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# *Taxomyces andreanae*: A source of anticancer drug

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**Abstract**
Plant endophytic fungi spend the whole or part of their life-cycle colonizing inter or intra-cellularly inside the healthy tissues of the host plants without causing any apparent harm to the host plant. *Taxomyces andreanae,* a hyphomycetous endophytic fungus is found symbiotically associated with its host plant, *Taxus brevifolia* and produces anticancer bioactive compound Taxol (paclitaxel). Biosynthesis of Taxol in this endophytic fungus starts with the cyclization of terpene, geranylgeranyl diphosphate and involves enzymes from different classes which are located in different cellular compartments. Taxol, a highly-oxygenated diterpenoid natural product is therapeutic because acts as a mitotic spindle poison which inhibits uncontrolled cancerous growths. The aim of present study is to review the research status on Taxol-producing fungal endophyte, *Taxomyces andreanae* and the ongoing efforts to develop heterologous Taxol biosynthesis.

**Keywords:** Endophytic fungi, *Taxomyces andreanae*, Taxol, *Taxus brevifolia*, Plant-host interaction

## 1. Introduction
Plant endophytic fungi are one of the important components of plant micro-ecosystems [1, 2]. Plant endophytic fungi can be defined as the fungi which spend the whole or part of their life cycle colonizing inter or intra-cellularly inside the healthy tissues of the host plants, typically causing no apparent symptoms of any disease. Endophytes have been found in each plant species examined, and it is revealed that there are over one million fungal endophytes existed in the nature [3]. They have been recognized as an important and reliable resource of natural bioactive products with potential applications in medicine, agriculture and food industry [4-6].
Since the "gold" bioactive compound Taxol (paclitaxel) discovered from the endophytic fungus *Taxomyces andreanae* in 1993 [7], many scientists have been showing their keen interests in studying fungal endophytes as potential producers of biologically active compounds. *Taxomyces andreanae* is a hyphomycetous endophytic fungus which is found symbiotically associated with its host plant, *Taxus brevifolia* and produces anticancer bioactive compound Taxol. During the past two decades, numerous valuable bioactive compounds from *T. andreanae* with antimicrobial and anticancerous properties have been successfully isolated. These bioactive compounds may be classified as alkaloids, terpenoids, steroids, quinones, lignans, phenols and lactones [8]. During the long period of co-evolution, a symbiotic relationship was progressively established between each endophytic fungus and its host plant. The host plant can supply ample source of nutrition and easeful niche for the survival of its endophytes. On the other hand, the endophytes can produce a sufficient amount of bioactive compounds for helping the host plants to cope with biotic and abiotic stresses, and promoting the host growth in return [9]. This study would be beneficial for us in studying the existing relationships between the endophytes and their host plants, and to develop a substitutable approach for efficiently producing these rare and valuable bioactive compounds [10].
This chapter mainly describes the research progress on the fungus, *Taxomyces andreanae* for producing bioactive compounds such as Taxol, which are also produced by their host plants *e.g. Taxus brevifolia, T. baccata, T. chinensis, T. canadensis* etc.

## 2. Classification
| | |
|---|---|
| Domain | : Eukarya |
| Super-kingdom | : Opisthokonta |
| Kingdom | : Fungi |
| Phylum | : Ascomycota |
| Class | : Hyphomycetes |
| Order | : Hyphomycetales |
| Genus | : *Taxomyces* |
| Species | : *andreanae* |

## 3. Source of *Taxomyces Andreanae*
Endophytic fungus, *Taxomyces andreanae* is found symbiotically associated living within a species of yew tree, *Taxus brevifolia* [7]. However it has long been known that yew trees produce an important anticancer compound, Taxol, *Taxomyces andreanae* is the first known isolated fungus to produce specifically this compound at a higher concentration than its host itself [11, 12]. *Taxus brevifolia* is a small to medium-sized shade tolerant evergreen tree, growing 10–15 m tall and with a trunk up to 50 cm diameter. It grows in varying types of environments, however, in drier environments it is generally limited to stream side habitats, whereas in moist environments it will grow up onto slopes and ridge tops [13].

## 4. Structure and Metabolism
Taxomyces andreanae is considered as a novel endophytic fungus found associated with the inner bark of T. brevifolia. Like many other Ascomycota genera, this fungus has septate and multinucleated hyphae of various sizes, ranging from 1.2 to 3.75 µm in diameter. To develop clumps of loosely constructed bulbil-like cells is the characteristic feature of this genus. These clumps are of various shapes and sizes typically ranging from 5×5 to 16×30 µm in diameter and length. The

nucleated cells in these clumps appeared to be loosely packed in the bulbil and are incapable of germination. This fungal endophyte can be easily isolated and grown in a lab on many common media by covering a plate with its mycelium in roughly 3-4 days. It generally lacks clamp connections and dolipore septations. Its teleomorph is found to be unknown [14].

## 5. Mechanism of Taxol Production

Taxol, a highly-oxygenated diterpenoid natural product isolated from the pacific yew tree (Taxus brevifolia), is undoubtedly one of the most successful anticancer drugs of all time [15]. The word Taxol (with a capital T) is a registered trademark name for a drug formulated with the paclitaxel. The mechanism for biological production of Taxol in Taxus plants have been mentioned in Fig. 1. Several key steps required to produce Taxol have been characterized at the biochemical and genetic levels involving 20 or more enzymatic reactions [16, 17]. The biosynthetic pathway starts with the cyclization of geranylgeranyl diphosphate to form taxa-4(5), 11(12)-diene and involves enzymes from several different classes that are located in different cellular compartments, including the plastid, endoplasmic reticulum and cytosol.

While horizontal gene transfer has long been proposed for the biosynthesis of Taxol in Taxomyces andreanae, it has been recently revealed that the endophyte genomes did not contain any sequences

With significant homology to the Taxol biosynthetic genes from *Taxus spp*. Heinig *et al* [18] indicating independent Taxol biosynthesis in endophytes from its plant host.

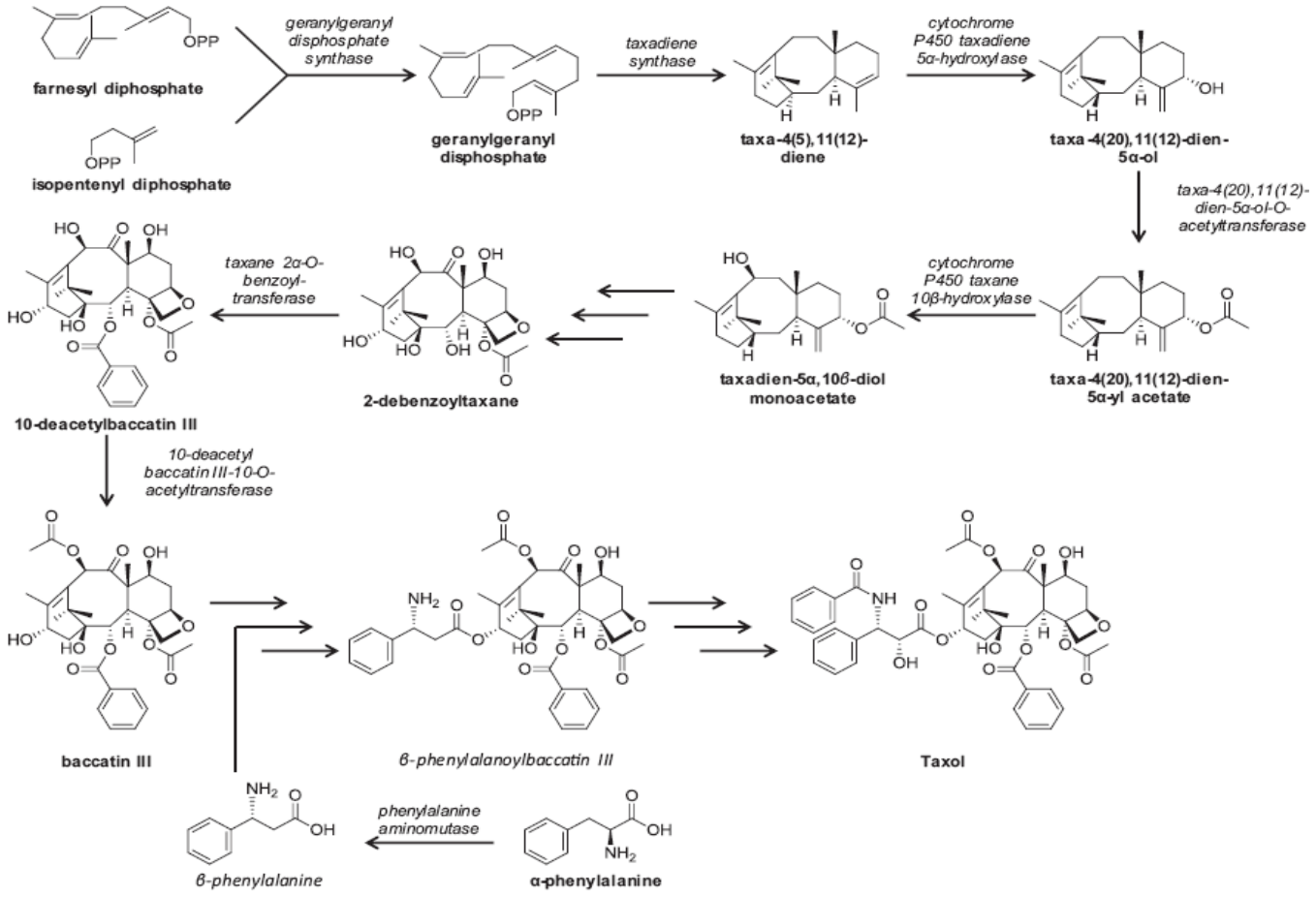


**Fig 1**: Graphical representation of Taxol biosynthesis in *Taxus spp.* (adapted from Walker & Croteau,) [19]. Multiple arrows indicate several as yet undefined steps.

Microbial Taxol and taxane biosynthesis was found in several other different genera of fungi including *Alternaria, Aspergillus, Cladosporium, Fusarium, Monochaetia, Pestlotia, Pestalotiopsis, Pithomyces, Penicillium* and *Xylaria*, which were isolated from yew and non-Taxus plants [20].

## 6. Significance

In the history of natural products from endophytes is the production and extraction of multibillion-dollar anticancer drug Taxol. This particular compound was initially obtained from the Pacific yew tree, *Taxus brevifolia* [21], a traditional medicinal plant used by Native Americans [22]. Since then, many other species of the genus *Taxus* have been reported to produce Taxol. These plants are generally slow-growing with isolated geographical distribution. Studies of endophytes from this plant revealed that the fungus, *Taxomyces andreanae*, also produce the exact same compound [23]. Taxol is a therapeutic because it is a mitotic spindle poison which inhibits uncontrolled cancerous growths (i.e. the spindle apparatus which aligns chromosomes during cell division (mitosis) is disrupted by preventing cancer cells from reproducing). Cancer is the second most frequent cause of death in industrialized countries, and subsequent treatments are urgently needed.

Taxol, an anticancer drug, finally got FDA (Food and Drug Administration) approval to treat ovarian cancer in 1992 and breast cancer in 1994 [24], although, its use was deemed impractical due to its low yield from yew trees. To treat a single patient of cancer with 5% saline solution, the bark of three yew trees was required [12]. Therefore, the discovery of *Taxomyces andreanae* in the early 1990s by Andrea and Donald Stierle, and their 1994 patent for Taxol production from this fungal endophyte, represented a remarkable step in the field of biotechnology and chemotherapy. Significant amount of Taxol have been extracted from *Taxomyces andreanae* when allowed to grow in a semi-synthetic liquid medium and subjected to a fermentation process [7]. Many other fungal species have been found to produce Taxol within and outside of the *Taxomyces* genera, however, often with a much lower yield and less stability when compared to *Taxomyces andreanae* [11].

## 7. Plant-Host Interaction

As an endophyte, *Taxomyces andreanae* exists entirely contained within the host without either member being harmed in this mutual interaction. Despite being a parasite, *Taxomyces andreanae* is not found to be pathogenic to the Pacific yew tree. Endophytes are unique in their ability to synthesize bioactive natural products, often which benefit their host in some or other way [18]. The bioactive compounds produced by *T. andreanae*, such as Taxol, benefit the yew tree by being toxic to predators by antagonistic mechanism or fulfilling some other vital needs of the host plant. It is revealed by the "balanced antagonism" hypothesis that the interaction between host plant and endophyte is a delicate balance between virulence and defense. It is found that the endophytes are not always symbiotically associated with host plant but may act as latent pathogens, which may become virulent under certain environmental conditions. However, recent studies indicate that this interaction is a complicated and tightly regulated system of communication and association rather than a delicate balance [25]. To explain the co-existence of endophytes within plants, several hypotheses have been proposed. Considering these studies, it is now well known that plants are complex communities of different endophytes that are mutually influencing the metabolic pathways and survival of all interacting organisms, including the host.

## 8. Conclusion

Plant endophytic fungi, as a novel and abundant secondary metabolites resource, owning their special ability to produce or secrete the same or similar compounds originated from their host plants, as well as other bioactive compounds, have increased many investigators interesting in both basic research and applied fields. The discovery of *T. andreanae* have raised greater interest in the prospects of microbial Taxol (paclitaxel) production. Endophytes like *Taxomyces andreanae* are of high interest to researchers for the production of medicinally important compounds that have applications as antibiotics and chemotherapeutic drugs.

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## 10. Conflict Of Interest

The authors hereby declare that there is no conflict of interest

## 11. References

1. Zhang HW, Song YC, Tan RX. Biology and chemistry of endophytes. Natural Product Reports, 2006; 23:753-771.
2. Rodriguez RJ, White JF, Arnold AE, Redman RS. Fungal endophytes: diversity and functional roles. New Phytologist 2009; 182:314-330.
3. Petrini O. Fungal endophytes of tree leaves. Microbial Ecology of Leaves, 1991, 179-197.
4. Strobel G, Daisy B, Castillo U, Harper J. Natural products from endophytic microorganisms. Journal of Natural Products. 2004; 67:257-268.
5. Gunatilaka AAL. Natural products from plant-associated microorganisms: distribution, structural diversity, bioactivity, and implications of their occurrence. Journal of Natural Products. 2006; 69:505-526.
6. Verma VC, Kharmar RN, Strobel GA. Chemical and functional diversity of natural products from plant associated endophytic fungi. Natural Product Communications, 2009; 4:1511-1532.
7. Stierle A, Strobel G, Stierle D. Taxol and Taxane Production by *Taxomyces andreanae*, an Endophytic Fungus of Pacific Yew. Science, 1993; 260:214-216.
8. Xu L, Zhou L, Zhao J, Jiang W. Recent studies on the antimicrobial compounds produced by plant endophytic fungi. Natural Product Research and Development, 2008; 20:731-740.
9. Silvia F, Sturdikova M, Muckova M. Bioactive secondary metabolites produced by microorganisms associated with plants. Biologia, 2007; 62:251-257.
10. Zhou L, Zhao J, Xu L, Huang Y, Ma Z, Wang J, *et al*. Antimicrobial compounds produced by plant endophytic fungi. *Fungicides: Chemistry, Environmental Impact and Health Effects*. New York, 2009, 91-119.
11. Guo BH, Wang YC, Zhou XW, Hu K, Tan F, Miao ZQ, *et al*. An Endophytic Taxol-prducing Fungus BT2 Isolated from *Taxus chinensis var. mairei*. African Journal of Biotechnology. 2006, 857-877.
12. Saupe, Stephen G. An Introduction to Plant Taxonomy. Plant Taxonomy (BIOL308), 2008, 29.
13. Wheeler NC, Jech K, Masters S, Brobst SW, Alvarado AB, Hoover AJ, *et al*. Effects of genetic, epigenetic, and environmental factors on Taxol content in *Taxus brevifolia* and related species. Journal of Natural Products. 1992; 55:430-440.
14. Strobel G, Stierle A, Stierle D, Hess WM. *Taxomyces andreanae*, a Proposed New Tax on for a Bulbilliferous Hyphomycete Associated with Pacific Yew (*Taxus brevifolia*). Mycotaxon, 1993; 47:71-80.
15. Brown DT. Preclinical and clinical studies of the taxanes. Taylor & Francis, London, 2003, 387-435.
16. Croteau R, Ketchum R, Long R, Kaspera R, Wildung M. Taxol biosynthesis and molecular genetics. Phytochem Rev, 2006; 5:75-97.
17. Jennewein S, Wildung MR, Chau M, Walker K, Croteau R. Random sequencing of an induced Taxus cell cDNA

library for identification of clones involved in Taxol biosynthesis. Proc Natl Acad Sci U S A, 2004; 101:9149-9154.
18. Heinig U, Scholz S, Jennewein S. Getting to the bottom of Taxol biosynthesis by fungi. Fungal Diversity, 2013; 60:161-70.
19. Walker K, Croteau R. Taxol biosynthetic genes. Phytochemistry, 2001; 58:1-7.
20. Zolia RFB, Ordun F, Cardenas AM, Cotera LBF. Microbial paclitaxel: advances and perspectives. The Journal of Antibiotics. 2010; 63:460-467.
21. Wani MC, Taylor HL, Wall ME, Coggon P, McPhail AT. The isolation and structure of Taxol, a novel antileukemic and antitumor agent from *Taxus brevifolia*. Journal of the American Chemical Society. 1971; 93:2325-7.
22. Gunther E. Ethnobotany of Western Washington. University of Washington Publications in Anthropology. 1945; 10:1-62.
23. Stierle A, Strobel G, Stierle D, Grothaus P, Bignami G. The search for a Taxol-producing microorganism among the endophytic fungi of the Pacific yew, *Taxus brevifolia*. Journal of Natural Products, 1995; 58:1315-24.
24. Goodman Jordan, Walsh V. The story of Taxol: nature and politics in the pursuit of an anti-cancer drug. Cambridge University Press, 2001.
25. Kusari Souvik, Hertweck C, Spiteller M. Chemical Ecology of Endophytic Fungi: Origins of Secondary Metabolites. Chemistry & Biology, 2012; 19:792-798.