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Abstract

Interactions of Diatom *Cyclotella meneghiniana* and Citrate Coated Silver Nanoparticles †

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Silver nanoparticles (AgNPs) are extensively utilized engineered nanomaterials that inevitably find their way into the aquatic environment [1]. A significant amount of research has been conducted to assess their potential toxicity to aquatic biota [2]. However, the underlying cellular mechanisms involved in the toxicity and tolerance of diatoms to AgNPs are still poorly understood. The present work aimed to gain better insight into the response of diatom *Cyclotella meneghiniana* to AgNP exposure, as a representative model for the lower trophic organisms in a freshwater environment, and its underlying mechanisms. *C. meneghiniana* was exposed to various concentrations of Cit-AgNPs (ranging from 0.001 mg/L to 5 mg/L) for up to 72 h, and the biological responses were compared with those induced by dissolved Ag^+. The response of diatoms to Cit-AgNP and Ag^+ was characterized in terms of diatom growth, membrane permeability, photosynthesis alterations, and morphological changes. The stability of the Cit-AgNPs in the exposure medium was also investigated by determining their dissolution, surface charge and hydrodynamic size. The DLS and SPR-UV-vis results showed a shift in the size distribution of cit-AgNPs towards higher values, which was related to aggregation/agglomeration processes. The dissolution of cit-AgNP in the exposure media increased over time and was concentration-dependent. The calculated 72 h-EC50 values, based on growth inhibition, were 0.348 ± 0.038 mg/L and 0.019 ± 0.001 mg/L for cit-AgNP and Ag^+, respectively, suggesting a higher toxicity of Ag^+ compared to cit-AgNP for *C.meneghiniana*. Short-term exposure (24 h) to Cit-AgNPs and Ag^+ resulted in reduced chlorophyll autofluorescence and impaired membrane integrity in *C. meneghiniana*. Furthermore, the photosystem II was affected, as indicated by a decrease in the quantum yield (Fv/Fm) and an increase in non-photochemical quenching (NPQ). Cells exposed to Cit-AgNPs and Ag^+ exhibited higher levels of proline accumulation compared to the control, implying an activation of the antioxidant mechanisms in diatom, since proline plays a role in ROS scavenging [3]. Additionally, the SEM-EDS analysis revealed an increased presence of polyphosphate bodies (PPB) in both the Cit-AgNP- and Ag^+-treated cells, in response to metal toxicity and stress. Indeed, polyphosphates are known as a chelator of cations, and their accumulation is linked to abiotic stress [4]. This study demonstrates that synergistic mechanisms are adopted by *C. meneghiniana* to deal with toxic levels of silver in both its ionic and nanoparticulate forms.

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