

## 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 (CH<sub>4</sub>) production, which leads to environmental problems. In this study, we aimed to determine the CH<sub>4</sub> production potential (CH<sub>4</sub>-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 CH<sub>4</sub>-PP. The average amount of CH<sub>4</sub>-PP in the organic rice field profile was the highest among all the samples (1.36 µg CH<sub>4</sub>/kg soil/day). However, the CH<sub>4</sub> oxidation potential (CH<sub>4</sub>-OP) is high as well, as this was a chance of mitigation options should focus on increasing the methanotrophic activity which might reduce CH<sub>4</sub> emissions to the atmosphere. The factor most influencing CH<sub>4</sub>-PP is soil C-organic (C<sub>org</sub>). C<sub>org</sub> and CH<sub>4</sub>-PP of the top soil of organic rice fields were 2.09% and 1.81 µg CH<sub>4</sub>/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<sub>org</sub> reached a maximum point of CH<sub>4</sub>-PP at various time after incubation (20, 15, and 10 days for the highest, medium, and the lowest amounts of C<sub>org</sub>, respectively). A high amount of C<sub>org</sub> provided enough C substrate for producing a higher amount of CH<sub>4</sub> and reaching its longer peak production than the low amount of C<sub>org</sub>. These findings also provide guidance that mitigation option reduces CH<sub>4</sub> emissions from organic rice fields and leads to drainage every 10–20 days before reaching the maximum CH<sub>4</sub>-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<sub>4</sub> is 21 times greater than from CO<sub>2</sub> (NIEDER & BENBI 2008). IPPC (2013) stated that the GWP value of CH<sub>4</sub> is 28 times higher than that of CO<sub>2</sub>.

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

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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 CH<sub>4</sub> 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 CH<sub>4</sub>. The CH<sub>4</sub> production potential (CH<sub>4</sub>-PP) and CH<sub>4</sub> oxidation potential (CH<sub>4</sub>-OP) were determined by measuring the amount of CH<sub>4</sub> production from soil samples with a flood incubation treatment in the laboratory. The results will be useful in order to find mitigation options for CH<sub>4</sub> 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 semi-organic 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<sub>2</sub>O pH (soil : water = 1 : 2.5; pH meter), C-organic (1 N K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub> oxidation), total of N (concentrated H<sub>2</sub>SO<sub>4</sub> destruction), C/N ratio, available P (0.5 M NaHCO<sub>3</sub> extraction), total of P (HCl 25% extraction), available K (1 M NH<sub>4</sub>OAc percolation), total of K (HCl 25% extraction), cation exchange capacity (percolation 1 M NH<sub>4</sub>OAc and NaCl 10%), and base saturation (1 M NH<sub>4</sub>OAc

percolation) (EVIATI & SULAEMAN 2009). Microbial biomass C was observed using the CHCl<sub>3</sub> fumigation method, with 0.5 M K<sub>2</sub>SO<sub>4</sub> extraction after 0.05 M K<sub>2</sub>SO<sub>4</sub> pre-extraction (MUELLER *et al.* 1992). The difference between fumigated and non-fumigated C was the number of microbial biomass C.

**CH<sub>4</sub>-PP and CH<sub>4</sub>-OP.** CH<sub>4</sub>-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<sub>0</sub>) and hour 24 (C<sub>24</sub>). The measurements were taken eight times, at five-day intervals.

After getting the concentration at times C<sub>0</sub> and C<sub>24</sub>, the gas production (in µg CH<sub>4</sub>/kg soil/day) was calculated using the following formula:

$$\text{CH}_4 \text{ production} = (C_{24} - C_0) \times \frac{V_{hs}}{WS} \times \frac{MW}{V_m} \times \frac{T_{st}}{(T_{st} + T)}$$

where:

C<sub>0</sub> – CH<sub>4</sub> concentration at hour 0 (ppm)

C<sub>24</sub> – CH<sub>4</sub> concentration at hour 24 (ppm)

V<sub>hs</sub> – headspace volume (ml)

WS – weight of soil sample (g)

MW – molecular weight of CH<sub>4</sub> (16.123 g)

V<sub>m</sub> – CH<sub>4</sub> volume at standard conditions (273.2 K) = 22.41 l

T<sub>st</sub> – temperature standard conditions (273.2 K)

T – air temperature of incubation (°C)

The measurement of CH<sub>4</sub>-OP used the same procedure as the measurement of CH<sub>4</sub>-PP above, excluding the addition of acetylene into the samples (WATANABE *et al.* 1995, 1997; CHAN & PARKIN 2000). The difference between CH<sub>4</sub> production with and without the inhibitor is the amount of CH<sub>4</sub> oxidized (CH<sub>4</sub>-OP).

**Statistical analysis.** The correlations between soil properties, CH<sub>4</sub>-PP, and CH<sub>4</sub>-OP were determined using the correlation analysis (STEEL & TORIE 1980), the Pearson's correlation coefficient was calculated as well. The differences between soil properties,

CH<sub>4</sub>-PP, and CH<sub>4</sub>-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

**CH<sub>4</sub>-PP and CH<sub>4</sub>-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, CH<sub>4</sub>-PP, and CH<sub>4</sub>-OP. The horizons of P1 profile (organic) have the highest average CH<sub>4</sub>-PP and CH<sub>4</sub>-OP followed by P2 (semi-organic), P4 (teak forest), and P3 (conventional).

From the result, CH<sub>4</sub>-PP and CH<sub>4</sub>-OP have a significant correlation ( $r = 0.92$ ,  $P < 0.001$ ,  $n = 19$ ). It

means that the soil has a high activity of methanogens (CH<sub>4</sub> production) and methanotrophs (CH<sub>4</sub> oxidation). This correlation is quite obvious as the first group (methanogens) produces a substrate (CH<sub>4</sub>), which is used by the second group (methanotrophs) (JOU LIAN *et al.* 1997). BRZEZIŃSKA *et al.* (2012) also found that if the soil has a high level of CH<sub>4</sub> production, it also has a high level of CH<sub>4</sub> consumption (oxidation). Soil that has a high potential of CH<sub>4</sub> production will not necessarily release high emissions of CH<sub>4</sub>. 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 CH<sub>4</sub> was oxidized in the soil, while almost all of it was oxidized around a root area (ZANG *et al.* 2013). By employing the methanotroph bacteria through some oxidation mechanisms, CH<sub>4</sub> could be converted into CO<sub>2</sub> (NIEDER & BENBI 2008; THAURER *et al.* 2008). CH<sub>4</sub> is oxidized by O<sub>2</sub> into

Table 1. Horizons P1–P4, selected soil properties, methane (CH<sub>4</sub>) production potential (CH<sub>4</sub>-PP), and oxidation potential (CH<sub>4</sub>-OP)

Profile	Horizon (notation)	Soil depth (cm)	Epipedon Endopedon soil classification*	Organic C	Mean	Microbial biomass C	Mean	CH <sub>4</sub> -PP	Mean	CH <sub>4</sub> -OP	Mean
				(%)		(µg/g soil)		(µg CH <sub>4</sub> /kg soil/day)			
P1	I (Apg)	0–18/22		2.09		498.39		1.81		0.73	
	II (Bt)	18/22–52/58	Umbric Argillic/Sombric	2.00	1.67	418.01	277.91	1.67	1.36	0.66	0.61
	III (Bw1)	52/58–78/100	Umbric Epiaqualf	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		1.78		475.42		1.23		0.52	
	II (Bw1)	19/22–46/62	Umbric Cambic	1.65	1.62	199.82	292.26	0.98	0.92	0.48	0.42
	III (Bw2)	46/62–91/108	Humic Epiaquept	1.56		172.26		0.82		0.39	
	IV (Bw3)	> 150		1.48		321.54		0.66		0.31	
P3	I (Apg)	0–27/32		1.08		367.48		0.84		0.43	
	II (Bw1)	27/32–56/75	Umbric Cambic	1.00		257.23		0.59		0.25	
	III (Bw2)	56/75–75/96	Umbric Cambic	1.01	0.91	303.17	291.23	0.63	0.65	0.37	0.34
	IV (Bw3)	75/96–102/108	Humic Epiaquept	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		1.83		468.53		1.21		0.51	
	II (Bw1)	23/33–50/70		1.88		443.27		1.12		0.47	
	III (Bw2)	50/70–80/95	Umbric Cambic	1.63	1.58	271.02	324.99	0.80	0.85	0.36	0.40
	IV (Bw3)	80/95–100/123	Typic Humudept	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

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$\text{CO}_2 + \text{H}_2\text{O}$  (NIEDER & BENBI 2008), by  $\text{NO}_3^-$  into  $\text{N}_2 + \text{CO}_2$ , and by  $\text{SO}_4^{2-}$  into  $\text{CO}_2 + \text{H}_2\text{S}$  (THAUER *et al.* 2008).

**CH<sub>4</sub>-PP and soil C<sub>org</sub>.** The soil property which has the closest relationship with CH<sub>4</sub>-PP is C-organic (C<sub>org</sub>) ( $r = 0.80$ ,  $P < 0.001$ ,  $n = 19$ ) (see Figure 1, left). When the C<sub>org</sub> in soil increases, the potential CH<sub>4</sub> 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 CH<sub>4</sub>-PP is strongly influenced by C<sub>org</sub>. Higher C<sub>org</sub> leads to the higher amount of CH<sub>4</sub>-PP.

The decomposition of organic matter through the oxidation-reduction process is terminated by the formation of  $\text{CO}_2$  and  $\text{CH}_4$ :  $\text{C}_6\text{H}_{12}\text{O}_6 \rightarrow 3\text{CO}_2 + 3\text{CH}_4$  (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  $\text{CO}_2$ ,  $\text{NO}_3^-$ ,  $\text{SO}_4^{2-}$  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  $\text{CO}_2$  and  $\text{CH}_4$ .

The group of methanogens in the soil can produce  $\text{CH}_4$  from either the reduction of  $\text{CO}_2$  and  $\text{H}_2$  into  $\text{CH}_4$ , the fermentation of  $\text{CH}_3\text{COOH}$  (acetic acid) into  $\text{CH}_4$  and  $\text{CO}_2$  (NIEDER & BENBI 2008; THAUER *et al.* 2008), or from the reduction of  $\text{CH}_3\text{OH}$  (methanol) into  $\text{CH}_4$  (REDDY & DELAUNE 2008). Figure 1 (left) shows that the CH<sub>4</sub>-PP has a quadratic function with C<sub>org</sub>. When C<sub>org</sub> increases, the CH<sub>4</sub>-PP significantly rises, which is defined by the equation Y function  $(\text{CH}_4\text{-PP}) = 0.91x^2 - 1.82x + 1.53$

+ 1.53 ( $R^2 = 0.81$ ). BRZEZIŃSKA *et al.* (2012) proved that there is a significant correlation between C<sub>org</sub> and CH<sub>4</sub> production through the equation model  $Y = 0.127x^{5.07}$  and  $R^2 = 0.90$ .

P1 has the highest average of C<sub>org</sub> content (1.67%) and CH<sub>4</sub>-PP (1.36  $\mu\text{g CH}_4/\text{kg soil/day}$ ) if compared to other soil profiles (see Table 1). C<sub>org</sub> 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<sub>org</sub> in organic fields if compared to semi-organic, conventional, and teak forest areas.

**CH<sub>4</sub>-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<sub>org</sub> 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<sub>org</sub> 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<sub>4</sub>-PP ( $r = 0.56$ ,  $P < 0.05$ ,  $n = 19$ ), but not with CH<sub>4</sub>-OP ( $r = 0.35$ ,  $P > 0.05$ ,  $n = 19$ ). Even though the correlation between microbial biomass C and CH<sub>4</sub>-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<sub>4</sub>-OP. The soil profile with a high population of microbial biomass C has

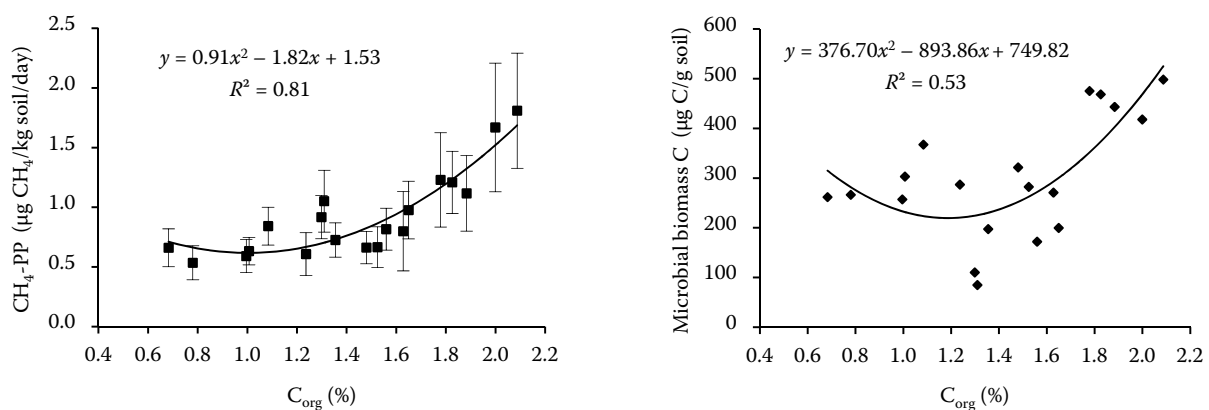


Figure 1. The line relationship of C<sub>org</sub> with CH<sub>4</sub> production potential (CH<sub>4</sub>-PP) (left) and microbial biomass C (right)

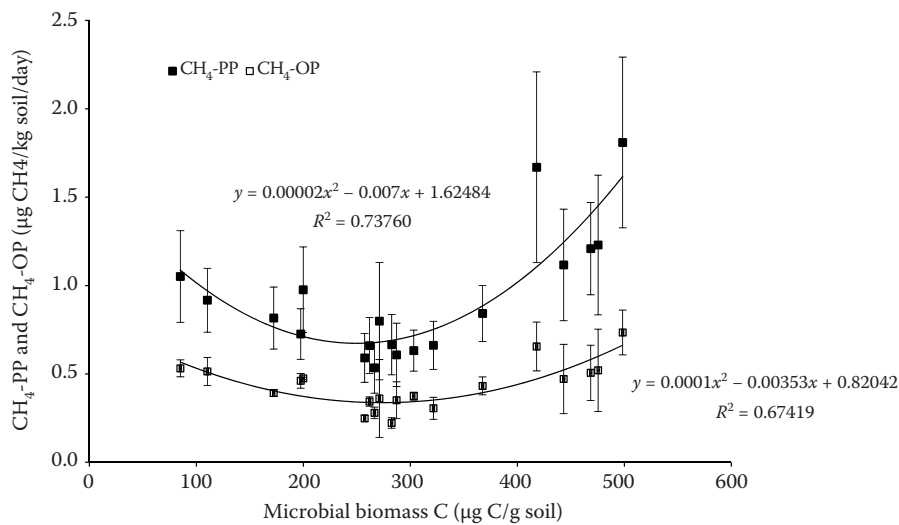


Figure 2. Regression lines of microbial biomass C with  $\text{CH}_4$  production potential ( $\text{CH}_4$ -PP) and  $\text{CH}_4$  oxidation potential ( $\text{CH}_4$ -OP)

high  $\text{CH}_4$ -PP and  $\text{CH}_4$ -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  $\text{C}_{\text{org}}$  and a lower amount of microbial biomass C (Figure 1, right). This condition caused the amount of  $\text{CH}_4$ -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  $\text{CH}_4$ . DAR *et al.* (2008) argue that methanogens have the range of 10% of the total microbes in the laboratory scale bioreactor experiments.

**$\text{CH}_4$ -PP and soil depth.** There was no significant difference between  $\text{CH}_4$ -PP and  $\text{C}_{\text{org}}$  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  $\text{CH}_4$ -PP and  $\text{C}_{\text{org}}$  between all horizon depths analyzed, microbial biomass C correlates significantly with  $\text{C}_{\text{org}}$  ( $r = 0.52$ ,  $P < 0.05$ ,  $n = 19$ ) and  $\text{CH}_4$ -PP ( $r = 0.56$ ,  $P < 0.05$ ,  $n = 19$ ), as  $\text{C}_{\text{org}}$  correlates significantly with  $\text{CH}_4$ -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  $\text{CH}_4$ -PP through its influence on the  $\text{C}_{\text{org}}$  and microbial biomass C. In the upper horizon,  $\text{C}_{\text{org}}$  and microbial biomass C were high as it would increase the  $\text{CH}_4$ -PP.

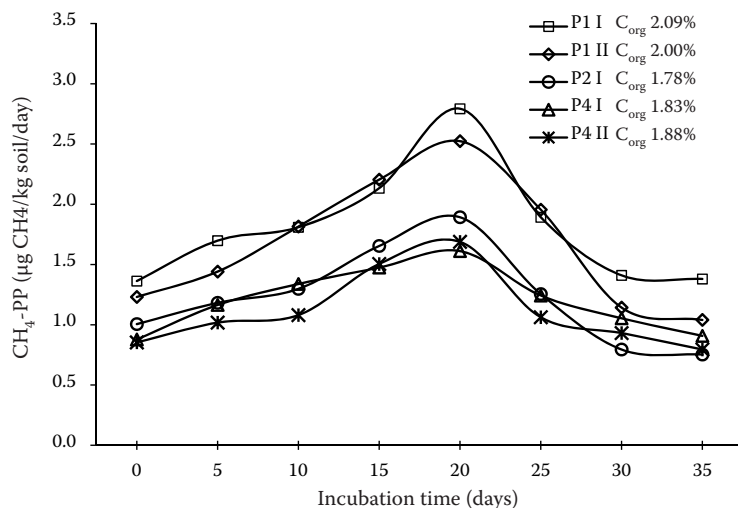


Figure 3.  $\text{CH}_4$  production potential ( $\text{CH}_4$ -PP) patterns of soil with high  $\text{C}_{\text{org}}$

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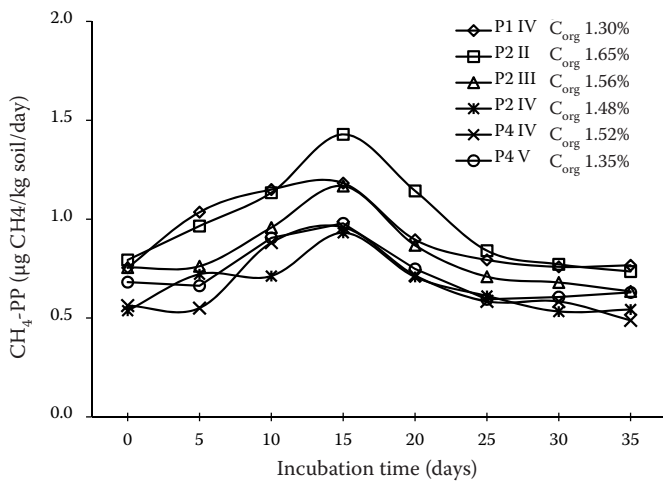


Figure 4. CH<sub>4</sub> production potential (CH<sub>4</sub>-PP) patterns of soil with moderate C<sub>org</sub>

WHALEN and REEBURG (2000) and BRZEZIŃSKA *et al.* (2012) investigated the correlation between soil depth, C<sub>org</sub>, and CH<sub>4</sub>-PP. C<sub>org</sub> content in the upper horizon was higher than in the lower horizon, so as the CH<sub>4</sub>-PP. FREITAG *et al.* (2010) concluded that the depth of the soil is one of the factors that control the amount of CH<sub>4</sub> emissions.

**CH<sub>4</sub>-PP and incubation time.** The level of C<sub>org</sub> determines both CH<sub>4</sub>-PP and CH<sub>4</sub> production patterns based on the time of flood incubation. The soil with high C<sub>org</sub> (P1 horizon I and II, P2 horizon I, and P4 horizon I and II), moderate C<sub>org</sub> (P1 horizon IV, P2 horizon II, III, and IV, and P4 horizon IV and V), and low C<sub>org</sub> (P3 horizon II, III, IV, and V, and P4 horizon VI) reached the maximum average of CH<sub>4</sub> 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<sub>org</sub> has enough C substrates to produce high amounts of CH<sub>4</sub> and it will reach a longer

peak production period. Meanwhile, soil with low C<sub>org</sub> has less C substrate, produces lower amount of CH<sub>4</sub>, 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<sub>4</sub> production after flooding the soil samples. Soil with higher C<sub>org</sub> experienced a longer peak time of production than soil with lower C<sub>org</sub>.

### CONCLUSION

The CH<sub>4</sub> production in soil is reflected in its chemical, physical, and biological properties. This study showed that the CH<sub>4</sub> production potential in organic rice fields is high because of the presence of high amounts of C<sub>org</sub>. In consequence, the mitigation options need more efforts than required in other cultivation (farming) systems. The CH<sub>4</sub> oxidation

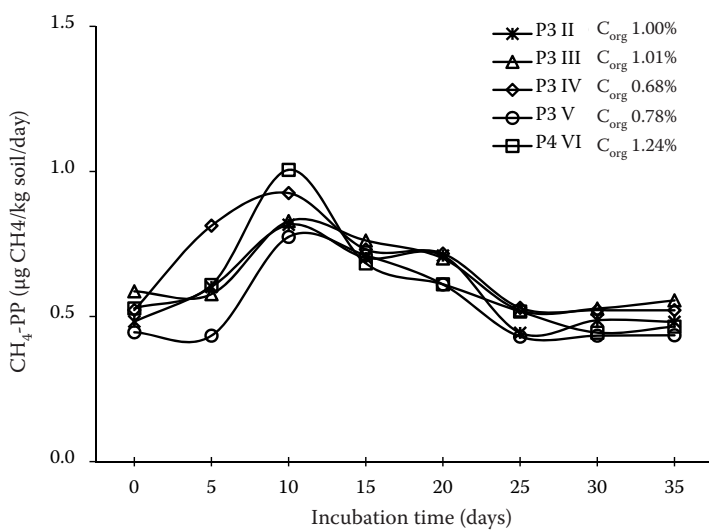


Figure 5. CH<sub>4</sub> production potential (CH<sub>4</sub>-PP) patterns of soil with low C<sub>org</sub>

potential in organic rice fields is high as well, as the mitigation options should focus on increasing the methanotrophic activity which might reduce CH<sub>4</sub> emissions to the atmosphere. Because C<sub>org</sub> has an important role in the production of CH<sub>4</sub>, it determines the dynamics of CH<sub>4</sub>-PP and CH<sub>4</sub>-OP. So using particular kinds of organic matter as C<sub>org</sub> sources would manage the composition of methanogens and methanotrophs so as to reduce CH<sub>4</sub>-PP and to increase CH<sub>4</sub>-OP.

C<sub>org</sub> also affected the pattern of the CH<sub>4</sub> production potential by the time of flood incubation. High amounts of C<sub>org</sub> reached a maximum CH<sub>4</sub>-PP on day 20 after incubation, while moderate C<sub>org</sub> on day 15, and low C<sub>org</sub> on day 10. Higher amounts of C<sub>org</sub> provide enough C substrates to produce higher amounts of CH<sub>4</sub> and reach longer peak times of production than lower C<sub>org</sub>. This finding provides guidance that mitigation option reduces CH<sub>4</sub> emissions from organic rice fields and experiences drainage every 10–20 days before reaching the maximum CH<sub>4</sub>-PP.

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