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Impact of Pesticides on Decomposition of Wheat Litter

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Abstract

Quantitative variation and changes in the composition of wheat litter fungi, exposed to natural condition were studies. Altogether 20 species of Deuteromycetes, 8 species of Ascomycetes and one member of Phycomycetes were isolated. Addition of benomyl and dithane-M-45 to plant litter exhibited decrease in the population dynamics of saprophytic fungi and also their composition.

The normal litter decomposition in terms of loss in dry weight was noticeably inhibited by the addition of pesticides as stated above. The data analysis on impact of pesticides on physiology of phylloplane fungi revealed that under the influence of pesticides could have been due to the qualitative and quantitative changes in the litter saprophytic microflora rather than due to any changes in their state of physiology.

Keywords: Phylloplane Microbes, Litter, Fungi, Pesticides.

Introduction

The non-target phylloplane microbes either give additional supports to certain metabolic process of host plants (Ruinen, 1965; Buckley and Pugh, 1971; Fokkema etal, 1979) or remain non effective during Ontogeny of leaves. When the leaves become senescent, the litter is decomposed by these microbes or other saprophytic organisms (Hering, 1967; Dickinson and Pugh, 1974; Harper and Lynch, 1985) that help increasing the soil fertility (Chang, 1967; Hudson, 1968; Garrett, 1981; Shri Niwas, 1988). The process of decomposition of litter mainly depends either on composition of saprophytes or the type of vegetation. There is evidence that quantitative and qualitative variation in bacteria (Cambell, 1985). However, in this regard the litter decomposition in agriculture field is very poorly understood.

So, the present set of experiments was designed to know precisely, the quantitative and qualitative nature of litter microflora and its contribution to decomposition process, if any. In addition, attempt was made to know the wide application of various pesticides on litter docomposition.

Aim of Study

The Aim of present research work is to improve the soil fertility by degradation of agro waste biomass. Use of pesticides in agriculture in harmful for the beneficial microorganisms like litter fungi and Bacteria. In Present work wheat litter used as agro waste for decomposition process in natural and controlled condition under pesticides treatment.

Materials and Methods

Cultivation of wheat plants and Litter Samplings

The cultivation of wheat plants and designing of experimental plots carried out carrier. The first sampling of litter was done in the first week of April, 1986 and subsequent samplings were made with an interval of one month till the second week of September, 1986. The leaf litter samples were kept inside small bags (10 x 5cm) made of mesh nylon cloth (0.1 mm^2). Known amount of litter (10 gm) on dry weight basis was kept in each bag and the bags were exposed to natural field conditions for decomposition. With an interval of one month litter sampling was done to study the loss in dry weight of litter and its mycoflora.

Determination of Dry weight

The loss in litter weight was estimated on dry weight basis. The litter samples were dried at 60^0C for 48 hrs to constant weight.

Composition of wheat Biomass

The composition of wheat biomass was determined by estimating the cellulose, hemicellulose and lignin by chemical analysis (Harper and Lynch, 1981).

Isolation of microfungi from Decomposing litter Biomass

Microfungi were isolated from wheat litter biomass by dilution plate technique.

Litter Decomposition under Controlled Conditions

Fresh litter biomass samples from control and pesticides treated plots were collected and oven dried as stated earlier. The dry litter biomass was ground to powder by means of electric grinder. One gram of powdered biomass sample was taken in sterile watch glass and was kept inside Petri dishes containing 25ml of sterile water. The purpose of taking water inside Petri dishes was to maintain humidity inside Petri dishes. Four sets of experiments were maintained. The first set was made free from microorganisms and in the second set various types of wheat litter decomposing fungi were inoculated, individually. The biomass sampling for third set and fourth set were from benomyl and dithane-M-45 treated experimental plots. The inoculation of fungi in the third and fourth set was carried out as said in case of the second set. Five species of fungi viz. Aspergilus flavus, A.niger, Penicillium chrysogenum, Rhizopus nigricans and Trichorma viride were taken for litter biomass decomposition studies as said above. In order to know the synergetic impact of fungi on biomass decomposition an additional sample was maintained by inoculating mixture of the above said fungi. The inoculation of fungi on biomass was done under asceptic conditions. Soon after the inoculation, the Petri dishes were covered with sterile aluminum foils and inserted inside small polythene bags. Including control, all Petri dishes were incubated under constant temperature (25^0C) for a week. After seven days of incubation, samples were examined for loss in biomass and cellulass enzyme activities.

Assay of Cellulase

The enzyme was extracted by mixing pre-chilled 0.05M citrate buffer (pH 5.0) with the biomass of fermented broth. Twenty ml of buffer was added to the fermented biomass and slowly stirred for 30 min. The resulting anzyme extract was then centrifuged at 10,000 g for 5 min at 0^0C. The supernatant was transferred to a pre-chilled centrifuge tube and again centrifuged at 15,000 g for 15 min at 0^0C. Cellulase enzyme in the supernatant was assayed by filter paper decomposition method as followed byy Bisaria and Ghose (1981). Specific activity of anzye was expressed as cellulase activity per 100 u gram of crude enzyme protein.

Estimation of Protein

The TCA insoluble protein of anzyme was measured by the procedure as described by Lowry et al (1951). BSA (Sigma fraction V) was used as standard.

Results

Quantitative and Qualitative Analysis of Litter Mycoflora

Qualitative variation and changes in the composition of mycoflora of wheat litter, exposed to natural conditions, has been given in the Table-1. Altogether 19 species from Deuteromycetes, 8 species from Ascomycetes and an individual member from Phycomycetes were isolated and identified (Table-1) from wheat litter during its decomposition process. From the literature survey, it has been noted that out or these 28 species of microfungi, five members i.e. Aspergillus flavus, A.niger, Penicillium chrysogenum, Rhizopus nigricans, and Trichoderma viride even with species variation were of most industrial importance Grueger and Grueger, 1984) due to their cellulose decomposing potential as compared with the other remaining fungi, available during litter decomposition.

In general, the effect of both the pesticides was noticed to be very much prominent. However, the broad spectrum pesticide i.e. dithane-M-45 was comparatively more effective than the systemic fungicide, benomyl. During last phase of decomposition, only three species from Ascomycetes and two species from Deuteromycetes were noticed in the benomyl and dithane-M-45 treated samples, respectively.

The population of Alternaria humicola, A.longipes, A.tenuissima, Chaetomium globossum, Cladosporium herbarum, Curvularia lunata, Drechslera hawaiiensis, D.spicifer, D.sp., Fusarium moniliforme and Helminthosporium sp. observed to be maximum during the third/fourth month of decomposition and disappeared completely in the fifth month of decomposition. Except Alternaria tenuissima, all the remaining 10 fungi, as stated above, were completely eliminated by both the pesticides. Aspergillus candidus, Drechslera australiensis and D.halodes were observed infrequently for a very short period and disappeared much before the last phase of decomposition of litter.

The treatment of benomyl and dithane-M-45 brought gradual reduction in the intensity of population of these two fungi. Only three members i.e. Aspergillus candidus, Penicillium chrysogenum and Trichoderma virede showed increase in the intensity of population with the increase in the decomposition time period. However, except Trichoderma viride, the remaining two fungi, as showed above, completely disappeared by both the pesticides during last phase of decomposition. Two members i.e. Acremonium sp. and Phoma glomerata from deuteromycetes appeared only during the first month of decomposition and did not appear, till the last phase of decomposition. These two members were very much susceptible to both the pesticides.

Changes in Litter Biomass

The changes in the biomass during litter decomposition under natural and control conditions are given in the Table-5. The changes in biomass during decomposition process under natural condition were noticed in an increasing order with respect to time. During the first 3 months, the loss in biomass of water spread sample was about 40-48%. But during late phase of decomposition the changes in the loss of biomass was higher as compared with the earlier phase of litter decomposition. During fifth moth of decomposition around 90% loss in biomass was noticed. It has been observed that the application of pesticides reduced the process of decomposition. The effect of benomyl and dithane-M-45 on phase of

decomposition around 60% loss in biomass was noticed in both the pesticide treated samples.

Table-3 shows the loss in weight litter biomass caused by individual fungi. Penicillium chrysogenum and Trichoderma viride were found to be the most potential fungi for wheat biomass decomposition. Around 50% loss in wheat biomass was caused by these two fungi. The litter decomposition potential of Aspergillus niger was observed to be minimum as compared to the members listed in Table-3. Around 40% loss in the biomass was done by this fungus. The synergetic effect of the five fungi (Table-3) on reduction in biomass was more than that by individual fungus Table-4 shows the cellulase activities of various Litter decomposing fungi both under control and natural conditions. Under control conditions Rhizopus nigricans, shows maximum filter paper decomposition activity followed by Trichoderma virude, Aspergillus flavus and A.nigeer respectively.

The enzyme extract from biomass exposed to natural condition showed about two-fold enzyme activities than that extracted from Rhizopus nigricans grown biomass sample.

Discussion

There is information on decomposition of litter biomass (Brinson, 1977; Anderson and Danell, 1982; Campbell, 1985) in terrestrial eco-system (Dickinson and Pugh, 1974; Mason, 1977; Swift et al, 1979). The process has also been well correlated with various atmospheric conditions i.e. temperature, humidity and rainfall those generally occur during seasonal changes (Harmon et al, 1986). However, analysis of present data showed that not only the physical environmental factors but also the bio-chemical changes in the litter biomass were responsible for the qualitative and quantitative variation of mycoflora during litter biomass decomposition.

The rapid rate of biomass decomposition under controlled conditions either by individual fungi or a group of fungi as compared to that under natural condition, could be possible due to the impact of hydrocabons decomposing fungi under maximum favourable condition like optimum pH, critical temperature and maximum humidity (Harmon el al, 1986)

The reduction in the process of decomposition of wheat biomass under natural conditions by pesticides treatment might be due to the qualitative and quantitative changes in the litter, microflora as observed by earlier workers (Alexender, 1969; Anderson, 1978; Cambell, 1985) However, the impact of systemic fungicide and broad spectrum pesticide was noticed to a same.

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Table – 1
Composition of litter microfungi (colonies cm^{-2}) on decomposing litter Biomass of wheat under natural conditions.

Name of fungi	Period (days) of litter decomposition treated with water (W), benomyl (B) and dithane-M-45 (D)														
	30			60			90			120			150		
	W	B	D	W	B	D	W	B	D	W	B	D	W	B	D
PHYCOMYCETES															
Rhizopus nigricans	27	-	-	26	-	-	83	-	-	-	-	-	-	-	-
ASCOMYCETES															
Acnaetomium globossum	28	-	-	55	-	-	56	-	-	-	-	-	-	-	-
Aspergillus candidus	-	-	-	6	-	-	6	-	-	-	-	-	-	-	-
A.flavus	27	20	-	28	-	-	90	55	30	138	-	-	166	-	-
A. lucnuenais	60	56	55	83	56	3	83	56	3	167	83	27	128	83	22
A. nidulans	28	28	-	84	-	-	-	-	-	-	-	-	-	-	-
A. niger	28	25	15	28	27	3	-	-	-	-	-	-	-	-	-
Chaetomium globossum	-	-	-	27	-	-	84	-	-	111	-	-	-	-	-
Penicillium chrysogenum	-	-	-	56	28	-	180	114	110	189	30	27	194	-	-
DEUTERONYCETES															
Acremonium	27	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Alternaria alternate	83	3	-	138	5	3	166	83	28	194	83	27	167	28	-
A.humicola	8	-	-	16	-	-	111	-	-	-	-	-	-	-	-
A.longipes	3	-	-	13	-	-	83	-	-	96	-	-	-	-	-
A. tenuissima	56	56	-	194	28	22	250	112	28	250	28	-	-	-	-
A.ap	6	-	-	56	-	-	3	-	-	-	-	-	-	-	-
Cladosporium cladosporioides	4	-	-	27	-	-	28	-	-	6	-	-	-	-	-
C.herbarum	11	-	-	56	-	-	-	-	-	-	-	-	-	-	-
Curvularia lunata	28	-	-	28	27	-	83	-	-	111	-	-	-	-	-
C. pallescens	-	-	-	-	-	-	-	-	-	-	-	-	56	-	-
Drechslera autraliensis	-	-	-	83	27	-	-	-	-	-	-	-	-	-	-
AUSTRALIENSIS															
D. spicifer	56	27	-	83	-	-	-	-	-	-	-	-	-	-	-
D.halodes	-	-	-	3	-	-	-	-	-	-	-	-	-	-	-
D.hawaiiensis	5	-	-	6	-	-	-	-	-	-	-	-	-	-	-
D.sp	3	-	-	11	-	-	28	-	-	-	-	-	-	-	-
Fusarium moniliforme	56	-	-	222	-	230		-	-	272	-	-	-	-	-
Helminthosporium sp	-	-	-	166	116	-	-	-	-	-	-	-	-	-	-
Phoma glomerata	83	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Trichoderma viride	83	-	-	86	-	-	111	10	-	122	-	-	156	28	6

Table - 2
Composition of Wheat Biomass

Content	Stem (%)	Leaf (%)
Cellulose	40+3.2	31+2.5
Hemicellulose	29+1.5	37+3.0
Lignin	14+1.2	13+1.28
Extractives	3	8

Table – 3
Loss In Weight of Litter Biomass During Growth of Various Microfungi. Each Figure Reoresebts An Average of Five Experiments

Name of fungi	Loss of biomass in mg/gm dry biomass		
	Control	Benomyl	Dithane-M-45
Aspergillus flavus	395+15	390+13	388+12.5
A.niger	360+12	372+10	362+15
Penicillium chrysogenum	530+15	525+9	535+11.5
Rhizopus nigricans	495+5	490+7	493+8
Trichoderma viride	510+22	500+12	485+17.5
Mixture of fungi	515+19	495+22	480+17.5
Control	0	0	0

Table – 4
Cellulase Enzyme Activity of Various Fungi Grown on Wheat Litter Biomass. Each Figure Represents An Average of Five Experiments

Name of fungi	Filter paper decomposition in mg/100 μg of enzyme protein		
	Control	Benomyl	Dithane-M-45
Aspergillus flavus	2.0+0.1	2.35+0.15	2.15+0.1
A.niger	1.6+0.05	1.75+0.13	1.57+0.1
Penicillium chrysogenum	1.6+.05	1.45+0.07	1.55+0.05
Rhizopus nigricans	3.0+0.2	2.78+0.5	2.9+0.17
Trichoderma viride	2.5+0.15	2.5+0.10	2.25+0.5
Mixture of fungi	2.3+0.17	2.15+0.15	2.0+0.15
Control	0	0	0
Natural condition Mixture of fungi available on the biomass	6.3+0.75	5.95+1.5	3.65+2.1

Table – 5
*** Channges in biochemical composition of litter during decomposition**.

Percentage of biochemicals					
Litter Decomposition Period	Cellulose	Hemicellulose	Lignin	Total Reducing Sugar	Water Soluble Protein
0 month	100	100	100	100	100
1 month	95	100	100	175	120
2 months	85	90	90	250	175
3 months	55	75	80	350	200
4 months	43	55	70	150	155
5 months	25	45	55	125	125

*The initial values i.e. 100μ for cellulose 475 mg/gm any weight, hemicellulose 375 mg/gm dry weight, lignin 250 mg/gm dry weight, reducting sugar 0.147 mg/gm dry weight and water soluble protein 0.105 mg/gm dry weight of decomposing litter biomass.