

Consequences of habitat heterogeneity for microbial biomass in a dry tropical forest of Vindhyan hill, India

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Abstract. Seasonal and spatial dynamics of microbial C, N and P in response to organic matter accumulation in a matrix of troughs and flats on the floor of a dry tropical forest have been studied. Troughs had significantly higher microbial C, N and P than all other microsites. Flats had the minimum microbial C, N and P in all the seasons. Release of nitrogen during first four weeks of the rainy season from the microbial biomass was 85-118 $\mu\text{g g}^{-1}$ in the patchy microsites and 56-96 $\mu\text{g g}^{-1}$ in the non-patchy microsites. Release of phosphorus from microbial biomass during that same period was 47-67 $\mu\text{g g}^{-1}$ in the patchy microsites and 24-56 $\mu\text{g g}^{-1}$ in the non-patchy microsites. The study reveals that the habitat heterogeneity due to topographic depressions has led to the formation of hot spots (troughs or patchy microsites) on the forest floor with greater potential for sustaining microbial biomass than adjacent non-patchy microsites (flats).

Key words: Biomass C/N ratio, biomass C/P ratio, hot-spots, microbial C, N & P; N & P release, patchy microsites

Introduction

Microbial biomass is a major source of plant nutrients in dry tropical forest (Singh *et al.* 1989). Jenkinson (1987) stated that microbial biomass is the "eye of the needle through which all natural organic materials that enter must pass often more than once". Microorganisms have been considered as the most active fraction of soil organic matter which plays the central role in carbon flow and nutrient cycling in ecosystems. They constitute a transformation matrix for all the natural organic materials in the soil and act as a labile reservoir for plant available N, P and S (Jenkinson and Ladd 1981). Microbes are also the major nutrient sink during immobilization (Paul and Voroney 1980). Microbial populations are sensitive to change in the soil environment (Doran 1980). Powlson *et al.* (1987) opined that soil microbial biomass responds much more rapidly than the total organic matter to any change in organic inputs.

Measurement of microbial biomass is a valuable tool for understanding and predicting the long term effects of changes in soil conditions (Srivastava and Singh, 1991).

Microbial biomass C constitutes about 2-4 % of the total organic carbon. Plants even if fertilized, obtain a part of their N requirement through the microbial mineralization from various organic sources. The rapid turnover of microbial biomass (1-3 years) (Jenkinson and Powlson 1976; Paul and Voroney 1980, Schnurer *et al.* 1985) makes it an important source of plant nutrients. In a modelling exercise, Parton *et al.* (1983) found that most of the microbial N came from the fractions of soil organic matter which had the shortest turnover time. Productivity in most of the terrestrial ecosystems is N limited. Myrold (1987) reported that the microbial biomass contains a pool of N and other nutrients that are readily available to plants. Recognition of the importance of the microorganisms in the functioning of ecosystems has led to an increasing interest in measuring the nitrogen held in their biomass (Adams and Laughlin 1981, Ayanaba *et al.* 1976, Brookes *et al.* 1985 a, b; Carter and Rennie 1984 a, b; Inubushi and Watanabe 1986, 1987; Inubushi and Wada 1988; Ladd *et al.* 1981, 1983; Myrold 1987; Powlson *et al.* 1987; Ross 1987, 1988; Singh *et al.* 1989; Shen *et al.* 1984; Van Veen *et al.* 1987; Voroney *et al.* 1981; Voroney and Paul 1984; Amoto and Ladd 1980).

The present chapter elucidates the seasonal and spatial dynamics of microbial C, N and P in response to organic matter accumulation in a matrix of Troughs and Flats on the forest floor.

Study Area

The study sites are located near Kotwa in the district of Mirzapur (24°55' to 25°10' N lat. and 82°30' to 82°45' E long) within the Marihan range of East Mirzapur Forest Division on Vindhyan Mountain range in Uttar Pradesh supporting a large tract of the mixed dry deciduous forest. The climate is dry tropical showing marked seasonality such as rainy (mid June - September), winter (November - February) and summer (April - mid June). October and March comprise transition periods between rainy and winter and between winter and summer, respectively. About eight months of the year are dry and four months moist, the later receives about 86% of total annual rainfall. Mean annual temperature is 24°C with mean

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monthly values range between 17°C and 38°C. Annual rainfall is around 821 mm; monthly values range from 2.0 to 238.8 mm. Mean monthly values of temperature and rainfall for 9 years (1984 - 1992) are plotted in Fig. 1. Data were collected at Berkachha farm of the Banaras Hindu University and Kotwa Forest Research Meteorological observatory, both located within 6 km of the study sites. The soils at the study sites on Vindhyan hills are ultisols derived from Kaimur sandstone (Dhandraul Orthro-quartzites) and reddish brown in colour, coarse sandy loam in texture and slightly acidic in reaction. pH varies between 6.2 and 6.3 and soil moisture ranges from 2.5% during summer to 20.0% during rainy season (Raghubanshi 1992).

Two study sites representative of the region's vegetation were selected based on a repeated reconnaissance of the area. One study site is comprised by the hill plateau and the other by the hill slope.

The vegetation of these study sites have been reported by Singh and Singh (1991). *Boswellia serrata* dominates the hill top with basal cover of 8.9 m² ha⁻¹ followed by *Acacia catechu* with a basal cover of 1.4 m² ha⁻¹. In mid slope, *Acatia catechu* is the dominating species with a basal cover of 1.45 m² ha⁻¹ along with *Lannea coromandelica* with a basal cover of 1.88 m² ha⁻¹.

The forest floor in both sites is characterised by a variety of topographic depressions leading to a matrix of flats (non-patchy microsities) and troughs (patchy microsities). The troughs averaged 0.8 m² in size and 6 cm in depth. Troughs are different in appearance from the flats due to accumulation of litter and other organic matter. The soils from the patchy microsities had significantly greater amounts of C, N, P, Ca and K than that from the non-patchy microsities. Soil C/N ratio was 14.2 in the patchy microsities and 14.8 in the non-patchy microsities (Roy and Singh 1994).

For the present study one 200m x 200 m permanent plot was located on the hillslope and another plot of the same size on the hill top. The patchy microsities and non-patchy microsities were identified and marked in each plot.

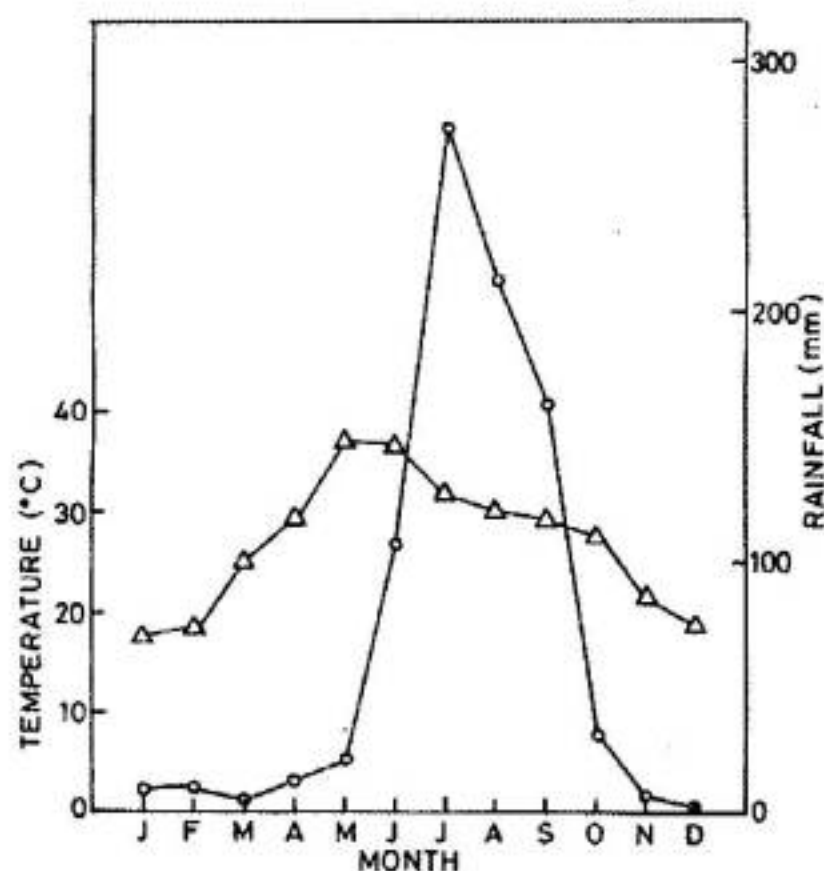


Fig. 1. Temperature and rainfall values for the study area based on 9 years data (1984-1992). Circles represent rainfall, triangles represent temperature.

Methods

Soil sampling

Soils were collected from the upper 10 cm layer randomly from the patchy (troughs) and adjoining non-patchy (flats) microsities of the hill top and the hill slope during summer (April/May), winter (December/January) and rainy (August) seasons. Eight soil samples were collected from each type of microsite separately for the hill top and the hill slope sites. Large pieces of plant material were removed and the soil samples were mixed together and from this composite stock five sub-samples for each microsite type were drawn for further analysis.

Microbial biomass C estimation

Microbial biomass C was estimated on the field moist soil samples. Sieving could have some effect on biomass estimation (Lynch and Panting 1981). The samples were stored for 7 - 10 days at room temperature (25 - 28°C) to settle down respiration (Srivastava and Singh 1988). Soil microbial biomass C (MB-C) was estimated using chloroform fumigation incubation method (Jenkinson and Powlson 1976). Liquid chloroform (CHCl₃) was used (Srivastava and Singh 1988), subsequently removed, and the soil samples were incubated with 1 g unfumigated soil from the respective stock and were adjusted to 50 - 60% of their water holding capacity. The samples were subsequently held at 27±2°C in airtight and leakproof aluminium cabinets, each of which contained 2 beakers one having 50 ml 1N NaOH and the other 20 ml distilled water to compensate for the drying effect of alkali.

Carbon dioxide (CO₂) evolution from the fumigated soils was estimated for 0 - 10 (X) and 10-20 (Y) days by titrating the residual alkali (Tinsley *et al.* 1951). The carbon dioxide evolved from incubated samples during 10 - 20 days after fumigation was taken as the control (Chaussod and Nicolardat 1982). To calculate microbial C a Kc factor of 0.45 was used to convert net CO₂ - C production to biomass C. Factor signifies the proportion of microbial C mineralized during the first 10 days after fumigation (Jenkinson and Ladd 1981). MB-C was calculated as X-Y/Kc.

Microbial biomass N estimation

The microbial N was estimated by the CHCl₃ fumigation-extraction method (Brookes *et al.* 1985a, b). However, liquid CHCl₃ was used instead of vapour (Srivastava and Singh 1989). 25 g field-moist soil was saturated with purified liquid CHCl₃ for 18-20 hours. After fumigation the soil was extracted with 0.5 molar K₂SO₄ (1:4 soil: extractant) for 30 minutes. Unfumigated soil was also extracted in the same way. Soil extract (fumigated and unfumigated) was analysed immediately through Kjeldahl digestion and distillation. Soil extract (25 ml) was taken in a 300 ml digestion flask and to this was added CuSO₄ solution (0.2 M, 1 ml) and concentrated H₂SO₄ (10 ml). The solution was digested for 2-3 hours. 25 ml of H₂O was added and 50 ml 10 N NaOH was poured into it. Distillation was done in 5 ml 2% boric acid solution

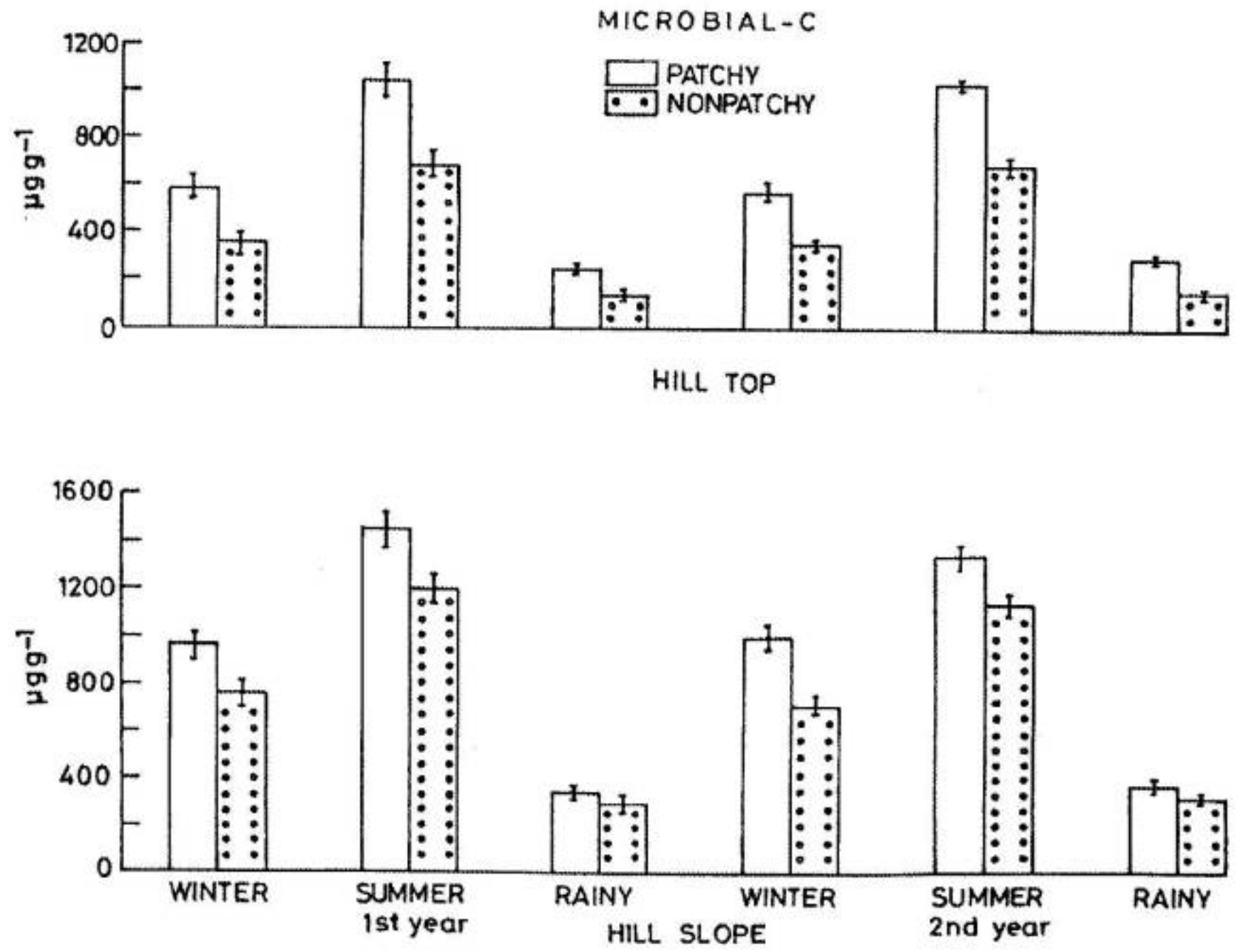


Fig. 2. Soil microbial C in the patchy and non-patchy microsites of the hill top and the hill slope sites in different seasons. (Bars represent ± 1 S.E.).

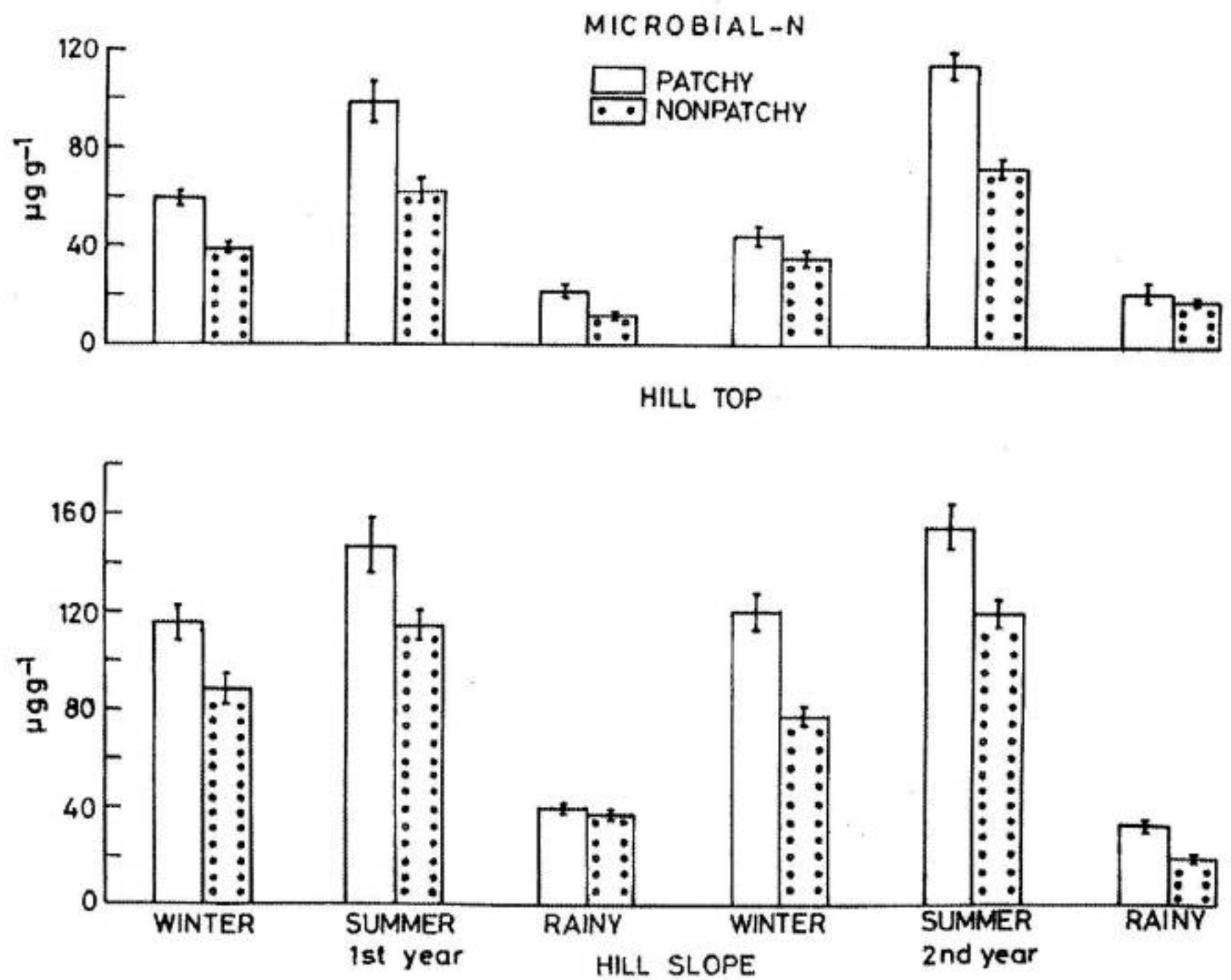


Fig. 3. Soil microbial N in the patchy and non-patchy microsites of the hill top and the hill slope sites in different seasons. (Bars represent ± 1 S.E.).

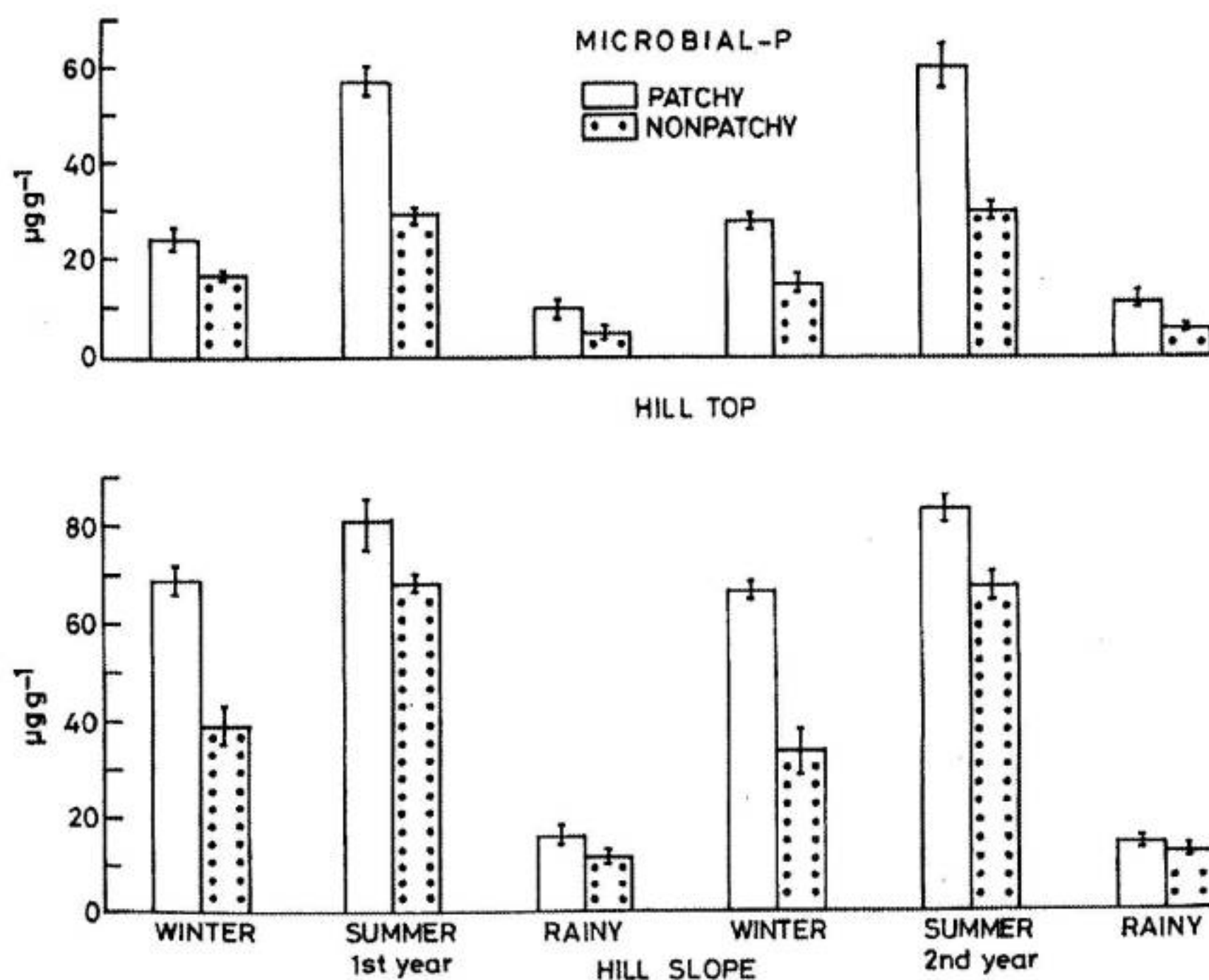


Fig. 4. Soil microbial P in the patchy and non-patchy microsites of the hill top and the hill slope sites in different seasons. (Bars represent ± 1 S.E.).

and titrated with 0.005 N H_2SO_4 using mixed indicator (Jackson 1958). Microbial biomass N was calculated as: $Biomass\ N = X - Y / K_n$, where X = total N in K_2SO_4 extract of fumigated soil, Y = total N in K_2SO_4 extract of unfumigated soil and K_n = fraction of biomass N extracted after $CHCl_3$ treatment. A K_n value of 0.54 (Brookes *et al.* 1985b) was taken by assuming that 54% of the microbial N was extracted in K_2SO_4 by $CHCl_3$ treatment.

Microbial biomass P estimation

Microbial biomass P was measured by chloroform fumigation-extraction method (Brookes *et al.* 1982), on the same field moist soil stock. Liquid chloroform was used (Chauhan *et al.* 1981; Headley and Stewart 1982; Srivastava and Singh 1988). Both fumigated and unfumigated soils were extracted in 100 ml 0.5 M $NaHCO_3$ solution (pH 8.5) for 30 min. Pi was determined by the ammonium molybdate-stannous chloride method (Olsen *et al.* 1954; Sparling *et al.* 1985). Microbial P was calculated as extra inorganic P released in fumigated soil (Pi released in fumigated soil - Pi released in unfumigated soil) divided by a Kp factor of 0.40, because it was assumed that 40% of P in the biomass is released as Pi. Phosphorus fixation during $NaHCO_3$ extraction was corrected by measuring the recovery of exogenously added Pi ($20\mu g\ g^{-1}$ soil).

All the results are expressed on an oven dry soil basis ($105^\circ C$ for 24 hr).

Results and Discussion

The seasonal pattern of soil microbial C, N and P was similar across the sites. Maximum biomass was measured during dry period and minimum in wet period (Table 1 and Fig. 2, 3 and 4). Patchy microsites of the hill slope had significantly higher microbial C, N and P than all other microsites. Non-patchy microsites of the hill top had the minimum microbial biomass C, N and P in all seasons. Analysis of variance revealed that the differences in the quantity of microbial C, N and P due to microsites and seasons were significant at $P < 0.05$.

Seasonal averages (Table 1) indicate that microbial C in the patches of the hill top ranged from 269-1042 $\mu g\ g^{-1}$ dry soil. The non-patchy microsites of hill top had much lower value (141-677 $\mu g\ g^{-1}$ dry soil), for microbial C. Patchy microsites of the hill slope contained 361-1394 $\mu g\ MB-C\ g^{-1}$ while the non-patchy soils of the hill slope had significantly lower values (313-1175 $\mu g\ MB-C\ g^{-1}$ dry soil).

Microbial biomass N in soils of the patchy microsites ranged from 21-106 $\mu g\ g^{-1}$ on the hill top and from 32-150 $\mu g\ g^{-1}$ on the hill slope sites. Non-patchy microsites contained 12-68 $\mu g\ g^{-1}$ on the hill top and 24-120 $\mu g\ g^{-1}$ on the hill slope. Microbial N was highest in the patchy microsites of the hill slope and lowest in the non-patchy microsites of the hill top. Mean microbial biomass in the dry forest and savanna ranged from 31-88 $\mu g\ g^{-1}$ (Singh *et al.* 1989). In solonchic soils in Canada the biomass N ranged

from 15-68 $\mu\text{g g}^{-1}$ (Carter 1986b). Myrold (1987) in Oregon forests observed microbial biomass N in the range of 28.5-218 $\mu\text{g g}^{-1}$ dry soil.

Microbial P followed the same pattern as microbial C and N in all the microsites. It ranged from 11 - 56 $\mu\text{g g}^{-1}$ dry soil in the patchy microsites of the hill top. On the hill slope it was 15 - 82 $\mu\text{g g}^{-1}$ in the patchy microsites. In the non-patchy microsites MB-P ranged from 6 - 30 $\mu\text{g g}^{-1}$ on the hill top and 12-68 $\mu\text{g g}^{-1}$ on the hill slope. Maximum MB-P was thus recorded for the patchy microsites of the hill slope and minimum for the non-patchy microsites of the hill top. Srivastava (1989) reported that microbial P ranged from 9 $\mu\text{g g}^{-1}$ in 5-yr old mine spoil to 28 $\mu\text{g g}^{-1}$ in mixed forest of dry tropical environment.

The data showed that the calculated release of nitrogen during first four weeks of the rainy season from the microbial biomass was in the following order: 118 $\mu\text{g g}^{-1}$ in the patchy microsites of the hill slope, 96 $\mu\text{g g}^{-1}$ in the non-patchy microsites of hill slope, 85 $\mu\text{g g}^{-1}$ in the patchy microsite of the hill top and 56 $\mu\text{g g}^{-1}$ in the non-patchy microsite of the hill top.

Calculated release of phosphorus from microbial biomass was in the following order: 67 $\mu\text{g g}^{-1}$ in the patchy microsites of the hill slope, 56 $\mu\text{g g}^{-1}$ in the non-patchy microsite of the hill slope, 47 $\mu\text{g g}^{-1}$ in the patchy microsites of the hill top, 24 $\mu\text{g g}^{-1}$ in the non-patchy microsites of the hill top. Release of N and P was calculated following Srivastava and Singh (1991). Microbial C ($\mu\text{g g}^{-1}$) was significantly correlated with microbial N ($\mu\text{g g}^{-1}$) and P ($\mu\text{g g}^{-1}$) and also microbial N ($\mu\text{g g}^{-1}$) was significantly correlated with microbial P ($\mu\text{g g}^{-1}$) according to the following equations:

Patchy microsites of the hill top

$$\text{Microbial N} = -7.016 + 0.109 (\text{Microbial C}) \\ (r=0.99, P<0.01)$$

$$\text{Microbial P} = -7.269 + 0.62 (\text{Microbial C}) \\ (r=0.9, P<0.01)$$

$$\text{Microbial P} = -2.796 + 0.56 (\text{Microbial N}) \\ (r=0.98, P<0.01)$$

Non-patchy microsites of the hill top

$$\text{Microbial N} = -1.74 + 0.102 (\text{Microbial C}) \\ (r=0.99, P<0.01)$$

$$\text{Microbial P} = -0.59 + 0.044 (\text{Microbial C}) \\ (r=0.99, P<0.01)$$

$$\text{Microbial P} = 0.37 + 0.428 (\text{Microbial N}) \\ (r=0.99, P<0.01)$$

Patchy microsites of the hill slope

$$\text{Microbial N} = -5.56 + 0.116 (\text{Microbial C}) \\ (r=0.98, P<0.01)$$

$$\text{Microbial P} = -6.254 + 0.067 (\text{Microbial C}) \\ (r=0.97, P<0.01)$$

$$\text{Microbial P} = -2.270 + 0.57 (\text{Microbial N}) \\ (r=0.99, P<0.01)$$

Non-patchy microsites of the hill slope

$$\text{Microbial N} = 1.57 + 0.103 (\text{Microbial C}) \\ (r=0.94, P<0.01)$$

$$\text{Microbial P} = -4.207 + 0.06 (\text{Microbial C}) \\ (r=0.98, P<0.01)$$

$$\text{Microbial P} = -2.0142 + 0.56 (\text{Microbial N}) \\ (r=0.97, P<0.01)$$

Mean annual values for MB-C, N and P are given in Table 2. Maximum MB-C, N and P occurred in the patchy microsites of the hill slope and minimum in the non-patchy microsites of the hill top. In the present study MB-C was 3.47% of the total C, MB-N was 4.77% of the total N, and MB-P was 11.03% of the total P, in the patchy microsites of the hill top. In the non-patchy microsites of the hill top MB-C was 5.98% of the total C, MB-N was 8.67% of total N and MB-P was 8.94% of total P. In the hill slope patchy microsites, MB-C was 2.97% of the total C, MB-N was 4.76% of the total N, and MB-P was 15.27% of the total P. In the non-patchy microsites of the hill slope, MB-C was 4.58% of total C, 6.97% of total N was MB-N and 13.7% of total P was MB-P.

Microbial C was 1.1-2.7% of the total organic C (Lynch and Panting 1980a, b) in a clay soil. Srivastava (1992) reported that microbial C was 2.2-5.0% of the total organic C in a range of tropical soils. Jankinson and Ladd (1981) reported that microbial biomass C constitutes about 2-4% of the total soil organic carbon. Microbial N generally accounts for 2-6% of the total soil N (Brookes *et al.* 1985b); Carter (1986) indicated a variation of MB-N from 1.4-7% of the total N. Powlson *et al.* (1987) reported that MB-N varied from 0.9-2.1% of the total N for certain temperate arable soils. Srivastava and Singh (1989) reported that 10.4-20.9% of total soil organic P as biomass P in a range of tropical soils. Williams and Sparling (1984) found 7.0-22.0% of total soil P as biomass P in organic soils of Scotland.

Microbial biomass C/N ratio ranged from 9.7 in the non-patchy soils of the hill slope to 10.17 in the patchy microsites of the hill top sites. Srivastava (1989) reported a range for C/N ratios of 9.1-9.7 for tropical forest soils. Dalal and Mayer (1987) for Australian arable soils reported biomass C/N ratios from 8.7-13.3. Dalal and Mayer (1987) argued that C/N ratios of microbial biomass are not always comparable because the proportion of microbial biomass N mineralized during incubation (K_n) varied from 0.2-0.3 (Voroney and Paul 1984) to 0.68 (Shen *et al.* 1984). In the present measurement K_n value of 0.54 was used (Srivastava 1989; Brookes *et al.* 1985b).

Biomass C/P ratios ranged from 16.58-23.23 in the present study. Biomass C/P ratios ranged from 16.6-30.6 in a range of tropical soils (Srivastava 1989). Brookes *et al.* (1984) reported biomass C/P ratios of 10.6-35.9 in fifteen soils from grassland and cultivated fields in U.K. West *et al.* (1986) reported C/P ratios in the range of 5.2-21.7 (biomass P from 45-76 $\mu\text{g g}^{-1}$) for certain silt loam soils under grazed pastures in New Zealand. Srivastava and Singh (1988) found 2.1-5.5% in biomass.

The study reveals that in the present study sites the habitat heterogeneity has resulted in the formation of hot spots (patchy microsites or troughs) on the forest floor with greater potentiality for sustaining microbial biomass than the adjacent non-patchy microsites (flats). Brady (1984) stated that microorganisms are sensitive to their chemical environment. The soil with higher organic matter has higher microbial biomass (McGill *et al.* 1981; Van Veen and Frissel 1981; Bolton *et al.* 1985; Powlson *et al.* 1987).

In these troughs nutrient is also trapped through litter accumulation and *in situ* decomposition (Roy

Microsites	Winter	Summer	Rainy
Microbial-C			
Patch top	583±27	1042±31	269±13
Non-patch top	366±13	677±30	141±9
Patch slope	982±39	1394±42	361±22
Non-patch slope	739±36	1175±38	313±18
Microbial-N			
Patch top	58±3	106±5	21±2
Non-patch top	37±2	68±3	12±1
Patch slope	118±5	150±7	32±2
Non-patch slope	83±4	120±5	24±2
Microbial-P			
Patch top	26±1	58±4	11±1
Non-patch top	16±1	30±1	6±1
Patch slope	68±3	82±3	15±1
Non-patch slope	41±2	68±2	12±1

Table 1. Microbial C, N and P in patchy and non-patchy microsites (2 years average, $\mu\text{g g}^{-1}$ soil \pm 1 SE)

	Hill top		Hill slope	
	Patchy	Non-patchy	Patchy	Non-patchy
MB-C ($\mu\text{g g}^{-1}$)	631	395	912	742
MB-N ($\mu\text{g g}^{-1}$)	62	39	100	76
MB-P ($\mu\text{g g}^{-1}$)	32	17	55	40
MB-C/organic C (percent)	3.47	5.98	2.97	4.58
MB-N/total N (percent)	4.77	8.67	4.76	6.97
MB-P/total P (percent)	11.03	8.94	15.27	13.7
MB-C/MB-N	10.17	10.12	9.12	9.7
MB-C/MB-P	19.72	23.23	16.58	18.55

Table 2. Mean annual (3 season x 2 years) soil biomass in the patchy and non patchy microsites. MB-C=microbial C; MB-N=Microbial N; MB-P=microbial P.

and Singh 1994). Microbial biomass as releases greater plant available N and P during rainy season in these hot spots than in the flats makes it more significant in forest nutrient cycling. Singh *et al.* (1989) showed that the microbial biomass acts as the dynamic source of nutrients in dry tropical forest. The flush of nutrients from microbial biomass during first four weeks of rainy season is much greater than the release of nutrients from litter during the whole rainy season (Raghubanshi *et al.* 1990; Roy 1992), in this forest. Greater nutrient

availability in troughs from microbial biomass as well as it's greater potentiality to sustain microbes indicate the importance of the troughs in dry tropical forest floor where the general soil condition is extremely nutrient poor and the vegetation manifests ravenous nutrient demand during rainy season.

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