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## Nitrogen and phosphorous mineralization and soil microbial biomass carbon, nitrogen and phosphorous in a humid subtropical forest ecosystem of North Eastern India

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### Abstract

The humid subtropical forest ecosystem in Meghalaya, north eastern India is a closed canopy forest with high diversity of vascular plants, particularly, the impact of tree species on net N and P mineralization, and soil properties beneath their canopy were studied. The reduction of N and P in *Myrica* plot (First plot), could be due to sandy nature and lower organic matter and microorganisms that help in retention of  $\text{NH}_4^+\text{-N}$ ,  $\text{NO}_3^-\text{-N}$  and  $\text{PO}_4^{3-}\text{-P}$ . High MBC in *Rhododendron* plots (Second plot) favoured the growth of microbial population and accumulation of microbial biomass. The higher MBN in *Neolitsea* plots (Third plot) and mixed (Fourth plot) could be due to higher nutrient return. Significantly higher MBN in these two plots could be due to the higher TKN, soil pH and lower soil C/N and other micro-environmental factors. The high nitrification, net N mineralization and net P mineralization under *Neolitsea* and mixed plots corresponds to the chemical properties of soil as evident from high pH, TKN, Av.P, extractable inorganic P, intermediate SOC, inorganic N and lowest C/N ratio. The differences among the plots suggest that N and P transformation processes on the forest floor varied depending on the distribution pattern of dominant trees in the patch.

**Keywords:** Dominant trees, mineralization, soil microbial biomass

### Introduction

There is growing interest in understanding the linkages between forest dynamics and ecosystem processes because human induced environmental changes are likely to alter forest composition, net primary productivity and patterns of nutrient cycling. Changes in plant diversity and community composition could influence the composition of microbial communities' through various direct and indirect mechanisms (Kamei, 2010) [19]. Differences in tree species can directly affect soil microbial communities through variations in litter quality (e.g. N, P, lignin, C:N), root activities, complexity, and amount of organic input to soils, so microbes receive organic matter of varying quality across stands of different tree species. Plant characteristics also vary and can indirectly affect microbes via changes in the soil environment (Hooper *et al.*, 2000) [17]. Soil microbes associated with different tree species often have variable amounts of microbial biomass (Templer *et al.*, 2003) [37], rates of nitrification (Bruggemann *et al.*, 2005) [10], N mineralization (Bruggemann *et al.*, 2005) [10] and P mineralization.

Soil microbial biomass is related to other factors such as climate (Dyer *et al.*, 1990) [13], soil moisture (Taylor *et al.* 1999) [36], soil texture (Bauhas *et al.*, 1998) [5], plant productivity (Zak *et al.*, 1994) [44] and organic matter quality (Zak *et al.*, 1990) [43]. Soil microbes affect the availability of soil nitrogen and phosphorus for uptake or loss through the process of mineralization and immobilization. A tight coupling between these processes could limit the pool size of soil mineral N and P and accordingly reduce the potential nutrient loss from soils. The balance between mineralization and immobilization is related to the quality of soil organic matter as well as the attributes of soil microbial community. Some experiments have found positive effects of plant diversity on soil microbial processes (Stephan *et al.*, 2000) [34]. However, recent findings are supportive of the view that the effects of individual plant species are important determinants of ecosystem properties of organic matter decomposition

and nutrient recycling (Hooper and Vitousek, 1997) [18]. Many workers studied in a humid subtropical forest ecosystem under tree species diversity regarding, litter dynamics (Kamei, *et al.* (2008) [20], impact on soil properties, and nitrogen and phosphorous mineralization (Kamei, *et al.* 2009a) [21] and interspecific variation in leaf litter production, decomposition, nitrogen and phosphorous loss from decomposing leaves (Kamei, *et al.*, 2009b) [22]. The present study was conducted to examine the effect of tree diversity on soil microbial biomass C, N and P and N and P mineralization pattern in humid subtropical forest of Meghalaya.

## Materials and Methods

### Soil Microbial biomass

Microbial biomass in soils was determined from the upper soil layer (0-10 cm) at seasonal basis. During each sampling period, three samples were collected using soil borer (10 cm diameter) from each of the permanent plots. The samples were sealed in polythene bags in field and brought to laboratory. They were bulked to obtain composite samples for each plot. They were sieved through 2 mm-mesh screen to remove stones, roots and plant debris and used in field moist condition for the determination of microbial biomass carbon, nitrogen and phosphorus.

### Analysis

Microbial Biomass Carbon (MBC) was determined by chloroform fumigation extraction method (Vance *et al.*, 1987) [43]. The organic C in the extracts of fumigated and non-fumigated soil samples was determined by digesting 4 ml filtered extract with 0.0667 M  $K_2Cr_2O_7$  (1 ml) and 5 ml of  $H_2SO_4$  (98% acid) for 30 minutes. The digested sample was titrated with acidified ferrous ammonium sulphate solution using 0.3 ml (3 - 4 drops) of indicator (o-phenanthroline monohydrate and ferrous sulphate hexahydrate). The MBC was calculated as

$$MBC = 2.64 Ec$$

Where, Ec is the difference between the amount of organic C in the  $K_2SO_4$  extract of fumigated and non-fumigated soils, both expressed as  $\mu g g^{-1}$  dry soil and 2.64 is the relationship between biomass C as measured by fumigation incubation method and amount of C extracted by 0.5 M  $K_2SO_4$  after chloroform treatment.

Microbial Biomass Nitrogen (MBN) was determined by fumigation extraction method (Brookes *et al.*, 1985) [9] slightly modified by Okalebo *et al.* (1993) [28].

$$MBN (\mu g g^{-1} \text{ dry soil}) = N_f - N_o$$

Where,  $N_f$  = biomass N of fumigated sample;  $N_o$  = biomass N of non-fumigated sample)

Microbial Biomass Phosphorous (MBP) was determined by chloroform fumigation extraction method (Brookes *et al.*,

1982) [7]. The extracts were analyzed for inorganic P using ammonium- molybdenum blue method (Allen *et al.*, 1974) [1].

$$MBP = b-a/0.40$$

where, a = the amount of inorganic P ( $\mu g g^{-1}$ ) extracted from unfumigated soil

b = the amount of inorganic P ( $\mu g g^{-1}$ ) extracted from fumigated soil 0.40 = the fraction of biomass P mineralized and extracted in 0.5 M  $NaHCO_3$

### Nitrogen and Phosphorus mineralization

Nitrogen and Phosphorus mineralization were studied *in situ* by buried bags technique (Eno, 1960) [14] on monthly basis for one annual cycle during September 2004 to September 2005. Initial soil moisture content (SMC),  $NH_4^+$ -N,  $NO_3^-$ -N and  $PO_4^{3-}$ -P concentrations were determined within 24 hours of sample collection following the method outlined by Allen *et al.* (1974) [1]. Nitrification rates were calculated based on the changes in  $NO_3^-$ -N concentrations by subtracting the initial concentration from their respective final concentration. Net nitrogen mineralization was calculated as the sum of changes in the extractable  $NH_4^+$ -N and  $NO_3^-$ -N over one month period. The increase in the concentration of  $PO_4^{3-}$ -P during the field exposure is referred to as net phosphorus mineralization.

### Statistical analysis

The data was analyzed using two-way ANOVA to test the effect of seasons/months and tree species on soil microbial biomass, available nutrients and mineralization rates. Fisher LSD test was carried out to compare the mean values. Correlation analysis was carried out (Zar, 1974) [45].

## Results

### Microbial biomass carbon (MBC)

The MBC concentration ranged from 446 - 922  $\mu g g^{-1}$  in *Rhododendron*, 347 - 709  $\mu g g^{-1}$  in *Neolitsea*, 258 - 463  $\mu g g^{-1}$  in mixed and 315 - 541  $\mu g g^{-1}$  in *Myrica* plots (Table 1). The mean MBC was significantly ( $p < 0.001$ ) high in *Rhododendron* (700  $\mu g g^{-1}$ ) as compared to other three plots. However, no significant ( $p < 0.001$ ) variation was observed between mixed (390  $\mu g g^{-1}$ ) and *Myrica* plots (387  $\mu g g^{-1}$ ) (Table 5). The MBC showed a marked seasonality ( $p < 0.001$ ) in all the experimental plots. The values peaked during winter and trough during rainy season (Table 1). The percentage contribution of MBC to SOC (Soil Organic Carbon) ranged between 1.37 - 2.26%. Among the experimental plots, maximum contribution was observed in *Rhododendron* (2.26%) and minimum in the mixed plots (1.37%) (Table 5). However, the contribution of MBC to SOC was not significant ( $p < 0.001$ ) between *Myrica* and mixed plots.

**Table 1:** Seasonal variation in soil microbial biomass carbon ( $\mu g g^{-1}$ ) in different experimental plots ( $\pm$  SE).

Experimental plots	Autumn	Winter	Spring	Rainy	Autumn
<i>Myrica</i>	336.78 $\pm$ 22.98	541.38 $\pm$ 45.86	390.20 $\pm$ 27.00	314.93 $\pm$ 33.24	349.87 $\pm$ 30.74
<i>Rhododendron</i>	487.11 $\pm$ 24.29	922.17 $\pm$ 149.54	882.10 $\pm$ 39.40	446.48 $\pm$ 23.56	766.88 $\pm$ 76.32
<i>Neolitsea</i>	471.11 $\pm$ 26.19	708.80 $\pm$ 43.29	573.46 $\pm$ 38.22	346.96 $\pm$ 45.87	507.85 $\pm$ 38.18
Mixed	404.90 $\pm$ 29.49	462.76 $\pm$ 53.78	445.05 $\pm$ 38.35	257.92 $\pm$ 7.58	380.30 $\pm$ 36.82

**Microbial biomass nitrogen (MBN)**

The MBN concentration ranged from 21 - 89  $\mu\text{g g}^{-1}$ , and based on the values the experimental plots can be arranged in the order of mixed  $\geq$  *Neolitsea*  $\geq$  *Rhododendron*  $>$  *Myrica* (Table 5). There was no significant variations ( $p < 0.001$ ) between mixed and *Neolitsea* plots, as well as *Neolitsea* and

*Rhododendron* plots. The MBN concentration showed a similar seasonal pattern to that of MBC, with high values during winter and low during rainy season (Table 8.2). The percentage contribution of MBN to TKN (Total Kjeldahl Nitrogen) ranged between 0.44% and 0.47% and did not vary significantly ( $p = 0.77$ ) across the plots (Table 5).

**Table 2:** Seasonal variation in soil microbial biomass nitrogen ( $\mu\text{g g}^{-1}$ ) in different experimental plots ( $\pm$  SE).

Experimental plots	Autumn	Winter	Spring	Rainy	Autumn
<i>Myrica</i>	23.12 $\pm$ 3.06	37.31 $\pm$ 2.47	28.79 $\pm$ 1.46	21.49 $\pm$ 2.26	21.90 $\pm$ 3.72
<i>Rhododendron</i>	22.71 $\pm$ 2.93	55.96 $\pm$ 6.44	41.37 $\pm$ 5.06	27.58 $\pm$ 4.93	26.77 $\pm$ 3.72
<i>Neolitsea</i>	22.71 $\pm$ 2.66	62.86 $\pm$ 7.04	47.85 $\pm$ 1.62	24.34 $\pm$ 2.43	26.77 $\pm$ 2.81
Mixed	27.58 $\pm$ 3.25	89.21 $\pm$ 0.81	42.18 $\pm$ 2.15	21.09 $\pm$ 2.93	29.20 $\pm$ 4.87

**Microbial biomass Phosphorus (MBP)**

The MBP concentration varied between 3.45 - 23.43  $\mu\text{g g}^{-1}$  and the experimental plots showed a similar distribution pattern as MBN (Microbial Biomass Nitrogen) in the following order: mixed  $\geq$  *Neolitsea*  $>$  *Rhododendron*  $>$  *Myrica* (Table 5). The seasonality of MBP was similar to that of MBC and MBN in all the experimental plots, with

higher values during winter and minimum during rainy (Table 3). The percentage contribution of MBP to available phosphorus varied between 3.11% and 4.23%. The maximum contribution was observed in mixed plots (4.23%) while the contribution did not vary significantly ( $p < 0.001$ ) between the other three plots (Table 5).

**Table 3:** Seasonal variation in soil microbial biomass phosphorus ( $\mu\text{g g}^{-1}$ ) in different experimental plots ( $\pm$  SE).

Experimental plots	Autumn	Winter	Spring	Rainy	Autumn
<i>Myrica</i>	7.80 $\pm$ 0.54	10.61 $\pm$ 1.49	8.43 $\pm$ 0.51	3.45 $\pm$ 0.43	6.56 $\pm$ 1.25
<i>Rhododendron</i>	8.76 $\pm$ 0.76	14.93 $\pm$ 1.17	13.71 $\pm$ 2.80	8.33 $\pm$ 2.25	9.18 $\pm$ 0.6
<i>Neolitsea</i>	17.24 $\pm$ 1.89	22.28 $\pm$ 1.11	17.50 $\pm$ 0.74	11.39 $\pm$ 1.59	17.10 $\pm$ 1.69
Mixed	18.10 $\pm$ 2.78	23.43 $\pm$ 3.55	19.63 $\pm$ 3.97	8.23 $\pm$ 0.95	16.68 $\pm$ 4.07

**Table 4:** Two-way ANOVA showing effects of seasons and experimental plots on soil microbial biomass carbon (MBC  $\mu\text{g g}^{-1}$ ), microbial biomass nitrogen (MBN  $\mu\text{g g}^{-1}$ ) and microbial biomass phosphorus (MBP  $\mu\text{g g}^{-1}$ ).

Parameters	Source of variance	Degree of freedom	Calculated F value	Tabulated F value	Significance level
MBC	Seasons	4	49.43	4.6	<0.001
	Plots	3	94.11	5.4	<0.001
MBN	Seasons	4	75.10	4.6	<0.001
	Plots	3	15.28	5.4	<0.001
MBP	Seasons	4	41.16	4.6	<0.001
	Plots	3	90.52	5.4	<0.001

**Table 5:** Mean soil microbial biomass-carbon (MBC  $\mu\text{g g}^{-1}$ ), -nitrogen (MBN  $\mu\text{g g}^{-1}$ ) and -phosphorus (MBP  $\mu\text{g g}^{-1}$ ) and their contribution (%) to soil organic carbon (SOC), total nitrogen (TKN) and total available phosphorus (Av.P) in different experimental plots (each value is a mean of 45 replicates across 5 seasons).

Experimental plots	Mbc	Mbc /soc Ratio (%)	Mbn	Mbn/ Tkn Ratio (%)	Mbp	Mbp/ Av.p Ratio (%)
<i>Myrica</i>	386.63 <sup>a</sup>	1.48	26.52	0.46	7.37	3.11
<i>Rhododendron</i>	700.95	2.26	34.23 <sup>a</sup>	0.44	10.98	3.11
<i>Neolitsea</i>	521.64	1.83	36.42 <sup>a, b</sup>	0.45	17.10 <sup>a</sup>	3.44
Mixed	390.19 <sup>a</sup>	1.37	41.85 <sup>b</sup>	0.47	17.21 <sup>a</sup>	4.23
LSD ( $p < 0.001$ )	55.68		5.93		1.86	

<sup>a, b</sup> Values with similar superscripts in the column are not significant.

**Available inorganic nitrogen ((NH<sub>4</sub><sup>+</sup>- N + NO<sub>3</sub><sup>-</sup>-N)****Ammonium nitrogen concentration (NH<sub>4</sub><sup>+</sup>- N)**

The NH<sub>4</sub><sup>+</sup>- N concentration was significantly ( $p < 0.001$ ) high in *Rhododendron* while there was no significant variation between the other three plots. The average NH<sub>4</sub><sup>+</sup>-N values ranged from 10.22 to 11.40  $\mu\text{g g}^{-1}$  (Table 7). Marked seasonality was observed in all the plots with high concentration recorded in dry period (13 -17  $\mu\text{g g}^{-1}$ ) and low during wet period (4 -7  $\mu\text{g g}^{-1}$ ) (Figure 1).

**Nitrate nitrogen concentration (NO<sub>3</sub><sup>-</sup>-N)**

The NO<sub>3</sub><sup>-</sup>-N concentration varies from 7.24 - 8.63  $\mu\text{g g}^{-1}$  (Table 7). The values did not differ significantly ( $p < 0.001$ ) between *Rhododendron* and *Neolitsea*, and mixed and

*Neolitsea* plots. The value was significantly low in *Myrica* plots. A similar seasonality to that of NH<sub>4</sub><sup>+</sup>- N was also observed in all the plots (Figure 2).

**Nitrification**

Among the experimental plots the mean nitrification rate ranged from 8.60 - 10.45  $\mu\text{g g}^{-1}\text{month}^{-1}$  and did not vary significantly ( $p < 0.001$ ) between *Rhododendron*, *Neolitsea* and mixed plots. However, *Myrica* plot (8.60  $\mu\text{g g}^{-1}\text{month}^{-1}$ ) recorded lowest nitrification rate (Table 7).

ANOVA of the result showed a marked ( $p < 0.001$ ) differences in nitrification rates across the months (Table 6). It was higher in all the experimental plots during September

(16 - 22  $\mu\text{g g}^{-1}\text{month}^{-1}$ ) except for *Rhododendron* plot during May (18.58  $\mu\text{g g}^{-1}\text{month}^{-1}$ ) and low during February (0.74 - 1.8  $\mu\text{g g}^{-1}\text{month}^{-1}$ ) in all the plots (Figure 3).

**Net N mineralization**

Amongst the experimental plots, net N mineralization was strongly affected by tree species ( $p<0.001$ ) ranging from 18.83 - 22.14  $\mu\text{g g}^{-1}\text{month}^{-1}$ . Net N mineralization varies

among the plots in the same order as net nitrification: low activities of N mineralization occurred under *Myrica* plot and the high rates occurred under *Rhododendron*, *Neolitsea* and Mixed plots (Table 7). The seasonal pattern of N mineralization was similar to nitrification, being maximum in the rainy (29 - 53  $\mu\text{g g}^{-1}\text{month}^{-1}$ ) and minimum in the winter season (1.65-3.60  $\mu\text{g g}^{-1}\text{month}^{-1}$ ) (Figure 4).

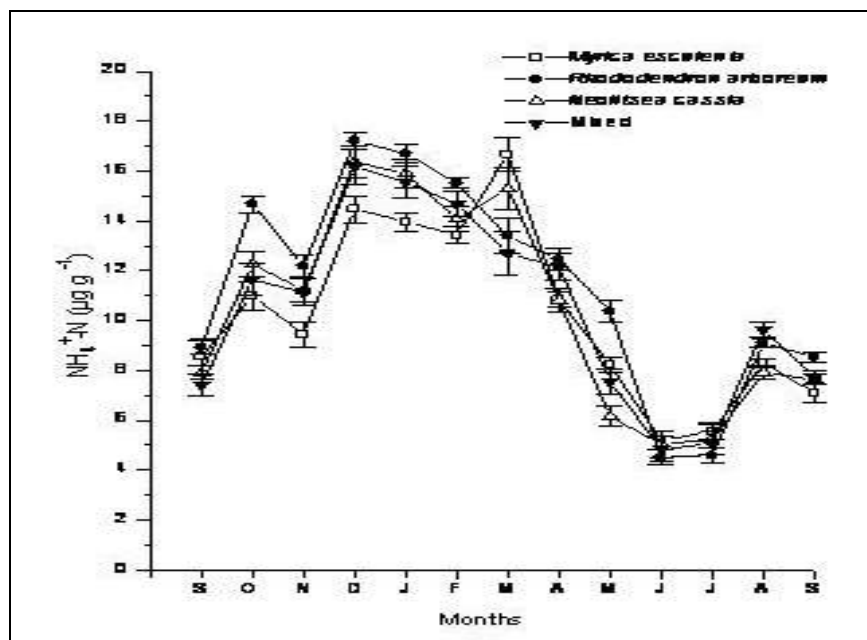


Fig 1: Variation in ammonium nitrogen (NH<sub>4</sub><sup>+</sup>-N) in the soil in different experimental plots.

Table 6: Two way ANOVA showing the effects of months and different experimental plots on ammonium nitrogen (NH<sub>4</sub><sup>+</sup>-N,  $\mu\text{g g}^{-1}$ ), nitrate nitrogen (NO<sub>3</sub><sup>-</sup>-N,  $\mu\text{g g}^{-1}$ ), phosphate phosphorus (PO<sub>4</sub><sup>3-</sup>-P,  $\mu\text{g g}^{-1}$ ) and N and P mineralization rate ( $\mu\text{g g}^{-1}\text{month}^{-1}$ ).

Parameters	Source of variation	Degrees of freedom	Calculated F value	Tabulated F value	Significance Level (P)
NH <sub>4</sub> <sup>+</sup> -N	Months	12	197.67	2.7	<0.001
	Plots	3	12.01	5.4	<0.001
NO <sub>3</sub> <sup>-</sup> -N	Months	12	672.26	2.7	<0.001
	Plots	3	43.63	5.4	<0.001
Nitrification	Months	12	218.59	2.7	<0.001
	Plots	3	12.68	5.4	<0.001
N mineralization	Months	12	315.59	2.7	<0.001
	Plots	3	12.31	5.4	<0.001
PO <sub>4</sub> <sup>3-</sup> -P	Months	12	372.49	2.7	<0.001
	Plots	3	149.23	5.4	<0.001
P mineralization	Months	12	981.66	2.7	<0.001
	Plots	3	48.76	5.4	<0.001

Table 7: Mean concentration of ammonium nitrogen (NH<sub>4</sub><sup>+</sup>-N,  $\mu\text{g g}^{-1}$ ), nitrate nitrogen (NO<sub>3</sub><sup>-</sup>-N,  $\mu\text{g g}^{-1}$ ), phosphate phosphorus (PO<sub>4</sub><sup>3-</sup>-P,  $\mu\text{g g}^{-1}$ ) and N and P mineralization rate ( $\mu\text{g g}^{-1}\text{month}^{-1}$ ) in different experimental plots [values are the means of 13 months across the year (n=117)].

Parameters	Myrica	Rhododendron	Neolitsea	Mixed	LSD(p<0.001)
NH <sub>4</sub> <sup>+</sup> -N	10.22 <sup>a,b</sup>	11.40	10.45 <sup>b,c</sup>	10.49 <sup>a,c</sup>	0.55
NO <sub>3</sub> <sup>-</sup> -N	7.24	8.15 <sup>a</sup>	8.63 <sup>b</sup>	8.39 <sup>a,b</sup>	0.34
Nitrification	8.60	9.84 <sup>a,b</sup>	10.45 <sup>b,c</sup>	10.22 <sup>a,c</sup>	0.85
N mineralization	18.83	21.88 <sup>a,b</sup>	22.14 <sup>b,c</sup>	22.01 <sup>a,c</sup>	1.66
PO <sub>4</sub> <sup>3-</sup> -P	6.57	7.42	8.45 <sup>a</sup>	8.42 <sup>a</sup>	0.27
P mineralization	4.54	5.24	5.87 <sup>a</sup>	5.74 <sup>a</sup>	0.32

<sup>a,b,c</sup> Values with similar superscript alphabets a, b and c in the rows are not significant.

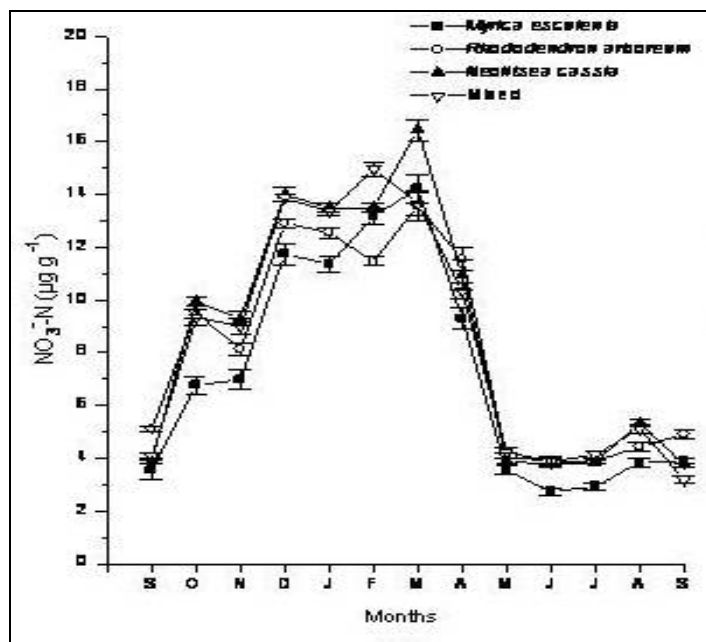


Fig 2: Variation in nitrate nitrogen (NO<sub>3</sub>-N) in the soil in different experimental plots.

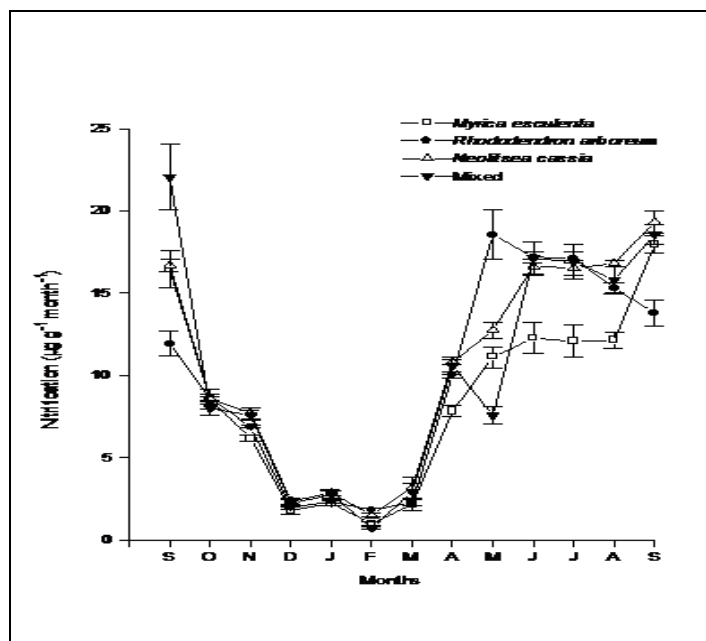


Fig 3: Net nitrification rate in different experimental plots.

**Inorganic phosphorus (PO<sub>4</sub><sup>3-</sup> - P)**

Two way ANOVA revealed a significant ( $p < 0.001$ ) difference in phosphate phosphorus (PO<sub>4</sub><sup>3-</sup> - P) concentration across the months and experimental plots (Table 6). The values ranged from 6.57-8.45 µg g<sup>-1</sup>month<sup>-1</sup> and based on the PO<sub>4</sub><sup>3-</sup>-P concentration the experimental plots can be arranged in the order: *Neolitsea* ≥ mixed > *Rhododendron* > *Myrica* (Table 7). The seasonal dynamics was similar to inorganic nitrogen (NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N) in all the plots (Figure 5). Highest values were recorded during the dry, winter period (9 -14 µg g<sup>-1</sup>month<sup>-1</sup>) and lowest during the wet, rainy period (3.62 - 4.92 µg g<sup>-1</sup>month<sup>-1</sup>).

**P mineralization**

The P mineralization rate ranged from 4.54 - 5.87 µg g<sup>-1</sup>month<sup>-1</sup>. Amongst the experimental plots the mean P mineralization rate was in the order of *Neolitsea* ≥ mixed > *Rhododendron* > *Myrica* (Table 7). The seasonal pattern of P mineralization was similar to N mineralization, being maximum in the rainy (12.51 - 17.09 µg g<sup>-1</sup>month<sup>-1</sup>) and minimum in the winter (0.24 - 0.36 µg g<sup>-1</sup>month<sup>-1</sup>) (Figure 6) season in all the plots.

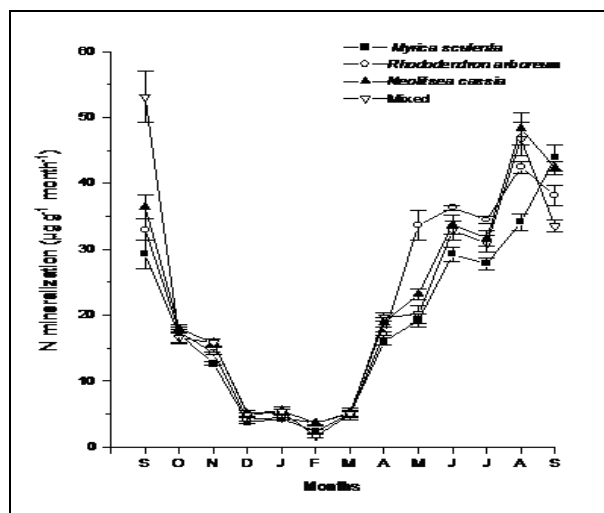


Fig 4: Net N mineralization rate in different experimental plots.

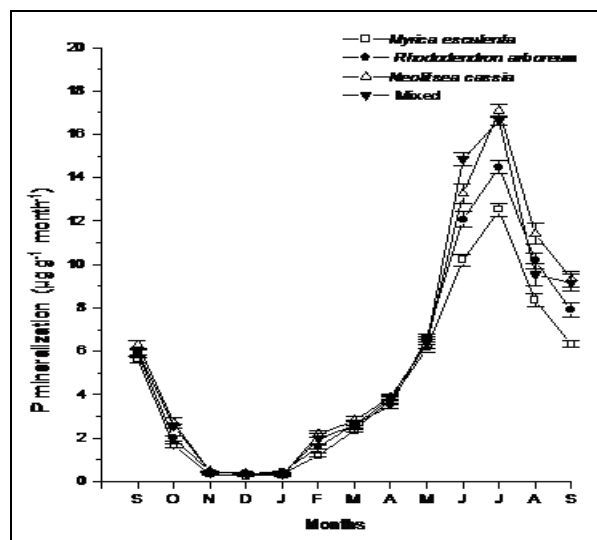


Fig 6: Net P mineralization rate in different experimental plots.

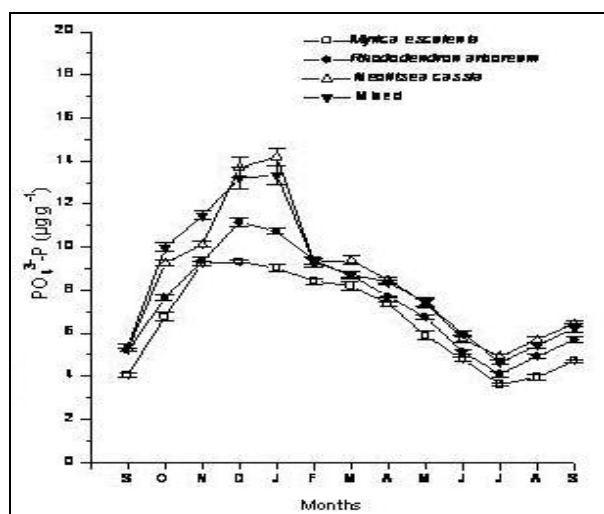


Fig 5: Variation in inorganic phosphorus ( $PO_4^{3-}$ -P) in the soil in different experimental plots.

**Discussion**

The seasonality of microbial biomass C, N and P with peak during winter and trough during rainy season is similar with the findings of subtropical forest from this region (Maithani *et al.*, 1996 [24]; Upadhaya, 2002 [40]; Upadhaya *et al.*, 2006[41]). The drop in microbial biomass in the beginning of the rainy season may be attributed to lysis and microbiorevory. During rainy season, temperature and soil moisture conditions were favourable for the growth of microbes indicated the period of rapid mineralization in soil and relatively greater demand for nutrients by plants. These lowered the microbial biomass values during rainy season. However, microbial biomass peaked during winter coincides with low soil moisture, air and soil temperature which indicates period of low microbial activity and greater nutrient retention in the soil microbial biomass. This is further supported by a strong negative correlation observed between SMC and soil microbial biomass C, N and P (Table 8). Also plant uptake of nutrients is greatly reduced on account of the death of majority of annuals and dormant growth phase of perennial species; nutrients are either immobilized in microbial biomass or accumulate in the soil.

The concentration of MBC obtained in the present study ( $257.92 - 922.17 \mu g g^{-1}$ ) is within the reported range ( $61 - 2000 \mu g g^{-1}$ ) for various temperate and tropical forest soils Vance *et al.*, 1987 [42]; Henrot and Robertson, 1994 [16]; Diaz-Ravina *et al.*, 1995 [11]. Maximum MBC in *Rhododendron* plots as compared to the other plots could be the result of greater input of organic matter and nutrients through litter and fine roots, which might have favoured the growth of microbial population and accumulation of microbial biomass. This difference in MBC could be attributed to the differences observed in the soil organic carbon which is evident from significant positive correlations between MBC and SOC ( $r = 0.35, p = 0.001$ ). Similar results were reported by Upadhaya (2002) [40] and Arunachalam *et al.* (1996) [2] and attributed it to an increase in organic matter content in forest stands owing to the greater accumulation of plant derived organic matter and microbial products.

Diaz-Ravina *et al.* (1988) [12] have reported that soil with low organic C usually has less microbial biomass and vice versa. Low soil organic carbon in mixed and *Myrica* plot could well explain the low MBC. Bauhas *et al.* (1998) [5] reported an effect of tree species on soil microbial biomass with concentrations of MBC and MBN lower beneath conifers than deciduous tree species, probably due to differences in foliage litter quality as microbial biomass decrease with increasing C/N or lignin/N ratios. Contrary to this, MBC showed a strong positive correlation with litter quality (lignin, C/N and lignin/N ratios) which may be due to lower litter quality in *Rhododendron* plots as compared to the other plots.

The contribution of MBC to SOC in the present study (1.37 - 2.26%) is well within the range reported from other tropical forest (1.5 - 5.3%; Theng *et al.*, 1989[38] and Luizao *et al.*, 1992[23]), tropical dry deciduous forest (1.6 - 3.6%; Srivastava and Singh 1989[33]), temperate forest soils (1.8 - 2.9%; Vance *et al.*, 1987[42]), pinewood and oakwood forests, (0.5 - 2%; Diaz-Ravina *et al.*, 1995 [11]. The concentration of MBN ( $21.09 - 89.21 \mu g g^{-1}$ ) in the present study was lower than that of soil of evergreen forest ( $42 - 242 \mu g g^{-1}$ ; Martikainen and Palojarvi, 1990[26], pinewood and oakwood forests ( $42 - 191 \mu g g^{-1}$ ; Diaz-Ravina *et al.*, 1995[11]). However, it was comparable to the values reported

from subtropical forest regrowth (37.79 - 123.85  $\mu\text{g g}^{-1}$ ; Maithani *et al.*, 1996 [24]) and tropical forest (27 - 93  $\mu\text{g g}^{-1}$ ; Barbhuiya, 2006 [4]) of this region.

The observed difference in concentration of MBN between the experimental plots could be related with the total Kjeldahl nitrogen (TKN) which is evident from the positive correlation (Table 8.8). The higher MBN in mixed and *Neolitsea* plots could be the result of higher nutrient return through litter and fine roots. Higher concentration of MBN in these two plots could be attributable to the higher TKN, pH and lower soil C/N. Although the resource quality of litter in mixed and *Neolitsea* plots are high, a strong correlation between litter quality (lignin, C/N and lignin/N ratios) and MBN were not observed (Table 8.8). The contribution of MBN to TKN was much lower (0.44 - 0.47%) compared to a range of forest soils (3.4 - 5.9%; Martikainen and Palojarvi 1990 [26]), pinewood and oakwood forests, (1.5 - 4.5%; Diaz-Ravina *et al.*, 1995 [11]), forest regrowth (7.3 - 8.3%; Maithani, *et al.*, 1996 [24]) and forest disturbance regime (1.3 - 1.7%; Barbhuiya, 2006 [4]). The MBP obtained in different experimental plots (3.45 - 23.43  $\mu\text{g g}^{-1}$ ) was lower compared to the reported range (5 - 67  $\mu\text{g g}^{-1}$ ) for woodland soils (Brookes *et al.*, 1984) [8]. However, the values were closed to dry

tropical ecosystems (9 - 28  $\mu\text{g g}^{-1}$ ; Srivastava, 1992 [32]). The higher concentration of MBP in mixed and *Neolitsea* plots could be due to the higher soil available P and pH as evident from the positive correlation (Table 8.8). Strong correlations between litter quality of tree species (lignin, C/N and lignin/N ratios) and MBP were observed (Table 8.8). This indicates that the effect of tree species on MBP is determined by the litter quality. The percentage contribution of MBP to available phosphorus (3.11 and 4.23%) is close to those reported by Brookes *et al.* (1984) [8] from deciduous woodland (4.7%), grassland (2 - 4.3%) and Arunachalam *et al.* (1996) [2] from a humid subtropical forest (1.4- 4.7%). Thus differences in the soil organic matter quantity and quality and soil pH mediated through a change in species composition in the plots may drive the differences in microbial biomass, given that greater concentrations of microbial biomass are generally found in soils of higher soil organic matter and pH (Blagodatskaya and Anderson, 1998[6], Bååth and Anderson, 2003 [3]) and that soil pH appears to influence microbial community composition (Bååth and Anderson 2003[3], Malmivaara-Lämsä and Fritze, 2003 [25]). This is supported by the strong correlation between soil properties and soil microbial biomass C, N and P (Table 8).

**Table 8:** Relationship between MBC, MBN and MBP and soil properties, microclimate and litter quality in the experimental plots.

Variable	Regression Equation	Degree of freedom	Correlation co-efficient (r)	P level
MBC vs. soil properties				
SOC	Y= -9.24 + 176.72X	179	0.35*	<0.001
TKN	Y= 381.93 + 152.92X	179	0.14*	<0.05
Av.P	Y= 2.95 + 0.002X	179	0.27*	<0.001
C:N	Y=537.26 - 0.41X	179	-0.11	0.14
pH	Y=-763.31+ 282.22X	179	0.44*	<0.001
vs. microclimate				
ST	Y= 994.95 - 31.23X	179	-0.54*	<0.001
SMC	Y=731.93 - 5.11X	179	-0.27*	<0.001
vs. litter quality				
N	Y= 887.47 - 279.03X	179	-0.41*	<0.001
P	Y= 507.72 - 148.18X	179	-0.01	0.87
C/N	Y=163.27 + 9.58X	179	0.41*	<0.001
Lignin	Y= 249.95 + 9.23X	179	0.35*	<0.001
MBN vs. soil properties				
SOC	Y= 14.86 + 7.00X	179	0.15*	<0.05
TKN	Y= 7.88 + 35.22X	179	0.35*	<0.01
Av.P	Y= 25.54 + 2.43X	179	0.18	0.02
C:N	Y= 57.22 - 5.64X	179	- 0.29*	<0.001
pH	Y=-199.55 + 52.35X	179	0.85*	<0.001
vs. microclimate				
ST	Y= 101.51 - 4.21X	179	-0.76*	<0.001
SMC	Y=79.03 - 0.97X	179	-0.56*	<0.001
vs. litter quality				
N	Y= 23.02 + 8.44X	179	0.13	0.07
P	Y= 16.59 + 341.59X	179	0.27*	<0.001
C/N	Y= 44.07 - 0.26X	179	-0.11	0.10

Lignin	Y= 42.77 – 0.29X	179	-0.12	0.10
MBP vs. soil properties				
SOC	Y= 2.66 + 0.02X	179	0.27*	<0.001
TKN	Y= 1.07 + 15.69X	179	0.46*	<0.001
Av.P	Y= 3.32 + 2.52X	179	0.55*	<0.001
C:N	Y= 21.38 – 2.08X	179	- 0.32*	<0.001
pH	Y=-41.77+ 12.27X	179	0.59*	<0.001
vs. microclimate				
ST	Y=28.12 – 0.94X	179	-0.50*	<0.001
SMC	Y=17.13 – 0.08X	179	-0.15*	<0.001
vs. litter quality				
N	Y= -2.65 + 11.39X	179	0.52*	<0.001
P	Y= -0.97 + 265.96X	179	0.63*	<0.001
C/N	Y= 26.10 – 0.36X	179	-0.48*	<0.001
Lignin	Y= 24.16 – 0.40X	179	-0.48*	<0.001

**Table 9:** Relationships between nitrification, N and P mineralization ( $\mu\text{g g}^{-1} \text{month}^{-1}$ ) and microclimate, soil properties, litter quality and microbial biomass in the experimental plots (n=36,  $p<0.001$ ).

Parameters	Regression Equation	Correlation co-efficient (r)	Significance level (p)
Nitrification vs. microclimate			
ST	Y= 26.31 – 1.07X	-0.80*	<0.001
SMC	Y= 4.19 + 0.13X	0.69*	<0.001
vs. soil properties			
pH	Y= -16.31 + 5.74X	0.75*	<0.001
C/N	Y= 14.64 – 1.26X	-0.79*	<0.001
SOC	Y= 6.32 + 1.20X	0.35*	<0.05
TKN	Y= 5.66 + 5.34X	0.78*	<0.001
Av. P	Y=7.11 + 0.68X	0.78*	<0.001
vs. litter quality			
N	Y= 8.00 + 1.28X	0.42*	<0.01
P	Y= 7.50 + 42.84X	0.73*	<0.001
C/N	Y= 11.14 – 0.03X	-0.36*	<0.05
Lignin	Y= 11.13 – 0.05X	-0.42*	<0.05
vs. MBN	Y= 7.23 + 0.07X	0.59*	<0.001
N mineralization vs. microclimate			
ST	Y= 52.30 – 2.02X	-0.79*	<0.001
SMC	Y= 10.13 + 0.26X	0.71*	<0.001
vs. soil properties			
pH	Y= -30.62 + 11.42X	0.78*	<0.001
C/N	Y= 30.05 – 2.31X	-0.76*	<0.001
SOC	Y= 12.16 + 3.14X	0.48*	<0.05
TKN	Y= 13.12 + 10.51X	0.81*	<0.001
Av. P	Y=16.25 + 1.27X	0.76*	<0.001
vs. litter quality			
N	Y=18.80 + 1.73X	0.30	0.07
P	Y= 17.35 + 72.71X	0.65*	<0.001
C/N	Y= 23.00 – 0.05X	-0.25	0.13
Lignin	Y= 22.37 – 0.05X	-0.24	0.15
vs. MBN	Y= 16.63 + 0.13X	0.56*	<0.001
P mineralization vs. microclimate			
ST	Y= 17.36 – 1.07X	-0.92*	<0.001
SMC	Y= 2.05 + 0.08X	0.64*	<0.001
vs. soil properties			
pH	Y= -12.22 + 3.87X	0.80*	<0.001
C/N	Y= 8.66 – 0.86X	-0.86*	<0.001
SOC	Y= 4.37 + 0.34X	0.15	0.37
TKN	Y= 2.86 + 3.22X	0.75*	<0.001
Av. P	Y=3.60 + 0.45X	0.80*	<0.001
vs. litter quality			
N	Y= 3.68 + 1.19X	0.61*	<0.001
P	Y= 3.51 + 34.49X	0.92*	<0.001
C/N	Y= 6.77 – 0.04X	-0.61*	<0.001
Lignin	Y= 6.49 – 0.04X	-0.57*	<0.001
vs. MBP	Y= 4.10 + 0.09X	0.79*	<0.001



### Available nutrients

The mineralized N and P are either immobilized in microbial biomass or accumulate in the soil as inorganic-N and P, resulting in a greater pool of mineral-N and P during dry period as compared to the wet period. Massive uptake of nutrients by vegetation from soil during the active growth season lowers the soil nutrient pool during the wet period of the year, leaching and runoff may also contribute to this reduction of nutrient pool. Schmitt and Randall (1994) [29] suggested that lower nitrate-N during rainy season could be because of  $\text{NO}_3^-$  losses via leaching and denitrification. Singh *et al.* (1991) [31] reported that plant uptake is high during wet period and immobilization in microbial biomass is low compared to dry period.

### Nitrification, N and P mineralization rates

The composition of the leaf litter of tree species of respective plots; mainly the C/N ratio as well as lignin/N ratio are important factors governing the soil microbial processes involved in decomposition and transformation of C, N and P containing compounds (Gower and Son, 1992 [15], Stump and Binkley, 1993 [35], Scott and Binkley, 1997 [30]). Lignin plays an important role in N transformation, thereby additionally binding inorganic nitrogen compounds and making them unavailable for plants and microbes (Thomas and Prescott, 2000) [39]. Litters with high ratios of lignin/N tend to decompose slowly (Melillo *et al.*, 1982) [27] and soils produced by those litters have been reported to have low N mineralization rates (Scott and Binkley, 1997) [30]. However, strong correlations between litter quality (N, P, lignin, C/N ratio) and nitrification and P mineralization rates were observed (Table 9). This indicates that the tree species litter quality alone do not determine the mineralization rate but there could be other factors like the micro environmental conditions, microbial biomass and soil nutrient status.

Canopy openness resulting in warm soil condition and low moisture content as observed in *Myrica* plots could be the result of high evaporation from the soil. Further, a strong correlation between microclimatic variables (soil moisture content and soil temperature) and ecosystem process such as nitrification, N and P mineralization rates suggests that these differences in the microclimatic conditions of the plots mediated through a change in species composition might be one of the potential causes for species specific differences in nitrification, N mineralization and P mineralization. Differences among experimental plots in MBN and MBP could be attributable to explain the variation in N and P transformation which is evident from the linear relationship between MBN and MBP with N and P mineralization respectively.

The high nitrification, net N mineralization and net P mineralization under *Neolitsea* and mixed plots corresponds to the chemical properties of soil as evident from high pH, TKN, Av. P, extractable inorganic P, intermediate SOC, inorganic N and lowest C/N ratio. These soil parameters showed intermediate values in *Rhododendron* plots as compared to high values in *Neolitsea* and mixed plots and low in *Myrica* plots. Thus tree species can exert a strong control on soil N and P transformation in forest ecosystems primarily through the quantity and quality of soil organic matter. The differences among the plots suggest that N and P transformation in forest floor vary from place to place

depending on the distribution pattern on the dominant trees in the patch.

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