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Effect of biogas digested slurry-based biochar on methane flux and methanogenic archaeal diversity in paddy soil

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ABSTRACT

A microcosm-based study was conducted to evaluate the effects of varying concentrations of biogas digested slurry-based biochar on CH_4 flux, associated methanogenic archaeal community, soil chemical and microbial properties, and plant biomass in paddy soil. Our results showed that biochar application significantly increased the CH_4 flux compared to untreated soil, and it increased with the increase in biochar concentration. The NH_4^+-N concentration also increased significantly in biochar treated soils due to more reducible conditions in the soil which prevented NH_4^+-N conversion to NO_3^--N. The application of biochar significantly increased soluble organic carbon, microbial biomass carbon and soluble total nitrogen content of the soils without affecting microbial biomass nitrogen contents. Paddy plant biomass such as shoot weight, panicle numbers and weight of panicles were also higher in biochar treated soils. Denaturing gradient gel electrophoresis revealed all methanogenic groups with the dominance of Methanosaetaceae and Methanocellales groups, whereas no differences were found in methanogenic archaeal diversity among biochar treated and non-treated soils. Thus, the increase in CH_4 flux with the application of varying biochar concentration is possibly due to the variation in the soil variables and plant biomass without affecting methanogenic archaeal diversity.

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1. Introduction

The increase in energy demand has driven the need to consider biofuels as an alternate to fossil fuels (Singla et al., 2012). The increase in biofuel production has simultaneously increased the formation of byproducts (Alotaibi and Schoenau, 2013) such as the digested slurry that remains after biogas production. This slurry contains essential plant nutrients (Sasada et al., 2011). Thus, it can be used as a soil amendment especially after its conversion into digested liquid (DL) and biochar (Singla and Inubushi, 2013, 2014a).

After carbon dioxide (CO_2), methane (CH_4) is a major greenhouse gas (GHG) with almost 34-folds higher global warming potential than CO_2 (IPCC, 2013). The concentration of CH_4 in the atmosphere has increased from the pre-industrial level of 715 ppb to about 1803 ppb in 2011 (IPCC, 2013). Among various sources, rice cultivation is considered as one of the most important anthropogenic sources that accounts for 10–15% of the global CH_4 emission to the atmosphere (Cheng et al., 2008). The global average CH_4 emission from rice cultivation is approximately 60 Tg $CH_4\,yr^{-1}$ which may increase further due to the expansion of rice cultivation for the rising world population (Cheng et al., 2008).

Rice productivity and C storage in paddy soils are the major areas of concern which mainly depends on the field management practices such as tillage operations, crop residue management, soil amendments with desirable nutrients, fertilizer applications and irrigation water, which ultimately affect CH_4 emission (Balesdent et al., 2000). Biochar, which is the product of the thermal degradation of organic materials in the absence of air (pyrolysis), is used as a soil amendment (Lehmann et al., 2011). Many studies have reported the beneficial effects of biochar in improving the soil quality and crop productivity (Ali et al., 2013; Saarnio et al., 2013; Singla and Inubushi, 2013, 2014a; Wang et al., 2012). The renewed interest in biochar comes primarily from its potential role as a long-term C sink in the soil, due to the recalcitrance of its microbial decomposition and chemical degradation (Schneider et al., 2011). There has also been increasing attention to the possibility of mitigating climate change by diverting C into the agricultural soils through the application of biochar (Lehmann et al., 2011). Its effect is found to be more significant for soils with low fertility and/or with low pH value, because of their lower nutrient retention capacity (Lehmann, 2007). Asai et al. (2009) and Wang et al. (2012)

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Table 1
Chemical properties of biogas digested liquid.

pH (H_2O)	EC (H_2O) ($mS\,m^{-1}$)	Total C (%)	DOC (%)	Total N (%)	NH_4^+-N ($\mu g\,ml^{-1}$)	NO_3^--N ($\mu g\,ml^{-1}$)	P_2O_5 ($\mu g\,ml^{-1}$)	K_2O ($mg\,ml^{-1}$)
6.24	12,320	0.71	0.22	1.49	14,900	0.30	11.20	37.40

reported decreased yield of rice by the application of biochar without N-fertilizer, thus indicating that the addition of N-source with biochar seems to be essential for the crop improvement.

Most of the studies suggest that the addition of biochar can increase CH_4 emission (Ali et al., 2013; Knoblauch et al., 2011; Wang et al., 2012; Zhang et al., 2010). However, some of the published reports also state that the application of biochar may decrease CH_4 emission (Feng et al., 2012; Liu et al., 2011) or may not bring any significant change in CH_4 production (Xie et al., 2013). It has also been reported that the range of CH_4 emission in rice paddies after biochar application in a single soil could vary not only with biochar application rates (Zhang et al., 2010) but also with years (Zhang et al., 2013). It appears that the amount of CH_4 emission will depend on the physical and chemical properties of the biochar, the type of the soil, the microbiological circumstances, and the water and fertilizer management (Cai et al., 1997; Zwieten et al., 2009). Many available reports used either single concentration of biochar (Saarnio et al., 2013; Singla and Inubushi, 2014a) or used very high doses of biochar (Feng et al., 2012; Zhang et al., 2010, 2013) under varying concentrations of biochar (C content of biochar: N content of applied N-fertilizer, 15.55–62.26). Moreover, it has been reported that biochar may enhance CH_4 emission even at C content of biochar: N content of N-fertilizer, 5–6 (Ali et al., 2013; Singla and Inubushi, 2014a). Another area of limitation from previous studies is the effects of biochar on soil microbial biomass in paddy fields especially under varying concentrations of biochar. In a recent study, Chen et al. (2013) reported that wheat straw-based biochar significantly increased gene copy numbers of bacterial 16S rRNA (28–64%) and decreased fungal 18S rRNA (35–46%) copy number in a slightly acid paddy soils (Chen et al., 2013).

On the basis of our previous study and the most of other available reports, it was hypothesized that the addition of biochar to the paddy vegetated soils may increase CH_4 emission and it should depend on the amount of biochar used. Microcosm or laboratory model is used to simulate and predict the function of a natural ecosystem and facilitates determination of the ecological role (s) of pivotal factors on the respective process (Lin and Patel, 2008; Grenni et al., 2012). Therefore, a microcosm experiment with rice (*Oryza sativa* var. Koshihikari) was conducted to evaluate the effects of varying concentrations of biochar on (a) CH_4 flux and plant biomass, (b) soil chemical and microbial properties, (c) any changes in methanogenic archaeal community. Thus, results of the present study may strengthen the knowledge for effects of varying concentrations of biochar on not only CH_4 flux, plant biomass, soil chemical properties but also on the soil microbial properties.

2. Materials and methods

2.1. Experimental design

The experiment was conducted at the soil science experimental field, Graduate School of Horticulture, Chiba University, Matsudo, Japan. The soil was collected from the paddy field of Kujukuri, Chiba, Japan and had following physico-chemical properties: texture: sandy; sand 97.3%; silt 2.7%; and clay <0.01% (Singla and Inubushi, 2013); pH (H_2O) (soil:water, 1:2.5) 6.6; electrical conductivity (EC) (H_2O) (soil:water, 1:5) 21.4 $mS\,m^{-1}$; total C (TC) 1.19%; total N (TN) 0.11%; ammonium-N (NH_4^+-N) 6.5 $\mu g\,g^{-1}$ dry soil (ds); and nitrate-N (NO_3^--N) 5.4 $\mu g\,g^{-1}$ ds (Singla et al., 2013). Biochar and digested liquid (DL) were obtained from Yamada Biomass Plant, Chiba, Japan. The production procedure and pyrolysis temperature for biochar production was kept similar with our previous reports (Singla and Inubushi, 2013, 2014a) and it was prepared by the carbonization of solid portion of digested slurry which is left after biogas production. The carbonization temperature was kept at around 330 °C which was followed by an output process temperature at around 370 °C Singla et al., 2014c. The carbonization was done for 6–7 min. Separated liquid of digested slurry was distilled to remove excess water and it formed DL as similar with previous report (Singla et al., 2013). It was stored at 4 °C until the application. The chemical properties of the DL and biochar are mentioned in Table 1 and Table 2, respectively.

Plastic containers (42 cm length × 32 cm width × 30 cm depth) were filled with about 32 kg soil to make a soil depth of 20 cm. Four treatments in triplicate were as follows: (1) control: DL as N source (110 mL $container^{-1}$) at 120 kg N ha^{-1}; (2) biochar low (BL): DL same as control + biochar (7.72 g $container^{-1}$) at 180 kg C ha^{-1} (C/N ratio 1.5); (3) biochar medium (BM): DL same as control + biochar (15.44 g $container^{-1}$) at 360 kg C ha^{-1} (C/N ratio 3.0); (4) biochar high (BH): DL same as control + biochar (30.88 g $container^{-1}$) at 720 kg C ha^{-1} (C/N ratio 6.0). Calcium superphosphate was used as P source to make a total concentration of P at 120 kg P ha^{-1}in each treated soil except for no external application of P in BH treated soils (Table 2). The whole quantity of biochar was applied as basal dose whereas DL was applied in 3 split doses: basal dose as 40% of the total N application, 2nd dose as 40% of the total N application 46 days after transplantation (DAT), 3rd dose as 20% of the total N application 82 DAT. The basal dose of each treatment was applied 2 days prior to the seedling transplantation. After 2 days of basal application, each container was flooded with water and then a total of 12 rice seedlings (21 days old) in 4 hills, each having 3 seedlings, were transplanted to each container (Singla and Inubushi, 2014b) and it was considered as 0 DAT. All the containers were kept under continuous flooded conditions, and drained one week prior to the harvest.

2.2. Gas sampling and analysis

The gas samples from each container at 6–10 days intervals were collected by closed chamber method using a chamber (height 100 cm; and diameter 17.4 cm) (Singla and Inubushi, 2014a,b). To avoid the soil surface disturbances in containers, a chamber base

Table 2
Chemical properties of biogas digested slurry based biochar.

Parameters	Value	Parameters ($mg\,kg^{-1}$)	Value
pH (H_2O)	8.81	Cd	0.4
EC (H_2O) ($mS\,m^{-1}$)	386	Cr	3.7
Total C (%)	31.72	Cu	76
H (%)	2.5	Pb	2.5
H/C ratio	0.08	Hg	0.1
SOC ($mg\,g^{-1}$)	20	Mo	0.02
Total N (%)	3.4	Ni	2.1
NH_4^+-N ($\mu g\,g^{-1}$)	19.90	Zn	317
NO_3^--N ($\mu g\,g^{-1}$)	2.20	B	5
P_2O_5 (%)	6.4	Cl	10
K_2O (%)	2.2	Na	25
Total Mg (%)	2.2	As	0.5

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