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# YEAST VOLATILE ORGANIC COMPOUNDS INHIBIT OCHRATOXIN BIOSYNTHESIS BY ASPERGILLUS CARBONARIUS AND A OCHRACEUS

Zahoor Ul Hassan*, Maria Farbo, Pietro Urgeghe, Angela Marcello, Samir Jaoua, Quirico Migheli

Department of Biological & Environmental SciCollege of Arts and Sciences, Qatar University,P.O. Box 2713, Doha - Qatar
* zahoor@qu.edu.qa

Ochratoxin A (OTA) has hepatotoxic, teratogenic, nephrotoxic and carcinogenic effect in mammals and it is classified as a group 2B carcinogen by the World Health Organization. The European Union has set the maximum OTA level at 2 mg/kg in wine, grape juice, and other grape products, and at 3 mg/kg for all products derived from cereal, including cereal products and cereal grains for human consumption. Some species of Aspergillus are the main source of OTA in warm and tropical regions, and in particular Aspergillus carbonarius (Bainier) Thom is considered one of the most relevant OTA producers in food and feed. Inhibiting the growth of OTA-producing fungi on sensitive commodities is by far the most reliable method to prevent OTA contamination of food and feed. Aim of this study was to evaluate the biocontrol ability of selected yeast strains against OTA producing Aspergillus carbonarius and Aspergillus ochraceus. In a previous report, two non-fermenting (Cyberlindnera jadinii 273 and Candida friedrichii 778) and two low-fermenting (Candida intermedia 235 and Lachancea thermotolerans 751) yeast strains have shown a significant antagonistic behaviour against a virulent strain of A. carbonarius on grape berries as well as in in vitro experiments, while the filtrated and autoclaved culture broth of the yeast strains had no significant effect on pathogen growth.This biological effect was at least partly due to the release of volatile organic compounds (VOCs), since growth inhibition was observed without contact between yeast and Aspergillus spp.. Aspergillus colonies exposed to yeast VOCs did not sporulate, and were characterized



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by a white mycelium; the colony border was undefined, with elongated and scattered hyphae compared to unexposed control. Single hyphal tips and mycelium fragments were then transferred on PDA and after 5 days of growth at 25°C, typical dark sporulating colonies were evident, suggesting that the anti-sporulating effect is reversible. Aiming to further characterise the effect of VOCs produced by biocontrol yeast strains, we observed that, beside vegetative growth and sporulation, the volatile compounds significantly reduced the production of OTA by both A. carbonarius and A. ochraceus isolates. Exposure to yeast VOCs also affected gene expression in A. carbonarius, as confirmed by downregulation of polyketide synthase, non-ribosomal peptide synthase, and the regulatory genes laeA and veA. The main compound of yeast VOCs was 2-phenylethanol, as detected by Headspace-Solid Phase Microextraction-Gas Chromatography-Tandem Mass Spectrometry (HS-SPME-GC-MS) analysis. Yeast VOCs represent a promising tool for the containment of growth and development of mycotoxigenic fungi, and a valuable aid to guarantee food safety and quality. Further studies will aim at testing single purified VOCs in order to identify the most effective compounds responsible for the inhibition of fungal growth and OTA production by Aspergillus spp. in preventive food safety strategies.