

**NAPHTHOQUINONES PRODUCED BY THE FUNGUS
MONACROSPORIUM AMBROSIUM FROM TEA (*CAMELLIA
SINENSIS*) IN CULTURE MEDIA; BIOLOGICAL ACTIVITY,
EFFECT ON CAFFEINE AND FUNGAL GROWTH IN THE
PRESENCE OF TRACE METAL IONS.**

C.L. Kehelpannala

Postgraduate Institute of Science, University of Peradeniya, Peradeniya, Sri Lanka
National Institute of Fundamental Studies, Kandy, Sri Lanka

Monacrosporium ambrosium, ectosymbiote of shot hole borer beetle (SHB), *Xyleborus fornicatus* was isolated from SHB beetle on potato dextrose agar (PDA). A study was carried out to investigate the effect of liquid cultures of *M. ambrosium* on caffeine and the biological properties of the ethyl acetate (EtOAc) extracts of the culture broth. The effect of selected metallic ions on fungal growth was also studied.

Caffeine, incorporated into cultures of *M. ambrosium* in glucose-yeast extract-peptone medium, did not cause visible changes in the fungal culture. High performance liquid chromatography of EtOAc extracts of the culture broth with caffeine, revealed that caffeine was unchanged, and that theophylline and theobromine, are not produced by a biotransformation of caffeine.

Cu^{2+} , Cd^{2+} and $\text{Hg}^+/\text{Hg}^{2+}$ (0.25 mM and 0.5 mM) were incorporated into potato dextrose broth (PDB) with *M. ambrosium*. The studies showed that of the three ions tested Cu^{2+} is the least toxic for fungal growth while $\text{Hg}^+/\text{Hg}^{2+}$ is the most toxic, with fungal growth completely inhibited at 0.5 mM concentration. Preliminary thin layer chromatography (TLC) studies indicated that additional metabolites were not produced by the fungus in the presence of these cations.

The EtOAc extract of liquid cultures of *M. ambrosium* in PDB was found to possess antifungal, phytotoxic activities, and was positive in the brine shrimp lethality assay. Activity guided fractionation of the EtOAc extract of *M. ambrosium* culture broth yielded six compounds, identified as anhydrojavanicin, dihydroanhydrojavanicin, 7-acetyl-5,8-dihydroxy-6-methyl-1,4-naphthoquinone, javanicin, anhydrofusarubin and solaniol by analysis of ^1H and ^{13}C NMR spectroscopic data, FAB-MS and comparison with literature data.

The antifungal activity of the EtOAc extract of *M. ambrosium* containing naphthoquinones was seen by its effect on the endophytic fungi *Pestalotiopsis camelliae* and *Phoma multirostrata* isolated from TRI 2023 tea stems. The EtOAc extract of *M. ambrosium* cultures (500 ppm and 1000 ppm) completely inhibited growth of the endophytic fungi *P. camelliae* at 1000 ppm, while growth of *P. multirostrata* was inhibited (40%) at 500 - 1000 ppm. The antifungal activity of naphthoquinones secreted by *M. ambrosium* did not affect the growth endophytic fungi, *Bipolaris sorokinana*, *Daldinia eschscholtzii* and *Glomerella magna* from other plant sources, when grown together with *M. ambrosium*, on PDA plates.