

Full Length Research Paper

Evaluation of soil biological properties of 9- and 15-year-old stands in the oil palm plantation in Perak, Malaysia

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Opening land for oil palm cultivation provokes many debates around the world regarding on the fate of biodiversity. A study was conducted to evaluate and compare soil biological properties of 9-year-old (P1) and 15-year-old (P2) stands of an oil palm plantation in Bikam, Perak, Malaysia. Composite samples were collected at depths of 0-15 cm (topsoil) and 15-30 cm (subsoil) located within six subplots (20 m × 20 m). The microbial population count was estimated using a spread-plate technique, and the Fluorescein diacetate (FDA) hydrolysis assay was used to measure microbial enzymatic activity. A rapid ethanol-free chloroform fumigation extraction technique was used for microbial biomass extraction, and the extracts were respectively analyzed by wet dichromate oxidation and Kjeldahl digestion for biomass carbon (C) and nitrogen (N). At the 0-15 cm depth, the microbial biomass C and N contents in the soils from both plots were significantly different ($P < 0.05$). At the 15-30 cm depth, only microbial enzymatic activity was significantly different between plots. Although the addition of fertilizers to the soil is believed to be a predisposing factor, no significant differences in P1 and P2 plots for the biomass C and N in soils at the 15-30 cm depth were observed. Variations in the MBC/MBN ratio in soils of the P1 and P2 plots indicate that changes occurring in the soil microbial composition are due to the availability of soil organic substrates and N. Thus establishment of an oil palm plantation does contribute to changes in soil biological properties.

Key words: Oil palm, different ages, microbial population, enzymatic activity, biomass C and N, rapid chloroform-fumigation extraction technique.

INTRODUCTION

Oil palm production has increased in many Association of Southeast Asian Nations (ASEAN) countries, especially

in Malaysia and Indonesia, because it is an important commercial product for domestic and international use

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(Laurance, 2007). As such, clearing of forest areas for oil palm cultivation has resulted in massive deforestation. In Malaysia, the area of oil palm plantations was about 4 million ha in 2005 and had an annual growth of 10.06% (Basiron, 2007). Butler and Laurance (2009) reported that expansion of oil palm cultivation contributes to the loss of biodiversity in lowland and peat swamp forests. Meanwhile, the members of The European Commission were trying to uphold a ban on the import of fuel crops, which includes oil palm, that are cultivated in areas on the endangered list, such as natural forests (Koh and Wilcove, 2008). In contrast, Basiron (2007) stated that Malaysia's oil palm industry is the country's best-organized sector, and many programs are in place to educate communities about the management, conservation and community services provided for the environment and people.

Therefore, it is crucial for scientists working on oil palm to provide essential information on the land management of the estate rather than just focusing on the yield of oil palm. Through such practice, we can predict the degree of disturbance caused by the plantation to the environment. We all are aware of the high levels of fertilizer application to this plant; hence, we need to quantify the effect of it on soil degradation and biodiversity. Lalfakzuala et al. (2008) suggested that heavily cultivated agricultural areas disrupt soil productivity due to depletion of soil organic matter. However, Henson (1999) believed that oil palm cultivation contributes little to environmental damage. He also clarified that negative effects only occur at the beginning of forest clearing operations.

One of the approaches used to determine the soil quality of a particular plantation is to analyze soil biological properties. Sánchez-Monedero et al. (2008) found that microbial enzymatic activity is an important aspect of the decomposition of organic materials, such as humus; in the degradation of pollutants; and in the transformation of nutrients suitable for plant uptake. In addition, Lalfakzuala et al. (2008) suggested that it is important to study biological properties as well because they are a sensitive indicator that can be used to quantify soil fertility and quality. Moreover, examination of microbial enzymatic activity gives instant view of the organic matter turnover in soil (Joergensen and Emmerling, 2006). Ajwa et al. (1999) also stated that soil microbial activity is a sensitive indicator that proportionally changes with the other changes or disturbances occurring in the soil of particular area. The fertility of the soil is proportional to the amount and the number of times fertilizer is applied to enhance oil palm growth (Phosri et al., 2010).

Gaspar et al. (2001) reported that soil microbial biomass contributes 1 to 4% of organic carbon and 2 to 6% of organic nitrogen in the soil. Furthermore, microbial biomass C and N are important constituents in soil organic matter and are the main nutrients stored for plant uptake. Furthermore, Ajwa et al. (1999) and Rice et al. (1996) found that environmental changes (e.g., weather

changes, physical disturbances and chemical toxicity) influence the activities of microbial biomass in forest soils. All of these factors make soil microbial biomass a good, sensitive indicator of soil quality and fertility (Islam and Weil, 2000).

In this study, the Fluorescein diacetate (FDA) hydrolysis assay described by Sánchez-Monedero et al. (2008) was chosen for the evaluation of microbial enzymatic activity because of its rapid estimation of overall enzymatic activity. In addition, Schnürer and Rosswall (1982) claimed that FDA possesses the ability to rapidly hydrolyze a wide range of enzymes including esterases, lipases and proteases.

Soil microbial biomass C and N are two important components of soil organic matter because plant or tree species in a particular area obtain nutrients from nutrient storage in organic matter (Barbhuiya et al., 2004). A higher level of organic matter in the soil indicates that a larger proportion of C and N are available. Studies of the relationship between soil microbial biomass and selected soil properties, such as organic matter and total nitrogen, will give us a clear view of the current fertility and quality of cultivated land. Soil acidity and moisture content can affect the distribution of microorganisms in the soil. Behera and Sahani (2003) reported that most bacteria cannot withstand acidic or dry soil conditions. Highly acidic soil conditions inhibit microbial activity in the soil, which subsequently affects nutrient cycling in the soil (González-Pérez et al., 2006). However, Shamshuddin and Che Fauziah (2010) reported that oil palm trees were able to achieve good growth performance in highly acidic soil.

To our knowledge, studies of the soil biological properties of oil palm plantations are lacking. Therefore, the objective of the current study was to evaluate and compare the soil biological properties of 9-year-old and 15-year-old oil palm stands in Bikam Plantation, Perak, Malaysia.

MATERIALS AND METHODS

Description of study site

A 9-year-old (N 03°.99'862" E 101°.24'220", approximately 12 m above sea level) (P1) and a 15-year-old (N 03°.99'811" E 101°.24'110", ± 9 m a.s.l) (P2) oil palm stand in a plantation in Bikam, Perak were selected as study plots. The mean annual rainfall and temperature are 2,417 mm and 24.5°C, respectively. The soils in this study area are classified as Ultisols; these soils are characterized as highly weathered because they contain a large amount of low-activity clays associated with high Al saturation (Arifin et al., 2008). Both plots were managed independently by local farmers. The basic fertilizers annually applied to these crops were N:P:K (15:15:15) fertilizers. The distance of the palm stand from each other was 9 m × 9 m.

Soil sampling

At each plot, six subplots were established that were each 20 m

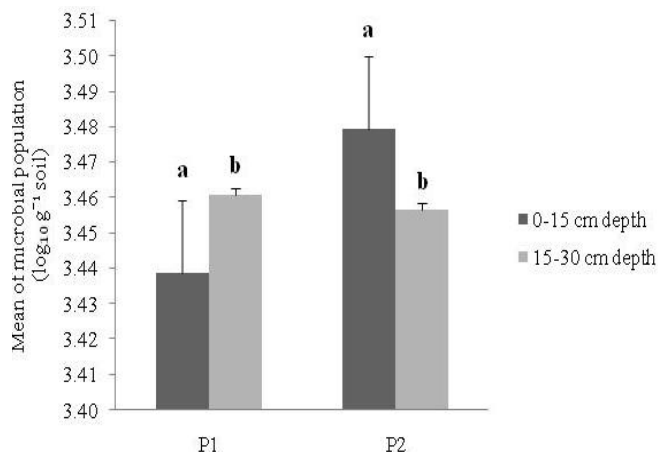


Figure 1. Means of the soil microbial populations at 9-year-old (P1) and 15-year-old (P2) oil palm plots. Different letters indicate significant differences between the means of the same soil depths at the 9-year-old (P1) and 15-year-old (P2) oil palm plots using the Student's t-test ($P < 0.05$).

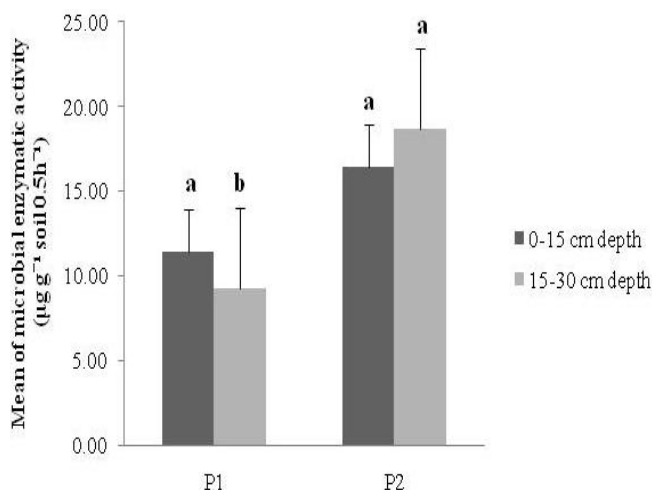


Figure 2. Means of the microbial enzymatic activity at 9-year-old (P1) and 15-year-old (P2) oil palm plots. Different letters indicate significant differences between the means of the same soil depths at the 9-year-old (P1) and 15-year-old (P2) oil palm plots using the Student's t-test ($P < 0.05$).

× 20 m in size. At each plot, a composite sample was obtained after mixing six soil samples that were collected randomly from 0-15 cm and 15-30 cm depths. A total of 24 composite samples were obtained from the plots. All composite samples collected were wrapped in UV-sterilized polyethylene bags and stored in ice-filled polystyrene boxes before being analyzed in the laboratory.

Soil analyses

A number of parameters were selected for determining the soil biological properties of P1 and P2. The microbial population count was estimated via the spread-plate technique (Sleytr et al., 2007).

FDA was used for microbial enzymatic activity analysis. The rapid ethanol-free chloroform fumigation extraction approach was used for the extraction of soil microbial biomass (Witt et al., 2002). The extract was subjected to wet dichromate oxidation to determine microbial biomass C (Vasquez-Murrieta et al., 2007; Vance et al., 1987) and to Kjeldahl digestion to determine microbial biomass N (Simmone et al., 1997; Brookes et al., 1985). Selected soil physico-chemical analyses were also performed. Organic matter and organic C were elucidated via the loss-on-ignition technique (Ahmadpour et al., 2010); total N was determined using Kjeldahl digestion. Soil pH was determined using a glass electrode and a soil: distilled water ratio of 1:2.5 (w/w) (Akbar et al., 2010). Bulk density was determined using disturbed soil sample technique as described by Gupta (2007). Gravimetric method was performed to elucidate soil moisture content for each sample.

Data analyses

The mean values obtained from the same soil depths in P1 and P2 plots were analyzed using the Student's t-test. Pearson correlation analyses were performed using SPSS (version 16.0) to detect any linear relationships between microbial biomass C and organic matter and between microbial biomass N and total N from the same soil depths at both sites.

RESULTS

Microbial population

For both plots, no significant differences ($P < 0.05$) were observed in microbial population counts of soil at the same soil depths (Figure 1). The means of the microbial population counts for the soils at 0-15 cm and 15-30 cm depths in P1 were $3.44 \pm 1.97 \log_{10} \text{g}^{-1} \text{soil}$ and $3.46 \pm 0.05 \log_{10} \text{g}^{-1} \text{soil}$, respectively; for the P2 plot, the means were $3.48 \pm 0.03 \log_{10} \text{g}^{-1} \text{soil}$ and $3.46 \pm 0.04 \log_{10} \text{g}^{-1} \text{soil}$ at the 0-15 cm and 15-30 cm depths, respectively.

Microbial enzymatic activity

The microbial enzymatic activity in the soils of P2 ($9.27 \pm 1.71 \mu\text{g g}^{-1} \text{soil } 0.5\text{h}^{-1}$) was significantly higher ($P < 0.05$) than that in the soils of P1 ($18.69 \pm 1.97 \mu\text{g g}^{-1} \text{soil } 0.5\text{h}^{-1}$) at the 15-30 cm depth (Figure 2). In contrast, no significant difference ($P < 0.05$) in microbial enzymatic activity was observed at the 0 to 15 cm depth for the P1 ($11.39 \pm 3.01 \mu\text{g g}^{-1} \text{soil } 0.5\text{h}^{-1}$) and P2 ($16.44 \pm 1.28 \mu\text{g g}^{-1} \text{soil } 0.5\text{h}^{-1}$) plots.

Microbial biomass C (MBC)

The MBC was found to be significantly higher ($P < 0.05$) in P1 ($409 \pm 142 \mu\text{g g}^{-1} \text{soil}$) compared to that in P2 ($82 \pm 33 \mu\text{g g}^{-1} \text{soil}$) at the 0-15 cm depth (Figure 3). However, no significant difference ($P < 0.05$) in MBC was found in the soil at the 15-30 cm depth for either plot. The means of the soil MBC for the P1 and P2 plots at the 15 to 30 cm depth were 330 ± 78 and $421 \pm 312 \mu\text{g g}^{-1} \text{soil}$, respectively.

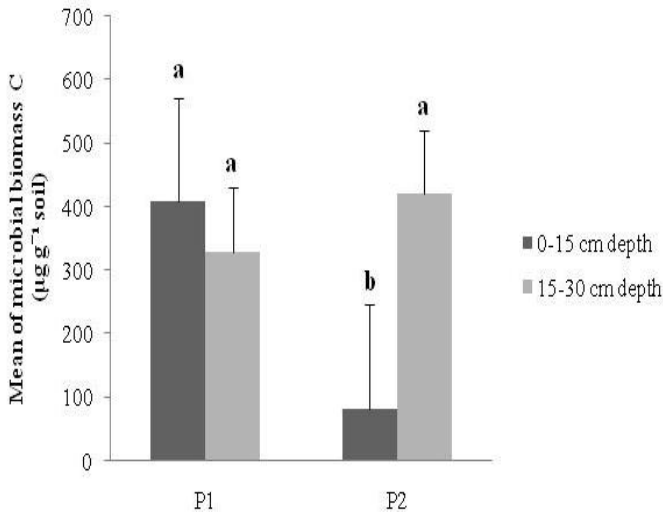


Figure 3. Means of the soil microbial biomass C at the 9-year-old (P1) and 15-year-old (P2) oil palm plots. Different letters indicate significant differences between the means of the same soil depths at the 9-year-old (P1) and 15-year-old (P2) oil palm plots using the Student's t-test ($P < 0.05$).

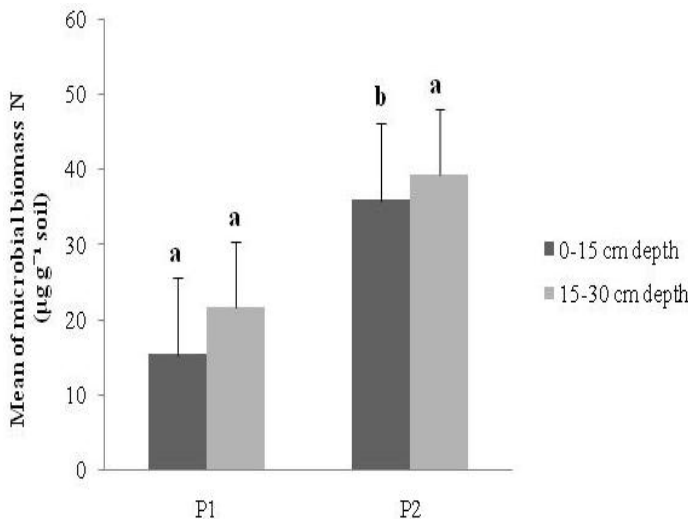


Figure 4. Means of the soil microbial biomass N at the 9-year-old (P1) and 15-year-old (P2) oil palm plots. Different letters indicate significant differences between the means of the same soil depths at the 9-year-old (P1) and 15-year-old (P2) plots using the Student's t-test ($P < 0.05$).

Microbial biomass N (MBN)

The mean of the soil MBN was found to be significantly lower ($P < 0.05$) in P1 ($15 \pm 3 \mu\text{g g}^{-1}$ soil) than in P2 ($22 \pm 4 \mu\text{g g}^{-1}$ soil) at the 0-15 cm depth (Figure 4). On the other hand, the means of the soil MBN at the 15-30 cm depth for both the P1 ($36 \pm 8 \mu\text{g g}^{-1}$ soil) and P2 ($39 \pm 12 \mu\text{g g}^{-1}$ soil) plots were not significantly different ($P < 0.05$).

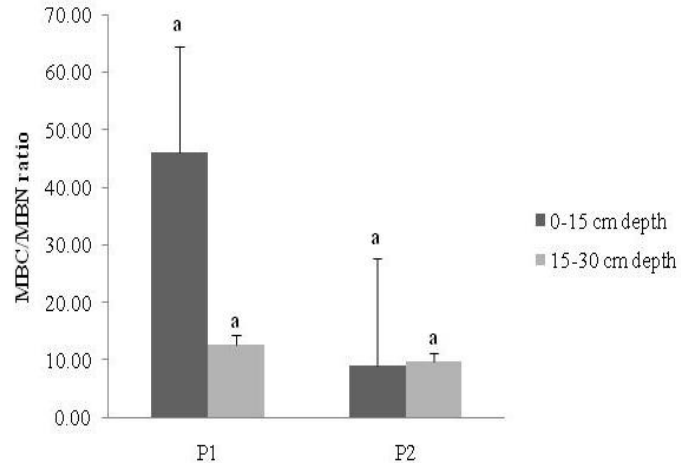


Figure 5. Ratios of microbial biomass C to microbial biomass N (MBC/MBN) at the 9-year-old (P1) and 15-year-old (P2) oil palm plots.

Microbial biomass C to microbial biomass N ratio (MBC/MBN)

The ratios of MBC/MBN in P1 at the 0-15 cm and 15-30 cm depths were higher compared to the P2 ratios (Figure 5). The values of the MBC/MBN ratio for P1 at the 0-15 cm and 15-30 cm depths were 46.03 ± 2.99 and 12.77 ± 2.95 , respectively. In contrast, the values of the MBC/MBN ratio for P2 at the 0-15 cm and 15-30 cm depths were 9.08 ± 2.28 and 9.71 ± 2.23 , respectively.

Organic matter, organic C, total N and soil acidity

Table 1 shows the soil organic matter, organic C, total N and pH of the P1 and P2 plots. The organic matter and organic C were significantly higher ($P < 0.05$) in the soils of P1 compared to those of P2. However, no significant difference in total N ($P > 0.05$) in soil was found between the plots. The soil of P1 exhibited a significantly different pH value compared to the soil of P2 ($P < 0.05$). P2 possessed higher ($P < 0.05$) bulk density compared to P1 for both soil depths. In contrast, P1 showed significantly higher ($P < 0.05$) moisture content compared to P2.

Pearson correlation analysis

Pearson correlation analysis indicated that no strong relationship exists between biomass C and organic matter or between biomass N and total N for the same soil depths in either of the plots (Table 2). Correlation analysis done between organic matters with MBC/MBN ratio showed no strong relationship. Hence, these data showed that organic matter and total N did not directly impact the microbial biomass C and N availability in the soil.

Table 1. Selected soil physico-chemical properties of the 9-year-old (P1) and 15-year-old (P2) plots.

| Parameter | P1 | P2 | P value |
|------------------------------------|---------------------------|---------------------------|----------|
| 0-15 cm depth | | | |
| Organic matter (%) | 5.18 ± 0.49 ^b | 3.19 ± 0.11 ^c | 0.002644 |
| Organic carbon (%) | 3.00 ± 0.28 ^b | 1.85 ± 0.06 ^c | 0.002644 |
| Total nitrogen (%) | 0.54 ± 0.04 ^b | 0.54 ± 0.02 ^b | 0.888409 |
| pH-H ₂ O | 4.47 ± 0.09 ^b | 4.80 ± 0.04 ^c | 0.006851 |
| Bulk density (g cm ⁻³) | 1.43 ± 0.03 ^b | 1.52 ± 0.02 ^c | 0.016433 |
| Moisture content (%) | 19.50 ± 0.72 ^c | 17.83 ± 2.39 ^c | 0.518794 |
| 15-30 cm depth | | | |
| Organic matter (%) | 6.30 ± 0.73 ^b | 11.27 ± 0.78 ^c | 0.007515 |
| Organic carbon (%) | 3.66 ± 0.43 ^b | 6.54 ± 0.45 ^c | 0.003105 |
| Total nitrogen (%) | 0.64 ± 0.17 ^a | 0.49 ± 0.05 ^a | 0.392245 |
| pH-H ₂ O | 4.47 ± 0.11 ^b | 4.84 ± 0.07 ^c | 0.015383 |
| Bulk density (g cm ⁻³) | 1.45 ± 0.02 ^a | 1.54 ± 0.01 ^b | 0.005173 |
| Moisture content (%) | 22.50 ± 1.96 ^a | 15.33 ± 1.58 ^b | 0.017503 |

Different letters within each row indicate significant differences between the means of soil properties at both depths at the 9-year-old (P1) or 15-year-old (P2) oil palm plots using the Student's t-test ($P < 0.05$).

Table 2. Pearson correlation analysis results comparing microbial biomass C (MBC) with organic matter (OM), microbial biomass N (MBN) with total N (TN) and OM with MBC/MBN ratio for both plots at the same soil depths.

| Soil depth (cm) | MBC versus OM | | MBN versus TN | | OM versus MBC/MBN ratio | |
|-----------------|---------------|----------------|---------------|----------------|-------------------------|----------------|
| | P value | r ² | P value | r ² | P value | r ² |
| P1 (0-15) | 0.193 | 0.616 | 0.346 | 0.470 | 0.168 | 0.644 |
| P1 (15-30) | 0.476 | -0.365 | 0.868 | 0.088 | 0.627 | -0.254 |
| P2 (0-15) | 0.248 | -0.565 | 0.059 | 0.794 | 0.324 | -0.490 |
| P2 (15-30) | 0.322 | -0.492 | 0.373 | 0.448 | 0.526 | -0.335 |

P1, 9-year-old oil palm stand; P2, 15-year-old oil palm stand.

DISCUSSION

Significant differences in microbial population counts for both P1 and P2 plots were detected; this result could be due to the adequate application of fertilizer. An adequate supply of nutrients helps microbes to enhance nutrient cycling within soils. It has been found that differences in the slope gradient contribute to the uneven distribution of soil nutrients that leads to the uneven distribution of soil microorganisms. However, both the P1 and P2 plots possess the same type of topography, which partly explains the similarities in the distributions of the microbial populations in these plots. The high rate of microbial enzymatic activity in the soils from the 15-30 cm depth of P1 could be due to the effects of fertilizer addition to the soil. Addition of fertilizer causes certain soil bacteria to become inactive due to the excessive supply of fertilizer in the soil (Klose et al., 1999; Deng and Tabatabai, 1997). After 12 years of planting, farmers normally apply less fertilizer. Hence, the reduced nutrient availability in the soil triggers or forces soil bacteria,

especially nitrogen-fixing bacteria, to reactivate nutrient cycling. In addition, Klose et al. (1999) stated that soil enzymes are vital tools for assessing soil quality because soil biota rapidly respond to changes in the soil environment.

In the topsoil, the MBC was higher in P1 compared to P2; this phenomenon was enhanced by the availability of high levels of organic C. Hu et al. (1997) illustrated that the turnover and changes occurring in soil organic C and biomass C of agricultural soils were influenced by management practices. Organic matter content in the oil palm soil triggers soil microbial activities because it is one of the vital nutrient sources for microbes. In addition, large amounts of organic matter and C in the soil provide an ideal medium for the growth of soil microorganisms. Moreover, the decomposition of soil organic matter by soil microbes contributes the high levels of humus and organic C to the soil; oil palm can then take up these nutrients. In contrast, low biomass C in P2 could be due to the lack of monitoring of the ground cover availability. The P2 area was monitored less than P1 because of the

age of the oil palm; this stand is 15 years old and does not require as much attention as the younger palm stand. On the other hand, the MBN in the soil at the 15 to 30 cm depth was higher in P2 than in P1. Low N availability in the soils of P1 could be due to the decomposition of litter (Barbhuiya et al., 2004). Besides low N availability, Omay et al. (1997) claimed that N fertilization and cropping systems used for long-term production affect certain soil physico-chemical properties and nutrient cycles. Murwira and Kirchmann (1993) found that immobilization of N in soil was due to the addition of certain types of N fertilizer and helped to increase crop yield.

Behera and Sahani (2003) reported that variations in the MBC/MBN ratio indicate a qualitative change in the microbial composition of soil. The MBC/MBN ratios at both soil depths were higher for the P1 than for the P2 soils. This result could be due to the reduced soil accumulation of N in P1 compared to P2. Furthermore, most of the total N will be absorbed through plant roots for oil palm growth; hence, this process results in lower N availability in the soil. In addition, Arunachalam and Pandey (2003) reported that fungal domination of microbial biomass in the soil of a disturbed forest potentially results in soil nutrient retention and conservation. The higher MBC/MBN ratio found in P1 soil is also due to the reduced microbial N availability that results from the slow rate of organic matter accumulation in fine roots. Arunachalam and Pandey (2003) also stated that deforestation or land opening for agricultural cultivation affects the restoration of total N.

From the Pearson correlation analysis, no strong relationship was detected between microbial biomass C and organic matter available in the soil of the oil palm plantation. This condition was probably due to the fact that organic matter content at oil palm plantation is relatively low due to no litter or forest ground (Haron et al., 1998) at the respective plot. In addition low contribution of organic matter on microbial biomass C through the study done by Lalfakzuala et al. (2008) by which the high rate of fertilizer application to soils of oil palm farms subsequently over took the function of organic matter as the nutrient reservoir in the soil. Furthermore, Patzel et al. (1999) stated that there was little organic matter available in agricultural land. Total N was also uncorrelated with microbial biomass N at both plots in the current study. This finding proves that the soil conditions were altered by the high or excessive amounts of nitrogen fertilizers in the soil, provided that excessive fertilizer application or available nitrogen in the oil palm plantation is the crucial factor that influences soil microbial biomass N. Large amounts of nitrogen in the soil reduce the ability of microbes to convert organic nitrogen into inorganic nitrogen (Murwira and Kirchmann, 1993; Omay et al., 1997).

Conclusion

Land opening for an oil palm plantation affects the

distribution of selected soil biological properties, including microbial enzymatic activity and biomass C and N in different age groups of oil palm plots. Land management practices for oil palm cultivation include fertilizer applications; these applications result in soil alterations that enhance or negatively impact soil microbial components. Hence, it is important to include soil biological properties in the evaluation of plantation soils. Soil biological properties, such as microbial enzymatic activity and biomass, are important for describing nutrient sustainability and regeneration that are used to assess land management of the oil palm plantation.

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