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Effects of Sewage Sludge Application on Rice Growth, Soil Properties, and N Fate in Low Fertile Paddy Soil

Naomi Asagi, Hideto Ueno and Azza Ebid
University Farm, Faculty of Agriculture, Ehime University, 498 Ko, Hattanji, Matsuyama, Ehime 799-2424, Japan

Abstract: The effects of the application of sewage sludge (SS) on the growth indices, yield and nutrient uptake in rice (*Oryza sativa* L. cv. Koshihikari) grown in a low fertility soil were investigated and were compared with the effects of the application of chemical fertilizer (CF) and no fertilizer (NF). The application of SS increased plant growth indices in comparison with the NF treatment; however, at harvest, the dry weight of the plants grown in the SS-treated soil was 30% lower than that of plants in the CF-treated soil. The amounts of N uptake by rice from CF, SS and the soil were determined using the A value method. The amounts of N uptake from the fertilizer and soil in the CF treatment were 0.137 and 0.054 g pot^{-1}, respectively and those in the SS treatment were 0.130 and 0.017 g pot^{-1}, respectively. The N use efficiencies of the plants in the CF- and SS-treated soils were 68.3 and 43.0%, respectively. Therefore, the relative efficiency of SS to CF was 62.9%. In comparison to the NF and CF treatments, the application of SS increased the soil microbial activity; this was determined by assaying the fluorescein diacetate esterase activity. At harvest, the pH of the SS-treated soil was higher than that of the soils in the NF and CF treatments and the electrical conductivity (EC) of the CF-treated soil was higher than that of the soils in the NF and SS treatments.

Key words: ^{15}N, A value, nitrogen, paddy, rice, sewage sludge

INTRODUCTION

In Japan, the amount of sewage sludge (SS) generated each year is continuously increasing; in 2002, it reached 428 M m^{-3} (Japan Sewage Works Association, 2003). The development of recycling techniques for SS has decreased the rate of landfill, sea dumping and incineration and has promoted the use of sludge as a fertilizer and a construction material. Since SS is generally rich in N, P and other plant nutrients, it is desirable to utilize SS in farmlands from the viewpoint of conserving resources and the environment.

Several studies have reported an increase in microbial and enzymatic activities in surface soil after a single or repeated application of organic wastes (Albiach *et al*., 2000; Zaman *et al*., 2002). The application of SS may also have beneficial effects on soil physiochemical and biological properties. However, the application of SS to farmlands has received considerable attention since it not only contains nutrients for plants but also sometimes has a significant level of heavy, potentially toxic metals such as Zn, Cu, Pb and Cd; these pose a risk to human health through accumulation in the soil and food chain (Wilkinson *et al*., 2001). It has also been reported that heavy metal concentrations in SS depend on regional characteristics. Thus, SS from a region with a low concentration of heavy metals can be applied to farmlands with less risk.

Corresponding Author: Naomi Asagi, University Farm, Faculty of Agriculture, Ehime University, 498 Ko, Hattanji, Matsuyama, Ehime 799-2424, Japan Tel: +81-89-993-1636 Fax: +81-89-993-3173

With regard to fertilization, organic materials can be potentially important sources of N in crop production. N is often the most limiting element for plant growth and quality. Therefore, it is important to estimate the amount of N supply and plant N uptake from the applied organic materials. However, in comparison to chemical fertilizers, it is difficult to appropriately control the amount of nutrient supply to crops from organic materials during cultivation; this is because this amount depends on the rate of decomposition and the mineralization processes of organic materials through which nutrients are made available to plants in inorganic or low-molecular-weight forms. Mineralization is a well-known complex process that is regulated by many environmental factors such as soil physicochemical properties, temperature, soil moisture and biota (Oyanagi *et al*., 2002). Several studies have been conducted on N mineralization in SS (Matsuoka *et al*., 2006; Zaman *et al*., 2002, 2004), N and P uptake (Moritsuka *et al*., 2006) and plant N uptake separately from soil and SS by using $\delta^{15}N$ (Goto *et al*., 1996). However, information on the rate of plant N uptake from SS (N use efficiency) is limited.

In this study, we investigated and discussed (1) the fate of N derived from the applied SS and soil separately in the cultivation of rice by using the indirect ^{15}N approach and (2) the effects of SS on the growth indices of rice, microbial activity and soil chemical properties.

MATERIALS AND METHODS

Soil, Treatment and Plant Cultivation

A green house experiment was conducted at the University Farm, Ehime University, Matsuyama, Japan. Wagner pots with an area of 0.02 m^2 were used. Each pot contained 3.37 kg of dry soil (Soil Taxonomy: Typic Udorthents; 1.6% clay, 3.5% silt, 94.9% sand; pH (H_2O) 6.2; electrical conductivity (EC): 0.065 dS m^{-1}; total carbon (TC): 1.32%; total nitrogen (TN): 0.09%; Bray-2 extractable P: 30 mg kg^{-1}; exchangeable K: 0.13 cmol kg^{-1}; exchangeable Ca: 18.1 cmol kg^{-1} and exchangeable Mg: 0.75 cmol kg^{-1}).

The experiment included three N fertilization treatments: (1) no fertilizer (NF) application, (2) application of only the chemical fertilizer (CF) NH_4Cl and (3) application of sewage sludge (SS) fertilizer). SS was obtained from Nishida Industry Inc., Ehime, Japan and its chemical properties were as follows: pH (H_2O) 6.1, 5.8 C/N, 42.3% TC, 7.3% TN, 2.5% P, 0.5% K, 0.8% Ca, 0.9% Mg, 49 mg kg^{-1} Zn, 720 mg kg^{-1} Cu, 0.43 mg kg^{-1} Hg, 5.6 mg kg^{-1} As and 0.4 mg kg^{-1} Cd.

In CF-treated pots, N was applied to the soil at the rate of 4, 3 and 3 g m^{-2} of N as $^{15}NH_4Cl$ (0.942 atom%) at the basal fertilization (June 10, 2004), tillering (June 29, 2004) and young panicle formation stages (July 27, 2004), respectively. On June 10, P and K were applied to the soil at the rate of 4 g m^{-2} of P as fused magnesium phosphate and 8 g m^{-2} K as KCl. In SS-treated pots, SS was applied to the soil at the rate of 208 g m^{-2} (= 15, 5 and 1 g m^{-2} of N, P and K, respectively) and a solution containing 0.1 g m^{-2} of $^{15}NH_4Cl$ (99.8 atom%) was carefully injected into the soil as ^{15}N tracer. K was supplemented to the soil at the rate of 8 g m^{-2} of K as KCl.

On June 10 in 2004, after the application of CF, SS was mixed well in the soil and the pots were flooded with tap water; then three seedlings per pot of rice-*Oryza sativa* L. cv. Koshihikari-were transplanted in each pot. The cultivation was conducted in the greenhouse with eight replicates per treatment.

Sampling and Chemical Analysis

The rice growth (plant height, number of tillers and leaf chlorophyll content index) was measured at 0, 17, 30, 48, 58, 74 and 116 days after transplanting (DAT). The chlorophyll content index was measured with a chlorophyll meter (SPAD-502; Minolta Co. Ltd., Japan). Total microbial activity that was evaluated by measuring the hydrolysis of fluorescein diacet ate (FDA) was measured at 0, 7, 15, 34, 66 and 106 DAT as described by Adam and Duncan (2001).

Plant tops and roots were collected at harvest, oven-dried at 70°C and ground into powder. The total N concentration in the subsamples of the rice plants in the NF treatment was determined using an NC analyzer (Sumigraph NC-80; SCAS, Japan). The total N concentration and abundance of ^{15}N in the subsamples of the plants in the CF and SS treatments were determined using a stable isotope mass spectrometer (ANCA-SL; Europa Scientific Co. Ltd., Cheshire, UK). Soil pH (H_2O) and EC were measured with a pH and conductivity meter (D-24; Horiba Ltd., Japan), respectively, using a suspension of soil and deionized water in a ratio of 1:5 (shaken on a reciprocal shaker for 30 min).

To measure P, K, Ca, Mg, Zn and Cu concentrations, the subsamples of the soil and plants were digested with HNO_3 and $HClO_4$. After digestion, the filtrate was subjected to P analysis by using the molybdate blue method and to K, Ca, Mg, Zn and Cu analyses by atomic absorption spectrometry (AA-6200; Shimadzu Co. Ltd., Japan).

Estimation of N Dynamics by Using the Indirect ^{15}N Method

To estimate the fate of N derived by the rice plants from SS, we employed the A value method-one of the indirect ^{15}N approaches. It is assumed that the available nutrient in the soil (A value) definition implies that when two sources of a given nutrient are present in the soil, the plant will absorb from each of these sources in proportion to the respective quantities available (Fried and Dean, 1952). The *A* value is a time-integrated estimate of the plant-available N pool (Stevenson *et al*., 1998). In the CF treatment (control), ^{15}N-labeled CF was applied. In the organic matter treatment (SS), ^{15}N tracer and unlabeled organic matter were applied.

The A value of the soil, i.e., A_S, in the CF treatment can be determined from Eq. 1.

$$A_S = \frac{100 - \%Ndfcf}{\%Ndfcf} \times A_{CF} \quad (1)$$

where: % Ndfcf is the percentage of plant N uptake from CF applied in the CF treatment and calculated as follows:

$$\%Ndfcf = (^{15}N \text{ atom\% excess in plant}/^{15}N \text{ atom\% excess in CF}) \times 100 \quad (2)$$

The A value of the chemical fertilizer (A_{CF}) is the amount of ^{15}N-labeled fertilizer applied in the CF treatment.

The A value of the soil and organic matter (A_{S+OM}) in the SS treatment can be determined from Eq. 3.

$$A_{S+OM} = \frac{100 - \%Ndft}{\%Ndft} \times A_T \quad (3)$$

where %Ndft is the percentage of plant N uptake from the ^{15}N tracer in the SS treatment and is calculated as follows:

$$\%Ndft = (^{15}N \text{ atom\% excess in plant}/^{15}N \text{ atom\% excess in } ^{15}N \text{ tracer}) \times 100 \quad (4)$$

The A value of ^{15}N tracer (A_T) is the amount of ^{15}N tracer applied in the SS treatment.

It is assumed that the A value of the soil is constant, regardless of the quantity of material added to the soil. Therefore, the A value of the organic matter (A_{OM}) is calculated by subtracting A_S from A_{S+OM}.

$$A_{OM} = A_{S+OM} - A_S \quad (5)$$

The percentage of plant N uptake from the organic matter applied in the SS treatment (%Ndfom) can be calculated from Eq. 6.

$$\%\text{Ndfom} = \frac{\%\text{Ndft}}{A_T} \times A_{OM} \quad (6)$$

The fertilizer use efficiency (FUE) is the recovery rate of N by rice from the applied N source. The FUE rates of the CF (FUE_{CF}) and organic matter (FUE_{OM}) were calculated from Eq. 7 and 8, respectively.

$$FUE_{CF}(\%) = \frac{\text{Ndfcf (mass)}}{\text{Mass of chemical fertilizer (N added)}} \times 100 \quad (7)$$

$$FUE_{OM}(\%) = \frac{\text{Ndfom (mass)}}{\text{Mass of organic matter (N added)}} \times 100 \quad (8)$$

We also computed the relative efficiency-the relative uptake rate of organic matter N to that of CF N-according to Nishida *et al.* (2004) as described in Eq. 9.

$$\text{Relative efficiency } (\%) = \frac{FUE_{OM}}{FUE_{CF}} \times 100 \quad (9)$$

Statistical Analysis

First, the normality of the data was assessed using the chi-square test for goodness of fit and the differences among means were then analyzed by Tukey's test or the Steel-Dwass test by using the software KyPlot (KyensLab Inc., Japan).

RESULTS

Effects on Rice Growth and Dry Weight

The plant height, number of tillers and leaf chlorophyll content index in the SS and CF treatments were higher than those in the NF treatment during cultivation and significant differences were observed (Fig. 1). The plant height in the SS treatment was significantly lower than that in the CF treatment at 17, 48, 58, 74 and 116 DAS, while the number of tillers in the SS treatment was significantly lower than that in the CF treatment at 17 and 58 DAT. The change in the leaf chlorophyll content index in the SS treatment was similar to that in the CF treatment. Eventually, at harvest, the dry weights of the plants (tops and roots) in the CF and SS treatments were significantly higher than those in the NF treatment by 12.3- and 8.4-fold, respectively (Table 1). This value in the SS treatment was significantly lower than that in the CF treatment by 0.7-fold. These results indicate that the application of SS and CF apparently increased the growth indices and yield of rice, although the application of SS in this study resulted in lower levels of growth and yield than in the CF treatment. This was despite the fact that the amount of N applied in SS was 1.5 times that in CF. The dry weight of plant tops and roots differed widely among the three treatments and showed significant differences. The ratios of the dry weight of the plant top to that of the root in the NF, CF and SS treatments were 2.2, 2.0 and 2.8, respectively. This ratio was the highest in the SS treatment.

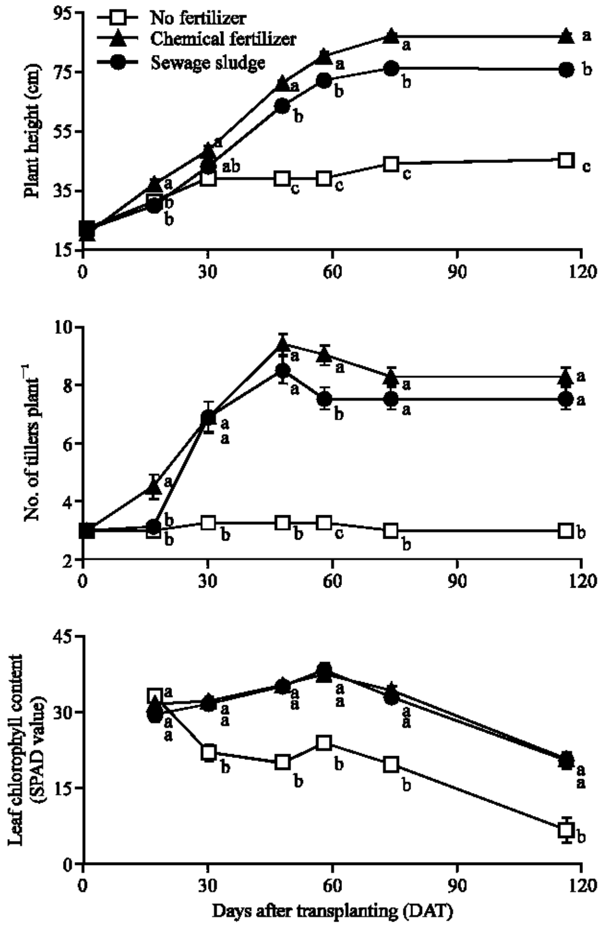

Fig. 1: Change in the rice growth (plant height (A), No. of tillers (B), leaf chlorophyll content index (C)) Period bars denote standard error. Symbols with the same letter in each time indicate no significant differences (p = 0.05)

Table 1: Comparison of the rice (*Oryza sativa* L. cv. Koshihikari) dry weight and N uptake from fertilizer and soil at harvest

	Dry weight average (g pot^{-1})				N uptake average (g pot^{-1})		
Treatments	Top	Root	Total	T/R	Total	Ndff†	Ndfs
No fertilizer	1.7c	0.8c	2.5c	2.2ab	0.009c	-	0.009 (100.0)b
Chemical fertilizer	20.5a	10.3a	30.8a	2.0a	0.191a	0.137 (71.7)a	0.054 (28.3)a
Sewage sludge	15.3b	5.6b	20.9b	2.8b	0.148b	0.130 (88.2)a	0.017 (11.7)b

†Ndff and Ndfs refer to N derived from the applied chemical fertilizer or sewage sludge, from the soil, respectively. Values in parentheses indicate percentages of N uptake from the respective pools to the total plant N. Values with the same letter in each column indicate no significant differences (p = 0.05)

Plant N Uptake from the SS, Cf and Soil

We calculated the amount of N taken up by rice from the CF, SS and soil (Table 1), by using the indirect ^{15}N method. The amount of plant N uptake in the CF and SS treatments was significantly higher than that in the NF treatment; the value in the SS treatment was significantly lower than that in the CF treatment. These were in accordance with the results of dry weight (Table 1). In the CF treatment, N uptake from CF (0.137 g pot^{-1}) was higher than that from the soil (0.054 g pot^{-1}). In the

SS treatment, N uptakes from SS and soil were 0.130 and 0.017 g pot^{-1}, respectively. N uptakes from the soil were lower than those from the fertilizers in the CF and SS treatments. The ratios of N uptake from the soil to the total plant N were 28.3% in the CF and 11.7% in the SS treatments.

The amount of N uptake from SS (0.130 g pot^{-1}) was 0.95 times lower than that from CF (0.137 g pot^{-1}), but no significant difference was observed. On the other hand, N uptake from the soil in the CF treatment was significantly higher than that in the NF and SS treatments and the amounts of N taken up from the soil in the NF, CF and SS treatments were 0.009, 0.054 and 0.017 g pot^{-1}, respectively. The ratios of N uptake from the soil in the CF and SS treatments to that from the soil in the NF treatment were 5.97 and 1.92, respectively.

Table 2 shows the A value and the FUE estimated using the indirect ^{15}N method. The A values of the soil, CF and SS applied were estimated to be 79, 200 and 588 mg pot^{-1}, respectively. The A values of CF and SS were greater than that of the soil by 2.53- and 7.44-fold, respectively. The available N in SS during cultivation was 2.94 times higher than that in CF.

The rates of N uptake derived from the A values of the CF, SS and soil differed widely among all treatments (Table 2). The rates in the NF, CF and SS treatments were 11.9, 68.3 and 22.2, respectively and showed significant differences. The rate of N uptake in the SS treatment was 0.33- and 1.87- fold that in the CF and NF treatments, respectively.

The N use efficiencies of CF and SS were 68.3 and 43.0 of the applied N, respectively (Table 2). The rate in the CF treatment was significantly higher than that in the SS treatment. Therefore, the relative efficiency of SS to CF was 62.9%.

Fate of Fertilizer N

Nitrogen distributions from CF and SS at rice harvest are illustrated in Table 3. The N in CF and SS were distributed to plant uptake (68.3 and 43.0%), N that remained in the soil (5.1 and 14.1%) and unaccounted for N (26.6 and 42.9%). The rate of N uptake from SS was lower than that from CF. The rates of N that remained in the soil and unaccounted for N in the SS treatment were higher than the corresponding rates in the CF treatment. Therefore, the rates of plant N uptake, N that remained in the soil and unaccounted for N in the SS treatment were 0.63, 2.76 and 1.61 times greater, respectively, than the corresponding rates in the CF treatment.

Table 2: The soil and fertilizer A values, the ratio of rice (*Oryza sativa* L. cv. Koshihikari) N uptake and N use efficiency, and the relative efficiency of the chemical fertilizer, sewage sludge, and no fertilizer treatments

	A value average±SE (mg pot^{-1})					
Treatments	Fertilizer	Soil	Total	N uptake/A value† Average±SE (%)	N use efficiency‡ Average±SE (%)	Relative efficiency§ (%)
No fertilizer	-	79±3	79.0±3c	11.9±0.7c	-	-
Chemical fertilizer	200±0.0b	79±3	279.0±3b	68.3±3.8a	68.3±3.8a	100
Sewage sludge	588±12a	79±3	668.0±13a	22.2±1.3b	43.0±2.2b	62.9

†Percentage of the amount of N uptake in the *A* value (fertilizer + soil) of no fertilizer, chemical fertilizer and sewage sludge treatments. ‡Percentage of the amount of N uptake from the applied N. §The relative value of the N use efficiency rate in the sewage sludge application to that in the chemical fertilizer application. Values with the same letter in each column indicate no significant differences (p = 0.05)

Table 3: Comparison of N distribution from the chemical fertilizer and sewage sludge at harvest

	Distribution (%)		
Fertilizers	Plant	Soil	Unaccounted
Chemical fertilizer	68.3a	5.1a	26.6b
Sewage sludge	43.0b	14.1a	42.9a

Values with the same letter in each column indicate no significant differences (p = 0.05)

Microbiological and Chemical Properties of the Soil

The effect of application of SS on soil microbial activity was evaluated by measuring the hydrolysis of FDA. In each soil treatment, microbial activity at a later growth stage (from the maximum tiller number stage to the ripening stage 34, 66 and 106 DAT; Fig. 2B) was higher than that at the early growth stage (initial growing stage -7 and 15 DAT; Fig. 2A). The activity in the SS-treated soil tended to be higher than that in the NF- and CF-treated soil during cultivation and the value in the SS-treated soil at the later growth stage was significantly higher than that in the NF-treated soil. The SS application activated soil microorganisms and their metabolisms.

The pH of SS-treated soil (7.51) was higher than those of NF- (7.16) and CF-treated (7.02) soils and they were significantly different from each other (Fig. 3). The EC of CF-treated soil (0.780 dS m^{-1})

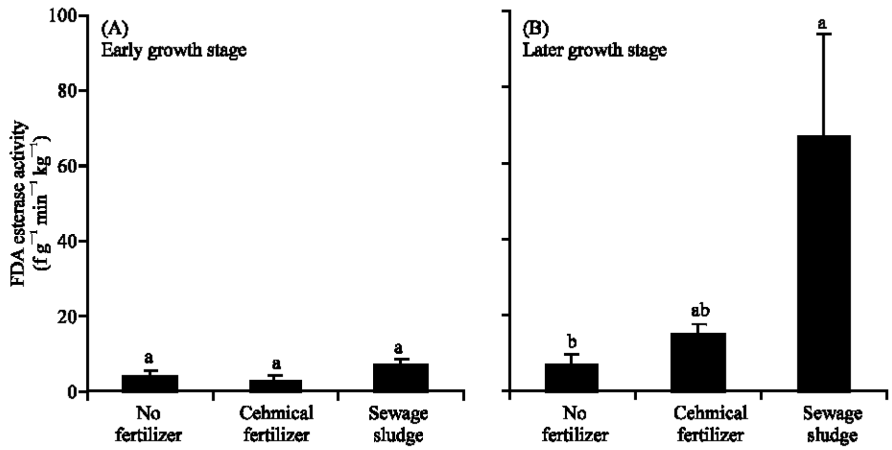

Fig. 2: Comparison of soil microbiological activity determined by fluorescein diacetate esterase activity. Means of the activities are separately indicated in (A) at early growth stage (7 and 15 days after transplanting (DAT)) and in (B) at later growth stage (34, 66 and 106 DAT). Bars denote standard error. Columns with the same letter in each stage indicate no significant differences (p = 0.05)

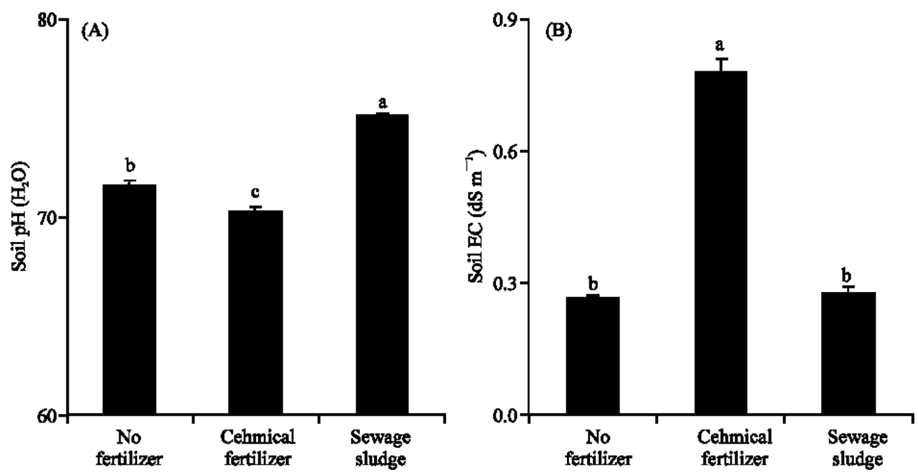

Fig. 3: Comparison of soil pH (A) and electrical conductivity (EC) (B) at harvest. Bars denote standard error. Columns with the same letter indicate no significant differences (p = 0.05)

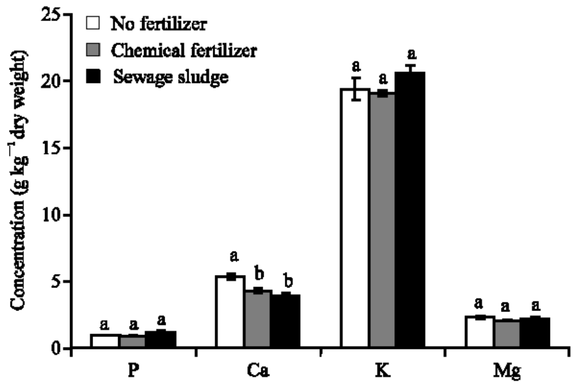

Fig. 4: Comparison of the mineral concentration in the rice stems and leaves at harvest. Bars denote standard error. Columns with the same letter in each mineral indicate no significant differences (p = 0.05)

was significantly higher than those of NF- (0.263 dS m^{-1}) and SS-treated (0.275 dS m^{-1}) soils. The EC value of the CF-treated soil was 2.97 and 2.84-fold the EC values of the NF- and SS-treated soils, respectively. There was no significant difference in the EC values between the NF- and SS-treated soils.

At harvest, no significant differences in the concentrations of P, Ca, K, Mg, Zn and Cu in the soil were observed among the three treatments (data not shown). The concentration of Ca in the stems and leaves of rice in the NF treatment was significantly higher than that in the CF and SS treatments (Fig. 4), although the concentrations of P, K, Mg, Zn and Cu in the rice stems and leaves collected at harvest showed no significant difference among the three treatments. In this study, the contamination of soil and rice stems and leaves with Cu and Zn derived from SS was negligibly low.

DISCUSSION

Plant Growth and Fate of Fertilizer N

Because of the low fertility of the soil, the rice growth and N uptake in the NF treatment were the lowest among the three treatments (Fig. 1 and Table 1). It is assumed that in the NF treatment, macronutrient limitation led to the poor growth of roots and possibly resulted in an extremely low amount of N uptake. Based on the above assumption, the A value and N uptake by rice in the SS treatment were calculated from the distribution of tracer N in the CF but not in the NF treatment.

The plant growth, dry weight and the amount of N uptake in the SS treatment were markedly and significantly higher than those in the NF treatment. Therefore SS could supply sufficient amounts of nutrients to the crop. However, the number of tillers in the SS treatment was significantly lower than that in the CF treatment at 17 DAT (Fig. 1A and B). This may be caused by a slower N mineralization from the SS than that from the CF at the early growth stage of rice or by the decrease in the redox potential of soil following harmful metabolite production during soil microbial fermentation under anaerobic conditions as described by Tanaka and Ono (2000). Moroyu ***et al***. (1981) also reported that the increase in rice height and tiller number was retarded by the rice or barley straw applications.

The A value of SS (588 mg pot^{-1}) was higher than that of CF (200 mg pot^{-1}); however, the rate of N uptake in the A value of SS (22.2%) was significantly lower than that in the A value of CF (68.3%) (Table 2). Consequently, the amount of N uptake in the SS treatment was significantly lower than that in the CF treatment (Table 1). This indicates the relative lower capability of the plant to take up available N in the SS-treated soil than that in the CF-treated soil. This may be caused by the negative effects of the organic matter application on root growth in the SS-treated soil, which is demonstrated by the results that the dry weight of root was lower and the top/root ratio was higher in the SS-treated soil than in the CF-treated soil (Table 1). On the other hand, the relative higher capability of the plant to take up available N in the CF-treated soil may be caused by the positive effects of the CF split application on N use efficiency.

The N use efficiencies of CF and SS were estimated to be 68.3 and 43.0%, respectively (Table 2). Therefore, the relative efficiency of SS to CF was 62.9%. This result suggested from a simple calculation that 1.59 times of N application by SS would be required when compared with that by the CF in order to equalize the uptake amount of N from CF and SS. Recent studies reported widely diverse relative efficiencies of organic fertilizers in paddy soil determined by the ^{15}N direct method, i.e., 70% in rape cake (Uenosono *et al*., 2004), 16-20% in cattle manure compost, 81% in poultry manure compost and 71% in swine feces (Nishida *et al*., 2004).

In this study, the recovery rates of N by rice from the applied CF and SS were 68.3 and 43.0%, respectively (Table 3). The recovery rates of N by rice from applied ammonium sulphate and *Sesbania aculeata* L. were 12.5 and 19.3%, respectively (Azam, 1990). Further, the recovery rates of N by rice were 5.6-7.1% from cattle manure compost, 29.2% from poultry manure compost and 25.4% from swine feces (Nishida *et al*., 2004) and 24.6% from rapeseed pod-wall, 19.1% from rice straw, 14.9% from cattle manure compost and 7.7% from rice straw compost (Ueno and Yamamuro, 2001). The higher recovery rates of fertilizer N in our experiment may be attributed to the fact that the used soil had considerably low fertility, as shown in the NF-treated pot.

The rates of unaccounted for N may be attributed to losses from the soil-plant system due to NH_3 volatilization and/or denitrification. N losses from SS were higher than those from CF. This may be because the higher soil pH (7.51) in the SS treatment led to the higher rate of NH_3 volatilization.

Microbiological and Chemical Properties of the Soil

The microbiological activity in the SS-treated soil tended to be higher than that in the CF-treated soil (Fig. 2). This was in agreement with the results of previous studies on soil enzyme activity (Albiach *et al*., 2000; Zaman *et al*., 2002) and biomass specific respiration (Aoyama *et al*., 2006). The pH of SS-treated soil after harvest was higher than those of the NF- and CF-treated soils (Fig. 3A). The application of SS probably increased the release amount of ammonia, exchangeable Ca and Mg and resulted in the higher pH. The EC of CF-treated soil after harvest was significantly higher than that of SS-treated soil (Fig. 3B). It was considered that the excess amount of CF remained in the soil as inorganic forms. This result was in agreement with the result of a previous study (Zaman *et al*., 2004).Care should be taken to avoid pollution of water resources from the nitrate and nutrient salts derived from the CF.

The concentrations of P, K, Mg, Zn and Cu in the rice stems and leaves at harvest showed no significant differences among the three treatments, although the concentration of Ca in the stems and leaves of the plants in the NF-treated soil was significantly higher than that in the plants in the CF- and SS-treated soils (Fig. 4). The reason for the differences between the plants with regard to Ca uptake is not clear, but it may be related to the relative concentration of Ca in the soil, mycorrhizal effects, or to physiological changes in the root.

Based on these findings, we conclude that SS with a low concentration of heavy metals can be regarded as a useful organic fertilizer from the viewpoint of nutrient supply for soil fertilization and nutrient recycling in the environment; however, further studies are necessary such as the effect of SS on root activity and the evaluation of soil chemical and biological properties in long-term experiments with several soils, crops, climates and soil management techniques, e.g., split application.

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