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Title : TOXIC VOLATILES AS TOOLS FOR THE SELECTIVE ISOLATION OF SOIL FUNGI AND MODULATING THEIR METABOLISM

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The fungal kingdom forms an important source for a broad range of secondary metabolites with widely different chemical structures, as well as diverse biological activities. In the present work, the main objective was to isolate fungi from local soil samples and identify bioactive metabolites that they could produce. To this end, soil samples were collected from a biological forest reserve, at UiTM's Puncak Alam Campus, Malaysia. The isolation from soils can proceed through many different techniques. Here, a new chemotechnique based on the use of five volatile compounds (dimethylsulphoxide, naphthalene, cineole, petroleum and formaldehyde) was used. As a result, a total of 82 fungi were isolated from soil samples taken at three different depths. The pure cultures were inoculated and fermented in the presence or in the absence of volatile compounds. A total of 100 crude extracts were analysed by HPLC and evaluated for antimicrobial activity against pathogenic microorganisms using the MTT assay. Twenty out of 100 crude extracts showed significant antibacterial activity against *Escherichia coli*, 17 extracts were active against *Enterococcus faecium*, 15 against *Pseudomonas aeruginosa* and 25 against *Staphylococcus aureus*. Twenty six crude extracts showed antifungal activity against *Candida albicans*. From the analysis of the above data, 8 out of the 82 fungal isolates were selected for further study. These include *Aspergillus nomius, A. terreus, Byssochlamys nivea, Talaromyces aculeatus, P. commune, Pseudallescheria minutispora, Trichoderma citrinoviride*, and *T. virens*, which were fully identified by morphological and genetic techniques. Their metabolites were purified by semi-preparative HPLC and identified by spectroscopic (MS, NMR, UV/Vis) and X-ray diffraction techniques. From Aspergillus nomius, four compounds were identified, namely kojic acid, aflatoxins B1 and G1, and 3-O-methylsterigmatocystin. Terrein was isolated from *A. terreus*. From the *Penicillium* and *Talaromyces* species, three compounds were identified, including penicillic acid, 6-methoxymellein and vermistatin. From *Pseudallescheria minutispora*, three rare pseurotins A, A1 and A2 were identified. From *Trichoderma citrinoviride*, hydroxyheptelidic acid and gliocladic acid were identified. Rare MK-8383 was obtained from *Byssochlamys nivea.* Finally, from *Trichoderma virens*, viridiol was identified, while the plane structure was established for a new metabolite named trichovirenic acid. Scanning electron microscopy (SEM) revealed marked effects when the *Talaromyces aculeatus* isolate was exposed to DMSO: (i) hyphae showing hairy spinulose appendages or nodules, (ii) hair-like appendages arising from penicillus phialides and long dense spines on the conidia, (iii) hyphae with wart-like growths, (iv) smooth conidia, (v) presence of mycelial coils, (vi) vesicle-like formations. Other less dramatic effects such as altered growth rate and discoloration were observed with some volatiles for some isolates. Significant changes in secondary metabolic profiles were observed with volatile exposure. Thus, the use of toxic volatile compounds presents a novel selective isolation procedure and allows for metabolic manipulation of fungi.