

## Environmental engineering Aplinkos inžinerija

# A REVIEW OF AQUATIC PLANT BIOMASS PRETREATMENT METHODS FOR BIOGAS PRODUCTION

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**Abstract.** The increasing global demand for renewable energy sources has led to interest in generating biogas from aquatic plant biomass. This research examines the pretreatment methods of biomass, aiming to enhance the efficiency of biogas production processes. The analysis presents mechanical, chemical, biological, or combined pretreatment techniques, assessing their respective advantages, limitations, and potential synergies. The review extends beyond individual methodologies to identify potential synergies or sequential applications that could maximize biogas production efficiency. This study ultimately facilitates moving the field forward, contributes to the transition to more sustainable energy solutions, identifies gaps in existing researches, paving the way for future investigations and combat to solve upcoming energy problems.

**Keywords:** anaerobic digestion, biogas, microalgae, pretreatment, aquatic biomass, bioreactor.

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## 1. Introduction

Among many renewable energy options, biogas production provides a solution that is both environmentally friendly and efficient. Biogas production from algae by Anaerobic digestion has been the subject of research for more than 60 years, several key obstacles to biogas production remain, especially limitations in the biodegradability of algal biomass. Such bioprocessing obstacles reduce the economic feasibility for moving the technology forward (Bohutskyi et al., 2014).

Aquatic plants find application in the production of various types of biofuels, including biodiesel through oil extraction processes, hydrothermal liquefaction and pyrolysis, and biogas through anaerobic fermentation. These methods enable the efficient conversion of microalgae energy resources into alternative fuels, opening up new prospects for a sustainable energy future (Xiao et al., 2020). Microalgae are a diverse group of microorganisms, including both unicellular and simple multicellular forms, spanning prokaryotic (cyanobacteria) and eukaryotic species, including green algae (Chlorophyta), red algae (Rhodophyta) and diatoms (Bacillariophyta) (Zabed et al., 2020). Chlorophytes (including micro- and macroalgae) are the largest group of algae with applications in bioremediation, water purification, food supply, pharmaceuticals and energy production (Saad et al., 2019). These organisms

have shown significant potential for biogas generation due to their unique biological and technological properties (Zabed et al., 2020). The use of microalgae as a biofuel feedstock not only helps reduce dependence on fossil fuels, but also offers an environmentally friendly alternative given its low carbon footprint and the ability to use land unsuitable for agriculture. Aquatic plant biomass, with its abundant availability and rapid growth rate, is an excellent candidate for biogas production (Dębowski et al., 2013). Bioreactor operating conditions can affect CO<sub>2</sub> availability, capacity, share ratio and light exposure. An important factor is the gas transport supplying CO<sub>2</sub>, NO<sub>x</sub> and SO<sub>x</sub> (sources of C, N and S), pH control through appropriate CO<sub>2</sub> concentrations and internal mixing. Internal mixing prevented nutrient concentrations, gradients, also reduced excessive dissolved oxygen and promoted light exposure for all cells (Jankowska et al., 2017) However, the path from raw aquatic biomass to energy-rich biogas is fraught with difficulties, the main one being the complexity of pretreatment processes (Passos et al., 2014).

The main purpose of pretreatment is to maximize biogas production by increasing the efficiency of anaerobic digestion. Pretreatment increases the amount of methane produced from a given amount of biomass, thereby increasing the overall energy efficiency of the process (Babatunde et al., 2023).

Current pretreatment methods used to convert aquatic plant biomass into a digestible substrate for biogas production. The biomass, containing organic components such as plants, algae and various types of microorganisms, is capable of being transformed into biogas – a composition consisting of methane (CH<sub>4</sub>) and carbon dioxide (CO<sub>2</sub>), as a result of the process of anaerobic digestion (Abusweireh et al., 2023).

The world moves toward a more sustainable future, understanding the role of aquatic plant biomass in biogas production is a crucial point for renewable energy. Thus, this review lays the foundation for further research and innovation in the field of biogas technologies.

## 2. Pretreatment methods applied to aquatic plant biomass

Pretreatment techniques categorized into thermal, mechanical, chemical, biological and combined methods. Various methods can be used depending on the type of algae biomass, specific process condition, efficiency, economic feasibility and environmental impact (Babatunde et al., 2023) (Figure 1). Each category includes a distinct set of processes designed to change the physical or chemical properties of a material in order to prepare it for subsequent use. The most important needs of pretreatment methods are to maintain the total organic matter content and to prevent the formation of inhibitory substances.

### 2.1. Thermal pretreatment

Currently, thermal and mechanical methods are considered to be more effective in disrupting microalgal cells, and thermal pretreatment is considered to be the most widely studied method. This method has not only been used in continuous reactors but has also played an important role in clean energy production (González-Fernández et al., 2012a). The effectiveness of thermal methods lies in their ability to change cell structures through heat, increasing the release of valuable cellular components. Meanwhile, mechanical pretreatments, which use physical forces to disrupt cell walls, complement thermal methods to optimize cell disruption. This synergy highlights progressive advanc-

es in biofuel technology to maximize energy production from microalgae (González-Fernández et al., 2012b).

Thermal methods involve the application of high temperatures such as steam explosion, boiling. This pretreatment involves heating the biomass to below 100 °C at atmospheric pressure. The hydrothermal method involves applying heat at temperatures above 100 °C with a gradual release of pressure after pretreatment. A steam explosion is a sudden drop in pressure after pretreatment at temperatures above 100 °C (Kendir & Ugurlu, 2018).

From numerous studies, it is clear that to improve the efficiency of methane production from microalgae biomass, the optimal pretreatment temperature should range from 55–170 °C. This temperature range is critical for the decomposition of organic components of microalgae, which contributes to increased methane yield, highlighting the importance of fine-tuning pretreatment conditions in bioenergy processes (de Oliveira et al., 2022). Biogas production by anaerobic digestion of algae is significantly increased by thermal pretreatment at 90 °C with a maximum increase of 41% per unit volatile solids added. Dannon modeling shows that only 40 minutes of pretreatment is needed to achieve 90% of maximum biogas production (Marsolek et al., 2014).

### 2.2. Mechanical pretreatment

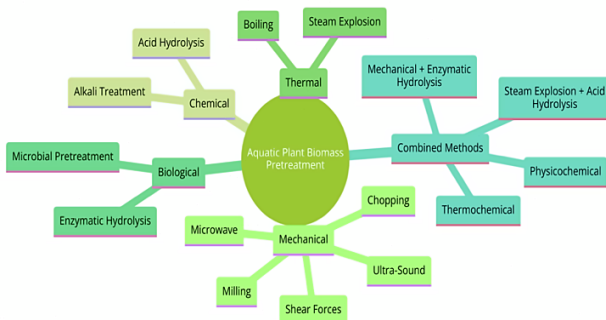
Mechanical methods rely on the application of physical forces such as ultrasound, microwave, chipping, milling or shear forces to reduce the crystallinity of cellulose, disrupting its structure, increasing the surface area of organic matter, and depolymerizing the microalgal cell wall containing hemicellulose (Kendir & Ugurlu, 2018). Among these methods, mechanical grinding and ultrasonic treatment are the most common.

Ultrasound at different energy levels was studied as a pretreatment in terms of organic matter solubilization and cell wall disruption to elucidate its effect on anaerobic biodegradability. Due to the increase in temperature during ultrasonication, this treatment was also compared with heat pretreatment without ultrasonication (González-Fernández et al., 2012).

### 2.3. Chemical pretreatment

Chemical treatment takes advantage of the destructive nature of chemicals such as acid hydrolysis, alkali treatments. Important to mention that chemical pretreatment has the disadvantages such as solubilized compounds can be toxic to methanogens and secondly, it can increase the production of hydrogen sulfide or ammonia nitrogen instead of methane.

Studies of the solubilization of cell wall components and intracellular organic compounds such as carbohydrates, lipids and proteins by chemical pretreatment have revealed that key parameters influencing the effectiveness of this method include the concentration of reagents, volume of biomass, temperature and duration of treatment.



**Figure 1.** Pretreatment methods for algae biogas production (Passos et al., 2014)

These factors play a critical role in optimizing the process of extracting valuable components from biomass, thereby improving the subsequent biochemical conversion and increasing the overall efficiency of bioenergy production (de Oliveira et al., 2022).

### 2.4. Biological pretreatment

Biological pretreatment is based on the use of enzymes' ability to hydrolyze, whereas thermal treatment is based on the use of heat to cause destruction (Kendir & Ugurlu, 2018). Key factors influencing the effectiveness of pretreatment include enzyme concentration, temperature conditions and exposure time. For each specific enzyme, the ideal temperature regime and pH level are determined, which lie within the limits of its optimal activity.

Despite this, the specificity of enzyme-substrate interactions, the wide variety of composition and structure of microalgae cell walls, as well as the high costs of enzyme production are significant obstacles that need to be addressed before introducing this technology at an industrial level in the biofuel sector (Passos et al., 2014).

### 2.5. Combined pretreatment

It is considered that one of the most common combined pretreatment methods is physicochemical or thermochemical treatment.

These methods combine pretreatment techniques improve the extraction of lignin, cellulose, and hemicellulose from lignocellulosic biomass. Other thermochemical pretreatment methods shown in the Figure 2 include liquid hot water pretreatment, steam explosion, ammonia fiber expansion, CO<sub>2</sub> explosion, ammonia recycle percolation, and oxidative pretreatment (Babatunde et al., 2023).

Energy-efficient pretreatment uses minimal or negligible amounts of electricity and results in higher biomass dilution. Biological and combined pretreatments often require less energy than other pretreatment methods. Choosing an energy-efficient pretreatment will lead to profitable biomethane (Kavitha et al., 2017).

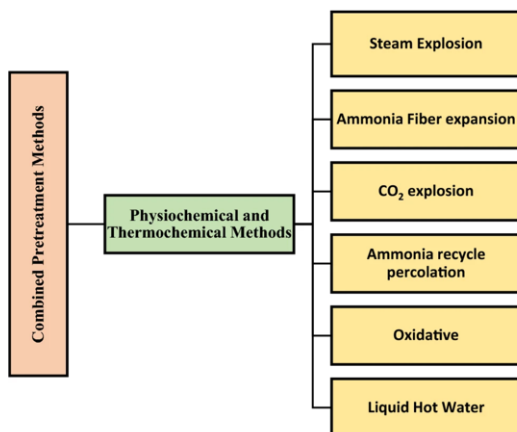


Figure 2. Combined pretreatment methods for biogas production using algae biomass

## 3. Pretreatment methods selection process and requirements

Each pretreatment method can be applied to specific or all types of microalgae biomass. However, the selection of a suitable pretreatment method mostly depends on their cost-effectiveness (Figure 3). While choosing a pretreatment method, low capital and operating costs are important parameters (Dębowski et al., 2013). Additionally, pretreatment methods that have a positive energy balance are advantageous for making anaerobic digestion an economically viable process. Technological applicability of the pretreatment process from laboratory to pilot scale is another important factor when choosing a suitable method (Babatunde et al., 2023).

Moreover, the selection of pretreatment methods is determined by the chemical composition of the algae, as using low-energy processing techniques results in lower yield, the process' energy balance has to be positive for it to have a practical impact (Abusweireh et al., 2023).

The use of chemicals causes the formation of toxic organic acids, which contaminate the product and modify the pH level in the reactor. It negatively affects the methanogenesis process and can lead to malfunction of the digestion equipment. To increase the activity of microorganisms and improve the process of anaerobic decomposition, alkaline substances are introduced into the pre-treated liquid from manure before fermentation, the purpose of which is to neutralize acidity (Thompson et al., 2019).

Biogas plants require a continuous supply of bio-raw materials which is substrate, therefore the possibility of the uninterrupted supplying for the substrate should be carried out in the first stage of the design. It is also considered that the most optimal is to place the biogas plant near the source of the produced substrate.

Research into the use of ultrasound to improve methane yield and solubilize microalgae biomass has revealed a variety of results ranging from negative to positive effects. These studies, which focus on the physical aspect of pretreatment, demonstrate that ultrasound is not the only method for preparing microalgae cells for subsequent use. This variety of results emphasizes the complexity of



Figure 3. Pretreatment methods advantages and disadvantages of different pretreatment methods (Kendir & Ugurlu, 2018)

**Table 1.** Limitations and solutions of pretreatment method

Pretreatment method	Limitation	Solution
Mechanical	High energy consumption Equipment wear and tear Expensive equipment repair	Optimization of process parameters (e.g., duration, intensity) Integration with low-energy processes
Chemical	Chemicals can be costly and corrosive pH imbalance Inhibition of the methanogenic microorganisms Environmental pollution from waste chemicals Potential inhibition of microbial activity in the anaerobic digestion process	Chemicals can be costly and corrosive Environmental pollution from waste chemicals Potential inhibition of microbial activity in the anaerobic digestion process
Thermal	High energy/heat demand Potential degradation of valuable compounds	Energy recovery systems to utilize waste heat Combination with other low-energy pretreatment methods
Biological	Slow process Cost of enzymes	Genetic engineering to produce cost-effective enzymes Use of mixed microbial cultures to enhance breakdown of cell walls Integration with mechanical or chemical pretreatment to reduce process time

Note: In a comprehensive review by Passos et al. (2014); Kendir and Ugurlu (2018), the authors explored various pretreatment methods with their own limitations, necessitating innovative solutions to overcome these challenges.

the mechanisms of interaction of ultrasound with micro-biological biomass and indicates the need for a deeper understanding of these processes (Paul et al., 2023). It is important to consider that the effectiveness of ultrasound pretreatment can vary significantly depending on the specific experimental conditions, including the type and condition of microalgae biomass, ultrasound parameters and treatment environment. This confirms the need for an integrated approach to the selection of pretreatment methods, based on specific purposes and characteristics of the starting material.

Furthermore, the selection of pretreatment methods is determined by the chemical composition of the algae, since the use of low energy processing techniques results in lower yield, the energy balance of the process must be positive for it to have a practical impact (Kendir & Ugurlu, 2018).

Table 1 provides an overview of common pretreatment methods for algae biogas production, highlighting their limitations and the solutions being developed to address these challenges. Solutions often involve optimizing process parameters, integrating different pretreatment methods, and developing new technologies to reduce costs and environmental impact.

#### 4. Developments in pretreatment method

Interest in producing biogas from aquatic plant biomass has grown significantly in recent years due to their high productivity and ability to treat wastewater while reducing pressure on agricultural land. Innovations in biomass pretreatment are aimed at increasing the efficiency of the anaerobic digestion process, increasing biogas yield, and reducing overall production costs.

The choice of pretreatment method and type of algae is key to significantly increasing methane production. Unlike lignocellulose-rich feedstocks, aggressive processing of algae biomass is not required due to its low lignin content. This circumstance today determines the preference for mechanical pretreatment methods due to their simplicity and high efficiency. However, according to this review, the full potential of algal biomass pretreatment has not been fully realized (Montingelli et al., 2015).

Researchers have developed various cost-effective pretreatment methods for the complete conversion of microalgae into various bioenergy products. The initial focus was on extracting the oil from the microalgae biomass to produce biodiesel and processing the remaining solids to produce high-energy biogas. This conversion route has a positive impact on researchers because it is more profitable than direct transesterification or the microalgae anaerobic digestion process. As a result, many researchers have directed their research interests towards improving various aspects of this conversion path (Kannah et al., 2021).

As mentioned earlier, pretreatment methods have problems such as high cost, environmentally harmful by-products, low biogas yield and high energy requirements persist. To further address these challenges, some innovative non-pretreatment strategies are aimed at improving the utilization of readily available and inexpensive feedstocks for biogas production. Strategies discussed include integrated biogas production, innovative digester design, co-digestion and bio-additives. The integrated biogas production strategy solves the cost problem, the innovative digester design strategy solves the inhibition problem, and the co-digestion strategy solves both the low yield and inhibitors problem, and the biomagnification strategy solves the inhibitor problem (Patinvoh et al., 2017).

Therefore, there are some challenges to improve pretreatment technologies for algal biomass for anaerobic digestion. An important aspect is to consider the specific characteristics of different types of microalgae, based on their unique chemical composition. In this context, attention needs to be paid to the development and testing of new reagents, as well as the exploration of combined pretreatment approaches that could include less energy-intensive technologies (de Oliveira et al., 2022).

## 5. Conclusions

1. Structural characteristics and type of microalgae significantly influence the productiveness of diverse pretreatment techniques.
2. Literature studies indicated that an optimal pretreatment method applicable to all microalgae varieties does not exist.
3. The review provided pros and cons of different pretreatment methods, and discussed the impact of each method of biogas production.
4. The thermal and mechanical methods are considered to be more effective in disrupting microalgal cells, where thermal pretreatment is considered to be the most widely studied method.
5. Biological and combined pretreatments require the least energy than other pretreatment methods therefore it considered as the most cost-effective.
6. The innovative non-pretreatment strategies offer promising solutions for enhancing biogas production efficiency by addressing cost, inhibition, low yield, and inhibitor issues through integrated production, digester design, co-digestion, and bio-additives.

## References

- Abusweireh, R. S., Rajamohan, N., Sonne, C., & Vasseghian, Y. (2023). Algae biogas production focusing on operating conditions and conversion mechanisms – A review. *Heliyon*, 9(7), Article e17757. <https://doi.org/10.1016/j.heliyon.2023.e17757>
- Babatunde, E. O., Gurav, R., & Hwang, S. (2023). Recent advances in invasive aquatic plant biomass pretreatments for value addition. *Waste and Biomass Valorization*, 14, 3503–3527. <https://doi.org/10.1007/s12649-023-02186-5>
- Bohutskyi, P., Betenbaugh, M. J., & Bouwer, E. J. (2014). The effects of alternative pretreatment strategies on anaerobic digestion and methane production from different algal strains. *Biore-source Technology*, 155, 366–372. <https://doi.org/10.1016/j.biortech.2013.12.095>
- de Oliveira, M. C., Bassin, I. D., & Cammarota, M. C. (2022). Microalgae and cyanobacteria biomass pretreatment methods: A comparative analysis of chemical and thermochemical pretreatment methods aimed at methane production. *Fermentation*, 8(10), Article 497. <https://doi.org/10.3390/fermentation8100497>
- Dębowski, M., Zieliński, M., Grala, A., & Dudek, M. (2013). Algae biomass as an alternative substrate in biogas production technologies. *Renewable and Sustainable Energy Reviews*, 27, 596–604. <https://doi.org/10.1016/j.rser.2013.07.029>
- González-Fernández, C., Sialve, B., Bernet, N., & Steyer, J. P. (2012a). Thermal pretreatment to improve methane production of *Scenedesmus* biomass. *Biomass and Bioenergy*, 40, 105–111. <https://doi.org/10.1016/j.biombioe.2012.02.008>
- González-Fernández, C., Sialve, B., Bernet, N., & Steyer, J. P. (2012b). Comparison of ultrasound and thermal pretreatment of *Scenedesmus* biomass on methane production. *Bioresource Technology*, 110, 610–616. <https://doi.org/10.1016/j.biortech.2012.01.043>
- Jankowska, E., Sahu, A. K., & Oleskowicz-Popiel, P. (2017). Biogas from microalgae: Review on microalgae's cultivation, harvesting and pretreatment for anaerobic digestion. *Renewable and Sustainable Energy Reviews*, 75, 692–709. <https://doi.org/10.1016/j.rser.2016.11.045>
- Kannah, R. Y., Kavitha, S., Karthikeyan, O. P., Rene, E. R., Kumar, G., & Banu, J. R. (2021). A review on anaerobic digestion of energy and cost effective microalgae pretreatment for biogas production. *Bioresource Technology*, 332, Article 125055. <https://doi.org/10.1016/j.biortech.2021.125055>
- Kavitha, S., Subbulakshmi, P., Banu, J. R., Gobi, M., & Yeom, I. T. (2017). Enhancement of biogas production from microalgal biomass through cellulolytic bacterial pretreatment. *Biore-source Technology*, 233, 34–43. <https://doi.org/10.1016/j.biortech.2017.02.081>
- Kendir, E., & Ugurlu, A. (2018). A comprehensive review on pretreatment of microalgae for biogas production. *International Journal of Energy Research*, 42(12), 3711–3731. <https://doi.org/10.1002/er.4100>
- Marsolek, M. D., Kendall, E., Thompson, P. L., & Shuman, T. R. (2014). Thermal pretreatment of algae for anaerobic digestion. *Bioresource Technology*, 151, 373–377. <https://doi.org/10.1016/j.biortech.2013.09.121>
- Montingelli, M. E., Tedesco, S., & Olabi, A. G. (2015). Biogas production from algal biomass: A review. *Renewable and Sustainable Energy Reviews*, 43, 961–972. <https://doi.org/10.1016/j.rser.2014.11.052>
- Passos, F., Uggetti, E., Carrère, H., & Ferrer, I. (2014). Pretreatment of microalgae to improve biogas production: A review. *Biore-source Technology*, 172, 403–412. <https://doi.org/10.1016/j.biortech.2014.08.114>
- Patinvoh, R. J., Osadolor, O. A., Chandolias, K., Horváth, I. S., & Taherzadeh, M. J. (2017). Innovative pretreatment strategies for biogas production. *Bioresource Technology*, 224, 13–24. <https://doi.org/10.1016/j.biortech.2016.11.083>
- Paul, R., Silkina, A., Melville, L., Suhartini, S., & Sulu, M. (2023). Optimisation of ultrasound pretreatment of microalgal biomass for effective biogas production through anaerobic digestion process. *Energies*, 16(1), Article 553. <https://doi.org/10.3390/en16010553>
- Saad, M. G., Dosoky, N. S., Zoromba, M. S., & Shafik, H. M. (2019). Algal biofuels: Current status and key challenges. *Energies*, 12(10), Article 1920. <https://doi.org/10.3390/en12101920>
- Thompson, T. M., Young, B. R., & Baroutian, S. (2019). Advances in the pretreatment of brown macroalgae for biogas production. *Fuel Processing Technology*, 195, Article 106151. <https://doi.org/10.1016/j.fuproc.2019.106151>
- Xiao, C., Fu, Q., Liao, Q., Huang, Y., Xia, A., Chen, H., & Zhu, X. (2020). Life cycle and economic assessments of biogas production from microalgae biomass with hydrothermal pretreatment via anaerobic digestion. *Renewable Energy*, 151, 70–78. <https://doi.org/10.1016/j.renene.2019.10.145>
- Zabed, H. M., Akter, S., Yun, J., Zhang, G., Zhang, Y., & Qi, X. (2020). Biogas from microalgae: Technologies, challenges and opportunities. *Renewable and Sustainable Energy Reviews*, 117, Article 109503. <https://doi.org/10.1016/j.rser.2019.109503>



## **VANDENS AUGALŲ BIOMASĖS PIRMINIO APDOROJIMO BIODUJŲ GAMYBAI METODŲ APŽVALGA**

**Z. Kazizova, A. Zagorskis**

Santrauka

Pasaulyje didėjanti atsinaujinančių energijos išteklių paklausa paskatino susidomėjimą biodujų gamyba iš vandens augalų biomasės. Šiame darbe nagrinėjami biomasės pirminio apdorojimo metodai, kuriais siekiama padidinti biodujų gamybos procesų efektyvumą. Analizėje pateikiami mechaniniai, cheminiai, biologiniai arba kombinuoti pirminio apdorojimo būdai, įvertinami atitinkamųjų pranašumai, trūkumai ir galima sinergija. Darbas apima atskirų metodų apžvalgą, kad būtų galima nustatyti galimas sinergijas arba nuoseklius pritaikymus, kurie galėtų padidinti biodujų gamybos efektyvumą. Šis darbas prisideda prie energijos gamybos sektoriaus pažangos ir tvaresnių energijos sprendimų, išryškina esamų taikymo metodų spragas bei atveria kelią sprendžiant būsimas energetikos problemas.

**Reikšminiai žodžiai:** anaerobinis skaidymas, biodujos, mikrodumbliai, pirminis apdorojimas, vandens biomasė, bioreaktorius.