

Recent Advances in Microalgal Biorefineries

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The increase in the world population leads to the generation of high amounts of wastewater and the requirement for more energy to satisfy the population's needs. Mainly based on fossil fuels, energy consumption is associated with the emission of greenhouse gases and other pollutants with negative impacts on climate and human health. It is vital to find a solution to mitigate the effects of human activities on the environment. In this context, microalgal biorefineries seem to be a sustainable approach for wastewater treatment and energy production problems. Since microalgae are photosynthetic organisms, they can capture CO₂ from the atmosphere or industrial gaseous streams and produce organic matter and energy. Moreover, microalgae are microorganisms that can accumulate different compounds during their growth, such as proteins, lipids, carbohydrates, and pigments. The lipids and carbohydrates can be used to produce biofuels, and the remaining biomass can also be used to produce biogas. Combining biofuel production with wastewater treatment is possible since microalgae can uptake nutrients (required for their growth) from these effluents, promoting their treatment. Also, the production costs of microalgae (lesser nutrient requirement) and the environmental impact (no freshwater used) are reduced.

This Editorial presents a comprehensive view of the potential use of microalgae for biofuel production and wastewater treatment by applying an energy biorefinery concept. The use of different types of wastewaters to grow microalgae, the application of stress factors to promote compound accumulation, and the economic viability of a biorefinery process were also included in this Editorial.

Mehariya, et al. [1] presented a review study regarding the current (cultivation systems, downstream biomass processing technologies) and future scenarios for integrating wastewater treatment and bioenergy production with microalgae. Wastewater treatment with microalgae can achieve higher pollutant removal efficiencies than most traditional systems. In terms of energy, the potential for biodiesel and biogas production through microalgal biomass was also assessed. González, et al. [2] evaluated the potential use of microalgae to produce biofuels (biodiesel and bioethanol through lipid and carbohydrate analysis) and biogas. A microalgal consortium (*Scenedesmus* sp., *Chlorella* sp., and *Nannochloropsis* sp.) was cultivated in two exterior photobioreactors (a 100 L column and a 400 L flat panel). The biomass productivities achieved were 0.116 g L⁻¹ d⁻¹ with the 100 L column and 0.266 g L⁻¹ d⁻¹ with the 400 L flat panel. The growth rate values ranged between 0.360 (100 L PBR) and 0.312 d⁻¹ (400 L PBR). The produced biomass was collected and used for biogas production in a batch-type digestion process. The CH₄ yield achieved was 296 ± 23 L CH₄ kgVSS⁻¹ with an additional 50% reduction in volatile solids and chemical oxygen demand. The lipid and carbohydrate contents were also analysed to evaluate the potential for their transformation into biofuels. Since the lipid and carbohydrate contents obtained in both PBRs were low, the authors suggest the application of stress factors to



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promote lipid and carbohydrate accumulation as future work. Hawrot-Paw, et al. [3] evaluated the effect of nutrient starvation on microalgal growth, lipid accumulation, and fatty acid profile in the growth of several microalgae species (*Chlorella vulgaris*, *Chlorella fusca*, *Oocystis submarina*, and *Monoraphidium*). The highest biomass concentrations were achieved with a 100% dose of nutrients in all microalgae species, ranging between 292 mg L⁻¹ (for *C. fusca*) and 525 mg L⁻¹ (for *Monoraphidium*), while with a 50% dose of nutrients, the biomass concentration decreased. Moreover, nutrient starvation increased lipid production and reduced the ash content in microalgal cells. Under nutrient stress conditions, the highest lipid content values were 31% (with *C. vulgaris*) and 90% (with *O. submarina*) higher. The authors concluded that the nutrient limitation stress positively affected the functional properties of the biodiesel that could be produced from the biomass; however, the stress effect depends on the species used. As a strategy to improve biomass productivity and lipid content, Nie, et al. [4] first tested different light intensities (10,000, 15,000, 20,000, and 25,000 lux) on the growth of *Golenkinia* SDEC-16. The highest biomass concentrations (5.18 and 5.27 g L⁻¹) were obtained with the highest light intensities (20,000 and 25,000 lux). However, the highest lipid contents (between 41.8% and 42.3%) were achieved with the lowest light intensities (10,000 and 15,000 lux). In order to improve biomass productivity and simultaneously enhance lipid accumulation, the authors introduced another stress factor. Two initial NaNO₃ concentrations (1 mM and 9 mM) were tested using BG-11 as a basal medium under 10,000 lux and 25,000 lux. The authors concluded that the optimal conditions were 25,000 lux using 9 mM of NaNO₃ since the lipid content increased, reaching 44.6%, with no effects on biomass concentration. Salinity stress was also tested by using NaCl concentrations of 20, 160, 320, and 640 mM to enhance lipid production even further under the previously defined optimal conditions. The authors concluded that the optimal conditions to enhance both biomass productivity and lipid accumulation were 25,000 lux, 9 mM NaNO₃ concentration, and 20 mM NaCl concentration. Under these conditions, the biomass concentration obtained was 6.65 g L⁻¹ and the lipid content was 54.4%.

Several authors have evaluated the potential of combining microalgal wastewater treatment with bioenergy production. Spennati, et al. [5] used a co-culture of *Chlorella vulgaris* and *Arthrospira platensis* to treat winery wastewaters and produce biomass as a potential feedstock for biodiesel. Wastewaters were collected from three different stages of the wine production process, diluted with Bold's Basal Medium up to 10%, 20%, 50%, and 100% (v/v), and used for microalgal growth in 200 mL photobioreactors. The authors observed the highest biomass productivity (0.66 g L⁻¹ d⁻¹) when using 20% (v/v) wastewater collected from a second washing tank. Wastewater retrieved from the filtration equipment and diluted to 50% (v/v) resulted in the highest lipid content (22%). Moreover, the co-culture was able to promote a chemical oxygen demand and polyphenol removal of over 92% and 50%, respectively, in all experimental conditions. Therefore, the authors concluded that winery wastewaters could be employed as an economic culture medium for microalgae, resulting in biomass that can be used as a feedstock for sustainable biofuel production. Hawrot-Paw, et al. [6] evaluated the growth of *Monoraphidium* in saline aquaculture-derived wastewater. Microalgae from this genus were cultured in a 14 L tubular photobioreactor, yielding 0.46 g L⁻¹ d⁻¹ biomass productivity and 18.53% lipid content. Moreover, this microalga efficiently removed nitrogen and phosphorus from the wastewater, with 82.6% and 99.06% removal efficiencies, respectively. A control assay was conducted using F/2 culture medium, leading to a higher lipid content but much lower biomass concentrations. Hence, the authors highlighted that a compromise between biomass production and lipid content is important for biodiesel production. In summary, the results suggest great potential in combining biomass production for valorisation into biodiesel with nutrient removal from aquaculture wastewater. In a large-scale process, Díez-Montero, et al. [7] used a mixture of 90% (v/v) agricultural runoff with 10% (v/v) domestic wastewater as a growth medium for a mixed microalga culture dominated by cyanobacteria (*Synechococcus* sp. and *Pseudanabaena* sp.) and green algae. Microalgae were cultured in 11.7 m³ tubular photobioreactors, and the biomass was used for anaerobic digestion and biogas production in a

demonstration-scale biorefinery plant. Biomass productivities varied between $7 \text{ g m}^{-3} \text{ d}^{-1}$ in winter and spring and $43 \text{ g m}^{-3} \text{ d}^{-1}$ during summer and at the beginning of autumn. The produced biomass was collected and pre-treated through a thermal process ($75 \text{ }^\circ\text{C}$ for 10 h) to increase the soluble organic matter and improve the anaerobic digestion rate and extent, consequently increasing the methane yield. The produced biogas had a methane content of 76%, yielding up to $0.38 \text{ L CH}_4 \text{ gvs}^{-1}$. In a final step, biogas was upgraded to biomethane in an adsorption column fed with microalgal broth from the photobioreactors, resulting in a methane content of up to 98.8%. In conclusion, the results from this study indicate that microalgal cultures can be utilised in large-scale biorefinery processes, in which wastewater is used as a nutrient source, biogas is produced and upgraded to biomethane, and the resulting digestate can be applied as a biofertiliser.

To evaluate the economic viability of microalgal production, Valdovinos-García, et al. [8] performed a techno-economic analysis (energy consumption and operating costs) of microalgal biomass production, analysing the CO_2 uptake from gaseous effluents of a thermoelectric plant. Several scenarios were defined and simulated, considering microalgal cultivation, harvesting, and drying. Taking into account 1 ha of cultivation area and productivity of $12.7 \text{ g m}^{-2} \text{ d}^{-1}$, 102 tons of CO_2 can be captured per year. The estimated operating costs were $\$4.75\text{--}6.55/\text{kg}$ of dry biomass.

Finally, to enhance the economic viability, process integration is essential to reduce production costs. Also, the extraction of several compounds from the produced biomass (biorefinery concept) increases the profitability and sustainability of the process. Solis, et al. [9] developed a multiobjective optimisation model that aims to reduce the costs and the environmental impact of an algal biorefinery, adopting the principle of resource recovery and recirculation. This model was validated with a case study: a biorefinery that produces biodiesel, glycerol, biochar, and fertiliser. The sensitivity analysis showed that demand fluctuations and process unit efficiencies strongly impact the optimal results.

Microalgae have several advantages that make them valuable feedstocks. Therefore, it is important to understand the operational and environmental conditions that affect microalgal growth and productivity. The use of stress factors is gaining increasing interest since it promotes the accumulation of compounds of interest. Also, further research regarding the optimisation of lipid and carbohydrate extraction for biodiesel and bioethanol is needed to make the process more cost-effective. By applying the biorefinery concept, it is possible to make the most of microalgae's potential by treating wastewaters and simultaneously producing biofuels and biofertilisers, as well as capturing CO_2 present in the atmosphere or industrial gaseous streams, making the process more profitable.

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References

1. Mehariya, S.; Goswami, R.K.; Verma, P.; Lavecchia, R.; Zuurro, A. Integrated approach for wastewater treatment and biofuel production in microalgae biorefineries. *Energies* **2021**, *14*, 2282. [[CrossRef](#)]
2. González, J.F.; Cuello, T.B.; Calderón, A.J.; Calderón, M.; González, J.; Carmona, D. Cultivation of autochthonous microalgae for biomass feedstock: Growth curves and biomass characterization for their use in biorefinery products. *Energies* **2021**, *14*, 4567. [[CrossRef](#)]
3. Hawrot-Paw, M.; Ratomski, P.; Koniuszy, A.; Golimowski, W.; Teleszko, M.; Grygier, A. Fatty acid profile of microalgal oils as a criterion for selection of the best feedstock for biodiesel production. *Energies* **2021**, *14*, 7334. [[CrossRef](#)]
4. Nie, C.; Jiang, L.; Hou, Q.; Yang, Z.; Yu, Z.; Pei, H. Heuristic Optimization of Culture Conditions for Stimulating Hyper-Accumulation of Biomass and Lipid in *Golenkinia* SDEC-16. *Energies* **2020**, *13*, 964. [[CrossRef](#)]
5. Spennati, E.; Casazza, A.A.; Converti, A. Winery wastewater treatment by microalgae to produce low-cost biomass for energy production purposes. *Energies* **2020**, *13*, 2490. [[CrossRef](#)]
6. Hawrot-Paw, M.; Koniuszy, A.; Gałczyńska, M. Sustainable production of *Monoraphidium* microalgae biomass as a source of bioenergy. *Energies* **2020**, *13*, 5975. [[CrossRef](#)]
7. Díez-Montero, R.; Vassalle, L.; Passos, F.; Ortiz, A.; García-Galán, M.J.; García, J.; Ferrer, I. Scaling-up the anaerobic digestion of pretreated microalgal biomass within a water resource recovery facility. *Energies* **2020**, *13*, 5484. [[CrossRef](#)]
8. Valdovinos-García, E.M.; Barajas-Fernández, J.; Olán-Acosta, M.d.l.Á.; Petriz-Prieto, M.A.; Guzmán-López, A.; Bravo-Sánchez, M.G. Techno-economic study of CO₂ capture of a thermoelectric plant using microalgae (*Chlorella vulgaris*) for production of feedstock for bioenergy. *Energies* **2020**, *13*, 413. [[CrossRef](#)]
9. Solis, C.M.A.; San Juan, J.L.G.; Mayol, A.P.; Sy, C.L.; Ubando, A.T.; Culaba, A.B. A multi-objective life cycle optimization model of an integrated algal biorefinery toward a sustainable circular bioeconomy considering resource recirculation. *Energies* **2021**, *14*, 1416. [[CrossRef](#)]