BIODEGRADATION OF THE FUNGICIDE METALAXYL BY ZYGOMYCETES

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A large number of fungicides, such as metalaxyl [methyl N-(methoxyacetyl)-N-(2,6-xylyl)-DL-alaninate], are used against Oomycete species that cause downy mildew in agricultural crops, including grapes. However, these compounds are potentially harmful for terrestrial and aquatic environments, and to human health due to carcinogenic and mutagenic properties. There is an increase in interest in using filamentous fungi, such as members of the Zygomycetes which degrade xenobiotic compounds using nonspecific extracellular enzymes for the bioremediation of pesticides in polluted soils.

In this study, several *Zygomycetes* including *Gongronella, Absidia, Circinella* and *Rhizopus* species, were used to screen degradation of metalaxyl using solid and liquid cultures. In order to identify and characterise these taxa a polyphasic approach including morphology characterization, molecular fingerprint M13-PCR and Matrix Assisted Laser Desorption Ionization Time of Flight Intact Cell Mass Spectrometry (MALDI-TOF ICMS) was used. After selective enrichment on solid medium containing a fungicide gradient concentration of 0-100 mg.L-1, *Absidia glauca* (CBS 101.08) and *Rhizopus orizae* (CCMI 900) showed a capacity to tolerate high metalaxyl concentrations. These resistant strains were selected to perform liquid assays using Yeast Nitrogen liquid cultures supplemented with sucrose (5 g.L-1) and metalaxyl (100 mg.L-1). Biomass concentration was determined by dry weight. The *A. glauca* and *R. orizae* strains showed specific growth rates of 0.774 h-1 and 0.999 h-1, respectively. Sucrose was completely consumed within 5 d with a sucrose consumption rate of 0.93 and 0.84 g.L-1.day-1 for *A. glauca* and *R. orizae,* respectively. The non-degraded metalaxyl in liquid cultures was determined by UV-HPLC and evaluated periodically for 21 d. The metalaxyl degradation rate for *A. glauca* and *R. orizae* was 2.22 mg.L-1 .day-1 and 2.29 mg.L-1day-1, respectively.

Results suggest that *A. glauca* (CBS 101.08) and *R. orizae* (CCMI 900) can be used in soil bioremediation experimentation for metalaxyl degradation. These strains are now under study to determine the presence of extracellular enzymes involved in the process.

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