Effects of the Long-Term Application of Anaerobically-Digested Cattle Manure on Growth, Yield and Nitrogen Uptake of Paddy Rice (Oryza sativa L.), and Soil Fertility in Warmer Region of Japan

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Abstract: The suitability of anaerobically-digested manure (ADM) from a biogas plant, as an alternative to chemical fertilizers for rice cultivation was evaluated by a long-term study. At the standard nitrogen (N) application rate (10 g m⁻²), the aboveground biomass, N uptake, and grain yield in rice plots with ADM application (MF) were not significantly different from those in the plots treated with chemical fertilizer (CF). Split application of ADM improved the apparent N efficiency. The N application rate corresponding to maximum grain yield was approximately 15 g m⁻² by a split application, and more N application by using ADM saturated grain yield due to decrease in ripened grain ratio and individual grain weight. On the whole, the soil total-C, total-N and available N in the MF plot were not significantly different from those in the CF plot. The available phosphate (P) levels was lower in the MF plot than in either the CF plot or plot without N application (NF), mainly due to lower content of P in ADM. However, the P level remained much higher than the fatal threshold level for the growth of paddy rice. These findings suggest that under appropriate fertilization conditions, ADM is a valuable organic resource, and can be used continuously as an alternative to chemical fertilization for rice cultivation, without substantial changes in soil C and N fertility.

Key words: Digested manure, Long-term experiment, Nitrogen, Paddy rice, Soil fertility, Yield.

Due to the rapid expansion of the dairy farm size in Japan, the disposal of wastes has become a prominent problem in the area. This problem may eventually emerge in other Asian countries where the demand for meat and dairy products has been increasing due to rapid economic development and changes in dietary habits (Dong, 2006). For instance, in China a total of 3,200 megatons of animal waste is estimated to be produced by 2020, a volume of waste approximately 1.7 times greater than the level in 2001 (Galloway et al., 2008). Animal manure has been applied to arable land to aid crop growth and to retain or improve soil fertility. If managed properly, manure can be a valuable resource (Holm-Nielsen et al., 2008). However, the increase in the amount of animal waste may result in excessive application of manure, which may cause environmental pollution and threaten human health.

Anaerobic digestion of organic wastes is an effective method to utilize these wastes as resources. During the anaerobic digestion process, organic compounds are decomposed releasing high levels of methane-containing biogas that can be used as an energy resource (Nishio and Nakashimada, 2007). Traditionally, small anaerobic digesters for households or farm units have been used in Europe and Asia. In the past few decades, however, much larger biogas plants have been designed for centralized anaerobic digestion.

The utilization of the large amount of anaerobically-digested manure (ADM) generated from the large-scale biogas plants is an important issue. In some European countries especially represented by Germany and Denmark, ADM has been returned to grassland or cropland as a valuable liquid fertilizer, as it contains a
substantial amount of nutrients essential for plant growth, such as N, P and K (De Boer, 2008). This application has contributed to biomass recycling within agroecosystems. Studies on upland crop cultivation using ADM have been widely conducted throughout the world (Morris and Lathwell, 2004; Matsunaka et al., 2006; Loria et al., 2007). However, the major land use associated with the monsoon climates in most Asian countries is paddy fields and studies on the application of ADM to paddy fields have scarcely been conducted. Recently, numerous Asian countries experiencing rapid economic growth have begun constructing large biogas plants in order to treat dairy and municipal organic waste (Li et al., 2009). Therefore, in Asia, a large amount of ADM is expected to remain after application to the uplands. Treatment of this excess ADM is costly, and an inadequate procedure may result in the escape of a considerable amount of nutrients into the environment, implying inefficient recycling of the waste material. Utilization of ADM into rice cultivation in Asian countries may help solve these problems.

Li et al. (2005) examined the effectiveness of ADM application on a paddy field and concluded that it could be used as an alternative to chemical N fertilizer for rice cultivation. Paddy soils generally accumulate more organic matter compared with upland soils due to the decrease in the decomposition rate of organic matter by the submerged condition during the growing season of rice (Kyuma, 2004). Therefore, it is necessary to examine the effects of ADM application from multiple perspectives and with a long-term approach because ADM contains undigested organic matter as well as inorganic nutrients.

The aim of this study is to investigate the long-term effects of ADM application on the plant growth and nutrient uptake in rice plants, and to analyze the effects on grain yield and major soil properties in a temperate region of Japan where the major agricultural land is mainly paddy field. The results of this study will be useful for introducing ADM into rice cultivation in temperate areas throughout the world.

Materials and Methods

1. Experimental site and fertilization treatments

The field study commenced in 2002, at a paddy field in the Experimental Farm of Kyoto University located in the Osaka Prefecture. The soil at the site was characterized as a sandy grey lowland soil, classified as a Gleysol according to the Food and Agriculture Organization of the United Nations (FAO’s) soil taxonomy, with a particle composition of 68.4% sand, 17.5% silt, and 14.1% clay. Annual mean temperature and precipitation around the experimental farm are 17.0°C and 1260 mm, respectively. In 2001, rice was cultivated uniformly all over the field to eliminate any residual effects of the previous crop in 2000.

The N treatments were no application of N (NF), application of chemical N (CF) and application of ADM (MF). Inorganic N was applied annually at the following rates: 0 g m
-2 in NF, 10 g m
-2 in CF (designated as CF-10) and 5, 10, 15 and 20 g m
-2 in MF-5, MF-10, MF-15 and MF-20, respectively. The fertilizer was applied in a single application (only basal application) or split application (basal-N 70%, topdressing-N 30%). The rice variety Hinoikari, widely cultivated throughout southwestern Japan, was used in this study. The recommended N application rate for cultivation of this variety around the region where the study was undertaken is approximately 10 g m
-2. The MF application rates 5, 10, 15 and 20 g m
-2 corresponded to 50, 100, 150 and 200%, respectively, of the recommended N application. In total, 11 treatments with four replicates each were arranged in the paddy field in a randomized block design. Each plot area was approximately 20 m
-2 (5 by 4 m) and plots were separated with plastic sheets to avoid cross-contamination of nutrients among plots. In the CF plots, ammonium sulfate ((NH4)2SO4) was applied as a N source, and in the MF plots, ADM from the Nantan City Yagi Bioecology Center, Nantan city, Kyoto Prefecture, was used. Table 1 shows the concentration of major nutrients in ADM (average values over 7 yr). It is evident that ADM contains a considerable amount of inorganic N in the form of ammonium (NH4+)
. Although ADM also contains undigested organic matter (described as “Total solid” in Table 1), the levels fluctuate considerably as illustrated by the high value of the total solid coefficient of variance (CV) (39%), the highest variation observed among all of the nutrients measured. For each year, the application amount of ADM was determined according to the NH4-N content in the ADM. In the NF and CF plots, P and K (chemical fertilizer) were applied as a basal application. At the basal ADM application in 2007 and 2008, chemical fertilizer (ammonium sulfate) of approximately 15% of inorganic N to be applied was exceptionally substituted for ADM. The annual application rates of N, P and K are summarized in Table 2.

When harvested, the total rice plant biomass excluding the stubble and underground portions was removed from the field, which was then kept fallow until the next cropping season.
2. Field management in growing period, plant sampling and analysis

In mid-June, three rice seedlings per hill were transplanted with a planting density of 33 by 21 cm (14.4 hills m\(^{-2}\)). A few days after transplanting, the basal application of ADM and chemical fertilizer was conducted, followed by topdressing at the end of July, approximately 25 d before the heading of rice. All experimental plots were kept submerged at 3−5 cm water depth until the heading stage, and intermittent irrigation was then employed thereafter. Pest management practices were conducted in accordance with neighboring farmers’ practices.

Plant samples were collected annually at maturation stage, which occurred approximately 120 d after transplanting. Within each plot, 15−20 hills of rice showing average growth were harvested. Each harvested sample was then equally divided for N analysis and grain yield surveys. Samples analyzed for N were dried at 80ºC for 48 hr, weighed to determine dry weights and ground to pass through a 2 mm sieve. Total N content in these ground samples was measured colorimetrically after the Kjeldahl digestion. The applied N efficiency (NE) for each treatment was calculated using the following equation:

\[
NE = \frac{(NF - NNF)}{NAP}
\]

Where, \(NF\) represents N uptake by the rice plant in each fertilized plot, \(NNF\) represents N uptake in the NF plots, and \(NAP\) represents the amount of inorganic N applied.

Grain yield was determined using the remaining half of the original harvest sample. Fully-ripened grains were separated from the threshed grains with 1.06 g mL\(^{-1}\) saline solution. The yield was defined as the weight of the fully-ripened brown rice with a water content of 15.5%.

3. Soil sampling and analysis

Prior to transplanting in 2002 and 2009, the first and last years of the experiment, three soil sub-samples were collected from the plow layer (0 to 15 cm) of each plot. Samples were air-dried, ground to pass through a 2 mm soil sieve and stored at room temperature under dark condition until analysis. Total carbon and nitrogen contents were measured using the dry combustion method. The available N content was calculated by the anaerobic incubation method: it was calculated from the difference between the extracted NH\(_4\)-N content before and after the incubation of the soil samples at 30ºC for 4 wk. The concentration of NH\(_4\)-N extracted by 10%-KCl solution was determined using the indophenol-blue method. The Bray-2 extraction method was used to determine available P. Phosphate in the extract was measured by the molybdenum-blue method devised by Murphy and Riley (1962).

4. Data analysis

The effects of continuous N application were evaluated by bisecting the dataset into two 4 yr periods with a 1 yr overlap. The periods in data analysis corresponded to the early half (2002–2005) and the latter half (2005–2008) of the study period. Statistic analysis was conducted using STATISTICA for Windows ver. 5.0j (StatSoft Inc., Japan). Fisher’s LSD test was used to detect significant difference among treatments.

Results

1. Weather during the growth period of rice in the seven-year study period

Table 3 shows the monthly fluctuations of the mean
temperature and the sum of precipitation measured during the rice cropping period. Although CV of the mean temperature was generally small, low temperatures in the early growth stages (June to July) influenced the growth of the rice plants. Larger fluctuations of monthly precipitation were observed, particularly in July, August and October, due to the considerable amount of precipitation by rainy front in summer and/or by typhoon in autumn.

2. Rice plant growth and nitrogen uptake

The aboveground biomass of the rice plants at the maturation period is shown in Table 4. Herein, the basal CF-10 without topdressing is shown as CF10-0, and the basal CF-7 followed by topdressing CF-3 is shown as CF7-3, and so on. The average biomasses for the total 7 yr and for the first and last 4 yr are shown for each treatment. Biomass values in the first 4 yr were lower than those in the last 4 yr. Fertilization with N was found to contribute to the increase in plant biomass. According to the LSD test, this effect was most apparent in the plots treated with the standard (10 g m\(^{-2}\)) or larger amount of N. In the split MF application plots, no obvious effect of N application rate on plant biomass was observed as demonstrated by the low CV value of 10%.

The nitrogen content of paddy rice at the maturation stage is shown in Table 5. Herein, the basal CF-10 without topdressing is shown as CF10-0, and the basal CF-7 followed by topdressing CF-3 is shown as CF7-3, and so on. The average nitrogen contents for the total 7 yr and for the first and last 4 yr are shown for each treatment. Nitrogen values in the first 4 yr were lower than those in the last 4 yr. Fertilization with N was found to contribute to the increase in plant nitrogen. According to the LSD test, this effect was most apparent in the plots treated with the standard (10 g m\(^{-2}\)) or larger amount of N. In the split MF application plots, no obvious effect of N application rate on plant nitrogen was observed as demonstrated by the low CV value of 10%.
application decreased as the experiment progressed. The positive effect of split application of N was particularly noticeable when applying larger amounts of N in the form of ADM. Fluctuations in N content with the year (CVs) were larger than those in plant biomass. Regardless of the N application method, the apparent N efficiency (NE) in MF treatments was lower than in CF treatments (Fig. 1). However, the NE in MF treatments, excluding the MF5-0 plot, appeared to increase as the experiment progressed, while NE in CF treatments appeared to decrease slightly. The split application of chemical N fertilizer and ADM appeared to increase the NE value. The results demonstrate that excess application of ADM, above the standard level (MF-10), does not contribute to the increase of NE.

3. Grain yield and yield trend

The yield response to fertilization throughout the 7 yr is shown in Table 6. In general, grain (brown rice) yield was significantly higher in the fertilized plots (MF and CF) than in NF plots. However, ADM application at a low rate, particularly by a single application, did not significantly increase the yield. No significant differences were observed between MF and CF at the standard application plot, but split N application appears to increase grain yield, more greatly than single application except for the MF14-6 plot. Yield fluctuations (CV in Table 6) decreased as the experiment progressed, except for the case of the MF10.5-4.5 and MF14-6 plots.

The relationship between grain (brown rice) yield and N uptake is shown in Fig. 2. Irrespective of the MF application method, grain yield increased with increasing N uptake, peaking at 15 g m$^{-2}$, and further increase in N application decreased as the experiment progressed. The positive effect of split application of N was particularly noticeable when applying larger amounts of N in the form of ADM. Fluctuations in N content with the year (CVs) were larger than those in plant biomass.

Regardless of the N application method, the apparent N efficiency (NE) in MF treatments was lower than in CF treatments (Fig. 1). However, the NE in MF treatments, excluding the MF5-0 plot, appeared to increase as the experiment progressed, while NE in CF treatments appeared to decrease slightly. The split application of chemical N fertilizer and ADM appeared to increase the NE value. The results demonstrate that excess application of ADM, above the standard level (MF-10), does not contribute to the increase of NE.

### Table 6. Comparison of brown rice yield among treatments.

| Treatments | Whole experimental period | 2002–2005 | 2005–2008 |
|------------|---------------------------|------------|------------|
| NF         | 431 d (12.2)              | 439 b (15.5)| 436 d (9.0)|
| CF10.0     | 544 ab (10.0)             | 541 ab (13.4)| 538 ab (5.9)|
| CF7.3      | 553 a (9.1)               | 554 a (11.6)| 552 ab (5.6)|
| MF5.0      | 458 cd (15.5)             | 464 ab (19.5)| 463 cd (10.6)|
| MF3.5-1.5  | 495 bc (11.4)             | 488 ab (16.0)| 506 bc (2.6)|
| MF10.0     | 526 ab (9.1)              | 514 ab (12.2)| 535 ab (3.7)|
| MF7.3      | 544 ab (8.7)              | 535 ab (12.0)| 550 ab (3.1)|
| MF15.0     | 540 ab (10.5)             | 535 ab (14.9)| 544 ab (1.9)|
| MF10.5-4.5 | 578 a (8.7)               | 565 a (10.9)| 571 a (9.5)|
| MF20.0     | 555 a (8.3)               | 550 a (11.4)| 563 a (2.6)|
| MF14-6     | 535 ab (11.5)             | 520 ab (15.2)| 518 abc (15.2)|

Each mean value is shown as involving 15.5% of moisture. In each column, values with different letters denote that they are significantly different at the $p<0.05$ by Fisher’s LSD. Value in parentheses after average denotes coefficient of variance (CV) (%).
uptake levels did not increase the yield. The relationship between N uptake by rice and yield components in the MF plots is shown in Fig. 3. Regardless of the method of N application, the number of spikelets tended to increase with increasing N uptake and more spikelets tended to be generated by split application than by single application (Fig. 3A). The increased number of spikelets corresponded to a decreased ripened grain ratios and 1000 grain weights. This trend was more noticeable in split application of ADM (Figs. 3B and 3C).

Trends in brown rice yield over the 7 yr in each treatment are shown in Fig. 4. To account for yearly fluctuations in yields more accurately, we used a moving average method. Yield variations in NF, CF10-0, CF7-3, MF5-0 and MF20-0 treatments were relatively small, with the difference between minimum and maximum values being less than 30 g m$^{-2}$. The MF14-6 plot showed the largest variation in yield (72.2 g m$^{-2}$).

4. Changes in soil property

Changes in the basic soil properties from the first (2002) to the last (2009) year of the study is shown in Table 7. Of all of the variables analyzed in this study, soil fertility increased from the first to the last years ($\Delta$). Although the amount of Total-C (TC) in each treatment increased from 2.8 to 6.2 g kg$^{-1}$ over the experimental period, these increases did not significantly vary with the N treatment. The increase in Total-N (TN), which ranged from 0.15 to 0.55 g kg$^{-1}$ appeared to be greater in the MF plots than in the NF and CF plots. Available N (AN) increased from 7.9 to 55.3 mg kg$^{-1}$, the increase being greater in fertilized plots (CF and MF). Increases in the amount of available P (AP) were larger in NF and CF plots (433 to 480 mg kg$^{-1}$). In MF plots, AP tended to increase with increasing ADM application rate.

Discussion

Among organic materials, ADM from biogas plants contains a large amount of inorganic N particularly NH$_4$N (Sutton et al., 1978). This composition makes ADM an appropriate alternative to chemical fertilization in rice cropping because rice plants prefer NH$_4$N rather than...
uptake and yield, compared with the NF plots (Tables 4, 5 and 6). However, MF5-0 plot did not increase these values significantly (p<0.05). Thus, this plot displayed apparently lower NE, which may be a consequence of inadequate N uptake during the early growth stage of rice. Takahashi et al., (1976) showed that the N content of rice plant increased exponentially until the panicle differentiation stage of rice, and thereafter it increases linearly depending on the N fertility of the soil. Thus, in the MF5-0 plot most of the inorganic N applied would have been lost or immobilized, causing decreased growth and yields. The plant biomass and grain yield in the MF5-0 plot were improved by split application (MF3.5-1.5) probably because the root system had been substantially formed before topdressing, enabling the quick absorption of topdressed N. Although N loss described above would also have occurred in the other fertilization treatments, higher application rates of N may have compensated for the loss.

At the standard N application rate, no significant difference in rice growth and yield was observed between MF and CF treatments. N uptake in MF plots was slightly lower than that in CF plots, but the difference was not significant. The soil C and N properties observed in the standard MF plots after 7 yr of rice cultivation were not significantly different from those observed in the CF plots. Although the available P in the MF plots in 2009 was significantly lower than that in the CF plots, the amount of available P in the MF plots was still higher than the fatal P levels reported by Komoto (1971), 50–60 mg kg\(^{-1}\) (dry soil). These findings suggest that it would be possible to use ADM as an alternative to chemical N fertilizer.

Application of ADM at higher N application than the standard application (10 g m\(^{-2}\)) resulted in a higher N uptake and grain yield (Tables 5 and 6). Maximum grain

### Table 7. Soil properties sampled from plow layer (0–15 cm) before transplanting in 2002 and in 2009.

| Treatments | Total-C (g kg\(^{-1}\)) | Total-N (g kg\(^{-1}\)) | Available N (mg kg\(^{-1}\)) | Available P (mg kg\(^{-1}\)) |
|------------|-------------------------|-------------------------|-----------------------------|-----------------------------|
|            | 2002 | 2009 | Δ     | 2002  | 2009  | Δ     | 2002  | 2009  | Δ     | 2002  | 2009  | Δ     |
| NF         | 22.3 | 25.4 | 3.1 a | 1.95  | 2.14  | 0.19 b | 100.5 | 108.4 | 7.9 c | 1041  | 1305  | 264 ab|
| CF10-0     | 21.4 | 24.5 | 3.1 a | 1.97  | 2.12  | 0.15 b | 88.4  | 123.7 | 35.3 ab| 1053  | 1533  | 480 a |
| CF7-3      | 19.3 | 22.2 | 2.8 a | 1.76  | 1.97  | 0.21 b | 80.6  | 112.9 | 32.3 ab| 1055  | 1488  | 433 ab|
| MF5-0      | 18.7 | 22.2 | 3.5 a | 1.70  | 1.97  | 0.28 b | 82.6  | 111.1 | 28.5 bc| 1038  | 1255  | 236 c |
| MF3.5-1.5  | 21.1 | 26.2 | 5.1 a | 1.92  | 2.28  | 0.36 ab| 90.6  | 125.7 | 35.2 ab| 1054  | 1498  | 480 c |
| MF10-0     | 19.8 | 24.2 | 4.4 a | 1.84  | 2.15  | 0.31 ab| 90.0  | 131.9 | 41.8 ab| 1112  | 1337  | 225 c |
| MF7-3      | 20.2 | 24.6 | 4.5 a | 1.85  | 2.14  | 0.29 ab| 83.1  | 116.6 | 33.5 ab| 993   | 1175  | 182 c |
| MF15-0     | 22.9 | 29.1 | 6.2 a | 2.08  | 2.46  | 0.38 ab| 92.4  | 117.2 | 24.8 bc| 995   | 1238  | 243 c |
| MF10.5-4.5 | 23.5 | 27.9 | 4.4 a | 2.09  | 2.39  | 0.30 ab| 97.2  | 131.4 | 34.2 ab| 1026  | 1326  | 300 abc|
| MF20-0     | 22.5 | 27.8 | 5.3 a | 2.05  | 2.32  | 0.27 b | 90.7  | 117.7 | 27.0 bc| 993   | 1337  | 344 abc|
| MF14-6     | 20.7 | 26.8 | 6.1 a | 1.91  | 2.46  | 0.55 a | 90.0  | 145.3 | 55.3 a | 1057  | 1334  | 277 bc|

Means followed by same letter do not differ significantly at the p<0.05 level by Fisher’s LSD.
yields were not recorded in the MF-20 plot due to the low ripening rates and light 1000 grain weights (Fig. 3). Rank growth and occasional severe lodging were also observed in MF-20 plots (data not shown). The negative effects associated with intense ADM application were largely due to the increased N application rate, since inorganic N supply from soil was not considered to contribute greatly, except for the MF14-6 plot. These results are not surprising, because the rice variety used in this study, “Hinohikari”, is not expected to grow under excessive fertilization as most of the other varieties currently cultivated in Japan. High N application rates, such as 20 g m$^{-2}$, would be unfavorable except for forage rice or high-yielding rice varieties, with higher yield performance and stronger lodging resistance under high levels of fertilization.

Generally, N topdressing is important since it increases spikelet number and maintains photosynthetic activity during the ripening stage of rice (Yoshida, 1981). Since the study was conducted in a sandy paddy field with a relatively lower nutrient-holding capacity, split N application is more effective at delivering N. In this study, indeed, values of applied NE by split application were higher than those by a single application, regardless of the N source or N application rate (Fig. 1). Increased numbers of spikelets were also generated by split ADM application (Fig. 3A).

In plots with high ADM application, NE plateaued or even decreased, implying that immobilization of N and/or substantial N loss may have occurred. At the time of transplanting, rice seedlings are so small that their capacity to absorb N is limited, consequently the ratio of N lost from the field by ammonia volatilization, leaching and percolation is high. Hou et al. (2007) reported that total N lost through ammonia emissions from ADM, intensely applied to forage rice, ranged from 29.6% to 51.7%, which was much higher than that observed in chemical fertilization (12.2%). This result was due to the increase in the pH of floodwater caused by the mass application of ADM that often show weakly alkaline pH (around 8). High N application using ADM should therefore be limited, both to reduce N loss and to aid environmental protection.

Grain yield in MF and CF plots was positively correlated with N uptake within a low to middle N uptake range (Fig. 2). The N content corresponding to the highest estimated rice plant yield was 14.8 g m$^{-2}$ in the single ADM application plots and 14.4 g m$^{-2}$ in the split ADM application plots. In the single application plots, the increases in spikelet numbers reached saturation at about 15 g m$^{-2}$ N uptake (Figs. 2 and 3). Considering this saturation and lower N efficiency, more yields by single application would be unfeasible, which lead to more nutrient loss and environmental impacts. In the split application, the MF10.5-4.5 treatment appears to deliver the optimal N level (Table 5) while levels of N application above this value would even reduce yield due to the lower ripened ratio and 1000 grain weight, and would also increase the risk of lodging.

The small variation in grain yield over the 7 yr study period in most of the treatments was mainly due to preservation of soil fertility, especially TN and AN (Table 7). Growth of the rice plant depends on inorganic N mineralized from the soil, which sometimes accounts for 60–70% of the total N uptake during the growing period (Koyama, 1975). In this study, plant residues were not returned to the experimental field, which resulted in small differences in soil fertility between successive treatments. On the other hand, a sharp drop in grain yield or a decline in soil N was not observed for NF plots during this study, implying that there is a considerable N supply from the soil system. Biological N fixation by N fixing microorganisms (Yoshida, 1971; Ono and Koga, 1984) may contribute to the observed N availability without N application.

The soil properties suggest that yearly ADM application had only a small effect on soil organic matter and potential inorganic N uptake by the rice plants. In this study, accumulation of soil N during the application of ADM or chemical N fertilizer was quite limited. Generally, the input of the organic matter by the application of ADM is smaller than that by application of fully-matured animal manure. In fact, at the standard application rate, the input of dry matter weight within ADM calculated from the data in Table 1 is 0.26 kg m$^{-2}$, which is approximately half of that commonly applied well-matured cow manure 1 kg m$^{-2}$ (assuming the moisture content; approximately 52%; LEIO, 2005). This is considered to be the minimum application rate to maintain soil fertility. On the other hand, soil total carbon and total nitrogen values in MF plots gradually increased in comparison with those in the NF and CF plots. Gutser et al. (2005) reported that the biodegradability of undigested organic matter in biogas slurry (equivalent to ADM) was lower than that of other organic materials. It is therefore unlikely that organic N contained in the solid organic matter in ADM is mineralized during the year of the ADM application. Meanwhile, if ADM was applied, particularly at a high application rate, were to continue, soil fertility and N mineralization rate would increase because even soil organic matter in soil is degraded slowly. Indeed, we observed the tendency that N uptake rate in standard MF application plots was higher than that in CF plots during growth period from heading stage to maturing stage in fourth and fifth year of the experiment (Nishikawa et al., 2007), which probably reflects the slow N release from solid organic N matter of ADM. These facts also suggest that readjustment of fertilization quantity should be conducted to reduce the negative effects of ADM such as lodging risk. N loss by ammonia volatilization, percolation and nitrate leaching.
Significant differences were observed in the increase in the available P between MF plots with low and standard application rates and the plots with chemical P fertilization (CF and NF). Considering the low P content of ADM and the absence of P fertilizer in MF plots (Tables 1 and 2), this result is reasonable. As mentioned above, however, the available P in MF plots in the experimental field was much greater than the fatal level for rice, which implies the possibility of saving P fertilizer. In instances when ADM with low P content is applied onto a soil with strong phosphate immobilization, such as an Andosol, additional application of P from other sources would then be essential for the normal growth of rice plants.

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References

De Boer, H.C. 2008. Co-digestion of animal slurry can increase short-term nitrogen recovery by crops. J. Environ. Qual. 37: 1968-1973.
Dong, F. 2006. The outlook for Asian dairy markets: the role of demographics, income, and prices. Food Policy 51: 250-271.
Galloway, J.N., Dentener, F.J., Marmer, E., Cai, Z., Abrol, Y.P., Dadhwal, V.K. and Vel Murugan, A. 2008. The environmental reach of Asia. Annu. Rev. Environ. Resour. 33: 461-481.
Gutser, R., Ebertseder, T., Weber, A., Schraml, M. and Schmidhalter, U. 2005. Short-term and residual availability of nitrogen after long-term application of organic fertilizers on arable land. J. Plant Nutr. Soil Sci. 168: 439-446.
Holm-Nielsen, J.B., Al Scadi, T. and Oleskowicz-Popiel, P. 2009. The future of anaerobic digestion and biogas utilization. Bios. Techn. 100: 5478-5484.
Hou, H., Zhou, S., Hosomi, M., Toyota, K., Yosimura, K., Mutou, Y., Nisimura, T., Takayanagi, M. and Motobayashi, T. 2007. Ammonia emissions from anaerobically-digested slurry and chemical fertilizer applied to flooded forage rice. Water Air Soil Pollut. 183: 37-48.
Komoto, Y. 1971. Growth and yield of rice plants in low phosphorus soils. Jpn. Agric. Res. Q. 6: 63-67.
Koyama, T. 1975. Practice of determining potential nitrogen supplying capacities of paddy soils and rice yield (nitrogen problems with special reference to paddy rice cultivation). J. Sci. Soil Manum. Jpn. 46: 260-269*
Kyuma, K. 2004. Long-term chemical and morphological changes induced by alternating submergence and drainage of paddy soils.

In Paddy Soil Science. Kyoto University Press. Kyoto. 115-131.
LEIO. 2005. Annual report of the institute of livestock industry’s environmental technology. 43-52*
Li, K., Inamura, T. and Umeda, M. 2003. Growth and nitrogen uptake of paddy rice as influenced by fermented manure liquid and squeezed manure liquid. Soil Sci. Plant Nutr. 49: 463-467.
Li, R., Chen, S., Li, X., Lar, J.S., He, Y. and Zhu, B. 2009. Anaerobic codigestion of kitchen waste with cattle manure for biogas production. Energ. Fuel. 23: 2225-2228.
Loria, E.R., Sawyer, J.E., Barker, D.W., Lundvall, J.P. and Lorimor, J.C. 2007. Use of anaerobically digested swine manure as a nitrogen source in corn production. Agron. J. 99: 1119-1129.
Matsunaka, T., Sawamoto, T., Ishimura, H., Takakura, K. and Takekawa, A. 2006. Efficient use of digested cattle slurry from biogas plant with respect to nitrogen recycling in grassland. Int. Congr. Ser. 1293: 242-252.
Morris, D.R. and Latham, D.J. 2004. Anaerobically digested dairy manure as fertilizer for maize in acid and alkaline Soils. Commun. Soil Sci. Plant Anal. 35: 1757-1771.
Murphy, J. and Riley, J.P. 1962. A modified single solution method for the determination of phosphate in natural waters. Anal. Chim. Acta. 27: 31-36.
Nishikawa, T., Inoue, H., Umeda, M., Yamasue Y. and Inamura T. 2007. Growth and nitrogen uptake of rice at the paddy field continuously applied with fermented liquid manure (for five years). Jpn. J. Crop Sci. 76 (Extra issue 1): 40-41*
Nishio, N. and Nakashimada, Y. 2007. Recent development of anaerobic digestion processes for energy recovery from wastes. J. Bioso. Bioeng. 103: 105-112.
Oji, Y. and Izawa, G. 1974. Studies on the absorption and assimilation of inorganic nitrogen in intact plants (part 4): Metabolic backgrounds for the differences in utilization of ammonium and nitrate by rice and cucumber plants. Jpn. J. Soil Sci. Plant Nutr. 45: 341-351*
Ono, S. and Koga, H. 1984. Natural nitrogen accumulation in a paddy soil in relation to nitrogen fixation by blue-green algae. Jpn. J. Soil Sci. Plant Nutr. 55: 465-470*
Sutton, A.L., Nelson, D.W., Mayrose, V.B. and Nye, J.C. 1978. Effects of liquid swine waste applications on corn yield and soil chemical composition. J. Environ. Qual. 7: 325-333.
Takahashi, J., Wada, G. and Shoji, S. 1976. The fate of fertilizer nitrogen applied to the paddy field and its absorption by rice plant VI. Influence of a thermal factor on the soil ammonium nitrogen and the absorption of nitrogen by rice plant. Proc. Crop. Sci. Soc. Jpn. 45: 213-219*
Yoshida, S. 1981. Fundamentals of Rice Crop Science. IRRI. Los Baños. p.209.
Yoshida, T. 1971. Soil Microbiology. IRRI Annu. Rep. 24:50.

* In Japanese.