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EXTRAÇÃO DE BIOPRODUTOS A PARTIR DE MICROALGAS: METODOLOGIAS E ESTRATÉGIAS DE SCALE-UP

EXTRACTION OF BIOPRODUCTS FROM MICROALGAE: A REVIEW OF METHODOLOGIES AND SCALE-UP STRATEGIES

EXTRACCIÓN DE BIOPRODUCTOS DE MICROALGAS: METODOLOGÍAS Y ESTRATEGIAS DE AMPLIACIÓN

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Resumo

O presente estudo teve como objetivo revisar metodologias de extração de bioprodutos a partir de microalgas, com ênfase em alternativas sustentáveis e estratégias de escalonamento para aplicação industrial. Foi realizada uma revisão integrativa da literatura em bases de dados científicas nacionais e internacionais, priorizando publicações a partir de 2021. As microalgas destacam-se pelo elevado potencial biotecnológico, gerando compostos de alto valor agregado, como pigmentos, ácidos graxos poli-insaturados e polissacarídeos, além de apresentarem viabilidade em processos de tratamento de efluentes. Entre as metodologias analisadas, destacam-se a extração assistida por micro-ondas (MAE), a por enzimas (EAE) e a por ultrassom (UAE), associadas ao uso de solventes verdes, como alternativas mais eficientes e ambientalmente seguras. Foram discutidos parâmetros críticos que influenciam o rendimento, como a razão líquido-sólido, a temperatura, a potência e o tempo de extração. Quanto ao scale-up, destacaram-se técnicas como cavitação hidrodinâmica, microfluidização em alta pressão e o uso de fotobiorreatores, capazes de otimizar a produtividade e a qualidade da biomassa. Conclui-se que a integração de processos de cultivo eficiente, intensificação de extração e aplicação de tecnologias sustentáveis é essencial para viabilizar economicamente a produção de bioprodutos de microalgas em larga escala, contribuindo para o avanço da bioeconomia e da economia circular.

Palavras-chave: Biocompósitos; Solventes verdes; Reatores biológicos; Processos integrados.

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Pages: 1-32

Abstract

This study aimed to review bioproduct extraction methodologies from microalgae, with a focus on sustainable alternatives and scale-up strategies for industrial applications. An integrative literature review was conducted across national and international scientific databases, focusing on publications from 2021 onward. Microalgae stand out for their high biotechnological potential, producing high-value compounds such as pigments, polyunsaturated fatty acids, and polysaccharides, and have shown feasibility in wastewater treatment processes. Among the analyzed methodologies, microwave-assisted extraction (MAE), enzyme-assisted extraction (EAE), and ultrasound-assisted extraction (UAE) were highlighted, especially when combined with green solvents, as more efficient and environmentally safe alternatives. Critical parameters influencing yield, such as liquid-to-solid ratio, temperature, power, and extraction time, were discussed. Regarding scale-up, techniques such as hydrodynamic cavitation, high-pressure microfluidization, and photobioreactor use were emphasized for their potential to optimize biomass productivity and quality. It is concluded that integrating efficient cultivation processes, intensifying extraction, and applying sustainable technologies is essential to economically enabling large-scale microalgae bioproduct production, thereby advancing the bioeconomy and circular economy.

Keywords: Biocomposites, Green solvents, Biological reactors, Integrated processes.

Resumen

Este estudio tuvo como objetivo revisar las metodologías de extracción de bioproductos a partir de microalgas, con énfasis en alternativas sostenibles y en estrategias de escalamiento para su aplicación industrial. Se realizó una revisión bibliográfica integradora en bases de datos científicas nacionales e internacionales, priorizando las publicaciones publicadas a partir de 2021. Las microalgas destacan por su alto potencial biotecnológico, al generar compuestos de alto valor, como pigmentos, ácidos grasos poliinsaturados y polisacáridos, además de su viabilidad en procesos de tratamiento de aguas residuales. Entre las metodologías analizadas, la extracción asistida por microondas (MAE), la extracción asistida por enzimas (EAE) y la extracción asistida por ultrasonidos (UAE), combinadas con el uso de solventes verdes, se destacan como alternativas más eficientes y ambientalmente seguras. Se discutieron parámetros críticos que influyen en el rendimiento, como la relación líquido-sólido, la temperatura, la potencia y el tiempo de extracción. En cuanto al escalamiento, se destacaron técnicas como la cavitación hidrodinámica, la microfluidización a alta presión y el uso de fotobiorreactores, que permiten optimizar la productividad y la calidad de la biomasa. La conclusión es que la integración de procesos de cultivo eficientes, la intensificación de la extracción y la aplicación de tecnologías sostenibles son esenciales para que la producción de bioproductos de microalgas a gran escala sea económicamente viable, contribuyendo al avance de la bioeconomía y la economía circular.

Palabras clave: Biocomposites; Disolventes verdes; Reactores biológicos; Procesos integrados.

1. Introduction

The expansion of efficient technologies that integrate socio-environmental and economic factors has become a central topic in global discussions. Coordinated efforts at the international level, aimed at ensuring the integrity of the planet and the

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Pages: 1-32

well-being of future generations, led to the proposal of the 17 Sustainable Development Goals (SDGs) by the United Nations (UN) (Silva et al., 2022). Within this context, biotechnology has gradually gained credibility as a strategic tool due to its ability to leverage biodiversity to generate renewable energy, advance sustainable agriculture, and develop less environmentally harmful chemical processes (Sutherland et al., 2021).

In this scenario, microalgae have garnered increasing attention for offering considerable biotechnological potential and the capacity to produce various economically significant compounds (Rizwan et al., 2018). The biomass produced by cultivating these microorganisms is rich in lipids and carbohydrates, enabling the production of third-generation biofuels. Moreover, this biomass contains natural high-value components, such as polyunsaturated fatty acids, carotenoids, and vitamins. Due to their metabolic diversity, microalgae can more readily adapt to climate change and variations in culture media, thereby enhancing their resistance to abiotic factors and xenobiotic substrates (Kokabi et al., 2019). These characteristics, combined with their suitability for production in bioreactors, which allow for better monitoring of biochemical and physicochemical parameters, enhance their capacity to supply bioactive compounds on a larger scale. Additionally, the abundance of high-value elements, such as pharmaceuticals and nutraceuticals, has attracted industrial interest, as has the potential to use microalgae as biocatalysts for exogenous substrates and their multifunctionality as biofertilizers (Victor et al., 2024).

REMUNOM
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Pages: 1-32

Today, the continuous global population growth is evident. Especially in developing countries, urbanization is advancing faster than city planning, leading to sanitation challenges that can cause public health issues and contaminated effluents (Iribarnegaray et al., 2018). In this context, standardized wastewater treatment techniques still exhibit gaps in effectiveness, as they often require high energy consumption, produce significant carbon dioxide emissions, and demand large amounts of chemical inputs. Therefore, the search for alternative technologies with lower environmental impacts, such as microalgae, has become essential. These microorganisms can remove organic carbon, nitrogen, and phosphorus from wastewater. Compared to conventional treatments, microalgae-based methods are more efficient and cost-effective, and generally result in effluents with a higher oxygen content when discharged into water bodies, enabling the recovery of protein and lipid compounds from the resulting biomass, as well as phosphorus and nitrogen (Arbib et al., 2014). One example is the treatment of domestic sewage using phytoremediation with the microalga *Chlorella vulgaris*, which has been proven in previous studies to remove 71% of total nitrogen and 67% of total phosphorus (Gani et al., 2016). Thus, it is worth noting that microalgae used in wastewater treatment could be employed to produce even pharmaceutical compounds, provided they undergo purification, rigorous decontamination, and quality control processes, ensuring compliance with sanitary and safety regulations for human use.

Furthermore, due to climate change and the increasing presence of pollutants and pesticides in ecosystems, various studies have demonstrated that microalgae, when exposed to these compounds, enhance their antioxidant responses and

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Pages: 1-32

increase enzymatic activity, such as catalase and superoxide dismutase. The scientific community may further explore this phenomenon to optimize the extraction of high-value components (Du et al., 2022). Additionally, various studies have investigated the health benefits of microalgal biomass, including its potential role in treating diseases such as Alzheimer's and diabetes, with promising results (Odenthal et al., 2024).

Therefore, microalgae can be considered a renewable and strategic resource, suitable for applications in various biotechnological fields and with growing potential for integration into the circular economy (Kholssi et al., 2021). Given these highlighted functionalities, this review aims to present the principal methodologies for extracting bioactive compounds from microalgae and to discuss scale-up strategies for industrial applications.

Thus, this study aims to review the literature on methodologies for extracting microalgae-derived bioproducts, evaluate alternative methods with potential for large-scale industrial applications, and assess their economic feasibility. It seeks to investigate both conventional and innovative techniques, highlighting their advantages, limitations, and the factors influencing the efficiency of bioactive compound recovery, including solvent choice, extraction type, and process conditions. Furthermore, the study aims to advance knowledge of the utilization of microalgae residues to produce high-value compounds, thereby promoting sustainable practices across various industrial sectors.

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Pages: 1-32

2. Materials and Methods

An integrative literature review was employed to develop this document. The integrative review methodology is a systematic approach that enables the inclusion of experimental and quasi-experimental studies, combining theoretical and empirical literature to provide a deeper understanding of a specific topic (Mendes, Silveira, & Galvão, 2008). This methodology involves synthesizing data from various sources to develop concepts, identify potential gaps in research areas, reassess theories, and examine research methods for a specific subject. Furthermore, integrative reviews are essential for discussing methodologies and their results, evaluating the feasibility of future studies, minimizing uncertainties in practical recommendations, offering more precise generalizations, and supporting decision-making for effective and economically viable interventions (Souza, Silva, Carvalho, 2010).

Following the methodology above, the literature review was structured into five stages:

- Formulation of the guiding question;
- Literature search;
- Data collection;
- Critical analysis of the studies;
- Discussion of the results.

REMUNOM
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DOI: 10.61164/sgdfdc52
Pages: 1-32

Based on this, the guiding question used was: "What are the key variables that influence the optimization of the bioproduct extraction process from microalgae and its scale-up?"

To explore insights into alternative methodologies for extracting high-value compounds related to biorefinery and scale-up strategies, data sources included the websites Scientific Electronic Library Online (SciELO), Web of Science, ScienceDirect (Elsevier), Springer, and Google Scholar, with a preference for publications from 2021 onwards. Some articles included in this discussion precede the specified period due to the scarcity of research and analytical results within the previously established timeframe. Full-text articles written in both Portuguese and English, and available on the platforms mentioned above, were included. To ensure thematic focus and methodological rigor, the following exclusion criteria were applied during article selection: (1) lack of quantitative data on extraction or scale-up parameters; (2) use of raw materials unrelated to microalgae; (3) conventional methods without comparison to emerging techniques.

Based on the selected articles, the titles, keywords, and abstracts were reviewed to identify studies on alternative microalgae extraction methods, comparisons of techniques, economic feasibility assessments, and industrial-scale applications. Only full-text articles available on the databases above were evaluated. In total, 41 scientific papers were selected, from which categories emerged that fostered reflections on biorefinery-related research. The findings were discussed in

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Pages: 1-32

light of the scientific literature, and the analysis supported the foundation of the literature review.

3. Results and discussion

3.1 Microalgae as a Biorefinery

Microalgae exhibit a wide range of morphological, physiological, and genetic diversity, spanning prokaryotic organisms such as cyanobacteria to eukaryotic organisms, including chlorophytes. The growth of these microorganisms is primarily dependent on three factors: water, a carbon source, and energy (Victor et al., 2024). Their life cycle can be briefly described as shown in Figure 1.

Figure 1: Life cycle of microalgae – growth stages and biomass production

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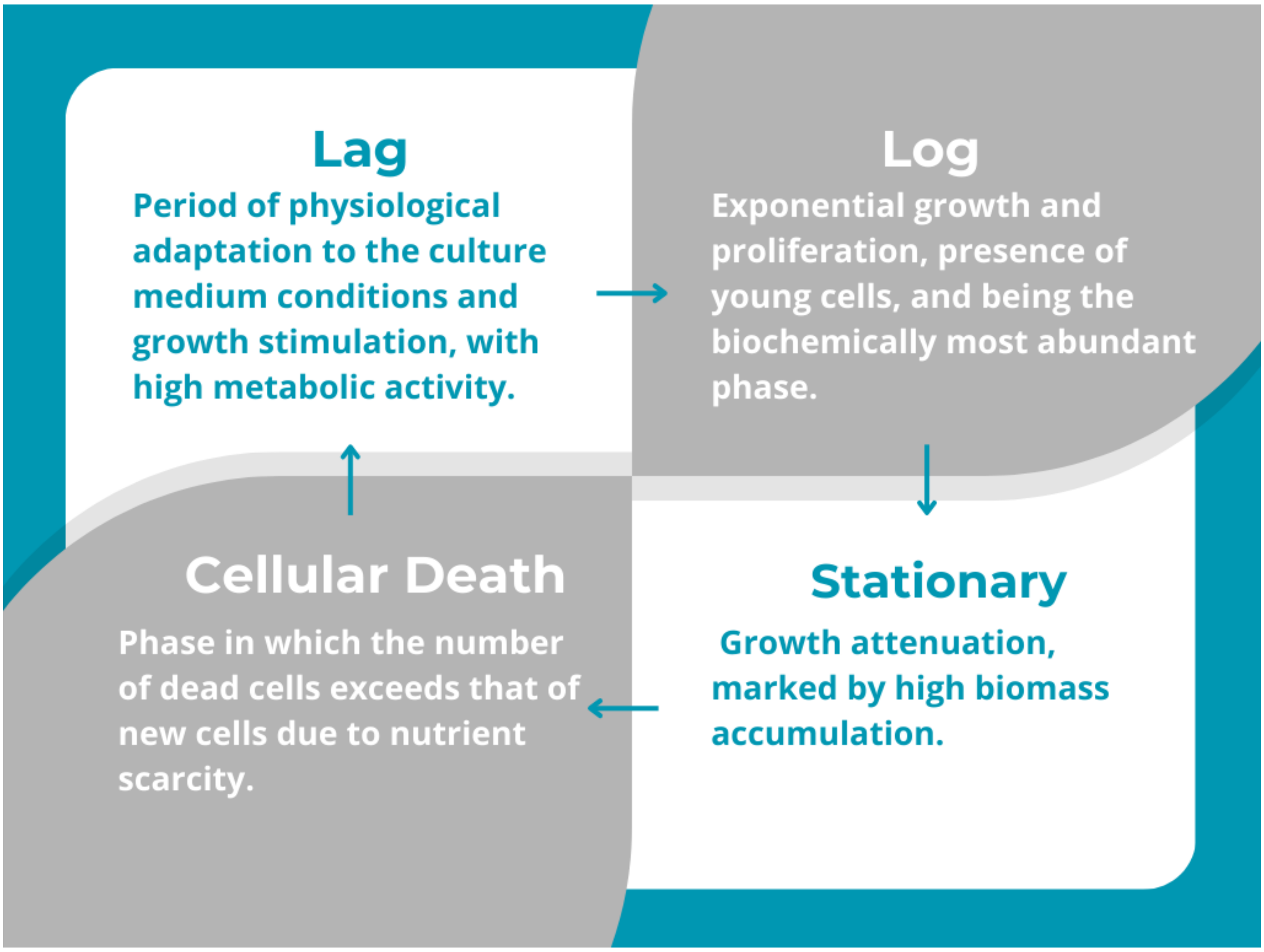

Source: Adapted from Andrade et al., 2014.

Although the primary metabolic pathway for microalgal development is predominantly photoautotrophic, their metabolic flexibility allows them to use alternative energy sources. For instance, in heterotrophic organisms, organic compounds are generally used as both energy and carbon sources. In mixotrophic conditions, energy can be obtained through photosynthesis or via the oxidation of organic compounds (Victor et al., 2024). Therefore, the adaptability of microalgae to various cultivation modes is a highly relevant factor in the biotechnological domain.

In this context, the large-scale production of microbial algae has the potential to foster the growth of companies based on sustainable resources, aiming to produce

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Pages: 1-32

high-value-added inputs that are both economically viable and aligned with biomass production. Hence, microcultivation techniques must ensure efficient utilization of biomass. Another essential condition for increased productivity is the use of different types of bioreactors. The design of the bioreactor system can be tailored according to the target species and the desired product profile. In large-scale scenarios, microalgae are often cultivated in open ponds; however, this approach is highly susceptible to contamination (Bhattacharya & Goswami, 2020).

Regarding methodologies used to enhance microalgal production, strengthening carbon fixation within the cells—by adjusting their metabolic pathways—enables greater extraction of bioproducts from their biomass (Chen et al., 2018). The use of synthetic substances as metabolic modulators has demonstrated effects such as regulating biosynthetic pathways, stimulating oxidative stress responses, and increasing the availability of metabolic precursors. For example, one bioproduct with modulated production is docosahexaenoic acid (DHA), a fatty acid component of omega-3 fatty acids, which has attracted interest from the nutraceutical industry due to its neuroprotective properties. Several studies on *Chlorella sorokiniana* cultures supplemented with 1-naphthaleneacetic acid and 2-phenylacetic acid have shown increases in lipid-rich biomass growth of up to 104%. Similarly, methodologies involving the addition of malic acid to *Schizochytrium sp.* culture media resulted in increases of 35–60% in DHA content within the total fatty acid composition (Figure 2) (Victor et al., 2024). Thus, optimizing microalgal cultivation has become crucial for improving biomass composition and facilitating its application in biotechnological processes.

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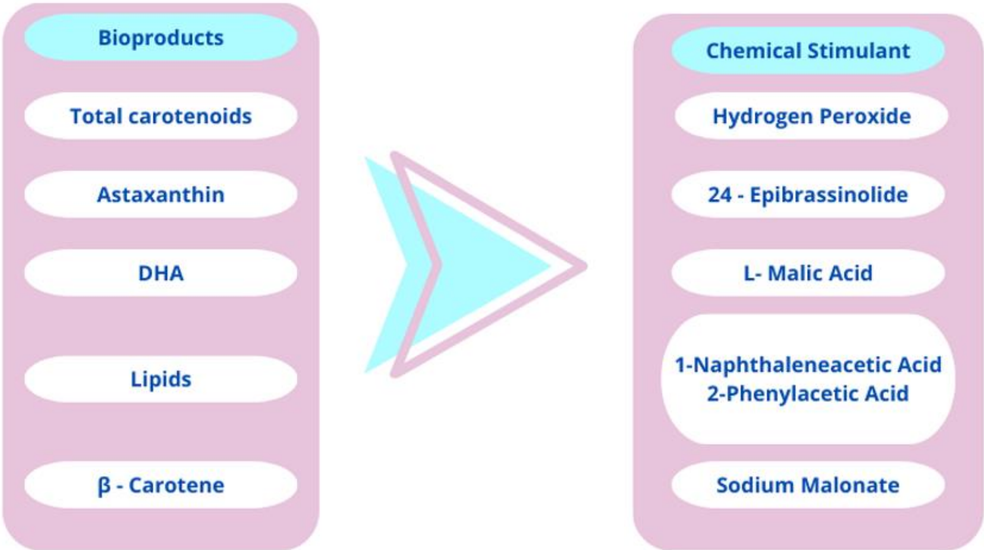
Pages: 1-32

Figure 2: Bioproducts and their respective chemical stimulants aimed at optimizing growth and accumulation.



Source: Own elaboration.

Finally, among the various high-value bioproducts synthesized by microbial algae are carotenoids, docosahexaenoic acid (DHA), eicosapentaenoic acid (EPA), bioactive and natural pigments, natural dyes, polysaccharides, and even cancer-preventing agents. Additionally, over the past 20 years, research on microbial algae has been primarily focused on four types: a) *Spirulina* (*Arthrospira platensis*), b) *Chlorella vulgaris*, c) *Dunaliella salina*, and d) *Haematococcus pluvialis*. The metabolic regulation imposed in these media enables targeted, enhanced production of bioproducts with diverse commercial interests (Table 1).

Table 1: Examples of production of bioproducts with diverse commercial interests.

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Species / Group	Products	Field of Application
Spirulina platensis	C-Phycocyanin	Cosmetics, nutraceuticals
Chlorella vulgaris	Ascorbic acid	Food, dietary supplements, animal feed
Haematococcus pluvialis	Astaxanthin	Nutraceuticals, cosmetics, animal feed
Dunaliella salina	β-Carotene	Nutraceuticals, supplements, animal feed
Crypthecodinium cohnii	DHA (Docosahexaenoic Acid)	Infant brain development, health, and nutrition
Spirulina maxima	Protein, Vitamin B12	Antioxidants, immune system support
Odontella aurita	Fatty acids, EPA (Eicosapentaenoic Acid)	Pharmaceuticals, cosmetics, anti-inflammatories
Porphyridium cruentum	Polysaccharides	Pharmaceuticals and cosmetics
Isochrysis galbana	Fatty acids	Nutrition, biofuel production

Source: Own elaboration.

In summary, the type of extraction to be adopted is directly related to the physicochemical characteristics of the bioproduct to be obtained, such as lipids, proteins, pigments, or polysaccharides, requiring specific approaches to maximize process efficiency.

3.2 Emerging Methodologies: Green Solvents as Sustainable Alternatives

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Pages: 1-32

Currently, the most common techniques for recovering bioactive components are steam distillation, hydrodistillation, and mechanical pressing. However, low extraction yields, long extraction times, and the large volumes of organic solvents required are common characteristics of these techniques, which hinder process efficiency and the commitment to a sustainable economy (Vo et al., 2024). Therefore, it is evident that there is a need to seek and optimize more efficient methodologies that are aligned with sustainable development and primarily use less harmful solvents.

The use of solvents is essential in extraction methodologies, as they dissociate the soluble fraction of interest from the algal matrix and purify the obtained extracts. On the other hand, the widespread use of organic solvents raises environmental concerns, including toxicity, volatility, and the potential for harmful residues. Consequently, it is necessary to explore methods that reduce the use of organic solvents or replace them with greener alternatives (Hashemi et al., 2022). Furthermore, it is essential to note that for a solvent to be classified as "green," it must be biodegradable, non-toxic, and derived from renewable sources. Among the prominent examples of green solvents used are water, in both its conventional and subcritical forms, for the extraction of hydrophilic and low-polarity compounds.

Similarly, eutectic solvents and their natural derivatives, such as choline and lactic acid, exhibit solvating power and are effective in extracting phenolic compounds and flavonoids. Biological solvents, derived from renewable raw materials such as

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Pages: 1-32

agricultural residues—including terpenes like d-limonene, esters, and alcohols—also can replace conventional organic solvents.

3.2.1 Microwave

The microwave-assisted extraction (MAE) process involves the direct introduction of electromagnetic energy into microalgae, primarily through two molecular-level energy transfer mechanisms: dipolar rotation and ionic conduction (Mao et al., 2023). Polar molecules in algae and green solvents retain microwave energy, generating thermal energy. Furthermore, the dipolar rotation mechanism predominates in distilled water, whereas ionic conduction becomes prevalent in saline, acidic, and alkaline solutions. This method provides rapid, effective heating of compounds, thereby enhancing extraction efficiency while maintaining the integrity of target molecules, including pigments, polyphenols, and fatty acids.

Several studies highlight that the liquid-to-solid ratio (LSR) is a key parameter in microwave applications, as it directly affects microwave heating efficiency. Researchers have observed that very low LSR values hinder the diffusion of bioactive compounds out of cells. In contrast, excessively high values lead to overheating that can degrade the bio-compounds. As reported by Safari et al. (2015) in the optimization of MAE to obtain bioproducts from *Chaetomorpha* sp., a Central Composite Design model was employed to investigate the antioxidant activities of these compounds. Under optimal conditions, 25% ethanol concentration, 300 W microwave power, and 8 minutes of extraction, the phenolic content and reducing

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Pages: 1-32

power were 1.09 mg tannic acid equivalent/g and 0.12 mg tannic acid equivalent/g, respectively (Safari et al., 2015).

Other essential factors for MAE effectiveness include applied power and temperature. High powers can induce thermal degradation, but when used appropriately, they also enhance compound diffusion. Regarding temperature, Xiao et al. (2010) observed a significant drop in flavonoid extraction efficiency above 110 °C, highlighting the importance of thermal control for thermosensitive compounds. Other critical conditions include extraction time and the number of cycles. Lucía Cassani et al. (2024) extracted phenolic compounds, flavonoids, and fucoxanthins from Ascophyllum nodosum under optimal conditions of 10.4 bar and 46.8% ethanol for approximately 3 minutes, yielding 87.55, 15.72, and 2.55 mg/g, respectively (Cassani et al., 2024).

Additionally, fractionating the technique into cycles with solvent reintroduction can be effective in preventing degradation and maintaining yield. Bárbara C. Jesus et al. (2023) utilized eutectic solvents and MAE to extract approximately 26.69 ppm of salicylic acid from Sargassum muticum at 60°C for 6 minutes, demonstrating the effectiveness of this process as an intensification technique. Finally, Lourenço-Lópes et al. (2023) analyzed MAE extraction of fucoxanthin from *Undaria pinnatifida*, using ethanol concentration, time, and pressure as variables, achieving a final concentration of 58.83 mg of fucoxanthin in 3 minutes at 2 bar under optimized conditions.

3.2.2 Enzyme-Assisted Extraction

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Vol: 19.03
DOI: 10.61164/sgdfdc52
Pages: 1-32

Enzyme-assisted extraction (EAE) is a methodology for extracting bioactive compounds (BCs) in which enzymes degrade the algal cell wall, which is composed of polysaccharides such as pectin, agar, cellulose, hemicellulose, alginate, proteins, and fucoidans (Velazquez-Lucio et al., 2018). These structural compounds also act as barriers to the release of BCs bound to the cellular matrix. Therefore, the introduction of enzymes allows for the breakdown of these structures, facilitating access to target compounds and maximizing extraction yields. Commonly used enzymes include α-amylase, cellulase, pectinase, and protease, which enable the extraction of phenolic compounds, flavonoids, pigments, and fatty acids (Jha et al., 2022).

When discussing enzyme concentration, it is considered one of the most critical factors affecting EAE effectiveness. High enzyme concentrations can either saturate the substrate or lead to enzyme aggregation, potentially reducing interaction efficiency. In a recent study supporting this premise, Hoang Chinh Nguyen et al. (2024) applied EAE to extract phenolic compounds from *Padina gymnospora*. The optimized conditions—0.32% enzyme, 60.5 °C, LSR of 61.31 mL/g, and 1.95 h extraction time—resulted in approximately 97.6% recovery of total phenolic content.

The pH of the solutions also significantly impacts enzymatic activity, affecting enzyme conformation and stability. Values outside the optimal range reduce the interaction between the cell wall and enzymes, thereby hindering the release of compounds. Moreover, pH fluctuations compromise the dissociation of functional

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DOI: 10.61164/sgdfdc52
Pages: 1-32

groups, such as amines and hydroxyls, thereby negatively affecting extraction yield (Ezeilo et al., 2020).

Other parameters influencing both enzyme integrity and compound diffusivity include temperature and incubation time. In a methodology tested with *Caulerpa lentillifera*, Rahmawati et al. (2020) used papain (a papaya-derived protease) at 60 °C for 16 hours at a 0.5% concentration to enhance the recovery of polyunsaturated fatty acids, such as omega-6 and omega-9. The results highlighted the need to balance temperature, time, and enzyme stability to prevent thermal degradation losses. Regarding the liquid-to-solid ratio (LSR), low concentrations have been shown to increase medium viscosity, hindering enzyme-substrate contact. In contrast, high concentrations raise operational costs without a proportional gain in yield (Vo et al., 2024).

3.2.3 Ultrasound

Ultrasound-assisted extraction (UAE) is a more sustainable and innovative process, characterized by reduced toxic waste and the feasibility of using environmentally friendly solvents, such as deep eutectic solvents. Based on acoustic cavitation induced by ultrasonic waves, this method can generate physical effects such as microjets, shock waves, and cell fragmentation, which disrupt the algal cell wall and facilitate the release of bioactive compounds (BCs). According to Jinggui Nie et al. (2021), this technique was employed to extract fucoxanthin from *Sargassum fusiforme*, using limonene and vegetable oils as solvents. The optimized conditions were 40 mL/g (LSR), 75 °C, and 53% ultrasonic amplitude for 27 minutes.

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Pages: 1-32

Ultrasonic power is one of the most critical parameters for effective UAE. Different studies have reported that powers between 20 and 900 W are generally used, resulting in efficient cavitation and extraction; however, excessively high power values can cause bubble collisions and reduce efficiency. Olfat et al. (2024) operated at 80 W using NADES as solvent, LSR of 29.13 mL/g, and 30 minutes, achieving approximately 88.31 mg/100 g of phenolic compounds from *Hypnea flagelliformis*.

Parameters such as ultrasonic pulsing and exposure time were investigated by Putra et al. (2022), who optimized the methodology for red algae to extract phenolic compounds, employing 100% power, a pulse duty cycle of 1 s^{-1}, 52.5 °C, 50% ethanol, and LSR of 30 mL/g, resulting in about 0.4 mg of gallic acid (mg GAE/g). Pulsing is effective because it produces intense, homogeneous cavitation, enhancing yield while reducing energy consumption. Regarding extraction time, prolonged durations can degrade target compounds and, therefore, require careful optimization.

Temperature control has also proven essential for extractions. Moderate temperatures, such as 50–75 °C, promote BC diffusion without causing thermal degradation. Jinggui Nie et al. (2021) highlighted that excessively high temperatures initially maximize extraction, but beyond a certain point, fucoxanthin degradation occurs. Their kinetic analysis revealed that extraction time, temperature, and amplitude directly influence both the initial extraction rate and the final yield. The optimized parameters were 75°C, 53% amplitude, and 27 minutes, resulting in 696.85 µg/g of fucoxanthin. These results reinforce the UAE's potential as a sustainable and practical approach.

REMUNOM
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Pages: 1-32

Although supercritical fluid extraction (SFE) is widely studied in the literature due to its sustainability and selectivity, investigations have reported lower yields than emerging methodologies such as EAE, UAE, and MAE. For this reason, SFE still requires further research to optimize operational parameters and adapt to different types of biocompounds; therefore, it was not addressed in depth in this work.

Table 2: Comparison of Extraction Methodologies, Green Solvents Used, and Compounds Extracted

Method	Green Solvent	Study / Author (Year)	Main Parameters	Extracted Compounds
MAE	25% Ethanol	Safari et al. (2015)	300 W, 8 min	Phenolic compounds, reducing power
MAE	46.8% Ethanol	Cassani et al. (2024)	10.4 bar, 3 min	Phenolics (87.55 mg/g), flavonoids (15.72 mg/g), fucoxanthin (2.55 mg/g)
MAE	Ethanol	Lourenço-López et al. (2023)	3 min, 2 bar	Fucoxanthin (58.83 mg)
MAE	Eutectic Solvents (DES)	Jesus et al. (2023)	60 °C, 6 min	Salicylic acid (26.69 ppm)
EAE	Acidified Water	Shannon & Abu-	pH 4.5, 3.05 h, 5.4% algae/water ratio	Fucoxanthin

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Pages: 1-32

		Ghannam (2018)		
EAE	Water	Xiaoyan Zhao et al. (2019)	pH 5, 40 °C, one h	Astaxanthin (60.93% yield)
EAE	Water	Habeebullah et al. (2020)	50 °C, 20 h, 1:100 mL/g ratio	Polyphenols (8.25 mg GAE/g), antibacterial peptides
EAE	Water	Hoang Chinh Nguyen et al. (2024)	0.32% enzyme, 60.5 °C, 1.95 h, LSR 61.31 mL/g	Phenolic compounds (97.6% yield)
EAE	Water	Rahmawati et al. (2020)	0.5% papain, 60 °C, 16 h	Omega-6, Omega-9 fatty acids
UAE	NADES	Olfat et al. (2024)	80 W, 29.13 mL/g, 30 min	Phenolics (88.31 mg/100 g)
UAE	NADES	Obluchinskaya et al. (2023)	23 min, LSR 12:1, 30% water	Phlorotannins (137 mg/g)
UAE	50% Ethanol	Putra et al. (2022)	100% power, 1 s pulses, 52.5 °C, 30 mL/g ratio	Phenolics (~0.4 mg GAE/g)
UAE	DES (Octanoic + Decanoic Acid)	Yang et al. (2023)	50 °C, 49 min, LSR 66.2 mL/g	Canthaxanthin (70.4 µg/mL)

REMUNOM
ISSN 2178-6925
Received: 15/07/2025 - **Accepted:** 27/10/2025
Vol: 19.03
DOI: 10.61164/sgdfdc52
Pages: 1-32

UAE	Vegetable Oils / Limonene	Nie et al. (2021)	40 mL/g, 75 °C, 53% power, 27 min	Fucoxanthin (696.85 µg/g)
UAE	Water / NADES	Krishnamoorthy et al. (2023)	80 kHz, 15 min	Lipids (17.9% yield)

Source: Own elaboration.

4 Methodologies For Scale-Up

The scale-up of techniques for obtaining bioinputs from microalgae represents one of the main challenges for establishing an industrial-scale biorefinery. Reference studies highlight that the downstream stage, occurring after cultivation and focused on effective recovery and quality preservation, stands out as the most costly part of the production chain, encompassing harvesting, drying, cell disruption, and extraction, all of which require substantial energy and resources (Korsah et al., 2025). Additionally, the upstream process, which involves the preceding productive stages, includes cultivation, strain selection, nutrient optimization, biomass enhancement, illumination, and management of environmental conditions. Therefore, proper coordination between these stages is crucial to maintain economic viability and ecological sustainability. Another challenge for scale-up is the variability of microalgae, as each type may possess distinct characteristics and varying levels of bioactive compounds, complicating the standardization of methodologies and necessitating product-specific treatments (Jacob-Lopes et al., 2024).

REMUNOM
ISSN 2178-6925
Received: 15/07/2025 - **Accepted:** 27/10/2025
Vol: 19.03
DOI: 10.61164/sgdfdc52
Pages: 1-32

Investigations have examined the role of hydrodynamic cavitation (HC) in large-scale compound extraction. This process occurs when a liquid is accelerated through a device such as a Venturi tube or an orifice, producing pressure oscillations that generate vapor bubbles. These bubbles collapse energetically, releasing heat, shockwaves, and microjets that can penetrate and break microalgal cell walls, thereby releasing target compounds (Sun et al., 2020). For example, a vortex-based device applied to Spirulina under optimal conditions of 150 kPa and 90 passes achieved a low energy consumption of ~0.06 kWh/kg of biomass and a yield of 52 mg phycocyanin per gram, indicating energy efficiency up to two orders of magnitude higher than conventional methods (Chegukrishnamurthi et al., 2025). Moreover, studies show that HC provides higher energy efficiency than ultrasound, being approximately 6–10 times more effective, and does not require additional chemicals (Arya et al., 2023).

Another industrial scale-up innovation is high-pressure microfluidization. Despite operating at pressures of around 120 MPa, this technique forces the microalgal suspension through narrow microchannels, generating shear forces and hydraulic shock that efficiently disrupt the cells. Ke et al. (2023) reported that using Chlorella pyrenoidosa, this approach achieved up to 98 % cell disruption, increased soluble solids content, and enhanced protein recovery by approximately 20 %, while also improving lipid content and chlorophyll levels. This method is compatible with biomass, does not cause significant thermal degradation, and is effective for extracting biocompounds (Ke et al., 2023).

REMUNOM
ISSN 2178-6925
Received: 15/07/2025 - **Accepted:** 27/10/2025
Vol: 19.03
DOI: 10.61164/sgdfdc52
Pages: 1-32

Photobioreactors (PBRs) are another prominent technological strategy. These closed systems promote microalgal growth under highly controlled conditions, optimizing parameters such as light, temperature, pH, nutrient availability, and CO_2 supply. PBRs can be designed in various formats, including tubular systems, flat panels, and bubble columns, each offering specific advantages in light efficiency, mass transfer, and operational simplicity. Tubular PBRs circulate culture media through transparent horizontal or vertical tubes. Flat panels utilize stacked transparent plates to maximize light capture, and bubble columns mix and aerate cultures via air injection, thereby enhancing CO_2 transfer and cultivation uniformity (Korsah et al., 2025).

Research shows that PBRs can achieve higher biomass productivity than open systems. Flat-panel and tubular designs have achieved productivity rates of nearly 1 g L^{-1} day^{-1}, depending on the species and operating conditions. In comparison, open ponds typically yield no more than 0.5 g L^{-1} day^{-1} (Jacob-Lopes et al., 2024). Despite higher initial and operational costs, these systems offer effective light capture, higher cell density, and improved area utilization, enabling scale-up for the production of high-value bioproducts. Precise environmental control also reduces contamination risks and enhances biomass composition. For instance, a bubble column PBR with a working volume of 28.5 L was used to cultivate *Scenedesmus almeriensis* under monitored LED illumination, a maintained temperature of 28 °C, and constant gas mixture injection (O_2, N_2, CO_2), ensuring stable conditions (±1 °C, stable pH) and supporting continuous modeling of growth and metabolic response for both upstream and downstream integration in a biorefinery (Molino et al., 2019).

REMUNOM
ISSN 2178-6925
Received: 15/07/2025 - **Accepted:** 27/10/2025
Vol: 19.03
DOI: 10.61164/sgdfdc52
Pages: 1-32

Numerical simulation of fluid dynamics has become an essential tool for PBR scale-up, allowing the optimization of flow, mixing, mass transfer, and light distribution within the culture substrate. This approach enables testing of reactor profiles and operational parameters while minimizing physical prototyping, costs, and project timelines. Modeling also identifies dead zones, improves hydrodynamics, and enhances energy efficiency, all of which are critical for maintaining biomass productivity and quality (Oro et al., 2025). Thus, integrating cultivation, harvesting, and extraction is essential for maximizing the economic performance of microalgae biorefineries. Continuous processes combining HC, microfluidization, and green solvents represent a promising route for industrial scale-up.

Calculating reactor capacity and conducting techno-economic analyses are vital for successful scale-up. A recent review in Bioresource Technology (2023) examined the challenges in scaling up microalgal biorefineries, including harvesting, downstream processing, and financial sustainability. The authors reported biomass production costs ranging from US$2.5 to US$10 per kg, depending on the technological approach (flocculation versus centrifugation), cultivation system (PBRs or open ponds), and extraction method.

Chapter 24 of Jacob-Lopes et al. (2025) systematically addresses reactor design units, emphasizing the management of parameters like cell growth rate, volumetric productivity, nutrient conversion, and liquid residence time. The chapter also covers economic and financial quantification, including operational and financing costs, production cost per kilogram of biomass, payback period, and internal rate of

REMUNOM
ISSN 2178-6925
Received: 15/07/2025 - **Accepted:** 27/10/2025
Vol: 19.03
DOI: 10.61164/sgdfdc52
Pages: 1-32

return (IRR). Strategies to minimize expenses include gas recirculation, utilization of residual heat, optimized LED lighting with adjustable spectra, and the selection of cost-effective reactor materials, such as reinforced plastics or borosilicate glass.

Therefore, successful scale-up depends on integrating bioprocess engineering, biochemistry, and economic principles. Establishing versatile and viable technological platforms requires adapting reactor designs to target compounds and regional production conditions. In conclusion, the industrial application of microalgal biocompound extraction innovations requires integrated approaches that combine efficient cultivation, process intensification, and environmentally friendly solvents. Continuous optimization and cost-reduction studies are essential to ensure sustainable, economically viable processes.

Conclusion

In the methodology, the guiding question addressed which key variables influence the optimization of bioproduct extraction from microalgae and its scale-up. Based on the reviewed literature, the main extraction parameters identified were the liquid-to-solid ratio, temperature, extraction time, applied power, and solvent choice, preferably a green solvent. These factors must be adjusted based on the physicochemical properties of the target compound to maximize efficiency, prevent degradation, and preserve bioactivity. Additionally, upstream steps such as harvesting and cell disruption directly affect compound accessibility and yield.

Regarding scale-up, variables such as the type of cultivation system, integration between upstream and downstream processes, and the adoption of

REMUNOM
ISSN 2178-6925
Received: 15/07/2025 - **Accepted:** 27/10/2025
Vol: 19.03
DOI: 10.61164/sgdfdc52
Pages: 1-32

intensification technologies—such as hydrodynamic cavitation, high-pressure microfluidization, and photobioreactors are critical to ensure technical, economic, and environmental feasibility. Therefore, the combination of optimized laboratory parameters with effective scale-up strategies is essential to enable competitive microalgae biorefineries, reducing costs, minimizing environmental impacts, and meeting the growing demand for high-value bioproducts.

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