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Isolation and Characterization of Fungal Endophytes from Medicinal Plant *Deverra tortuosa*

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Abstract: The study aims to isolate and identify endophytic fungus from the medicinal herb Deverra tortuosa obtained from the Egyptian desert.. Endophytic fungi were isolated from three parts of the plant - root, stem, and flower. Five endophytic fungal species were purified and identified based on morphological characteristics - *Alternaria alternata, Trichoderma viride, Aspergillus flavus, Penicillium chrysogenum,* and *Rhizobus solani*. Some fungi develop colorful colonies, while others are irregularly shaped or circular. Specialized phrases define colony color, form, height, and boundary. The isolated fungal strains have been reported in previous studies to have beneficial bioactivities like plant growth promotion, antimicrobial properties, etc. Endophytic fungi have a significant ecological impact due to their interactions with host plants.. They also produce bioactive secondary metabolites that can have pharmaceutical and agricultural applications.The study concludes that the medicinal plant *Deverra tortuosa* provides a niche for diverse endophytic fungi adapted to arid conditions. Further studies are recommended to evaluate the bioactivities and applications of the isolated fungal strains.

keywords: Endophytic fungi, *Deverra tortuosa*, Wild plants; Desert; Isolation.

1. Introduction

Endophytic fungi are a group of microorganisms that live asymptomatically within the internal tissues of plants without causing any apparent disease. This relationship can be mutualistic, where both the fungus and the plant benefit [1,2]. Endophytic fungi exhibit an elevated level of diversity, and numerous species have been identified. They can belong to various taxonomic groups, including Ascomycota and Basidiomycota. Endophytic fungi contribute to the overall biodiversity of ecosystems. They are found in various plant species across different habitats, and their diversity adds to the complexity of microbial communities [3].

The ecological importance of endophytic fungi extends beyond their interactions with plants. They play a role in nutrient cycling, decomposition, and maintaining ecosystem balance [4]. Endophytic fungi are known to produce a wide range of bioactive secondary metabolites. These compounds can have pharmaceutical, agricultural, or industrial applications and may play a role in defending the plant against pests and diseases [5]. Due to their ability to produce bioactive compounds, endophytic fungi are often explored for their bioprospecting potential. Scientists investigate these fungi for novel natural products that could have applications in medicine, agriculture, and other industries.

The long-standing interaction between plants and fungi involves several fungal species. Fungi are a substantial group of plant pathogens that cause the bulk of plant ailments, accounting for less than 10% of all known fungi [6]. Fungus that are connected to plants make up a small percentage of the overall number of fungi. Most fungi are decomposers, feeding on the remains of dead plants and other living things. Other types of partnerships include fungi acting as decomposers, beneficial symbionts, and endophytes secretive plant colonists [7, 8].

Deverra tortuosa, also referred to as "Shabat El-Gabal," is widely distributed across various

phytogeographical regions in Egypt, especially in desert wadis, sandy areas, and stony plains [9, 10]. This aromatic and glabrous shrub features dichotomously branched stems and caducus leaves with distinct striations [11]. Apart from its multifaceted uses, including as fuelwood, food source, and for medicinal and aromatic purposes, the plant is notably attractive to livestock, particularly camels [12]. It serves as a crucial range plant during the summer months. The plant's shoots find application as a condiment and are employed in treating asthma and intestinal cramps [13]. Furthermore, the essential oil derived from the plant serves as a valuable source of antibiotics effective against certain pathogenic microorganisms [14]. The study aims to isolate and identify endophytic fungus from the medicinal herb *Deverra tortuosa* obtained from the Egyptian desert.

2. Materials & Methods

2.1. Plant material

Deverra tortuosa specimens were gathered from the northern region of the eastern desert of Egypt during the blooming stage, specifically for the purpose of isolating endophytes.

The samples consist of robust and thriving *Deverra tortuosa*. plants, which have been split into four distinct parts: shoot, leaf, flower, and root. Every individual sample was put into an individual sterile bag and sent to the laboratory for further processing. The plant species were identified, verified, and preserved at our laboratory.

2.2. Isolation and Purification of endophytic fungi

The samples obtained from the shoot and root were divided into many pieces, each measuring about 5 mm in diameter, and then rinsed under a continuous flow of water. The pieces were subjected to a 1-minute immersion in 70% ethanol, followed by a 3-minute exposure to sodium hypochlorite (2% available chlorine v/v), and finally a 30-second immersion in 70% ethanol to achieve surface sterilization. Subsequently, they underwent two rounds of washing in sterile distilled water .

Each Petri dish included ten specimens of each organ, coupled with a concentration of 250 mg/l of chloramphenicol. Five replicates of each organ sample were prepared on Petri plates. Subsequently, the plates were placed in an incubator set at a temperature of 26±2°C [15].

After one week, the growing hyphae of the segments were transferred to a new PDA Petri plate for purification. The endophytic fungi, which were kept in isolation, were subculture monthly and maintained on a Potato Dextrose Agar (PDA) slant. The slants were incubated for 5 days at a temperature of 28 °C, and then transferred to storage at a temperature of 4 °C.

3. Results and Discussion

Even though endophytic fungi are one of the most important components of plant micro-ecosystems and should have a considerable influence on the growth and development of host plants, our knowledge of the specific interactions that occur between endophytic fungi and the plants that they are attached to is still restricted.

3.1. Isolation of endophytic fungi

The isolation of fungi is a fundamental step in various scientific and industrial processes, enabling researchers to explore their diversity, functions, and applications across different fields. Fungi that are endophytic were recovered from samples of *Deverra tortuosa* that were growing in the interior desert of Egypt and seemed to be in good condition. It is shown in Figure 1 that the isolation process was carried out on three different components of three different fresh plant samples: the root, the stem, and the flower.

3.2. Purification of endophytic fungi

Purifying fungal cultures helps in obtaining a single, well-defined strain. This is crucial for accurately characterizing the morphology, physiology, and genetic traits of a specific fungal species or strain. Five species of endophytic fungi (Table 1) were isolated from the three parts of *Deverra tortuosa* collected from different habitats (inland desert) as shown in Figures 2. Elbermawi *et al*. [16] isolated *Alternaria* sp. from the leaves of desert plants growing in Egypt (*Lycium schweinfurthii*, *Pancratium maritimum* and *Cynanchum acutum*). Selim *et al*. [17] identified over one hundred fungal endophyte strains from eighteen medicinal plants such as *Alternaria alternate*,

Aspergillus flavus, *Penicillium chrysogenum*, *Penicillium chrysogenum*, etc.

Table 1. Number of fungi endophytes isolated from *Deverra tortuosa* collected from different habitats.

Isolates	*Deverra tortuosa*		
	Root	**Stem**	**Flower**
Alternaria alternata	1	-	-
Trichoderma viride	-	-	1
Aspergillus flavus	-	1	1
Penicillium chrysogenum	-	1	-
Rhizobus solani	-	1	-
Total	**1**	**3**	**2**

3.3. Morphological characterization of isolated endophytic fungi

Endophytic microorganisms have a role in the development of plants and the restoration of plant health via a variety of mechanisms. These mechanisms include the regulation of phytopathogens and the release of phytohormones such gibberellic acids, cytokinin's, and indole acetic acid. Colony morphology is a method that is used by researchers to determine the characteristics of a particular colony of fungus that grows on agar in a Petri dish under controlled conditions. It is possible to make use of it to aid in discovering them.

Various kinds of fungus will form colonies that appear different from one another; some colonies may be colored, while others may be irregularly shaped or round. Common colony characteristics such colony colour, shape, height, and border are described using specialized terms, as illustrated in Table 2.

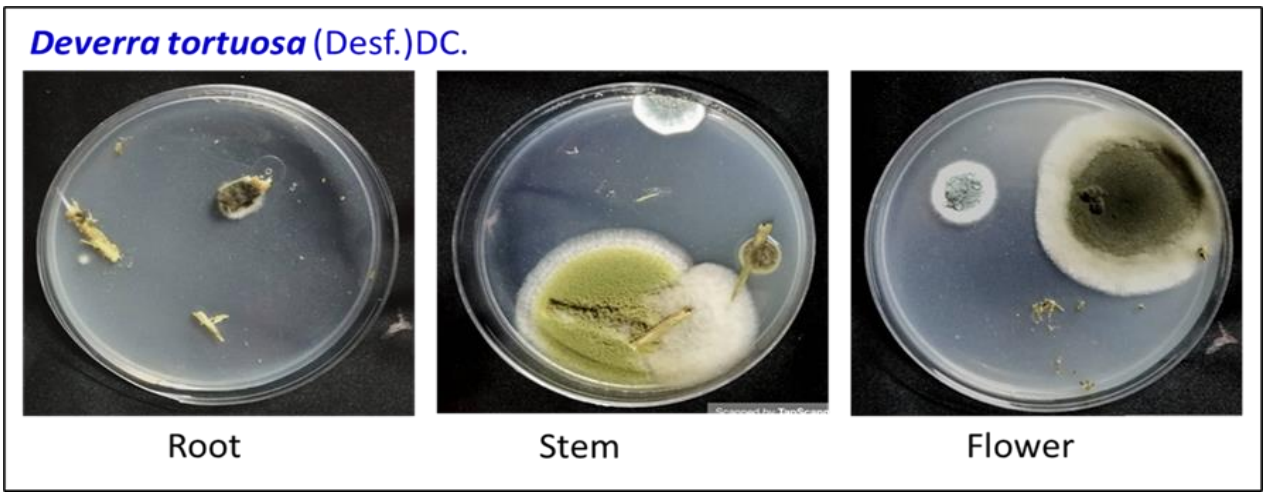

Figure 1. Isolation of endophytic fungi from *Deverra tortuosa* collected from inland desert

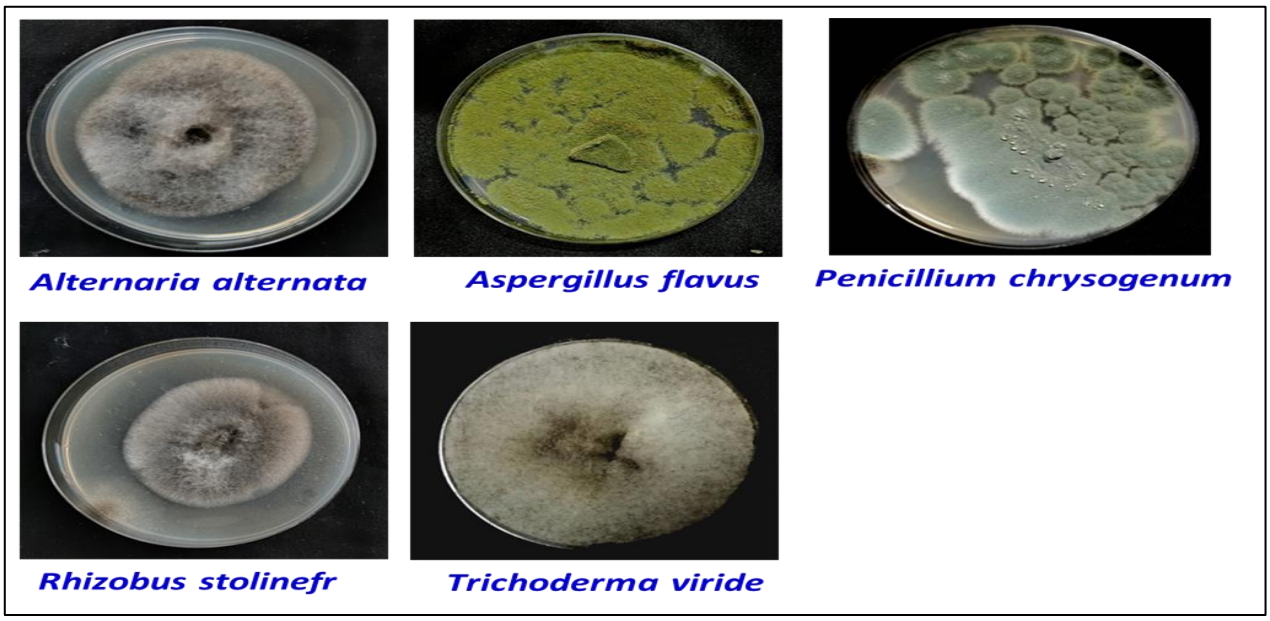

Figure 2. Purification of endophytic fungi isolated from *Deverra tortuosa* in inland desert habitats.

Table 2. Description of fungi endophytes isolated from *Deverra tortuosa* collected from different habitats.

No.	Fungus name	Colony tint		Form	Elevation	Margin
		Surface	Reverse			
1	Alternaria alternata	Greenish Olive	Dark olive	Irregular	Raised	Curled
2	Trichoderma viride	Greenish blue	Yellowish	Irregular	Corvex	Lobate
3	Aspergillus flavus	Yellow	Yellow	Filamentous	Raised	Filiform
4	Penicillium chrysogenum	Dark green	Orange	Irregular	Umbonate	Lobate
5	Rhizobus solani	Whitish pink	Green	Filamentous	Raised	Filiform

4. Conclusion

According to the findings of the present research, the medicinal plant *Deverra tortuosa*, which is indigenous to dry and semi-dry habitats, provides an ecological niche for a wide range of endophytic fungi. Through the process of morphological characterization, it was determined that five different fungal strains were found in healthy portions of *Deverra tortuosa*, including the root, stem, and flower. An examination of the relevant literature revealed that the fungal strains that were found exhibited biological activity in a variety of activities, including agricultural, industrial, and medicinal ones. It is recommended that more experimental research be conducted to evaluate the viability of isolated fungal strains for intended uses, as well as their safety risks.

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