

Short Communication

Exploring Endophytic Fungi of *Suaeda maritima* (L.) Dumort, for Eco-Friendly Bioremediation of Chromium-Contaminated Tannery Sites

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Abstract

The present study explores the range of endophytic fungi associated with *Suaeda maritima* (L.) Dumort., a halophytic plant thriving in tannery effluent-infected soils of Dindigul District, Tamil Nadu. Plant samples have been accumulated from three infected regions, and fungal endophytes had been isolated through trendy surface-sterilization and culture-established techniques. A total of 45 fungal isolates had been recovered, exhibiting colonization frequencies starting from 32% to 54%. Morphological characterization and ITS rDNA-primarily based molecular identification found out 11 wonderful fungal taxa, predominantly belonging to the genera *Aspergillus*, *Penicillium*, *Fusarium*, and *Alternaria*. Diversity indices indicated mild version across websites, reflecting selective variation of endophytic fungi to saline and heavy-steel-harassed environments. These findings highlight the ecological resilience of *S. Maritima* associated fungi and their capability roles in bioremediation, strain tolerance, and bioactive metabolite manufacturing.

Keywords-*Suaeda maritime*, Endophytic fungi, Tannery effluent, ITS sequencing, Fungal diversity, Bioremediation

Introduction

Endophytes, derived from the Greek words endo (within) and phyton (plant), are microorganisms that inhabit inner plant tissues without inflicting seen disorder signs and symptoms (1st Baron Beaverbrook & White, 2000). These institutions are historical, dating returned over 400 million years (Krings et al., 2007). Endophytes encompass fungi, bacteria, and *actinomyces* that colonize almost all plant species and ecosystems—from arid to alpine and tropical environments (Arnold, 2007; Hyde & Soytong, 2008).

Endophytic fungi play important roles in plant fitness with the aid of generating growth-selling and pressure-relieving metabolites consisting of indole acetic acid, ACC deaminase, and phosphate-solubilizing enzymes (Ripa et al., 2019). They also help vegetation in tolerating biotic and abiotic stresses, consisting of heavy metal toxicity (An et al., 2015; Khan et al., 2017). Their

symbiotic dating complements host resilience and contributes to pollutant degradation, nutrient mobilization, and ecological stability (Wen et al., 2022).

In recent years, endophytic fungi were explored as potent agents for bioremediation—the system of transforming or detoxifying pollutants into much less harmful forms. Endophyte-assisted phytoremediation combines the plant's herbal uptake capacity with fungal metabolic ability to remediate contaminated environments effectively (Hussain et al., 2018). This eco-friendly method offers benefits along with low operational price and minimum ecological disturbance

Among commercial pollution, chromium contamination from tanneries remains a extreme environmental subject. India hosts approximately 3000 tanneries, with Tamil Nadu being a major hub (Council for Leather Exports, 2010). Districts like Dindigul, Erode, and Vellore face intense soil chromium accumulation (Murali & Rajan, 2012), adversely affecting crop productivity and soil health.

S. Maritima., a halophytic plant of the own family Amaranthaceae, flourishes in such contaminated web sites and is understood for its adaptability and biological interest. The presence of endophytic fungi may additionally allow this plant to tolerate heavy metals and live to tell the tale in polluted environments. However, confined statistics exists on the variety and function of endophytic fungi in *S. Maritima* inhabiting tannery-polluted soils.

Therefore, the present observe aimed to isolate and symbolize fungal endophytes associated with *S. maritime* growing in chromium-contaminated tannery soils. The isolated endophytic fungi had been diagnosed the use of both molecular and morphological equipment for correct taxonomic classification. Furthermore, the phylogenetic range and taxonomic relationships among the recovered isolates had been assessed, and the range and distribution patterns of endophytic fungi throughout one of a kind tannery effluent-infected sites had been in comparison to apprehend the ecological have an effect on of pollution pressure on endophytic community composition and their ability roles in plant boom promotion and bioremediation.

Materials and Methods

Study Area and Plant Selection

The study turned into performed in tannery effluent-contaminated areas of Dindigul District, Tamil Nadu, India (10°05'–10°09' N; 77°30'–seventy eight°20' E), a prime leather-based-processing hub with >eighty tanneries. Three web sites (Kutiyapatti, Chinnapalapatti, and Begampur), about 1.5 km aside, were selected for sampling (Figure 1). The halophytic plant *S. Maritima*. (Amaranthaceae) become recognized and authenticated on the Rapinat Herbarium, St. Joseph's College, Tiruchirappalli, Tamil Nadu, and voucher specimens have been deposited

Sample Collection and Surface Sterilization

Healthy leaf and stem samples have been amassed during September 2022 from 3 floras in keeping with web page (triplicates). Samples have been washed under strolling faucet water and double-distilled water, followed through surface sterilization (95% ethanol – 10 s, 0.5% NaOCl – 2 min, 70% ethanol – 2 min). Samples have been rinsed thrice with sterile distilled water and air-dried aseptically. Sterilized tissues were cut into 0.5 cm stem segments and leaf halves.

Isolation of Endophytic Fungi

Segments had been inoculated onto Potato Dextrose Agar (PDA) supplemented with 20 mg/L chloramphenicol to suppress bacterial boom. Five tissue portions were positioned in keeping with plate and incubated at 37°C below a 12 h mild/darkish cycle for 30 days. Emerging fungal colonies had been purified through sub-culturing and maintained on PDA slants at 4°C and in 15% glycerol at –20°C.

Molecular Identification

Genomic DNA was extracted the usage of the NucleoSpin® Plant II Kit (Macherey-Nagel) following the producer's protocol. DNA quality turned into tested with the aid of 0.8% agarose gel electrophoresis. The ITS location turned into amplified the use of regular primers ITS1-F (five'-TCCGTAGGTGAACCTGCGG-three') and ITS4-R (five'-TCCTCCGCTTATTGATATGC-three'). PCR reactions had been finished in a complete extent of 10 µL below the following conditions: 98°C for 30 s, observed by means of 40 cycles of 98°C for 5 sec, 58°C for 10 sec, and 72°C for 15 sec, with a very last extension at 72°C for 1 min. Amplicons had been visualized on 1.2% agarose gels (Data Not Shown)

Sequencing and Data Analysis

PCR products have been purified the use of ExoSAP-IT (GE Healthcare) and sequenced the usage of BigDye Terminator v3.1 chemistry on an ABI 3500 DNA Analyzer. Sequences had been edited and in comparison with NCBI GenBank statistics the usage of BLASTn for taxonomic identification. Sequences were deposited in GenBank beneath accession numbers SJC01–SJC11.

Statistical and Diversity Analysis

Endophytic colonization frequency (CF %) was calculated as the wide variety of segments colonized divided via general segments X 100 (Hata & Futai, 1995). Relative isolation frequency (RF%) turned into computed as the percentage of isolates in line with host relative to total isolates (Su et al., 2010; Yuan et al., 2010) is indicated in Table 1

Results

General Overview

The study observed and aimed to evaluate the diversity as well as the distribution of endophytic fungi associated with *S. maritima* gathered from tannery effluent-infected soils in Dindigul District, Tamil Nadu. Leaf and stem tissues have been selected for isolation, as preceding reviews imply a better colonization frequency in leaves due to their larger floor place (Chareprasert et al., 2006; Mishra et al., 2012; Wu et al., 2019).

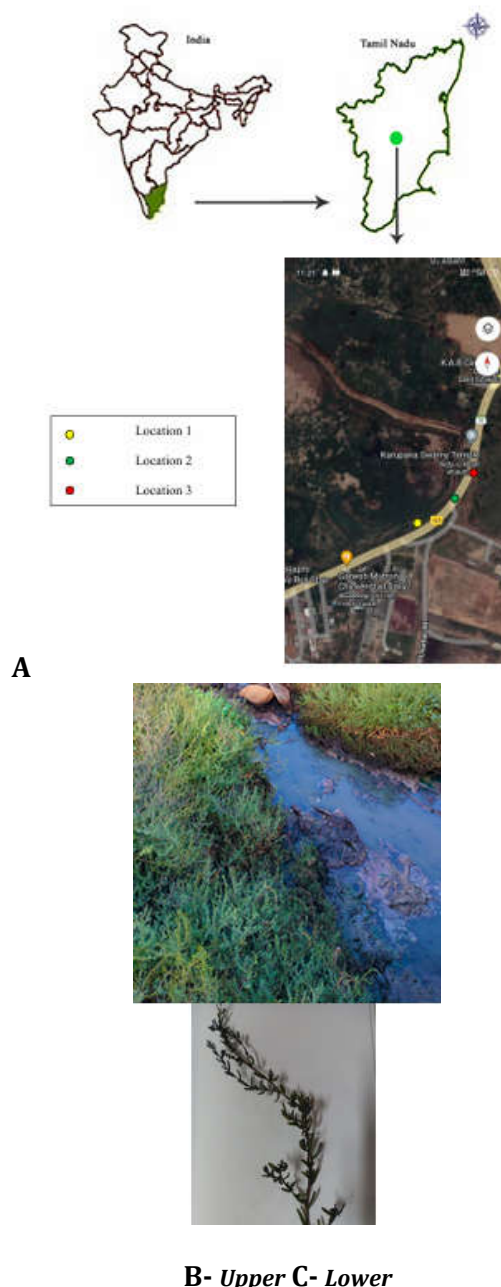


Figure-1 A Map showing the sampling sites of *S. maritima* in tannery effluent-contaminated areas of Dindigul District, Tamil Nadu, India. The upper panels depict the geographic location of Tamil Nadu within India, and the lower panel represents the specific sampling locations identified along the effluent-affected region. Location 1 (yellow) indicates the proximal point near the effluent discharge source, Location 2 (green) represents the intermediate zone, and Location 3 (red) denotes the distal site farther from the contamination origin.

Figure 1 B&C- *S. maritima*. growing in tannery effluent-contaminated soil of Dindigul District, Tamil Nadu, India. (B) Field view showing the effluent-polluted habitat with visible chromium-rich sludge and vegetation adapted to saline and metal-stressed conditions. (C) Collected specimen of *S. maritima* showing its characteristic succulent leaves and branching morphology used for endophytic fungal isolation

A total of 300 isolates were acquired from 150 plant tissue segments accumulated from 3 sampling locations. Based on cultural and microscopic traits, the isolates have been classified into 10 taxa. The most dominant species were *Irpex lacteus* (24%), *Trichoderma harzianum* (18%), *Alternaria tenuissima* (16%), and *Irpex sp.* (15%)

Molecular identity the usage of *ITS* rDNA sequencing changed into executed for morphologically unidentifiable isolates. The GenBank accession numbers and sequence similarities are listed. Out of 12 unidentified isolates, 3 exhibited >98% series similarity to acknowledged taxa, while the final isolates confirmed lower similarity values, suggesting the presence of potentially novel endophytes related to *S. maritima*

Colonization Frequency

The mean colonization extent become highest in Location 3 (81.1%), followed by using Location 1 (62.2%) and Location 2 (47.5%). Among all isolates, *I. lacteus* recorded the maximum colonization volume (16%), observed via *T. harzianum* (12%), *A. tenuissima* (11%), and *Irpex sp.* (10%).

Colonization became determined to vary extensively between tissues. Stem tissues exhibited higher colonization (86.66%) than leaves (40.74%), indicating tissue-particular variation in endophyte distribution.

Table 1. Colonisation extent (% CE) of two tissues of *S. maritima*. isolated from three tannery effluent-contaminated locations in Dindigul District, Tamil Nadu.

Plant part	Location 1	Location 2	Location 3	Total
Leaf	24.4	35.0	62.2	40.53
Stem	100.0	60.0	100.0	86.66
Total	62.2	47.5	81.1	63.59

Mean colonisation extent (% CE) of endophytic fungi in leaf and stem tissues of *S. maritima* collected from three sampling sites. Location 1 represents the proximal site near effluent discharge, Location 2 the intermediate zone, and Location 3 the distal site farther from contamination origin.

Isolation Extent and Taxonomic Composition

The isolation extent of endophytic fungi ranged from 0.006 to 0.106. The maximum isolation quantity become discovered in *I. lacteus* (0.106), accompanied by way of *T. harzianum* (0.08) and *A. tenuissima* (0.07). Fungal species belonging to *Basidiomycota* showed

better isolation volume than the ones of *Ascomycota*. However, *Ascomycetous* taxon, which includes *Trichoderma*, *Colletotrichum*, and *Xylaria*, were greater frequently recovered typical (Figure 2).

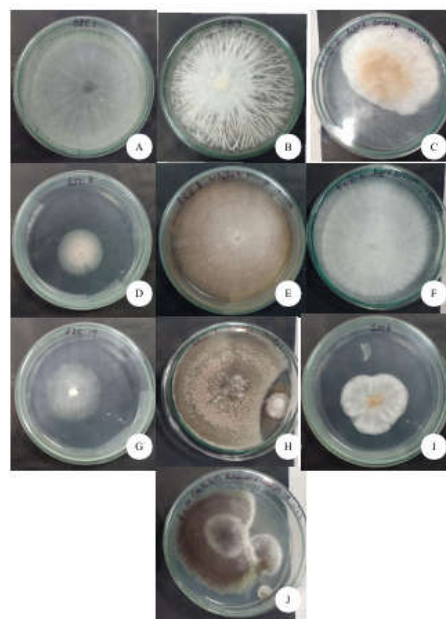


Figure 2- Colony morphology of endophytic fungi isolated from *S. maritima*. growing in tannery effluent-contaminated soils. (A) *Trichoderma harzianum*, (B) *Xylaria sp.*, (C) *Trichoderma effusum*, (D) *Colletotrichum karstii*, (E) *Colletotrichum sp.*, (F) *Phlebiopsis magnicystidiata*, (G) *Irpex sp.*, (H) *Alternaria tenuissima*, (I) *Gilbertiella sp.*, and (J) *Irpex lacteus*

Discussion

The findings of this look at demonstrate that *S. maritima* supports a various range of endophytic fungi even below the strain of tannery effluent infection. The imply colonization and isolation extents suggest slight to high fungal abundance, suggesting that the plant-endophyte affiliation may also confer adaptive benefits underneath steel-pressured environments.

The higher colonization located in stem tissues contrasts with previous studies that pronounced extra endophyte diversity in leaves (Arnold & Lutzoni, 2007; Mishra et al., 2012). This reversal can be attributed to higher heavy metallic accumulation in leaves, inhibiting endophyte status quo (Monnet et al., 2001; Sun et al., 2011). Environmental elements together with salinity, pH, and effluent attention are also acknowledged to persuade endophyte colonization (Naik et al., 2009; Eschen et al., 2010).

Molecular identity through *ITS* sequencing supplied taxonomic resolution for morphologically ambiguous isolates, corroborating earlier reports that emphasize the significance of molecular equipment for endophytic fungal studies (Guo et al., 2001; Wang et al., 2005; Ovaskainen et al., 2010). Notably, seventy five% of the isolates exhibited low collection similarity to recognised taxa, indicating the capacity lifestyles of novel endophytic fungi in *S. maritima* growing in saline, chromium-wealthy environments.

The coexistence of Ascomycota and Basidiomycota taxa indicates that both agencies make a contribution to host version beneath stress. Basidiomycetes including *Irpex lacteus* may additionally aid in lignocellulosic degradation and detoxification, even as Ascomycetes like *Trichoderma* and *Colletotrichum* are regarded for producing plant boom-selling and protective secondary metabolites (Gond et al., 2012; Kharwar et al., 2011).

These consequences spotlight the ecological significance of endophytic fungi in enhancing host tolerance and offer perception into their potential packages in phytoremediation of chromium-contaminated soils.

Conclusion

The study examination highlights the range of endophytic fungi inhabiting *S.maritima* growing in tannery effluent-infected soils of Dindigul District, Tamil Nadu. The isolation and molecular identity of fungal endophytes revealed genera predominantly tailored to saline and pollutant-rich environments, reflecting ecological resilience and host-microbe specificity. The mild range indices and location-clever variations suggest that effluent publicity affects endophytic network shape. These isolates, specifically *Aspergillus* and *Penicillium* species, may additionally serve as promising candidates for biotechnological exploitation in bioremediation and stress-resilient bioactive compound production. Further useful characterization of those endophytes may need to elucidate their position in plant model to infected habitats and their ability applications in environmental and industrial biotechnology.

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