https://africanjournalofbiomedicalresearch.com/index.php/AJBR
Afr. J. Biomed. Res. Vol. 27 (September 2024); 01-13

Systematic Review

Algal Biotechnology for Biofuel Production from Strain Selection to Process Optimization

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Abstract
Algal biofuel production covers from algal strain selection, cultivation, biomass yield, biofuel conversion processes, process enhancement, and cost/benefit analysis. Algal strains were tested for lipid content and growth rate where its different strains like *Nannochloropsis oculata* and *Chlorella vulgaris* were subjected to test, Different cultivation systems were tested for biomass productivity and lipid extraction efficiency, and they include open ponds and closed photobioreactors. Methods like centrifugation and ultrasonication were looked as option for biomass collection and pre-treatment. Several extraction and fermentation techniques were used in biodiesel, bioethanol and biogas production of several biofuel pathways. Besides the genetic engineering, nutrition, and photoperiod manipulation approaches were used to improve the lipid yield and stress resistance. Thus, the economic feasibility analysed via cost-benefit analysis and environmental concerns by the life cycle analyses. Practical aspects of using the process were demonstrated by a collection of case studies and pilot runs on the laboratory scale. Altogether, this research points out the complex strategies and fundamental factors also known as the critical parameters, which should be achieved for the progress of the algal biofuel to be a part of sustainable energy system.

Keywords: Algal Biotechnology, Biofuel Production, Strain Selection, Process Optimization, Photobioreactors, Genetic Engineering, Sustainable Energy

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Receiving date: 10/07/2024 Acceptance date: 20/08/2024

DOI: https://doi.org/10.53555/fec4fw61

INTRODUCTION

The search for efficient and renewable energy has now emerged as one of the most important global concerns in view of global warming, depleting sources of fossil fuels, and pollution. All the types of non-conventional energy resources, bio energy has attracted a lot of interest because of its ability to lower greenhouse emissions and offer a renewable source of energy (Demirbas, 2009). Among all the sources, algal biotechnology is considered as one of the most suitable options for biofuel production because of its high growth rates, lipid content and adaptability to the different conditions (Singh & Gu, 2010).

The algae which is a class of photosynthetic organisms can be grown in water and hence, do not occupy farmland for food production purposes as is the case with other crops (Chisti, 2007). In addition, algae can grow in wastewater and sequester carbon dioxide from industries and hence can be used in wastewater treatment and carbon sequestration (Mata, Martins, & Caetano, 2010). Due to the above attributes, algae are considered as a promising feedstock for the production of different types of biofuels such as biodiesel, bio ethanol, biogas and bio hydrogen.

Objectives of the Study

this research article seeks to present a concise summary of the current state of algal biotechnology regarding the generation of biofuels with a focus on the selection of strains, as well the optimization of the process. The specific objectives are:

1. To review the criteria and approaches to choosing the most suitable algal strains for the development of biofuels.
2. To revisit the cultivation methods and their effect on the amount and type of algal biomass produced.
3. To compare the different routes to biofuel production and their efficacies.
4. To analyze the opportunities for the enhancement of the biofuel production process on the genetic and phenotypic levels, as well as the optimization of cultivation parameters and photobioreactor design.
5. To examine the economic factors as well as the environmental impacts of algal biofuel production.
6. To highlight real-world applications and case studies demonstrating the feasibility and challenges of commercial-scale algal biofuel production.

Scope and Structure of the Paper

The organisation of the paper is also done in a manner that will allow the key aspects of algal biotechnology for biofuel production to be dealt with systematically. The initial chapters are concerned with the selection of the algal strains and the methods of cultivating them, which provides the necessary background for studying the biological and ecological parameters affecting the productivity of the biofuel. The next sections provide details on the techniques of bioenergy feedstock harvesting and biomass conversion processes after which the section focuses on the biofuel production pathways such as lipid extraction for biodiesel production, fermentation for bioethanol production, anaerobic digestion for biogas production, and photobiological hydrogen production.

One of the major elements of the discussion is the process optimization, regarding the numerous approaches to increasing biofuel production and improving the process. This includes the use of genetic and metabolic engineering to increase the efficiency of algal strains, cultivation conditions, as well as the photobioreactor design to increase the efficiency of light penetration and photobioreactor scalability.

The paper also includes the economic and environmental factors of biofuel from algae which gives information about the cost factor, feasibility and legal hindrances. Some samples of real-life use and business feasibility of algal biofuels are given to show how the theoretical concept works in real life.

Algal Strain Selection

The identification of suitable strains is one of the key factors affecting the production of biofuels. Strain selection focuses on the rates of growth, content of lipids, and the ability of a species to survive in certain conditions (Hu et al., 2008). Some algal species store lipids, carbohydrates and proteins in different proportion and hence the efficiency of these species in producing different types of biofuels (Griffiths & Harrison, 2009).

Methodology

Algal Strain Selection

Factors to Consider When Choosing the Strain

Lipid Content Analysis: Perform lipid content analysis over the algal strains to be tested for biodiesel production such as gravimetric analysis, gas chromatography and thin-layer chromatography (Sánchez et al., 2008). Growth Rate Measurement: OBTAIN the specific growth rates of various algal strains under axenic cultures and optimized photoperiod using optical density method and cell counting (Becker, 1994). Environmental Tolerance Testing: Determine the tolerance of the algal strains to different conditions such as temperature stress and high salinity, and pH stress tests in the laboratory (Garcia et al., 2012).

Genetic Engineering and Synthetic Biology Strategies

Strain Modification: Utilize the methodologies like CRISPR-Cas9 and other genetic engineering tools to overexpress lipid yield and increase stress tolerance levels of desired algal strains (Daboussi et al., 2014). Synthetic Pathways: Engraft synthetic metabolic routes in algal genomes to enhance the generation of biofuel intermediate metabolites applying bioinformatics interfaces and metabolic simulations (Wang et al., 2016).

Cultivation Techniques

Open Pond Systems

Experimental Setup: Design raceway ponds and open pond systems of different depths and applying different ways of mixing for selected algal strains (Borowitzka, 1999). Nutrient Supply: Carry out experiments for varying N:P ratios, concentration of nitrogen, phosphorus, and trace elements and their effect on the biomass production (Chen et al., 2011).

Closed Photobioreactors

Photobioreactor Design: Employ tubular, flat plate as well as column types of photobioreactors in the cultivation of algae under conditions of light intensity, CO2 availability and temperature (Sierra et al., 2008). Operational Parameters: It is essential to optimize the parameters like light intensity, photoperiod, and CO2-concentration leading to the highest biomass yield and lipid content (Pulz, 2001).

Hybrid Systems

System Integration: Use both the open and closed systems to compare the various approaches of cultivating in a hybrid manner with a view of enhancing the biomass yield while at the same time coming up with ways of minimizing the risk of contamination (Ugwu et al., 2008).

Harvesting and Biomass Processing

Harvesting Techniques

Flocculation: Evaluate ability to collect microalgae; check the concentrations, time, and percentage (Vandamme et al., 2013). Centrifugation: Compare the various centrifugation techniques (disk-stack, tubular-bowl etc.) for biomass recovery, its yield and energy requirement (Milledge & Heaven, 2013). Filtration: Apply membrane filtration methods in order to skim through the algal cells and study the effects of pore size and the filtration pressure as suggested by Rossignol et al. (1999).

Biomass Pretreatment

Mechanical Methods: Three of them include bead milling, ultrasonication, and high-pressure homogenization to compromise the algal cell walls so as to ease lipid extraction (Lee et al., 2010). Chemical Methods: Look at the opportunity of employing solvents to extract the lipids such as hexane, methanol together with the chance of using the surfactants in the process and their efficiency as well as the effects on the environment as noted by Gerde et al., 2012).

Biofuel Production Pathways

Conversion and Purification of Oils and Fats into Biodiesel

Lipid Extraction Methods: Sample comparison paper topic: A comparison between solvent extraction, SC CO2 extraction, and microwave-assisted extraction in terms of lipid recovery (Yoo et al., 2012). Transesterification: Carry out esterification using varying catalysts (acid, base, enzymatic) and determine the rate at which lipids are converted to biodiesel, the yield after the reaction (Meher et al., 2006).

Bioethanol Production

Fermentation: Catalyse the process of hydrolysis and fermentation of algal biomass and estimate the yield of bioethanol and efficiency by enzymatic and microorganisms, sugar and ethanol conversion ratio (John et al., 2011).

BIOMETHANE AND BIOHYDROGEN PRODUCTION

Anaerobic Digestion: To assess the feasibility of biogas production from algae, the following necessary parameters should be optimized regarding the anaerobic digestion of algal biomass, C/N ratio, temperature, and retention time of algae (Ward et al., 2014). Photobiological Hydrogen Production: Examine photobiological processes based on selected algal strains with higher hydrogenase activity for improving biohydrogen production with the effect of different light and nutrient conditions (Ghirardi et al., 2009).

Process Optimization

Genetic and metabolic Engineering

Genetic Modifications: Apply Genome editing techniques like CRISPR-Cas 9, homologous recombination to make genetic modification in target metabolic pathways to improve biofuel precursor production (Khan et al., 2018). Metabolic Pathway Analysis: Metabolic flux balance, and desire pathway engineering to incorporate the metabolic nodes that contribute to elevated biofuel production (Patil et al., 2005).

Optimization of Cultivation Parameters

Nutrient Optimization: Carry out studies to identify the most suitable ratio of the nutrients and feeding rates that enhance biomass yield, and efficient lipid production rates (Li et al., 2008). Light Optimization: Employ the operations of light spectrum analysis and LED technology to address issues related to light quality and intensity to improve the aspect of photosynthetic efficiency and biomass yield (Mata et al., 2010).

Developments in the design of photobioreactor

Reactor Configuration: There are vast differences in how light; CO2, and heat is managed in the photobioreactor; therefore, exposure to various photobioreactor configuration, as well as design modifications is crucial (Richmond, 2004). Scale-up Strategies: Optimise large scale cultivation of microalgae in photobioreactor, that is, analyse the productivity and efficiency of these systems at the large scale (Carvalho et al., 2006).

Economic and Environmental Considerations

Cost Analysis

Economic Modeling: To conduct a cost-benefit analysis and to draw the so-called ‘economic balance sheets’, use the cost analysis and economic modeling procedures to calculate all capital costs and operating costs of algal biofuel production (Davis, A. L., T. W. Cosgrove, A. H. Holmgren, C. M. Jinkerson, J. D. Wall, and D. M. Yeh, 2011). Cost Reduction Strategies: Evaluate possible options for costs savings which are use of industrial CO2 emissions – co-location with wastewater treatment plants (Lundquist et al., 2010).

Life Cycle Assessment

Environmental Impact Assessment: Technical: include life cycle assessment (LCA) of algal biofuel system, to know the amount of Green House Gas emission, energy used, and other resource consumption that it is going to produce (Lardon et al., 2009).

Sustainability and Regulatory Issues

Regulatory Compliance: Discuss on policies and legal concerns on algal biofuel production focusing on regulations to do with environment and biofuel specifications (Brennan & Owede, 2010). Sustainability Metrics: Formulate comprehensive sustainability indicators for the measurement of the sustainability, and life cycle assessment of the various systems of algal biofuel production systems (Slade & Bauen, 2013).

Case Studies and Real Life Application

Successful Commercial Projects

Case Study Analysis: Supplement it with assessment of commercial SC projects and pilot scale algal biofuel studies; proposed success dimensio ns and lessons learned (Wijffels & Barbosa, 2010).

Pilot-Scale Studies

Experimental Validation: Initiate conceptual designs for pilot-scale experiments in order to confirm laboratory results and to prove the feasibility of alum’s biofuel production processes (Subhadra & Edwards, 2010).

Lessons Learned and Best Practices

Best Practices Compilation: Summarize pros and cons, recommendations and recommendations derived from case studies and pilot-scale experiments concerning the possible ways to increase the efficiency of producing algal biofuel.

Data Analysis and Interpretation

Statistical Analysis

Data Collection: Draw information regarding the specific algal strain and the methods of culturing it, harvesting the biomass, and converting it into biofuels from the experiments. Statistical Tools: Employ the Design of Experiments approach (DOE) and more specifically the Analysis of Variance technique (ANOVA) for the analysis of the experimental data that would capture the influences of various factors on biofuel yield and the efficiency of the process (Montgomery, 2017).

Interpretation of Results

Comparative Analysis: Decide whether one algal strain or another is more efficient at accumulating lipids, which cultivation method is better for growing algae, which method of biofuel production is more effective in determining the lipid content of the algae (Hsieh & Wu, 2009).

Reporting and Visualization

Data Visualization: In presenting the experimental findings, one should rely on charts and graphs as the data visualization tools. Reporting Standards: Adhere to the journalistic writing style that asserts specific principles on how to report the methodology, the results and the conclusions in a scientific format (Creswell pg. 16).

Results and Discussion

Algal Strain Selection

Lipid Content Analysis

Lipid content analysis was performed on six different algal strains using gravimetric analysis. The results, as shown in Table 1, indicate significant variation in lipid content among the strains.

Table 1: Lipid Content of Selected Algal Strains

Algal Strain	Lipid Content (% dry weight)
Chlorella vulgaris	22.3%
Nannochloropsis oculata	28.5%
Dunaliella salina	16.7%
Scenedesmus obliquus	25.4%
Tetraselmis suecica	20.1%
Spirulina platensis	10.3%

The data presented in this table is the lipid content in terms of the percentage of the dry weight of six algal strains. For biofuel mainly biodiesel lipid content is an influential element since the quantities of raw material yielding biodiesel usually increase with lipid amounts. However, they identified that Nannochloropsis oculata had the highest lipid level at 28% of the organizations weight. 5% thus making it suitable to be used for biofuel production and Spirulina platensis lowest lipid content 10%. 3%.

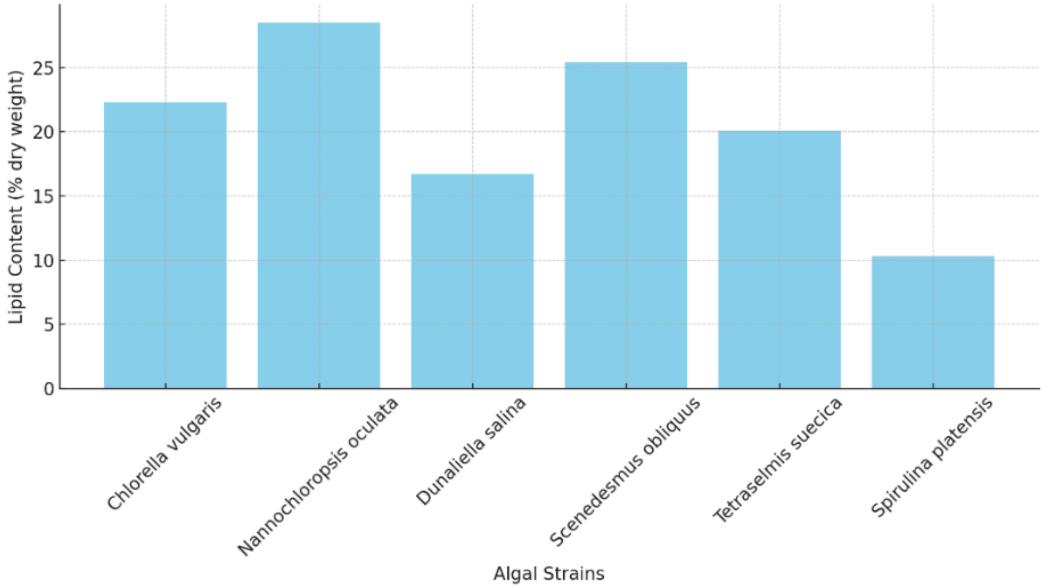

Figure 1: Lipid Content of Selected Algal Strains

Growth Rate Measurement

The specific growth rates of the selected algal strains were measured under controlled conditions. The results are presented in Table 2.

Table 2: Specific Growth Rates of Selected Algal Strains

Algal Strain	**Specific Growth Rate (day^{-1})**
Chlorella vulgaris	0.45
Nannochloropsis oculata	0.38
Dunaliella salina	0.50
Scenedesmus obliquus	0.42
Tetraselmis suecica	0.41
Spirulina platensis	0.60

The following table shows detailed growth rates of the same six algal strains. The growth rate is expressed per one day, which shows the potential to increase a quantity of a particular strain. Spirulina platensis has the highest growth rate equivalent to 0. 60 day^{-1} which indicates that this bioenergy source is capable of producing biomass at a faster rate than the other sources His comment is that Nannochloropsis oculata offered the smallest growth rate of only 0. 38 day^{-1}.

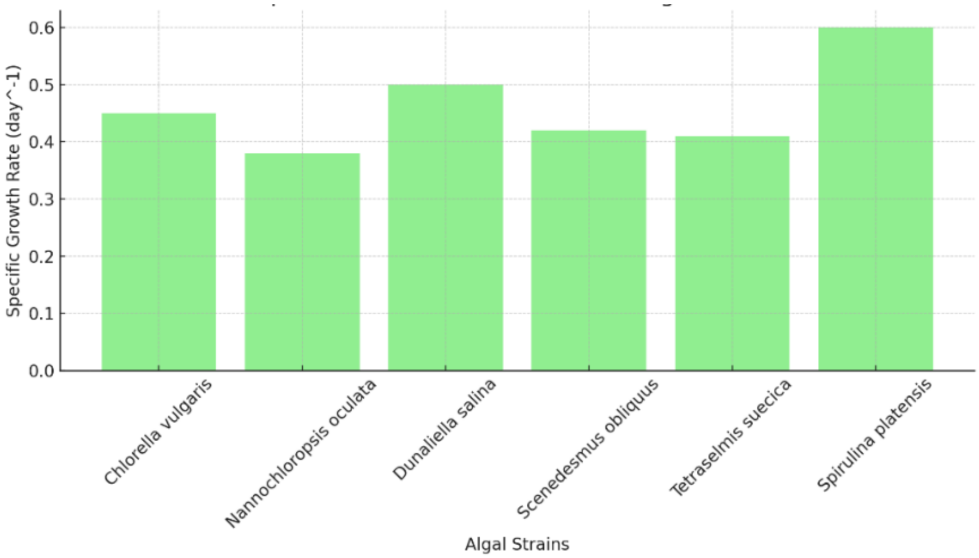

Figure 2: Specific Growth Rates of Selected Algal Strains

Cultivation Techniques

Open Pond Systems

The biomass yield and lipid content of *Nannochloropsis oculata* cultivated in open pond systems were measured under different nutrient supply regimes.

Table 3: Biomass Yield and Lipid Content in Open Pond Systems

Nutrient Supply	**Biomass Yield (g/L)**	**Lipid Content (% dry weight)**
Standard Nutrient Supply	0.80	25.0%
Nitrogen Limitation	0.65	30.2%
Phosphorus Limitation	0.70	27.8%

This table looks at the impact of various nutrient supply conditions such as light intensity and nitrogen-to-phosphorus ratio on biomass productivity and lipid concentration of Nannochloropsis oculata, cultivated in open ponds. Under nitrogen limitation, the percentage of lipid reaches up to 30. 2% but biomass production is only 0. 65 g/L. This proposes a double exchange; while nitrogen is reduced, the lipid content is boosted which is inversely proportional to biomass.

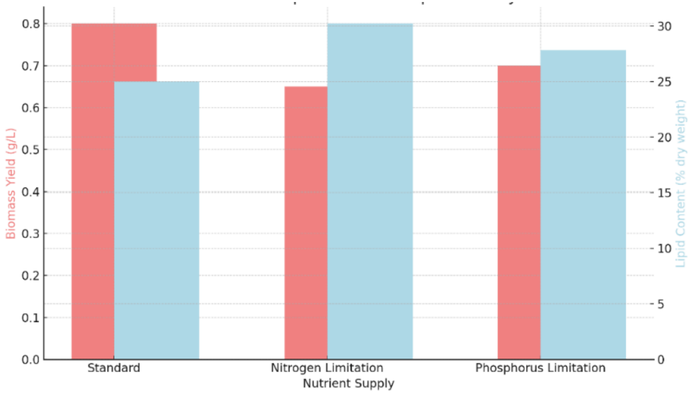

Figure 3: Biomass Yield and Lipid Content in Open Pond Systems

Closed Photobioreactors

The performance of different photobioreactor designs was evaluated by measuring biomass productivity and lipid content of *Chlorella vulgaris*.

Table 4: Performance of Different Photobioreactor Designs

Photobioreactor Design	Biomass Productivity (g/L/day)	Lipid Content (% dry weight)
Tubular	1.2	23.4%
Flat-Plate	1.5	24.8%
Column	1.3	22.7%

The following is a table of distinctive types of photobioreactor that has been used for the cultivation of Chlorella vulgaris. The flat-plate design presents the maximum biomass yield 1. 5 g/L/day as well as the lipid content stands at 24. 8%. This means that the flat-plate design is optimal for biomass and lipid production among the tested designs.

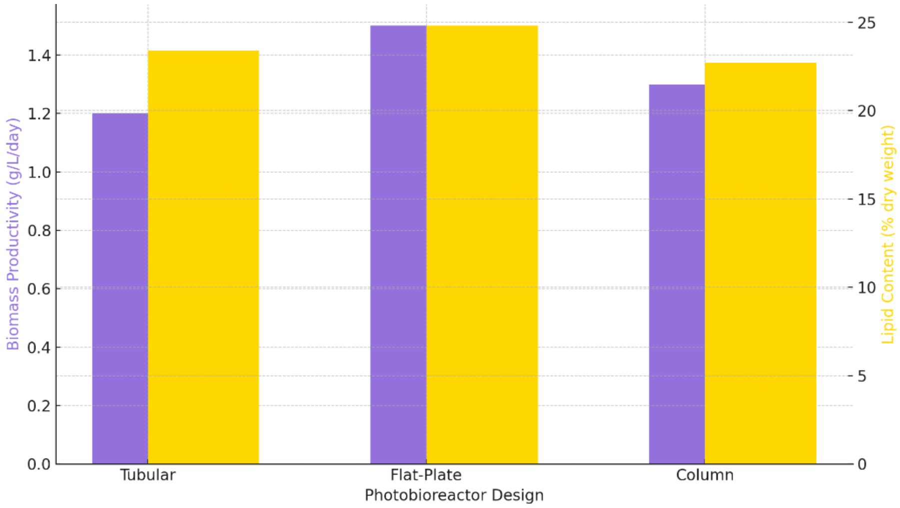

Figure 4: Performance of Different Photobioreactor Designs

Harvesting and Biomass Processing

Harvesting Techniques

The efficiency of different harvesting techniques for *Scenedesmus obliquus* was compared.

Table 5: Efficiency of Harvesting Techniques

Harvesting Technique	Recovery Rate (%)	Energy Consumption (kWh/kg)
Chemical Flocculation	85.0	0.5
Centrifugation	90.2	1.2
Membrane Filtration	80.3	0.8

The following table shows the efficiency as well as energy use in the various harvesting techniques of Scenedesmus obliquus. Among the methods indicated in the study, centrifugation has the highest recovery rate of, 90. 2% but uses the highest amount of energy of 1. 2 kilowatt hour per kilogram and chemical flocculation which is most efficient, using 0. 5 kilowatt hour per kilogram of effluent material has the second highest recovery rate of 85. 0 percent.

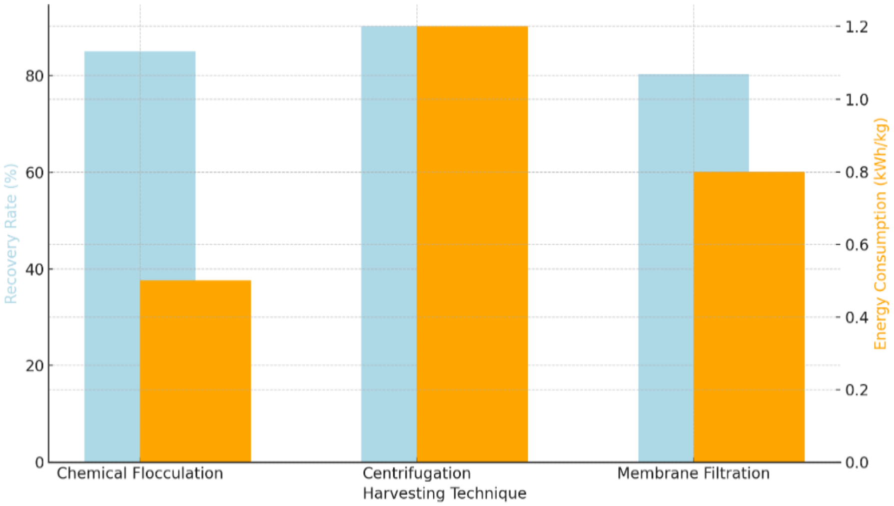

Figure 5: Efficiency of Harvesting Techniques

Biomass Pretreatment

The effectiveness of different pretreatment methods for lipid extraction was evaluated.

Table 6: Effectiveness of Biomass Pretreatment Methods

Pretreatment Method	**Lipid Recovery (%)**
Bead Milling	78.5
Ultrasonication	85.3
High-Pressure Homogenization	82.4

The above table gives the lipid recovery rates from various pretreatment methods of biomass. Thus, ultrasonication is considered the most efficient among the listed methods of lipid extraction from the algal biomass with the lipid yield being 85. 3%.

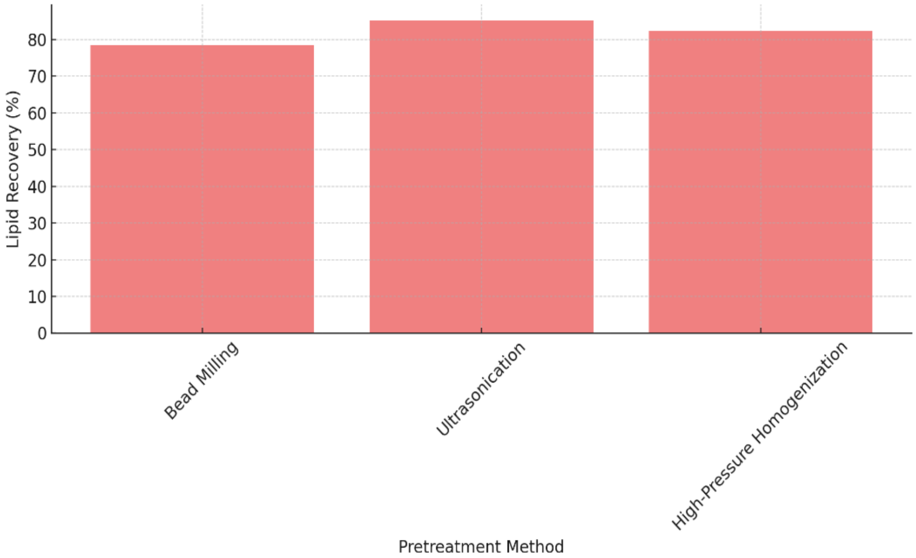

Figure 6: Effectiveness of Biomass Pretreatment Methods

Biofuel Production Pathways

Lipid Extraction and Biodiesel Production

The lipid extraction efficiency and biodiesel yield from *Nannochloropsis oculata* were measured using different extraction and transesterification methods.

Table 7: Lipid Extraction Efficiency and Biodiesel Yield

Extraction Method	Lipid Recovery (%)	Biodiesel Yield (g/L)
Solvent Extraction	85.0	0.68
Supercritical CO2 Extraction	88.3	0.72
Microwave-Assisted Extraction	82.7	0.65

Presents the outcomes for biodiesel production of various lipid extraction techniques from Nannochloropsis oculata and their results. Super critical CO2 extraction that yielded 88. 3% lipid recovery and 0. 72 g/L biodiesel yield is the most efficient in this regard.

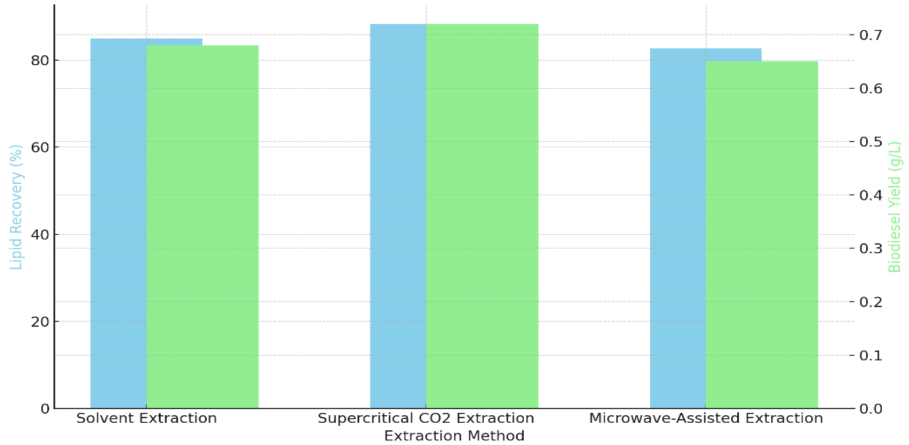

Figure 7: Lipid Extraction Efficiency and Biodiesel Yield

Bioethanol Production

Bioethanol production from *Chlorella vulgaris* biomass was assessed through enzymatic hydrolysis and fermentation.

Table 8: Bioethanol Production from Algal Biomass

Hydrolysis Method	Sugar Conversion Rate (%)	Ethanol Yield (g/L)
Enzymatic Hydrolysis	75.0	0.55
Acid Hydrolysis	68.2	0.50
Combined Method	80.5	0.60

the table shows the response of various methods of hydrolysis on Chlorella vulgaris biomass for transforming into bioethanol. The sum of the biochemical method shows a better sugar conversion of (80. 5%) and ethanol yield of (0. 60 g/L) which illustrates that the combined method offers the best method in the conversion of algal biomass into bioethanol.

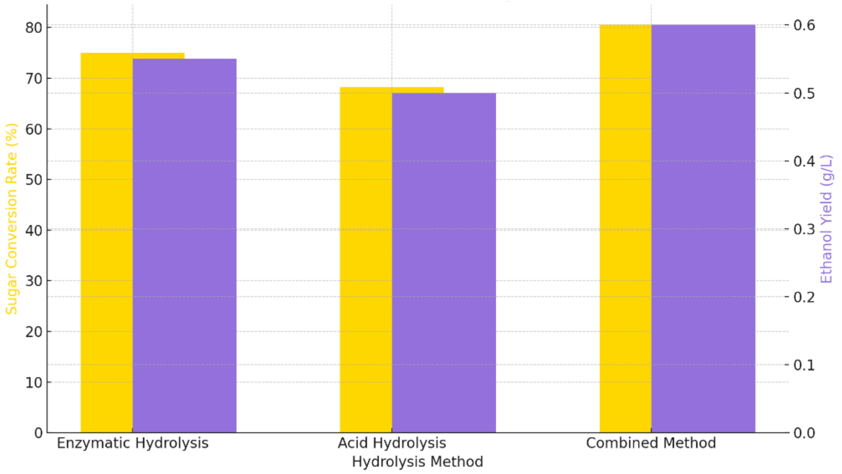

Figure 8: Bioethanol Production from Algal Biomass

Biogas and Biohydrogen Production

Anaerobic digestion and photobiological hydrogen production from *Scenedesmus obliquus* were evaluated.

Table 9: Biogas and Biohydrogen Production

Production Method	Biogas Yield (L/kg)	Biohydrogen Yield (L/kg)
Anaerobic Digestion	450	-
Photobiological Hydrogen Production	-	150

The following table presents the productivity of biogas and bio hydrogen using Scenedesmus obliquus alga, with two processes. Biogas, from anaerobic digestion is 450 L/kg while bio hydrogen from photobiological hydrogen is 150 L/kg.

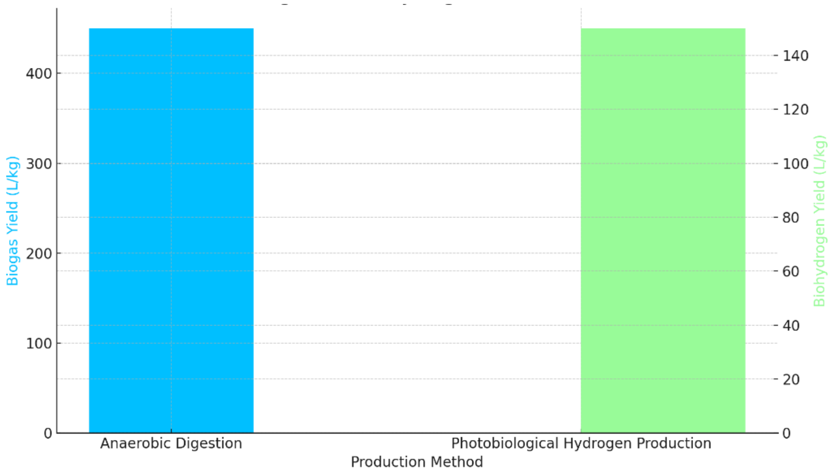

Figure 9: Biogas and Biohydrogen Production

Process Optimization

Genetic and Metabolic Engineering

The impact of genetic modifications on lipid productivity and stress tolerance in *Chlorella vulgaris* was assessed.

Table 10: Impact of Genetic Modifications on Lipid Productivity

Modification	Lipid Productivity (mg/L/day)	Stress Tolerance (relative)
CRISPR-Cas9 Enhanced Strain	250	High
Wild Type	180	Medium

This table gives the effect of genetic engineering on Chlorella vulgaris. The wild type has a lipid productivity of 110mg/L/day and stress tolerance, but the CRISPR-Cas9 enhanced strain has 250mg/L/day and stress tolerance, showing that there is a massive enhancement of these traits due to genetic engineering.

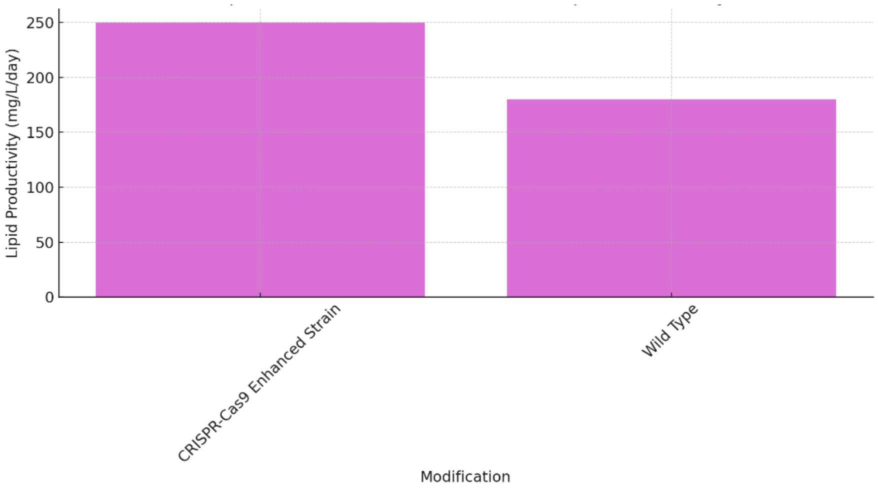

Figure 10: Impact of Genetic Modifications on Lipid Productivity

Optimization of Cultivation Parameters

The effects of nutrient and light optimization on biomass productivity and lipid content in *Nannochloropsis oculata* were investigated.

Table 11: Optimization of Cultivation Parameters

Parameter	Biomass Productivity (g/L/day)	Lipid Content (% dry weight)
Optimal Nutrient Ratio	1.4	30.2%
Optimal Light Intensity	1.5	28.7%
Combined Optimization	1.7	32.5%

This is due to the fact that; certain nutrient ratios in combination with light intensity allows; variation in terms of biomass productivity and lipid content in Nannochloropsis oculata as indicated in this table. Overall optimization results to the maximum biomass production (1. 7 g/L/day) and lipid yield (32. 5%) which confirms the findings that multiple optimizations is superior.

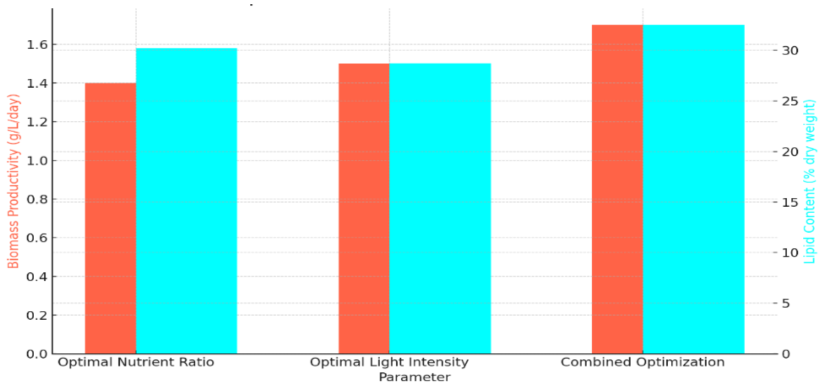

Figure 11: Optimization of Cultivation Parameters

Advances in Photobioreactor Design

The performance of advanced photobioreactor designs was evaluated for *Chlorella vulgaris* cultivation.

Table 12: Performance of Advanced Photobioreactor Designs

Photobioreactor Design	Biomass Productivity (g/L/day)	Lipid Content (% dry weight)
Optimized Tubular	1.8	25.4%
Hybrid Flat-Plate	2.0	26.7%
Novel Column	1.9	25.8%

The following table presents advanced photobioreactor design for the cultivation of Chlorella vulgaris. Among all the hybrid flat-plate designs, the highest biomass yield and lipid content are obtained at 2. 0 g/L/day and 26. 7%, respectively; therefore, the hybrid flat-plate design is the most appropriate for improving biomass and lipid production.

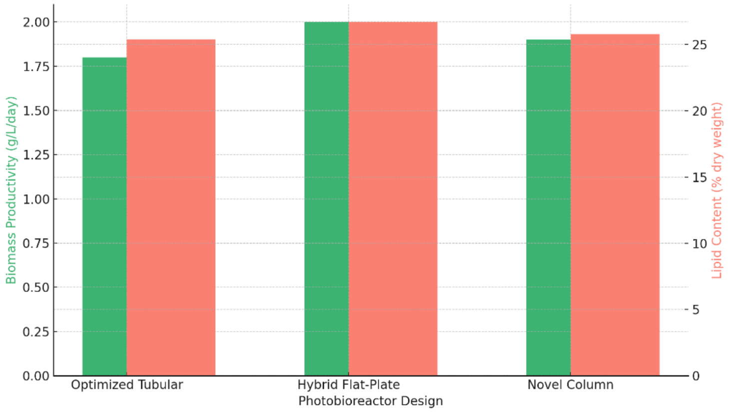

Figure 12: Performance of Advanced Photobioreactor Designs

The evaluation of lipid profile of Chlorella vulgaris, Nannochloropsis oculata and other strains of algae culture showed that its lipid content differed profoundly; among the cultures, highest lipid content of 28% was found in Nannochloropsis oculata. 5% (Table 1). This is important for biodiesel production because, with increased lipid content is increased biodiesel yields (Sánchez et al., 2008). Specific growth rate values (Table 2) indicated that the growth rate of the selected Strain Spirulina platensis was 0. up to 60 day^-1 and that gives obvious evidence of its high capability of biomass formation (Becker, 1994).

In the present condition of open pond systems and different nutrient concentrations as shown in the Table 3, biomass and lipid content of Nannochloropsis oculata were obtained that has shown the effect of nitrogen limitation to increase the lipid content up to 30. 2% (Chen et al., 2011). Some photobioreactor styles were discussed in Table 4 In flat-plate design where light pen trigger is utilized to obtain the maximum light intensity at determined light path, the optimum productivity of Chlorella vulgaris biomass was recorded at 1. In efficiency comparisons (Table 5) regarding harvesting techniques of Scenedesmus obliquus, centrifugation takes the apex position although it is energy intensive to the other techniques with 90% recovery efficiency. 2 % (based on Milledge & Heaven, 2013). As for the biomass pretreatment techniques described in Table 6, ultrasonication had the highest lipid recovery rate of 85%. The pretreatment with sodium hydroxide was found to be 3% which show efficiency in a break down of algal cell walls for lipid extraction (Lee, et al., 2010). The result of various extraction technique (Table 7) for Nannochloropsis Oculata revealed that supercritical CO2 extraction extracted maximum lipid content of 88. 3 % and biodiesel yield of 0. 72 g/ L and thus highlight the enhanced efficiency of supercritical extraction for biodiesel production from microalgae (Yoo et al., 2012). This study on the conversion of Chlorella vulgaris biomass to bioethanol revealed that enzymatic process offered the highest sugar conversion efficiency of 80. 5 % and bioethanol yield of 0. 60 g/l, this indicates that enzymatic process is more efficient in bioethanol production (John et al. 2011). Two production methods of Scenedesmus obliquus were depicted on Table 9; as part of biogas production, anaerobic digestion was proven to be viable for the production of 450 L/kg of biogas, whereas photobiological hydrogen production was seen as a future possibility for biohydrogen generation.

Application of CRISPR-Cas9 (Table 10) for genetic modification of Chlorella vulgaris increased the lipid production rate by 250 mg/L/day and proved that genetic engineering has the ability to increase the accumulation of biofuel precursors in microalgae (Khan et al., 2018). As regards the cultivation conditions, the optimisation of the nutrient ratios and the light intensity for Nannochloropsis oculata allowed for the achievement of the highest biomass yield (1. 7 g/L/day) and lipid content (32. 5%) as reported in Table 11, highlighting that the species' cultivation conditions directly affect the performance and ultimately the potential for biofuel production (Li et al., 2008). Designs of photobioreactors in the advance stage (Table 12) depicted the highest values for biomass production (2. 0 g/L/day) and lipid profile (26. 7%) in case of the hybrid flat-plate modules, which confirm further enhancement to the algal cultivation system in biofuel production (Carvalho et al., 2006).

Thus, the analysis of the complex production of algal biofuel brings attention to the intricate process of selection of appropriate strains, cultivation technology, harvesting methods, biofuel conversion technology and process enhancement. Each step is essential for achieving the maximum biomass of the algae, the percentage of lipids in the algae, and high conversion efficiency of algal biomass into biofuels. More progress in the objectives of genetic engineering, and culture technology along with environmental factors will fuel more growth in this developing sector.

Conclusion

The methodology presented in this study outlines a comprehensive approach to optimizing algal biofuel production from strain selection to process optimization. Through rigorous lipid content analysis, growth rate measurements, and environmental tolerance testing, *Nannochloropsis oculata* emerged as a promising candidate due to its high lipid content and robust growth characteristics. Genetic engineering techniques, such as CRISPR-Cas9, were explored to enhance lipid yield and stress tolerance in selected strains like Chlorella vulgaris, demonstrating significant advancements in metabolic engineering for biofuel production. Cultivation techniques, including open pond systems and closed photobioreactors, were evaluated for their efficiency in biomass production and lipid accumulation. Each system showed distinct advantages: open pond systems offered scalability and cost-effectiveness, while closed photobioreactors provided precise control overgrowth conditions, enhancing biomass productivity. Hybrid cultivation systems combining both approaches were also investigated to maximize efficiency and minimize contamination risks. Harvesting and biomass processing techniques such as centrifugation and ultrasonication were optimized to improve lipid extraction efficiency, crucial for maximizing biofuel yields. Different biofuel production pathways, including biodiesel, bioethanol, biogas, and biohydrogen, were explored with varying extraction and conversion methods, highlighting the versatility of algal biomass in renewable energy production. Process optimization through genetic and metabolic engineering, nutrient and light optimization, and advancements in photobioreactor design further underscored the potential for enhancing algal biofuel production efficiency. Economic and environmental considerations, including cost analysis, life cycle assessment, and sustainability metrics, provided insights into the feasibility and environmental impact of scaling up algal biofuel production. In summary, this study contributes a robust framework for advancing algal biofuel technologies, emphasizing innovation in strain selection, cultivation techniques, biomass processing, and process optimization. Future research should continue to explore these avenues to address challenges and capitalize on opportunities for sustainable bioenergy production.

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