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Growth and Metabolic Responses of *Microcystis aeruginosa* to Titanium Dioxide Nanoparticles

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INTRODUCTION

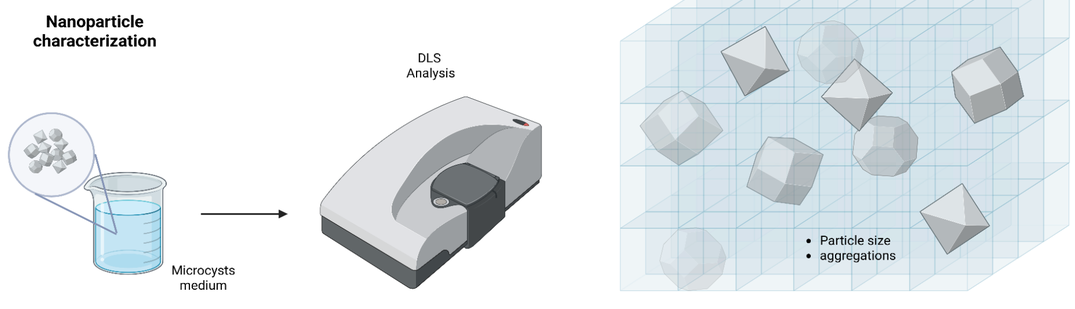
Microcystis aeruginosa is a freshwater cyanobacterium capable of forming massive blooms and producing microcystins, hepatotoxins with adverse effects on aquatic organisms and human health. Among the emerging contaminants in aquatic environments, titanium dioxide (TiO_2) in nanoparticle (NP) form has garnered increasing attention due to its widespread industrial application and growing presence in natural water bodies. Its impact on the physiology and secondary metabolism of photosynthetic microorganisms remains insufficiently understood.

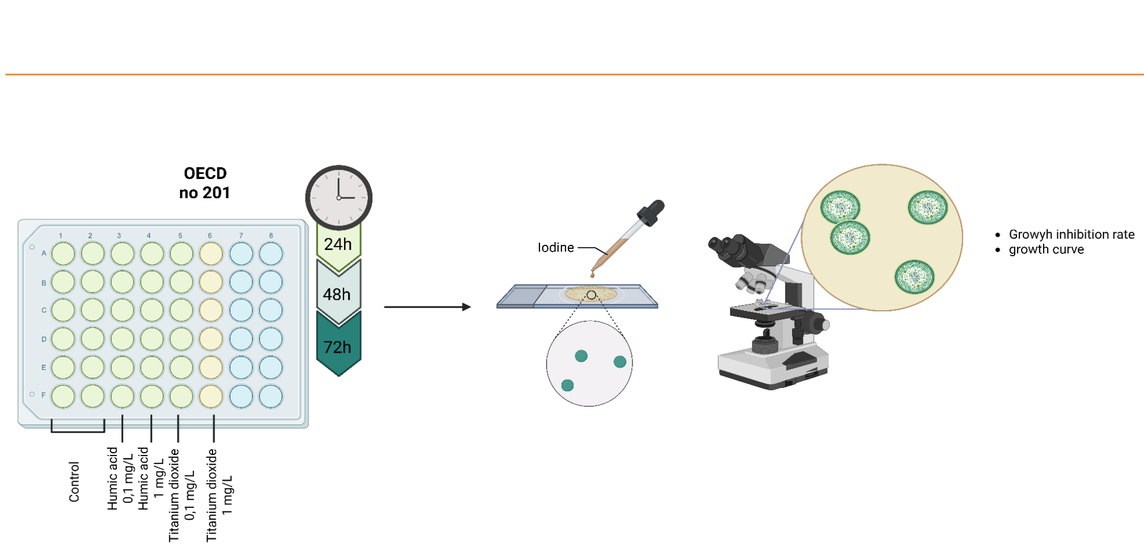
OBJECTIVE

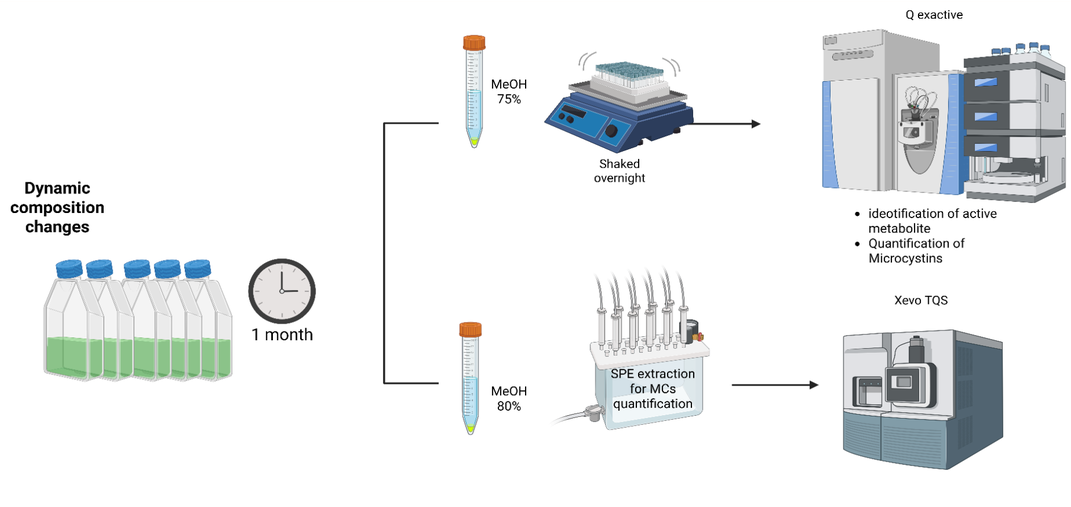
To investigate the growth of *M. aeruginosa* under exposure to TiO2 NPs and to assess chemical alterations on the production of microcystins.

METHODOLOGY

M. aeruginosa (LEGE-91096) was cultured in the presence of TiO_2 NPs (commercial P25) at concentrations of 0.1 and 1 mg/L. Control treatments included humic acid (HA) as surface coating (1:1 ratio). The nanoparticles were characterized by dynamic light scattering (DLS), cytotoxicity was assessed following the OECD 201 guideline, and a metabolomic analysis was conducted using UHPLC-MS/MS.







RESULTS & DISCUSSION

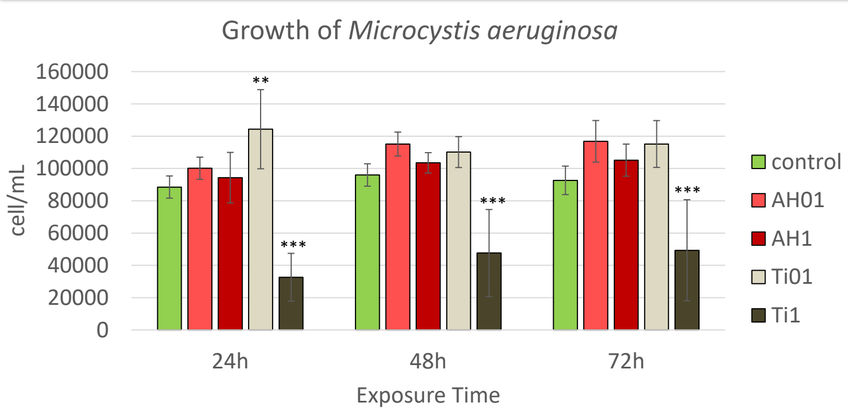


Figure 1. Number of *M. aeruginosa* cells/mL after 24, 48, and 72 hours of exposure to humic acid (HA) and titanium dioxide (TiO_2) nanoparticles at concentrations of 0.1 and 1 mg/L according to OECD 201 guideline.

While HA had no significant effect on growth, TiO_2 NPs induced significant alterations on *M. aeruginosa* growth (**fig. 1**):

- at 0.1 mg/L, the TiO_2 NPs caused an increase at 24h in cell numbers
- at 1 mg/L, the TiO_2 NPs induced a marked decrease in cell density between 24h and 72h.

The analysis of the chromatogram of *M. aeruginosa* exposed to TiO_2 NPs showed six different microcystins congeners (**fig. 2**) suggesting diverse production of toxic compounds.

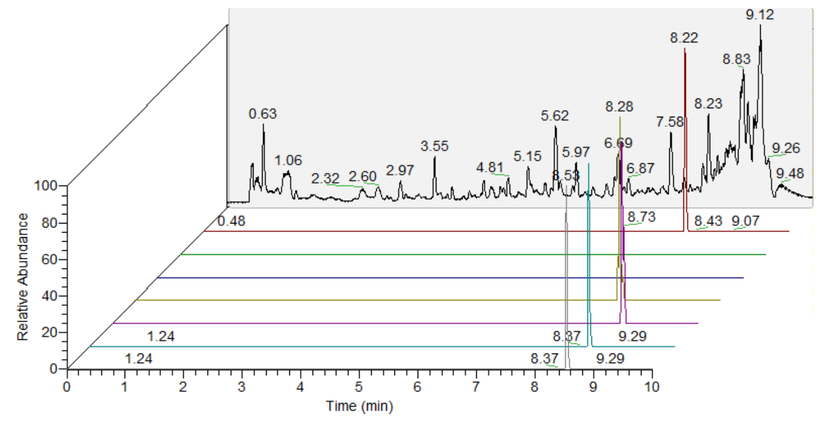


Figure 2. Chromatogram of UHPLC-MS/MS: MC-LR, MC-RR, MC-YR, MC-HilR, MC-LL, and MC-LA. Retention times are indicated above each peak, and extracted ion chromatograms (EICs) are shown in different colors corresponding to each microcystin variant. The sample was obtained from a *M. aeruginosa* culture exposed to TiO_2 NPs.

CONCLUSION

- Exposure to TiO_2 NPs, particularly at 1 mg/L, significantly reduced *M. aeruginosa* viability, indicating a clear cytotoxic effect.
- In addition to growth inhibition, metabolomic analysis revealed an increase in toxic compound production under nanoparticle-induced stress.
- These findings support the hypothesis that stressed *Microcystis* cells may respond by producing a greater quantity or variety of potentially harmful secondary metabolites, posing increased ecological and health risks.

FUTURE WORK

- Investigation of transcriptomic and proteomic responses of the microalgae.
- Evaluation of the effects of nanoparticle-induced stress on the production and toxicity of secondary metabolites and cyanotoxins.

ACKNOWLEDGEMENTS

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