The BioScan 5 (2) : 243-245, 2010

SEASONAL OCCURRENCE OF ENDOPHYTIC MYCOFLORA OF INNER BARK OF MEDICINAL PLANT *ACACIA CATECHU* WILLD

T. G. NAGARAJA* AND P. G. DEVKAR
Department of Botany, The New College, Kolhapur - 416 012
Department of Agrochemicals and Pest Management
Shivaji University, Kolhapur - 416 004
E-mail: tgnagaraja @Rediffmail.Com

INTRODUCTION

Endophytic microorganisms are those that inhabit interior of plants especially in branches, bark and stems showing no apparently harm to host (Azevedo, 1998). Endophytes include fungi that have one or more of a variety of interaction with their host plant, some fungi are wide spread and are found on many different plant species and others are highly specific to single hosts in single environment.

*Acacia catechu* Willd (Mimosaceae) is moderate sized deciduous tree growing naturally in Konkan and western ghats. The bark of this medicinal plant is used in infusions or decoctions as astringent, antidysenteric and antipyretic. The most important product is catechu or katha, useful in melancholia, conjunctivitis, haemoptysis and skin diseases. Various plant parts are used in sore mouth, pain in chest, asthma, colicky pain, gravel phthisis etc., gum resin used in masticatories has been reported from this plant (Joshi, 2000).

Since *Acacia catechu* Willd like all other plants harbour endophytic fungi. We become interested in a possible contribution of fungal diversity in the bark. Endophytic fungi have been isolated from leaves, stem and roots of many woody plants in the tropics (Bettucci et al., 1999). Endophytes have a capacity to protect their hosts against insect pests, pathogens and even domestic herbivores, animals and production of phytohormones (Ma et al., 2004) and other compounds of biotechnological interest (Strobel and Daisy, 2003). Taxol, an potent anti cancer agent was isolated from an endophyte *Pestalotiopsis microspora* in a Yew tree and Gibberlic acid an growth hormone from an endophyte *Gibberella fujikuroi* harbouring rice plant advocates endophytes are potential source of useful metabolites.

The present study was carried out to isolate and identify fungal endophytes in the bark of *Acacia catechu* Willd or katha, which is indigenous medicinal plant in India.

MATERIALS AND METHODS

Fresh bark samples were harvested from tree growing in Shivaji University, campus, Kolhapur on morning hours, in three different seasons viz., rainy, winter and summer seasons with the help of ethanol disinfected knife, bark pieces were cut at 1-2 meter above the ground level and the depth of 1-1.5 cm in the trunk region. The collected bark pieces were brought to the laboratory and surface sterilized by dipping the sample pieces in 70 percent alcohol (v/v) for 1 minute followed by 1-2 minutes in 3-5 % sodium hypochlorite solution (v/v) in a 500 mL beaker. Soon after bark pieces were cut in to the size of 4 X 4 cm for experimental study, later bark pieces were washed thoroughly with distilled water with several times. The excess of water was blotted to dry with blotting paper. The outer skin was removed slowly with ethanolic disinfected knife and inner portion containing cortex segments were cut and are plated on water agar medium (20 g L\(^{-1}\)) mixed with septran (100 mg L\(^{-1}\)) and incubated in a chamber for 21 days at 24ºC. The petriplates were allowed to grow endophytic fungi monitored regularly. From the sterilized bark pieces of hyphal tips started growing slowly, such hyphal

### KEY WORDS

- Endophytes
- *Acacia catechu*
- *Aspergillus niger*
- *A. flavus*
- *Fusarium oxysporum*
- *Chaetomium globossum*

### ABSTRACT

Endophytic fungi from inner bark of medicinal plant *Acacia catechu* Willd were studied in different seasons during 2007-2008. A total of 32 endophytes were recorded during rainy season followed by 29 endophytes in winter season and 19 endophytes in summer season. *Aspergillus niger*, *A. flavus*, *Trichoderma viridi* and *Penicillium* sp., were found to be dominant endophytes followed by *Fusarium oxysporum*, *Chaetomium globossum* and *Verticillium* sp.
masses were transferred to PDA. Each fungus was assigned a number and stored properly. The endophytic mycoflora were identified based on morphological characters using standard manual, some isolates were documented at the Department of Botany, Shivaji University, Kolhapur.

The percent frequency of occurrence of endophytic mycoflora was calculated by the number of bark segments of Acacia catechu Willd colonizing a specific fungus divided by total number of segments plated on agar multiplied by 100 (Fisher and Petrini, 1987). The dominant endophytes were measured as percentage colony frequency divided by sum total percentage of colony frequency of all endophytes multiplied by 100.

RESULTS AND DISCUSSION
The results have been depicted in Table 1, 2 and 3. only a few studies have been carried out endophytic mycoflora of tropical trees (Mahesh et al., 2005). Genera like Bipora punctata, Geotrichium acacii, Cephalosporium sp., Trichothecium sp., Torula sp., Verticillium sp., etc., have been recorded for the first time as endophytes (Table 1, 2 and 3). During rainy season (Jun 2007 to Oct 2007) a total of 32 species belonging to Ascomycotina and Hypomycotina fungi were recorded in the bark of Acacia catechu Willd (Table 1). During winter Trichoderma viridi was found to be dominant species, followed by Aspergillus flavus. Whereas in summer season (Feb 2008 to Mar 2008) Penicillium sp., were found to be dominant. Thus Aspergillus flavus, A. niger, Fusarium oxysporum, Drechslera sp., Chaetomium globosum and Trichothecium roseum were dominant endophytes isolated from inner bark of Acacia catechu Willd. A similar mycoflora was documented by Mahesh et al., (2005) in inner bark of Azadirachta indica.

The endophytes such as Acremonium sp., Phialocephala sp., commonly have been isolated from tropical and sub tropical plants (Koga et al., 1997). Whereas Fusarium oxysporum, Drechslera sp., are pathogenic to crops, but some times, they may get modified by mutation and grow into useful non pathogenic endophytes.

Aerobasidium sp., Populari sp., Thielopsis sp., was found during rainy season. Cladosporium sp., Cephalosporium sp., Choanephora sp., was isolated during winter season. Whereas Phoma sp., Penicillium sp., Trichothecium sp., was recorded in summer season. This seems occurrences of these endophytes are influenced by several environmental variation (Ahholm et al., 2002).

The toxic products synthesized by endophytic fungi in woody plants and that were able to modify growth and death rates in larvae of the spruce bud worm C. fumiferana feeding on balsam fire (Calhoun et al., 1992). The endophytes in this case were identified as Phyllosticta and Hormonema dematioides and the toxic compounds were mainly heptelidic acid and regulosine, even tremorgenic toxins in tropical woody plant infected with an endophytic fungus from the genus Phomopsis (Bills et al., 1992). Penicillium sp., have been found to produce important antibiotics, which kill bacteria and other microorganism. Antibiotic Phomol was isolated from fermentation of Phomopsis sp., endophytic fungus from Erythrina cristagalli (Weber et al., 2004). Thus endophytes providing protection against pathogens as well as they are potential biocontrol agents and could be utilized to protect tissue culture plants before they are transplanted to the field. Again we are currently studying the secondary metabolites synthesized by endophytes, which may capable of protecting plants from pest and disease attack.

ACKNOWLEDGEMENT
The authors are thankful to Co-ordinator Department of Agro Chemicals and Pest Management, Shivaji University, Kolhapur for providing laboratory facilities.

REFERENCES


