เชื้อราในดินและรากไม้ในป่าดิบแล้งสะแกราช

Soil and Root Fungi in Sakaerat Dry Evergreen Forest

Poonpilai Wongseenin and Malee Sundhagul

Department of Biology, Kasetsart University and the Applied Scientific Research Corporation of Thailand, respectively.

ABSTRACT

Studies were made on soil and root fungi of the two different communities, open and dense, of a dry evergreen forest at the Applied Scientific Research Corporation of Thailand's Sakaerat Experiment Station in Northeastern Thailand.

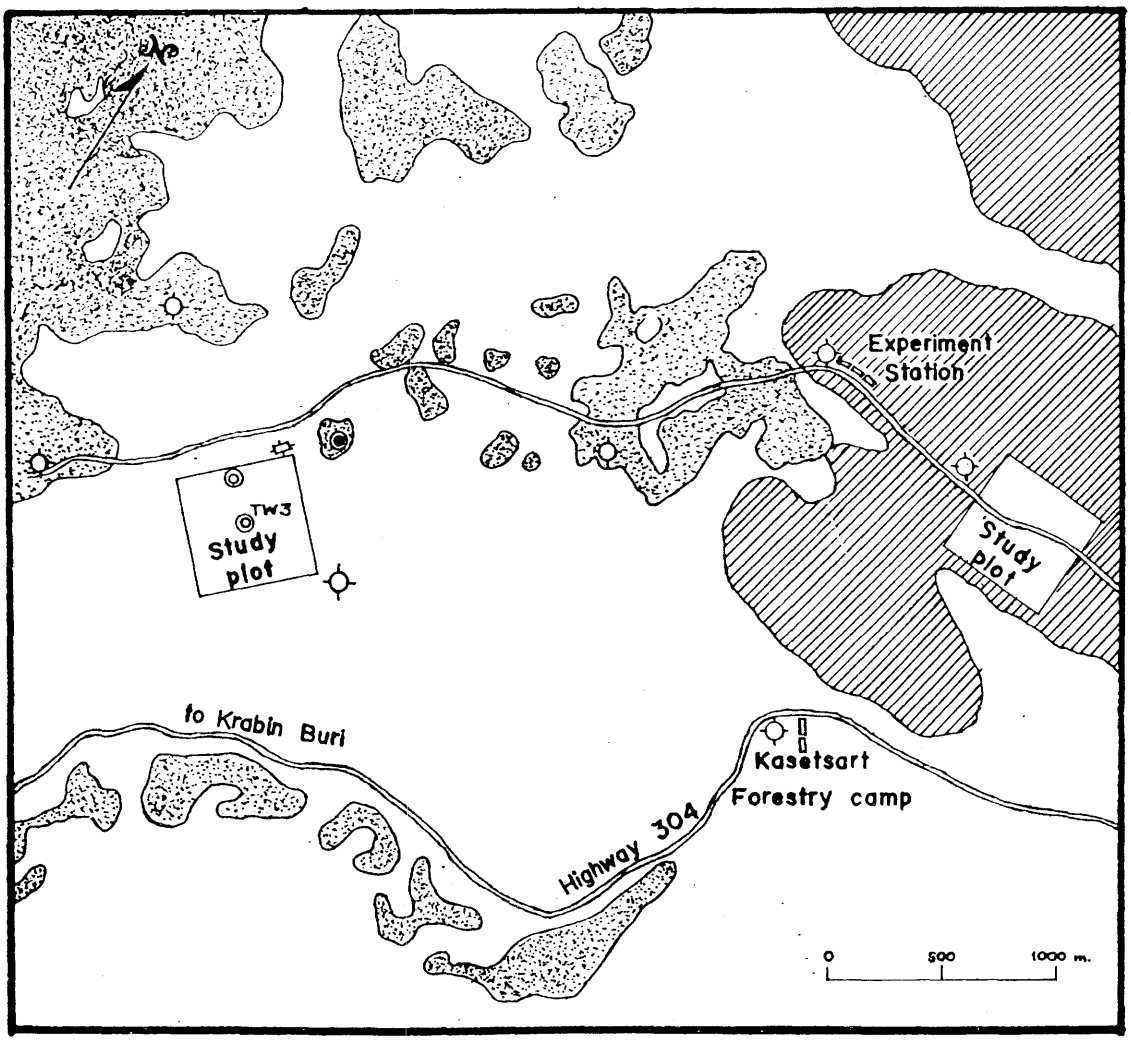
Fungal population of the root system, the rhizosphere and non-rhizosphere soil were determined. Isolation and identification of the fungi were made. Soil were also analysed for their physical and chemical characteristics. Attempts were also made to study the relationship between the fungal characteristic and the two type of the forest communities.

It was found that the two communities of the forest differed in fungal density as well as in fungal composition. The fungal numbers were higher in the dense community where the soil were richer in nutrients. Most of the fungi were found to be common to both types of forests. They are *Penicillium*, *Aspergillus*, *Trichoderma*, *Gliocladium*, *Gongronella* and *Scopulariopsis*. Certain other fungi, such as *Absidia* and members of Mycelia sterilia, were found to confine in the dense forest soil while *Cylindrocladium* were confined in the open forest soil.

Much attention has been given to the role of microorganisms in bringing about soil fertility and nutrient cycle. Ecological studies of fungi have been carried out by several workers. Warcup (17) reported that the number of fungal species and colonies in soil profile decreased with depth. Similar results were obtained by Eicker (4). He also found that the rate of decreasing of fungi in the soil were correlated with soil moisture. Correlation between fungal distribution and organic carbon content of the soil was reported by Mclennon (10).

The common species of fungi isolated from glassland soil, as reported by Warcup (17), were members of the genera *Penicillium* and *Mortierella*, follow by members of *Absidia*, *Thilaria*, *Trichoderma*, *Cephalosporium*, *Fusarium*, *Gliomastix*, *Mucor* and *Zygorrhynchus*. Mclennon (10) reported that *Mortierella raminiana* was the commonnest fungi, and *Penicillium* with it multiplicity of species was the most dominant type. On the other hand Christensen (3) reported that the high proportion of the species encountered were *Penicillium pavilli*, *Paecilomyces carneus*, *Gliocladium roseum*, *Penicillium steckii*, *P. brivicompectum*, *P. janthinellum* and *P. palitans*.

There are many factors which influences the members and types of organisms colonizing the immediate environ of the root known as rhizosphere (6). The spectra of such commonly occuring substances as amino acids, sugars, and organic acids in root exudates differed greatly among species and varieties (14). Part of



Grassland

Open forest

Dense forest

Tower in grassland

Micromet building

Living quarter

Mesomet station

Tower in forest

Fig. 1 Location of study plots within Sakaerat Experiment Station.

exudates found in soil is from sloughed-off root cap, injured cells, root hair and the autolysis of epidermal cells (15). Peterson (11) found that the number of microorganisms at the root zone increased as the soil moisture decreased. Plant age and soil type were also known to have influence upon the nature of the fungal flora of plant root.

The present study is conducted to determine the interrelationship between the environmental factors within two forest ecosystems of a dry evergreen forest in Northeast Thailand, the dense and the open forest, and the soil microflora, especially those inhabiting the roots and their immediate environ. The study area is within the Sakaerat Experimental Station of The Applied Scientific Research Corporation of Thailand, an area of 80 sq. km. The station is 60 km. south of Nakhon Ratchasima province. Two study plots, 500 meters by 500 m., were orbitrarily chosen to represent the dense and the open forest communities. The study plot in the dense forest has the center at tower 3 and study plot in the open forest is one km. from the living quarter along the road to the high way 304.

The location of the study plots within the station were shown in Figure I.

Materials and Methods

Soil samples were collected twice during the study period, the first collection was made in December 1969 and the second one in February 1970. Samples were collected from 16 collecting sites located within each of the two 500 by 500 m. study plots, one representing the dense community and the other the open community. At each site, soils were collected at two different levels, one at the depth of between 0—10 cm. (level A) and the other 10—20 cm. (level B).

The 16 samples collected from each level of each study plot were then deduced into four working samples. All working samples, after pooling, were then processed to determine the root's fungal density. Roots were first carefully shaken to remove adhering soil clumps (rhizosphere soils) before being treated with ten percent chlorox solution (13) to destroy adhering rhisosphere fungi. The cleaned roots were aseptically cut into several small pieces of five mm. in length. One hundred pieces were randomly picked and placed on Martin's medium agar plates, (8), twenty pieces to a plate.

All plates were incubated at ambient room temperature of 30±2 C. Pieces showing fungal growth were recorded as positive. Counts were made after six days of incubation. All fungi showing morphological differences were later picked and transferred to potato dextrose agar plates for further study and identification. Tentative identification of the fungal isolates were made according to Barron (1), Gilman (5), Mccallum (9) and Raper (12). The numbers of the rhizosphere and non-rhizosphere soils were made by the dilution plate method (16)

Soil moisture content was determined by drying the soil samples in a drying oven at 100 C over night. The pH was determined with a Beckman pH meter. Weight determination of root and soil from each working sample were separetely made from which the percentage of root was calculated. Chemical analysis for organic matter, phosphorus, potassium and the determination of water holding capacity of the soils were kindly carried out by Division of Agricultural Chemistry, Department of Agriculture, Ministry of Agriculture. The methods used were according to Jackson (7) and the procedure of Walkley Black (2).

Results and Discussion

Physical and chemical characteristics of the forest observation of the structure of the dry evergreen forest at Sakaerat Experiment Station revealed that, it composes of two distinct types of communities, the dense forest and the open forest. In the open forest trees are

sparsingly distributed and the forest floor is covered mainly with grasses and only some seedlings. In the dense forest, large number of seedlings as well as shrubs were observed on the forest floor underneath the densly covered canopy. The profile is slightly deeper in the dense forest than the open forest.

In the soil of the open forest, the root content was found to be higher than that in the soil of the dense forest. Roots in the open forest soil were fine roots while those in the dense forest soil were the mixture of fine, medium and thick roots. The high content of roots in the open forest soil might be attributable to the thick growth of grasses on the forest floor. The physical and chemical characteristics of the two types of forest are shown in Table 1.

Fungal Population. Soil fungi determination of total numbers of fungi in the soils of the dense and the open forests revealed that the fungal population was relatively denser in the dense forest than in the open ferest. However, the total number is changed in relation with time, being low in December and high in February. The drought period before the second collection in February probably induced high rate of sporulation and, consequently, resulted in increasing number of fungi. Total fungal counts of the soils of the two types of forests are given in Table 2.

Table 1. The physical and chemical characteristics of the soils.

Type of forest	Month	Level	Waterholding capacity (%)	pH	Organic matter (%)	K (ppm.)	P (ppm)	Moisture content (%)	Root content (% by wt.)
Dense	December	A	60.34	4.4	4.14	106.5	0.6	10.95	0.64
		B55.	55.28	4.8	2.88	78.0	1.1	11.22	0.25
	February	A	55.42	4.1	4.41	86.5	1.9	9.44	0.67
		B	52.88	4.3	2.37	75.5	1.6	9.70	0.29
Open	December	A	47.11	5.3	2.32	86.2	1.6	5.02	1.24
		B	51.14	5.6	2.18	94.0	1.0	6.85	0.64
	February	A	49.24	5.2	3.39	104.5	2.9	4.77	1.28
		B	49.65	5.1	1.97	82.0	1.3	6.98	0.76

Table 2. Number of soil fungi of dense and the open communities of the dry evergreen forest.

Type of forest	Month	Level	No. of fungi per gram dry soil				
			1	2	3	4	Average
Dense	December	A	14,400	75,500	88,000	159,600	84,275
		B	6,400	60,200	19,900	152,000	59,625
	February	A	220,300	95,700	283,900	160,400	190,000
		B	986,900	100,900	237,800	217,000	385,650
Open	December	A	8,900	37,200	34,000	52,000	33,025
		B	4,900	52,100	9,300	15,000	20,325
	February	A	26,700	82,800	187,900	150,500	111,975
		B	56,900	139,800	126,400	90,500	103,400

Rhizosphere Fungi. Determination of the number of fungi in the vicinity of the root surface revealed that, within equal amount of forest soils, there were more fungi in the rhizosphere region of the dense forest than in the open forest. This was found to be true for both soil levels and both collecting periods, December and February. (Table 3)

The root content alone did not appear to have direct relationship with the fungal density since it was found that there were more roots in the open forest soils than in the dense forest soil (Table 1). It may be said that the root system in the dense forest soils has more influence upon the fungal density than that in the open forest soils since it has been known that the substance released from roots may directly affect their root surface population (3).

Root Inhabiting Fungi. Studies were made on the fungi which live in the root without injuring them. The ratio of the fungal inhabited pieces of roots to sterile pieces was higher for the roots from the dense forest indicating that the large portion of the root system of the dense forest soil was inhabited with fungi (Table 4). While it was found that the total population of fungi in the soils generally decreases with depth such correlation was not observed in fungal density of the root system. Studies at different times of year showed that during the period of February there was a higher content of fungi in the root system than that observed during the month of December.

Fungal Characteristic. The total of 277 isolates were obtained during the study period (Table 5). In general, it was observed that the fungi of the dense forest soil were more diversified in morphological characteristic. Those isolates were tentatively classified into 18 morphological groups, most of the fungal strains were found to be common in both forest soil types. Only members of *Cylindrocladium* were found to be confined within the open forest soil (Table 6).

Table 3. Number of rhizosphere fungi of the dense and the open forests.

Type of forest	Month	Level	No. of fungi per gram dry soil				
			1	2	3	4	Average
Dense	December	A	15.7	31.8	65.1	347.1	114.9
		B	14.5	119.3	87.8	880.3	275.5
	February	A	215.	150.8	28.3	173.0	93.4
		B	20.3	33.2	28.8	45.0	31.8
Open	December	A	3.6	90.3	73.3	21.4	47.2
		B	33.8	93.5	5.1	14.9	36.8
	February	A	23.1	82.4	62.1	34.2	50.4
		B	7.3	20.5	22.9	11.9	15.6

Table 4 The percentage of root inhabited with fungi.

Type of forest	Month	level	1	2	3	4	Average	Ratio of inhabited to non-inhabited pieces
Dense	December	A	24.54	29.91	31.91	12.32	24.68	1.0 : 3.0
		B	14.16	39.47	23.43	40.31	29.34	1.0 : 2.5
	February	A	81.50	96.20	90.70	69.00	84.35	5.5 : 1.0
		B	73.80	3.60	96.40	89.00	65.70	2.0 : 1.0
Open	December	A	24.39	14.40	1.62	0.91	10.33	1.0 : 8.6
		B	7.63	33.33	5.40	10.63	14.25	1.0 : 6.0
	February	A	70.50	16.80	94.20	62.00	60.88	1.5 : 1.0
		B	68.20	52.20	46.00	60.00	56.60	1.3 : 1.0

Fungi frequently isolated from the open forest soil environment were members of *Penicillium*, *Aspergillus*, *Gliocladium* and *Trichoderma*. Common fungi from the dense forest soils included members of *Gongronella*, *Absidia* and *Cylindrocladium*. Sterile mycelia were found in both types of forest soils, and associated mainly with the root system. The frequency of occurence of those fungi is shown in Table 7.

Table 5. Number of fungal isolates obtained from the dense and the open forests.

Type of forest	Level	No. of fungal isolates			
		Soil	Rhizosphere	Root	Total
Dense	A	30	23	24	77
	B	34	23	14	71
Open	A	25	21	21	67
	B	27	19	16	62
Total		116	86	75	277
Percent		41.8	31.0	28.1	100.0

Table 6. Distribution of fungi within the soils of the Sakaerat forest.

Fungi	Dense forest	Open forest
Absidia sp.	+	–
Aspergillus terreus	+	+
Aspergillus fumigatus	+	+
Cylindrocladium scoparium	+	–
Gliocladium sp.	+	+
Gongronella sp.	+	+
Mycelia sterilia (1)	+	–
Mycelia sterilia (2)	+	+
Penicillium sp.	+	+
Penicillium sp.	+	+
Penicillium sp.	+	–
Penicillium sp.	+	+
Penicillium sp.	+	+
Penicillium sp.	+	+
Penicillium lilacinum	+	+
Penicillium sp.	+	+
Scopulariopsis sp.	+	+
Trichoderma sp.	+	+

Table 7. The occurrence of fungi in the dense and the open forests.

	Dense forest						Open forest					
	Soil		Rhizosphere		Root		Soil		Rhizosphere		Root	
	A	B	A	B	A	B	A	B	A	B	A	B
Penicillium sp.	1	3	2	1	—	1	—	—	—	1	—	1
Penicillium sp.	1	—	—	—	2	1	—	1	1	3	2	2
Gongronella sp.	2	2	—	2	—	—	—	1	—	—	1	1
Penicillium sp.	1	—	2	—	1	—	—	—	—	—	—	—
Aspergillus terreus	1	2	—	—	—	—	—	—	2	—	1	—
Scopulariopsis sp.	—	1	—	—	—	—	—	1	—	—	1	—
Penicillium sp.	—	1	1	—	—	1	—	—	1	—	—	—
Mycelia sterilia	—	—	—	—	4	2	—	—	—	—	—	—
Mycelia sterilia	—	2	—	—	1	2	—	—	—	—	1	—
Penicillium sp.	1	1	1	1	1	1	1	—	—	—	—	—
Penicillium sp.	—	1	—	—	—	—	1	—	1	1	—	1
Aspergillus fumigatus	1	—	—	—	1	—	1	—	—	—	—	—
Absidia sp.	—	—	2	1	—	—	—	—	—	—	—	—
Penicillium lilacinum	—	—	1	1	—	—	—	1	—	—	—	—
Gliocladium sp.	—	—	—	—	1	—	1	—	—	1	1	2
Cylindrocladium scoparium	—	—	—	—	1	1	—	—	—	—	—	—
Penicillium sp.	—	2	—	—	1	—	1	—	—	—	—	—
Trichoderma sp.	2	2	—	—	—	—	2	2	—	—	—	—

1, 2, 3 and 4 designate the numbers of working samples containing the fungi. Fungi occured only one of the four samples was recorded as 1 and those occured in two as 2.

Literature Cited

1 BARRON, L.G. 1968. The genera of hyphomycetes from soil. Williams and Wilkins Co., Baltimore. 364 p.

2 CHAPMAN, H.D. and F.P. Parker. 1961. Method of analysis for soil plants and waters. Univ. of California. 309 p.

3 CHRISTENSEN, M. and M.P. Backers. 1961. New and noteworthy Penicillia from Wisconsin soil. Mycologia. 53:451-463.

4 EICKER, A. 1970. Vertical distribution of fungi in Zululand soil. Trans. Brit. Mycol. Soc. 55 : 45-57.

5 GILMAN, C.J. 1957. A manual of soil fungi. Iowa State Univ. Press, Iowa. 450 p.

6 HARLY, J.L. 1969. The biology of mycorrhiza, Plant Science monographs. Leonard Hill, London. 25 p.

7 JACKSON, M.L. 1962. Soil chemical analysis Prentice Hall, Inc., Eaglewood Cliffs, New Jersey. 498 p.

8 MARTIN, J.P. 1950. Use of acid, rose bengal and streptomycin in the plate method for estimating soil fungi. Soil Sci. 69:215-265.

9 MCCALLUM, E.G. 1963. A laboratory guide to fungi in polluted water and sewage water in treatment systems, Public Health Service Publ. No. 999 W-P-I, Ohio. 132 p.

10. MCLENNON, E.I. and C.D. Sophie. 1954. The ecology of soil fungi of an Australia heathland. Australian J. Bot. 2:220-245.

11 PETERSON, E.A., et al. 1965. Microorganism in root zone in relation to soil moisture. Can. J. Microbiol. 2:483-489.

12 RAPER, K.B. and I.D. Funnel. 1965. The genus *Aspergillus*, Williams and Wilkins Co., Baltimore. 686 p.

13 RIKER, E.J. and S.R. Regina. 1936. Introduction to research on plant disease. J.S. Swiff, St. Louis. 117 p.

14 ROVIRA, A.D. 1965. Interaction between plant roots and soil microorganisms. Ann. Rev. of Microbiol. 19:241-266.

15 Schroth, M.N. and D.C. Hildebrand. 1964. Influence of plant exudates on root infecting fungi. Ann. Rev. Phytopath. 2:101-132.

16 WALTER, G. William, et al. 1960. Laboratory manual general bacteriology. Brown Co., Debugee, Iowa. 113 p.

17 WARCUP, J.H. 1951. The ecology of soil fungi. Trans. Brit. Mycol. Soc. 38: 376-399.