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Evaluation of nanoatrazine bioaccumulation in green microalga *Raphidocelis subcapitata*: characterization, stability and uptake analysis

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The use of nanoencapsulated pesticides, such as nanoatrazine (nATZ), has been proposed as a promising strategy to enhance herbicidal efficiency and reduce environmental toxicity. However, concerns remain regarding their environmental fate and bioaccumulation potential in non-target organisms, especially within aquatic food webs. This study aims to investigate the behavior and initial bioaccumulation of nATZ encapsulated in poly(ϵ -caprolactone) (PCL) nanoparticles in the fresh-water microalga *Raphidocelis subcapitata*, as a primary step toward understanding its trophic transfer.

The nATZ-PCL nanoparticles were synthesized via nanoprecipitation and characterized using dynamic light scattering (DLS). The

resulting particles exhibited an average size of 226 nm, a polydispersity index (PDI) of 0.12, and a zeta potential of -25 mV, indicating a stable and monodisperse suspension. Additionally, stability assessments in different exposure media confirmed that the physicochemical properties of the nanoparticles were maintained under experimental conditions.

In the bioaccumulation assays, *R. subcapitata* cultures were exposed for 7 days to a defined concentration of 0.103 mg/L of nATZ-PCL, corresponding to the previously determined effective concentration (EC₂₅). Quantification of intracellular atrazine was performed via high-performance liquid chromatography (HPLC), revealing a concentration of 1.68 μ g/g dry weight, confirming cellular uptake of the nano-encapsulated herbicide. These preliminary findings suggest that nATZ-PCL is bioavailable to primary producers and can potentially enter the aquatic trophic chain.

These results highlight the importance of monitoring the fate and effects of nanoformulations in aquatic systems. Further experiments involving *Daphnia magna* and *Danio rerio* are currently underway to assess trophic transfer and potential sublethal effects. Biochemical responses, including antioxidant enzyme activities, are being proceeded to evaluate biomarkers effects. Additionally, exposure scenarios will include the presence of natural organic matter (NOM) to better simulate environmentally relevant conditions.

This research is expected to contribute to the establishment of more accurate environmental risk assessment strategies for nanopesticides, supporting regulatory advances and sustainable practices in agriculture.

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