

NITRIFICATION POTENTIAL OF A TROPICAL PEAT SOIL UNDER OIL PALM (*ELAEIS GUINEENSIS* JACQ.) CULTIVATION AT DIFFERENT OPERATIONAL ZONES AND SOIL DEPTHS

Mohd Rizal Ariffin^{1*}, Osumanu Haruna Ahmed², Halimi Mohd Saud³, Isharudin Md Isa¹ and Mohd Nizar Khairudin⁴

¹Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia.

²Department of Crop Science, Faculty of Agriculture and Food Science, Universiti Putra Malaysia Bintulu Sarawak Campus, 97008 Bintulu, Sarawak, Malaysia.

³Department of Agriculture Technology, Faculty of Agriculture, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia.

⁴Oil Palm Agronomy, Tun Razak Agriculture Research Centre, FGV R & D Sdn. Bhd, 26400 Jengka, Pahang, Malaysia.

Abstract

The factors driving nitrification under oil palm (*Elaeis guineensis*.Jaq) cultivation in peat soil provide fundamental knowledge on managing available nitrogen (N) from losses. Characterization of operational zones and depths that are sensitive for N transformation is crucial for site specific N management. N fertilization in oil palm cultivation caused the inorganic N susceptible to losses through leaching and gasses emission. In order to understand nitrification in peat oil palm cultivation, specific area and depth that are susceptible to nitrification need to be characterized. Peat soil from three operational zones namely weeded circle (WC); frond heap (FH) and harvesting path (HP) was sampled up to six depths (0-10, 10-20, 20-30, 30-45, 45-60 and >60 cm) in an oil palm plantation in Perak, Malaysia. The samples was analyzed for potential nitrification rate (PNR), ammonium (NH₄⁺), nitrite (NO₂⁻) and nitrate (NO₃⁻) concentration. Results showed nitrification was found to be concentrated in the top soil WC zone as shown by the PNR (0.367 - 0.48 mg N kg⁻¹ day⁻¹). Deeper soil layer (>30cm for WC and >10 cm for both HP and FH) unable to show positive nitrification in PNR. Therefore, it can be assumed that most nitrifiers community are concentrated on the topsoil. It is also assumed that nitrate availability in the subsoil originated from vertical movements from the topsoil. Therefore it is believed nitrification in cultivated peat soil was concentrated in surface and fertilized soil due to favorable condition -lower moisture content and available substrate.

Key words: Nitrification, peat soil, oil palm, potential nitrification rate (PNR), Inorganic Nitrogen.

Introduction

Nitrification is a predominant process of the nitrogen cycle responsible for the availability of inorganic N forms to plants and soil microorganisms. Nitrification rates in soils are generally unpredictable depending on nitrifiers population (density and diversity) and upon various soil condition factors such as substrate concentration pH, moisture content, temperature and oxygen availability (Sahrawat, 2008). Characterization of nitrification potential in soil is crucial for site specific N management as it provides indication of maximum capacity for nitrifier

*Author for correspondence : E-mail: ra_rizal@upm.edu.my

populations in soil to convert ammonium into nitrate under the optimal conditions (Norton and Stark, 2010).

Tropical peat soils accounted over 8% (33-49 Mha) of total world's peat soil (Maltby and Immirzi, 1993) and most of it (>70%) are located in Malaysia and Indonesia. In undisturbed condition, peat soils/or organic soils constitute 10% of the world's soil Nitrogen (N) pool where N is conserved in organic compounds and nitrification process is limited or absent due to anaerobic condition. However, when peat soil is converted to oil palm, peat soil water table tables were normally lowered and maintained at 60 to 80 cm from surface (Mutert *et*

al., 1999). This condition create different moisture regime in soil depth and created oxidized zones on the surface soil which stimulate aerobic process such as nitrification and mineralization (Macrae *et al.*, 2012). Recent studies in the tropics have indicated that nitrification occurred despite limited O_2 availability for certain periods due to fluctuating redox regimes (Pett-Ridge *et al.*, 2013).

In addition, oil palm cultivation consisted three different areas around the palm tree which are harvesting path (HP), frond heap (FH) and weeded circle (WC). These zones are distinctly different in terms of agronomic practices, fertilizers input and soil physicochemical properties. HP served mainly as an access road for the plantation workers to harvest the oil palm bunches. FH on the other hand served as area between palms where piles of pruned fronds are placed. Part of this area receives fertilizer during fertilizer application. Legume cover crops, ferns and other under storey vegetation are allowed to grow in this area and therefore can contribute to organic matter build up. Finally, WC is the the area where fertilizers are applied close to the trunk especially for young palm (1-2 years from planting) This area is cleared from any vegetation to ease plantation worker to harvest the oil palm bunches, with radius 1 to 2 m around oil palm trunks. Therefore, it is expected that these zones have various implications on inorganic N availability and thus directly affects nitrification.

The purpose of this study was to determine the influence of operational zones (HP, FH and WC) and their depths on nitrification potential and inorganic N availability in relation to soil physicochemical parameters in peat soil cultivated with oil palm. It is hypothesized that nitrification potential and inorganic N availability varies in difference operational zones due to different agronomic practices in that areas.

Materials and Methods

Site description and soil sampling

Soil samples were collected in Ladang Sg. Samak, Malaysian Palm Oil Board (MPOB) Research Station in Teluk Intan, Perak, Malaysia ((3.49° N, 101.06° S). The site has mean annual temperature ranging from 25 to 32°C and the annual rainfall are between 1,219 to 2,128 mm with the driest month is July (<100mm) and the wettest, November. The total annual rain days ranged between 96 to 229 days. The soil was classified as Penor Series, Terric Sulfisaprist (Soil Survey Staff, 2014). The study area is flat, receives a moderately high and uniformly distributed rainfall and has a high soil water table which was kept 60 cm from the surface. Conventional fertilization has been carried out in WC zone which received 1 kg urea, 1 kg christmas island rock phosphate (CIRP) and 5 kg muriate of potash (MOP) palm⁻¹ year⁻¹ since 2007 divided three time per year. The palm age was 6-years-old at sampling time. Three replicates of soil samples were collected at depths of 0-10 cm, 10-20 cm, 20-30 cm, 30-45 cm, 45-60 cm and above 60 cm in WC and FH zones. However, the soil samples were only collected at 0-10 cm, 10-20 cm and 20-30 cm depths for HP zone. This is because in the soil in HP zone was very compacted at the deeper layer.

Soil physicochemical analyses

The soil samples were analyzed for pH (10 g) (1:10 w/w) (Metson, 1971), moisture content (50 g) (24 hr oven dried at 60°C); total carbon (TC) and total nitrogen (TN) (5 g) (LECO TruMac CNS Analyzer, USA). The NH_{4}^{+} , NO_{2}^{-} and NO_{2}^{-} were extracted using 10 g of soil with 2M KCl using a 1:10 soil:extractant ratio and a 1 hour end-over-end shake followed by filtration (Keeney and Nelson, 1982). Concentrations of NH⁺, NO⁻ and NO⁺ in solution were measured using auto analyzer with Cadmium-Copper reduction column (Lachat Part No. 50277). The summation of $NH_4^+ NO_2^-$ and NO_3^- was referred to as N_i. Nitrification potential (NP) was determined by shaking 15 g of field-moist soil sample with 100 mL working solution containing 1.5 mM NH⁺ and 1mM PO₄³⁻ (pH 7.20) (Hart *et al.*, 1994). The slurry was collected at 2, 6, 20 and 24 h; filtered and analyzed for NH_4^+ and NO_3^- concentration using auto analyzer with Cadmium-Copper reduction column (Lachat Part No. 50277).

Statistical analysis

Statistical Analysis Systems (SAS, version 9.4, Raleigh, NC) was used for statistical analysis of data. The data were subjected to analysis of variance (ANOVA) followed by means separation using the Tukey's Test at p < 0.05. Pearson correlation analysis was carried out for all parameters at p<0.05.

Results and Discussion

Soil physicochemical properties

The average soil moisture of all soil depths and operational zones ranged from 51.9 % to 84.3%. Soil moisture was different among the topsoil (0-10 cm) of operational zones (Table 1). FH have the highest soil moisture followed by WC and HP. In FH and WC area, soil moisture increased with increasing depth. Soil moisture increased with depth as results of lowering water table and the closer the depth to water table the higher the soil moisture (Berglund and Berglund, 2011).

Soil pH did not differ amongst operational zones at

Operational zone	Depth (cm)	Soil properties								
		pH Moisture		TC	TN	C/N				
		water	(%)	(%)	(%)	ratio				
FH	0-10	3.6dA	66.2dA	47.4aA	1.3aA	35.3cB				
	10-20	3.8dA	74.1bcA	48.9aA	1.1bA	42.3bA				
	20-30	3.7dA	72.2bcA	49.6aA	1.2bA	42.7bA				
	30-45	4.0c	76.9b	31.5b	0.7c	46.3b				
	45-60	4.3b	70.1cd	9.8c	0.2d	58.4a				
	>60	4.5a	84.2a	9.9c	0.2d	54.6a				
WC	0-10	3.7cA	55.7cB	49.2ab	1.2aA	40.5bA				
	10-20	3.8bcA	64.9bB	A49.6aA	1.2aA	42.2abA				
	20-30	3.7cA	72.6aA	49.7aA	1.1abA	45.4aA				
	30-45	3.8c	73.4a	41.7ab	0.9abc	49.1a				
	45-60	4.2ab	73.4a	35.1b	0.7c	56.8a				
	>60	4.30a	72.6a	35.2b	0.7bc	50.1a				
HP	0-10	3.6abA	51.9bC	46.6aA	1.1aB	43.3aA				
	10-20	3.7aA	67.6aB	47.8aA	1.0aA	44.3aA				
	20-30	3.5bA	69.0aA	38.8aA	0.9aA	42.1aA				
Means within column in each operational zone with different letter (s) indicate significant difference using Tukey's test at $p < 0.05$. Small capitals represent comparison between soil depths in the zone. Upper case letter represent										
comparison of the depth between the zones.										

 Table 1: Comparison of soil properties in different operational zones in oil palm plantation.

0-10, 10-20 and 20-30 cm. The average pH of FH, WC and HP ranged between 3.60 and 4.56 (Table 1). However, pH in the lower depth (above 30 cm) of FH was higher (above 4) compared to that in the upper FH surface soil. As for WC area, the pH of 45-60 cm and >60 cm was higher compared to other WC depths. A lower pH in the topsoil can be due to the combination of high urea fertilizer and proton release from nitrification



Fig. 1: Ammonium content in operational zones at different depths. (Means among bars in each operational zone with different letter(s) indicate significant difference using Tukey's test at p<0.05). Upper case letter represent comparison of the depth between the zones).

of NH_4^+ to NO_3^- (Anuar *et al.*, 2008), as a results of urea hydrolysis after urea fertilization. However, limiting factors such as lower input of N, high C/N ratio and high moisture content were present in the deeper soil layer (>20 cm), thus providing an unfavorable condition for mineralization process.

There were no significant differences in TC among the operational zones at 0-10, 10-20 and 20-30 cm (Table 1). In HP zone, TC were lower in the 30-45 and >60 cm depths compared to other upper depths. In contrast, TC was lower in the 45- > 60 cm in WC zone compared to the other depth in that area. Similar pattern was observed in TN where most of the N are located on the top (0-30 cm). The value of C/N ratio in the 0-10 cm layer (40.5%) is lower than those in > 20 cm depths.

NH_4^+ , NO_2^- , NO_3^- , NP and interrelationship

The WC topsoil (0-10 cm) contained 8.5 μ g g⁻¹ soil NH₄⁺ and it was two-fold higher compared to all other depths and zones (Fig. 1). Similar results also obtained for NO₃⁻ where it was highest compared to all other depths and zones at WC 0-10 cm (Fig. 1). Nitrite was not present in all of the soil samples and it can be concluded that NO₂⁻ ions were unstable in acid soils and quickly transformed into NO₃⁻ or lost to the environment via



Fig. 2: Nitrate content in operational zones at different depths. (Means among bars in each operational zone with different letter(s) indicate significant different using Tukey's test at p<0.05. Upper case letter represent comparison of the depth between the zones).

gasses (Shen *et al.*, 2003). WC alone had more than two-fold (15.6 μ g g⁻¹) of N₁ at the top 30 cm compared to FH (7.73 μ g g⁻¹) and HP (6.26 μ g g⁻¹) (Fig. 3). However, the combination of NO₃⁻ concentration in top 30 cm showed that WC contained just slightly higher NO₃⁻ with 10.9 μ g g⁻¹ compared to 9.38 μ g g⁻¹ (FH) and 9.2 μ g g⁻¹ (HP). If compared in terms of percentage, NO₃⁻ form accounted 59 % of the total inorganic N for HP, 54.5% for FH and 41% for WC. In 0-10 cm depth alone, WC contained three-folds higher inorganic N (14.7 μ g g⁻¹) compared to FH (3.61 μ g g⁻¹) and HP (4.48 μ g g⁻¹). However, up to 64% of the inorganic in 0-10 cm FH were in NH₄⁺ form. This is followed by WC with 58% and HP with 35%.

Nitrification in WC zone was found to take place in low pH condition with C/N ratio ranging from 40.5 to 42.3%. In 0-10 cm FH zone, the PNR result showed positive value $(0.21 \,\mu g \, g^{-1} \, \text{soil day}^{-1})$ which half the amount of PNR value in 0-10 cm of WC zone (0.43 µg g⁻¹ soil day⁻¹) (Fig. 4). NP at HP 0-10 cm area (0.13 μ g g⁻¹ soil day⁻¹), was lower compared with FH and WC. In this study, soil depth of 0-10 and 10-20 cm in WC zone showed to be an active site for nitrification based on PNR and NO_3^{-} availability. Trumbore, (2000) believed that soil surface with high plant debris, root exudates, temperature and lower moisture content was favorable for microbial decomposition such as nitrification. WC area received organic matter from leaf leachate, root exudates and loose oil palm fruits that accumulated at the WC area. The mineralization and nitrification were then stimulated by addition of reactive N in form of urea.

Detection of nitrification process by PNR only showed that there was 0.48 and 0.37 μ g g⁻¹ soil day⁻¹ released in 0-10 and 10-20 cm in WC zone, respectively



Fig. 3: Inorganic N content in operational zones at different depths. (Means among bars in each operational zone with different letter(s) indicate significant difference using Tukey's test at p<0.05. Upper case letter represent comparison of the depth between the zones).

(Fig. 4). The PNR value of soil layer 20-30 cm was a negative value (-0.88 μ g g⁻¹ soil day⁻¹), indicating nitrification process is not detectable in the lower soil depth (Fig. 4). However, NO_3^{-1} were available in deeper soil layer and this could be attributed to leaching of to the lower soil layer. In comparison with WC, FH area which had high OM build up and fewer disturbances can be assumed to be close to natural condition. It was believed that this area would have high amount of soluble N contributed by the OM. However, poor nitrification rate was observed in this area. The results were consistent with Vernimmen et al., (2007) study in low land forest types where regardless the amount of OM, without any fertilization, nitrification was very low or non-existent. Study by Westbrook and Devito, (2004) also observed lower nitrification activity in uncut peatland compared to cut boreal peatland which indicated minimal disturbance would preserve N from being nitrified. In HP zone, soil moisture content is likely to be the sole factor in regulating nitrification as the other parameters such as TC, TN and C/N ratio showed no significant difference in all soil depths. The NH_4^+ and NO_3^- concentration at 0-10 cm were lowest compared with WC and HP. However, it was enough to support nitrification as prove by the measured PNR of 0.13 µg g⁻¹ soil day⁻¹. Low NH₄⁺in HP could be due to relatively free vegetation and no N fertilizer was applied. Anuar et al., (2008) described HP as an area of poor soil structure and high bulk density and the soil was compacted due to high traffic during harvesting and maintenance operations. Therefore, there was a possibility that nitrification was restricted by O₂ diffusion which limit the aerobic microbial activity (Schjønning et al., 2011).



Fig. 4: Potential nitrification rates (PNR) in operational zones at different depths. (Means within bar with different small letter(s) indicate significant different using Tukey's test at p<0.05. Upper case letter represent comparison of the depth between the zones).

	Moisture (%)	\mathbf{pH}_{water}	TC	TN	C/N ratio	\mathbf{NH}_4^+	NO ₃ .	N _i	PNR
Moisture (%)	1.000								
pH _{water}	0.2750.166	1.000							
TC	0.0030.989	0.3030.125	1.000						
TN	-0.0770.699	0.1980.322	0.719<.0001	1.000					
C/N ratio	0.1040.604	0.0930.646	0.2930.138	-0.4460.0196	1.000				
$\mathrm{NH_4^+}$	-0.3060.121	0.2160.532	0.1800.369	0.1630.416	-0.0260.898	1.000			
NO ₃ -	-0.2720.169	0.0160.936	0.0920.648	-0.0300.880	0.0980.625	0.732<.0001	1.000		
Ni	-0.3120.112	0.0860.669	0.1530.445	0.0870.664	0.0280.889	0.952<.0001	0.905<.0001	1.000	
PNR	-0.6550.0002	0.0390.847	-0.0890.661	0.2410.227	-0.4450.0199	0.3280.095	0.2400.227	0.3120.113	1.000

Table 2: Pearson correlation coefficient between soil physicochemical properties.

*All the samples used in this study were included (n=45). TC = Total carbon, TN = Total nitrogen, NH_4^+ = ammonium, NO_3^- = nitrate, N_i = inorganic nitrogen and PNR = Potential Nitrification Rates.

The PNR technique provides an optimum condition for nitrification depending on the availability of nitrifiers community. PNR is also indicative of the size of the AOA/ AOB community (Hart, 1994). It was assumed that most of the nitrifiers and nitrification are concentrated at the top soil and substrate available area. The same observation had also been recorded by Abbasi et al., (2001) and Eilers et al., (2012) where nitrification decreased with soil depth. In addition, Fierer et al., (2003) also discovered that most of the gram-negative bacteria, fungi and protozoa were higher at the soil surface and substantially lower in the subsurface. Several reasons may be causing this vertical distribution, but it was believed to be largely derived from substrate availability. At the same time, other studies also pointed that O₂ and soil moisture can contribute to the difference in microbial activity at different depths (Persson and Wirén, 1995; Sahrawat, 2008).

Using Pearson correlation, soil NO₃⁻ was positively correlated with NH₄⁺ (Table 2). In this case, it was assumed that the supply of reactive N in the form of urea was the main factor that stimulated nitrification. It can be observed from the different NO₃⁻ content and PNR among the operational zones and soil depths. It is assumed that substrate limitation of N ceased nitrification rate in FH and HP. Nitrification is affected by availability of NH₄⁺ to the population of nitrifying microorganism, which in turn influenced by the quality of soil OM, especially C/ N ratio. High C/N ratio, leads to immobilization of NH₄⁺ (Sahrawat, 2008).

PNR was negatively correlated with soil moisture content (R= -0.655) and C/N ratio (R= -0.445) (Table 2). The higher the soil moisture, the lower PNR reading. Similar results was observed in few studies including (Agehara and Warncke, 2005; Macrae *et al.*, 2012; Westbrook and Devito, 2004). Nitrification is an aerobic

process, therefore, the higher the soil moisture, the lower nitrification because of depleting O_2 (Pihlatie *et al.*, 2004). This interpretation is supported by the observation from few studies indicating that drained peat had higher nitrification rates due to lower water content and improved aeration compared to un-drained peat (Andert *et al.*, 2011; Furukawa *et al.*, 2005; Jauhiainen *et al.*, 2012; Russow *et al.*, 2013). Oxygen content in the soil were reduced at higher soil moisture as most of the pore spaces are occupied by water and higher soil moisture also restricted the diffusion of atmospheric air into the soil. Thus, optimum conditions for both moisture and aeration are critical for nitrification to take place in the soil (Sahrawat, 2008).

Conclusion

Nitrification was found to be higher in the topsoil of operational zones particularly the WC zone which received N supply and had lower soil moisture and C/N ratio. Further study on WC zone is required to increase N-use efficiency where the loss of applied N was high because of nitrification. Vertical movement of NO_3^- from upper to deeper soil layer was observed in lower soil depths where NO_3^- accumulated. Regardless of the high organic N contents throughout the operation zones, supply of reactive N via fertilization is the key factors that stimulate availability of N, and PNR.

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