



Diversity of Endophytic Fungi of *Catharanthus Roseus* L from Different Localities of Aurangabad District

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ABSTRACT

Endophytic fungi from the stems, roots, leaves, and flowers of *Catharanthus roseus* L. were isolated using standard isolation protocol. A total of 41 endophytic fungal isolates were obtained from seven different locations of the Aurangabad region. These endophytic fungal isolates were classified into 11 fungal genera. Most of these fungi belonged to *Alternaria* sp., *Aspergillus* sp., *Fusarium* sp., and *Collectotrichum* sp. and species. *Catharanthus roseus* L. Conservation of medicinally important plants through sustainable practices is one of the methods for their availability. Another alternative approach for the production of secondary metabolites or biopharmaceuticals is the search for fungal endophytes having medicinal importance.

Keywords: Endophytic fungi; *Catharanthus roseus* L.; *Alternaria*, *Aspergillus*, *Fusarium* and *Collectotrichum*.

I. INTRODUCTION

Medicinal plants are the reservoir of biopharmaceutical compounds traditionally used for the treatment of human ailments. Deforestation or the constant use of medicinal plants makes them threatened or endangered (Chinedu, 2017). Conservation of medicinally important plants through sustainable practices is one of the methods for their availability. Another alternative approach for the production of secondary metabolites or biopharmaceuticals is the search for fungal endophytes having medicinal importance. The long association of fungi with plants in mutuality's mode probably makes fungi rich sources for bioactive compounds (Pimentel, 2011).

Terrestrial plants *Catharanthus roseus* L. are known to harbor endophytic microbes, which in many cases produce bioactive molecules (Hyde KD et al.2008). Endophytes are microbes that live in the internal tissues of the host plant without showing any symptoms of disease (Bacon CW et al., 2000), however, recent studies have shown that they may become pathogenic during host plant senescence (Rodriguez R et al. 2008). Whilst living symbiotically with plants, many endophytes have developed the ability to mimic host chemical diversity and are also believed to be responsible for the production of biologically active metabolites once attributed to the plant host (Shweta Set all. 2010). Some of the chemically complex structures that are of dual endophyte/plant origin and which have been investigated in the last few decades include paclitaxel

(Yang X et al. 2004), vinca alkaloids (Ueda J et al. 2002), camptothecin (Shubha M et al. 2007), podophyllotoxin (Kour A et al. 2008) and hypericin (Kusari Set al. 2008). Therefore, endophytes are potential sustainable sources of plant-associated natural products

Catharanthus roseus L. (Apocynaceae) is a plant well known for the production of the anticancer alkaloids vincristine and vinblastine (Duflos A et al. 2002). The antimetabolic effect of these alkaloids is due to their ability to arrest cell division during metaphase through binding tubulin and inhibiting spindle fiber formation (Himes RH et al. 1976). The crude extracts of *Catharanthus roseus* L. are known for their cytotoxic activity against an array of tumor cell lines, including human fibrosarcoma cells, human cervix adenocarcinoma, human lung adenocarcinoma, murine colon carcinoma, murine Lewis lung carcinoma, and murine melanoma cells (Tung CY et al. 2002). In addition to *Catharanthus roseus* L., an endophytic *Fusarium oxysporum* strain isolated from *Catharanthus roseus* L. was also reported to produce vinca alkaloids (Tillet D et al. 2000). However, despite the pharmacological importance of endophytic compounds very few investigations have focused on isolating endophytes from *Catharanthus roseus* L. and exploring their chemical diversity and bioactivities (Lingqi Z et al. 2000 & Ravindra NK et al. 2008)

II. MATERIALS AND METHOD:

Collection of Plant Samples Healthy fresh and mature leaves of *Catharanthus roseus* L. (Apocynaceae) were collected from a healthy plant grown in different localities of Aurangabad, Maharashtra. The plant material was brought to the laboratory in sterile bags and processed within a few hours after sampling. Fresh plant materials were used for isolation work to reduce the chance of contamination.

III. ISOLATION OF FUNGAL ENDOPHYTES

Catharanthus roseus L. plants were collected from different areas of Aurangabad (Maharashtra, India) to determine the number of genera and species of endophytes present in leaf, stem, and root tissues. After plant selection, leaf, stem, root, and flower tissues were cut with the help of a sterile scalpel and placed in sterile plastic bags to store the material at 4°C until the isolation procedure was started. Endophytic fungi were isolated from the leaf, stem, and root tissues of *Catharanthus roseus* L. obtained from different areas of the Aurangabad District. The leaf, stem, and root tissues were cut into small pieces approximately (0.2 cm x 0.2 cm) and surface sterilized with 0.01% mercuric chloride (HgCl₂) solution for 90 seconds and washed thoroughly with sterilized distilled water. Residual water on their surface was removed by soaking on sterile blotting paper. Small pieces of leaves were placed on the surface of potato dextrose agar (PDA) poured into Petri dishes.

IV. IDENTIFICATION OF ENDOPHYTE ISOLATES

Purified endophyte fungal isolates were identified by the Slide culture technique in a moist chamber for morphological characterization using standard manuals described by (Alexopoulos and Beneke, 1961), Barnett, 1960.

V. RESULTS

Host plants and endophytic fungi are symbionts, in which hosts and endophytes benefit from each other. Plants provide nutrition and protection to their endophytes in return, endophytes excrete functional products and increase their host resistance to biotic and abiotic stresses. The living aerial parts of all plants may have mutualistic internal endophytic fungi, which exhibit a momentous part of fungal diversity. Endophytic fungi of the plant *Catharanthus roseus* L.

of the Aurangabad region exhibit high diversity. A total of 41 endophytic fungal isolates were obtained from seven different locations of the Aurangabad region. These endophytic fungal isolates were classified into 11 fungal genera. Most of these fungi belonged to *Alternaria* sp., *Aspergillus* sp., *Fusarium* sp., and *Collectotrichum* Sp. species. The maximum colonization was observed in *Fusarium solani*. Ecologically as a fungal endophyte., *Fusarium solani* diversified in most of the locations of this region in the plant *Catharanthus roseus* L. Mycelia sterile was a large group of fungi that failed to sporulate and was ubiquitous in plant endophytic isolation.

Endophytic fungi isolation from *Catharanthus roses* L.

Name of fungi	Root	Stem	Leaf	Flow er
<i>Aspergillus niger</i>	+	+	+	-
<i>Collectotrichum</i> Sp.	-	+	+	+
<i>Aspergillus flavus</i>	+	+	+	-
<i>Fusarium solani</i>	+	+	+	+
<i>Macrophomina phaseolina</i>	+	-	-	-
<i>Fusarium veticilliodes</i>	+	+	+	-
<i>Curvularia</i> sp.	+	-	+	-
<i>Alternaria alternata</i>	+	+	+	-
<i>Helmenthosporium</i> p.	-	+	+	-
<i>Talaromyces radius</i>	-	-	-	+
<i>Phoma</i> sp.	+	+	+	-
<i>Verticillium</i> sp.	-	-	-	-
<i>Trichoderma</i> sp.	+	+	+	-
<i>Colletotrichum</i> sp.	-	+	+	-
Mycelia sterile	+	-	+	+
<i>Aspergillus fumigatus</i>	+	-	-	-

+ Present, Absent –

VI. CONCLUSION

Endophytic fungi of the plant *Catharanthus roseus* L. of the Aurangabad region exhibit high diversity. A total of 41 endophytic fungal isolates were obtained

from seven different locations of the Aurangabad region. These endophytic fungal isolates were classified into 11 fungal genera. Most of these fungi belonged to *Alternaria* sp., *Aspergillus* sp., *Fusarium* sp., and *Collectotrichum* Sp. The maximum colonization was observed in *Fusarium solani*.

Conservation of medicinally important plants through sustainable practices is one of the methods for their availability. Another alternative approach for the production of secondary metabolites or biopharmaceuticals is the search for fungal endophytes having medicinal importance.

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