Proc. Indian natn. Sci. Acad. B 45 No. 6 pp 648-659 (1979)

Root Decomposition

R R MISHRA
Department of Botany, School of Life Sciences, North-Eastern Hill University, Shillong 793 003

(Received 16 October 1978; after revision 27 January 1979)

Fungal flora of roots of *Triticum aestivum*, *Hordeum vulgare*, *Paspalum scrobiculatum*, *Echinochloa crusgalli* and *Pennisetum typhoides* was studied at different stages of decomposition. A detailed investigation on the decomposition of roots of *T. aestivum* under different soil amendments was carried out. Biochemical analysis of the root samples for cellulose, hemicellulose and lignin at different stages of decomposition was done. Total soluble sugars, total free amino acids and presence or absence of phytotoxic substances were also determined. Germination of 9 frequently occurring fungal species from the different roots was studied in the root extract of all the plants.

Key Words: Root Decomposition, Microbe, Phytotaecins

Introduction

An understanding of the processes involved in the decomposition of the root system of the plants is essential to have an idea regarding the turnover of the underground root biomass. This is more important for the crop plants where the plant materials added to the field are mainly the roots after the crop is harvested. The root system left out in the field is main source of nutrient enrichment particularly where the artificial fertilization is neglected. The degree of decomposition is regulated by the spectrum of the microbial species associated with the root system and those in the soil.

Enough information is available regarding the decomposition of the aerial parts of the plants. Detailed and critical studies, however, on the succession of fungi on living and dead roots under different ecological conditions are limited. Waid (1957, 1974) has extensively investigated the root microflora and his studies may be considered as pioneer on the subject. The works of few more researchers like Chesters (1950 1960) and Marchant (1970) also need special mention.

Comparative study, however, on the succession of microfungi on different roots under indentical conditions in natural and

amended soil is lacking. This aspect has been taken up for the present study. The plants selected for the investigation being of great economic importance, the information presented in the paper, may be useful in understanding the nutrient release locked up in the plant material.

Materials and Methods

Fungal flora of roots of *Triticum aestivum*, *Hordeum vulgare*, *Paspalum scrobiculatum*, *Echinochloa crusgalli* and *Pennisetum typhoides* was studied at different stages of decomposition. Seeds of the above plants were sown in earthen pots in June 1972. Except for *P. typhoides*, the number of seeds sown per pot was six and 10 replicates were maintained for each plant. In *P. typhoides* only two plants were raised in each pot and 20 replicates were kept in this case. After the emergence of the seedlings the pots were transferred to a glass house at a temperature of 20±2°C. The pots were regularly supplied with equal amount of water. For the first 3 months the pots were watered at an interval of 4 days which was later changed to 6 days. The plants were left undisturbed for one year with regular watering. The moisture status of the pot soil in all the sets was maintained at 25% approx.

Monthly collection of the root samples from the different depths was done aseptically in sterilized containers. The samples of the same plant species from different depths were mixed to get a composite sample. The root pieces were cut aseptically into small fragments (1.0 cm). Equal number (50) of such pieces was introduced in sterilized distilled water and shaken on electric stirrer to get the suspension for the assessment of fungi. Dilution plate method was followed using Martin's medium. The above pieces were also inoculated on Czapek's + Dox Yeast medium and the fungi appearing on and in close proximity of the root pieces were recorded. The data presented in the paper have, however, been consolidated for the three stages of decomposition i.e. early (1–3 months), middle (4–9 months) and last (10–12 months).

Cellulose, hemicellulose and lignin contents of the root samples collected at three different decomposition stages were determined by the method described by Peach and Tracey (1955). Root samples of the plants were separately collected at different stages of decomposition for the collection of root extract. The roots were washed and crushed in 5 ml of water, filtered through paper and finally through Seitz filter. Root extract of the same plant collected throughout the year was mixed, concentrated under vacuum to 2 cc and the spore germination of 9 frequently occurring fungal species from the roots was studied. The extract was also analysed for total soluble sugar and total free amino acids (Peach & Tracey 1955) and phytotoxic substances (Smith 1960).

T. aestivum was selected for further detailed study of the root mycoflora and the decomposition of the root system under different soil amendments. Two sets were maintained in this case. Twenty plants raised in the pots were cut from the base after two months of growth and the roots were allowed to decompose without any disturbance. Roots from such plants were collected monthly for the assessment of fungal population. Roots were collected from 0–4 cm (Crown), 4–10 cm (middle) and 10–16 cm (distal) layers in separate sterilized containers. Simultaneously the roots were also collected from the plants with intact shoot system for comparison. The fungal flora in this set was isolated from root surface by dilution plate method and by plating root pieces as described above. In other set the root pieces after washing with sterilized distilled water were

teased aseptically into extra stelar and stelar regions and inoculated separately on Czapek's Dox + Yeast agar medium for fungi.

In other set of the experiment the roots of *T. aestivum* were collected from 4-month-old plants, washed thoroughly and dried. Equal amount of such roots (2 g) was packed in nylon bags (2 mm mesh size) and kept at a depth of 10 cm in the pot soil amended separately with sugar (Sucrose—S), nitrogen (Urea—N) and organic manure (OM). The amendment of the substances was done at the rate of 10g. kg^{-1} of soil. Twenty bags were kept in each case after ten days of amendment and control consisted of the soil with roots only. The pots were watered regularly to maintain the moisture status at approximately 25% and were placed in glass-house at 20 ± 2°C.

After one month of decomposition the root samples were collected from the nylon bags separately and the fungal population studied. The study was prolonged for a period of one year. In this case also the data have been compiled in three categories i.e. early, middle and last stages of decomposition as suggested earlier. In this case the cellulose, hemicellulose and lignin of the different samples were separately determined for three stages of decomposition by the methods suggested earlier.

After isolation of the fungi from different sets, 17 fungal species which were frequently isolated from the root of the plants were screened for pectinase, cellulase, saccharase and lignase activities (Peach & Tracey 1955).

Results

Fungal occurrence: The fungi associated with the decomposition of different roots differed remarkably (table 1). None of the species isolated was recorded from the roots of all the plants. *A. terreus* was most common and was cultured from four of the plant species. Phycomycetes were mostly obtained from *T. aestivum* roots. The spectrum of the fungal flora also varied with stages of decomposition. Except for Aspergilli which were obtained from early, middle, and last stages of decomposition other species were present either at one or two stages only.

Fungal population: Different plant species showed varying populations at the three stages of decomposition (table 2). In the early stage the maximum fungal population was noted in *P. typhoides*; in the middle stage in *P. scrobiculatum* and in the last stage in case of *T. aestivum*. The number of fungal species was generally more at the middle stage except in *P. typhoides* where the number was more in every stage of decomposition.

Cellulose, hemicellulose, and lignin contents (figure 1): The consolidated average figure for these show that in all the cases the contents were maximum in root of *P. typhoides*.

Fungal germination (table 3): Except for control set the germination percentage was

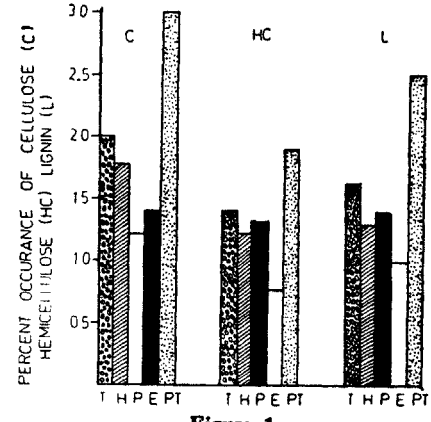

Figure 1

T=*T. aestivum* H=*H. vulgare*, P=*P. scrobiculatum*, E=*E. crusgalli*, PT *P. typhoiaes*

Table 1 *Record of fungi on the root of different plants (percent occurrence)*

Fungi	*T. aestivum*	*H. vulgare*	*P. scrobiculatum*	*E. crusgalli*	*P. typhoides*
Rhizopus nigricans	13.6	10.0	0	0	0
M. racemosus	13.0	0	0	0	0
Cunninghamella echinulata	10.4	0	0	0	0
Neocosmospora vasinfecta	0	8.0	0	0	0
Aspergillus niger	18.0	5.5	0	0	14.3
A. terreus	15.0	20.0	14.5	9.4	0
A. phoenicis	0	20.2	0	0	0
A. nidulans	0	0	11.6	8.4	0
A. versicolor	0	0	0	16.6	0
A. aculeatus	0	0	0	0	24.4
A. flavus	0	0	0	0	32.2
A. fumigatus	0	0	6.3	0	0
Penicillium chrysogenum	0	0	0	0	8.6
P. terrestre	0	0	5.3	0	0
Acrophialophora fusispora	0	4.2	4.4	0	0
Alternaria alternata	9.7	0	0	0	0
Curvularia lunata	8.2	7.4	11.5	0	0
Fusarium nivale	0	20.2	16.4	0	0
F. sp	0	0	0	16.4	0
White septate hyphae	4.4	0	0	40.2	0
Black septate hyphae	0	0	0	0	12.5
Other less significant fungi	7.7	6.5	30.0	9.0	8.5
No. of spp.	20	23	31	31	19

Table 2 *Fungal population (10^{-4} colonies. g^{-1} dry soil) and number of species (within bracket) at different stages of decomposition*

	T. aestivum	*H. vulgare*	*P. scrobiculatum*	*E. crusgolli*	*P. typhoides*
Early stage	7.2 (15)	6.9 (17)	2.4 (10)	2.3 (8)	23.0 (22)
Middle stage	22.0 (10)	12.9 (16)	24.2 (19)	23.2 (12)	11.0 (12)
Last stage	11.0 (6)	9.5 (9)	5.8 (12)	8.4 (9)	8.7 (10)

highest in the root extract of *P. typhoides* and the lowest in *T. aestivum*. Control set, however, always favoured maximum spore germination.

Soluble sugars/amino acids/phytotoxic substances (table 4): Maximum sugars and amino acids were recorded from *P. typhoides* roots and the amount was lowest in case of *P.*

Table 3 *Percent spore germination of fungi in the root extract*

	Control	*T. aestiyum*	*H. vulgare*	*P. scrobicnlatum*	*E. crusgalli*	*P. typhoides*
Rhizopus nigricans	89	29	42	32	43	76
Cunninghamella echinulata	92	10	39	16	24	82
Asp. terreus	84	36	83	79	76	78
A. niger	84	69	73	26	30	79
Penicillium terrestre	83	14	43	10	18	60
Acrophialophora fusispora	93	16	63	19	10	53
Curvularia lunata	86	43	76	83	80	73
Alternaria alternata	86	13	45	32	24	80
Fusarium nivale	96	12	85	32	18	39

Table 4 *Total soluble sugars (μg/g fresh root), total free amino acid (mμg/root) and phytotoxic substances in the root extract*

	T. aestivum	*H. vulgare*	*P. scrobiculatum*	*E. crusgalli*	*P. typhoides*
Total sugars	300	180	166	200	216
Total amino acids	330	220	200	200	280
Phytotoxins	0	+	+	0	+

+=Present, 0=absent

Table 5 *Fungi in the different regions of root of standing plants of* T. aestivum

	Crown region	Middle region	Distal region
Rhizopus nigricans	++	++	+
Mucor racemosus	+	++	++
Cnmninghamella echinulata	+	+	+
Aspergillus niger	+	++	0
A. terreus	+	++	0
Verticillium sp.	+	+	++
Fusarium sp.	+	+	++
Black septate hyphae	++	+	+
White septate hyphae	+	+	++
Other less frequent spp.	+	+	+
Total spp.	32	19	13
Fungi/g root	32008	22702	14900

++=Dominant, +=Subdominant, 0=absent

scrobiculatum. Phytotoxic substances were recorded from *T. aestivum*, *P. scrobiculatum* and *H. vulgare* sets only.

Most of the dominant and subdominant fungi of the crown, middle and distal regions of *T. aestivum* were of common occurrence. The population of fungi in the crown region was highest and the lowest number was from distal region (Table 5). The number of fungi cultured from crown, middle and distal regions of harvested crops was low as compared to standing plants (table 6). More fungi were isolated from extrastelar region in the early stage of decomposition. The number of species also decreased from crown to distal regions. The number of species varied in the extrastelar region at different root layers and also at the three stages of decomposition. The difference was, however, insignificant in the stelar region. A glance at table 6 reveals that phycomycetous forms were more common in the extrastelar region of early and middle stages of decomposition. Amongst the forms isolated only *A. terreus* and *Verticillium* sp. were cultured at all the three stages while others were of restricted occurrence. Septate sterile hyphae with clamp connection were restricted to the stelar region at the later stages only (table 6).

In figure 2 is presented the fungal population obtained from the root and the soil of the amended sets with sugar, nitrogen and organic manure. Simultaneously the data are also given for the control set without any amendment. The trend of variation in

Table 6 *Distribution of fungal species in different root regions of* T. aestivum *at the three decomposition stages of harvested set*

		Early stages		*Middle stage*		*Last stage*	
		Extrastelar (ES)	Stelar (S)	ES	S	ES	S
Rhizopus nigricans		Cr M	0	Cr M	0	0	0
Mucor racemosus		Cr M	0	Cr	0	0	0
Cunninghamella echinulata		Cr	0	0	0	0	0
Aspergillus terreus		Cr	Cr	Cr M	Cr	Cr MD	D
Memmoniella echinata		0	0	Cr M	0	Cr MD	MD
Alternaria alternata		Cr	0	Cr M	0	Cr MD	0
Fusarium roseum		Cr MD	MD	Cr MD	MD	0	0
Verticillium sp.		Cr MD	MD	Cr MD	MD	MD	D
White St. hyphae		0	0	0	Cr M	0	D
Black St. hyphae		0	0	0	0	0	D
Septate hyphae (with clamp connections)		0	0	0	Cr M	0	Cr MD
*Other less frequent Species.							
Total	Cr	18	5	10	6	11	4
Spp.	M	12	4	6	4	7	3
	D	7	4	6	4	4	4

Cr=Crown, M=Middle, D=Distal region, 0=absent

*Fungal species grouped in the category were irregularly distributed in the different region

the fungal population of soil and root was more or less the same. Generally the maximum population was recorded from nitrogen set followed by sugar, organic manure and least from control set. In the case of root the population was maximum between 18 December and 24 March. In soil. on the other hand, the condition was slightly different. In N and S sets the maximum was on 8 October and 8 February respectively. In OM set the maximum population was in the beginning itself and the population in all sets decreased with advance in decomposition stages. Qualitatively the fungal species were always higher in the soil. The number of species in the early and middle stages of decomposition in the case of root samples did not vary much but the difference was notable in the last stage where the number decreased drastically. The condition in the soil, however, was different where the number of species at the different stages did not differ much. (Table 7).

A glance at table 7 clearly indicates that a number of dominant species in all the sets were common. Aspergilli were frequently isolated with high frequency. *Rhizopus* was more frequent in the early and rare in the later stages. *Fusarium* sp., *A. niger*, *A. flavus* and *A. terreus*, were frequently cultured from the different sets of all the three stages. Certain forms like *Cladosporium*, *Cylindrocarpon* and *Myrothecium* were of restricted occurrence.

Figure 3 shows the cellulose, hemicellulose and lignin contents of the root of the amended and control sets at different stages of decomposition. The depletion in the quantity of the three substances was faster in N set followed by S, OM and control. Loss in weight of *T. aestivum* root at different stages of decomposition is plotted in figure 4. Out of the 17 species tested for the enzymatic activity, 13 responded to cellulase activity. Generally the species were

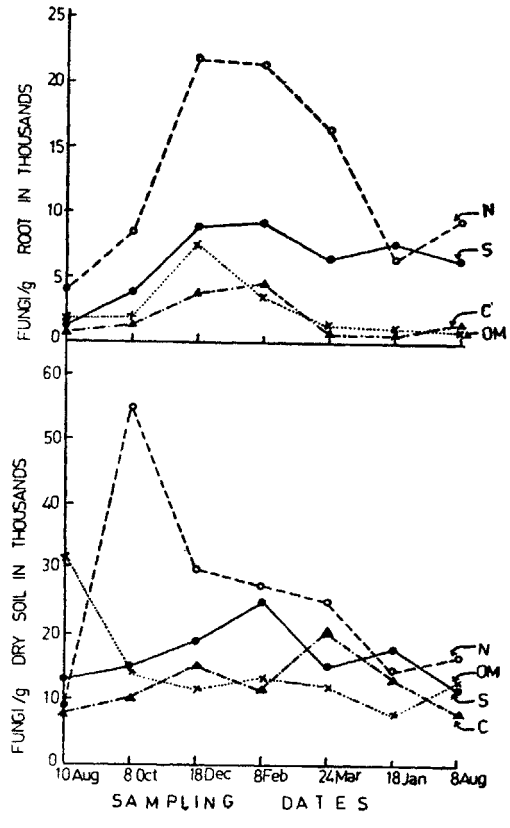

Figure 2
N=Nitrogen, S=Sugar, OM=Org manure, C=Control

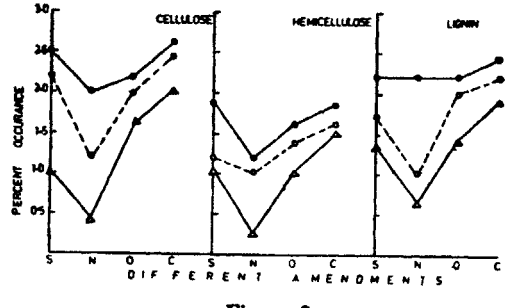

Figure 3
S=Sugar, N=Nitrogen, O=Org. matter, C=Control, Early stage=●——●, Middle stage=O——O, Last Stage=Δ—Δ

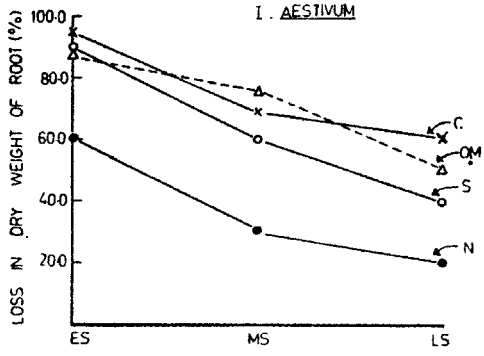

Figure 4
S=Early stage, MS=Middle stage, LS=Last stage, N=Nitrogen, S=Sugar. OM=Org. manure, C=Control

specific in their activities except *A. terreus* and sterile hyphae which exhibited wider activity. Phycomycetes responded well for saccharase activity. Two sterile forms (one with black coloured septate hyphae and other with clamp connection) were the only species which exhibited lignase activity (table 8).

Discussion

Spectrum of the fungal species associated with the root of the different species under identical ecological and edaphic conditions varied both in quality and quantity. Though Aspergilli contributed largely to the forms

Table 7 *Fungal species associated with different decomposition stages of root of* T. aestivum *and corresponding soil*

Amendments		Early stage (1—3 months)	Middle stage (4—9 months)	Last stage (10—12 months)
Sugar set	Root	*A. flavus*, *Fusarium* sp., *Curvularia* sp., *A. nidulans*, *A. terreus*, (Fungal spp. 20)	*A. flavus*, *A. terreus*, *Cladosporium* sp., *F.* sp. *A. sydowi* (F. spp. 36)	*A. terreus*, *Curvularia* spp. (F. spp. 10)
	Soil	*A. tamari*, *A. nidulans* (F. spp. 39)	*A. niger*, *A. nidulans*, *A. flavus*, *A. sydowi* (F. spp. 36)	*A. terreus* (F. spp. 32)
Nitrogen Set.	Root	*R. nigricans*, *A. niger* (F. spp. 14)	*R. nigricans* (F. spp. 13)	*Trichoderma* sp., *Cylindrocarpon*, (F. spp. 6)
	Soil	*A. niger*, *P. fusisporus*, *A. flavus*, (F. spp. 16)	*A. flavus*, *A. niger* *A. nidulans*, *A. terreus*, F. sp. (F. spp. 18)	*A. niger*, *Fusarium* sp. (F. spp. 20)
Org. Manure Set.	Root	*A. terreus*, St. Col., *A. flavus*, *Fusarium* sp. (F. spp. 24)	*A. terreus*, *Fusarium* sp., *A. niger*, *C.* sp. *Myrothecium* (F. spp. 26)	*Trichoderma* sp., *C. lunata*, (F. spp. 12)
	Soil	*Rhizopus*, *Fusarium* sp.	*A. niger*, *Myrothecium*, *Fusarium* sp. (F. 36)	*Fusarium* sp., *A. niger*/ (F. spp. 30)
Control	Root	*Pen.* sp., *Fusarium* sp. (F. spp. 12)	*Fusarium* sp., *A. terreus*, *C. herbarium* (F. spp. 16)	*A. niger*, *Fusarium*, *Trichoderma* sp., (F. spp. 12)
	Soil	*Rhizopus*, *A. niger*	*A. nidulans*, *Fusarium* *P. humicola*, (F. spp. 22)	*C. tetramera*, *P. humicola* (F. 24)

Figures in brackets indicate the number of fungal species

Table 8 *Enzymatic activity of certain fungi*

	Pectinase	Cellulase	Saccharase	Lighnase
Rhizopus nigricans	0	0	+	0
Mucor racemosus	0	0	+	0
Cunninghamella echinulata	0	+	+	0
Aspergillus niger	0	+	+	0
A. terreus	+	+	+	0
Trichoderma sp.	0	+	+	0
Gliocladium roseum	0	+	0	0
Acrophialophora fusispora	+	+	0	0
Penicillium sp.	0	+	0	0
Verticillium sp.	0	+	0	0
Memnoniella sp.	+	+	0	0
Alternaria alternata	0	+	0	0
Cladosporum herbarium	+	+	0	0
Fusarium sp.	+	+	+	0
Wt. Septate Col.	0	+	4	0
Bl. septate Col.	0	0	0	+
Sept. hyphae with clamp connection (Basidiomycete ?)	0	0	0	+

+=Present; 0=not detected.

associated with the roots, most of the species of the genus were of restricted association. Specificity of the forms may be explained in the light of the biochemical nature of the roots under investigation*. As evident from the data tabulated in table 4, the maximum nutrients available for the microbes were in the roots of *P. typhoides* where the fungal population was also generally recorded as maximum. This direct correlation, however, is not unquestionable because the root analysis sometimes may not prove very rewarding. Some of the substances present in the extract may not be available directly to the microorganisms associated with the root system. This may, however, give an indication regarding the nutrient budget of the root system and in absence of any direct approach to the problem, it may be of some help in understanding the problem. To substantiate this proposition, it was considered advisable to test the germination of the more frequent fungal species associated with the roots of the different plants in their root extract. In table 3 is summarised the data collected from the study. The root extract of *P. typhoides* favoured the spore germination of almost all the test fungi. The values obtained in this case for a number of species approached very near to the control. From a comparison of the data in the tables 1 and 3 it may be inferred that generally the forms which are more frequent in the root region of a particular species were also favoured in the root extract. Other species not favoured in the extract were either absent or isolated with less frequency in the root environment. This fact, therefore, minimises the doubt

*Rovira 1959, Vancura 1964

raised above regarding the suitability of the root extract analysis to explain the fungal behaviour.

With advancing decomposition depletion of cellulose,' hemicellulose and lignin was faster in case of *E. crusgalli*, *P. scrobiculatum* and *H. vulgare*. Initially there was not much difference in the amount of these substances in the roots of *T. aestivum*, *H. vulgare*, *P. scrobiculatum* and *E. crusgalli* but after a year of decomposition the amount of the substances differed considerably. This difference may possibly be due to the spatial distribution of the substances and the fungal species involved in the decomposition. The high fungal population in *P. typhoides* root is not directly proportional to the rate of decomposition in this case. I have no suitable explanation for this except to suggest that the root tissue comlexity may be one of the factors.

When the investigation was intensified with the roots of *T. aestivum* it was observed that in the standing plants the species spectrum of crown, middle and distal regions of the root varied. Dominant species were mostly common in the three regions but many less frequent forms were of course different. Drastic reduction in the number of the species was marked from crown to distal region. Marchant (1970) noted that the root of certain species were mostly devoid of fungi and bacteria. Srivastava (1969) and Kanaujia (personal communication) have also noted similar spatial distribution pattern of the mycoflora on the root surface. In the crown region the root system is a mixed one consisting of old and new roots. Older roots provide sloughed off tissue for decomposition and the younger roots enrich the nutritional status of the environ ment through root leakage. The sum total of the nutritional level of the region is comparatively much higher than the distal region where the root exudation is the major source of nutrition. The condition is intermediate in the middle region. This nutritional pattern affects the spatial distribution of the mycoflora. In crown region the forms associated with decomposition of the older roots coupled with those found on living root surface enrich the root region microflora. In distal region certain limited species specific to the region are encouraged to develop and the flora, therefore, is not very rich. Difference was also noticed when the extrastelar and stelar regions were examined for the mycoflora at the different stages of decomposition of the harvested plants. Cells of the extrastelar region being rich in the food harboured more fungal species than the stelar cells. Further, the sclerenchymatous cells of the stele being rich in phenolic substances probably did not favour the entry, subsequent establishment and growth of rich mycoflora. In the last stage of decomposition when the root tissue got exhausted of the food material the number of species decreased drastically. Sterile forms with and without clamp connection were the dominant species at this stage. The species rich in lignase activity were responsible for the decomposition of the left out plant tissue at this stage. Thornton (1965) also recorded darker hyphal forms being associated with older living roots of perennial rye grass and *Trifolium repens*. Brown septate hyphae were also recorded by Gadgil (1965) and Nicolson (1959) on the roots of grasses at the time of their sloughing off.

Decomposition of the root was affected differently in the standing and harvested plants. The latter set favoured the growth of lesser number of species. This may be explained in the light of food material reserved in the root system. Though not investigated in the present study, the food material may be expected to be more in the root of standing plants where photosynthe-

tic activity and storage processes are not hampered.

As summarised below the fungal species of group 5 were isolated almost from all the regions of the root and on the other extreme, forms of group 1 were restricted to stelar region. In between these two extremes were species belonging to groups 2, 3 and 4 which were of intermediate distribution.

Root surface	*Extrastelar region*	*Stelar region*
		1. St. White and black hyphae, septate hyphae with clamp connection (Basidiomy cetes)
	2. *Fusarium* *Verticillium*	
	3. *Rhizopus* *Mucor* *Cunninghamella* *Memnoniella* *Alternaria*	
4. Penicillia, Aspergilli, *Trichoderma*, *Cladosporium*, *Gliocladium*		
5. *A. terreus*, *Acrophialophora*		

Studies of Kreutzer (1972) have shown that the *Fusarium roseum* was less frequent in the inner region of certain grass roots as also reported by Waid (1957). A persual of the table 7 however, indicates that *F. roseum* was isolated from stelar region in the early and middle stages but was absent in the last stage of decomposition.

In the amended sets rate of decomposition was faster in the nitrogen set where the fungal population was also maximum (fig. 2). The number of fungal species, however, was less as compared to S and OM amendments. High population density of the forms possessing cellulase activity resulted in fast decomposition of the substrate. In S and OM sets also though most of the forms of N set were present but the decomposition rate was slow probably because of N deficiency.

Surprisingly enough throughout the course of investigation the members of ascomycetes and basidiomycetes could not be observed in any appreciable number. Forms like *Chaetomium* sp. and *A. nidulans* were isolated but their frequency of occurrence was very low. Only sterile hyphae with clamp connection are suspected to be basidiomycetous but in this case also it is with reservation only unless the fruiting bodies are made to develop. The role of the fungi of these two groups in decomposition of the roots should not be underestimated because

of their low occurrence, which may be due to the shortcomings of the techniques used and the media selected.

An overall picture of the study reveals that the distribution pattern of the fungal species on and within the decomposing roots, besides a number of other factors, is regulated by their enzymatic activities. Forms with saccharase activity were mostly the early colonizers and were followed by those with pectinase and cellulase activities. The last to colonize were those exhibiting lignase activity (table 8). The role of the species with these activities in the colonization and ultimately with decomposition of the plant material is very important. Pugh et al. (1972) have stressed the importance of such species on the colonization of the leaf surface and some of the species are common in this investigation also.

Though the effort has been made in this study to investigate the different parameters related to root decomposition under identical ecological conditions, a number of questions, however, still remain to be answered which will be looked into in future studies.

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