Studying the Microalgae Growth and Stable Nanoemulsion Production Systems for Environmental Mitigations

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ABSTRACT

Recently, environmental concerns have been rising in the world because of the greenhouse gases, such as CO_2, N_2O, CH_4, and fluorinated gases and industrial products, which in turn cause irreversible climate changes. Algal oil is considered to be one of the most promising alternatives of natural sources for energy production, but the lipid accumulation and lipid productivity of microalgae have been far from the desired levels. This research focuses on culturing spirulina algae in a tank in the laboratory with optimum growth conditions over a period of 40 days. Then, the lipid content was increased by growing microalgae in stressful conditions under low temperature, light or low nutrient supply. After the lipid extractions, produced algae oil was converted into nanoemulsion using surfactant and sonication process. The test results showed that the particle size of the emulsions was less than 10 nm and stable (without any separation) for at least two months.

Keywords: algae growth, harvesting, oil extraction, nanoemulsion, energy.

1 INTRODUCTION

The rapid extinction of fossil fuels, increasing population using crude oil and natural gas, and unevenly rising crude oil prices have contributed to one of the most challenging problems in the world [1]. Most of the world's energy essentials are for industrial use, transportation, and general household purposes. Electricity constitutes only 30% of world's energy, while nearly 70% depends on fossil fuels, which is considered to be a serious problem due to the greenhouse gasses, such as CO_2, N_2O, and CH_4 of the fossil fuels. The enormous increase of the world population is directly proportional to energy demands, which made a persistent dependence on fossil fuels for the generations of various energies [1-5].

The emission of CO_2 in the atmosphere was used to be around 280 ppm in the old days, and now it has increased to 420 ppm, which is almost an increase of 60% and being increased at an average rate of 2.4 % per annum. The significance of the high figures on world's energy consumption has become a major global issue. As an alternative to fossil fuels, biomass energy has been emerging as a standby of renewable energy sources. Biomass energy can be produced by algae, which are earthly crops for conversion into biocrude and biogases. The evolution of producing biofuel (e.g., ethanol and biodiesel) using corn began in early 1970s in the United States and Brazil and achieved remarkable results. Biofuels have become interestingly the major alternatives to all the challenges [6-10].

Oil contents of microalgae are usually between 20 and 50wt% on an average dry base, but some algae strains (e.g., Botryococcus braunii) have an oil content of up to 80% when stressed under appropriate conditions (e.g., freezing temperature, varying intensities of light sources, and lack of nutrition) [1,11]. The fatty acids attached to the triacylglycerols of algae oil can be long and short chain hydrocarbons. The shorter the chain length, the more they are ideal for the production of a biofuel. The long chains may have other beneficial uses, as well. Many species of algae have been examined for their production capabilities of fatty acids [12,13]. Table 1 provides the oil contents of some of the microalgae spices [1].

Table 1: Oil contents of some of the microalgae spices [1].

Microalgae	Oil Contents (% Dry Wt)
Botryococcus braunii	25-80
Chlorella protothecoides	23-30
Chlorella vulgaris	14-40
Cylindrotheca sp.	16-37
Dunaliella salina	14-20
Neochloris oleoabundans	35-65
Nitzschia sp.	45-47
Phaeodactylum tricornutum	20-30
Schizochytrium sp.	50-77
Spirulina maxima	4-9

In order to produce oil from the algae biomass, cellular lipids need to be extracted from the cell bodies. Extraction of the lipids requires rupturing the algal cells and then using various methods, such as mechanical pressing, chemical breakdown, solvent extraction, supercritical fluid extraction, heating/cooling, and filtering, oil can be produced from the algae biomass [1]. After the extraction process, the oil is purified for different industrial applications.

Nanoemulsions are often described as the dispersions of an oil phase in an aqueous phase which are obtained when the size of an emulsion are at nanoscales - commonly in the range 1 to 100 nm. Nanoemulsions usually cannot form readily by hand agitation, so they require a high intensity energy source for the emulsion which can be supplied by selected mechanical devices [1]. Ultra-sonication is one of the most used mechanical devices for nanoemulsion production and stabilization. This mechanical device generates high shear force to make finer size of emulsions. Besides the sonication, high speed mixer/agitation, homogenizers and fluidizers can be used to make nanoemulsions.

2 EXPERIMENT

2.1 Materials

Live algae (spirulina) culture and starter mix were purchased from the Algae Labs. Pure cells were cultured by growing them in a fish tank and plastic bottles under appropriate conditions. After fully grown, it was harvested by the natural method of pumping and dried under the sun. The dried biomass was extracted for oil production with chemicals (chloroform and methanol). Tanfloc for flocculation and sodium dodecyl sulfate (SDS) for nanoemulsion process were also purchased and used in the same process without any modifications.

2.2 Methods

Microalga spirulina platensis (class of Cyanobacteria) was cultured in pure Brita water using fluoresce light source, air bubbles, and starter mix solution provided from the manufacturer. The starter mix consisted of sodium carbonate, potassium nitrate, sea salt, ammonium phosphate, strong green tea, and iron sulfate. The temperature was between 30 and 33 °C (by heater) and culturing time was about 40 days. Filtration and flocculation (Tanfloc) techniques were used to harvest the algae from the water in the tank and bottle [1].

The harvested algae was dried, frozen for a few days, ground by a mortar grinder, and mixed with chloroform and methanol (2 to 1 ratio) for oil extraction. The mixture was refluxed for about 6 hours for the oil dissolution. After separating of algae biomass from the oil in a funnel, oil reach solvent was evaporated using a rotavapor under vacuum to get the algae oil. Extracted algae oil and pure water were sonicated in SDS solutions at different time and concentrations to produce algae oil-based nanoemulsion for different industrial applications. Optical microscope, UV-vis spectroscopy and dynamic light scattering (DLS) techniques were used to characterize the produced nanoemulsions [1]. Figure 1 shows the images of the algae cultivation in a tank, produced algae oil, and nanoemulsion of the algae oil after the sonication.

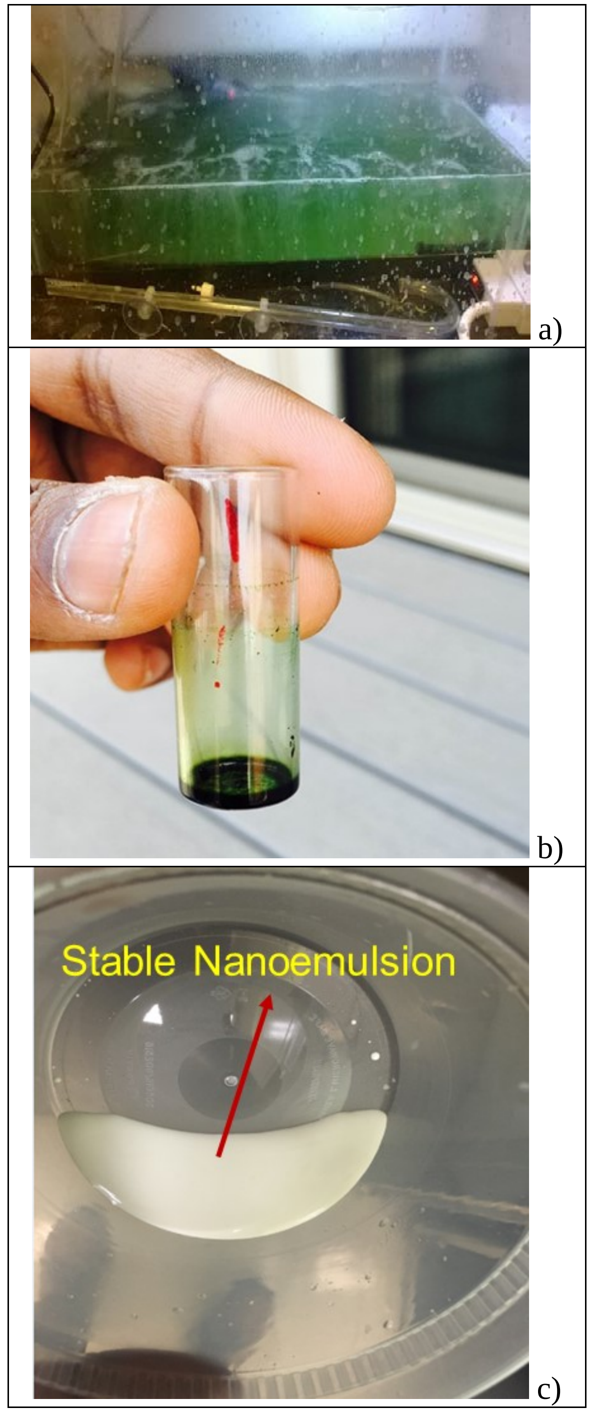

Figure 1: Images showing the a) algae cultivation in a tank, b) produced algae oil, and c) nanoemulsion of the algae oil.

3 RESULTS AND DISCUSSION

In order to make nanoemulsions from the algae oil, two sets of tests were conducted on the algae oils. In the first set of tests, 0wt%, 0.1%wt, 0.2%wt, and 0.4% SDS were added into pure water, mixed well, and the sonicated for 10 minutes to make stable nanoemulsions. In the second set of the tests, 0.2wt% of SDS was kept constant while changing the sonication times, including 2.5, 5, 10 and 20 minutes. It was clearly seen that at 0 wt% of SDS and 10 minutes of sonication, nanoemulsions were separated (not stable) and became two different phases (oil and water) in a short time period [1]. However, adding the SDS (up to 0.4wt%) into the emulsion process, stable nanoemulsions were obtained as is seen in Figure 1c. This indicates that algae oil requires a stabilizing agent for a better nanoemulsion production in pure water. Increasing sonication time from 2.5 to 20 minutes, while keeping the SDS constant of 0.2wt%, reduced the size of nanoemulsion from micron to nanosize [1].

Dielectric constant, reflective index, and viscosity values of the inclusions/nanoemulsions were found from our previous studies [1], while the absorption peaks of the prepared nanoemulsions were determined using the UV-Vis spectroscopy. The droplet sizes of the nanoemulsions were determined by the DLS technique using the parameters, such as viscosity, absorbance, and dielectric constant for each nanoemulsion sample. As detailed in [1], the average particle size of the nanoemulsion is 9.5 nm. These studies indicated that sonication time and stabilizing agent are dominating factors in order to produce nanoscale emulsions [1].

In general, micro emulsions are not sable and can separate during the storage time; however, nanoemulsions are usually sable or metastable, and can stay longer without any separation. Usually, sonication technique is used to rupture large microscale droplets into nanoscale droplets to obtain nanoemulsions. In spite of their nature, nanoemulsions can be used over many months or even years. The stable nature of the emulsions are due to the addition of surfactant SDS and longer sonication times [1]. Due to the environmental concerns, renewable energy systems have been gaining much attention worldwide. It is also stated that algae based biomasses and oils can be the major biomass sources of the renewable energies available in the world to address some of the major environmental problems.

4 CONCLUSIONS

Because of the environmental and health concerns of the conventional fuels, many studies have been focused on the renewable energy sources. Algae based biomass and their products can be the greatest opportunities for the energy demand of the world. In this study, spirulina microalga was cultured in a water tank under specific nutrition, temperature and air bubble, and harvested prior to the oil extraction process. Algae oil was extracted using solvents (chloroform and methanol) after stressing at freezing temperature. The obtained algae oil was converted into stable nanoemulsion using SDS surfactant, and different sonication times. Test results showed that sonication time and SDS concentrations had a major impact on the size of the nanoemulsions (less than 10 nm) and their stability. This study may open up new possibilities to extend this project for combustion engine and fuel burner uses for environmental remediation.

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