## Abstract

Fossil fuels are non-renewable energy sources, and different sources have estimated that with the current utilization rate, the earth is left with petroleum reserves to sustain for roughly 50 years. Humankind cannot stop the use of fossil fuels right away as economic growth and development are underpinned by a stable and reliable energy source. Finding a sustainable and reliable renewable alternative to petroleum is of prime importance for the survival of the global society and for avoiding significant catastrophes for human civilization.

Biofuel has similar physiochemical properties and energy density to petroleum fuels, and existing industrial infrastructure can easily switch to biofuels, making biofuels one of the most promising fuel sources capable of replacing petroleum. However, there are concerns about the environmental impact of producing biofuels using first- and second-generation feedstock, such as land-use changes, water use, and competition with food production. This has led researchers to focus on the 3<sup>rd</sup> and 4<sup>th</sup> generation of biofuels using microalgae as the feedstock.

Microalgae are unicellular photosynthetic microorganisms living in aqueous environments. Microalgae can grow in various environments, such as freshwater, saline water, sewage, etc. Microalgae have high photosynthesis efficiency, almost 50 times higher than some terrestrial plants. In optimal conditions, microalgae double its biomass within 24 hours. Microalgae can accumulate high lipid contents within their cells, up to 80% of their dry weight in certain species. The ability to accumulate high lipid content makes microalgae a very attractive feedstock for biodiesel production. All these properties make microalgae a preferred feedstock for biofuel in a bio-refinery approach.

However, the high production cost of microalgae biomass makes microalgal biofuel technologies commercially unacceptable. Amongst all the steps involved in microalgae biofuel technology, microalgae biomass production is one of the most cost-intensive processes. Over 90% of microalgae biomass is produced globally in open culture systems. Commercial microalgae biodiesel production is still not sustainable because of the low density of biomass in open culture systems, resulting in expensive processing costs. Researchers are actively exploring innovative cultivation techniques and optimizing growth conditions. Research in microalgae culture systems is crucial for advancing the sustainability of microalgae biofuel production. Developing efficient

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and economically viable microalgae biofuel production systems requires a deep understanding of the intricate biological and environmental factors influencing microalgae cultivation.

The current study is carried out with the following objectives to address the issues current largescale microalgae culture systems face.

- 1. To design and develop a photobioreactor (PBR) for mass-scale microalgae cultivation.
- 2. To optimize the culture conditions and improve the biomass and lipid productivity.
- 3. To analyze the algal biomass and biofuel properties produced from the microalgae cultured in the developed PBR.

The thesis is divided into five major chapters, with chapters 3 and 4 sub-divided into three chapters per the objectives. Synopsys of the chapters are presented below:

**Chapter 1** introduces the current global scenario and the objectives leading to this research. It presents the prevailing energy scenario and the scope and limitations of available renewable alternatives. Furthermore, it illustrates the significance of 3<sup>rd</sup> and 4<sup>th</sup> generation biofuels derived from microalgae feedstock, its advantages and limitations.

**Chapter 2** presents literature reviews related to this research work. A literature review was conducted to study the available microalgae culture systems and their pros and cons, followed by factors affecting the design of the microalgae culture. Subsequently, studies on various optimization methodologies to optimize the parameters of the developed system were carried out.

**Chapter 3A** demonstrates the methodology for designing and developing three microalgae culture systems. Finally, a photobioreactor capable of mass-scale microalgae culture was designed and developed, prioritizing biofouling prevention, which was a significant issue in the success of wide-scale implementation of closed photobioreactor systems. The developed microalgae culture systems were tested by culturing microalgae and then harvesting and characterizing the final products.

**Chapter 3B** demonstrates the methodology opted for optimization of culture parameters of the developed microalgae culture system. Face centered central composite design (FCCCD) approach of RSM was used with the independent variables, nitrogen concentration (sodium nitrate), light intensity, light duration, and air flow rate, to model and optimize for the highest possible resultant

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responses, i.e., biomass and lipid productivity of *Chlorella homosphaera*. FCCCD was chosen for this experiment as it requires only three levels, the highest, lowest, and midpoint of each experimental variable, making it simple and having the most negligible probability of error.

**Chapter 3C** demonstrates the methodology for characterizing the biomass and lipid produced in the developed microalgae culture system. The dry microalgae biomass was analyzed for its physiochemical properties like calorific value, ash, and lipid content. The lipid obtained was then further processed into biodiesel using transesterification, and the properties of the obtained biodiesel were investigated using <sup>1</sup>H NMP and GCMS.

**Chapter 4A** describes the design philosophies of the three microalgae culture systems being developed. The chapter also illustrates the advantages and limitations of the developed microalgae culture systems. Finally, the chapter demonstrates the findings of the scientific experimentations and analysis carried out in the developed microalgae culture systems.

**Chapter 4B** describes the optimization of performance or the biomass and lipid productivity of the developed Stacked Tray Automated Modular Photobioreactor (STAMP) achieved by optimizing the culture conditions like light intensity, light duration, light wavelength, airflow rate, and nitrogen content using Face Centered Central Composite Design approach of Response Surface Methodology. The optimization is detailed in this chapter.

**Chapter 4C** details the analytical findings to characterize the biomass and biofuels obtained from the STAMP systems developed during the current investigation. The calorific value, ash content, lipid content, lipid characterization using GC-MS, biofuel production estimation using <sup>1</sup>H-NMR spectroscopy, oil density, acid value, and carbon residue are characterized and compared with the global standards in this chapter.

**Chapter 5** summarizes the overall findings of the current research work. Further, it sheds light on the possible future scope of the current investigation.