

# Copper and zinc stress in aquatic fungi: the role of cellular antioxidant defences

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Aquatic hyphomycete species play an important role in biogeochemical cycles, being the main decomposer fungi of plant detritus in streams. Pollution by heavy metals in freshwaters is currently a worldwide problem, especially in industrial areas. Above threshold concentrations, metal ions are known to promote reactive oxygen species (ROS) formation that may overwhelm the cellular antioxidant defences. Our research aimed at elucidating the putative role of cellular antioxidant defences in stress induced by copper and zinc in aquatic hyphomycete species.

Two species were used, *Heliscus submersus* UMB 135.01 and *Varicosporium elodeae* UMB 142.01, isolated, respectively, from a polluted and a non-polluted stream in the Northwest of Portugal. Both species were grown 8 days in 1% malt extract either with or without adding copper and zinc at concentrations that inhibited growth by 50% and 80% (pH 5.0, 18 °C and 160 rpm). In addition, the fungi grown in non-amended medium were also exposed to copper or zinc for periods of 30 min and 14 h.

Cellular oxidative stress induced by copper and zinc was evaluated by epifluorescence microscopy with MitoTracker CMH<sub>2</sub>-XROS, a specific dye for intracellular ROS content. Our results point to different levels of ROS production triggered by the two heavy metals tested. Specifically, copper induced higher levels of ROS than zinc in *V. elodeae*. We also assessed the putative role of catalase, superoxide dismutase and glucose-6-phosphate dehydrogenase in the cellular anti-oxidative defences of these fungi against copper and zinc in both adapted and non-adapted mycelium. Results will be presented showing the influence of these enzymes in the global cellular anti-oxidant defences to heavy metals.

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