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ASSESSMENT OF THE MICROBIAL BIOMASS AND THE ENZYME ACTIVITY IN RELATION TO THE FERTILITY AND QUALITY OF PEAT FOR A SUSTAINABLE AGRICULTURE IN SEPANG, MALAYSIA

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ABSTRACT

Microbial biomass is an important parameter linking the plants to the soil and a source of plant nutrients. Soil microbes play important role in maintaining the soil fertility and they vary with locations due to environmental and human factors. In this study, we examined the effect of the microbial communities on the fertility and quality of the peat or organic soil for a sustainable agriculture. This study was conducted in a cassava farm located in Sepang district of Selangor, Malaysia, and the soil type is peat or organic (0-60 cm). A fresh peat samples (0-15 cm) was collected randomly (30 points) across the study area of about 9 ha, kept in refrigerator at 4°C for microbial biomass carbon, microbial biomass nitrogen and dehydrogenase determination using the appropriate methods. Results obtained ranged from 57.25 µgC/g – 1189.21 µgC/g, 1.46 µgN/g – 10.31 µgN/g, and 0.19 µgTPF/1g/24hrs - 6.23 µgTPF/1g/24hrs for microbial biomass carbon (MBC), microbial biomass nitrogen (MBN) and dehydrogenase (DHA) respectively. There was a moderate variation as the coefficient of variation (CV %) recorded were 67.82 %, 47.0 % and 60.12 % for MBC, MBN and DHA respectively. Results shows that the high organic content of the peat favors the growth of microbial population and increases the fertility of the soil in the study area. The microbial index and enzyme activity (MBC, MBN, DHA), showed correlation with other soil nutrients and shows that they can serve as a useful soil quality indicator with respect to the different land use and soil management.

Keywords: Peat; fertility; Microbial population; Microbial Biomass; Enzyme activity;

INTRODUCTION

The tons of organism in the soil plays an essential role in the nutrient cycling and the development of soil structure. The soil organisms continuously adapt to the changes in their environments therefore, they are regarded as a sensitive indicators of soil quality assessment. The measurement of the soil organisms is difficult to make and interpret due to their responsiveness to environmental changes and microbial environment can change over short distance and short period of time. Soil microorganisms are responsible for the decomposition of plant residue into humus and nutrients for optimum plant growth. Minerals and ions are immobilized by the microbial community until the organism dies before it will be released. The roles of the microorganism in soil are nitrogen cycling, promotes plant growth, degrading synthetic soil contaminants, improves drought tolerance of plant, improves soil aggregation and controls disease and insect pest (Linden et al. 1994).

Most soils throughout the earth contains millions of microorganisms that functions in increasing the soil quality or fertility and essential for plant growth (Christos et al., 2014). The microorganisms are sometimes regarded as agents of disease on the farmland, however, they have a positive effect on the soils by increasing the fertility status of the soil, because these microbes help to decompose the organic matter (Schulz et al., 2013). Most of the nutrients found in the soil due to the biological activities are mainly by the actions of the microorganism (Kiflu and Beyene, 2013). The nutrients status in the soil are measured by identifying the quality of the microorganism present in the soil in an agro-ecosystem (Lombard et al., 2011), and these helps the farmers to maintain the soil nutrients to obtain a better yield of crops. Previous studies (Grayston et al., 2004; Celik, 2005; Ibekwe et al., 2010; Strickland & Rousk,

2010) has revealed that microbial community structure are determined by chemical properties and environmental factors of the soil. The activities of the soil microorganisms will increase or decrease due to the effect of soil and land management, thereby, affecting the fertility status of the soil (Minerdi et al., 2001; Liu et al., 2010).

The microbial indicators as given by Kennedy & Papendick (1995), are Organic Carbon, Microbial Biomass Nitrogen and Carbon, Potentially Mineralizable Nitrogen (PMN), Soil Respiration, Enzymes, Ratio of Biomass Carbon to Total Organic Carbon, Ratio of Soil Respiration to Microbial Biomass, Microbial Community Fingerprinting (substrate utilization, fatty acid analysis, nucleic acid analysis, etc.). The microbial biomass represents about 1-5 % of the total organic carbon in the soil and it also gives a lot of information on the improvement and deterioration of the soil quality which result from the different soil managements and land use (Powlson & brooks, 1987). The activity of the microbial biomass is commonly used to determine the characteristics of the microbiological status of the soil (Nannipieri et al., 1990), and also to ascertain definitely the effect of cultivation (Anderson & Domsch, 1993) and field management (Perrott et al., 1992) on soil microorganism. Therefore, this present study aims at assessing the effect of the microbial biomass carbon (MBC), microbial biomass nitrogen (MBN) and dehydrogenase activity (DHA) on the fertility or quality of peat or organic soil for a sustainable agriculture, also to study the interrelationship between MBC, MBN and DHA with some selected chemical properties.

MATERIALS AND METHODS

This study was conducted in a cassava farm (TKPM Ulu Chucoh) located in Sepang district in the Southern part of Selangor State, Malaysia (latitude 02°45^{1} N and longitude 101°40^{1} E) with an elevation of 4m above water level. The study was conducted on a 9 ha land and the soil type in the study area comprises of both the peat (60 cm in depth) and admixture of peat and mineral soil underlying the peat (60-100 cm in depth). A fresh peat samples (0-15 cm) was collected randomly (30 points) across the study area, kept in refrigerator at 4^{o}C for microbial biomass analysis, and the enzyme activity determination using the appropriate methods. Other soil samples (chemical analysis) from the same sample points were collected, air-dried, ground using mortar and pestle, sieved to pass a 2 mm mesh sieve for further laboratory analysis. For chemical analysis, Soil pH, Soil Organic Carbon (SOC), Available P, Exchangeable cations (Ca, Mg, K), and for the biological analysis, microbial biomass carbon (MBC), microbial biomass nitrogen (MBN) and dehydrogenase activity (DHA) were determined. Soil pH (H_2O) was determined as proposed by Jones (2002). Determination of available P was done using the Bray and Kurtz 2 method (Kuo 1996). The Soil Organic Carbon was determined by Walkley and Black (1934) method. Exchangeable cations (Ca, Mg, K) were determined by leaching method using 1 N ammonium acetate at pH 7 (Ross & Ketterings 1995; Shamshuddin 2006). Microbial biomass analysis was carried out according to the methods proposed by Anderson & Ingram (1993), 0.5M of k_2SO_4 was used for the extraction. For MBC, the extract was transferred into a digestion tube, added 1ml of 0.167M of potassium dichromate ($K_2Cr_2O_7$), and 5 ml of conc. H_2SO_4, heated for 30 min at 150^{o}c, allow to cool and determine the concentration using the UV- spectrophotometer at 600 nm, MBC was calculated as proposed by Vance et al., (1987). MBN after extraction was transferred into the Kjeldah flask, added 5ml of conc. H_2SO_4, 1 g of kjeldah catalyst and then heat for 1 hour at 360^{o}c, let it cool, distil and titrate using 0.001N of HCl (Bremner 1965). MBN was calculated according to Brookes et al., 1985. Dehydrogenase activity (DHA) was determined using the Casida et al., 1964 by reducing 2,3.5 triphenyltetrazolium chloride (TTC), the samples were incubated for 24hours, followed by extraction using 5 ml of methanol (repeat 3 times), vortex, centrifuge, and separate the extract. Use the extract to determine the concentration of DHA in the soil sample using the spectrophotometer. Descriptive statistical measures such as Mean, Range, Standard Deviation (SD), Coefficient of variation (CV %) were used.

Analysis of variance (ANOVA) and the least significant difference (LSD) was used for mean separation at P≤0.05. Pearson correlation was done to investigate the interrelationships among the chemical properties and the biological index (MBC, MBN, DHA) (Yang et al., 2011).

RESULTS AND DISCUSSIONS

The values of soil pH obtained in the study area ranged from 3.90 to 5.03 (0-15 cm), having a mean value of 4.63 across the samples analyzes (30 points). The mean value recorded for the pH was low (<5) across the study site and this could be a result of injury caused by proton pressure to the root of the plants in the study area. The result showed a significant difference with varying points or sample locations at p≤0.05. It can be concluded that the soil in the study area is very strongly acidic to a strong acidic and the acidity of the soil in the study area indicates the presence of the exchangeable hydrogen, aluminum and organic compounds (organic acids) that contains fulvic and humic acid (Andriesse 1974). The coefficient of variation (CV %) of the pH was 5.94 % which means there are low variation across the study area Aweto (1982). Organic carbon in the study area has values which ranges from 11.34-26.40 %, with the mean value of 17.64 % recorded across the study area. There were significant differences with varying sample points at p ≤0.05. The high organic carbon content was a result of plant cycling and carbon inputs from plant roots as well as plant residues (containing high nitrogen and carbon) in the topsoil (Jobbágy & Jackson 2000) of the study area. The values recorded for the exchangeable bases in all the sample collected ranged from 1.37-17.10, 0.70-7.75 and 0.25-0.91 $cmol_+$/kg for Ca, Mg and K respectively. The mean value for the exchangeable bases across the study site are 9.49, 3.05 and 0.39 $cmol_+$/kg for Ca, Mg and K respectively. Results showed that Ca is significantly different with varying location or sample points but Mg and K showed no significant different with varying location or sample points across the study area at p ≤0.05. The values recorded for the exchangeable bases were classified as moderate to low due to the acid nature of the peat and high aluminum content in the soil solution which impede their availability to plants (Arifin *et al.,* 2008; Abdu *et al.,* 2008), also, it could be a result of leaching, because peats are susceptible to leaching due to low content of clay and absence of mineral matters (Ahmed *et al.,* 2015).

The result also showed that Ca, and Mg are strongly influenced by pH, and result from the study revealed that Ca occupied most of the exchange site which is similar to study by (Lucas 1982). Available phosphorus (P_{av}) content in all the samples collected were classified as moderate to very low ranging from 25.80-118.50 mg/kg, with the mean value for the available P across the study area recorded as 58.36 mg/kg. Reports from previous studies showed that available P is always high at top layers compared to other depths, and this was a result of high content of organic matter. The low available P in the study area could be caused by immobilization by specific adsorption of the nutrients by Fe and or Al compound (Fageria & Baligar 2008).

The activity of the microbial biomass is commonly used to determine the characteristics of the microbiological status of the soil (Nannipieri et al., 1990), and also to ascertain definitely the effect of cultivation (Anderson & Domsch, 1993) and field management (Perrott et al., 1992) on soil microorganism. The values recorded for the microbial biomass carbon (MBC) across the study area ranged from 57.25 µgC/g – 1189.21 µgC/g. The average mean for all sample points was recorded as 516.06 µgC/g. The values for MBC across the study area were found to be high i.e >200 µgC/g (except some few points), the high content of MBC in the study area shows a greater accumulation of plant litters and fine roots which favors the growth of the microbial population (Marschner et al., 2003; Gu et al., 2009). The high MBC could also be result of the effect of manure applied to the soil in the study area by the farmers as reported by (Ritz et al., 1997; Garcia-Gil et al., 2000). The result obtained for the MBC was similar to the findings by Hao et al., (2008), that microbial biomass is greater in soils rich in organic manure (peats). Results obtained for MBC across the study area revealed that the values were within the range reported in some previous studies (Jenkinson & Powlson, 1976; Lynch & Panting, 1980; Ross et al., 1980;

Srivastiva & singh, 1988). The values obtained for the microbial biomass nitrogen (MBN) across the study area ranged from 1.46 µgN/g – 10.31 µgN/g, with the mean value for MBN across the study area was 4.29 µgN/g. The values obtained for the microbial biomass nitrogen (MBN) across the study area was low compared to the values recorded for the microbial biomass carbon (MBC). Jekinson & Ladd, (1981), reported that the microorganisms tends to differ in their nitrogen content than that of carbon content due to the stages of growth of microorganism. The low content of MBN recorded in this study could be a result of organic manure in the field and changes in microbial species or population. Acidic nature of the soil in the study area could also be the reason why the values of the MBN in the study area was low, because optimum growth for most microbes in the soil is best at a neutral pH (Kumar et al., 2017). Previous studies (Naher et al., 2013; Shahid et al., 2016; Wang et al., 2019b), reported that the imbalance in some major soil nutrients could be associated to the low content of the MBN in the study area. Although, the organic matter content was high in the study area, but the high content of soil organic matter is not reliable to predict the N response as high organic matter soil may still produce significantly low N response if N mineralization is slow. This is most common in a very acidic soils and peats.

The soil enzyme i.e the dehydrogenase activity (DHA) across the study area has values ranging from 0.19 µgTPF/1g/24hrs - 6.23 µgTPF/1g/24hrs, and the mean value for the DHA across the study area was recorded as 2.9 µgTPF/1g/24hrs. The dehydrogenase activity (DHA) represents the viable cells and it reflects the total range of oxidative activity of the soil microflora. It is also considered as a good indicator of microbial activities (Nannipierie et al., 1990). This study revealed that the values for DHA was low across the study area (30 points).

The low content of the DHA may be due to the effect of organic manure present in the field on the dehydrogenase because of the more decomposable components of manure on the metabolism of soil microorganism. It could also be a result of the acidic nature of the soil (Kumar et al., 2017). The result from the study did not show any consistent increase or decrease in the values of the microbial and enzyme activities across the study area. The fluctuation could be a result of the differences in the management practices by individual farmers owning different plots. Also the inconsistency could be related to the variation in the soil moisture, temperature, stages of plant growth and nutrient supply (Campbell et al., 1999).

The result for the microbial biomass (MBC and MBN), and the soil enzyme activity (DHA) across the study area showed a moderate variation as the coefficient of variation (CV %) recorded were as 67.82 %, 47.0 % and 60.12 % for microbial biomass carbon (MBC), microbial biomass nitrogen (MBN) and dehydrogenase activity (DHA) respectively. Results obtained in this study showed that the microbial biomass carbon (MBC) had a positive correlation with some major soil nutrients such as pH (r=0.37938, p=0.181), soil organic carbon (r=0.6039, p=0.222), Mg (r=0.5268, p=0.0532), Ca (r=0.24421, p=0.043), and Avail. P (0.15563, p=0.0595). Microbial biomass nitrogen (MBN) also showed a positive correlation with pH (r=0.14453, p=0.622), Ca (r=0.21689, p=0.0456), Mg (r=0.13580, p=0.0425), Avail. P (r=0.17668, p=0.054), but a negative correlation with soil organic matter (r=-0.6039, p=0.0222), and K (r=-0.19809, p=0.0497). The MBC and MBN showed a correlation (r=0.30157, p=0.2947). The correlation of the microbial biomass with the soil nutrients shows that microbial biomass can serve as a useful soil quality indicator with respect to the different land use and soil management. The positive correlation of the MBC and soil organic matter agrees with Yao et al., (2000) and cookson et al., (2007).

CONCLUSION

The cultivated Peat of the study area in Sepang was analyzed to assess and investigate their (MBC, MBN, DHA) effects on the peat quality of the study area for a sustainable agriculture. The results from this study showed some variations in chemical properties (pH, SOC, avail.P, Exchangeable bases) and biological properties (MBC, MBN, DHA) with varying points or locations across the study area. The pH of the peat in the study area was classified as very strongly acidic to strong acidic with the pH (H_2O) values ranging from 3.30 to 4.64 which is

expected of peat. The exchangeable bases (Ca, Mg, K) were classified to be low to moderate in the study area which was due to the acidity of the peat showing that they are also pH dependent.

High content of avail P at the top layer (0-15 cm) was related to the high organic matter content. The microbial carbon was high in the study area due to a greater accumulation of plant litters and fine roots which favors the growth of the microbial population, while the microbial biomass nitrogen and dehydrogenase activity were low across the study site, this was related to the acidic nature of the soil in the study area because optimum growth for most microbes in the soil is best at a neutral pH. The microbial index and enzyme activity (MBC, MBN, DHA), showed correlation with other soil nutrients and shows that they can serve as a useful soil quality indicator with respect to the different land use and soil management.

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Table 1: Descriptive statistics of the chemical properties across the study area (TKPM Ulu Chucoh)

	pH	SOC	Ca	Mg	K	P
plots		**%**		**cmol+/kg**		**mg/kg**
1	4.45	11.34	1.37	0.70	0.26	36.00
2	4.66	19.04	3.49	0.86	0.25	31.50
3	3.98	22.20	2.07	0.79	0.25	48.00
4	4.6	15.38	4.20	0.94	0.29	106.50
5	4.6	16.26	4.14	0.97	0.53	118.50
6	4.9	18.68	17.10	3.03	0.40	98.60
7	4.6	16.13	16.65	3.48	0.26	82.25
8	4.9	19.25	11.92	4.82	0.31	52.01
9	5.03	16.96	8.77	7.57	0.38	30.83
10	4.8	19.35	8.57	7.75	0.43	52.71
11	4.9	15.51	12.54	3.21	0.36	55.68
12	4.6	26.40	15.02	3.32	0.91	38.78
13	4.52	13.56	15.52	3.72	0.48	39.94
14	4.21	16.95	11.51	1.64	0.36	25.80
Mean	4.63	17.64	9.49	3.05	0.39	58.36
Stdev	0.27	3.56	5.41	2.28	0.17	29.37
Min	3.98	11.34	1.37	0.70	0.25	25.80
Max	5.03	26.40	17.10	7.75	0.91	118.50
CV %	5.94	20.17	57.06	74.55	42.98	50.32
p-value	<0.0001	<0.0001	0.0485	0.4635	0.0071	<0.0001

Table 2: Descriptive statistics showing the microbial biomass and enzyme activities across the study area (TKPM Ulu Chucoh)

	MBC	MBN	DHA
Plots	**µgC/g**	**µgN/g**	**µgTPF/1g/24hrs**
1	665.39	4.37	2.14
2	221.54	4.27	6.23
3	214.77	2.95	3.94
4	1104.27	5.83	0.89
5	140.00	4.32	1.88
6	508.30	5.89	4.41
7	602.31	4.27	5.82
8	566.29	4.37	2.93
9	883.45	4.47	3.10
10	1189.21	2.91	4.03
11	173.08	4.27	4.62
12	92.02	1.46	1.20
13	630.00	10.31	0.19
14	57.27	2.95	1.36
15	693.00	1.76	1.55
Mean	516.06	4.29	2.95
Stdev	350.01	2.02	1.78
Min	57.27	1.46	0.19
Max	1189.21	10.31	6.23
CV %	67.82	47.03	60.12

MBC- microbial biomass carbon, MBN- microbial biomass nitrogen, DHA- dehydrogenase