



## Identification of Griseofulvin and some derivates by LC-SPE-NMR from new species of saprophyte fungus.

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### Abstract

The Brazilian economy is based on agriculture's sector, mainly on citriculture. Our country is responsible for to the most production of orange and orange juice in the world. However, with this production emerges diseases, like Greening, Citrus Variegated Chlorosis, Gummosis, that attack the citrus and causes significant financial losses. The control of this pests and diseases is traditionally accomplished through the use of pesticides and fungicides [1] but the use indiscriminate are harmful to human health and the environment. In this context, the researchers have employed the saprophyte into the alternative control. One in special, founded in Brazil semi-arid soil, *Memnoniella levispora* (MEL), in preliminary study was shown potent promissory against the fungus that cause the Gummosis disease. Therefore, the aim of this study was to establish a fast chemical profile by LC-SPE-NMR to identify the metabolites responsible to this activity. The MEL fungi were grown in carrot-corn liquid culture media and kept in a growth chamber at 25°C, for 21 days. Before this, vacuum filtration and liquid-liquid separation with ethyl acetate was done. The chemical profile of the ethyl acetate extract from MEL was analyzed and optimized in LC-DAD-SPE-NMR (Agilent 1200). The chromatographic conditions at 254nm, using water (A) and acetonitrile (B) like solvents' elution are shown below in the table 1.

Table 1: Chromatographic conditions to LC-DAD-SPE-NMR analyses

TIME	0	5	40	41	51	52	57	61
PERCENTAGEM	80	80	32	16	0	0	80	80
A (%)								
B(%)	20	20	68	84	100	100	20	20

The LC/UV trace revealed 10 major peaks. All the peaks were trapped onto an SPE cartridge and after the drying step, it was eluted into the NMR flow probe and the <sup>1</sup>H NMR spectrum was acquired. It was possible identified compounds like griseofulvin (peak 2), dechlorogriseofulvin (peak 3), dehydrogriseofulvin (peak 4) and dechlorodehydrogriseofulvin (peak 5). Others compounds were trapped and acquired dates of NMR but not identified yet. Therefore, hyphenation of LC-SPE-NMR-MS allows us identified, 4 interesting compounds, in low concentration in a new species of fungus, especially griseofulvin, the first antifungal isolated, common used in treatments of fungi human disease, first related in literature for this species, and others isolated compounds with activity and structure are still unknown.

### References

- [1] P.F. de Lima, M.F. Furlan, F.A. de Lima Ribeiro, S.F. Pascholati, F. Augusto, In vivo determination of the volatile metabolites of saprotroph fungi by comprehensive two-dimensional gas chromatography, J. Sep. Sci. 38 (2015) 1924–1932.