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An Approach to Studying Microfungal Succession on Decaying Pine Needles Litter in Chandak Forest in the Presence of Cow Dung

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ABSTRACT

Fungi inhabiting pine needles were isolated by dilution plate technique. pH content of the decomposed Pine needles mixed with cow dung was 8.3 - 9.6. Soil moisture content was 68.7% in the month of September and minimum 24 % in the month of May. Eighty nine fungal species were recorded from litter samples examined throughout the year. Maximum fungal species 71 were found in the month of August and minimum 18 in the month of December.

Key words: microfungal, succession, Pine litter, needles, cow dung, decaying.

INTRODUCTION

Pinus roxburghii Sarg. (syn. Pinus longifolia Roxb.) (Pinaceae), commonly known as chir pine, is one of the five pines found in India - Pinus roxburghii Sarg, Pinus wallichiana Jackson, Pinus gerardiana Wall, Pinus kesiya Royle ex Gord and Pinus armandi French and the most widely occurring. It is also known as Himalayan long needle pine, long leaved Indian pine, Indian chir pine, chir or chil, is a tall tree with a spreading crown found in the Himalayan from Kashmir to Bhutan, Afghanistan and in southern Indian hills (Shuaib et al., 2013; Tiwari et al., 2016). Vegetation of different parts of northern area was described by various workers. Chaudhri (1960) described vegetation of Kaghan valley.

Fungi are some of the most important organisms in the world, because of their vital role in ecosystem function, influence on humus and human-related activities (Mueller and Bill, 2004). Fungi are not only beautiful but play a significant role in the daily life of human beings besides their utilization in industry, agriculture and medicine (Cowan, 2001). Macrofungi need moisture to develop. The peak mushrooms and macrofungi season for each region is different for each ecological climate (Arora, 1991). The number of fungi recorded in India exceeds 27,000 species, the largest biotic community after insects (Sarbhoy et al., 1996).

In recent time this tree is facing environmentalogist scornance. Even government stopped its plantation. People think that this tree does not support eco-friendly behavior. This tree is xerophytic in its habitat. It can remain not only in adverse climate but also in adverse soil. Due to its comeliness this tree is also used as ornamentation. One of the demerits of this tree is its oily leaves which take long time in decomposition. During summer months its dry leaves catch fire instantly and cause baneful consequences. It burns everything all pine associates in the forest. In present study the decomposition of pine litter along with micro fungal succession in hilly condition of Pithoragarh forest would be studied. Till date no attempt has been conducted in the region, Pithoragarh. In this paper, fungal succession on Pine needles in the presence of cow dung was studied on different seasons throughout the year.

MATERIALS AND METHODS

Study area and climate

The study was conducted in pure

Pine forest of Chandak at Pithoragarh District of Kumaun Himalayas at an altitude of 1600 m. Pithoragarh district is located between 29° 13' 50" N and 80° 11' 30" E to 29°58'1" N and 80°22'1" E with a geographical area of 7100 km2. The topography ranges from 500 m to 4000 m and consists of different forest types. 1000-1500 m represents semi-temperate type forest. Above 1500-2000 m represents sub temperate type forest and 2000-3000 m represents temperate type of forest. Mostly Quercus sp., Rhododendron sp. and Cedrus deodara are dominant species. The climatic data is given in the Fig.1.

Collection and treatment of Pine leaf litter

The pine litter (only fresh fallen leaf litter) was collected in the summer season (from April to June) in horizontally placed hanging net from Chandak Pine forest and brought to the laboratory for further research work. A total of 36 nylon litter bags of (25x25 cm) with 2 mm mash size were taken and filled with homogenously mixed cow dung and pine needles (1:1 ratio). All litter bags were carried to the study area and partially covered under the previously fallen litter on the forest floor. The litter bags were collected in the triplicate from and a total of 3 bags were taken every month. The process continues up to one year. pH if the litter and soil was estimated by pH meter. Moisture content of the litter sample were calculated by following formula-

$$\text{Mc}\ (\%) = \frac{(\text{Ww} - \text{Wd})}{\text{Ww}} \times 100$$

- Mc = moisture content (%) of material
- Ww = wet weight of the original sample, and
- Wd = weight of the dried sample.

Mycological observation

For fungal determination of litter dilution plate technique was used (Bisht, 2012). For mycoflora determination, 5 g of litter were chopped with a sterile knife and grinded; the fine litter was blended in 100 ml of sterile water and shaken in a mechanical shaker for 15-20 min to obtain a homogeneous dispersal. From this initial suspension, 1 ml of 1 × 10-3 serial dilution was pipette into each of four replicates of Potato Dextrose Agar (PDA) medium and Czapek's Dox Agar medium with streptomycin sulfate (300 µg/ml), which was cooled to 45°C, and poured into Petri dishes. The dishes were incubated at 25 ± 2°C near UV light. After 5-7 days examined for fungal growth. Identification was based on morphology following examination stereo and compound microscopes and scientist of FRI.

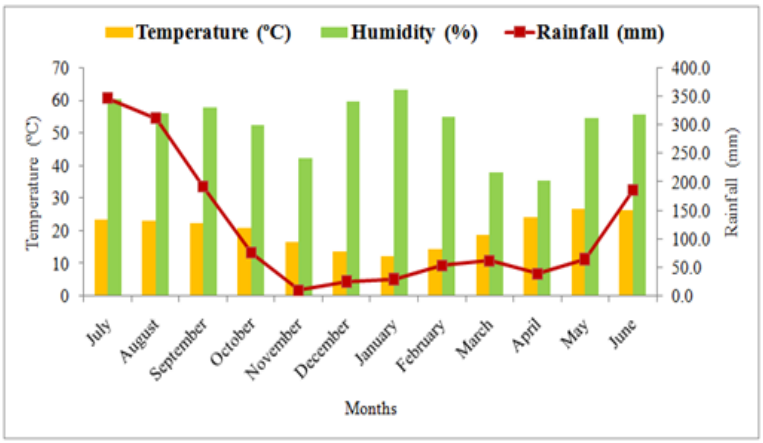
The data of each month and each parameter were collected in triplicate and averaged for analysis in excel 2010.

RESULTS

The pH of litter was between 8.3-9.6. Soil pH of the forest was between 5.55-7.79 and soil moisture content was 68.7 % in the month of September and minimum 24% in the month of May (Table.1). Eighty nine fungal species were recorded from litter samples examined throughout the year listed in the Table.2. Maximum fungal species 71 were found in the month of August and minimum 18 in the month of December. Whole year was divided into 4 seasons winter, summer, rainy and autumn. In the rainy season the fungal growth and their colonies were found highest than other seasons. Some species of higher fungi like mushrooms grew only in rainy season. Few species of Alternaria and Trichoderma remain throughout the year. The linear correlation between litter pH and growing mycoflora y = 8.3563x-35.224 and R^2 = 0.0568 on cow dung treated litter. The linear correlation between humidity and growing mycoflora was y= -0.5493x+69.982 and R^2 = 0.0706 on application of cow dung.

Table.1. Monthly litter and soil pH and soil moisture

Months	Cow dung treated litter pH	Soil pH	Soil Moisture (%)
July	8.6	5.65	68.3
August	8.4	5.85	68.5
September	8.3	5.55	68.7
October	9.6	6.65	38
November	9.5	6.55	36
December	9.3	6.75	32
January	9.6	6.55	30
February	9.4	6.65	28
March	9.5	6.55	36
April	9.5	6.45	29
May	9.6	6.79	24
June	8.4	6.05	36



(Data source: Meteorological department Pithoragarh)

Fig.1. Annual temperature, humidity and rainfall of the study area

Table.2. Mycoflora present in cow dung treated litter across the year

Species name	January	February	March	April	May	June	July	August	September	October	November	December
Absidia clavata			+	+			+	+	+	+	+	
Absidia spinosa			+	+				+	+			
Absidia tuneta		+	+				+	+	+	+		
Acremonium sp.			+	+				+	+			
Agaricus campastris							+	+	+			
Agaricus silvicola								+	+			
Alternaria tenuis	+	+	+	+	+	+	+	+	+	+	+	+
Alternaria alternate	+	+	+	+	+	+	+	+	+	+	+	+
Alternaria chlamydospora	+	+	+	+	+	+	+	+	+	+	+	+
Alternaria solani	+	+	+	+	+	+	+	+	+	+	+	+
Amanita chepangiana							+	+				
Amanita muscaria								+	+			
Amanita vaginata								+				
Arthrobotrys sp.							+	+	+			
Aspergillus candidus	+	+	+				+	+	+	+	+	+
Aspergillus flavus		+	+	+	+		+	+	+	+	+	
Aspergillus niger			+	+	+		+	+	+	+	+	
Aspregillus awamori	+	+	+					+	+	+	+	+
Aspregillus fumigates		+	+	+	+	+	+	+	+	+	+	
Aspregillus tamari	+	+	+				+	+	+	+	+	
Aspregillus terreus	+	+	+	+				+	+	+	+	+
Aspregillus ustus		+	+	+		+	+	+	+	+	+	
Aurobasidium sp.		+	+	+	+	+	+	+	+	+	+	
Biospora antennata						+	+	+				
Bipolaris sp.			+	+				+				
Botrytis sp.								+	+	+		
Cantharellus cibarius							+	+	+			
Cephalotrichum sp.		+	+					+	+	+		
Cercospora sp.	+	+	+	+								
Chetomium atrobrunnem		+	+						+	+		
Chetomium cupreum	+	+	+					+	+	+		
Chetomium globosum		+	+	+					+	+	+	
Chrysosporium keratinophilume	+	+	+								+	+
Cladosporium cladosporides			+				+	+	+	+	+	
Cladosporium herbarum	+	+	+				+	+	+	+	+	
Cladosporium oxysporum			+	+			+	+	+	+	+	
Cladosporium tennuissium	+	+	+	+			+	+	+	+	+	+
Coniella castaneicola							+	+	+			
Coprinus comatus							+	+				
Coprinus disseminatus									+	+		
Curvularia clavata			+	+	+	+		+	+	+		
Curvularia geniculata			+	+			+	+	+			
Curvularia lunata		+	+	+	+		+	+	+	+		
Drechslera gramineum		+	+		+	+				+	+	+
Drechslera rostrata		+	+						+	+		
Epicoccum nigrum												

Continued Table 2.....												
Fusarium longipes		+	+	+			+	+	+	+		
Fusarium moniliforme		+	+	+			+	+	+	+		
Fusarium oxysporium		+	+	+			+	+	+	+		
Genoderma lucidum							+	+	+	+		
Helminthosporium anomalum							+	+	+			
Humicola grisea	+	+							+	+	+	
Hydnum repandum								+	+	+		
Laccaria amenthystea								+	+			
Lactarius volemus								+	+			
Lycoperdon perlatum								+				
Mortierella subtilissima									+			
Mucor circinelloides			+	+			+	+	+			
Mucor hiemalis			+	+			+	+	+	+		
Mucor mucedo			+	+			+	+	+			
Mucor racemosus			+	+			+	+	+	+		
Myrothecium roridum				+	+	+	+	+	+			
Nigrospora saccharina		+	+				+	+	+	+		
Penicillium chrysogenum	+	+	+	+	+	+	+	+	+	+	+	+
Penicillium citrinum		+	+	+	+	+	+	+	+	+	+	
Penicillium expansum	+	+	+	+	+	+	+	+	+	+	+	
Penicillium notatum		+	+	+	+	+	+	+	+	+	+	
Phaeotrichum sp.	+	+	+							+	+	+
Phoma fimeti								+	+			
Phoma glomerata			+									
Pilaria sp.							+	+				
Piptocephalis sp.						+	+					
Pithomyces sp.										+		
Podospora sp.								+	+			
Ramaria stricta							+	+				
Rhizopus oryzae			+	+	+	+	+	+	+	+		
Rhizopus stolonifer				+	+		+					
Scleroderma geaster							+	+	+			
Sordaria fimicola								+	+			
Stachybotrys atra			+					+				
Stemphylium pyriforme	+	+									+	+
Thamnostylum sp.			+	+					+			
Trichoderma harzianum	+	+	+	+	+	+	+	+	+	+	+	+
Trichoderma koningii	+	+	+	+	+	+	+	+	+	+	+	+
Trichodrema viridae	+	+	+	+	+	+	+	+	+	+	+	+
Trichothecium roseum		+	+	+								
Trichurus sp.								+	+	+	+	
Verticillum alboatrum	+	+	+				+	+	+	+	+	+
Verticillum tenuissimum	+	+						+	+	+	+	+

DISCUSSION

Availability of 89 fungi species in decomposing litter revealed spore or mycelium availability as well as favorable conditions for fungal growth in the study site. The listed fungi of lower and higher groups (Table.2) revealed common species which have already been reported from different forest types in Uttarakhand and other Himalayan forest (Kumar and Singh, 2010; Guleri et al., 2010; Gaur and Kaushik, 2011; Chaturvedi et al., 2012; Thakur and Harsh, 2014). The present study revealed that the use of cow dung accelerated the fungal growth which may also increase the decomposition of pine needles because cow dung made the substrate rich in carbon and nitrogen (Douds et al., 1997; Muthukumar and Udaiyan, 2000; Tiwari et al., 2016).

In different seasons round the year, the occurrence of maximum fungi in rainy season revealed the best climatic condition for growth and development and the finding are supported by different reports (Tiwari, 1992; Bisht 2012; Lodge and Cantrell, 1995; Guadarrama and Alvarez-Sanchez, 1999; Codina et al.,2008) where the relative humidity in the air (Reponen et al., 1996), moderate temperature (Koske, 1987) and environmental factors (Walstad et al., 1970: McCune et al., 2002) are correlated with season growth of fungi in natural habitats. pH of the cow dung mixed litter was more than 7 which indicates that high pH is good for the fungal growth. Increase in pH is good for better growth of fungal species

(Rousk et al., 2010.). The best growth and development of the mycoflora was observed between pH 6.5 (Eraso and Gancedo, 1987; Gock et al., 2003) to pH 9.5 (Wheeler et al., 1991; Blagodatskaya and Anderson, 1998).

CONCLUSION

Present study concluded that cow dung can increase the fungal growth in Pine needles which can also increase its decomposition. Pine needles are highly acidic in nature and its decomposition is slow because it does not provide good substrate for fungal growth. Here a new approach was used to increase population and succession on fallen pine needles mixed with cow dung.

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