Microbiology and fertility in coconut-based mixed farming and coconut monocropping systems

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Soil microflora, enzyme activities and microbial biomass in the root zone and rhizosphere of coconut-based mixed farming and coconut monocropping systems were studied. Bacterial counts were higher in the rhizosphere of coconut and Napier grass of mixed farming than in coconut monocropping. The microflora and enzyme activities decreased with the increase in soil depth. Urease, dehydrogenase and phosphatase enzyme activities were greater in the rhizosphere soil than in the root zone soil. The soil microbial biomass and the concentration of N and available P and K were higher in the mixed farming system.

Keywords: Mixed farming; Rhizosphere; Root zone; Soil biomass; Soil enzymes; Soil fertility

In pure stands of plantation coconuts under normal spacing of 7.5 × 7.5 m, about 75% of the total areas is not being effectively utilized. There is great scope for growing several inter- or mixed crops in coconut gardens, as shown by Nelliat and Shama Bhat (1979). The mixed farming system aims at increasing the productivity of coconut plantings and to integrate livestock production. It is known that the nature and activity of microflora and microfauna in soil depend upon the crop grown and the management practices (Clark, 1969); under a perennial crop they are likely to be almost constant, but the introduction of other crops could change this equilibrium. The influence on soil microflora has been reported in coconut-based mixed farming system (Potty and Jayasankar, 1976; Potty *et al.*, 1977), but the studies did not include soil enzymes and soil microbial biomass. In the present investigation, in addition to soil microflora, soil enzyme activity profiles, microbial biomass and fertility of the soils in the root zone (three depths) and the rhizosphere of coconut mixed farming and coconut monocropping systems were studied.

Materials and methods

Site and soil

The mixed farming experiment was started in 1972 with Napier grass Var.NB-21 (*Pennisetum purpureum* Schum.) in a 60-year-old coconut garden at the Central Plantation Crops Research Institute, Kasaragod, India. The latitude, longitude and height at mean sea level (MSL) are 12° 30′, 74° 52′ and 10 m, respectively. The mean annual rainfall is 3700 mm and the relative humidity ranges 60–94%. During the investigation, the maximum and minimum temperature ranges were 29.0–33.2 and 21.4–25.3°C. The spacing adopted for planting of coconut was 7.5 × 7.5 m (175 palms ha^{-1}). The basin region of the coconut palms was left without grass. Black pepper (*Piper nigrum* L.) was trailed on the trunks of the coconut palms, which were fertilized with 500:320:1200 g NPK $palm^{-1}$ $year^{-1}$, applied in two equal dressings during March and September; the grass received 20 kg N, 8 kg P_2O_5 and 60 kg K_2O ha^{-1} after every cutting, in the form of urea, superphosphate and muriate of potash, respectively. The grass was cut at 30–40-day intervals to feed the dairy animals. The cowdung was placed in a biogas plant and the slurry was recycled into the coconut/grass mixed stand. During the dry period (December–May), sprinkler irrigation (2 cm weekly) was provided to ensure the steady growth of the grass. The soil is sandy loam with low water holding capacity (30–32%). The physical properties of the soils are given in Table 1.

Collection of soil samples

Rhizosphere soils were collected from the coconut/grass intercrops and from coconut monocrop plots (five samples each) in October 1984, according to the method of Nair and Subba Rao (1977). The roots were carefully removed from the block of soil with minimal tearing. Then the roots were cut into pieces 2.5 cm long with a sterile scalpel and transferred to polythene bags.

Non-rhizosphere soil was collected from the interspaces of coconut monocrop plots (five samples).

Root-zone soils were collected by auger core samples from various crops, depending on the root

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Table 1 Physical properties of soil at three depths, coconut soils

Soil depth (cm)	Mechanical composition			
	Coarse sand (%)	Fine sand (%)	Silt (%)	Clay (%)
0–25	75.6	10.2	1.0	12.8
26–50	62.2	8.8	2.0	16.0
51–100	86.8	2.0	3.2	10.9

spread. The samples were collected from three depths, 0–25, 26–50 and 51–100 cm. Soils were collected from four locations around the plant to obtain a composite sample. The root-zone soils around the coconuts were collected 60 cm away from the bole; in grass, soils were collected 10 cm from the base of the plant (three samples each). All samples were stored in polythene bags at 4°C for three weeks.

Soil microflora and enzyme activity

The soil dilution and plate count methods were employed to enumerate the rhizosphere and root-zone microflora (Pramer and Schmidt, 1964). The population counts of bacteria (10^5 and 10^6), fungi (10^3 and 10^4) and actinomycetes (10^4 and 10^5) were enumerated using soil-extract agar, Martin's Rose Bengal agar and Kuster's agar, respectively (three replicates for each dilution). The plates were incubated at 30°C and the counts of bacteria, fungi and actinomycetes taken after 48, 72 and 120 h of incubation, respectively. Their numbers were calculated gram^{-1} of dry soil. The urease, dehydrogenase and phosphatase activities were assayed for both the rhizosphere and root-zone soils.

Urease activity was determined by the procedure of Pancholy and Rice (1973), except that the ammonia evolved by hydrolysis of the urea in the reaction mixture was estimated by Nesslerization (Jackson, 1973). 10 g of the freshly collected soils (in duplicate) were placed in 250 ml Erlenmeyer flasks; 20 ml of phosphate buffer (pH 6.7) and 20 ml of 10% urea and 1 ml of toluene were added. The contents were shaken for 5 min and incubated at 30°C for 24 h. A control with 20 ml distilled water instead of the urea solution was run simultaneously. After filtering, the soil was treated with 1N KCl and the volume made up to 100 ml with distilled water. The amount of ammonia in one ml of filtrate was determined by Nesslerization.

Dehydrogenase activity was determined by the modified 2–3–5 triphenyl tetrazolium chloride (TTC) reduction procedure described by Casida *et al.* (1964). 10 g of freshly collected soil was dispensed in each of two sets of 100 ml test tubes; 200 mg of $CaCO_3$ was added, followed by 1 ml of 1.5% solution of TTC and 9 ml of distilled water to each tube. The content were mixed well, plugged with a rubber stopper and incubated at 32°C for 24 h. The 2–3–5 triphenyl formazon (TPF) formed was extracted with methanol and the amount read at 485 nm on a spectrophotometer.

Phosphatase activity was assayed by the procedure of Tabatabai and Bremner (1969). To 1 g of soil (2 mm sieve) in a 50 ml Erlenmeyer flask, 4 ml of citrate phosphate buffer (pH 6.5), 0.25 ml toluene and 1.0 ml of 0.115M aqueous solution of disodium *p*-nitrophenyl phosphate were added and incubated at 32°C for 1 h. The amount of *p*-nitrophenol released upon incubation was determined spectrophotometrically at 420 nm.

Soil microbial biomass

The soil microbial biomass was estimated by the chloroform fumigation technique (Jenkinson and Powlson, 1976). 50 g of soil, adjusted to field capacity, in a 100 ml beaker were incubated at 30°C prior to fumigation, carried out in large desiccators with chloroform. The control soils were kept in disiccators lined with moist filter paper. The soils were transferred to 500 ml glucose drip bottles. Fresh soils (300 mg) of corresponding soils were added as inoculum to all bottles, into which 10 ml of 0.5N NaOH in 25 ml test tubes was introduced to absorb the CO_2. After 10 days, the CO_2 evolved was determined by titrating the NaOH against 1N HCl. The bottles were reset with NaOH and incubated for another 10 days to determine biomass. The biomass C was determined by dividing the flush of CO_2-C by a K_c factor of 0.45 (Jenkinson and Ladd, 1981).

Soils were analysed for organic carbon, pH, total nitrogen, phosphorus (total and available) and potash (total and available) (Jackson, *ibid.*).

Results and discussion

The root zone and rhizosphere microflora of coconut mixed farming and coconut monocropping systems are presented in Table 2. The population density of the bacteria was higher in the rhizosphere of coconut/Napier grass of mixed farming than in the rhizosphere of monocropped coconut. The actinomycetes numbers were significantly greater in the coconut rhizosphere in the mixed farming system. The bacterial numbers were greater in the root zone of Napier grass than in the root zone of monocropped coconut. The rhizosphere regions recorded more numerous microorganisms compared with the root-zone soils, possibly due to root exudates in the rhizosphere region. The cultivation of fodder legumes and Napier grass in coconut gardens increased the soil microflora (Potty and Jayasankar, *ibid.*; Potty *et al.*, *ibid.*). It has been shown that a coconut/cacao mixed cropping also increased proliferation of microorganisms (Nair and Subba Rao, *ibid.*). In general, the reductions in bacterial, fungal and actinomycetes populations were observed with increased soil depth in the root zone of coconut and grass in mixed and monocropping systems.

Soil enzyme activity in the rhizosphere in most cases was higher than in the root zone soils (Table 3). Urease activity was greater in the rhizosphere of coconut/Napier grass in comparison with coconut monocropping. Phosphatase activity was higher only in the coconut rhizosphere in the mixed farming system. The root-zone soils of Napier grass recorded higher urease and dehydrogenase activities than for coconut in mixed farming and monocropping system, decreasing with soil depth. Similar observations were reported in rye grass and native pasture soils (Hoult and McGarity, 1986). The enzyme activities were low in the interspace soils of monocropped coconuts.

The soil microbial biomass estimation indicated a

Table 2 Microflora (CFU g^{-1} soil) in the root zone and rhizosphere of coconut mixed farming (MF) and coconut monocropping systems

Crop	Depth (cm)	Root zone[a] Bacteria ($\times 10^6$)	Root zone[a] Fungi ($\times 10^3$)	Root zone[a] Actinomycetes ($\times 10^5$)	Rhizosphere[b] Bacteria ($\times 10^6$)	Rhizosphere[b] Fungi ($\times 10^3$)	Rhizosphere[b] Actinomycetes ($\times 10^5$)
Coconut (MF)	0–25	8.03	9.33	9.50	12.25	4.55	2.05
	26–50	3.23	8.55	2.89			
	51–100	3.03	2.45	2.50			
Napier grass (MF)	0–25	8.73	3.22	5.23	9.28	2.92	0.30
	26–50	1.87	2.33	2.45			
	51–100	1.30	1.56	1.11			
Coconut, monocrop	0–25	2.27	2.27	3.67	8.87	4.87	0.31
	26–50	2.73	3.03	4.00			
	51–100	1.43	1.07	1.67			
Coconut, monocrop interspace[c]	0–25	1.27	0.47	4.33			
	26–50	0.67	0.77	2.33			
	51–100	0.37	0.77	2.00			
LSD ($P = 0.05$)					5.50	4.49	0.39
Crop		0.54	0.72	1.34			
Depth		0.47	0.62	1.16			
Crop × depth		0.94	1.24	2.32			

[a] Average of three replications; [b] average of five replications; [c] serial dilution for bacteria and actinomycetes is 10^5

Table 3 Soil enzyme activities in the root zone and rhizosphere of coconut-based mixed farming (MF) and coconut monocropping systems

Crop	Depth (cm)	Root zone[a] Urease[1]	Root zone[a] Dehydrogenase[2]	Root zone[a] Phosphatase[3]	Rhizosphere[b] Urease[1]	Rhizosphere[b] Dehydrogenase[2] Without glucose	Rhizosphere[b] Dehydrogenase[2] With glucose	Rhizosphere[b] Phosphatase[3]
Coconut (MF)	0–25	7.61	2.74	147.67	9.04	2.61	6.13	161.7
	26–50	6.55	1.28	59.67				
	51–100	4.78	–	29.67				
Napier grass (MF)	0–25	8.42	3.38	81.33	8.54	4.71	12.5	136.0
	26–50	6.32	1.95	38.00				
	51–100	2.55	–	15.67				
Coconut, monocrop	0–25	7.67	2.87	69.33	5.41	2.82	6.04	133.6
	26–50	5.44	1.46	54.33				
	51–100	2.04	–	34.67				
Coconut, monocrop interspace	0–25	3.62	1.89	29.83				
	26–50	3.80	0.83	24.00				
	51–100	1.96	–	15.17				
LSD ($P = 0.05$)					3.29	2.77	2.10	30.42
Crop		0.96	0.21	17.79				
Depth		0.83	0.15	15.40				
Crop × depth		1.66	0.29	30.81				

[a]Average of three replications; [b]average of five replications; [1], urease as µg of NH_4^+ g^{-1} of soil h^{-1}; [2], dehydrogenase as µg of Formazon g^{-1} of soil h^{-1}; and [3], phosphatase as µg *p*-nitrophenol g^{-1} of soil h^{-1}

higher biomass carbon in the root zone of coconut/Napier grass in the mixed farming than in coconut in the monocropping system (Table 4). The greater microbial biomass is due to the increased microbial load in the mixed farming system. The chemical analysis of soil samples, pH, organic carbon, total N, P, K and available P and K are given in Table 5. The organic carbon and total N contents of the soil were higher in the mixed farming system than in the coconut monocropping system. The addition of slurry from the biogas plant added considerable organic matter into the system, thereby increasing the various microbial and biochemical properties of the soil. The total P and available P were slightly

Table 4 Soil microbial biomass in the root zone of coconut-based mixed farming (MF) and coconut monocropping systems

System	Soil biomass (µg C g^{-1} soil)[a] 0–10 days	Soil biomass (µg C g^{-1} soil)[a] 10–20 days
Coconut (MF)	187.80	27.80
Napier grass (MF)	177.95	41.88
Coconut monocropping	111.22	19.46
LSD ($P = 0.05$)	*70.74*	*28.50*

[a]Average of five replications

Table 5 Chemical composition of soils of coconut-based mixed farming (MF) and coconut monocropping systems*

Crop	Depth (cm)	pH	Organic C (%)	Total N (%)	Phosphorus		Potassium	
					Total P (%)	Available P (μg g^{-1} soil	Total K (%)	Available K (μg g^{-1} soil)
Coconut (MF)	0–25	6.50	0.71	0.068	0.50	22	0.15	88
	26–50	5.10	0.50	0.053	0.36	18	0.23	97
	51–100	5.00	0.26	0.029	0.21	16	0.23	50
Napier grass (MF)	0–25	5.35	0.36	0.038	0.30	17	0.13	23
	26–50	5.30	0.19	0.021	0.35	18	0.23	32
	51–100	5.43	0.18	0.018	0.12	2	0.20	24
Coconut, monocrop	0–25	5.55	0.22	0.020	0.42	13	0.18	60
	26–50	5.28	0.15	0.015	0.31	12	0.19	56
	51–100	4.44	0.10	0.011	0.28	8	0.22	40
Coconut, monocrop interspace	0–25	5.48	0.18	0.018	0.30	10	0.17	27
	26–50	4.93	0.09	0.010	0.52	1	0.21	13
	51–100	4.80	0.06	0.006	0.40	1	0.17	23

*Average of three replications

higher in the mixed farming system.

Soil fertility depends to a large extent on the microbial profile of a soil. The organic recycling influences the microflora of the rhizosphere and root zones. Jacob Matthew and Mohamed Shaffee (1979) found that mixed farming in coconut had synergistic effects, increasing the coconut yield compared with coconut monocropping. The enhanced microbiological and biochemical properties in the mixed farming system may be one of the factors contributing to increased productivity. The recycling of cowdung from the biogas plant in the mixed farming system helps to maintain soil fertility at levels higher than in the coconut monocropping system. The greater root volume of crops per unit volume of soil adds more organic matter by way of dead roots. Our investigation has revealed the more abundant microflora and greater enzyme activities in the coconut-based mixed farming system than in the coconut monocropping system. The build-up in soil organic carbon, N and P indicates the improvement in the soil fertility in the mixed farming system.

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