

ABSTRACT

Fitra Ari Aditya. 24020121140148. *Enhancement of Lipid Production in Chlorella vulgaris as a Biofuel Feedstock through Hematite Nanoparticle (α -Fe₂O₃) Supplementation with In Vivo Nile Red Staining Analysis.* Supervised by Prof. Drs. Sapto Purnomo Putro M.Si., Ph.D dan Dra. Riche Hariyanti, M.Si.

Microalgae *Chlorella vulgaris* is a promising candidate for third-generation biofuel feedstock due to its rapid growth, high biomass productivity, and substantial lipid accumulation. This study aimed to evaluate the effects of hematite nanoparticles (α -Fe₂O₃) on the growth, lipid accumulation, and biofuel productivity of *C. vulgaris*, using Nile Red (NR) staining as a rapid in vivo lipid quantification method. Five α -Fe₂O₃ concentrations (0, 10, 25, 50, and 75 mg/L) were tested with two replicates. Parameters measured included optical density (OD₆₈₀), dry biomass, lipid content (%), lipid yield (g/L), lipid productivity (g/L/day), fluorescence intensity (RFU), and cell morphology. Results indicated a significant dose-dependent increase ($p < 0.05$) in both growth and lipid accumulation, with the highest biomass observed at 75 mg/L (44.705 g/L), representing a 430% increase compared to the control (8.417 g/L). Lipid content rose from 15.2% in the control to 37.2% at 75 mg/L, while the highest lipid yield and productivity were 16.63 g/L and 1.66 g/L/day, respectively, accompanied by an estimated biofuel potential of 14.97 mL/L/day. Fluorescence analysis confirmed enhanced intracellular lipid accumulation, with RFU values peaking at 4395 ± 105 under 75 mg/L treatment, alongside an increased mean cell diameter (1.336 μ m). NR staining revealed strong, uniform yellow-orange fluorescence in treated cells, indicating broad lipid distribution without impairing photosynthetic functionality. These findings demonstrate that α -Fe₂O₃ supplementation significantly enhances both growth and lipid biosynthesis in *C. vulgaris*, highlighting its potential as an effective strategy for scalable biofuel production.

Keywords: *Chlorella vulgaris*, α -Fe₂O₃, lipid, Nile Red, biofuel.