

## Process Study on Microbial Conversion of Kitchen Waste into Biodiesel

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Journal of Energy Bioscience, 2024, Vol.15, No.5 doi: [10.5376/jeb.2024.15.0027](https://doi.org/10.5376/jeb.2024.15.0027)

Received: 19 Jul., 2024

Accepted: 29 Aug., 2024

Published: 17 Sep., 2024

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### Preferred citation for this article:

Wang J.J., Lin Q., and Zhou W., 2024, Process study on microbial conversion of kitchen waste into biodiesel, Journal of Energy Bioscience, 15(5): 289-300 (doi: [10.5376/jeb.2024.15.0027](https://doi.org/10.5376/jeb.2024.15.0027))

**Abstract** This study explores the microbial process of converting kitchen waste into biodiesel, with a focus on identifying efficient microbial strains and optimizing the conversion process. The study identified several key findings. First, the filamentous fungi *Mortierella isabellina* NRRL 1757 demonstrated high lipid productivity and versatility when grown on various waste substrates, including glycerol, orange peel extract, and ricotta cheese whey, with lipid productivities of 0.46, 1.24, and 0.91 g/(L d), respectively. Additionally, the fatty acid profile of the produced lipids was highly compatible with biodiesel production, similar to commonly used palm and Jatropha oils. Another significant discovery was the use of the algae strain *Golenkinia* sp. SDEC-16, which showed the highest power density, biomass concentration, and total lipid content when used in microbial fuel cells with kitchen waste anaerobically digested effluent, achieving a lipid content of 38%. Furthermore, the bacterium *Klebsiella variicola* TB-83 was found to produce ethanol efficiently from biodiesel-derived glycerol under alkaline conditions, with a maximum ethanol production of 9.8 g/L. The findings of this study suggest that microbial conversion of kitchen waste into biodiesel is a viable and sustainable approach. The identified microbial strains, particularly *Mortierella isabellina* NRRL 1757 and *Golenkinia* sp. SDEC-16, show great potential for high lipid production, making them suitable candidates for biodiesel manufacturing. Additionally, the ability of *Klebsiella variicola* TB-83 to produce ethanol from biodiesel waste further supports the feasibility of integrating waste-to-energy processes.

**Keywords** Microbial conversion; Kitchen waste; Biodiesel; *Mortierella isabellina*; *Golenkinia* sp.; *Klebsiella variicola*; Lipid production; Ethanol production

## 1 Introduction

Kitchen waste, a significant component of municipal solid waste, is rich in organic materials that can be repurposed for various applications. The valorization of kitchen waste not only addresses waste management issues but also provides a renewable resource for biofuel production. The organic content in kitchen waste, such as carbohydrates, proteins, and lipids, makes it an ideal substrate for microbial processes aimed at producing biodiesel (Cheirsilp and Louhasakul, 2013; Carmona-Cabello et al., 2020; Zhang et al., 2020).

The global increase in waste generation poses substantial challenges for waste management systems. Traditional disposal methods, such as landfilling and incineration, contribute to environmental pollution and greenhouse gas emissions. Concurrently, the reliance on fossil fuels for energy production exacerbates climate change and depletes finite resources. Integrating waste management with renewable energy production, such as converting kitchen waste into biodiesel, offers a sustainable solution to these intertwined challenges (Almeida et al., 2012; Zheng et al., 2012; Li et al., 2013).

Biodiesel is a biodegradable, renewable fuel that can be used in conventional diesel engines with minimal modifications. It offers several environmental benefits, including reduced emissions of carbon monoxide, particulate matter, and unburned hydrocarbons. Biodiesel production from waste materials, such as kitchen waste, further enhances its sustainability by utilizing low-cost feedstocks and reducing waste disposal issues. The fatty acid profiles of microbial lipids derived from waste are comparable to those of traditional plant oils, making them suitable for biodiesel production (Cheirsilp and Louhasakul, 2013; Srivastava, 2019; Carmona-Cabello et al., 2020).

This study aims to explore the microbial processes involved in the conversion of kitchen waste into biodiesel, with a focus on analyzing the processes and their potential advantages. The research will examine the efficiency of different microbial strains in lipid accumulation and analyze the transesterification process of these lipids into biodiesel, striving to contribute to the development of sustainable waste-to-energy technologies. The scope of the study includes a review of existing methods, experimental results, and a discussion on the scalability and industrial applicability of the proposed process.

## 2 Characteristics of Kitchen Waste

### 2.1 Composition of kitchen waste

Kitchen waste (KW) is a significant component of municipal solid waste, characterized by high moisture content, organic matter, and variability in its chemical composition. Studies have shown that KW typically contains a mix of food residues, including fats, oils, and grease (FOG), proteins, carbohydrates, and minerals. For instance, household kitchen waste (HH) and kitchen waste from Chinese restaurants (CR) have been found to have higher crude protein content (26%) and considerable amounts of minerals, making them nutritionally suitable for various recycling processes (Ho and Chu, 2018). Additionally, the lipid content in waste cooking oils (WCOs) ranges from 73% to 84.5%, with significant amounts of saturated and unsaturated fatty acids (Sharma et al., 2021).

### 2.2 Suitability of kitchen waste for biodiesel production

Kitchen waste, particularly waste cooking oils and FOG, is highly suitable for biodiesel production due to its high lipid content and the presence of various fatty acids. The diverse nature of fatty acids in WCOs, such as C16 and C18, makes them ideal candidates for biodiesel production, complying with international standards (Sharma et al., 2021) (Figure 1). Moreover, FOG-derived biodiesel has shown better characteristics concerning oxidative stability, flash point, cetane number, and total emissions compared to other feedstocks (Abomohra et al., 2020). The economic analysis also supports the use of FOG as a cost-effective alternative to conventional biodiesel feedstocks.

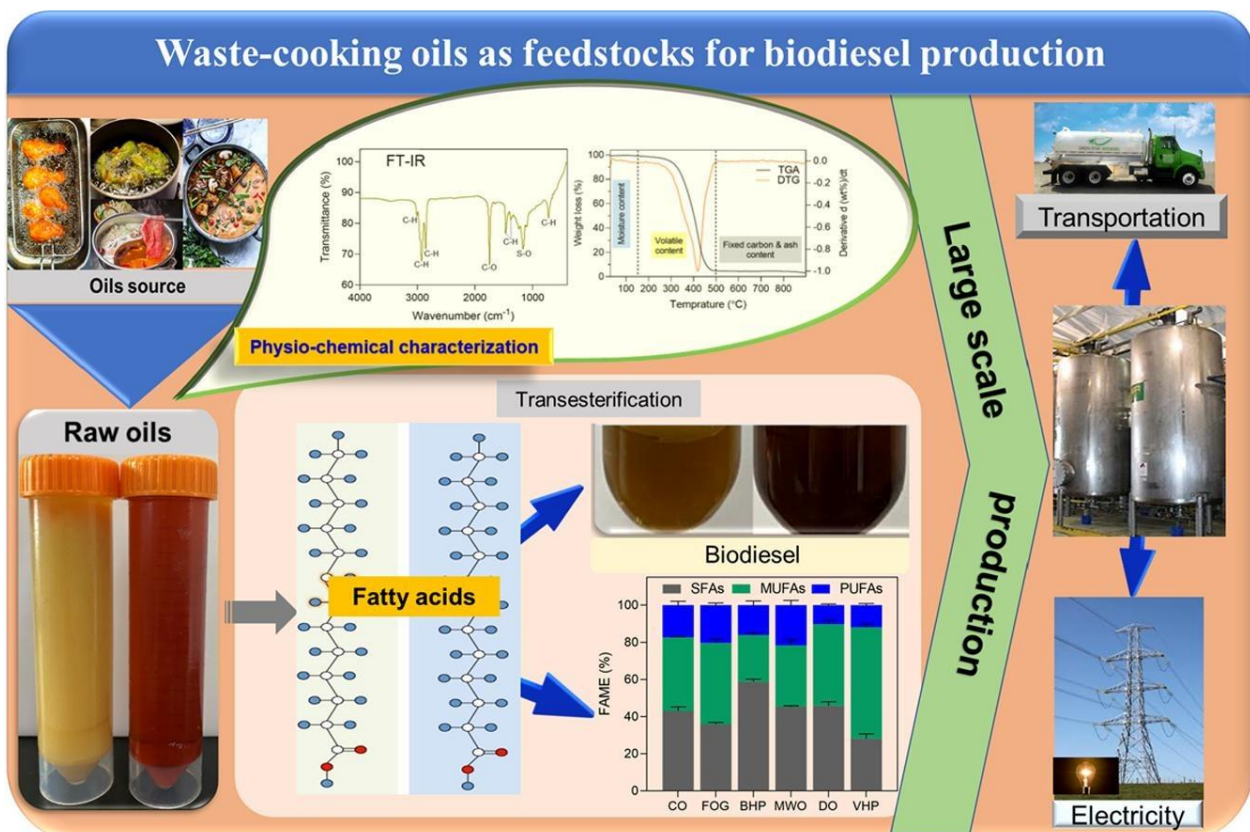


Figure 1 Waste cooking oils as feedstocks for biodiesel production: characterization and conversion process (Adapted from Sharma et al., 2021)

### 2.3 Variability in waste components (fats, oils, and grease content)

The composition of kitchen waste, particularly the FOG content, can vary significantly depending on the source and type of waste. For example, post-consumption food wastes have higher dry matter (>26%) and fat content (>13%), making them more reliable feedstocks for biodiesel production due to their lower temporal variability (Ho and Chu, 2018). The levels of free fatty acids (FFAs) and moisture in waste cooking greases can also vary widely, with FFAs ranging from 0.7% to 41.8% and moisture from 0.01% to 55.38% (Çanakçı, 2007). This variability necessitates efficient pretreatment methods to optimize the conversion of these wastes into biodiesel (Çanakçı, 2007; Abomohra et al., 2020).

## 3 Microbial Conversion Process Overview

### 3.1 Introduction to microbial fermentation in biodiesel production

Microbial fermentation is a pivotal process in the production of biodiesel, particularly when utilizing waste materials as feedstock. This biological method leverages the metabolic capabilities of microorganisms to convert organic substrates into valuable biofuels. Unlike traditional chemical methods, microbial fermentation can operate under milder conditions and can process a variety of feedstocks, including lignocellulosic biomass and industrial organic wastes (Zhang et al., 2020; Adegboye et al., 2021). The process typically involves the breakdown of complex organic materials into simpler compounds, which are then converted into lipids by oleaginous microorganisms. These lipids can subsequently be transesterified into biodiesel (Wahlen et al., 2011; Carmona-Cabello et al., 2020).

### 3.2 Types of microorganisms involved in the process

A diverse array of microorganisms is employed in the microbial fermentation process for biodiesel production. These include bacteria, yeasts, and microalgae, each contributing uniquely to the conversion of substrates into lipids. For instance, oleaginous yeasts like *Rhodospiridium toruloides* are known for their high lipid accumulation capabilities (Carmona-Cabello et al., 2020). Bacterial species such as those from the families Clostridiaceae and Ruminococcaceae play significant roles in the anaerobic fermentation process, aiding in the efficient breakdown of complex organic materials (Martínez-Burgos et al., 2020). Microalgae, such as *Nannochloropsis* sp., are also utilized due to their high lipid productivity and ability to grow on non-arable land (Wahlen et al., 2011; Shi et al., 2021).

### 3.3 Metabolic pathways for lipid production from waste

The metabolic pathways involved in lipid production from waste materials are complex and involve several key steps (Figure 2). Initially, the organic waste is hydrolyzed into simpler sugars and fatty acids through enzymatic actions. These simpler compounds are then taken up by microorganisms and funneled into metabolic pathways such as glycolysis and the tricarboxylic acid (TCA) cycle. The intermediates from these pathways are subsequently directed towards lipid biosynthesis. Metabolic engineering can further enhance these pathways to increase lipid yield and productivity (Zhang et al., 2020; Adegboye et al., 2021). For example, genetic modifications can be made to increase the flux through the acetyl-CoA pathway, a critical precursor for lipid biosynthesis (Adegboye et al., 2021).

### 3.4 Advantages of Using Microbes Over Conventional Methods

Using microbes for biodiesel production offers several advantages over conventional chemical methods. Firstly, microbial processes can utilize a wide range of feedstocks, including waste materials, which reduces the overall cost and environmental impact (Carmona-Cabello et al., 2020; Zhang et al., 2020). Secondly, microbial fermentation operates under milder conditions, which can lower energy requirements and improve safety (Adegboye et al., 2021; Nanda et al., 2023). Additionally, the use of engineered microorganisms can lead to higher yields and more efficient conversion processes (Adegboye et al., 2021). Microbial methods also allow for the production of biodiesel with favorable properties, such as a high content of saturated fatty acids, which can enhance fuel stability and performance (Wahlen et al., 2011; Shi et al., 2021). Finally, the integration of microbial processes into a biorefinery approach can facilitate the production of multiple biofuels and biochemicals,





pre-treatment has been found to be more effective than acid and alkaline treatments in increasing biohydrogen generation from kitchen waste (Jais et al., 2023). Similarly, the combination of physical and chemical methods, such as ultrasonication with hydrogen peroxide, has demonstrated superior results in enhancing hydrogen production from organic wastes (Salem et al., 2018). The optimization process also involves adjusting the volatile solid concentrations and mixing ratios of different waste types to achieve the highest hydrogen production rates (Nam, 2023).

#### 4.3 Challenges in scaling pre-treatment processes

While pre-treatment methods have shown promising results at the laboratory scale, several challenges remain in scaling these processes for industrial applications. One of the primary challenges is the economic feasibility of the pre-treatment methods. For instance, although freeze-thaw pre-treatment has been identified as the most profitable process, the costs associated with maintaining low temperatures can be prohibitive (Ma et al., 2011). Additionally, the energy requirements for thermal and chemical pre-treatments can be substantial, potentially offsetting the benefits of increased biohydrogen yields (Jeyakumar et al., 2020). Another challenge is the variability in the composition of kitchen waste, which can affect the consistency and efficiency of the pre-treatment process (Pagliaccia et al., 2019). Addressing these challenges requires further research into cost-effective and energy-efficient pre-treatment methods, as well as the development of robust systems that can handle the heterogeneity of kitchen waste.

### 5 Lipid Accumulation by Microorganisms

#### 5.1 Microbial strains used for lipid accumulation (bacteria, yeast, fungi)

Various microbial strains, including bacteria, yeast, and fungi, have been explored for their lipid accumulation capabilities. For instance, the oleaginous yeast *Yarrowia lipolytica* has been extensively engineered to enhance lipid production, achieving lipid contents as high as 61.7% in bioreactor fermentations (Tai and Stephanopoulos, 2013). Similarly, the yeast *Rhodospiridium toruloides* has shown significant lipid accumulation when cultivated in bioethanol wastewater, reaching a lipid content of 34.9% (Zhou et al., 2013). Bacterial strains such as *Bacillus cereus* have also been used in co-culture systems with yeast to optimize lipid production and wastewater treatment (Karim et al., 2021).

#### 5.2 Mechanisms of lipid biosynthesis

Lipid biosynthesis in microorganisms involves several key enzymatic steps. In *Yarrowia lipolytica*, overexpression of diacylglycerol acyltransferase (DGA1) and acetyl-CoA carboxylase (ACC1) has been shown to significantly enhance lipid production by driving the synthesis of triglycerides (TAGs) and fatty acids (Tai and Stephanopoulos, 2013; Qiao et al., 2015). The process involves the conversion of acetyl-CoA to malonyl-CoA by ACC1, followed by a series of reactions leading to the formation of fatty acids, which are then esterified to glycerol to form TAGs. In *Schizochytrium* sp., adaptive laboratory evolution (ALE) has been used to enhance lipid biosynthesis under stress conditions, improving both lipid yield and antioxidant defenses (Sun et al., 2018).

#### 5.3 Process parameters affecting lipid yield (temperature, pH, etc.)

Several process parameters significantly influence lipid yield in microbial cultures. Temperature and pH are critical factors; for example, optimal lipid accumulation in a co-culture of *Lipomyces starkeyi* and *Bacillus cereus* was achieved at a pH of 6.5 and a temperature of 32.5 °C (Karim et al., 2021). In *Schizochytrium* sp., low temperature and high salinity were used to enhance DHA content and lipid accumulation while preventing lipid peroxidation (Sun et al., 2018). Additionally, nutrient availability, such as carbon and nitrogen sources, plays a crucial role. Feeding strategies, such as the addition of glucose during the cultivation of *Rhodospiridium toruloides*, have been shown to significantly increase both biomass and lipid production (Zhou et al., 2013).

#### 5.4 Case studies of high lipid-producing microbes

Several case studies highlight the potential of high lipid-producing microbes. In one study, *Yarrowia lipolytica* was engineered to overexpress key enzymes in the lipid biosynthesis pathway, resulting in a lipid content of 41.4% in a 2-L bioreactor fermentation (Tai and Stephanopoulos, 2013). Another study demonstrated that

*Schizochytrium* sp. could achieve a DHA yield of 38.12 g/L under optimized conditions using a cooperative two-factor ALE strategy (Sun et al., 2018). Additionally, the oleaginous yeast *Rhodospiridium toruloides* was able to produce 3.8 g/L of biomass with a lipid content of 34.9% when cultivated in bioethanol wastewater (Zhou et al., 2013). These examples underscore the effectiveness of genetic and process optimization strategies in enhancing lipid production for biofuel applications.

## 6 Transesterification of Microbial Lipids into Biodiesel

### 6.1 Chemical process of transesterification

Transesterification is a chemical reaction where triglycerides react with alcohol (usually methanol or ethanol) in the presence of a catalyst to form fatty acid methyl esters (FAMES), commonly known as biodiesel, and glycerol as a byproduct. This process can be carried out using various catalysts, including acid, base, and enzyme catalysts. For instance, the use of Fe<sub>2</sub>O<sub>3</sub> nanocatalysts has shown to improve biodiesel yield significantly when compared to conventional acid (HCl) and base (NaOH) catalysts, achieving up to 86% yield under optimized conditions (Banerjee et al., 2019). Similarly, homogeneous acid catalysis using H<sub>2</sub>SO<sub>4</sub> has been found effective, with a maximum methyl ester yield of 60% (Mathimani et al., 2015).

### 6.2 Role of catalysts in biodiesel conversion

Catalysts play a crucial role in the transesterification process by lowering the activation energy and increasing the reaction rate. Different types of catalysts, such as homogeneous (acid and base) and heterogeneous (solid acid and base), have been explored. For example, graphene oxide (GO) as a solid acid catalyst has demonstrated high efficiency, achieving a lipids conversion efficiency into FAMES of 95.1% in microwave-assisted transesterification reactions (Cheng et al., 2016). Additionally, enzyme catalysts like immobilized lipase have been used, providing a biodiesel yield that is seven times higher compared to alkaline-based transesterification (Teo et al., 2014).

### 6.3 Yield optimization strategies for biodiesel production

Optimizing the yield of biodiesel involves fine-tuning various parameters such as the methanol-to-lipid ratio, catalyst concentration, reaction temperature, and time. Response Surface Methodology (RSM) has been employed to optimize these parameters, achieving a maximum biodiesel yield of 89.583% with H<sub>2</sub>SO<sub>4</sub> catalyst under specific conditions (Chamola et al., 2019). Concurrent extraction and reaction processes have also been developed to enhance yield and reduce energy consumption, achieving yields higher than 90 wt.% (Im et al., 2014). Supercritical methanolysis is another effective method, with yields reaching up to 99.32% under optimized conditions (Shirazi et al., 2017).

### 6.4 Challenges in ensuring biodiesel quality

Ensuring the quality of biodiesel involves addressing several challenges, including the removal of impurities, achieving the desired fatty acid profile, and meeting international standards such as ASTM and European norms. For instance, the biodiesel produced from *Chlorella* sp. BDUG 91771 was characterized to have a suitable Degree of Unsaturation (DU), Long Chain Saturated Factor (LCSF), and Cold Filter Plugging Point (CFPP), aligning with prescribed standards (Mathimani et al., 2015). Additionally, the supercritical transesterification process has been shown to improve the quality of biodiesel by reducing the proportion of polyunsaturated fatty acids (Jazzar et al., 2015). However, maintaining consistent quality across different batches and feedstocks remains a significant challenge.

## 7 Process Efficiency and Economic Feasibility

### 7.1 Energy balance and process efficiency in microbial conversion

The energy balance and process efficiency of biohydrogen production from marine algae are critical factors in determining the viability of this biofuel. Algae, particularly macroalgae, are considered efficient sources of biomass for biohydrogen production due to their high-energy yield and sustainable nature (Kumar et al., 2021). The pretreatment of algae is essential to enhance the hydrolytic process during dark fermentation, although it can lead to the formation of inhibitory substances that need to be controlled through detoxification techniques.

Additionally, the energy conversion characteristics and environmental impacts of biohythane production via two-stage anaerobic fermentation from microalgae and food waste have been evaluated, showing a net energy input to output ratio of 0.24, indicating a favorable energy balance (Sun et al., 2019).

## 7.2 Cost analysis of biodiesel production from kitchen waste

The economic feasibility of biodiesel production from kitchen waste has been explored through various studies. For instance, the use of microalgae in biodiesel production has shown that while the process is technically feasible, the high costs associated with cultivation, harvesting, and extraction remain significant barriers (Anto et al., 2020; Branco-Vieira et al., 2020; Li and Zhou, 2024). The production cost for microalgae biomass is estimated at 2.01 €/kg, and for biodiesel, it is 0.33 €/L, with a return on investment (ROI) of 10% and a payback time of 10 years (Branco-Vieira et al., 2020). Despite these costs, the use of kitchen waste as a substrate for biodiesel production can potentially reduce overall expenses and improve economic viability (Hou et al., 2016).

## 7.3 Potential for commercial scalability

The potential for commercial scalability of biohydrogen production from marine algae is promising but faces several challenges. The large-scale production of algal biomass for biofuel applications is not yet widespread due to the complexities in balancing ecological and economic concerns (Dębowski et al., 2020). However, advancements in photobioreactor technologies and the integration of microalgae-based H<sub>2</sub> production processes offer potential routes for commercialization (Goswami et al., 2020). The scalability of these systems depends on overcoming the high production costs and improving the efficiency of biomass conversion processes (Dębowski et al., 2020; Goswami et al., 2020).

## 7.4 Comparison with other biodiesel feedstocks

When comparing marine algae to other biodiesel feedstocks, several factors come into play. Algae are considered a third-generation biofuel feedstock with high productivity and lipid yields, making them attractive for biodiesel production (Williams and Laurens, 2010). However, the cost of producing biodiesel from algae remains higher compared to traditional feedstocks like plant oils and fish waste (Anto et al., 2020; Prasanna et al., 2023). Fish waste, for example, is a significant source of biodiesel and offers a cost-effective alternative due to its abundance and the environmental benefits of waste management (Prasanna et al., 2023). Additionally, the biochemical composition of algal biomass influences the economics of biodiesel production, with higher lipid content reducing the availability of other valuable compounds (Williams and Laurens, 2010).

# 8 Environmental and Societal Benefits

## 8.1 Reduction in kitchen waste volume through microbial conversion

The utilization of kitchen waste for biohydrogen production through microbial conversion presents a significant opportunity for waste reduction. Studies have demonstrated that specific algae species, such as *Golenkinia* sp. SDEC-16, can effectively treat kitchen waste anaerobically digested effluent (KWADE), leading to substantial reductions in chemical oxygen demand (COD) and total nitrogen (TN) levels. This process not only mitigates the volume of kitchen waste but also enhances the efficiency of bioelectricity and lipid production, thereby contributing to a more sustainable waste management system (Hou et al., 2016).

## 8.2 Carbon footprint and lifecycle analysis of biodiesel from waste

The production of biodiesel from biological waste, including fish waste, offers a promising alternative to traditional fossil fuels. The lifecycle analysis of biodiesel derived from fish waste indicates a lower carbon footprint compared to conventional diesel. This is primarily due to the utilization of waste materials that would otherwise contribute to environmental pollution. The conversion of fish waste into biodiesel not only addresses waste disposal issues but also reduces greenhouse gas emissions, making it a more environmentally friendly option (Prasanna et al., 2023). Additionally, biohydrogen production from green algae and cyanobacteria has been identified as a clean energy source, further reducing the carbon footprint associated with hydrogen production from fossil fuels (Mona et al., 2020).

### 8.3 Socio-economic impacts: waste management, energy independence

The socio-economic impacts of biohydrogen production from marine algae are multifaceted. Effective waste management practices, such as the conversion of kitchen and fish waste into biofuels, can lead to significant economic benefits. These include reduced waste disposal costs and the creation of new revenue streams from biofuel production. Moreover, the development of biohydrogen and biodiesel from waste materials can enhance energy independence by providing a renewable and sustainable energy source. This shift towards renewable energy can reduce reliance on imported fossil fuels, thereby improving national energy security and fostering economic stability (Hou et al., 2016; Mona et al., 2020; Prasanna et al., 2023). Furthermore, the deployment of microalgal cultivation technologies in industrial and domestic settings can optimize the use of non-arable land and waste resources, contributing to a more sustainable and resilient energy infrastructure (Dębowski et al., 2020).

## 9 Challenges and Future Prospects

### 9.1 Technical challenges in microbial conversion and biodiesel synthesis

The production of biodiesel from marine algae faces several technical challenges that need to be addressed to improve efficiency and commercial viability. One of the primary issues is the complexity of lipid extraction and conversion processes. Traditional methods involve multiple steps, including the use of organic solvents for lipid extraction followed by transesterification to produce biodiesel. This multi-step process is not only time-consuming but also costly and environmentally unfriendly (Wahlen et al., 2011; Anto et al., 2020).

Moreover, the optimization of reaction conditions for simultaneous extraction and conversion of lipids from microalgae is still under research. Recent studies have shown that it is possible to achieve high yields of biodiesel through optimized single-step processes, but these methods need further refinement and scaling up (Wahlen et al., 2011). Additionally, the genetic and metabolic engineering of microalgae to enhance lipid productivity and growth rates is another area that requires significant research and development (Zhang et al., 2020).

### 9.2 Research gaps and future directions

Despite the progress made in the field, several research gaps remain. One major gap is the lack of comprehensive understanding of the metabolic pathways involved in lipid accumulation and conversion in microalgae. This knowledge is crucial for the genetic engineering of algal strains with higher lipid content and faster growth rates (Show et al., 2019).

Another area that needs attention is the development of cost-effective and efficient cultivation systems. While photobioreactors and open pond systems have been extensively studied, there is still a need for innovative designs that can reduce contamination risks and improve biomass productivity (Shen, 2014; Anto et al., 2020). The integration of algae cultivation with wastewater treatment and CO<sub>2</sub> bio-fixation offers a promising approach to make the process more sustainable and economically viable (Shen, 2014; Mohan et al., 2015).

Future research should also focus on the development of advanced pretreatment methods for lipid extraction. Techniques such as hydrothermal processing, microwave-assisted extraction, and the use of supercritical solvents have shown potential but require further optimization and scaling up (Anto et al., 2020).

### 9.3 Policy and regulatory considerations for waste-to-biodiesel initiatives

The successful implementation of waste-to-biodiesel initiatives depends not only on technological advancements but also on supportive policy and regulatory frameworks. Governments need to provide incentives for research and development in this field, as well as subsidies for the initial setup of biodiesel production facilities (Prasanna et al., 2023).

Regulations should also be put in place to ensure the sustainable sourcing of raw materials, such as marine algae, and to promote the use of waste materials for biodiesel production. This can help in reducing the environmental impact and making the process more economically viable (Mohan et al., 2015; Prasanna et al., 2023).



Furthermore, international collaboration and standardization of biodiesel production processes can facilitate the sharing of knowledge and technologies, thereby accelerating the development and commercialization of algal biodiesel (Demirbaş, 2010).

## 10 Concluding Remarks

The research on biohydrogen production using marine algae has demonstrated significant potential for sustainable energy solutions. Key insights from the studies include the high efficiency of carbon conversion in marine algae-bacteria consortia, which can reach up to 6.3% under optimal conditions. The integration of microbial cell factories in algal biorefineries has shown promise in producing various value-added products, including biohydrogen, due to the carbohydrate-rich and lignin-lacking properties of algae. Additionally, the optimization of fermentative hydrogen production through various pretreatment methods and the use of mixed microbial cultures has been highlighted as crucial for enhancing biohydrogen yields.

Microbial conversion plays a pivotal role in the sustainable production of biohydrogen. The use of mixed microbial cultures, as opposed to pure cultures, has been shown to improve the efficiency of hydrogen production by providing a diverse range of metabolic pathways for the decomposition and hydrogenation of biomass. The interactions within these microbial communities are essential for maintaining ecosystem functionality and optimizing the bioprocesses involved in biohydrogen production. Furthermore, the use of microbial consortia in anaerobic digestion processes has been effective in recycling nutrients and reducing environmental impacts, thereby supporting the sustainable growth of microalgae and enhancing overall biohydrogen production.

While the primary focus of this study is on biohydrogen production, the insights gained can also be applied to the broader context of biodiesel production from kitchen waste. The principles of microbial conversion and optimization of fermentation processes are equally relevant to biodiesel production. The use of waste substrates, such as kitchen waste, for microbial conversion into biodiesel can significantly reduce feedstock costs and environmental impacts. The successful application of microbial consortia in biohydrogen production suggests that similar strategies could be employed to enhance the efficiency and sustainability of biodiesel production from kitchen waste. The integration of these processes into a circular bioeconomy framework could further promote the adoption of biodiesel as a viable alternative to fossil fuels.

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## Acknowledgments

We would like to extend our special thanks to our colleagues, especially Rain Wang and Sheengh Yu. Your guidance and collaboration have not only enhanced the quality of our research but also helped us take a significant step forward in our academic journey.

## Conflict of Interest Disclosure

The authors affirm that this research was conducted without any commercial or financial relationships that could be construed as a potential conflict of interest.

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