Effect of long-term application of manure and mineral fertilizers on nitrogen mineralization and microbial biomass in paddy soil during rice growth stages

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ABSTRACT

Net N mineralization rate (NMR), net N consumption rate (NCR), microbial biomass carbon (MBC) and nitrogen (MBN), potentially mineralizable N (PMN) and mineral N ($N\text{-}NH_4^+$ and $N\text{-}NO_3^-$) were measured in paddy soil at five growth stages of rice to determine the effect of long-term fertilization in subtropical China. The studied long-term treatments included CK (no fertilization), N, NP, NPK and NPK + OM (NPK plus organic manure). The NPK + OM treatment gave the highest values of the measured variables among all treatments. There was no significant difference in other treatments except for mineral N and PMN at early growth stages. All these variables were generally highest at transplanting stage as two thirds of fertilization was applied as basal fertilizers and the rice uptake was low. Then they decreased or leveled off with the rice growth stages except for MN in all treatments. Stepwise regression revealed that NMR was significantly correlated with MBC and $N\text{-}NH_4^+$ ($R^2 = 0.954$, $P < 0.01$) at all rice growth stages. So, mineral plus manure fertilizer application and more mineral fertilizer as topdressing were recommended in subtropical paddy soil.

Keywords: paddy soil; *in situ*; net N mineralization; soil microbial biomass; potential mineralizable N

Nitrogen mineralization in soil is a key process needed to be fully understood and taken into account when meeting the N demand of crops (Smith et al. 1977). The main sources of N used by crops are from: (i) mineralization of soil organic N; (ii) decomposition of plant residues or organic amendments; and (iii) application of mineral N (Mikha et al. 2006). Nitrogen mineralization produces $N\text{-}NH_4^+$ and $N\text{-}NO_3^-$, which are uptaken by plants and then control the supply and magnitude of mineral N from soil to plants (Ghosh and Kashyap 2003).

Accurate estimation of N mineralization is essential for determining the rate of fertilizer application, optimizing N use efficiency and minimizing adverse impacts of excessive N on the environment (Mikha et al. 2006, Sharifi et al. 2007). An insight into the dynamics of net N mineralization in the field and its influencing factors will provide knowledge for optimizing crop management and improving nutrient use efficiency. Although the *in situ* net N mineralization in paddy soil was intensively studied (Angus et al. 1994, Yan et al. 2006), microbe and to regulate the dyhan tests to plant nutrient N mineralization has received less attention as to its dynamics at different growth stages. Meanwhile, little relevant information including soil microbial biomass is available in paddy soil in long-term fertilizer experiment.

Long-term experiments could provide insight into the N supply and soil fertility response to fertilizer management. The *in situ* net N mineralization rate, N consumption rate, soil microbial biomass, potentially mineralizable N and mineral N were chosen to analyses in this study, which were all related to N mineralization, and may be used as an index of soil function together with other microbiological or biochemical properties. So, the objectives of the study were to evaluate the effects of long-term fertilizer application and rice growth stages on the net N mineralization, microbial biomass and related parameters, and to determine the relationships between the N mineralization variables and selected soil parameters in subtropical paddy soil.

MATERIAL AND METHODS

Site description. The long-term experimental plots were established in 1981 to study the effects of mineral and manure fertilizers application on rice production and soil fertility at the Research Institute of Red Soil of Jiangxi, Jinxian County, Jiangxi Province of China (116°26′E, 28°37′N, about 26 m above sea level). The average annual rainfall is 1727 mm, about 37.8% and 14.4% of which falls during the first rice season (April to July) and the second rice season (July to October), respectively. Monthly mean temperature is lowest (5.5°C) in January and highest (29.9°C) in July.

The cropping system was rice-rice-fallow in winter. Five fertilization treatments were selected for this study, viz. no fertilization (control, CK), N (application of mineral N fertilizer), NP (application of mineral N and P fertilizers), NPK (application of mineral N, P and K fertilizers), and NPK + OM (application of mineral N, P, K plus OM fertilizers). The mineral fertilizer for each rice season was 90 kg N/ha, 20 kg P/ha, 62 kg K/ha applied as urea, calcium magnesium phosphate, and KCl respectively. The organic fertilizer was pig manure at a dose of 22.5 t/ha/year. All the fertilizers were applied as basal fertilizers except for urea, in which two thirds of urea was applied as basal fertilization and one third as topdressing. The treatments were laid out in a completely randomized block design with three replicates, having a plot area of 46.67 m^2 in each replicate. The plots were separated with concrete plates to avoid interference between plots. The soil was derived from Quaternary red clay and classified into a typical Stagnic Anthrosols (IUSS Working Group, WRB, 2006). It has a typical soil profile of Ap-P-Bw1-Bw2. The field was used for paddy cultivation for more than 100 years before the experiment. The initial soil properties of plough horizon (0–15 cm) before the experiment were as follows: soil bulk density of 1.17 g/cm^3, pH (H_2O) of 5.4, soil organic carbon of 16.31 g/kg, total N, P, K of 1.48, 0.54 and 10.39 g/kg, respectively.

Soil sampling and analysis. Soil samples were collected in surface layer (0–15 cm) in each plot at five rice growth stages during the second rice cropping period, *viz.* transplanting stage (GS1, 7 days after transplanting), tillering stage (GS2, 24 days after transplanting), jointing stage (GS3, 45 days after transplanting), grain filling (GS4, 75 days after transplanting) and maturing stage (GS5, 100 days after transplanting) in 2007. The soil was sampled with a soil shovel from 5 random points within each plot, and then mixed to make a composite sample.

Immediately after sampling, excessive water was drained off on the ground. Visible root fragments and stones were removed manually. Samples were transported to the laboratory in polyethylene bags, then spread on paper sheets and air-dried for 12 h before further measurement. Each composite soil sample was sieved (2-mm mesh) and divided into two parts: one part was used to measure potentially mineralizable N, the other for microbial biomass and mineral N (N-$NH_4^+$and N-NO_3^-). Soil moisture content of individual sample was determined gravimetrically in 20 g portions after drying at 105°C for 24 h. Furthermore, soil chemical properties were analyzed at the maturing stage.

Soil pH in distilled water (1:2.5 w/v) was measured using a pH meter equipped with a glass electrode. Soil organic carbon (SOC) was determined by wet digestion method (Snyder and Trofymow 1984). The labile soil organic matter (LSOC) was measured by a spectrophotometer after oxidation of 333 mmol/l $KMnO_4$ (Lefroy et al. 1993). The soil total N (TN) was analyzed by Kjeldahl digestion-distillation method. Soil bulk density (SBD), MN (N-NH_4^+ and N-NO_3^-), total P (TP), total K (TK), available P (AP) and available K (AK) were measured by using routine methods (Lu 1999).

Microbial biomass C and N were determined by the fumigation extraction method (Brookes et al. 1985, Vance et al. 1987). Soil microbial biomass C and N were calculated by dividing the difference of total extractable C (or N) between fumigated and unfumigated samples with conversion factors of 0.38 for biomass C and 0.54 for biomass N.

Potentially mineralizable N (PMN) was determined by the 7-day waterlogged incubation method (Keeney 1982). Soil moist samples (6 g) were placed in a 100 ml centrifuge tube, saturated with 10 ml of deionized water, and incubated at 40°C for 7 days. Then, 40 ml of 2.5 mol/l KCl was added to the tube and shaked for 1 h, then it was filtered through acid-washed filter paper. Finally, the N-NH_4^+ was determined before and after the incubation by the same procedure and PMN was calculated from the difference between two analyses.

***In situ* measurement of net N mineralization.** The *in situ* net N mineralization was measured at the five growth stages by using an improved closed-top solid cylinder method (Angus et al. 1994, Yan et al. 2006). This method involved driving 10 PVC tubes (6 cm in internal diameter and 25 cm in depth) to a depth of 15 cm and leaving the enclosed soil undisturbed for about 2–4 weeks. The water surface in the tube was covered with a 2 mm thick layer of liquid paraffin to inhibit denitrification losses. The top of each tube was then covered with rubber stopper to prevent overflow and the effect of light and rainfall. The incubation began at transplanting stage and sequential samplings (according to the rice growth stage) were made 4 times during the whole incubation period. To represent the starting value, a soil sample was collected before incubation in an adjacent undisturbed patch of soil. A sub-sample was analyzed for N-NH_4^+ and N-NO_3^-. After each sampling, the tubes were driven into an adjacent undisturbed patch of soil. A soil sample was collected again before the next incubation period.

The *in situ* net N mineralization rate (NMR) was calculated as the difference between mineral N values at the end and beginning of incubation period to the time interval. The net N consumption rate (NCR) was determined as the difference between mineral N values in inner and outer of PVC tubes at the sampling time to the time interval.

Statistical analysis. One-way analysis of variance (ANOVA) was used to compare differences between mean soil values. Two-way ANOVA was performed to determine the effects of fertilizer treatments, rice growth stages and their interactions on N mineralization parameters. Least significant difference test (LSD) was applied to determine whether means differed significantly. Correlation and regression analyses were used to examine the relationships between N mineralization, microbial biomass and other soil parameters. All the statistical analyses were conducted using the statistical package SPSS 11.5.

RESULTS AND DISCUSSION

Physical and chemical soil characters

The physical and chemical properties of soils sampled at the maturing stage were presented in Table 1. Compared with the initial soil, 26-year fertilization increased SOC by 19.2%–58.2% in all treatments including CK treatment. Generally, SOC contents in paddy soils became stable after 30 years of rice cultivation in subtropical China (Li et al. 2003). So, this increase of SOC could be attributed to the increased underground biomass due to the introduction of modern rice varieties as well as increased nutrient inputs through rainfall and irrigation water.

The NPK + OM treatment was greater in SOC, as well as the related properties such as TN, TP and pH among the treatments. But for SOC, TN and pH, there was no significant difference between the CK and mineral fertilization treatments. The results indicated that the increase of SOC was mainly attributed to the input from pig manure or increased root biomass (Kundu and Ladha 1995). The similar results were also reported in Nepal by Regmi et al. (2002).

The increase in SOC resulted in the decrease of soil bulk density (SBD) in the NPK + OM treatment; however, there was no significant difference in SBD within all treatments. The N application increased MN and the magnitude of the increase was greater in NPK + OM than in other N fertilizer treatments. The treatments without P application (CK and N) had the lowest TP and AP. The K application significantly increased the AK, but not the TK.

Table 1. Selected soil properties in the long-term fertilizer experiments at the maturing stage of late rice in 2007 in Jinxian, China

Treatments	pH	SBD (g/cm^3)	SOC	LSOC	TN	TP	TK	MN	AP	AK
			(g/kg)					(mg/kg)		
CK	4.92^b	1.07^a	19.44^b	2.44^c	2.02^b	0.54^d	9.68^a	10.61^d	10.09^c	50.93^c
N	4.93^b	1.04^a	19.70^b	2.92^c	2.06^b	0.49^d	9.61^a	12.52^c	10.53^c	50.33^c
NP	5.01^b	1.03^a	20.11^b	2.61^c	2.11^b	0.69^c	9.97^a	13.60^b	25.10^b	50.73^c
NPK	5.05^b	1.05^a	20.91^b	3.33^b	2.09^b	0.84^b	10.18^a	14.49^b	31.47^b	63.05^b
NPK + OM	5.25^a	0.99^a	25.81^a	4.02^a	2.64^a	1.31^a	10.11^a	26.30^a	108.35^a	71.78^a

Means, in a column, followed by a common letter are not significantly different at $P < 0.05$ based on Fisher's LSD test, pH, soil:water = 1:2.5; SBD, soil bulk density; SOC, soil organic carbon; LSOC, liable soil organic carbon; TN, soil total N; TP, soil total P; TK, soil total K; MN, mineral N; AP, soil available P; AK, soil available K

Table 2. ANOVA tests of between-subjects effects of N-NH_4^+, N-NO_3^-, NMR, NCR, MBC, MBN and PMN during rice growth stages

Source	Variable	N-NH_4^+	N-NO_3^-	NMR	NCR	MBC	MBN	PMN
Fertilization	*F*-values	32.44	48.93	43.36	35.32	308.47	77.84	113.38
	significance	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Growth stage	*F*-values	33.56	8.70	11.84	38.29	20.96	56.84	16.18
	significance	0.00	0.005	0.006	0.00	0.00	0.00	0.001
Fertilization × Growth stage	*F*-values	9.81	10.84	0.25	3.08	3.04	1.67	4.86
	significance	0.00	0.00	0.98	0.02	0.01	0.14	0.00

NMR, net N mineralization rate; NCR, net N consumption rate; PMN, potentially mineralizable N; MBC, microbial biomass C; MBN, microbial biomass N

Dynamics of mineral N

Both N-NH_4^+ and N-NO_3^- in soil were significantly affected by rice growth stage and fertilization treatment, depending on N form as well (Table 2, Figure 1).

N-NH_4^+ content decreased generally with the rice growth stage (Figure 1) and was greater in the NPK + OM treatment than in other treatments at all growth stages except for the jointing stage, at which stage N-NH_4^+ content was not significantly different among the fertilization treatments. N-NH_4^+ content was over 13.9 mg/kg at the transplanting stage in all treatments and was greater than 24.6 mg/kg in the NPK + OM treatment at the transplanting and tillering stages,

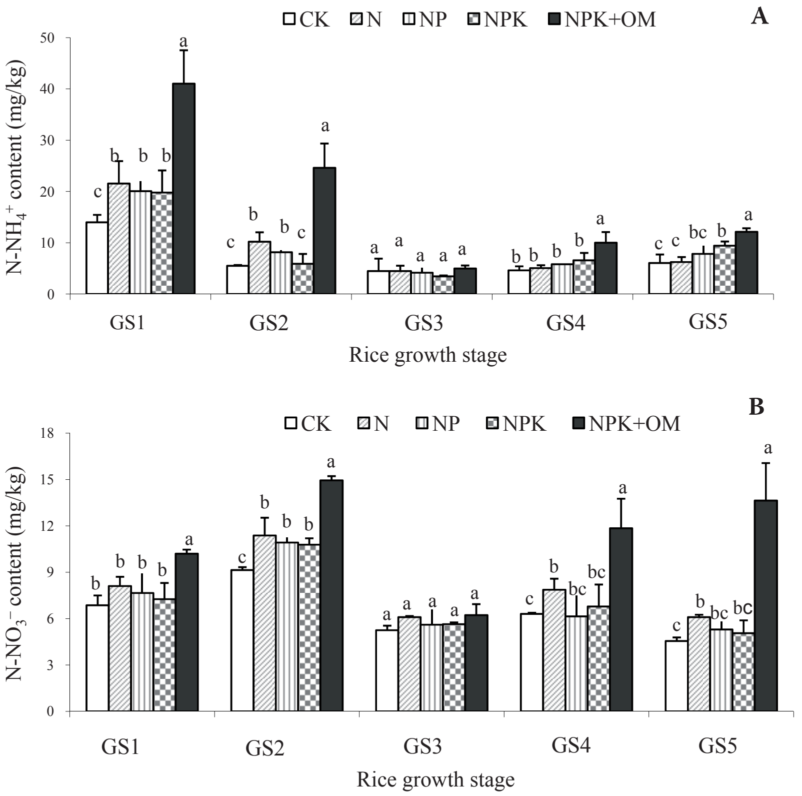

Figure 1. Dynamics of N-NH_4^+ and N-NO_3^- contents during rice growth stages under long-term fertilizer experiment. Bars represent standard deviation (S.D.) of three replicates. Different letters indicate significant differences among the fertilzer treatments at the same rice growth stage. GS1 = transplanting stage, GS2 = tillering stage, GS3 = jointing stage, GS4 = grain filling stage, GS5 = maturing stage

while it was less than 12.2 mg/kg at the following stages. Moreover, the N-NH_4^+ content showed no significant difference between CK and mineral fertilization treatments at the jointing and grain filling stages.

Different from N-NH_4^+, N-NO_3^- content was greatest at the tillering stage, followed by the transplanting stage and lowest at grain filling stage. N-NO_3^- content was also greatest in the NPK + OM treatment at all growth stages except for the jointing stage, at which stage N-NO_3^- content was not significantly different among the fertilization treatments. N-NO_3^- content in the CK treatment was significantly lower than the mineral fertilization treatments at the tillering, grain filling and maturing stages, while N-NO_3^- content in the mineral fertilization treatments was not significantly different at all growth stages.

A large proportion of basal fertilizers transform into N-NH_4^+ and thus lower rice uptake can result in the higher N-NH_4^+ content at the early growth stages. The decrease of N-NH_4^+ with time was due to preferential uptake of N-NH_4^+ by rice plant. Less variation in N-NO_3^- during the rice growth stages was found, because it was transformed from N-NH_4^+ and not preferentially used by rice plant. Compared with CK, mineral N application increased N-NH_4^+ and N-NO_3^- at early growth stages and no significant difference was observed at late growth stages, suggesting the mineral N dose was not sufficient and its supply ability was not persistent.

In situ net N mineralization rate and N consumption rate

The net N mineralization rate and N consumption rate were also significantly affected by rice growth stages and fertilization treatments (Table 2). In general, both NMR and NCR decreased with the development of rice growth stages, which were greater in NPK + OM than other treatments and no significant difference among the mineral fertilization treatments (Figure 2). It suggested that the manure input could increase soil total N and

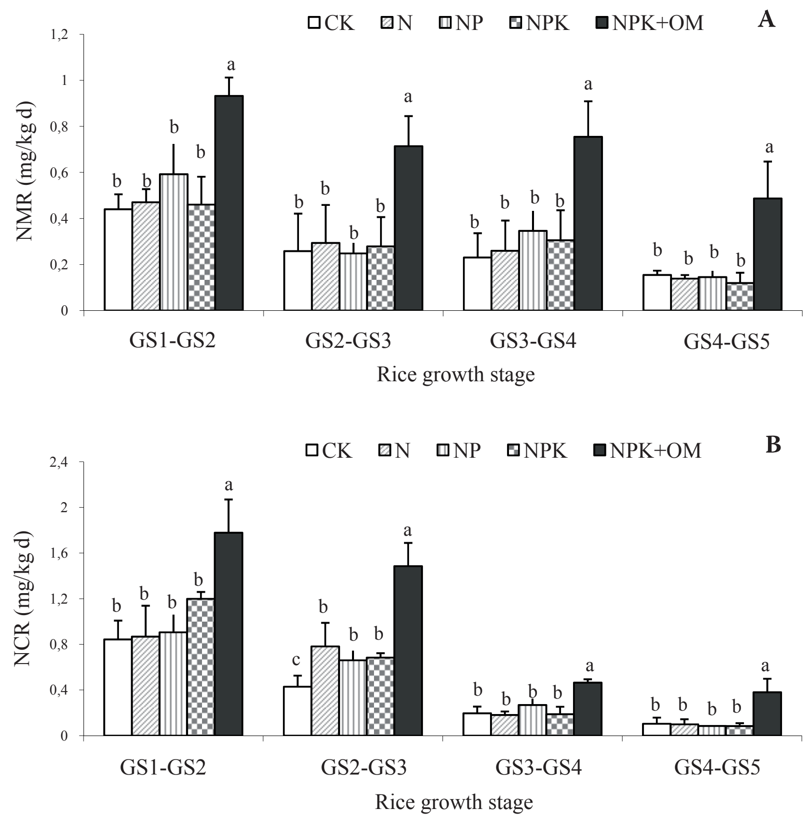

Figure 2. Dynamics of net nitrogen mineralization rate (NMR) and consumption rate (NCR) at different rice growth stages under long-term fertilizer experiment. Bars represent standard deviation (S.D.) of three replicates. Different letters indicate significant differences among the fertilization treatments at the same rice growth stage. GS1 = transplanting stage, GS2 = tillering stage, GS3 = jointing stage, GS4 = grainfilling stage, GS5 = maturing stage

improve the nutrient status of soil. While, NMR and NCR in CK treatment were not significantly different from those in mineral fertilization treatments except for NCR at the time from tillering to jointing stages. This means that N application was not sufficient in the experiment.

The highest NMR at early rice growth stages was probably caused by the mineralization of N from basal fertilizer application (Ghosh and Kashyap 2003). Decomposable organic material in the NPK + OM could also stimulate N mineralization in soil (Kundu and Ladha 1997). The similar levels of NMR at whole rice growth stages among the treatments except for NPK + OM could attribute to the similar level of SOC and TN (Table 1). The NMR was low at the end of rice growth stages, probably due to the decreased labile organic matter and microbial activity. In CK treatment, the NCR was lower than in the mineral fertilization treatments from tillering stage to jointing stage, suggesting that N uptake by rice crop during the period was not only from the indigenous soil but also from the mineral N transformation.

The NCR was significantly greater than NMR at tillering and jointing stages in all treatments, indicating that the mineralization N from soil was not sufficient for rice growing, if not considering the N input from the outside environment. So, more N fertilizer should be applied especially at the jointing stage.

Soil microbial biomass C and N

Soil MBC and MBN were greater in NPK + OM treatment than in other fertilization treatments at all rice growth stages. Soil MBC was lower in CK treatment than in other treatments only at the transplanting stage (Figure 3). Kaur et al. (2005) reported the similar results that the microbial biomass increased from no fertilized soils to mineral fertilized soils and to organic amended soils. Amendment of

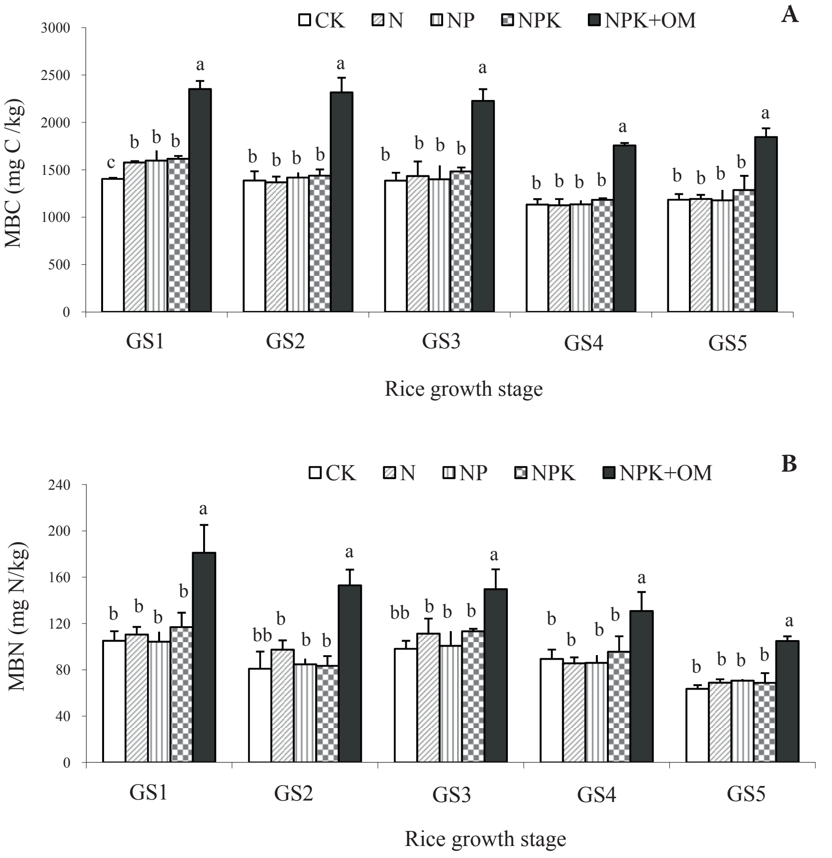

Figure 3. Dynamics of soil microbial biomass carbon (MBC) and microbial biomass nitrogen (MBN) during rice growth stages in paddy soil under long-term fertilizer experiment. Bars represent standard deviation (S.D.) of three replicates. Different letters indicate significant differences among the fertilization treatments at the same rice growth stage. GS1 = transplanting stage, GS2 = tillering stage, GS3 = jointing stage, GS4 = grain filling stage, GS5 = maturing stage

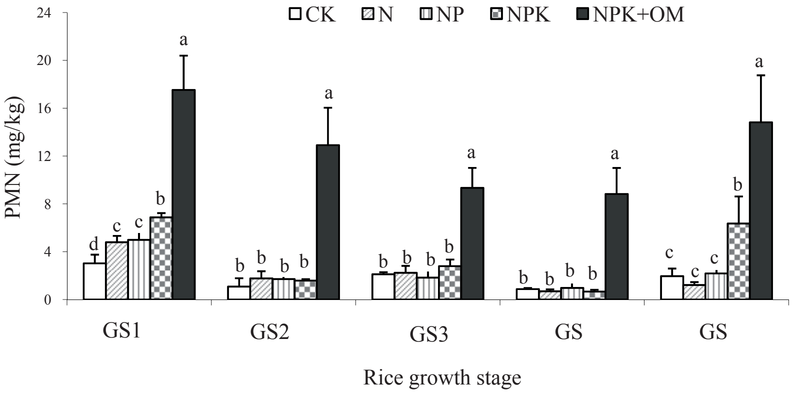

Figure 4. Dynamics of potentially mineralizable nitrogen (PMN) during rice growth stages in paddy soil under long-term fertilizer experiment. Bars represent standard deviation (S.D.) of three replicates. Different letters indicate significant differences among the fertilization treatments at the same rice growth stage. GS1 = transplanting stage, GS2 = tillering stage, GS3 = jointing stage, GS4 = grain filling stage, GS5 = maturing stage

organic materials input more organic carbon to the soil, which could stimulate the microbial growth and activity (Sastre et al. 1996, Kanchikerimath and Singh 2001).

Soil MBC and MBN were both decreased with the development of rice growth stages. The higher MBC and MBN at the transplanting stage in all treatments could attribute to exposed organic matter from puddling and more available nutrients for microbe growing. It was lower at the late growth stages, which might relate to the less root exudation (Aulakh et al. 2001) coupled with the soil drainage management at rice maturity.

The MBC/SOC ratio ranged from 5% to 9% with the rice growth stages in all treatments (data not shown). The similar result was found in other paddy soils in China (Yang et al. 2005). The mean MBC/SOC ratio was highest in NPK + OM treatment, which suggested that organic manure practices had a positive influence on this ratio. Furthermore, the larger ratio implied an increased availability of fresh substrates, while smaller one implied reduced availability (Anderson and Domsch 1989).

Potentially mineralizable N

Potentially mineralizable N (PMN) was affected largely by fertilization treatments and rice growth stages (Figure 4). The interactive effect of NPK + OM treatment and transplanting stage showed the highest PMN (17.53 mg/kg) in paddy soil. But there were no significant difference with PMN among CK, N, NP, and NPK treatments except for the PMN at transplanting and maturing stages. Meanwhile, the highest value (7.38 mg/kg) of mean PMN was at transplanting stage followed by maturing stage (5.72 mg/kg), and the lowest (2.41 mg/kg) at grain filling stage.

The anaerobic incubation method (measured as PMN) was recommended by Keeney (1982) for its satisfactory relationship between the result and other available N indices. Because the same biological processes that controlled the release of plant available N in the field were also responsible for production of mineral N in this method. The PMN in this study had significant correlation with NMR, MBC and MBN ($P < 0.01$) at whole rice growth stages (Table 3), suggesting that the PMN fraction in paddy soil was mediated by soil microbial biomass, which also acted as an important source for mineralizable N in soil.

Correlations

Correlation studies showed the existence of a positive significant relationship ($P < 0.01$) between the N mineralization variables and soil biochemical characters during rice growth stages except for N-NO_3^- (Table 3). Stepwise regression equations were developed to find that NMR was significantly associated with MBC and N-NH_4^+ ($R^2 = 0.954$, $P < 0.01$), and NCR was significantly associated with N-NH_4^+ ($R^2 = 0.834$, $P < 0.01$). The results suggested that the NMR was controlled by microbial size and activity, and the amount of N-NH_4^+ was mostly produced from N mineralization in soil and consumed by rice crop uptake.

The correlations between N mineralization variables and selected soil chemical parameters at maturing stage were shown in Table 4. It was

Table 3. Correlation (R^2 values) matrix for NMR, NCR, PMN, MBC, MBN, N-NH_4^+ and N-NO_3^- during rice growth stages

Variables	NCR	PMN	MBC	MBN	N-NH_4^+	N-NO_3^-
NMR	0.797**	0.946**	0.947**	0.902**	0.774**	0.410
NCR		0.762**	0.729**	0.660**	0.913**	0.611**
PMN			0.869**	0.739**	0.706**	0.583**
MBC				0.903**	0.671**	0.589**
MBN					0.669**	0.465*
NH_4^+-N						0.459*

NMR, net N mineralization rate; NCR, net N consumption rate; PMN, potentially mineralizable N; MBC, microbial biomass C; MBN, microbial biomass N; *, **indicate the significance of correlation at $P < 0.05$ and 0.01 ($n = 25$, data pooled from all five stages)

Table 4. Correlation (R^2 values) matrix between soil N mineralization variables and selected soil parameters at the maturing stage ($n = 15$)

Variables	SOC	LSOC	TN	MN	C/N	pH
NMR	0.737**	0.746**	0.774**	0.827**	0.102	0.830**
MBC	0.873**	0.801**	0.991**	0.936**	0.097	0.825**
MBN	0.894**	0.771**	0.956**	0.963**	0.090	0.796**
PMN	0.865**	0.617*	0.910**	0.895**	0.115	0.653*

NMR, net N mineralization rate; MBC, microbial biomass C; MBN, microbial biomass N; PMN, potential mineralizable N; SOC, soil organic carbon; LSOC, liable soil organic carbon; TN, soil total N; MN, mineral N; *, **marked correlations are significant at $P < 0.05$ and 0.01, respectively (all data pooled from five treatments)

observed that NMR, MBC, MBN and PMN were all significantly correlated with SOC, LSOC, TN, MN and pH ($P < 0.05$). The similar results were also found by Ireneo et al. (1996) in wetland rice soil.

The results demonstrated that both N mineralization variables and microbial biomass were significantly influenced by fertilizer treatments, rice growth stages and even their interactions. In general, the mineral fertilizer application (N, NP and NPK) did not significantly affect the N mineralization and soil microbial biomass compared with the CK under current fertilization conditions. So, mineral plus manure fertilizer application and more mineral fertilizer as topdressing were recommended in subtropical paddy soil.

REFERENCES

Anderson T.H., Domsch K.H. (1989): Ratios of microbial biomass carbon to total organic carbon in arable soil. Soil Biology and Biochemistry, *21*: 471–479.

Angus J.F., Ohnishi M., Horie T., Williams R.L. (1994): A preliminary study to predict net nitrogen mineralization in a flooded rice soil using anaerobic incubation. Australian Journal of Experimental Agriculture, *34*: 995–999.

Aulakh M.S., Wassmann R., Bueno C., Kreuzwieser J., Rennenberg H. (2001): Characterization of root exudates at different growth stages of ten rice (*Oryza sativa* L.) cultivars. Plant Biology, *3*: 139–148.

Brookes P.C., Landman A., Pruden G., Jenkinson D.S. (1985): Chloroform fumigation and the release of soil nitrogen: A rapid direct extraction method to measure microbial biomass nitrogen in soil. Soil Biology and Biochemistry, *17*: 837–842.

Ghosh P., Kashyap A.K. (2003): Effect of rice cultivars on rate of N-mineralization, nitrification and nitrifier population size in an irrigated rice ecosystem. Applied Soil Ecology, *24*: 27–41.

Ireneo J.M., Iwao W., Grace B.M., Jasper G.T. (1996): Nitrogen mineralization in tropical wetland rice soils: I. Relationship with temperature and soil properties. Soil Science and Plant Nutrition, *42*: 229–238.

Kanchikerimath M., Singh D. (2001): Soil organic matter and biological properties after 26 years of maize–wheat–cowpea cropping as affected by manure and fertilization in a Cambisol in semiarid region of India. Agriculture, Ecosystems and Environment, *86*: 155–162.

Kaur K., Kapoor K.K., Gupta A.P. (2005): Impact of organic manures with and without mineral fertilizers on soil chemical and biological properties under tropical conditions. Journal of Plant Nutrition and Soil Science, *168*: 117–122.

Keeney D.R. (1982): Nitrogen-availability indexes. In Page A.L. et al. (ed.) Methods of soil analysis. Part 2. 2nd ed. Agronomy Monogr. 9. ASA and SSSA Madison, WI. pp. 711–733.

Kundu D.K., Ladha J.K. (1995): Efficient management of soil and biologically-fixed N_2 in intensively cultivated rice fields. Soil Biology and Biochemistry, *27*: 431–439.

Lefroy R.D.B., Blair G., Strong W.M. (1993): Changes in soil organic matter with cropping as measured by organic carbon fractions and ^{13}C natural isotope abundance. Plant Soil, *155*/*156*: 399–402.

Li Z.P., Li D.C., Zhang T.L., Chen B.Y., Yin R.L., Shi, Y.Q., (2003): Dynamics of soil properties of paddy fields in red soil region. Acta Pedological Sinica, *40*: 870–878. (In Chinese)

Lu R.K. (1999): Analytical methods for soils and agricultural chemistry. Beijing: China Agricultural Science and Technology Press. (In Chinese)

Mikha M.M., Rice C.W., Benjamin J.G. (2006): Estimating soil mineralizable nitrogen under different management practices. Soil Science Society of America Journal, *70*: 1522–1531.

Regmi A.P., Ladha J.K., Pathak H., Pasuquin E., Bueno C., Dawe D., Hobbs P.R., Joshy D., Maskey S.L., Pandey S.P. (2002): Yield and soil fertility trends in a 20-year rice-rice-wheat experiment in Nepal. Soil Science Society of America Journal, *66*: 857–867.

Sastre I., Vicente M.A., Lobo M.C. (1996): Influence of the application of sewage sludges on soil microbial activity. Bioresour Technology, *57*: 19–23.

Sharifi M., Zebarth B.J., Burton D.L., Grant C.A., Cooper J.M. (2007): Evaluation of some indices of potentially mineralizable nitrogen in soil. Soil Science Society of America Journal, *71*: 1233–1239.

Smith S.J., Young L.B., Miller G.E. (1977): Evaluation of soil nitrogen mineralization potentials under modified field conditions. Soil Science Society of America Journal, *41*: 74–76.

Snyder J.D., Trofymow J.A. (1984): A rapid accurate wet oxidation diffusion procedure for determining organic and inorganic carbon in plant and soil samples. Communications in Soil Science Plant Analysis, *15*: 587–597.

Vance E.D., Brookes P.C., Jenkinson D.S. (1987): An extraction method for measuring soil microbial biomass C. Soil Biology and Biochememistry, *19*: 703–707.

Yan D.Z., Wang D.J., Sun R.J., Lin J.H. (2006): N mineralization as affected by long-term N fertilization and its relationship with crop N uptake. Pedosphere, *16*: 125–130.

Yang C.M., Yang L.Z., Zhu O.Y. (2005): Organic carbon and its fractions in paddy soil as affected by different nutrient and water regimes. Geoderma, *124*: 133–142.

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