

Determining the Role of n-Hexane in Multiple Phases in *Nannochloropsis* sp. and *Nitzschia* sp.

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Algae biofuels have great potential to become a viable fuel source and replace non-renewable hydrocarbons such as petroleum and coal. A major difficulty in producing algal fuel is the extraction of crude oil from the cell wall. Hexane solvent is one of the most common methods for extracting lipids, and this experiment addresses how multiple phases of solvent extraction affect the overall lipid yield from *nannochloropsis* sp. and *nitzschia*. This in turn can be utilized to develop more efficient and cost-effective ways of producing algae fuel. This study uses gravimetric analysis to demonstrate changes in the mass of a central algae biomass, as well as the masses of each extraction phase. In each phase, 40 mL of n-Hexane was added to a biomass with an initial mass of 2.00 g. The hexane dissolved and suspended the oil, and a centrifuge was used to separate the biomass and solvent solution. The results demonstrated that multiple phases of n-Hexane extraction are more effective with *Nannochloropsis* sp., showing a direct correlation between the percentage of biomass extracted and the number of phases with the average first phase extracting 29.41% of the total yield, second phase extracting 49.01% and the third phase extracting 21.58%. However, the preliminary solvent phase with *Nitzschia* sp. is the most effective overall, supported by an average first phase accounting for 76.52% of the total yield. Thus, the results between phases and a chi-square test indicate statistically significant results (p -values= 1.83×10^{-3} for *Nannochloropsis* sp. and <0.001 for *Nitzschia* sp., $\alpha=0.05$).