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Experiment on Effect of Interactions Between Soil Mycoflora and Fungivorous Microarthropods on Leaf Litter Decomposition and Fungal Reproductive Potential

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ABSTRACT

In this experiment 50 microcosms replicates were set up in a plastic chamber (8 x 8 cm high and diameter) with a 2 cm^2 hole in the lid covered with nylon mesh. Effect of fungivory on leaf litter decomposition was assessed by microscoms experiment. In this experiment a comparison of 4 treatments (T_2-Fungi only, T_3-Fungi+*Oppia yodai*,T_4-Fungi+*Onychiurus* sp.,T_5-Fungi+*Oppia yoda i*+ *Onychiurus* sp) with the Control(Sterilized and defaunted-T_1) were made. It has been revealed from the present experiment that the presence of both Acarina(*Oppia yodai*) and Collembola (*Onychiurus* sp) in treatment -5 resulted in significant loss of leaf litter weight .The means comparison test showed that the difference between control to T_2 and control to T3 were not significant. Treatment -4 showed slightly higher loss of weight than that of T_2 and T_3 but significantly lower than treatment-5.The effect of fungivory on fungal community structure was evaluated on the basis of reproductive potential of different species (*Alternaria alternata, Cladosporium cladosporioides, Fusarium chlamydosporium* and *Aspergillus flavus*) by the total number of recovered spore as well as total number of different spore recovered with ±95% confidence interval only. At the end of the experiment altogether 4 types of spore were identified from 90% of microcosms based on shape and appearance as well as hyphal fragments. Number of spores of *Alternaria alternata* showed significant increment over T_2 (12±1) in T_5 (23±2.5) and T_3 (18±1.5) but not in T_4(9±1).The number of recovered spore of *Cladosporium cladosporoioides* was higher(18±2) than that of *Alternaria alternata* in T_4.The spores of *Fusarium* and *Aspergillus* recovered from all the treatments were numerically lower than that of *Alternaria* and *Cladosporium*.

Key words *Experiment, Effect of Interactions, Soil, Mycoflora and Fungivorous Microarthropods, Leaf Litter Decomposition, Fungal Reproductive Potential*

Decomposition of plant organic matter is a process directly linked to soil fertility. Therefore, a main challenge for soil biologists is to understand the dynamics of the decomposition process and its relations to the origin and maintenance of soil fertility in natural and cultivated systems. In order to do this, it is necessary to understand the biology of decomposer organisms and their interactions with physical and biotic environments. The influence of soil fauna on soil biological processes is well documented (reviews by Seastedt, 1984, Ingham et al., 1985; Lussenhop, 1992). Microarthropods are among the most abundant decomposers in soil.Collembola, Acarines and enchytraeids are the major taxa belonging to this group. These animals live in the pore system of the soil and most of them preferentially feed on fungi; ingest decomposed plant material and mineral particles. These fungal grazing microarthropods affects microbial respiration (Bengtsson and Rundgren,1983; Kaneko et al.1998), decomposition rates(Cortet et al.2003),nutrient cycling, plant growth (Klironnomes and Kendrick,1995), fungal biomass ,fungal succession,the distributation of fungi in soils (Lussenhop, 1992) and the interaction between competing fungal species (Tiunov and Scheu ,2005). Interactions between micro arthropods and fungi are central to many processes in soil, such as decomposition and nutrient cycling. Fungi are an important group of decomposer organisms. As a group fungus degrade all kinds of organic matter and man-made products; they absorb by digesting with their filamentous hyphae, complex polymers such as carbohydrates, lipids, proteins and nucleic acids. They release their digestive enzymes into the surrounding and absorb the available nutrients (Swift et al.1979).Saprophytic fungi are able to degrade cellulose and lignin (Domsch and Gams, 1969; Dix and Webster, 1995) which enables them to decompose litter and wood. Fungi are the main decomposer group of the products of primary production, leaf litter, twigs, roots etc. Therefore, fungi are central to understanding the process of decomposition in terrestrial ecosystem (Rodin and Basilevic, 1967, 1968). In accordance, fungivory stands up as a key interaction to be study if we aim to understand the dynamics of the decomposition process (Bandyopadhyay et al.2009) and its implications to soil fertility. In relation to decomposition other studies have shown that fungivory increased the respiration rate of the micro-biota in decomposing leaf litter (Hedlund and Oehrn,2000).Along

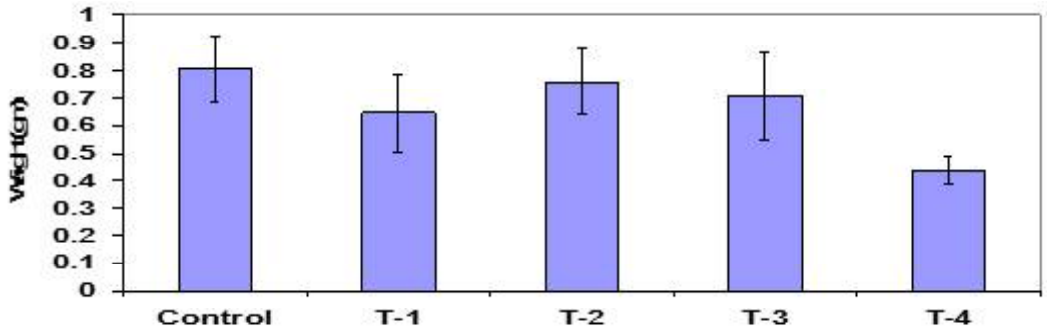

Fig 1. Leaf litter decomposition in microcosms experiments after 60 days of Exposure to different treatments

Table 1. One way Analysis of Variance (ANOVA) of Leaf litter decomposition in Microcosms experiments after 60 days of exposure to different treatments

Source of Variation	ss	df	MS	F	P-value	F crit*	F crit**
Between Treatments	0.404835	4	0.101209	29.09769	4.3808	2.866081	4.43069
Within Treatments	0.069565	20	0.003478				
Total	0.4744	24					

** F-Critical at 1% level * F-Critical at 5% level

with decomposition, fungi have several important functions in the ecosystems, such as symbiosis and parasitism. One-fourth of all described fungal species form lichens and almost all plant species are associated with mycorrhizal fungi (Smith and Read,1997). Interactions between micro arthropods and fungi are central to many processes in soil. Surprisingly, the possible mechanisms of these interactions, such as grazing, disturbance and dispersal, have been little studied. Grazing of soil animals on fungi may affect the competition between saprophytic and mycorrhizal fungi (Tiunov and Scheu, 2005), the recovery and succession of saprophytic fungi (Maraun et al.1998 b) and the dispersal of fungal propagules (Anderson, 1988, Renker et al.2005). Soil fungi provide the principal nutrient for the soil micro arthropods in the soil food web and this prey-predator relationship is significant in the decomposition process. The aim of the present work was to determine the effects of interactions between soil micro-arthropods (Collembola And Acarina) and mycoflora on soil fertility and impact of this fungus-fauna relationship on decomposition process.

MATERIALS AND METHODS

The present study has been carried out by Innovation Hub of Burdwan science Centre,NCSM,Govt.of India,Burdwan and Burdwan Raj College,Burdwan district in West Bengal, India.It is situated at a distance of about 6

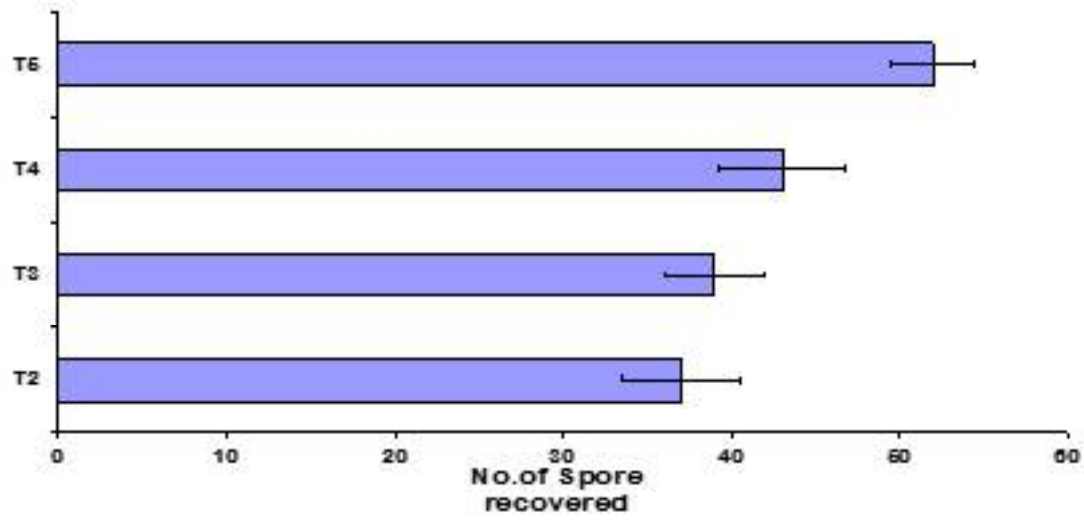

Fig. 2. Effect of fungivory on the total number of recovered Spore (Mean ± 95% confidence intervals

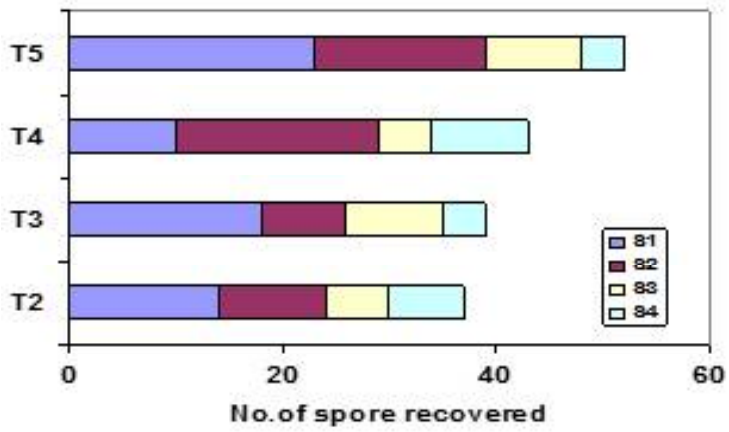

Fig 3. Effect of fungivory on the recovered spores of different fungi. (S1- *Altenaria alternata*, S2-*Cladosporium cladosporioides*, S3-*Fusarium chlamydosporum* and *Aspergillus flavus*).

km(approx) in the south –west of Barddhaman Railway station. These area falls between 23^{0}23'14"N latitude and 87^{0}86'15"E longitude at an altitude of 30m above sea level. The soil characteristics of the studied area are sandy loam to alluvial with moderate porosity. In the present study soil samples were collected from non-rhizosphere and rhizosphere of most dominant plant species of study sites at monthly interval over a period of two years (January 2014 to December 2015). Dhillon(1964) [11]described an apparatus for collection of soil samples, which was later, modified by Roy (1967)[12] has been applied in this sampling process. In the present investigation Tullgren funnel as modified by Murphy (1962)[13] was used for extraction of the arthropods. The fungal population was assessed by inoculating the soil solution of 10^{-3} and 10^{-4} dilutions. The conventional dilution plate method was followed using potato dextrose agar (PDA). This experiment was based on the methodology of Guevara et al., 2002. In this experiment 50 microcosms replicates were set up in a plastic chamber (8 x 8 cm high and diameter) with a 2 cm^2 hole in the lid covered with nylon mesh. 2 gm of wet leaf litter extracted from a bulk amount was added to each container. Leaf litter was collected at the study site (Site-I) from the surface area of randomly selected points and chopped into 1cm^2 pieces, wet and homogenized. Then, from the bulk chopped leaf litter were weighed (50 samples of 2 gm each) to a precision of 0.1ìg, taking each sample from the centre of the bulk to minimize variation in water content between samples. Then a suspension of spores of four fungi *Alternaria alternata, Cladosporium cladosporioides, Aspergillus flavus* and *Fusarium chlamydosporium,* commonly occurring in the soil and leaf litter of the study site was prepared. Each species were grown in a 5cm Petri dish(2% malt agar extract at 30^{0}C).After 10days the mycelium surface of each plate was washed with 120ml of distilled water and the spore as well as mycelium fragments were mechanically suspended. These suspensions were mixed and then added 5ml to the 40 chambers. By artificially increasing the relative dominance (spore number) of these 5 species, an attempt was taken to minimize potential differences in the composition of the micro-biota between microcosms. The micro arthropods fauna in all chambers were eliminated by fumigating them with Chloro benzene crystals for 5min and followed by ventilation for 30 min. Then out of these 50 chambers 10 chambers kept defaunted (T_2), 10 chambers was added with one adult oppid mite(*Oppia yodai)*-T_3;10chambers was added with one adult collembolan species *(Onychiurus* sp.)-T_4; 10 chambers was added with an adult *Oppia yodai* and one adult *Onychiurus* sp.-T_5.Leaf litter of remaining 10chambers was dried and stored as control for initial dry weight (T_1). The 40 experimental microcosms were placed in a green house for 60 days during September, 2014-November, 2015. Then leaf litter of each 40 microcosms dried at 70^{0}C for 3 days and the leaf litter weighed to a precision of 0.1ìg, together with the dried leaf litter of the 10 control chambers. The weight of the leaf litter after 60 days between treatments and the overall control were compared. The overall one-way ANOVA was analysed to detect significant differences between treatments.

After weighting the leaf litter as described above 10 ml of distilled water was added to the 40 microcosms (each of 10 microcosms of T_2 T_3,T_4 and T_5) and shook them vigorously before taking a 50il sample from each to classify and count all spores under a compound microscope (Magnus,Olympus). The overall number of recovered spore and the number of different types of spore recovered in all the treatments were compared

RESULTS AND DISCUSSIONS

In this study effect of fungivory on leaf litter decomposition was assessed by microscoms experiment. In this experiment a comparison of 4 treatments (T_2-Fungi only,T_3-Fungi+*Oppia yodai*,T_4-Fungi+*Onychiurus* sp.,T_5-Fungi + *Oppia yodai* + *Onychiurus* sp) with the Control(Sterilized and defaunted-T_1) were made. The weight of control was taken before 60 days of incubation. Loss of litter weight after 60 days of incubation was taken as a measure of decomposition rate. The weight of control was

taken as initial weight and final weight of the leaf litter was taken as the response variable. It has been revealed from the present experiment that the presence of both Acarina(*Oppia yodai*) and Collembola (*Onychiurus* sp) in treatment -5 resulted in significant loss of leaf litter weight (Fig:1) .The means comparison test showed that the difference between control(0.816 gm) to T_2(0.765gm) and control to T3(0.721gm) were not significant. Treatment -4 showed slightly higher loss of weight (0.645gm) than that of T_2 and T_3 but significantly lower than treatment-5(0.431gm).

One way ANOVA (Table: 1) showed significant differences between treatments (F=29.09; df 4.45 at $p<0.001$ and $p<0.05$).

Effect on Fungal reproductive potential

The effect of fungivory on fungal community structure was evaluated on the basis of reproductive potential of different species (*Alternaria alternata, Cladosporium cladosporioides, Fusarium chlamydosporium* and *Aspergillus flavus*) by the total number of recovered spore as well as total number of different spore recovered with ±95% confidence interval only. At the end of the experiment altogether 4 types of spore were identified from 90% of microcosms based on shape and appearance as well as hyphal fragments. The spores were corresponded to those species added at the beginning of the experiment. These are spores of *Alternaria alternata, Cladosporium cladosporioides, Fusarium chlamydosporium* and *Aspergillus flavus*. Comparisons between treatments showed significant differences between total number of spores recovered in T_2 (35±4) and T_5 (52±3.5) (Fig: 2). The number (Mean± 95 % confidence level) of four different types of spore recovered showed more or less same trend (Fig:3) except in treatment -4. Number of spores of *Alternaria alternata* showed significant incresement over T_2 (12±1) in T_5 (23±2.5) and T_3 (18±1.5) but not in T_4 (9±1). The number of recovered spore of *Cladosporium cladosporoioides* was higher (18±2) than that of *Alternaria alternata* in T_4. The spores of *Fusarium* and *Aspergillus* recovered from all the treatments were numerically lower than that of *Alternaria* and *Cladosporium*.

From the results of the present study it has been revealed that fungivory is one of the key interactions which accelerate decomposition. This results corroborates studies of Cortet et al. (2003) Hedlund and Oehrn (2000),. Beare et al. (1992) and several others. This may be probably due to consumption of detritus and grazing of microbes by diverse fauna which generally increases decomposition and the mineralization rate by increasing the surface area of decomposing substrates for microbial attack (Verhoef and Brussard, 1990) and can affect fungal growth by grazing and dispersal of fungal propagules (Renker et al.2005, Hanlon and Anderson, 1979, 1980). This present study was largely consistent with the findings of Setälä and Huhta(1990),Sulkava and Huhta(1998), Sulkava et al.(2001) that diverse fauna (T_5 of the present study) will affect the rate of decomposition process. However, the observed result may be due to the fact that fungi were differentially affected by fungivory. *Alternaria alternata* appeared to be more active (high spore production) in all the treatments except in T_4.*Cladosporium cladosporoioides* found to be more active in T_4. These findings indicate that fungivory affects the activity (Spore production) of litter fungi and fungivores performed different on a variety of offered fungi indicating distinct resource (food) qualities.Fungivory by the Acarines and Collembola i.e. a complex and diverse faunal make up has the potential to affect fungal community and the decomposition process by selectively feeding on a strong combative species. All these results were corroborate with the findings of Tiunov and Scheu (2005) Parkinson et al. (1979), Lussenhop (1992) and several others

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