N₂O AND CH₄ FLUXES FROM *Acacia mangium* PLANTATION SOILS IN RESPONSE TO NITROGEN APPLICATION AND FH LAYER REMOVAL

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Abstract: We measured N₂O fluxes and CH₄ uptake rates following NH₄Cl and KNO₃ (100kg N ha⁻¹) application with and without FH layer during a relatively dry season in an *Acacia mangium* plantation stand in Sumatra, Indonesia. High N₂O fluxes at control (no treatment) with FH (0.46 – 0.53 mg N m⁻² d⁻¹) suggested that *A. mangium* soils function as a larger source of N₂O than natural forest soils. In the relatively dry season, FH layers in the *A. mangium* plantation were not a direct source of N₂O, but appear to contribute to nitrogen cycling and the following N₂O production in mineral soils as a supplier of available carbon and nitrogen. Application of NO₃⁻ fertilizers significantly increased N₂O fluxes irrespective of the FH removal treatment, suggesting that increased NO₃⁻ availability enhanced N₂O emissions through the denitrification process and that anaerobic microsites can exist even in the relatively dry soils in the *A. mangium* plantation. CH₄ uptake rates at control with FH layer ranged between 0.70 – 0.84 mg C m⁻² d⁻¹, which is consistent with other natural tropical forest soils. *A. mangium* soils supplied with N rich litter do not appear to decrease the function as a sink for atmospheric CH₄ at least in a relatively dry season, though NH₄⁺ addition significantly reduced CH₄ uptake rates.

Key words: Acacia mangium, Fast wood plantation, Nitrous oxide, Methane, Denitrification, Nitrogen application

1. INTRODUCTION

Tropical forest soils are an important source of nitrous oxide (N₂O), and act as sinks for methane (CH₄) (Keller et al., 1986; Potter et al., 1996). The recent rapid increases in the atmospheric concentrations of these major greenhouse gases significantly contribute to global warming (IPCC, 2007). N₂O is mainly produced in soils by microbial processes of nitrification under aerobic conditions and denitrification under anaerobic conditions (Davidson et al., 2000). CH₄ is produced by methanogens under anaerobic conditions and is consumed by methanotrophs under aerobic conditions of soils (Le Mer and Roger, 2001).

The emission and uptake rates from soils of these greenhouse effect gases are strongly dependent on various environmental conditions that control microbial activities, such as temperature, soil moisture and substrate availability (Firestone and Davidson, 1989; Davidson et al., 2000). Especially, nitrogen availability indicated by the concentrations of available ammonium (NH_4^+) and nitrate (NO_3^-) , which are direct substrates of nitrification and denitrification, influences the N₂O emission rates significantly. Many field studies showed that the NH_4^+ and $NO_3^$ fertilization stimulates N2O emissions from agriculture soils (Skiba et al., 1993), grassland (Velthof et al., 1997) and forest (Keller et al., 1988). Moreover, N₂O emissions from soils have been shown to be largely influenced by the quality of the plant litter (Erickson et al., 2001). Presence of leguminous and other nitrogen (N)-fixing trees in forests may enhance N₂O emission from the soils, because they produce N rich litter through symbiotic N fixation, leading to high soil N availability and fast soil N cycling (Erickson et al., 2001). In the fast-growing leguminous tree plantations in tropical Asia, their soils have been demonstrated to be a significant source of N_2O as well (Arai et al., 2008; Konda et al., 2008). Therefore, it is required to elucidate the mechanisms of N_2O emissions from the leguminous tree plantation soils in order to develop methods to predict the emissions more accurately and to mitigate the emissions.

Litter layer in forests can function not only as a substrate supplier into the soils but also a direct source of N_2O . Some studies indicated that litter layers were the direct N_2O emission source (Dong et al., 1998; Tietema et al., 2007). Konda et al. (2008) also suggested the possibility that FH layers accumulated on the *A. mangium* plantation soils were a direct source of N_2O and we suspected that FH layers contributed to their N_2O emissions.

The effect of high N inputs on CH_4 dynamics is still controversial. Many studies have reported that the N fertilization of forest soils had an inhibitory effect on CH_4 oxidation rate (Steudler et al., 1989; King and Schnell, 1994), since NH_4^+ may compete with CH_4 oxidation enzymes (Steudler et al., 1989), however, the inhibitory effect was not observed by Castro et al. (1993). Leguminous tree plantations supply nitrogen in the form of N-rich litter to the soil, and may alter the soil function as a sink for atmospheric CH_4 .

In this study, we conducted the field experiments combining N application and FH layer removal treatments (1) to evaluate the effect of the increased nitrogen availability on N₂O and CH₄ emissions, (2) to estimate involved mechanisms controlling N₂O emissions and (3) to quantify the contribution of FH layer to the N₂O and CH₄ emissions in an *A. mangium* plantation.

2. MATERIAL AND METHODS

2.1. Area Description

The field experiment was conducted in August 2005 at an 8-year-old A. mangium plantation (3°52'40''S, 103°58'40''E) in South Sumatra, Indonesia. The experiment site locates within the large scale (about 1930 km²) plantation of A. mangium. In the stand, trees were planted with 2×4 -m intervals in 1997, and 85 g of phosphate fertilizer (SP-36) and 35 g of urea per tree were applied once on planting. The mean annual temperature and precipitation from 1991 to 2002 were 27.3°C and 2,750 mm, respectively (Hardjono et al., 2005). The period from June to September is relatively dry (average monthly precipitation < 150 mm (Hardjono et al., 2005)), and this study was conducted during the relatively dry season. The topography is undulating and the soils are Acrisols (ISSS Working Group RB, 1998) derived from Tertiary sedimentary rock.

2.2. Experimental design

Fifteen plots for replication were set up about 20m apart from each other within a $60m \times 100m$ area including different topographical elements. We established 9 plots at the upper plateau and 6 plots at the slope and valley bottom. At each plot, 8 square $(0.8 \text{m} \times 0.8 \text{m})$ subplots were established >0.1 m apart from each other and a 4×2 factorial design was imposed with N application and FH removal as factors. At a half of the subplots, the FH layer was remained (hearafter, +FH) and at the other half subplots, the FH layer was entirely removed in the subplot (hereafter, -FH). At +FH subplots, we prepared the control (control +FH), application of distilled water (water +FH), ammonium chloride (NH_4Cl) $(NH_4^+ + FH)$ and potassium nitrate (KNO_3) $(NO_3^- + FH)$, respectively. Also, at -FH subplots, we prepared the control (control -FH), application of distilled water (water -FH), NH₄Cl (NH₄⁺ -FH) and KNO₃ (NO₃⁻-FH), respectively.

N application treatment was conducted on 9 August (0 day) and FH removal treatment on 10 August (+1day). Before N application, L layer was removed temporarily from each subplot except for control subplots in order to apply N onto the FH layer. Two L of distilled water or 100 kg N ha⁻¹ of NH₄Cl or KNO₃ dissolved in 2 L of distilled water were applied evenly on top of the FH layer with a watering can at the water, NH_4^+ and NO_3^- subplots, respectively. Two L of water was equivalent to 3 mm of rain. After the application, we returned the L layer evenly to the each subplot treated. One day after the application (+1day), we removed FH layers from all subplots for the FH removal experiments no later than 1 hour before gas sampling. We removed L layer temporarily before the FH removal treatment and returned them evenly after the FH removal in order to minimize soil drying.

2.3. Gas and soil sampling and analyses

We measured N_2O and CH_4 fluxes using the static chamber method (Ishizuka et al., 2002; Konda et al., 2008) 1 day before the N application (-1 day) at control subplots and 1 day and 3 day after the application (+1day and +3day, respectively) at all subplots. Polypropylene chambers (22.2 cm upper diameter, 18.7 cm lower diameter, and 12.0 cm high) were inserted into the soil to a depth of 2 cm by 1 day before gas sampling. We inserted an extra chamber adjacent to each control +FH subplot and removed FH layers from the inside of chambers on -1day to determine gas fluxes from control -FH subplots on -1day. We took gas samples 0, 15 and 30 min after covering a chamber with a lid. The gas concentration was determined by two gas chromatographs (GC-14B-ECD and GC-14B-FID, Shimadzu Co. Ltd., Kyoto, Japan). We calculated the gas flux by linear regression because the increase in gas concentration in the chamber during this sampling period appeared linear. We calculated CH₄ fluxes as uptake rates. The methods of gas sampling and analysis are detailed in Konda et al. (2008). Total N₂O and CH₄ emissions from 0day to +3day were calculated by summing the daily fluxes within each subplot, assuming the fluxes on 0day to be the same as those on -1day and the flux on +2day by a linear change in emissions between +1day and +3day.

Litter and soil samples were collected after gas sampling on -1day, +1day and +3day. We collected FH layer samples from 0.059 m^2 area at each control +FH subplot on -1 day (n=15), and from 0.030 m^2 area at every + FH subplot on +1day and +3day (n=60 everyday). We took top 10 cm mineral soil at each control +FH subplot (n=15) using two 200-ml (5.1 cm in diameter, 10 cm in height) sampling cylinders on -1day, and at every subplot using one sampling cylinder on +1day and +3day (n=120 everyday). One cylinder soil sample (200 ml) on -1day was used for analyses of bulk density, expressed in an oven-dry basis (105 °C, 24 h). We used the bulk density at control subplots as those at other subplots in each replication plot. FH layer sample of each day and the soil samples of +1day and +3day in addition to another cylinder soil sample of -1day were homogenized and stored in a refrigerator at 4°C. Gravimetric water content of FH layer and soil samples were determined after drying subsamples at 105 °C for 24 h. We calculated water-filled pore space (WFPS) of soil using gravimetric moisture, bulk density and particle density (2.58 Mg m⁻³) determined by a pycnometer. Inorganic ammonium (NH₄-N) and nitrate (NO₃-N) were extracted with 10-fold 2M KCl for 7 g and 5 g of FH layer and soil samples by shaking for 1 h, respectively. The filtrate was stored in a freezer, and determined for NH₄-N and NO₃-N concentrations using a flow-injection analyzer (AQUA LAB Co., Ltd., Tokyo, Japan).

3. RESULTS AND DISCUSSION

FH removal treatment had no significant effect on soil properties at control and every application subplot. Water content of FH layers increased significantly about 20-30% after water and NO₃⁻

application on +1day, while WFPS of soils did not change after every application with and without FH layer (Table 1). NH₄-N and NO₃-N contents in the FH layers and soils increased significantly after NH_4^+ and NO_3^- applications, respectively (Table 1).

Lowercase superscript letters represent significant differences in the FH layer and soil properties among 4 subplots on each day at P value <0.05 level. Water content of FH layer and soil was expressed by gravimetric water content and WFPS, respectively.

Increase of NH₄-N contents after NH₄⁺ application at +FH subplots was 1.45 and 4.26 g N m⁻² in FH layer and soil, respectively, and increase of NO₃-N contents after the application at +FH subplots was 1.29 and 6.22 g N m⁻² in FH layer and soil, respectively (calculated by subtracting NH₄-N and NO₃-N contents at water subplot from that at NH₄⁺ and NO₃⁻ subplot in Table 1). These results mean 43 % and 25 % of applied NH₄⁺ and NO₃⁻ were lost by 1 day after the applications.

 N_2O fluxes at control +FH in this study, 0.46 – 0.53 mg N m⁻² d⁻¹, were consistent with the previous fluxes in *A. mangium* plantation soils during the relatively dry season (Arai et al., 2008; Konda et al., 2008). The fluxes were higher than 0.22 (±0.18) mg N m⁻² d⁻¹ observed by Ishizuka et al. (2005) in the natural forest soils in Jambi, Sumatra, Indonesia, during a relatively dry season.

 NH_4^+ addition significantly reduced CH_4 uptake rates on +3 day in both +FH and -FH subplots (ANOVA, *P*<0.05), though the reduction effect was not consistent on + 1day (Fig. 1). Inhibition of CH_4 uptake by NH_4^+ application was consistent with many previous studies (Steudler et al., 1989; King and Schnell, 1994). *A. mangium* trees supply N-rich litter to the overall soil surface. Hence, we suspected that high NH₄⁺ supply to the *A. mangium* soils through Nrich litter decomposition could weaken the soil CH₄ sink compared to other tropical forest soil. However, CH₄ uptake rates at our control subplots, 0.70 - 0.84mg C m⁻² d⁻¹, were comparable to $0.70 (\pm 0.35)$ mg C m⁻² d⁻¹ in the natural forest soils in the adjacent province on Sumatra during the relatively dry season (Ishizuka et al., 2005) and 0.63 - 1.22 mg C m⁻² d⁻¹ in the tropical rain forest soils in Australia (Kiese et al., 2003). Therefore, *A. mangium* plantations do not appear to decrease the function as a sink for atmospheric CH₄ in a relatively dry season.

FH layer removal did not change N₂O fluxes at control subplots significantly. This result indicated that FH layers in A. mangium plantation soils were not a direct source of N₂O at least in the drier season, or we could not detect inherently very low N₂O emissions from FH layers because of large spatial variability of the fluxes. This result contrasts with previous studies (Dong et al., 1998; Tietema et al., 2007), indicating that litter layers produced N₂O. On the other hand, at 10 NO_3^- subplots out of 15, 3 days' total N₂O emissions after NO₃⁻ application were lower in -FH subplots than in +FH subplots (P = 0.082, paired t-test). This result indicates that NO₃⁻ application possibly promoted denitrification and resulting N₂O emissions in the FH layers. Thus we consider that FH layers of A. mangim plantation may have potential for denitrification, though the reactions to the NO_3^- application were not consistent among 15 sites. CH₄ uptake was not observed in FH layers and suggested that it was mainly related to the mineral soil rather than in the surface litter layer (Tang et al., 2006).

Table 1. Mean (SD) of water content, NH₄-N and NO₃-N contents of FH layer and soil at each subplot with and without FH removal before and after the treatments.

		water content / WFPS (%)			NH4-N (g m ⁻²)			$NO_{3}-N (g m^{2})$		
	Subplot	- 1day	+ 1day	+ 3day	- 1day	+ 1day	+ 3day	-1day	+lda y	+3da y
FH	+FH									
laye	Control	169	123 ^b	98	0.19	0.20 ^ъ	0.33 ^ъ	0.05	0.04 ^ъ	0.08 ^b
r	Water	-	152 °	117	-	0.20 ^ъ	0.30 ^ъ	-	0.06 ^ъ	0.08 ^b
	NH₄⁺	-	151 ^{ab}	127	-	1.65°	1.93 °	-	0.06 ^ъ	0.06 ^ъ
	NO ₃	-	164 ^a	129	-	0.29 ^ъ	0.44 ^ъ	-	1.35 °	1.70 ª
Soil	+FH									
	Control	56.1	57.4	54.9 ^b	2.24	3.06 ^b	2.64 [°]	1.38	1.84 ъс	1.81 ^b
	Water	-	61.1	58.9 ^{ab}	-	3.00 ^ъ	2.68 ^b	-	1.74 °	2.04 ^b
	NH4 ⁺	-	59.3	60.8°	-	7.26°	6.50 °	-	2.61 ^b	2.87 ^b
	NO ₃	-	58.7	59.2 ^{ab}	-	3.14 ^b	3.06°	-	7.96°	8.27 °
	-FH									
	Control	-	58.0	56.3	-	3.03 ^b	2.74 ^b	-	1.82 ^b	2.05 ^b
	Water	-	57.0	59.6	-	3.14 ^b	3.02 ^ъ	-	1.56 ^b	2.08 ^b
	NH_4^+	-	59.6	58.5	-	6.77 °	8.39 °	-	1.92 °	2.16 ^b
	NO_3	-	59.2	58.4	-	3.12 ^b	2.92 ^b	-	7.68°	6.85°

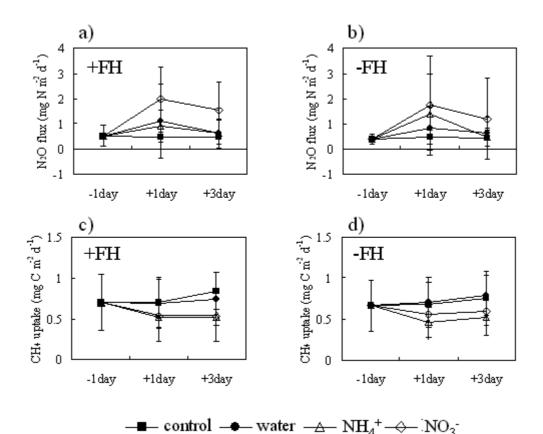


Figure 1. N₂O fluxes (a, b) and CH₄ uptake rates (c, d) on each day in response to N applications with or without FH layer (+FH and –FH, respectively). The fluxes and uptake rates on -1day were determined only at control subplots.

4. CONCLUSION

FH layers in A. mangium plantation were not a direct source of N₂O at least in the drier season, but appear to contribute to nitrogen cycling and following N₂O emissions in the soils as a supplier of available carbon and nitrogen into the soils. Because NO_3^{-1} application to the soils significantly increase N₂O fluxes, increased NO3⁻ availability could enhance denitrification contributing to N₂O emissions in A. mangium plantation soils. Since leguminous tree plantations supply N rich leaf litter to the soil surface, high nitrogen input through litter decomposition might magnify the variation of N_2O fluxes. Whereas NH_4^+ addition significantly reduced CH₄ uptake rates, the contribution of N rich litter of A. mangium do not appear to decrease the function as a sink for atmospheric CH₄ at least in a relatively dry season.

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