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Abstract

Ecotoxicological Study of the Pharmaceutical Active Substance Isotretinoin

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Pharmaceutical residues are increasingly recognized as environmental contaminants due to their continuous release into aquatic systems. Isotretinoin is a vitamin A derivative used in dermatology to treat severe acne. It is a biologically active micropollutant, yet its ecological effects and potential impact on aquatic organisms remain poorly understood. This thesis aimed to investigate the acute ecotoxicological effects of isotretinoin on the green alga *Desmodesmus subspicatus*. The study aimed to evaluate whether isotretinoin exerts inhibitory or stimulatory effects on algal growth and photosynthetic activity under standardized laboratory conditions.

The algal growth inhibition tests were conducted in accordance with OECD Guideline 201 (Freshwater Alga and Cyanobacteria, Growth Inhibition Test). Each experiment was performed three times independently to ensure reproducibility and reliability of the results. In this method, exponentially growing algal cultures are exposed to a range of test-substance concentrations for 72 hours. The test evaluates the substance's ability to inhibit algal growth relative to untreated controls. Algal growth inhibition was assessed by optical density at 750 nm, and chlorophyll-a content was additionally determined following ethanol extraction. Additionally, the

photosynthetic activity was evaluated using the FluoroMeter Module, which was applied as a complementary analysis outside the standard OECD 201 protocol to evaluate photosynthetic performance and stress responses.

The optical density results showed no inhibition of algal growth at any of the tested isotretinoin concentrations. A moderate growth stimulation was observed in the solvent control, indicating that ethanol contributed to increased biomass. Statistical analysis confirmed that the observed differences among treatment groups were primarily due to solvent effects rather than isotretinoin exposure. Therefore, isotretinoin did not cause measurable growth inhibition in *D. subspicatus* under the acute exposure conditions applied. Chlorophyll-a measurements revealed a more pronounced increase in pigment concentration, particularly at higher isotretinoin levels. This response suggests a physiological adjustment involving enhanced pigment synthesis rather than a toxic effect on biomass production. The observed pattern may indicate a mild stimulatory, hormetic response, characterized by low-dose enhancement of metabolic activity.

During the complementary photosynthetic activity measurements changes in key chlorophyll-a fluorescence parameters indicated slower excitation kinetics and reduced electron transfer efficiency at the highest tested concentration (10 mg/L). These results suggest that isotretinoin can influence the photosystem II (PSII) electron transport chain, reflecting a mild physiological stress rather than direct toxicity. As very limited ecotoxicological and environmental analytical data are available about isotretinoin, this study fills a gap in the environmental risk assessment of the active substance.