

P22: Ecotoxicity of Congo red, hematoxylin, safranine and neutral red dyes in green algae and cyanobacteria

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Introduction: Dyes from different chemical categories are frequent in effluents from textile production, food processing and paper mills. Dyes are also very frequently used in biology, biochemistry, histology and microbiology laboratory studies. Congo red ($C_{32}H_{22}N_6Na_2O_6S_2$) is a water-soluble secondary diazo dye. Hematoxylin ($C_{16}H_{14}O_6$) is a natural dye, extracted from the heartwood of the tree *Haematoxylum campechianum*. Safranine ($C_{20}H_{19}ClN_4$) is used as a biological stain commonly used as a counterstain in the Gram stain procedure and in histological studies. Neutral red ($C_{15}H_{17}ClN_4$), is a vital stain used in many staining methods.

Objectives: The objective of this work was to describe the effects of Congo red, hematoxylin, safranine and neutral red dyes in *Chlorella vulgaris* and *Microcystis aeruginosa* growth rate.

Materials and Methods: Toxicity tests followed the European Guideline for algal and cyanobacteria growth inhibition test. Cells from algae and cyanobacteria cultures in exponential growth phase, were incubated in a microplate, with five dye concentrations (0; 40; 4; 0.4; 0.04 and 0.004 $\mu\text{L/L}$), under continuous light and shaking at 22°C. Average growth rates were calculated from cell numbers based on OD determination at 24h period until 96h.

Results and Discussion: After 96h exposure and at the higher concentration tested the four dyes inhibited both *Microcystis* and *Chlorella* growth rate but with different patterns and intensities. Differences of sensitivity were found between the two species with different LC50 values, depending on the dye. In some cases, and at the lowest concentration, both species growth showed hormesis. The time related response was different between species.

Conclusion: The dyes studied have different negative effects in *Microcystis* and *Chlorella* growth rate. Interspecific differences of sensitivity were found.

References

1. OECD (2011), *Test No. 201: Freshwater Alga and Cyanobacteria, Growth Inhibition Test*, OECD Guidelines for the Testing of Chemicals, Section 2, OECD Publishing, Paris. DOI: <http://dx.doi.org/10.1787/9789264069923-en>