

# Fungal Endophytes Associated with the Indian laburnum (*Cassia fistula* L.)

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## Abstract

Endophytes are unknown symbionts that live in plant tissues without indicating their presence. The association of endophytes with plants is well established. They not only help in water and mineral uptake but also play an important role in metabolic activities of certain plants. Endophytes play an imperative role to maintain the health of plants, as they can protect or prepare the plant against abiotic and biotic stresses and help in enhancing growth and yields. In present investigation about endophytes in plants, Indian laburnum (*Cassia fistula* L.), a flowering plant found mostly in India and South Asia belonging to legume family of angiosperm was used. Traditionally the leaves of plant are used for skin diseases, burning sensation, dry cough, dysentery, inflammation, fever etc. Isolates from leaves collected from Aurangabad district of Marathwada region in India showed the presence of fungal endophytes. Fungal strains like *Phyllosticta fallopiae*, *Colletotrichum cobbittiiense* and *Diaporthe brasiliensis* was found.

**Key words:** Fungal Endophytes, Indian Laburnum, Bioactive compounds.

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## Introduction

More than 20,000 bioactive metabolites are of microbial origin (1). It has been estimated that there may be 1.5 million fungal species, while only about 100,000 species are presently known (2). The horizontally transmitted endophytes have been reported from all major groups of plants including algae (3-4), lichens (5) mosses and ferns (6), conifers (7) and angiosperms(8- 9), and may persist even in aseptically cul-

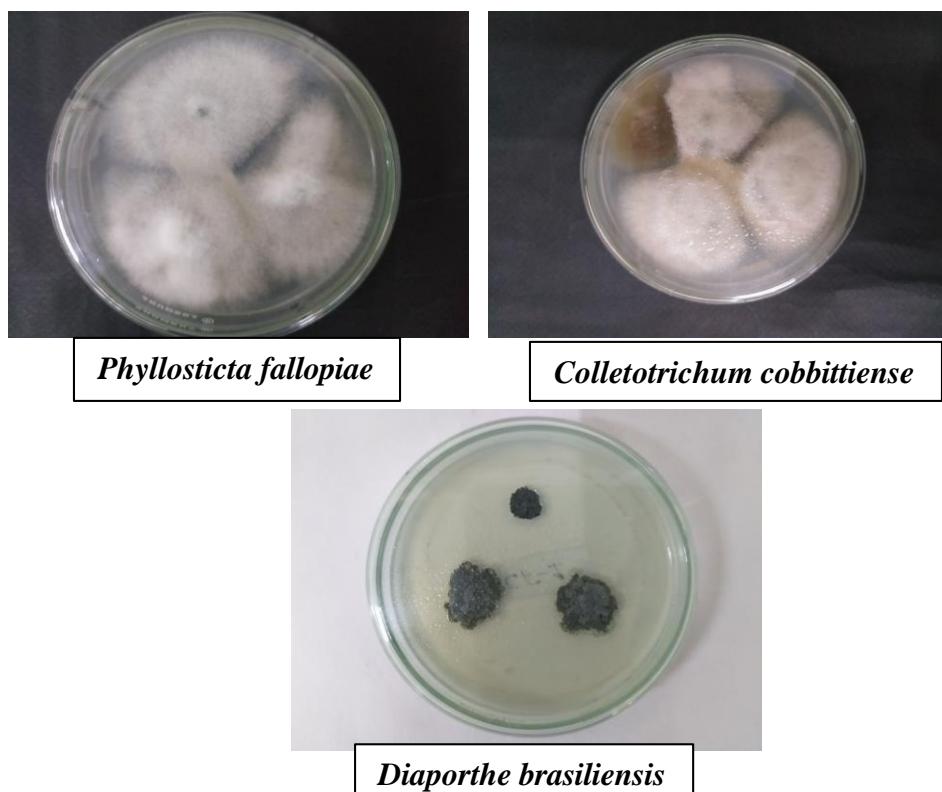
tured plants(10). Endophytes are the microbes that colonize living internal tissues of plants without causing any immediate overt symptoms (11). They are found in almost all plants studied, including liverworts, hornworts, mosses, lycophytes, equisetopsids, ferns and seed plants from arctic to the most biologically diverse tropical forests (12). Plant-associated microbes have also been recognized for their ecological roles influencing host populations, plant communities (13-14), biosynthesis, biotransformation and biodegradation (15-16). Individual plants can harbor dozens of endophytic fungal species (17) and these endophytes contribute to the hyper diversity of fungi (18). Medicinal herbs are an important group of hosts for endophytic fungi (19). Endophytes from Chinese medicinal plants show efficacy as pharmaceutical and agricultural compounds

(20-21). The various natural products produced by endophytic fungi possess unique structures and great bio-activities, representing a huge reservoir which offers an enormous potential for exploitation for medicinal, agricultural and industrial uses (22-23). Fungi are among the most important groups of eukaryotic organisms that are well known for producing many novel metabolites which are directly used as drugs or function as lead structures for synthetic modifications (24). In present investigation the plant leaf of Indian laburnum (*Cassia fistula* L.) was collected from different locations in and around Aurangabad city of Maharashtra state to discourse number of endophytes present in it. In the preliminary investigation endophytic fungi were evident that has been isolated, screened and identified.

## Material and Methods

The leaves of experimental plant which is a deciduous tree was collected from Dr. Babasaheb Ambedkar Marathwada University campus during

the months of July to October 2018 from Aurangabad district. They were brought to research lab in sterile polythene bags. Leaves were washed thoroughly in distilled water, blot dried, and first immersed in 70% ethanol (v/v) for one min followed by second immersion in sodium hypochlorite (3.5%, v/v) for three minutes (25). They were rinsed three times in changes of sterile distilled water and dried on sterile blotters under the airflow to ensure complete drying. 0.5cm x 0.5 cm size bits were excised with the help of a sterile blade and placed on PDA plates. Periodically the bits were examined for the appearance of fungal colony and each colony that emerged from segments was transferred to another plate of PDA for further identification. The morphological and molecular identification of the isolates was carried out by NCIM CSIR-NCL, Pune. All fungal mounts were made on microscopic glass slides in lactophenol-cotton blue and cultures which failed to sporulate were grouped as mycelia sterilia.



**Figure 1:** Leaf endophytes showed presence of fungi

## Results

A total of 3 isolates were obtained from 54 tissue fragments from Indian laburnum (*Cassia fistula* L.) plant. Extent of endophytes colonization varied in plant parts where leaf fragments recorded more endophytes than that of root fragments. Leaf endophytes showed presence of fungi like *Phyllosticta falllopiae*, *Colletotrichum cobbittense* and *Diaporthe brasiliensis*. Leaf fragments were rich in mycelia sterilia as they didn't show any kind of sporulation.

## Discussion

Efforts are on to isolate novel bioactive compounds from various endophytes that are living in the internal tissue of plants. Current investigation highlights preliminary examination in these regard and focuses on endophyte diversity in Indian laburnum (*Cassia fistula* L.). Further study will be elaborative which will be based on increased sample area from all over Marathwada region of Maharashtra and explore more parameters. Effect of seasonal variations will be considered on the nature of endophytes plant holds. Isolates will be subjected to determine presence of bioactive compounds to obtain newer antioxidants with therapeutic applications if any.

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## Disclosure

The authors report no conflicts of interest in this work. No violation of human rights and safety.

## References

1. Bérdy J. Bioactive microbial metabolites: a personal view. *J of Antibiotics*. 2005; 58: 1–26.
2. Hawksworth DL. Fungal diversity and its implications for genetic resource collections. *Studies in Mycology*. 2004; 50: 9–18.
3. Zuccaro A, Schoch CL, Spatafora JW, Kohlmeyer J, Draeger S, Mitchell J. Detection and identification of fungi associated with the brown seaweed *Fucus seratus*. *Appl Environ Microbiol*. 2008;74:931–941.
4. Suryanarayanan TS, Venkatachalam A, Thirunavukkarasu N, Ravishankar JP, Doble M, Geetha V. Internal mycobiota of marine macroalgae from the Tamilnadu coast: distribution, diversity and bio-technological potential. *Bot Mar*. 2010;53:457–468.
5. Suryanarayanan TS, Thirunavukkarasu N, Hariharan GN, Balaji P. Occurrence of non-obligate microfungi inside lichen thalli. *Sydowia*. 2005; 57:120–130.
6. Petrini O. Taxonomy of endophytic fungi of aerial plant tissues. In: Fokkema NJ, van den Heuvel (eds) *Microbiology of the phyllosphere*. Cambridge University press, Cambridge, 1986; pp 75–187.
7. Giordano L, Gonthier P, Varese GC, Miserere L, Nicolotti G. Mycobiota inhabiting sapwood of healthy and declining Scots pine (*P. sylvestris* L.) trees in the Alps. *Fungal Divers*. 2009; 38:69–8.
8. Saikkonen K. Forest structure and fungal endophytes. *Fungal Biol Rev*. 2007;21:67–74.
9. Tejesvi MV, Ruotsalainen AL, Markkola AM, Pirttilä AM. Root endophytes along a primary succession gradient in northern Finland. *Fungal Divers*. 2010;41:125–134.
10. Lucero ME, Barrow JR, Osuna P, Reyes I, Duke SE. Enhancing native grass productivity by co-cultivating with endophyte-laden calli. *Rangeland Ecol Manag*. 2008;61:124–130.
11. Petrini O. Taxonomy of endophytic fungi of aerial plant tissues. *Microbiology of Plant Microbe Interactions*. 1986;16: 580–587.
12. Bacon CW, White JF. *Microbial Endophytes*. New York: Marcel Decker INC, 2000; pp. 237–261.
13. Clay K, Holah J. Fungal endophyte symbiosis and plant diversity in successional fields. *Science*. 1999;285: 1742–1744.
14. Rudgers JA, Clay K. Endophyte symbiosis with tall fescue: how strong are the impacts on communities and ecosystems? *Fungal Biology Reviews*. 2007;21: 107–124.
15. Koide K, Osono T, Takeda H. Colonization and lignin decomposition of *Camellia japonica* leaf litter by endophytic fungi. *Mycoscience*. 2005;46: 280–286.
16. YuW, Dai CC. Endophytes: a potential resource for biosynthesis, biotransformation, and biodegradation. *Annals of Microbio*. 2011; 61: 207.
17. Arnold AE, Lutzoni F. Diversity and host range of foliar fungal endophytes: are tropical leaves biodiversity hotspots? *Ecology*. 2007;88: 541–549.
18. Hawksworth DL. The magnitude of fungal diversity: the 1.5 million species estimate revisited. *Mycological Research*. 2001;105: 1422–1432.
19. Huang WY, Cai YZ, Hyde KD, Corke H, Sun M. Biodiversity of endophytic fungi associated with 29 tradi-

tional Chinese medicinal plants. *Fungal Diversity*. 2008;33: 61–75.

- 20. Shentu XP, Chen LZ, Yu XP. Anti-fungi activities and cultural characteristics of gingko endophytic fungus No. 1028. *Acta Phytophyl Sin*. 2007;34:147–152.
- 21. Kusari S, Lamshöft M, Zühlke S, Spitteler M. An endophytic fungus from *H. perforatum* that produces hypericin. *J Nat Prod*. 2008;71:159–162.
- 22. Tan RX and Zou WX. Endophytes: a rich source of functional metabolites. *Natural Product Reports*. 2001;18: 448-459.
- 23. Zhang HW, Song YC and Tan RX. Biology and chemistry of endophytes. *Natural Product Reports*. 2006;23: 753-771.
- 24. Stadler M, Keller NP. Paradigm shifts in fungal secondary metabolite research. *Mycological Research*. 2008;112:127–130.
- 25. Schulz B, Wanke U, Draeger S, and Aust HJ. “Endophytes from herbaceous plants and shrubs: effectiveness of surface sterilization methods.” *Mycological Research*. 1993; 97(12):1447–1450.