N and PO₄³⁻-P concentration in soil was relatively higher in R5 (HTAC 250), R6 (HTAC 500) (Figure 5). Thus, enough nutrients were supplied to promote the spikelet growth on each panicle, though lower ripening rate of grain was observed in R5 and R6.

The rice growth in R3 (HTAC 60) was inferior compared with that in R2 (Che.60), which applied the same amount of nitrogen. While the rice growth in R4 (HTAC 180) showed a similar level with that in R2 (Che. 60). Compost contains abundant organic matters. The N form in HTAC exist in both inorganic and organic forms, while chemical fertilizer only contains inorganic N. As we known, inorganic N (NH₄⁺-N and NO₃⁻-N) can be directly absorbed by plant root. Though there are many researches demonstrated that some plants can use soluble organic N directly, the N uptake rate of organic N was lower than that of inorganic N (Franklin et al., 2017). Most of organic N need to be firstly decomposed by microorganisms in soil and then taken by plant as inorganic form. This might can explain why the rice growth was inferior when applying same amount of HTAC as chemical fertilizer. However, this does not mean the organic fertilizer is inferior to chemical one. Franklin et al. (2017) reported that nitrogen use efficiency can be significantly improved by applying organic N. Although the optimized fertilization amount was 457.5 kg N/ha by calculating from the modified quadratic equations. Considering both plant quality and rice yield, 180 kg N/ha, which gives 1.84 times (5.38 t/ha) higher rice yield compared with R0 (NONE, 2.93 t/ha), was deemed as the optimal fertilization amount of HTAC for rice growth. According to ministry of Agriculture, Forestry and Fisheries of Japan, the average rice yield in Ibaraki prefecture was 5.16 t/ha (MAFF, 2016), which is similar with rice yield at the optimal condition. Zhao et al. (2016) investigated four conditions with same level of N (300 kg N/ha) for rice planting: 1) control without fertilizer; 2) chemical fertilizers (conventional dosage); 3) 50% NPK fertilizer plus 6000 kg/ha pig manure; 4) 30% NPK fertilizer plus 3600 kg/ha pig manure organic-inorganic compound fertilizer. The rice yields in condition 2), 3) and 4) were 1.45, 1.47 and 1.54 times higher than in control, respectively. Higher increase rate of rice yield (1.84 times) was achieved in this study by using only HTAC of 180 kg N/ha, indicating the good quality of HTAC for rice growth.

4.2 Effect on Surrounding Environment

According to the environmental quality standard values for water in lakes and marshes (MOE, 2016), the TN and TP in effluent water should be less than 1 mg/L and 0.1 mg/L, respectively. The initial TN concentration in surface water rapidly increased due to simultaneous dissolution by irrigation. Then, it decreased by the uptake of floating plants and rice, or by leaching into the soil. The initial TN concentrations in surface water of R2 (Che. 60), R5 (HTAC 250), R6 (HTAC 500) were relatively higher than others (Figure 3). On day 14, the TN concentration in R2 (Che. 60) decreased to around 1.5 mg/L. However, they were still high in R5 (HTAC 250,

2.5 mg/L) and R6 (HTAC 500, 3.0 mg/L). On day 44, only the TN concentration in R5 (HTAC 250) and R6 (HTAC 500) were higher than 1 mg/L. The TP concentration variation trend was similar with TN. Thus, before the summer drainage, the N and P concentration in surface water in R5 (HTAC 250) and R6 (HTAC 500) exceeded the standard values. If the surface water leaked out, it would cause environmental burden. In the aspect of environmental friendly agriculture, the amount of HTAC less than 250 kg N/ha is recommended for rice growth.

Compared with the chemical fertilizer treatment, the rice yields were higher when the amount of HTAC more than 180 kg N/ha. However, when the amount of HTAC higher than 250 kg N/ha, the N and P concentration in surface water exceeded the standard values just before the summer drainage. Thus the amount of HTAC need to be within 250 kg N/ha, and it also would have little risk of rice lodging. Thus, 180 kg N/ha (R4) was deemed as the optimal fertilization amount of HTAC for rice growth.

5. Conclusion

This research first reported the effect of hyper-thermophilic aerobic compost (HTAC) on rice growth and surrounding environment. HTAC can enhance the rice yield significantly. Considering plant quality, rice yield and environmental friendliness, 180 kg N/ha (R4), which showed similar rice yield with that of chemical fertilizer, was deemed as the optimal fertilization amount of HTAC for rice growth. The N and P concentrations in the surface water at this fertilization amount also were lower than environmental quality standard value before discharging the water.

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