Methane Production Potential of Soil Profile in Organic Paddy Field

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Abstract

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The use of organic fertilizers in the organic paddy/rice field can increase methane (CH₄) production, which leads to environmental problems. In this study, we aimed to determine the CH₄ production potential (CH₄-PP) by a soil profile from samples using flood incubation. Soil properties (chemical, physical, and biological) were analyzed from soil samples of three different paddy farming systems (organic, semi-organic, and conventional), whilst soil from teak forest was used as the control. A significant relationship was determined between soil properties and CH₄-PP. The average amount of CH₄-PP in the organic rice field profile was the highest among all the samples (1.36 μ g CH₄/kg soil/day). However, the CH₄ oxidation potential (CH₄-OP) is high as well, as this was a chance of mitigation options should focus on increasing the methanotrophic activity which might reduce CH_4 emissions to the atmosphere. The factor most influencing CH_4 -PP is soil C-organic (C_{org}). C_{org} and CH_4 -PP of the top soil of organic rice fields were 2.09% and 1.81 µg CH₄/kg soil/day, respectively. As a consequence, here the mitigation options require more efforts than in the other farming systems. Soil with various amounts of C_{org} reached a maximum point of CH_4 -PP at various time after incubation (20, 15, and 10 days for the highest, $medium, and the lowest amounts of \ C_{org}, respectively). \ A \ high amount of \ C_{org} \ provided \ enough \ C \ substrate for \ constant \ constant$ producing a higher amount of CH₄ and reaching its longer peak production than the low amount of C_{org}. These findings also provide guidance that mitigation option reduces $\mathrm{CH_4}$ emissions from organic rice fields and leads to drainage every10-20 days before reaching the maximum CH₄-PP.

Keywords: emission; horizon; methane; mitigation; soil

As described by Mujiyo et al. (2016), a majority of paddy fields in Indonesia have a low level of organic matter (Sanchez 1976; Karama 2001; Syamsiyah & Mujiyo 2006), which affects the levelling off of productivity. Indonesian farmers have overcome these problems by implementing organic farming, which focuses on the use of organic fertilizers. However, it appears to increase the methane production (LE Mer & Roger 2001; Nieder & Benbi 2008), by providing C sources and decreasing the amount of oxidation-reduction potential (Eh) (Hou et al. 2000;

LI 2007). Methane is one of the greenhouse gases (GHG) contributing by around 16% to the total GHG production (IPPC 2014). Global warming potential (GWP) from the presence of CH $_4$ is 21 times greater than from CO $_2$ (NIEDER & BENBI 2008). IPPC (2013) stated that the GWP value of CH $_4$ is 28 times higher than that of CO $_2$.

The U.S. Environmental Protection Agency (USEPA 2006) noted that the projection of increasing methane emissions is primarily attributed to the increasing demand for rice, due to the rapid population growth in

rice-consuming countries. Ca. 78% of methane emissions produced in Indonesia, China, India, Thailand, Vietnam, and Myanmar stem from rice cultivation. Therefore, there is a need to do an assessment on GHG emissions from agricultural land with a high potential of methane emissions. The present study is a potential investigation, especially to determine the most appropriate mitigation options based on key factors.

Methane emissions are the result of $\operatorname{CH_4}$ cumulation from bottom-to-top horizon with the soil profile. It can reach the surface and eventually be emitted to the atmosphere. This study aims to determine the potential of each soil horizon to produce and oxidize $\operatorname{CH_4}$. The $\operatorname{CH_4}$ production potential ($\operatorname{CH_4}$ -PP) and $\operatorname{CH_4}$ oxidation potential ($\operatorname{CH_4}$ -OP) were determined by measuring the amount of $\operatorname{CH_4}$ production from soil samples with a flood incubation treatment in the laboratory. The results will be useful in order to find mitigation options for $\operatorname{CH_4}$ emissions, especially in organic paddy fields.

MATERIAL AND METHODS

Soil samples. The horizon was determined based on the Soil Survey Staff (2014). The soil was taken from the horizon in the soil profile of organic (P1), semi-organic (P2), conventional (P3) paddy field, and the teak forest land-use (P4) as a control. Soil samples were air-dried and sieved for diameter 0.5 mm and 2 mm. All soil samples were collected from soil profiles of organic paddy fields in Sukorejo, Sambirejo District, Sragen Regency, in Central Java. The sample organic rice fields have been certified organic by INOFICE (2008) as a producer of organic food (organic rice). The criterion for organic rice field selection is the use of organic fertilizers, for semiorganic the use of chemical and organic fertilizers, for conventional the use of most types of fertilizers (including both chemical and only occasionally organic fertilizers), and for the teak forest as a control it is the land-use type of teak forest.

Soil properties. The soil properties were analyzed by texture (pipette method), H_2O pH (soil: water = 1:2.5; pH meter), C-organic (1 N $K_2Cr_2O_7$ oxidation), total of N (concentrated H_2SO_4 destruction), C/N ratio, available P (0.5 M NaHCO $_3$ extraction), total of P (HCl 25% extraction), available K (1 M NH $_4OAc$ percolation), total of K (HCl 25% extraction), cation exchange capacity (percolation 1 M NH $_4OAc$ and NaCl 10%), and base saturation (1 M NH $_4OAc$

percolation) (EVIATI & SULAEMAN 2009). Microbial biomass C was observed using the CHCl $_3$ fumigation method, with 0.5 M K $_2$ SO $_4$ extraction after 0.05 M K $_2$ SO $_4$ pre-extraction (MUELLER *et al.* 1992). The difference between fumigated and non-fumigated C was the number of microbial biomass C.

 CH_a -PP and CH_a -OP. CH_a -PP was measured by incubating soil samples in the laboratory, then the gas produced was analyzed by a gas chromatograph (Shimadzu GC-14A; Shimadzu Corporation, Kyoto, Japan) equipped with a flame ionization detector (FID). Twenty grams of 0.5 mm diameter soil samples were air-dried, then put into a glass incubator, and added with 40 ml of distilled water, mixed gently, and then put into the incubator at 30°C. The gas production protocol was adopted from Susilowati (2007) with the modification of adding acetylene as an inhibitor of methane oxidation (WATANABE et al. 1995, 1997; CHAN & PARKIN 2000). Fifty ml of acetylene was added into each 1000 ml of air in the headspace room of the glass incubator (WATANABE et al. 1995). Gas samples were measured at hour 0 (C_0) and hour 24 (C₂₄). The measurements were taken eight times, at five-day intervals.

After getting the concentration at times C_0 and C_{24} , the gas production (in $\mu g\ CH_4/kg\ soil/day$) was calculated using the following formula:

$$CH_4 \text{ production} = (C_{24} - C_0) \times \frac{Vhs}{WS} \times \frac{MW}{Vm} \times \frac{Tst}{(Tst + T)}$$

where:

C₀ – CH₄ concentration at hour 0 (ppm)

C₂₄ - CH₄ concentration at hour 24 (ppm)

Vhs - headspace volume (ml)

WS - weight of soil sample (g)

MW - molecular weight of CH₄ (16.123 g)

Vm $- CH_4$ volume at standard conditions (273.2 K) = 22.41 l

Tst - temperature standard conditions (273.2 K)

T – air temperature of incubation (°C)

The measurement of $\mathrm{CH_4}$ -OP used the same procedure as the measurement of $\mathrm{CH_4}$ -PP above, excluding the addition of acetylene into the samples (Watanabe *et al.* 1995, 1997; Chan & Parkin 2000). The difference between $\mathrm{CH_4}$ production with and without the inhibitor is the amount of $\mathrm{CH_4}$ oxidized ($\mathrm{CH_4}$ -OP).

Statistical analysis. The correlations between soil properties, CH₄-PP, and CH₄-OP were determined using the correlation analysis (STEEL & TORIE 1980), the Pearson's correlation coefficient was calculated as well. The differences between soil properties,

 $\mathrm{CH_4} ext{-PP}$, and $\mathrm{CH_4} ext{-OP}$ between all horizon depths were determined using one-way analysis of variance (ANOVA) with the Duncan's Multiple Range test. All statistical analyses were performed with SPSS Statistics 17.0 software.

RESULTS AND DISCUSSION

 $\mathrm{CH_4} ext{-}\mathrm{PP}$ and $\mathrm{CH_4} ext{-}\mathrm{OP}$. The description of the soil profile shows that P1 consists of four horizons, P2 consists of four horizons, P3 consists of five horizons, and P4 consists of six horizons. Table 1 shows the horizons profile, selected soil properties, $\mathrm{CH_4} ext{-}\mathrm{PP}$, and $\mathrm{CH_4} ext{-}\mathrm{OP}$. The horizons of P1 profile (organic) have the highest average $\mathrm{CH_4} ext{-}\mathrm{PP}$ and $\mathrm{CH_4} ext{-}\mathrm{OP}$ followed by P2 (semi-organic), P4 (teak forest), and P3 (conventional).

From the result, CH_4 -PP and CH_4 -OP have a significant correlation (r = 0.92, P < 0.001, n = 19). It

means that the soil has a high activity of methanogens (CH₄ production) and methanotrophs (CH₄ oxidation). This correlation is quite obvious as the first group (methanogens) produces a substrate (CH₄), which is used by the second group (methanotrophs) (Joulian et al. 1997). Brzezińska et al. (2012) also found that if the soil has a high level of CH₄ production, it also has a high level of CH₄ consumption (oxidation). Soil that has a high potential of CH₄ production will not necessarily release high emissions of СН₄. ZHU et al. (2012) stated that around 50% of the methane produced in wetlands is consumed before it reaches the atmosphere. In the laboratory condition, less than a half of the CH₄ was oxidized in the soil, while almost all of it was oxidized around a root area (ZANG et al. 2013). By employing the methanothroph bacteria through some oxidation mechanisms, CH₄ could be converted into CO₂ (NIEDER & BENBI 2008; THAURER et al. 2008). CH₄ is oxidized by O₂ into

Table 1. Horizons P1–P4, selected soil properties, methane (CH_4) production potential (CH_4 -PP), and oxidation potential (CH_4 -OP)

Profile	Horizon (notation)	Soil depth (cm)	Epipedon Endopedon soil classification*	Organic C	Mean	Microbial biomass C	Mean	CH ₄ -PP	Mean	CH ₄ -OP	Mean
				(%)		(µg/g soil)		(μg CH ₄ /kg soil/day))
P1	I (Apg)	0-18/22	Umbric Argillic/Sombric Umbric Epiaqualf	2.09	1.67	498.39	277.91	1.81	1.36	0.73	0.61
	II (Bt)	18/22-52/58		2.00		418.01		1.67		0.66	
	III (Bw1)	52/58-78/100		1.31		84.98		1.05		0.53	
	IV(Bw2)	> 150		1.30		110.24		0.92		0.51	
P2	I (Apg)	0-19/22	Umbric Cambic Humic Epiaquept	1.78	1.62	475.42	292.26	1.23	0.92	0.52	0.42
	II (Bw1)	19/22-46/62		1.65		199.82		0.98		0.48	
	III (Bw2)	46/62-91/108		1.56		172.26		0.82		0.39	
	IV (Bw3)	> 150		1.48		321.54		0.66		0.31	
Р3	I (Apg)	0-27/32	Umbric Cambic Humic Epiaquept	1.08	0.91	367.48	291.23	0.84		0.43	0.34
	II (Bw1)	27/32-56/75		1.00		257.23		0.59	0.65	0.25	
	III (Bw2)	56/75-75/96		1.01		303.17		0.63		0.37	
	IV (Bw3)	75/96-102/108		0.68		261.83		0.66		0.34	
	V (Bw4)	> 150		0.78		266.42		0.53		0.28	
P4	I (Ap)	0-23/33	Typic Humadept	1.83	1.58	468.53	324.99	1.21	0.85	0.51	0.40
	II (Bw1)	23/33-50/70		1.88		443.27		1.12		0.47	
	III (Bw2)	50/70-80/95		1.63		271.02		0.80		0.36	
	IV (Bw3)	80/95-100/123		1.52		282.50		0.67		0.22	
	V (Bw4)	100/123-143/168		1.35		197.52		0.73		0.46	
	VI (Bw5)	143/168-180		1.24		287.09		0.61		0.35	

P1 – organic; P2 – semi-organic; P3 – conventional; P4 – teak forest; *soil classification according to Soil Survey Staff (2014); Sub group, data not shown

 $\rm CO_2 + H_2O$ (Nieder & Benbi 2008), by $\rm NO_3^-$ into $\rm N_2 + \rm CO_2$, and by $\rm SO_4^{2-}$ into $\rm CO_2 + H_2S$ (Thauer *et al.* 2008).

 ${
m CH_4}$ -PP and soil ${
m C}_{
m org}$. The soil property which has the closest relationship with ${
m CH_4}$ -PP is C-organic (${
m C}_{
m org}$) (r=0.80, P<0.001, n=19) (see Figure 1, left). When the ${
m C}_{
m org}$ in soil increases, the potential ${
m CH_4}$ production goes up. These results are in line with those obtained by Joulian *et al.* (1996), Oelbermann and Schiff (2008), and Liu *et al.* (2011) that ${
m CH_4}$ -PP is strongly influenced by ${
m C}_{
m org}$. Higher ${
m C}_{
m org}$ leads to the higher amount of ${
m CH_4}$ -PP.

The decomposition of organic matter through the oxidation-reduction process is terminated by the formation of CO_2 and CH_4 : $C_6H_{12}O_6 \rightarrow 3$ $CO_2 + 3$ CH_4 (Le Mer & Roger 2001; Nieder & Benbi 2008). Sanchez (1976) stated that the decompositions of organic matter in the flooded and unflooded soil are the same until the stage of the pyruvic acid formation. The pyruvic acid and the other intermediate products formed in unflooded soil further oxidize to the final product of CO_2 , NO_3^- , SO_4^{2-} and resistant humic material, and the pyruvic acid formed in flooded soil with anaerobic conditions is further reduced to alcohol, organic acids, etc., and finally to the final product of CO_2 and CH_4 .

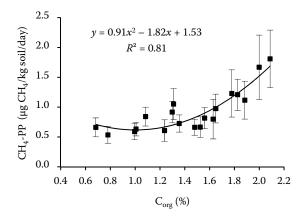
The group of methanogens in the soil can produce $\mathrm{CH_4}$ from either the reduction of $\mathrm{CO_2}$ and $\mathrm{H_2}$ into $\mathrm{CH_4}$, the fermentation of $\mathrm{CH_3COOH}$ (acetic acid) into $\mathrm{CH_4}$ and $\mathrm{CO_2}$ (Nieder & Benbi 2008; Thaurer et~al.~2008), or from the reduction of $\mathrm{CH_3OH}$ (methanol) into $\mathrm{CH_4}$ (Reddy & Delaune 2008). Figure 1 (left) shows that the $\mathrm{CH_4}$ -PP has a quadratic function with $\mathrm{C_{org}}$. When $\mathrm{C_{org}}$ increases, the $\mathrm{CH_4}$ -PP significantly rises, which is defined by the equation Y function ($\mathrm{CH_4}$ -PP) = $0.91x^2-1.82x$

+ 1.53 (R^2 = 0.81). Brzezińska *et al.* (2012) proved that there is a significant correlation between $C_{\rm org}$ and ${\rm CH_4}$ production through the equation model $Y=0.127x^{5.07}$ and $R^2=0.90$.

P1 has the highest average of $C_{\rm org}$ content (1.67%) and CH_4 -PP (1.36 µg CH_4 /kg soil/day) if compared to other soil profiles (see Table 1). $C_{\rm org}$ content in the soil profiles of organic fields, especially in horizons I and II (up to depths of ca. 50 cm), is assessed into moderate category (2–3%) and is significantly higher than in other soil profiles of the same horizon that are categorized as "low" and "very low" (< 2%). The organic rice system practices since 2001, with the average use of 6 t of manure during each planting season, are strongly suspected as the primary factor causing higher amounts of $C_{\rm org}$ in organic fields if compared to semi-organic, conventional, and teak forest areas.

 ${
m CH}_4$ -PP and microbial biomass C. The populations of methanogens and methanotrophs were observed by analyzing the microbial biomass C to describe the microbial population in the soil. The correlation between microbial biomass C and C_{org} is significant (r=0.52, P<0.05, n=19). Figure 1 (right) shows the quadratic regression of the equation ($R^2=0.53$). The higher the C_{org} content in the soil, the higher the microbial population observed. This result is supported by a previous research by DALAL *et al.* (2011).

Microbial biomass C correlates significantly with CH_4 -PP (r = 0.56, P < 0.05, n = 19), but not with CH_4 -OP (r = 0.35, P > 0.05, n = 19). Even though the correlation between microbial biomass C and CH_4 -OP is not significant, it has a tendency of increasing in the amount of microbial biomass C that is also followed by rising amounts of CH_4 -OP. The soil profile with a high population of microbial biomass C has



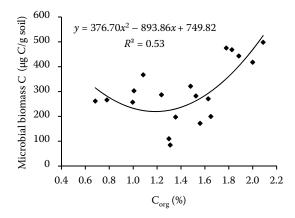


Figure 1. The line relationship of C_{org} with CH_4 production potential $(CH_4$ -PP) (left) and microbial biomass C (right)

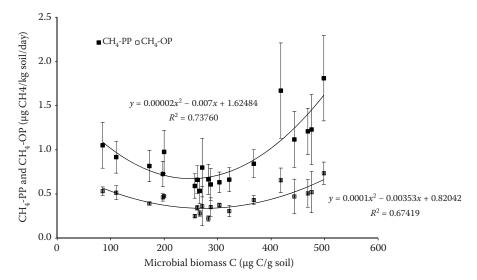


Figure 2. Regression lines of microbial biomass C with CH_4 production potential (CH_4 -PP) and CH_4 oxidation potential (CH_4 -OP)

high $\mathrm{CH_4}$ -PP and $\mathrm{CH4}$ -OP as well. These correlations are presented in quadratic regressions with the value of $R^2 = 0.74$ and 0.67 respectively (see Figure 2). Five soil horizons of P3 contain a low amount of $\mathrm{C}_{\mathrm{org}}$ and a lower amount of microbial biomass C (Figure 1, right). This condition caused the amount of $\mathrm{CH_4}$ -PP to decrease (Figure 2). However, it can be assumed that the increasing amount of microbial biomass C is followed by the number of microbes that produce and oxidize $\mathrm{CH_4}$. Dar *et al.* (2008) argue that methanogens have the range of 10% of the total microbes in the laboratory scale bioreactor experiments.

 ${
m CH_4}$ -PP and soil depth. There was no significant difference between CH₄-PP and C_{org} between all horizon depths observed (one-way ANOVA F=2.225, P>0.05; one-way ANOVA F=1.159, P>0.05, re-

spectively). However, there was a significant variation in microbial biomass C (one-way ANOVA F = 3.587, P < 0.05). Microbial biomass C in horizon I was the highest among all the other samples. Although there was no significant difference in CH₄-PP and C_{org} between all horizon depths analyzed, microbial biomass C correlates significantly with C_{org} (r = 0.52, P < 0.05, n = 19) and CH₄-PP (r = 0.56, P < 0.05, n = 19), as C_{org} correlates significantly with CH_4 -PP (r = 0.80, P < 0.001, n = 19). The significant variation in microbial biomass C between the horizon depths and the three correlations shows that there is a strong presumption significantly affecting the depth of horizon for CH₄-PP through its influence on the C_{org} and microbial biomass C. In the upper horizon, C_{org} and microbial biomass C were high as it would increase the CH₄-PP.

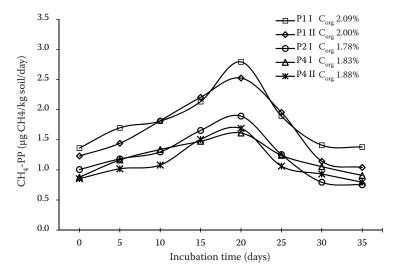


Figure 3. ${
m CH_4}$ production potential (${
m CH_4}$ -PP) patterns of soil with high ${
m C}_{
m org}$

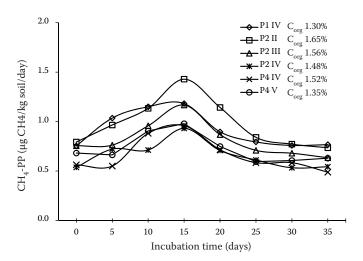


Figure 4. $\rm CH_4$ production potential (CH $_4$ -PP) patterns of soil with moderate $\rm C_{org}$

Whalen and Reeburg (2000) and Brzezińska et al. (2012) investigated the correlation between soil depth, $C_{\rm org}$, and CH_4 -PP. $C_{\rm org}$ content in the upper horizon was higher than in the lower horizon, so as the CH_4 -PP. Freitag et al. (2010) concluded that the depth of the soil is one of the factors that control the amount of CH_4 emissions.

 $\rm CH_4$ -PP and incubation time. The level of $\rm C_{org}$ determines both $\rm CH_4$ -PP and $\rm CH_4$ production patterns based on the time of flood incubation. The soil with high $\rm C_{org}$ (P1 horizon I and II, P2 horizon I, and P4 horizon I and II), moderate $\rm C_{org}$ (P1 horizon IV, P2 horizon II, III, and IV, and P4 horizon IV and V), and low $\rm C_{org}$ (P3 horizon II, III, IV, and V, and P4 horizon VI) reached the maximum average of $\rm CH_4$ production at various times after incubation, i.e. on day 20 (Figure 3), day 15 (Figure 4), and day 10 (Figure 5), respectively.

Soil with high C $_{\rm org}$ has enough C substrates to produce high amounts of CH $_{\rm 4}$ and it will reach a longer

peak production period. Meanwhile, soil with low $C_{\rm org}$ has less C substrate, produces lower amount of CH_4 , and shows faster peak production periods than the others. These results were strengthened by previous findings by Brzezińska et~al.~(2012) and Yuan et~al.~(2014), which measured CH_4 production after flooding the soil samples. Soil with higher $C_{\rm org}$ experienced a longer peak time of production than soil with lower $C_{\rm org}$.

CONCLUSION

The $\mathrm{CH_4}$ production in soil is reflected in its chemical, physical, and biological properties. This study showed that the $\mathrm{CH_4}$ production potential in organic rice fields is high because of the presence of high amounts of $\mathrm{C_{org}}$. In consequence, the mitigation options need more efforts than required in other cultivation (farming) systems. The $\mathrm{CH_4}$ oxidation

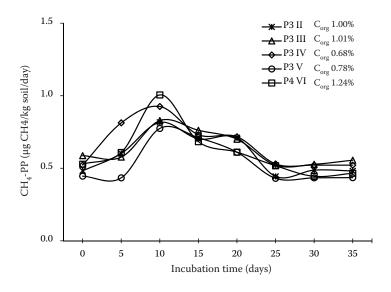


Figure 5. CH_4 production potential (CH_4 -PP) patterns of soil with low C_{org}

potential in organic rice fields is high as well, as the mitigation options should focus on increasing the methanotrophic activity which might reduce $\mathrm{CH_4}$ emissions to the atmosphere. Because $\mathrm{C_{org}}$ has an important role in the production of $\mathrm{CH_4}$, it determines the dynamics of $\mathrm{CH_4}$ -PP and $\mathrm{CH_4}$ -OP. So using particular kinds of organic matter as $\mathrm{C_{org}}$ sources would manage the composition of methanogens and methanotrophs so as to reduce $\mathrm{CH_4}$ -PP and to increase $\mathrm{CH_4}$ -OP.

 $\rm C_{org}$ also affected the pattern of the $\rm CH_4$ production potential by the time of flood incubation. High amounts of $\rm C_{org}$ reached a maximum $\rm CH_4$ -PP on day 20 after incubation, while moderate $\rm C_{org}$ on day 15, and low $\rm C_{org}$ on day 10. Higher amounts of $\rm C_{org}$ provide enough C substrates to produce higher amounts of $\rm CH_4$ and reach longer peak times of production than lower $\rm C_{org}$. This finding provides guidance that mitigation option reduces $\rm CH_4$ emissions from organic rice fields and experiences drainage every 10–20 days before reaching the maximum $\rm CH_4$ -PP.

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