



STUDIES ON ENDOPHYTIC FUNGI FROM MEDICINAL PLANTS OF MANGROVE ENVIRONMENT

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Abstract- Mangroves are the collection of plants in tropics and subtropics, which occur on the beachfront. The network of mangroves consists entirely of salt tolerant plants of the delicate and sumptuous mud, mostly trees and brooms, and large, weathered, evergreen leaves. Mangrove biological systems are supplement inadequate, particularly in nitrogen what's more, phosphorus. Mangrove Endophytic fungi are progressively perceived for their bioactive mixes' creation. Bioactive mixes detached have been found to have hostile to malignant growth, against diabetic and numerous different properties that were valuable in biomedical research and medication advancement.

Keywords: mangrove, Endophytic fungi, bioactive mixes, biomedical

I. INTRODUCTION

Mangroves are the collection of plants that take place in tropical and subtropical intertidal areas on the beachfront. The entire mangrove network consists of salt-tolerant plants of sensitive, swampy mud, most of them of trees and coves with broad, robust, perennial leaves. The supplementary missing ecosystems of the mangroves are phosphorus, particularly in nitrogen. Each ecosystem underpins human life by providing human administrations with immediate or roundabout benefits. One of the world's most profitable habitats is mangrove areas. As supervisors, they supply substantial biomass for their youth stock and structure. The Mangroves term refers to a collection of halophytic plant species, known as the ecosystem of salt-tolerant woodland, with a wide range of bio-financial products and administrations and a variety of other waterfront and marine habitats.

Mangrove species are classified as 'selective' and 'non-exclusive' species, which are essentially circulated in the earth or in the water, but which occur in addition in the mangrove setting, to mangrove conditions (as a mangrove, commit mangrove or genuine mangrove). Mangrove fungi form the microorganism's second largest natural collection. The data on mangrove-related fungi have increased considerably since they were first reported from mangrove establishments in Australia.

plants are generally colonized by a wide scope of Endophytic microorganisms, for example, microscopic organisms and fungi endophytes, a term initially presented by de Barry in 1866 (Carroll, 1986), to colonize inside plants tissues for in any event a piece of their life cycle without causing obvious illness side effects unmistakable all things considered. Albeit broad and assorted in both characteristic and rural ecosystems (Meyling and Eilenberg, 2007) and present nearly in all plant's organs (Stone et al., 2000), the job of endophytes in forming plants-condition collaborations, particularly plants-herbivorous creepy crawly collaborations, has not been satisfactorily valued.

Several studies have shown parasitary endophytes can be insured against herbivorous crawls (Azevedo et al., 2003), herbal diseases, or parasite nematodes (Arnold et al., 2003 and Hanlon et al., 2012). (Waweru et al., 2014). Endophysical contagenic species include the all-round Clavicipitaceous endophytes which, with regard to the vertical transmission of endophytic growth by seeds, form a private relationship with their host plants and produce explicitly harmful alkaloids to the bug class I Endophytic fungal products (Rodriguez et al., 2009).

The less specific endophytes which colonize either above or below ground host plant tissues are considerably lower than ground level. Most entomopathogens have two divisions in sphere - Zygomycota and Ascomycota - so far, entomopathogens have been regarded as endophytes in the application Hypocreales only in the latter division (Roy et al., 2006).

According to Rodriguez et al. (2009), Class II endophytes of entomopathogenic Endophytes should be delegated, since both above and underlying colonies of their individual Host Plants have been found to colonies (Tefera and Vidal, 2009 and Quesada-Moraga et al., 2014).

II. LITERATURE REVIEW

The worldwide decent variety of fungi has been talked about effectively for as long as 20 years, with uncommon regard for extrapolative appraisals of species extravagance dependent on have and geographic particularity (Hawksworth, 1991). Host generalize and an absence of discernible living space selectivity due to edaphic or timberland qualities seem to propose that high appraisals of contagious lavishness be treated with alert. In any case, albeit vague with respect to has, backwoods age, and soil conditions, the rich network of endophytes seen in these tropical grasses proposes that even host-and natural surroundings generalists can show neighborhood spatial structure that will contribute considerably to biodiversity at territorial viewpoints.

In different taxes, various fungi have been identified as endophytes. Caruso et al. (2000) disconnected from the *Taxus baccata*, distinguishing 25 unique genera. Five generations and three unknown fungi were found by Wang et al. (2008) in *T. mairei*. 26 genera in *T. chinensis*, detailed in 2009. Liu et al. In '*T. globosa*,' the number of genera found (26) was similar to *T. baccata* and *T. chinensis*, but higher than '*T. mairei*.' It should be noted that the genera of *Alternaria*, *Aspergillus*, *Colletotrichum* and '*Xylary*' were more than normal for all four species of *Taxus*.

The Endophytic fungi were recognized to 43 taxa, of which *Cephalosporium*, *Sirococcus*, *Penicillium* and *Aspergillus* were the prevailing genera. *Cephalosporium*, *Penicillium* and *Aspergillus* were the most widely recognized endophytes and have been announced as the prevailing contagious endophytes of different plants species in various situations (Bacon and Palencia, 2010; Vega et al., 2010).

The colonization pace of Endophytic fungi was fundamentally higher in the stems than in the leaves. A similar wonder was identified in *Acer truncatum*, *Saussurea involucre*, *Dracaena Cambodian* and *Aquilaria sinensis* (Lv et al., 2010 and Sun et al., 2011). This might be because of the way that the stems are steady, though the leaves are deciduous. The other conceivable explanation might be that the temperature at the testing locales changes extraordinarily with occasional and day-night varieties, which results in more noteworthy variances in the leaf temperature than in the stem temperature.

Endophytes with sheet foliage undergo various unique strain, undeniable from those facing xyl endophytes or tissue-related endophytes, such as inner bark and roots (Su et al., 2010).

III. MATERIALS AND METHODS OF THE STUDY

Endophytic fungi: In particular, *Avicennia marina*, *Suaeda monica*, and *Rhizophora mucronate* were collected from the mangrove plant species. The collected examples were purposely placed into polythene packs and used... The parasite was separated as 7, 8, 9 and 10. Cultivation in potato dextrose agar has not been adulterated.

Cultivation and sampling:

The test life was made in 2-liter containers of Erlenmeyer with 500 ml of PDB. The animal was inoculated and developed 21 days. After incubating the filtrate in lifestyle, four layers of cheese are separated and isolated from mycelia. At the time, three similar amounts of dissolvable ethanol were used for filtrate lifestyle. The normal phase was accumulated, and the dissolvable was then extracted with a pivoting vacuum evaporator by dissemination under reduced pressure at 45°C. The dry solid development was replenished in ethanol and the hard concentrates for their disease prevention properties were investigated.

Antioxidant Assays:

ABTS Radical Scavenging Activity:

Both action packages included ABTS 7.4 mM and persulphate 2.6 mM of potassium as shown by Arnao, Cano and Asota 11. The working course was mastered by combining the two inventory plans in the same amounts and enabling them to respond at dull room temperature for 12 hours. Mixing in with 1 ml ABTS game plan with 50 ml of methanol, to achieve 1.1 ± 0.02 units at 734 nm of absorbance has weakened the game design. Tests (1.5 ml) were combined with 2.850 ml of ABTS action and the blend remained for a 2-hour reduction at room temperature. At 734 nm the absorption was then assessed. With this state, the ability to look through ABTS radical has been resolved:

ABTS searching impact (%) = $[(A_0 - A_1 / A_0) \times 100]$

Where, A₀ was the absorption of the control reaction and A₁ the absorption in view. The half hindrance focusses fixation (EC₅₀) was calculated via the insertion from the direct relapse study.

DPPH Radical Scavenging Activity: The free radical concentrate quest was calculated with 1, 1-diphenyl-2-picryl-hydrazyl (DPPH). Separate centralization (0,1–20 mg/ml) was quickly mixed into 0,2 mm radical diphenyl-2, picrylhydrazyl (DPPH, Sigma) methane arrangement 1 ml with 0,2 mm of diphenyl radicals. A blend of 0,2 mm was enthusiastically shaken and left to reflect 30 minutes of obscurity, and absorbance determined to be 517 nm with a transparent 12. EC₅₀ esteem (mg/ml) is the convincing fixation in which radicals in DPPH have been investigated halfway through and the value has been obtained through a straight reciprocity analysis. For analysis, A-tocopherol was used. Using the accompanying state, the ability to rub DPPH radical was calculated:

DPPH rummaging impact (%) = $[(A_0 - A_1 / A_0) \times 100]$,

Where A₀ was the absorption of the control response and A₁ the absorbance of the example in view. Interjections from the straight reciprocal investigation obtained the focus attachment giving half-limit (EC₅₀).

Assurance of Antioxidant Component:

Complete Phenol:

The Taga, Miller and Pratt using the Folin-Ciocalteu's technique have been resolved to complete the phenolic mixes. 100 mg ethanol separate was used with 5 ml of 0.3 percent HCl in methanol/deionized water (60:40, v/v). Added 2 ml of a 2 percent sodium carbonate fluid from the subsequent blend (100 µl). The mixture was sprung 2 minutes. The absorbance was measured at 750 nm against clear to 100 µl of the half Folin-reagent Ciocalteu's and was brooded for 30 mins. The substance absolute phenol was measured using the bending of gallic corrosive, and the results were recorded as mg per g of gallic corrosive counterparts (GAEs).

Flavonoid:

The concentrate of parasites (250µl) were mixed with a refined water content of 1.25% and a NaNO₂ setting of 5%, 75µl. The AlCl₃.H₂O (10 percent, 150µl) schedule was included after 5 minutes. After 6 minutes the blend has been supplied with NaOH (1M, 500µl) and refined water (275µl). The action course was well mixed and the strength of the rose cover was assessed at 510 nm. The material of the flavonoid was determined depending on a quercetin alteration curve and the findings were transmitted as mg per g concentrate of quercetin.

FINDINGS AND RESULTS

Radical Scavenging Activity against ABTS: ABTS was used to investigate the radical rummaging effect of concentrates using the stable free radical with preservation of the label at 734 nm. The results showed that ABTS reacted with different ABTS focuses, ranging from 100, 200, 400, 800 and 1600µg/ml separately and readings were estimated to be reduced by ABTS to 734 nm by a radical cation decrease. At the highest centralization in Ec₅₀ estimates of 1600 µg/ml, the ethic concentrates of PHOMOPSIS amygdali showed 72.38 percent extremely high decolorization (Table 1). The wider decolorization decrease is legitimately related to the widening concentration centralization in Figure 1.

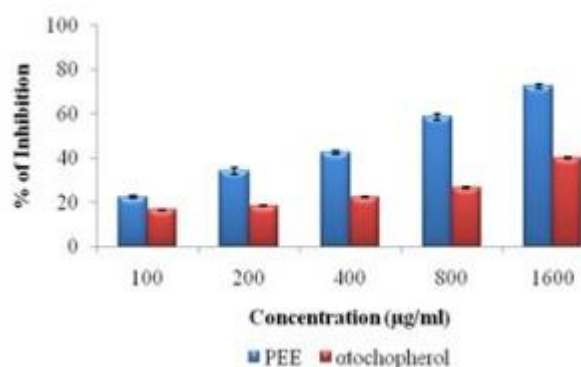


FIGURE 1: FREE RADICAL PHOMOPSIS SP ETHANOLIC EXTRACT (PEE) STILL RADIC SCAVENGE EFFECT (PEE)

ABTS assay is an amazing tool to determine the action of plant chemicals in the prevention of cancer. The cancer prevention agents' properties, removed from palatable basidiomycetes tested against the radical ABTS, have replied that the radicals will rummage.

Radical Scavenging Activity utilizing DPPH: To take into consideration radical rummaging impacts of concentrates, DPPH, a stable free radical with mark-ingestion in 570 nm, was used. As a proton for this radical cancer prevention agent, assimilation decreases. In the case of several fixations of (100 to 6400µg), the example has been tested against this radical and readings have been seen by decreasing the absorption as a demonstration of the level of radical rummaging property.

Together with the Regular α -Tocopherol the stunning effects of this example were evaluated. The DPPH-radical contagious concentrate was most extremely declined by 2.17% at the largest group of 6400 µg/ml and the EC50 was 2030.25%±0.81 µg/ml toward DPPH-radicals (Figure 2, Table 1). Phomopsis amygdali was presented with ethanol concentrates higher than the normal α -tocopherol which was in line with the previous review done by Duan et al,

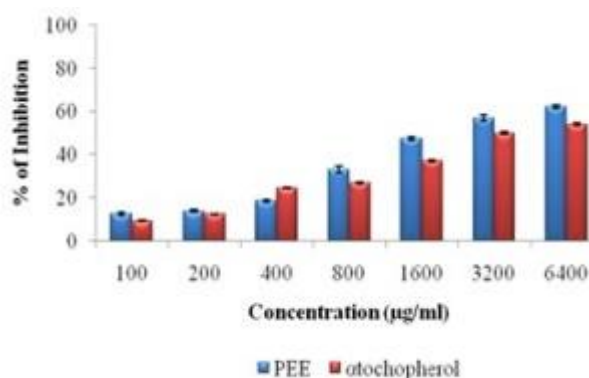


FIGURE 2: FREE RADICAL PHOMOPSIS SP SCAVENGING. AGAINST DPPH Radical ETHANOLITY (PEE)
TABLE 1: OBTAINED BIOACTIVE COMPOUNDS from PHOMOPSIS SP. EXTRACTED CULTURE FILTRATE WITH ETHANOL.

Sample	ABTS	DPPH	Phenol (mg/g)	Flavonoid (mg/g)
<i>Phomopsis sp.</i>	580.02±0.57	2030.25± 0.81	18.33	6.44

Assurance of Antioxidant Compounds: Phenolic and flavonoid compounds tend to have an important task in balancing lipid oxidation. The action relating to cancer prevention agents is presented in Table 1. All phenol outputs were 6.44±1.24µg/mg quercetin proportional, with a dry weight and flavonoid content of .3±0.68mg GAE/g. The results showed that *Phomopsis amygdali*'s ethanol concentrate contains large phenols and flavonoids. Liu et al. recorded absolute phenol content in the 54.51 mg/g range and 86.76 mg/g flavonoid content in the *Phomopsis amygdali* concentrate. In comparison with the territory of the cellular strengthening material, the current analysis showed the diversified extraction method, that the endophytic production of phenolic and flavonoid radiation can be seen to be a source of popular cancer prevention agents and show an incredible movement of anti-ABTS and DPPH radicals. In addition, it should also analyses the attributes of plant chemicals that have a property of cancer prevention agents.

REFERENCES

1. Aamir, S., Sutar, S., Singh, S.K. and Baghela, A., (2015). A rapid and efficient method of fungal genomic DNA extraction, suitable for PCR based molecular methods. *plantsPathol and Quarantine*; 5(2): 74–81.

2. Abbas Ahmed, Dahab, M.M., Taher Taha, N.A. and Fareed Hassan, S.M., (2015). Production, Purification and Characterization of L-Asparaginase from Marine Endophytic *Aspergillus* sp. ALAA-2000 under Submerged and Solid-State Fermentation. *Journal of Microbe. Biochem. Technol*; 7: 3-18.
3. Abe, K., (2006). Impact of *Aspergillus Oryzae* Genomics on Industrial Production of Metabolites. *Mycopathologia*; 162(3): 143-153.
4. Adebayo, E.A. and Ishola, O.R., (2009). Phytochemical and antimicrobial screening of crude extracts of *Terminalia glaucescens*. *Afr. J. Pharm. Pharmacol*; 3(5): 217-221.
5. Adeline, S.Y., Ting, Y.C., (2015). Endophytic L-asparaginase-producing fungi from plants associated with anticancer properties *Journal of Advanced Research*; 6: 869–876.
6. Ahmed, M., Khaleeq, A and Saghir, A., (2014). Antioxidant and Antifungal Activity of Aqueous and Organic Extracts of Liquorice. *World Applied Sciences Journal*; 30(11): 1664-1667.
7. Bruns, T.D., White, T.J. and Talyor, J.W., (1991). Fungal molecular systematic. *Annual Review of Ecology and Systematics*; 22: 525-564.
8. Bryan, G.T., Daniels, M.J. and Osbourn, A.E., (1995). Comparison of fungi within the *Gaeumannomyces-Phialophora* complex by analysis of ribosomal DNA sequence. *Appl. Environ. Microbiol*; 61: 681-689.
9. Carroll, G.C., (1986). The biology of endophytism in plants with particular reference to woody perennials. In *Microbiology of the Phyllo sphere* (eds Fokkema, N. J. and van den Huevel, J.), Cambridge University Press, Cambridge; 205–222.
10. Caruso, M., Colombo, A.L., Fedeli, L., Pavesi, A., Quaroni, S., Saracchi, M., (2000). Isolation of Endophytic fungi and actinomycetes. *Ann. Microbiol*; 50(1): 3–13.
11. Chaverri, P., Samuels, G.J. and Stewart, E.L., (2000). Convergent evolution of *Gliocladium* morphology in *Hypocrea*. *Abstract. Inoculum. Newsletter of the Mycological Society of America. Mycologia*; 51: 24.