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STUDY OF CELLULOLYTIC FUNGI ASSOCIATED WITH SOILS OF CASUARINAS FROM MAHARASHTRA

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ABSTRACT

Casuarinas are the important nitrogen fixing non-leguminous tree species. The study was conducted during 2018-19 from the soils of *Casuarina* plantations which were abundantly growing in coastal and interior parts of Maharashtra. Total of 5 cellulolytic fungal genera and 18 species from rhizosphere soils and 9 species from non rhizosphere like *Aspergillus, Chaetomium, Penicillium, Trichoderma* and *Rhizoctonia* were isolated from these soils. The abundance of cellulose degrading enzymes of cellulolytic fungi indicated the recycling of nutrients is done efficiently by use of enzymatic system of organisms and that will help in soil reclamation. Thus, Plants are important in agroforestry is because of the presence of these fungi in rhizosphere .

Key words: *Casuarina, Cellulolytic fungi, soil reclamation*

Article History

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➢ INTRODUCTION:

Casuarinas are the important nitrogen fixing non-leguminous tree species. It is a native of Southern hemisphere and comprise of a group of trees and shrubs of about 80 species. They are economically important plants grown as plantations under agro-forestry programmes for fuel and fodder all over the world (Crowley G.M 1994). Due to its capacity to improve infertile soils, it is introduced in arid and semiarid regions of the world successfully. The tree is multipurpose and helps to improve physical and chemical properties of soil, suppresses the weeds, helps in maintaining the fertility of soil. The success of the plantation is contributed to various properties of the plant and soil interactions.

Soil is an important medium for many microorganisms that help the plants and nature by providing them with different metabolites. The ability of Casuarinas to grow vigorously on poor soils is due to their symbiotic association with several microorganisms. These fungi are also well known agents of decomposition of organic matter and cellulosic substrates (Lind et.al 2002). The micro-flora includes one such groups-cellutolytic fungi that helps to break down polysaccharide cellulose like *Rhizoctonia, Fusairum, Penicillium, Chaetomium* etc. which have cellulolytic properties. The cellulolytic microbes secrete cellulolytic enzymes which play an important role in natural biodegradation processes in which they degrade lingo-cellulosic material (C.Coronado-ruiz et. al. 2018). The large number of fungal forms inhabits the soil and plays an important role in bio-gradation of organic matter. They are identified and described on the basis of colony characters, structure of mycelium, form, colour, texture, spore colour, fruit bodies etc. (Ainsworth et. al., 1973; Manoharachary; 1986). The Microfloral studies associated with the roots of *Casuarina* show that they are rich in fungal associations and help in plant growth and establishment. (Kulkarni , 1998)

The present study aims at isolation and identification of cellulolytic fungi associated with the soils of *Casuarina* plantations from different parts of Maharashtra and its effect on soil improvement.

➢ MATERIAL AND METHODS:

In the present investigation the rhizosphere and non-rhizosphere soil samples from Casuarinas were collected from various coastal and interior localities of Maharashtra state like Alibagh, Dapoli, Ratnagiri and Ahmednagar during October 2018-19. Soil samples were collected from 10-15 cms below the surface in clean polythene bags.

Isolation of soil fungi was done using soil dilution technique. The fungi were grown on medium like Jensen's Agar, Waksman Agar and Cellulose agar that were used to isolate different fungi from soil. Proper conditions of growth were maintained i.e. Temperature- 30-35^{0}c, pH 4 to 6 under dark conditions were maintained for seven days for growth of cellulolytic fungi. Cotton blue stain and lacto phenol mountant were used for study.

The fungal colonies were isolated in pure culture on the respective media and with the help of slide cultures identification was done. The different fungal forms were isolated from the soils and studied under the compound microscopically. The identification of the fungal forms is done as per the Handbook of Soil Fungi – New Delhi (Nagmani et. al 2006) .

➢ OBSERVATIONS:

Different cellulolytic fungal forms were isolated and maintained in pure cultures on Jensen's Agar medium. These include-

1. *Aspergillus* Mich. Ex Fr.

Aspergillus is a common imperfect fungi with its perfect state in ascomycetous fungi. It is ubiquitous in nature and consists of many species in nature. *Aspergillus* species are important in various industries. One of its role includes biodegradation of organic matter with the help of cellulolytic enzymes. The class nigri are efficient producers of hydrolytic enzymes like cellulases. (Ibatsem Khokhar et. al. 2012).

- Colonies – light to dark coloured, variously coloured depending on the spore colour. Colony cottony,loose, thread like.

- Fruit body – Condiophores mostly globose of varying sizes, chains of conidia. conidia black, green, orange, brown coloured.

2. ***Chaetomium*** Kunze and Schmidt

Chaetomium is one of the rapidly growing ascomycetous fungus on cellulosic substrate. It initially produces white cottony colonies which change their colour as it progress to maturity. It is commonly recognized due to ostiolate fruit bodies with membranous perithecial wall covered by hairs. It produces asci and pigmented ascospores.

- Colonies – White to Creamish, cottony thread like.
- Fruit body - Perithecia variously shaped either round to oval with hairy appendages containing oval to cylindrical asci and dark ascospores.

3. *Penicillium* Link

Penicillium is one of common imperfect fungi with its perfect state in ascomycetous fungi. It is of major importance in food spoilage, drug production and natural degradation. *Penicllium* shows cellulase activity by producing cellulolytic enzymes. It has great potential as an alternative source of enzymes for degradation of lignocellulosic biomass. It also produces xylanolytic enzymes while growing on cellulose as well as xylan.

- Colonies – Green, dark green, blue to orange in colour, textured.
- Fruit body – Erect condiophores. Brush like phialades present on the conidiophores bearing chains of conidia. Conidia are coloured.

4. Rhizoctonia Kuhn.-

Rhizoctonia is a strong saprophyte of the imperfect fungi with its perfect state in Basidiomycetous fungi. They are capable of surviving for extended periods of time in the absence of living host plants by feeding on decaying organic matter. The fruit bodies are black coloured sclerotia. They produce pectinolytic and cellulolytic enzymes which help them to degrade the organic matter.

- Colonies – white with black heads.
- Fruit body – Black coloured sclerotia.

5. Trichoderma Pers.

Trichoderma is an imperfect fungi with its perfect state in ascomycetous fungi. It efficiently degrades the plant cellulosic cell wall. The species are well known for their capacity to produce cellulolytic enzymes which help it to form endophytic interactions with plant roots.

- Colonies – smooth to ornamented, cottony, appear grey to brown in colour.
- Fruit body – branched conidiophores. 2-5 stalk like phialades present on the conidiophores bearing coloured conidia.

An observation table I shows the different isolated fungal forms.

TABLE I

Sr. No.	Species from Rhizosphere soils (RS)	Species from Non- Rhizosphere soils (NRS)
1.	*Aspergillus niger* Tiegh.	*Aspergillus niger* Tiegh.
2.	*Aspergillus flavus* Link.	*Aspergillus flavipus* (Bainier & Sartori)Thom & Church
3.	*Aspergillus terreus* Thom	*Aspergillus humicola* Chaudhuri & Sachar.
4.	*Aspergillus nidulansi*(Eidam) Wint.	*Aspergillus ochraceus* Wilh.
5.	*Aspergillus flavipus* (Bainier & Sartori)Thom & Church	*Chaetomium gracile* Udagawa
6.	*Aspergillus awamori* Nakaz.	*Chaetomium spirale* Zopf.
7.	*Chaetomium amberpetense* Rama Rao & Ram Reddy	*Penicillium glabrum* (Wehmer) Westling.
8.	*Chaetomium convolutum* Chivers.	*Penicillium purpurogenum* Stoll.
9.	*Chaetomium indicum* Corda.	*Trichoderma viride* Pers.
10.	*Chaetomium reflexum* Skolko & J. W Groves	
11.	*Penicillium chrysogenum* Thom.	
12.	*Penicillium citrinum* Thom.	
13.	*Penicillium glabrum* (Wehmer) Westling.	
14.	*Penicllium rubrum* Stoll.	
15.	*Rhizoctonia solani* Kuhn.	
16.	*Trichoderma harzianum* Rifai.	
17.	*Tricoderma fasiculatum* Bisset.	
18.	*Trichoderma viride* Pers.	

RESULT & DISCUSSION:

The results show that a variety of cellulolytic fungal genera inhabit the soils of Casuarina. They include 18 species isolated form rhizosphere soils and 9 species isolated from non rhizosphere soils. Four species are commonly found in both the soils. These species are rich in cellulolytic enzymes are therefore help to degradation of the organic matter and litter component of the soil. These results are showing same effect as experiments conducted by Stursova' M. et. al. 2012.

Thus it can be concluded that the *Casuarina* soils are rich in degrading fungal forms and can be used for degradation of cellulolytic components from soil that will help to improve the soil fertility. The *Casuarina*

plantations are done in coastal and barren lands so that these fungi help in cellulosic degradation and make the soil porous and usable. The study reveals a step towards soil reclamation in degraded lands.

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