

## Influence of copper on cell viability and reactive oxygen species (ROS) production in the marine microalgae *Dunaliella tertiolecta* and *Pavlova gyrans*

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There has been increasing concern over heavy metal pollution in the coastal areas due to human activities and the magnification of such metals within the aquatic food chain. Copper (Cu) is one of the most widely detected contaminants in the marine ecosystems. As microalgae are the primary producers and are sensitive to environmental changes, they can be used as biological tools to assess the toxicity of heavy metals in the marine ecosystems. The aim of this study was to assess the toxicity of Cu in the marine microalgae *Dunaliella tertiolecta* and *Pavlova gyrans*. The influence of Cu on growth, cell viability and reactive oxygen species (ROS) production was investigated. The 96h toxicity test was conducted using Cu concentrations ranging from 1.28, 2.56, 5.12, 10.24 to 20.48 mg/L based on absorbance at 620 nm (OD<sub>620</sub>). Cell viability was assessed after 0, 1, 2, 4, 6, and 24 h respectively by determining esterase enzyme activities in the cells stained with fluorescein diacetate (FDA) followed by flow cytometry analysis. The ROS production was determined using cell permeable 2', 7'-dichlorodihydrofluorescein diacetate (H<sub>2</sub>DCFDA) and flow cytometry. Based on the 96 h toxicity testing, *P. gyrans* (EC<sub>50</sub> = 3.986 mg/L) was more sensitive to Cu than *D. tertiolecta* (EC<sub>50</sub> = 8.422 mg/L). In the cell viability test, the cultures were exposed to Cu concentrations at EC<sub>50</sub> and 20.48 mg/L. The cell viability of *D. tertiolecta* decreased by 13.6 % and 51.6 % after exposure to 20.48 mg/L Cu for 4 and 6 h respectively, compared to the control. *Pavlova gyrans* showed a mark decrease in cell viability (87%) after exposure to 20.48 mg/L Cu for 2 h. The cell viabilities of the control and EC<sub>50</sub> were very similar, ranging from 84 - 90% within the duration of the test (0-24 h). The ROS production increased in both species when treated with Cu. This study showed that cell viability and ROS production could serve as additional endpoints to assess Cu toxicity in the two microalgae species.