Soil respiration and microbial population in tropical peat under oil palm plantation

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Abstract. Peatland clearance and drainage result in the change of anaerobic to aerobic condition and hence microbial activities that increase CO_2 emission. This study aimed to evaluate the rate of microbial respiration and microbial population from sapric and hemic peat under oil palm plantation. Research activities included measurement of soil respiration using potassium hydroxide (KOH) to capture the respired CO_2, and counting the population of microbes. Results of this study showed that the highest rate of respiration of 3.3 ± 0.8 mg CO_2 100 g^{-1} day^{-1} occurred from the 0-20 cm layer and it decreased to 2.1 ± 1.0 mg CO_2 100 g^{-1} day^{-1} from the 20-40 cm layer and 1.2 ± 0.9 mg CO_2 100 g^{-1} day^{-1} from the 40-60 cm layer in sapric peat. For the hemic peat the highest rate of respiration of 3.1 ± 0.4 mg CO_2 100 g^{-1} day^{-1} occurred from the 0-20 cm layer and it decreased to 2.0 ± 0.7 mg CO_2 100 g^{-1} day^{-1} from the 20-40 cm layer. Soil respiration decreased with peat depth and bacteria were the most dominant microbes in each peat depth, indicating that bacteria play a more important role in respiration than other microbes.

1. Introduction

Land use change of peatlands in Indonesia is relatively rapid, especially for the expansion of oil palm plantations. Approximately 20% of oil palm plantations on the islands of Sumatra, Kalimantan, and Papua are on peatland [1]. Peatland clearing, which in general conducted concomitantly with the construction of drainage canals results in increased temperatures and microbial activity. The loss of carbon from peatlands into the atmosphere occurs through decomposition of peat organic matter by microbes which are known as heterotrophic respiration [2].

Microbial activity greatly affects the amount of CO_2 emissions from peatlands. Microbes that use organic materials as an energy source are groups of heterotrophic microbes. The higher activity of heterotrophic microbes in the soil will lead to the higher CO_2 emissions. Both the population and activity of microorganisms in peat are important factors influencing the process of peat decomposition.

There was the hypothesis that the rate of peat respiration will decrease with the increasing rate of peat maturity. However, to our knowledge, there have not been any studies comparing peat respiration of mature (saprist) peat and the less mature (hemist) peat. The results of this study will be important as a basis of more rational estimation of the rate of CO_2 emission from microbial respiration or also known

as peat decomposition. This study aimed to (i) evaluate the rate of microbial respiration and microbial population from tropical peat samples taken from different depths and different peat maturity, and (ii) determine the relative importance of microbes in peat soil respiration.

2. Materials and method

2.1. Study site and sampling procedure

This study was conducted at Labuhan Batu Selatan District (N $2^0$0'48" E $100^0$16'7"), North Sumatera with sapric peat maturity and peat depth ranging from 100-200 cm, and at Muaro Jambi District (N $1^0$40'56.9" E $103^0$49'04.2"), Jambi, Indonesia with hemic peat maturity and peat depth ranging from 150-300 cm. In the North Sumatera location, peatland was opened in 1991 which is intended for transmigrants from Java island which began the planting of oil palm in 1992. Currently, the age of the oil palm is 25 year. In the Jambi location, peatland was opened in 2005 and it was planted to oil palm in 2007. This study was conducted from November 2016 to October 2017.

Soil samples for peat biological analyses (soil respiration and microbial population) were taken from the traffic interrow (TI) and the palm-frond pile ("dead") interrow (DI) at the surface layer (0-20 cm), and subsurface layers (20-40 cm and 40-60 cm) for the North Sumatra site. There were 9 samples at TI and 9 samples at DI. The distance between the points on TI and DI were 25 m, 50 m, and 100 m from the drainage canal. For the Jambi site, samples were taken from the identical positions, but only from the 0-20 cm and 20-40 cm depth. There were 6 samples at TI and 6 samples at DI. A peat auger (Ejkelkamp model) was used for sampling. For soil respiration and microbial population measurement, fresh soil samples were used. The samples were transferred into plastic bags and stored in the ice box for transportation from the field to the laboratory and placed in a chilled room at 4-16 ^{0}C before being analysed.

2.2. Soil respiration and microbial population measurement

The measurement of soil respiration and the microbial population was conducted at the Soil Biology Laboratory of Universitas Sumatera Utara, Medan, Indonesia. Soil respiration was measured with a chemical titration method adapted from Anas [3]. Fresh samples of 100 g, 5 mL of 0.2 N potassium hydroxide (KOH), and 10 mL of water were incubated in 1,000 mL vial for 2 weeks at 28-30^{0}C. At the end of the incubation period, 2 drops of phenolptalein were added into the KOH beaker until a red solution is formed. HCl was added into the red solution until the red color disappears and at that point, the volume of HCl used in the titration was recorded. Afterwards, two drops of metyl orange were added into the solution until the yellow color is formed. The solution, then was re-titrated with HCl to form a pink color and the required HCl volume was recorded. A control vial with no soil was included in the incubation to correct for the CO_2 in the jar at the initiation of the incubation. The amount of respiration (R) (in mg CO_2 100 g^{-1} day^{-1}) was calculated using Equation 1.

$$R = \frac{(a-b) \text{ x } t \text{ x } 120}{n} \quad (1)$$

Where:

a = The volume of HCl used for titration in mL
b = The volume of HCl used for titration of the blank/control in mL
t = The normality of HCl
n = the incubation time in the day

Alongside soil respiration measurement, the population of microbial groups of bacteria and fungi, was enumerated from each sample. Spread plate counting technique conducted the enumeration from a series of dilution of each sample. 10 g of fresh soil was put into a 250 mL Erlenmeyer containing 90 mL of physiological solution (8.5 g NaCl in 1 L distilled water) then shaken for 30 min. One mL of pure culture was put into a 10 mL test tube containing 9 mL of physiological solution (resulting in 100 times or 10^{-2} dilution,). It was shaken for 1minutes, and 1 mL of 10^{-2} dilution was taken for dilution of 10^{-2}. Dilution process was continued until the dilution 10^{-9} was reached. For the bacteria, one mL of the 10^{-7},

10^{-8}, and 10^{-9} diluted solution was added with 10 mL of nutrient agar at 50^0C; then the culture was incubated for 3 days. For the fungi, cellulolytic, and lignolytic microbes, one mL of the 10^{-4}, 10^{-5}, and 10^{-6} diluted solution was added with 10 mL of nutrient agar at 50^0C, then incubated for 3 days. After incubation, the number of microbes was counted using Quebec colony counter [3]. The number of microbes was calculated using equation 2.

$$\Sigma\, cell mL^{-1} = \Sigma \text{ colonies x dilution factor} \quad (2)$$

2.3. Data analyses

Data are presented as the mean ± standard deviation (SD) in each soil depth to evaluate the difference of soil respiration in the TI and DI as well as the mature (saprist) from the immature (hemist) peat. Regression analyses were used to test the relationship of microbial population and soil depth in each of the sampling position, peat maturity.

3. Results

3.1. Soil respiration

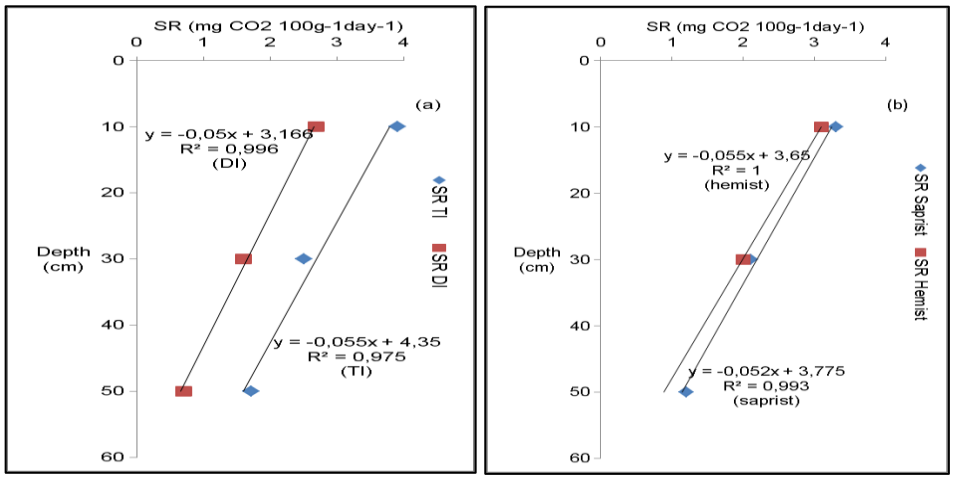


Figure 1. A relationship between soil depth and soil respiration (SR) on the traffic interrow (TI) and dead interrow (DI) (left) and SR by bottom of the saprist and hemist peat (right)

Soil respiration in the saprist and hemist maturity stages decreased significantly with increasing depth (Figure 1), but there was no significant difference in peat respiration between the 20-40 cm and the 40-60 cm layer (Table 1). The highest rate of breathing of the sapric (North Sumatra site) peat of 3.3 ± 0.8 mg CO_2 $100g^{-1}day^{-1}$ occurred from the 0-20 cm layer, and it decreased to 2.1 ± 1.0 mg CO_2 $100g^{-1}day^{-1}$in the 20-40 cm layer and to1.2 ± 0.9 mg CO_2 $100g^{-1}day^{-1}$ in the 40-60 cm layer. For the hemic (Jambi site) peat, the highest rate of respiration was 3.1 ± 0.4 mg CO_2 $100g^{-1}day^{-1}$ occurred from the 0-20 cm layer, and it decreased to 2.0 ± 0.7 mg CO_2 $100g^{-1}day^{-1}$ from the 20-40 cm layer. Soil respiration is highly variable and can fluctuate widely depending on substrate availability, organic matter and moisture content [4,5]

When comparing soil respiration in traffic interrow and "dead" interrow it showed a significant difference ($p = 0.007$) (Table 2). However, we did not find the significant difference in soil respiration between the saprist and hemist maturity peat ($p = 0.625$) (Table 3).

Table 1. Differences test analyses of soil respiration at the surface and subsurface layer.

Depth(I)	Compared to Depth(J)	N	Mean difference (I-J)	Std. Error Mean	p-value
0-20 cm	20-40 cm	6	1.2290	0.50835	0.029
	40-60 cm	6	2.1143	0.50835	0.001
20-40 cm	0-20 cm	6	-1.2290	0.50835	0.029
	40-60 cm	6	0.8853	0.50835	0.102
40-60 cm	0-20 cm	6	-2.1143	0.50835	0.001
	20-40 cm	6	-0.8853	0.50835	0.102

Table 2. Difference test analyses of soil respiration in the traffic interrow and the "dead" interrow.

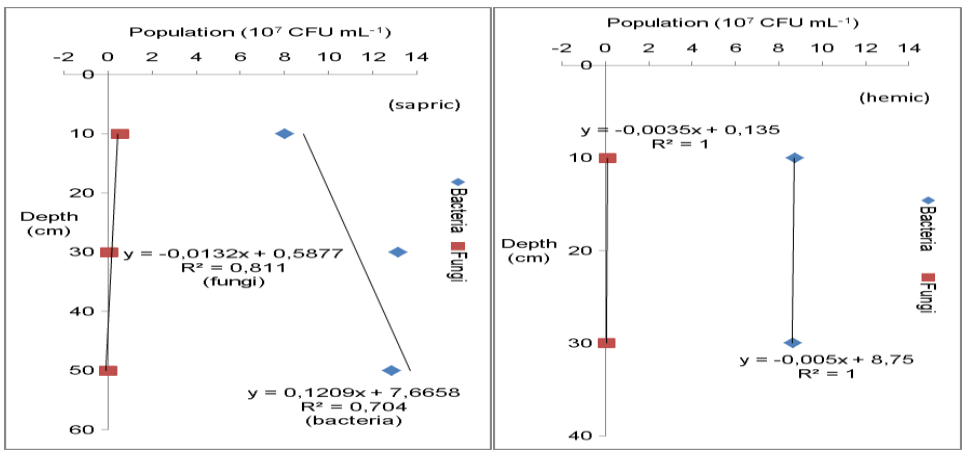
Sampling point	N	Mean	Std. Deviation	Correlation	t	Df	p-value
Traffic interrow	9	2.68578	1.277098	0.747	3.638	8	0.007
"Dead" interrow	9	1.65700	0.958356				

Table 3. Differences test analyses of soil respiration in saprist and hemist peat.

Peat maturity	N	Mean	Std. Deviation	Correlation	T	Df	p-value
saprist	12	2.67	1.06	0.345	0.503	11	0.625
chemist	12	2.51	0.78				

3.2. Microbial population

Bacteria were the dominant group of microbes both in the surface and subsurface layers and both in the saprist and hemist peat (Figure 2). For the saprist peat, the population of bacteria in the 0-20 cm layer was 8.0 x 10^7 CFU mL^{-1}, but somehow it was higher in the 20-40 cm layer reaching to 13.1 x 10^7 CFU mL^{-1}. For the fungi, it was 0.5 x 10^7 CFU mL^{-1} in the 0-20 cm layer and was about the same in the 20-40 cm layer. Lignolytic microbes in the 0-20 cm layer were 0.1 x 10^7 CFU mL^{-1} and were about the same in the 20-40 cm layer. Cellulolytic microbes in the 0-20 cm layer were 0.1 x 10^7 CFU mL^{-1} and were 0 in the 20-40 cm layer.



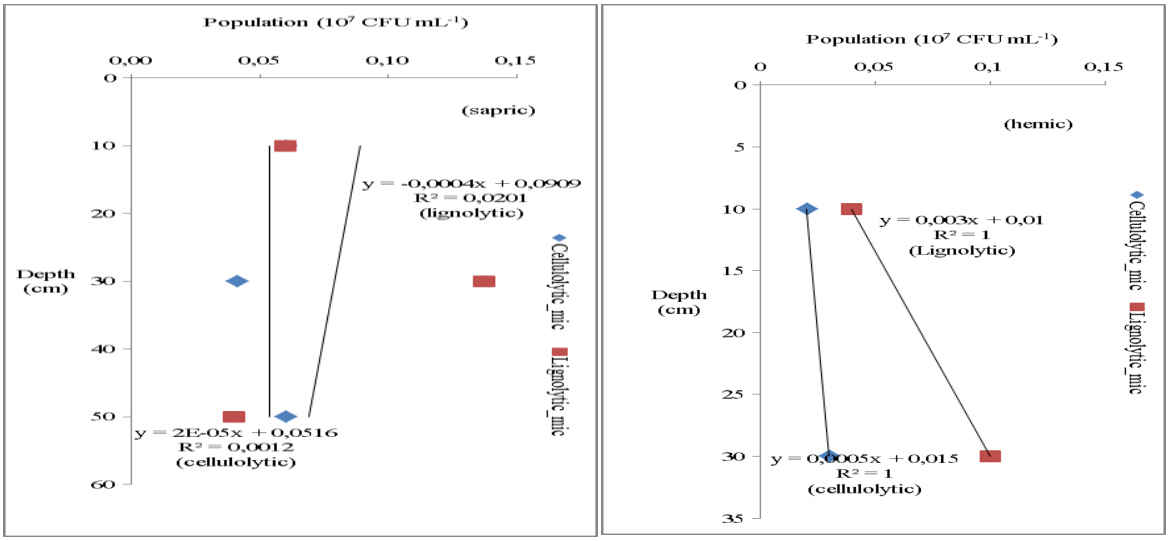


Figure 2. A relationship between soil microbial population and soil depth in saprist and hemist peat;bacteria and fungi (upper) and cellulolytic and lignolytic microbe (lower).

4. Discussion

4.1. The relationship of soil respiration and soil depth

Respiration rate in the thicker layer was lower than in surface layer in both saprist and hemist peat, which was confirmed by the negative relationship between soil respiration and depth in saprist and hemist peat (Figure 1). Microbial respiration affected by several factors such as moisture, temperature, oxygen, nutrient availability, and soil characteristic [6]. Generally, in the more profound layer nutrient availability is lower than the surface layer. This condition inhibits the activity of microorganism in the decomposition process. Besides microbial activity, amount and diversity of microbes also more economical in the deeper layer. The study by Naconieczna and Sypniewski [7] showed that the surface layers were the most active in respiration. The decreased of breath between the depths of 0-20 and 20-40 cm was between 14 and 30%. Another study by Liu et al. [8] reported the similar result that CO_2 emission at 0-20 cm was higher than the deeper layers. The decreased soil respiration with depth was observed both inorganic and in mineral soils [9]. Fresh litter and partially decomposed plant material of the upper peat decompose at a faster rate than older peat material located in the deeper part of the profile [10]. It is clear from this study that soil respiration, which is determined by microbial activity, was affected by soil depth and that it was higher in the surface layer and this seems to be attributed to a relatively high input of fresh and readily decomposable organic matter such as dead root [11].

Several studies have shown that microbes prefer soil influenced by root activity compared to bulk soil [12]. In the deeper layer where lignin content probably higher than in the surface layer also causes of lower respiration rate. Lignin is typically considered a resistant material that is resistant to microbial decomposition. Only specialised biota, predominantly fungi can synthesize extracellular enzymes that break down these structures into biologically usable forms [13], and lignin is known to limit microbial enzyme accessibility to these more labile cell-wall polysaccharides [14].

Statistical analyses (Table 2 and 3) demonstrate that there was a significant difference in soil respiration between traffic interrow (TI) and the dead interrow (DI). The mean value of TI (2.68 mg-CO_2 100g^{-1} d^{-1}) was higher than DI (1.65 mg-CO_2 100g^{-1} d^{-1}). In the field condition, TI was the way for harvesting and fertilising, so that the situation was less of covering than DI which is covering by cover crops and palm leaves. Land condition with less covering causes the higher temperature of TI. According to Pendal [15], rising temperatures directly spur the decomposition process by accelerating enzyme activity and chemical reactions.

When comparing soil respiration between saprist and hemist peat, the result showed that the mean value of respiration in saprist peat (2.67 mg-CO_2 100g^{-1} d^{-1}) was higher than hemist peat (2.51 mg-CO_2 100g^{-1} d^{-1}). This condition due to nutrient availability in saprist peat was higher than fibrist [16]. Substrate availability in saprist peat also higher than hemist peat which supported to microbial activity.

However, statistical analyses showed that there was no significant difference of respiration in saprist and hemist peat (Table. 3). It indicates that decomposition process in saprist peat (from north Sumatra location) was identic with hemist peat (from Jambi location). Another study reported the same result that saprist peat emitted CO_2 significantly higher than in chemist and fibrist peat [17,18]. Therefore, the opinion that CO_2 emission will decrease in more mature peat cannot be proven yet. The numbers of factor affecting the release of CO_2 emission from peatlands including environmental factors may cause the amount of soil respiration in this study to be different from other regions.

4.2. Microbial Population and their function in the decomposition process

Decomposition rate is mainly determined by the activity of microorganism in soil, and that action is determined by the microbial biomass and the environmental condition in the soil [19]. In this study, bacteria were the dominant groups of microbial population in both surface and subsurface layer in both saprist and hemist peat (Figure 2). Number of bacteria in hemist peat was higher than saprist (Figure 2). This is because of soil acidity of saprist peat was higher than hemist peat. A study by Husen et al [20] shows similar result with this current study that bacteria were the dominant group of microbial population in peat sample and the most responsible in peat respiration (decomposition). A study by Winsborough and Basiliko [21] suggested that bacteria could be more active than fungi across many peatland types.

Bacteria and fungi metabolically function as chemoorganotrophs. They are generally heterotrophic and obtain carbon and energy while degrading organic compounds added to soil, including plant residues and dead soil organisms. Those microorganisms secrete exoenzymes (extracellular enzymes) into their environment to initiate the breakdown of litter, which consists of compounds that are too large and insoluble to pass through microbial membranes. These exoenzymes convert macromolecules into soluble products that can be absorbed and metabolized by microbes [5]". A study by Hieber and Gessner [22] showed that fungal biomass and activity generally dominated microbial breakdown process of leaves in streams, while bacteria become relevant in advanced stages of decomposition process. When microbes die, their bodies become part of the organic substrate available for decomposition. Bacteria, and fungi are involved in both aerobic and anaerobic degradation of organic matter, but fungi are more efficient than bacteria in degradation of highly recalcitrant organic matter because they produce a wider range of extracellular enzymes than bacteria [23].

In the lower 20-40 cm and 40-60 cm layers lignolytic microbes had the higher population, because of lignin compound was more accumulate in the deeper layer, although their activities in respiration is lesser so than the total bacterial activities in the surface layer.

In this current study, we did not find significant difference of respiration between the hemist peat from Jambi and the saprist peat from North Sumatra. It seems the complexity of the influencing factors have not lead us to a confirmation of a hypothesis that the respiration slows down in the more mature peat. The analyses by depth in this laboratory study shows the more rapid respiration in the upper layer. The higher component of lignin and in the deeper layer seems to be a factor.

5. Conclusions

Soil respiration decreased with peat depth and this seems to be affected by the higher concentration of more recalcitrant lignin compounds in the deeper layer. Respiration from the traffic interrow was significantly different from that of the palm frond accumulation "dead" interrow. However, we did not found difference in respiration between the saprist (most mature) and hemist (less mature) peat. It means that decomposition process in saprist peat of north sumatera location is still active likes in less mature peat of Jambi location. Therefore, new innovations are needed in peatland management to minimize the release of CO_2 from peatland. So that the peatland life time can be last longer. Bacteria was the most dominant microbe in each depth of peat, indicating that bacteria play a more important role in respiration than other microbes like fungi.

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