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REVIEW ARTICLE

Microalgae-Based Treatment of Winery Wastewater

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Abstract

Winery wastewater (WWW) is a high-strength effluent characterized by high organic load, nutrient content, and seasonal variability, posing significant environmental challenges. This review critically evaluates microalgae-based systems for WWW treatment, addressing their performance, scalability, and role within circular bioeconomy frameworks. Analysis of recent studies shows that microalgae-based systems can achieve high pollutant removal efficiencies, including up to ~90–92% chemical oxygen demand removal, high removal of total organic carbon and nitrogen (typically above 80%), and up to 91–95% ammonium removal. These systems also enable the production of biomass rich in proteins (up to ~58.8% dry weight) and valuable compounds such as pigments, supporting multiple valorization pathways. A key finding is that, despite strong technical performance, large-scale implementation remains limited. The main constraints are associated with wastewater variability, presence of inhibitory compounds, and operational challenges in biomass recovery, particularly energy-intensive harvesting and downstream processing. Techno-economic and life cycle analyses indicate that standalone systems are rarely economically viable under current conditions, and that feasibility depends on integration into multiproduct biorefineries and existing infrastructures. Regulatory uncertainty and the need for stakeholder acceptance further constrain deployment. Overall, microalgae-based WWW treatment represents a promising but system-dependent solution, requiring integrated technological, economic, and policy frameworks to achieve scalable implementation.

Keywords Microalgae · Winery effluents · Circular economy · Bioproducts · Bioeconomy

Introduction

Wine production is carried out in rural regions worldwide, including Mediterranean and Atlantic areas (Italy, France, Spain, Portugal, Greece, Turkey), Central and Eastern Europe (Germany, Hungary, Romania, Bulgaria), as well as other major wine-producing regions such as Australia, New Zealand, South Africa, the United States of America, Chile, Argentina, and China. In 2023, global wine production reached 237.3 million hectoliters (MhL), according to the International Organization of Vine and Wine [1].

Wine production typically involves several stages, including grape maceration, pressing, alcoholic and malolactic fermentation (depending on the wine type), maturation, stabilization, filtration, and bottling. These steps require significant amounts of water, energy, fertilizers, and organic additives, and generate large volumes of waste streams (such as pips, skins, grape marc and wine lees), wastewater, and greenhouse gases (e.g., carbon dioxide and volatile organic compounds) [2].

Wine lees, consisting primarily of yeast cells, tartaric salts, and adsorbed organic compounds formed during fermentation and wine clarification processes, represent a significant by-product of the winemaking industry. Their management poses additional environmental and operational challenges, but they also constitute a potential resource for valorization within integrated biorefinery approaches [3].

WWW consists of spilled wine, washing water, cooling water, cleaning chemicals, and leachates from solid by-products. More than 60% of these effluents are generated during the harvest season, which typically lasts up to three months and places considerable stress on wastewater treatment systems. During the winemaking process, wastewater generation can reach up to 14 L per liter of wine produced, depending on the scale of the facility, the type of wine

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(e.g., red, white, or specialty wines), and the winemaking and cleaning technologies used—including water softener regeneration brine [4].

Wine residues provide nutrients that promote the proliferation of yeasts and bacteria, making deep cleaning and sanitation of tanks, pumps, hoses, floors, bottles, and transport boxes essential for preserving wine quality [5]. However, these operations also contribute to generating large volumes of wastewater, proportional to wine production scale.

If discharged without treatment, WWW can cause significant environmental damage, notably by promoting eutrophication in water bodies (e.g., wetlands, rivers, streams, ponds), due to rapid oxygen consumption that depletes dissolved oxygen levels and endangers aquatic and amphibious life. Although WWW can be reused for irrigation, its composition may alter soil properties such as pH, color, and electrical conductivity, due to the release of inorganic and organic ions [5, 6].

Conventional treatment of WWW typically involves physical, chemical, and biological processes to remove major pollutants, often supplemented by advanced oxidation processes for color and recalcitrant compound removal [7, 8]. The activated sludge process is commonly applied and effective at removing contaminants, but it has high operational costs due to significant sludge production and the energy required to maintain aerobic conditions. Moreover, the resulting by-products generally lack commercial value [9].

For these reasons, alternative approaches are being explored for the simultaneous treatment and valorization of WWW. Recent studies have demonstrated the successful application of microalgae in treating WWW showing this approach to be both effective and economically viable. Microalgae can grow on a variety of wastewaters—including WWW—requiring low energy input, effectively removing heavy metals, and generating minimal waste sludge [10, 11].

Table 1 Typical ranges of key physicochemical parameters found in WWW [2, 5, 8, 12–16]

Parameter	Unit	Reported range
pH	–	3.0–6.0
COD	mg O_2 L^{-1}	300–49,000
BOD_5	mg O_2 L^{-1}	1000–13,000
Total organic carbon (TOC)	mg C L^{-1}	500–7,000
Total nitrogen (TN)	mg L^{-1}	8–415
Total phosphorus (TP)	mg L^{-1}	2–280
Total solids (TS)	mg L^{-1}	5000–18,000
Suspended solids (SS)	mg L^{-1}	30–8,600
Volatile solids (TVS)	mg L^{-1}	6000–20,000
Electrical conductivity	mS cm^{-1}	0.3–5.6
Phenolic compounds	mg L^{-1}	0.5–3,500
Total sugars	mg L^{-1}	1000–8000

This review highlights the advantages of using WWW as a substrate for microalgae cultivation, offering a dual benefit of wastewater treatment and biomass production. It examines the cultivation strategies and system configurations applied in this context, as well as the potential for generating high-value bioproducts within an integrated biorefinery framework. Emphasis is placed on the full process chain—from WWW characteristics to biomass valorization—underlying the importance of holistic and sustainable approaches.

WWW Characteristics

Winery wastewater (WWW) characteristics are strongly influenced by multiple factors, including climate conditions, local water quality, grape variety, wine type, and operational practices within the winery, particularly cleaning procedures and the use of chemical agents [2, 8]. Additionally, seasonal variability plays a significant role in determining WWW composition. During the harvest and crushing period, typically occurring in autumn, large volumes of wastewater are generated with high organic load, elevated concentrations of suspended solids, and strong odors due to grape processing activities. In contrast, during the off-season, wastewater is mainly derived from cleaning and maintenance operations, resulting in lower volumes and reduced organic load [5, 8].

WWW is typically acidic, with pH values generally ranging between 3 and 6, mainly due to the presence of organic acids such as tartaric, malic, lactic, and citric acids [2, 8, 12]. It is characterized by a high organic content, with chemical oxygen demand (COD) typically ranging from 300 to 49,000 mg L^{-1} and biochemical oxygen demand (BOD_5) between 1,000 and 13,000 mg L^{-1} (Table 1) [2, 12, 13]. Nutrient concentrations are generally low to moderate, with total nitrogen (TN) typically ranging from 8 to 415 mg L^{-1} and total phosphorus (TP) from 2 to 280 mg L^{-1} [8, 12, 14]. In addition, WWW contains significant amounts of ethanol, sugars (1000–8000 mg L^{-1}), organic acids, and phenolic compounds (0.5–3,500 mg L^{-1}), contributing to its complex and variable composition (Table 1) [2, 12].

The environmental impact of WWW is primarily associated with its high organic load, mainly derived from readily biodegradable compounds such as sugars and ethanol, as well as more recalcitrant substances like polyphenols. These phenolic compounds are of particular concern due to their toxicity to microorganisms, plants, animals, and humans, even at low concentrations, and their resistance to biodegradation [2, 8]. Furthermore, the presence of suspended solids (30–8600 mg L^{-1}, Table 1) and the large volumes generated (typically 0.2–4 L per liter of wine produced) contribute significantly to its environmental impact [15, 16].

The acidic nature of WWW may also negatively affect soil quality when discharged without treatment, by altering nutrient availability—particularly phosphorus and calcium—and reducing populations of beneficial microorganisms. Additionally, improper management of WWW can lead to the generation of unpleasant odors and gaseous emissions, mainly due to the degradation of organic matter under uncontrolled conditions [2].

Discharging Untreated WWW in the Environment

Discharging WWW into the environment often causes serious issues like eutrophication and algal blooms due to its high nitrogen and phosphorus content. These nutrients act as a potent fertilizer for algae and other aquatic plants, leading to rapid and uncontrolled growth (Fig. 1). While algae are naturally present, a bloom can block sunlight from reaching deeper waters, killing submerged plants. The algae eventually die and are decomposed by bacteria, which consume large amounts of dissolved oxygen, promoting hypoxia or anoxia (oxygen depletion). This lack of oxygen can kill fish and other aquatic life in a phenomenon often referred to as a "dead zone" [17]. In addition, ethanol present in WWW can be directly toxic to aquatic organisms, and its degradation by microorganisms further depletes oxygen. Phenolic compounds are also toxic to many aquatic species, affecting their growth, reproduction, and survival. The combination of oxygen depletion and toxic compounds can decimate aquatic biodiversity, leading to the loss of sensitive species and a shift towards less diverse, more resilient communities [16].

Discharging WWW onto land can have a detrimental effect on soil health and groundwater quality, since winemaking process uses various salts for cleaning and processing. These salts, particularly potassium, accumulate in WWW. Repeated application of this wastewater to soil can lead to its salinization, inhibiting plant growth, alter soil structure, and reduce agricultural productivity [18]. Moreover, as above stated, WWW typically has a very low pH (between 3.0 and 5), making it highly acidic. Continuous discharge of this acidic effluent can lower the soil's pH, a process known as acidification. Soil acidification can reduce nutrient availability to plants and increase the solubility of toxic metals like aluminum and manganese, which can harm crop roots and contaminate groundwater [19].

WWW carries a high concentration of microorganisms, including bacteria and yeasts from the fermentation process. When discharged on land, these microbes can outcompete and disrupt native soil microbial communities essential for nutrient cycling and soil health. They can also degrade organic matter and consume oxygen in the soil, altering its physical and chemical properties and leading

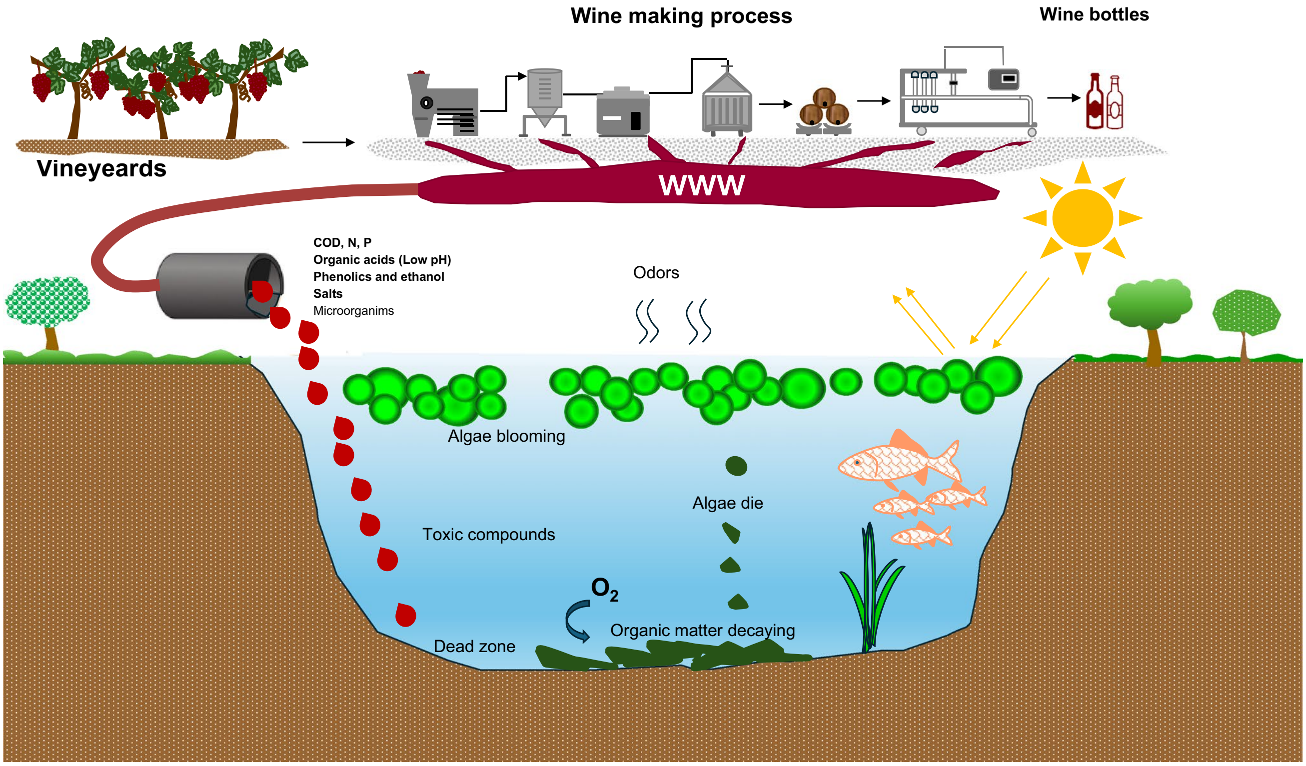

Fig. 1 Environmental impacts of discharging untreated WWW

to foul-smelling gases due to anaerobic decomposition. When released into lakes and rivers, the high concentration of microorganisms in WWW, along with the organic load, leads to rapid oxygen depletion as they decompose the waste. This can create "dead zones," killing fish and other aquatic life. The introduced microbes can also become invasive, disrupting the native microbial community structure and forming extensive biofilms [17].

Why Microalgae to Treat WWW?

Current methods for treating WWW include physical, chemical, and biological approaches. Physical methods often involve screens and filters to remove large solids, while chemical treatments use coagulation and flocculation with metal salts to remove suspended solids and phosphorus [20]. While these methods are effective, they can be costly and generate large amounts of chemical sludge that is difficult to dispose of. Biological methods, in contrast, are the most sustainable and efficient for this proposal [1]. They use microorganisms, like bacteria and microalgae, to degrade the high organic load and remove nutrients, such as nitrogen and phosphorus. The key benefits of biological treatment are its lower operational costs, the ability to recycle nutrients, and the production of a valuable biomass that can be used as a biofertilizer, a biostimulant or a biopesticide, transforming a waste product into a valuable resource and promoting a circular economy. Aerobic and anaerobic biological systems are usually employed. The anaerobic biological treatment uses anaerobic digestors which may work under high organic loading rates. Due to the absence of aeration and the low sludge production, anaerobic treatment generally incurs lower operational costs compared with aerobic processes and can handle higher organic loads, making it more suitable for concentrated wastewaters. In addition to treating effluents, anaerobic digestion also produces biogas (a mixture of methane and CO_2), enabling simultaneous energy recovery. However, undesirable odors may arise from the accumulation of volatile fatty acids (VFAs), which require careful pH control [21]. Furthermore, hydrogen sulfide (H_2S) can be generated through the reduction of sulfate and sulfite present in WWW by sulfate-reducing bacteria, leading to odor problems, equipment corrosion, and potential inhibition of methanogenesis, thus representing a significant operational challenge [22].

In contrast, aerobic digestion requires continuous aeration, making it energy-intensive, and produces large amounts of sludge that require further handling and disposal. Nonetheless, it is highly effective in removing nutrients, which makes it a widely adopted treatment method.

An emerging alternative that addresses several limitations of both systems is microalgae-based treatment. Microalgae can grow under autotrophic (using sunlight and CO_2) and mixotrophic or heterotrophic (utilizing organic matter) modes, all aerobic processes. This metabolic flexibility enables them to thrive in complex wastewaters such as winery effluents. Through photosynthesis, microalgae release oxygen that supports aerobic degradation of organic pollutants, while simultaneously assimilating nutrients such as nitrogen and phosphorus. Moreover, the resulting biomass may be enriched in proteins, pigments, and other valuable compounds, making it suitable for a wide range of applications such as biofertilizers, biostimulants, biopesticides, feed ingredients, or bioproduct precursors. This dual role—wastewater treatment and resource recovery—positions microalgae as a sustainable and economically attractive solution for WWW treatment [17, 23, 24]. In addition, microalgae cells assimilate nutrients directly for growth, being particularly efficient in removing nitrogen and phosphorus, which is especially valuable for WWW rich in organic nitrogen and phosphorus. Furthermore, since microalgae capture CO_2 during photosynthesis, they contribute to carbon sequestration, providing a means to mitigate CO_2 emissions from various industrial sources, including power generation and fermentation-based facilities. Microalgal cultivation systems primarily rely on light as the energy source for photosynthesis, although additional energy is required for aeration and agitation to ensure adequate mixing and gas exchange. These systems can be operated in open ponds or photobioreactors. The wastewater treatment by microalgae produces less sludge compared to conventional biological treatments, being the algal sludge easier to dewater and manage than the sludges produced in other biological treatments. Importantly, a few microalgal species can degrade or transform phenols, tannins, ethanol, and other organic compounds found in wastewater [25]. This capability offers significant advantages over WWW conventional treatments, which often struggle to remove these complex and recalcitrant compounds efficiently. Consequently, microalgal treatment can achieve improved pollutant removal with lower chemical inputs and reduced formation of harmful by-products, making it a more sustainable and cost-effective option for WWW management, compared to conventional treatment methods (Table 2).

Symbiotic relationships between microalgae and bacteria and yeasts present in WWW can enhance nutrient removal, particularly chemical oxygen demand (COD), through algal–bacterial consortia [24] or algal-yeast consortia [26] (Fig. 2). Such consortia offer promising potential to improve nutrient removal in wastewater treatment via mutualistic interactions and synergistic effects [27]. Indeed, photosynthetic microalgae produce dissolved oxygen as a byproduct,

Table 2 Comparison between biological treatment methods for WWW [8, 9, 12, 13]

Aspect	Anaerobic treatment	Aerobic treatment	Microalgae-based treatment
Energy requirement	Low (no aeration)	High (requires aeration)	Low (uses sunlight as energy source)
Sludge production	Low	High	Low (easier to dewater and manage)
Nutrient removal efficiency	Moderate	High	High (direct assimilation of N and P by microalgae)
Pollutant removal	Effective for COD	Effective for COD and nutrients	Effective for COD, N, P, polyphenols, tannins, ethanol
CO_2 capture/emission	CO_2 generation from biogas	Net CO_2 emitter	CO_2 uptake through photosynthesis
Bioproducts	Biogas	Sludge (limited valorization)	Biomass for biofuels, bioproducts, feed, fertilizers, bioplastics
Operational complexity	Moderate (requires pH/VFA monitoring)	High (requires continuous aeration and sludge handling)	High (requires light/CO_2 and harvesting control)
Odor issues	High (volatile organic acids formation)	Low	Low
System types	Anaerobic digesters	Activated sludge, biofilters	Open ponds, photobioreactors
Environmental footprint	Moderate (odor, GHG emissions, leakage risk)	High (energy-intensive aeration)	Low–moderate (CO_2 capture; energy for harvesting)
Integration potential	Moderate (Biogas recovery)	Moderate (nutrient polishing)	High (CO_2 capture, co-product recovery, integration with vineyards/wineries
Microbial synergies	Anaerobes (no synergy with algae)	Bacteria-dominated species	Algae–bacteria consortia (enhanced treatment and biomass production)
Sustainability outlook	Moderate (energy recovery potential)	Low–moderate (high energy demand)	High (resource recovery and circular bioeconomy)
Process stability	Moderate (sensitive to pH and toxic compounds)	Moderate (sensitive to pH and toxic compounds)	Moderate (sensitive to pH and toxic compounds)
Capital cost (CAPEX)	Moderate–high (digesters required)	Moderate–high (digesters required)	Moderate–high (digesters required)
Operational Cost (OPEX)	Low–moderate	Low–moderate	Low–moderate
Need for Skilled Operation	Moderate	Moderate to high	High (biological and operational complexity)
Monitoring Requirements	Moderate (pH, VFAs, alkalinity)	Moderate (DO, sludge age, nutrients)	Moderate (light, nutrients, contamination control)
Full-Scale Implementation	Widely implemented	Widely implemented	Limited full-scale implementation (mostly pilot-scale)

supporting aerobic degradation by heterotrophic microorganisms, which reduces energy costs and minimizes pollutant volatilization risks associated with mechanical aeration. In return, bacterial and yeast heterotrophic metabolism supplies inorganic carbon (e.g., CO_2) and essential secondary metabolites such as vitamins that promote autotrophic algal growth [28]. A symbiotic relationship between algae and bacteria has been described for *Chlorella* and *Scenedesmus* microalgae, which are vitamin B12-auxotrophic microorganisms that rely on other organisms, like bacteria, for their supply of this essential vitamin. The organic content in wastewater systems supports diverse bacterial populations, such as *Pseudomonas*, *Paracoccus*, and *Bacillus*, which produce and release vitamin B12 and other growth factors into the surrounding water which will be uptaken by the vitamin B12-auxotrophic microalgae, creating a symbiotic relationship where the microalgae can thrive [29]. However, designing effective consortia requires thorough understanding of their interaction dynamics, especially under varying wastewater compositions and operational conditions.

Several studies have concluded that microalgae-based treatment of WWW is an eco-friendly and renewable process [23, 30, 31]. Moreover, it can be integrated with solar energy and land use on-site (e.g., at vineyards or wineries), potentially reducing the ecological footprint of wine production facilities [31].

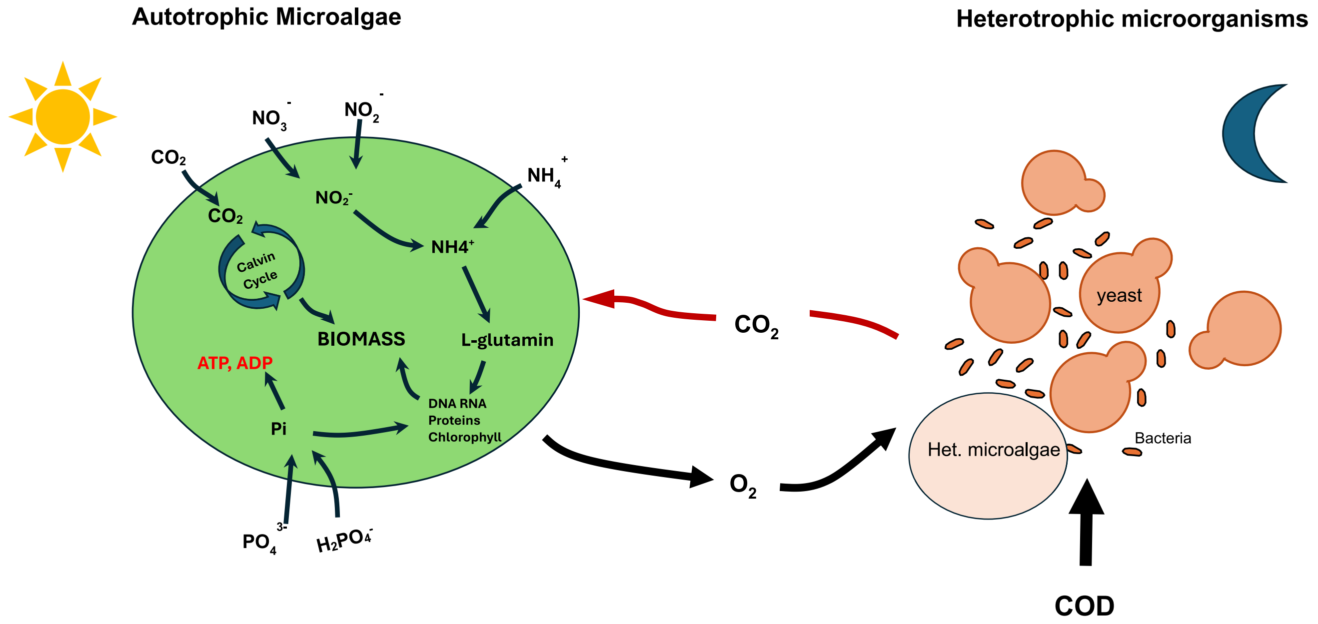

Fig. 2 Autotrophic-heterotrophic symbiosis in WWW treatment

Microalgae Species Used for WWW Treatment

Several microalgal species have been investigated for the treatment of WWW due to their capacity to remove nutrients and generate valuable biomass. However, only a limited number of studies have been published to date, highlighting the need for further systematic research to fully assess their potential and optimize large-scale applications. Species from the genera *Chlorella*, *Desmodesmus*, *Heterochlorella*, and *Arthrospira* have shown to be promising for this application (Table 3). Most of the studies reported to date have been conducted at laboratory scale, with only a limited number extending to pilot-scale systems, and no full-scale applications identified. In addition, relevant operational parameters such as hydraulic retention time (HRT), as well as the use of diluted or pretreated wastewater, vary significantly among studies and influence treatment performance.

Casazza et al. [32] investigated the growth of Arthrospira platensis and Chlorella vulgaris in diluted winery wastewater (20–60% v/v) under laboratory-scale batch conditions using vertical glass bubblers. The results demonstrated effective nitrogen removal (up to 90%) and highlighted the potential of WWW as a culture medium for microalgal biomass production, particularly under mixotrophic conditions.

Marchão et al. [33] used WWW as culture medium under laboratory-scale batch conditions, employing diluted effluents and, in some cases, synthetic media as control. Four species, including Chlorella vulgaris, were tested under mixotrophic and heterotrophic conditions. C. vulgaris showed the best performance under mixotrophic conditions, removing over 90% of total organic carbon (TOC) and total nitrogen (TN), with COD removal reaching 92%. The highest biomass productivity reached 147.5 mg L^{-1} d^{-1}, alongside reductions in polyphenols (49%) and phosphate (40%). Biomass grown on WWW also showed enhanced carotenoid content. However, as the treated effluent still did not meet discharge standards, a tertiary photocatalytic treatment (UV LEDs/TiO_2) was applied, achieving an additional 40% COD removal (of the residual load) and 80% polyphenol removal.

The use of mixed microalgal cultures has been described as a strategy for enhancing the efficiency and versatility of WWW treatment. Spennati et al. (2020 and 2022) [34, 35] explored the use of Chlorella vulgaris and Arthrospira platensis mixed cultures for WWW treatment in two complementary studies. Spennati et al. (2020) [34] used this co-culture to treat three different WWW from different steps of the wine production process, having reported biomass and lipid productivities of 0.66 g /Ld and lipids of 7.10 ± 0.22 g Lipid/100 Ld, when using 20% of second washing WWW after 4 days of treatment. COD and polyphenol content of the three different wastewaters were reduced by the co-culture by more than 92% and 50%, respectively. Spennati et al. [35] further investigated thesame co-culture in three different laboratory-scale systems: a tubular photobioreactor, a column photobioreactor, and an open pond, all operated in batch mode. Biomass concentrations of 5.54 g L^{-1}, 3.90 g L^{-1}, and 2.19 g L^{-1} were obtained, respectively. Protein and carbohydrate extraction yielded 10.95% (w/w) protein and 3.95% (w/w) carbohydrates. In addition, the system achieved removal efficiencies of approximately 84% for COD and 50–60% for phenolic compounds.

Table 3 Microalgal strains used for WWW treatment: biomass production, bioproducts, and nutrient removal

Culture medium	Microalgal strains	Cultivation conditions	Biomass	Other products	Nutrients removal (%)	References
WWW, diluted to 20, 40 and 60% (v/v);	*Arthrospira platensis* UTEX 1926*; Chlorella vulgaris* CCAP 211	Mixotrophic mode 250 mL vertical glass bubbler, batch mode, lab-scale	Concentration: 1.55 g L^{-1} Productivity: 0.103 g L^{-1} d^{-1}	Fatty Acid content: 13.6% w/w	Total C: 33 Total N: 90	[32]
WWW, diluted with Bold Basal medium (10, 20, 50 and 100% v/v)	*Chlorella vulgaris* CCAP 211and *Arthrospira platensis* UTEX 1926 mixed cultures	Mixotrophic mode 200 − mL bubblers; batch mode, lab-scale,	Productivity: 0.66 g L^{-1} d^{-1}	Lipids: 7.10 ± 0.22% (w/w)	COD: 92 Polyphenol: 50	[33]
Diluted WWW	*Scenedesmus obliquus* ACOI 204/07*; Chlorella vulgaris* INETI 58*; Auxenochlorella protothecoides* UTEX 25*; Arthrospira (Spirulina) maxima* LB 2342	Mixotrophic and heterotrophic modes Erlenmeyer flasks (100 mL-1 L), batch mode, lab-scale	Productivity: 147.5 mg L^{-1} d^{-1}	Carotenoids: 8,7 mg g^{-1}	COD: 92 Total N: 91 Phenols: 49 PO_4–P: 40	[34]
WWW, diluted with Bold Basal medium (20% v/v)	*Chlorella vulgaris* CCAP 211and *Arthrospira platensis* UTEX 1926 mixed cultures	Mixotrophic mode Tubular photobioreactor (5 L), open pond (0.5 L) and column photobioreactor (7 L), batch mode, lab-scale	Concentration: TP: 5.54 g L^{-1} CB: 3.90 g L^{-1} OP: 2.19 g L^{-1}	Protein: 10.95% (w/w) Carbohydrates: 3.95% (w/w)	COD: 84 Phenols: 50–60	[35]
WWW, diluted (50–70% v/v; acclimation and operation) and anaerobically pretreated (SBR)	*Chlorella sorokiniana* UTEX 1230	Batch mode (initial tests); sequencing batch reactor (SBR, 2 L), semi-continuous operation (HRT 4 d–36 h), lab-scale		Protein: 58.8% (w/w) Lutein: 1075 mg kg^{-1} β-carotene: 45.5 mg kg^{-1} Tocopherol: 131.2 mg kg^{-1}	COD: 78 ± 9 NH_4-N: 95 ± 9	[10]
Liquid fraction of anaerobic digestate from winery residues (wine lees) and activated sludge; not diluted	*Chorella vulgaris*	Mixotrophic mode; tubular photobioreactor (4 L; 1 L for HRT optimization), semi-continuous mode (HRT 10 d), lab-scale	Concentration: 2.28 g L^{-1}	Lipids: 20% (w/w) Starch: 6.40% Protein: 44.39%	COD: 33.01 NH_4-N: 84.58 Polyphenols: 38.06	[36]
Raw WWW	*Desmodesmus sp. MAS1; Heterochlorella sp. MAS3*	Mixotrophic mode Lab-scale: tubular photobioreactor (0.5 L) and aquaria (5 L), batch mode; Pilot-scale: High-Rate Algal Pond (50 L), semi-continuous mode (HRT 2–3 d	Concentration: 0.825 g L^{-1}		Total organic carbon: 81 Total N: 85 Total P: 73.5	[31]
Red wine effluents (pre-filtered), diluted to 1–30% (v/v); control: synthetic medium	*Chlorella vulgaris* A4F_Ma016	Mixotrophic mode Erlenmeyer flasks (250 mL), batch mode; photobioreactor (1.7 L), semi-continuous mode (incremental addition of red wine effluents at 1–30% v/v), lab-scale	Concentration: 0.68 g L^{-1}		Sugars: 78.3 Phenols: 98.0	[37]

Zkeri et al. [10] evaluated *Chlorella sorokiniana* for WWW treatment through both batch and sequencing batch reactor (SBR) experiments at laboratory scale. Diluted wastewater (50–70% v/v) was used during acclimation and operation, and anaerobically pretreated WWW was also assessed. Semi-continuous SBR systems operated with HRT values ranging from 4 d to 36 h. A two-stage configuration achieved 85% COD and 91% NH_4-N removal, while a one-stage system using pretreated WWW achieved 78% COD and 95% NH_4-N removal. Biomass analysis revealed high protein content (up to 58.8%) and valuable compounds such as lutein, β-carotene, and tocopherols.

Scarponi et al. [36] explored the use of the liquid fraction of anaerobic digestates derived from winery residues (lees and activated sludge) as culture medium for *Chlorella vulgaris*. Cultivation was carried out at laboratory scale in tubular photobioreactors operated in semi-continuous mode with an HRT of 10 d under mixotrophic conditions. Biomass concentrations reached 2.28 g L^{-1}, with a composition of 20% (w/w) lipids, 6.40% starch, and 44.39% protein. In addition, removal efficiencies of 84.58% for total ammonia, 33.01% for COD, and 38.06% for polyphenols were achieved. This approach highlights an alternative pathway integrating anaerobic digestion and microalgal cultivation for resource recovery.

Praveen et al. [31] assessed the performance of two acid-tolerant strains, *Desmodesmus* sp. MAS1 and *Heterochlorella* sp. MAS3, using raw, unfiltered and undiluted winery wastewater under mixotrophic conditions. Laboratory-scale batch experiments were conducted in aquaria and tubular photobioreactors, while pilot-scale operation was performed in a high-rate algal pond (HRAP) under semi-continuous conditions, with an HRT of 2–3 d. Biomass concentrations reached 0.825 g L^{-1}, and removal efficiencies of 81% for total organic carbon, 85% for total nitrogen, and 73.5% for total phosphorus were achieved. These results demonstrate efficient nutrient removal and highlight the scalability of microalgal systems under non-diluted conditions.

Sousa et al. [37] investigated the growth of *Chlorella vulgaris* A4F_Ma016 using pre-filtered red wine effluents diluted to 1–30% (v/v), under mixotrophic conditions. Experiments were conducted in Erlenmeyer flasks (batch mode) and in a photobioreactor operating in semi-continuous mode with incremental addition of effluents. Biomass concentration reached 0.68 g L^{-1}, with high removal efficiencies for sugars (78.3%) and phenolic compounds (98.0%).

These studies demonstrate the versatility and effectiveness of different microalgal species for WWW treatment under a wide range of operational conditions, including variations in scale, HRT, and wastewater conditioning (dilution and pretreatment). Overall, high removal efficiencies of organic matter, nutrients, and phenolic compounds, together with significant biomass production, highlight the potential of microalgal systems as integrated platforms for wastewater remediation and resource recovery. However, the strong variability in operational parameters across studies underscores the need for further standardization and optimization, particularly to support scale-up and full-scale implementation. Collectively, these findings reinforce the role of microalgae-based processes as promising solutions within circular bioeconomy frameworks, while emphasizing the importance of advancing research towards more robust, scalable, and economically viable systems.

Microalgae Role in WWW Treatment

As above mentioned, microalgae play a pivotal role in the treatment of WWW by facilitating nutrient removal, enhancing dissolved oxygen levels through photosynthetic activity, and generating valuable biomass. Their integration into sustainable or combined treatment systems contributes to improved water quality, reduced environmental impact, and the potential for resource recovery. Oxygenation is improved primarily through photosynthetic oxygen production. During daylight hours, microalgae perform photosynthesis, using sunlight to convert carbon dioxide (CO_2) and water (H_2O) into glucose and oxygen (O_2) (Eq. 1) [38]:

$$6\ \mathrm{CO_2} + 6\ \mathrm{H_2O}\ +\ \text{light} \rightarrow \mathrm{C_6H_{12}O_6} + 6\ \mathrm{O_2} \tag{1}$$

This oxygen is released directly into the surrounding water, significantly increasing dissolved oxygen (DO) levels in the media growth containing WWW.

However, during the night, in the absence of light, microalgae (like all aerobic organisms) shift to cellular respiration, a process where they consume oxygen to metabolize stored organic compounds (e.g., glucose), releasing CO_2 and reducing DO levels:

$$\mathrm{C_6H_{12}O_6}\ + 6\ \mathrm{O_2}\ \rightarrow 6\ \mathrm{CO_2} + 6\ \mathrm{H_2O}\ +\ \text{energy} \tag{2}$$

As a result, DO concentrations tend to drop during the dark period, which may lead to temporary hypoxic conditions, especially in dense cultures or poorly aerated systems. This dual (day–night) oxygen fluctuation is important to consider when designing and managing photobioreactors or wastewater treatment systems using microalgae, since high DO levels are crucial for an efficient WWW treatment [39]. Indeed, high DO levels support aerobic heterotrophic microorganisms which are the primary and fastest degraders of organic matter, thus contributing significantly to COD removal in WWW. These organisms break down organic compounds derived from grape residues and fermentation byproducts. Microalgae, being primarily photosynthetic, contribute to COD removal indirectly, mainly by releasing oxygen. In this microbial dynamic, the autotrophic microalgae provide the necessary oxygen for the aerobic respiration of the bacteria, yeast and heterotrophic microalgae (Fig. 2). This symbiotic relationship ensures a robust and efficient degradation of the organic load and prevents anaerobic conditions, which can lead to unpleasant odors, slower degradation, and the production of harmful compounds like hydrogen sulfide (H_2S) as above mentioned [40].

Microalgae play a key role in the bioremediation of WWW by efficiently assimilating inorganic nutrients such as nitrate (NO_3^-), ammonium (NH_4^+), and phosphate (PO_4^{3-})

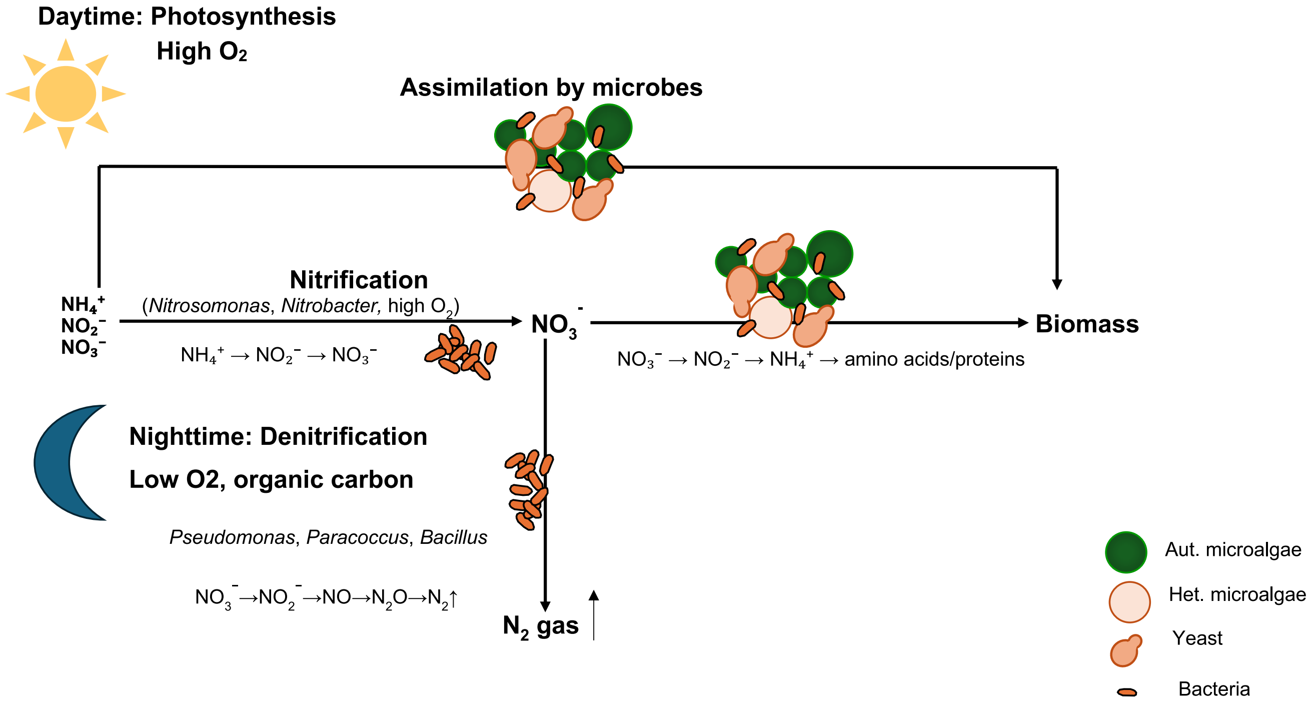

Fig. 3 Nitrogen removal mechanisms in WWW

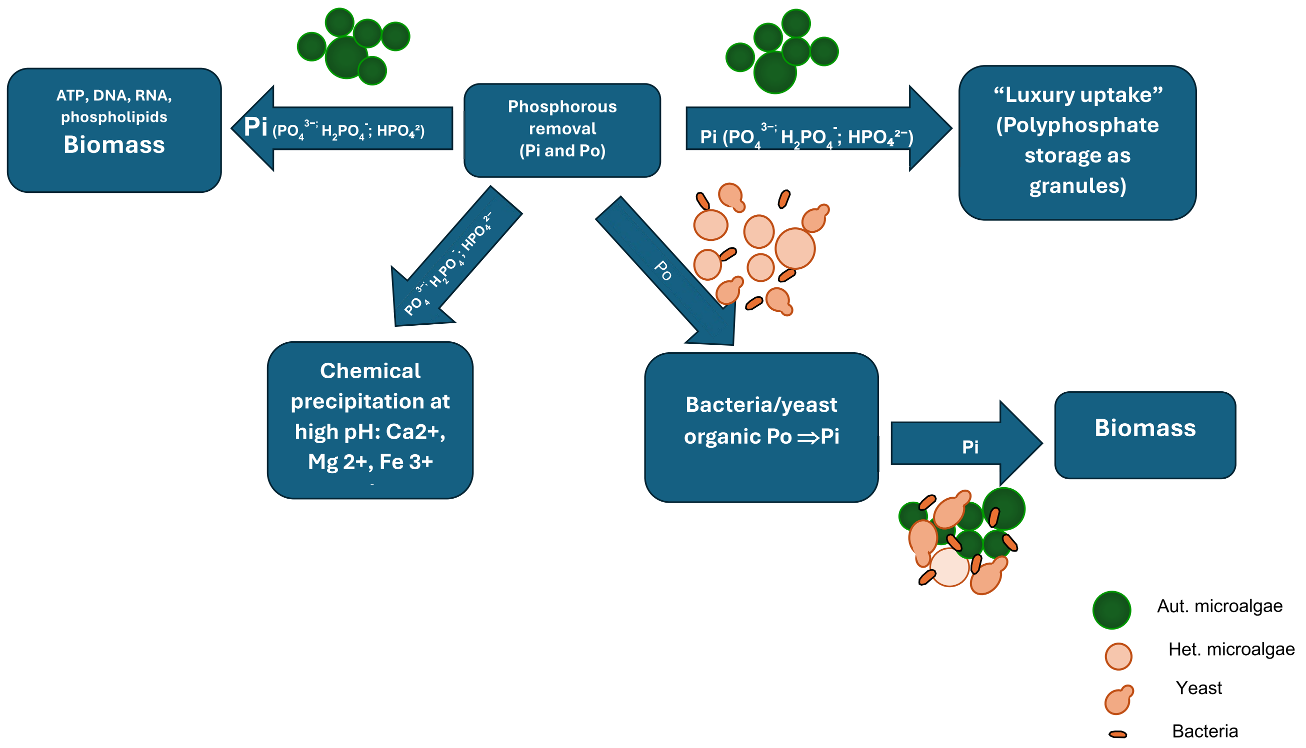

Fig. 4 Phosphorus removal mechanisms in WWW. Pi—inorganic P; Po—Organic P

(Fig. 3 and 4). These nutrients are typically present at high concentrations in WWW due to the decomposition of grape residues, the use of cleaning agents, and fermentation byproducts. Both autotrophic microalgae and heterotrophic microorganisms can assimilate these nitrogen forms (NH_4^+, NO_3^-, and NO_2^-) to support growth and biomass formation (Fig. 3). During daytime, photosynthetic activity by microalgae releases O_2, increasing the dissolved oxygen

(DO) levels in the medium. Elevated DO promotes aerobic microbial processes such as nitrification, primarily carried out by *Nitrosomonas* ($NH_4^+ \rightarrow NO_2^-$) and *Nitrobacter* ($NO_2^- \rightarrow NO_3^-$). The nitrate produced can either be assimilated by autotrophic or heterotrophic microbes for aminoacids, proteins and biomass synthesis or, under oxygen-limited conditions—typically during nighttime—undergo bacterial denitrification, predominantly mediated by heterotrophic bacteria such as *Pseudomonas*, *Paracoccus*, and *Bacillus* species, resulting in the reduction of nitrate to gaseous nitrogen species (N_2, N_2O) and achieving permanent nitrogen removal [41, 42].

Concerning the phosphorous removal, microalgae require inorganic phosphorus (as PO_4^{3-}, $H_2PO_4^-$, HPO_4^{2-}) for growth, being incorporated into ATP, nucleic acids, and phospholipids (Fig. 4). During exponential growth, microalgae assimilate inorganic phosphate directly from the medium. During phosphorus-rich conditions in WWW, microalgae can perform "luxury uptake", a mechanism in which phosphate is absorbed in excess of immediate metabolic needs and stored intracellularly as polyphosphate granules. This mechanism allows cells to continue growing later when external phosphate concentrations are low. In algal–bacterial consortia such as those present in WWW, organic phosphorus compounds (e.g., phosphoesters—major constituents of grape pulp, skins, seeds, and particularly of yeast cells that become non-viable after fermentation) are degraded by heterotrophic microbes, which mineralize these compounds into inorganic phosphate (Pi). The released Pi can then be assimilated by autotrophic microalgae and heterotrophic microbes to support cellular growth and biomass production [43]. An indirect way to remove phosphorous is the co-precipitation with metals. In wastewater systems, microalgal photosynthesis increases the pH (due to CO_2 uptake). Higher pH can cause chemical precipitation of phosphates with calcium, magnesium, or iron present in WWW, leading to abiotic phosphorus removal [44]. Therefore, in WWW, phosphorus removal occurs mainly by biological assimilation into algal biomass and luxury polyphosphate storage, with some contribution from precipitation at high pH.

Through photosynthetic and metabolic processes, microalgal cells absorb and utilize nutrients for cellular function and biomass production. N and phosphorus P are essential macronutrients involved in the synthesis of key biomolecules such as proteins, nucleic acids, enzymes, phospholipids, chlorophyll, and adenosine triphosphate. These compounds are crucial for microalgae cells' growth, energy transfer, and cellular replication [45]. The harvested algal biomass is then phosphorus-rich and nitrogen-rich, being often proposed as a biofertilizer or soil conditioner. It is clear that microbial communities present in WWW are a central key to an effective and symbiotic system for its treatment. Autotrophic microalgae facilitate the removal of COD by creating the necessary aerobic environment for heterotrophic microorganisms to efficiently break down organic pollutants. They are also crucial for other nutrient removal, directly assimilating nitrogen and phosphorus for their growth and, through their photosynthetic activity, driving the environmental conditions (e.g., pH shifts and oxygen production) that support bacterial nitrification and anoxic denitrification. By orchestrating these processes, autotrophic-heterotrophic consortia may transform WWW from a pollutant-rich effluent into a valuable resource, as the resulting nutrient-rich biomass can be repurposed, thus closing the nutrient loop and establishing a sustainable, circular economy model for the wine industry [46].

Monitoring Microalgae WWW Cultivations

The successful implementation of microalgae-based systems for WWW treatment hinges on the biological performance of the selected microalgal strains. WWW contains a complex mixture of organic compounds—such as acids, ethanol, and phenolics—that can inhibit cell growth and affect physiology [47, 48]. These inhibitory substances may impair cell viability, reduce nutrient uptake efficiency and, ultimately, compromise treatment effectiveness. Therefore, continuous and accurate monitoring of algal cultures is crucial to detect stress or inhibition at an early stage, and to optimize overall system performance.

Conventional monitoring methods typically involve periodic sampling followed by optical density (OD) measurements, dry weight determinations, chlorophyll content analysis, or nutrient assays. While these approaches provide valuable information on biomass growth and nutrient removal, they are often time-consuming, labor-intensive, and lack the resolution to differentiate live, dead, or stressed cells. Moreover, they do not capture rapid dynamic changes in microbial communities or population heterogeneity within microbial cultures—critical factors in WWW treatment due to the variability in wastewater composition associated with seasonality and winemaking operations.

In microalgae-based WWW treatment systems, cultures rarely consist of pure microalgal populations. Instead, microalgae coexist with bacteria and other microorganisms naturally present in wastewater. While these mixed consortia can enhance treatment performance through synergistic interactions [23], they also introduce significant challenges for monitoring, particularly regarding the accurate determination of microalgal biomass concentration. Conventional analytical methods such as optical density (OD), dry weight, or chlorophyll measurements may be affected by the presence of non-algal particles and heterotrophic microorganisms, leading to overestimation or misinterpretation of biomass. Therefore, distinguishing between algal and non-algal biomass is critical for reliable process monitoring and performance assessment.

Several online and at-line monitoring techniques can be employed to provide detailed, real-time information on microalgal cultivation in WWW systems such as:

(A) Spectroscopic Techniques

Chlorophyll fluorescence (Pulse-Amplitude Modulated Fluorometry) allows non-invasive assessment of photosystem II efficiency (Fv/Fm), which serves as a sensitive indicator of photosynthetic performance and stress [49]. This method is particularly useful in identifying physiological responses to WWW toxicity or nutrient limitation. Fourier Transform Infrared Spectroscopy (FTIR) provides information on the macromolecular composition (lipids, proteins, carbohydrates) of microalgal cells by detecting specific molecular vibrations [50, 51]. This technique enables tracking of biochemical shifts linked to environmental stress. Raman Spectroscopy allows the nutrient concentration quantification in culture media [42, 43] and its coherent extensions enable detection, spatial mapping, and relative quantification of carotenoids and lipids in microalgae [52, 53].

(A) Imaging Techniques

Confocal Laser Scanning Microscopy (CLSM) provides high-resolution, 3D visualization of microalgae, enabling detailed studies of their growth, structure, and health in situ, even within biofilms. By using a focused laser beam and an optical pinhole to eliminate out-of-focus light, CLSM produces sharp, clear images that allow for the differentiation of individual cells and extracellular matrix components. This non-invasive technique is crucial for monitoring microalgae in research, particularly for understanding their response to environmental stress factors, assessing ecotoxicity, and evaluating applications in wastewater treatment and biofuel production [54].

(I) Omics-Based Approaches

Transcriptomic tools (e.g., RNA-seq, qPCR) enable the detection of differential gene expression in response to specific stressors found in WWW, such as high organic load or phenolic compounds. Proteomics and metabolomics offer complementary insights into protein expression and metabolite accumulation under different environmental conditions. These tools are useful for identifying pathways involved in nutrient assimilation or stress responses [55].

(I) Biosensors and Microelectrodes

DO and CO_2 Sensors are essential tools for tracking metabolic activity, providing real-time indicators of respiratory and photosynthetic processes [56]. Ion-Selective Electrodes (ISEs) facilitate in situ monitoring of key nutrients (nitrate, phosphate, ammonium), which are critical for understanding nutrient uptake and potential limitations during cultivation [57]. Microfluidic systems and lab-on-a-chip devices integrate multiple monitoring modalities into compact platforms, allowing rapid screening of cell responses to wastewater conditions at microscale [58].

(A) Flow cytometry

Flow cytometry (FC) is a powerful tool for real-time, high throughput monitoring of microbial cultures, offering detailed insights into cell size, morphology, pigment content, viability, and physiological status through the use of specific fluorescent dyes such as propidium iodide, Sytox Green, carboxyfluoresceine diacete (CFDA), and $DiOC_6(3)$ which can be used to assess can assess membrane integrity, enzymatic activity, and membrane potential, respectively [59]. This technique allows for precise cell count and enables discrimination between microalgae and coexisting microorganisms, as well as between healthy, stressed, and dead cells, which is particularly important when cultivating microalgae in WWW, a complex and potentially inhibitory matrix. Indeed, an important advantage of FC is its ability to detect natural chlorophyll autofluorescence, enabling accurate identification of microalgae in heterogeneous environments where traditional methods such as OD or dry weight can be confounded by suspended solids and microbial contaminants [60].

Beyond cell viability and growth assessment, FC enables rapid quantification of intracellular compounds like lipids and carotenoids, which are relevant for biomass valorization. Unlike conventional extraction-based methods, FC provides fast, solvent-free analysis and can detect subpopulations, offering timely insights for process optimization and early corrective actions [61].

In the dynamic context of WWW treatment, FC serves as a powerful tool for robust process control. It allows for the rapid characterization of the microbial community, providing valuable information on the physiological state of different populations, including microalgae, bacteria, and yeast. This enables the early detection of stress responses and imbalances, ensuring that operators can optimize cultivation strategies in near real-time (at line) [59]. By providing deep insights into the complex symbiotic dynamics within the system, FC contributes directly to the development of more efficient and scalable microalgae-based wastewater treatment systems.

(A) Artificial Intelligence, Machine Learning, and Automated Monitoring Systems

Artificial intelligence (AI) and machine learning (ML) are increasingly being integrated into microalgae-based wastewater treatment systems to enhance process efficiency, robustness, and monitoring accuracy. As highlighted in a comprehensive review by Ning et al. [62], ML applications in algal biotechnology extend to strain identification, quantification, stress detection, and the optimization of cultivation conditions. These tools rely on algorithms capable of processing large and complex datasets, thereby providing predictive insights and automated decision-making support that go beyond conventional monitoring methods. Practical applications include the identification and quantification of algal strains, real-time diagnosis of cellular stress for improved process control, and the prediction and regulation of key cultivation parameters such as harvesting time, nutrient dynamics, and biomass composition. By enabling early detection of stress signals and offering predictive control over growth conditions, AI- and ML-driven monitoring systems can maximize treatment efficiency, biomass valorization, and overall sustainability. In the context of WWW treatment, where influent characteristics can fluctuate considerably in terms of organic load, phenolic compounds, and nutrient concentrations, the integration of AI and ML may provide a powerful framework to manage variability and optimize algal-based treatment performance. As emphasized by Imamoglu et al. [63] the adoption of AI- and ML-enhanced monitoring is expected to be pivotal for the development of smart, automated, and scalable wastewater treatment technologies in near future.

Microalgae Cultivations on WWW

As above stated, WWW is typically rich in organic carbon due to the presence of sugars, alcohols, and volatile fatty acids derived from grape residues and fermentation processes. However, the concentrations of essential nutrients such as nitrogen (N) and phosphorus (P) can vary significantly depending on winemaking practices, grape variety, and the stage of processing (e.g., fermentation, cleaning, or bottling). This imbalance can hinder microalgal growth and reduce treatment efficiency, as algae require balanced nutrient availability to support cell division and metabolic activity. Under nitrogen- or phosphorus-limited conditions, microalgae may enter a stress response, leading to reduced biomass productivity or undesirable shifts in community composition. To address these limitations, supplementation with inorganic nutrients (e.g., ammonium or phosphate salts) or dilution with nutrient-rich wastewater or freshwater can be employed to restore optimal stoichiometric ratios (e.g., Redfield ratio of C:N:P = 106:16:1) and prevent the accumulation of inhibitory compounds such as organic acids [32, 34, 35, 59]. Tailoring the nutrient profile of WWW to match microalgal requirements is therefore crucial for stable cultivation, efficient pollutant removal, and high-value biomass production.

Microalgae can be cultivated in either open or closed systems, each with distinct advantages and limitations. Open raceway ponds are widely used due to their low capital and operational costs, ease of construction, and suitability for large-scale operation. However, they are more vulnerable to contamination, environmental fluctuations (e.g., temperature, evaporation), and have lower productivity due to limited control over cultivation parameters [64]. Photobioreactors (PBRs) provide a controlled environment, allowing precise regulation of light, CO_2 supply, temperature, and mixing. They are more efficient in terms of biomass productivity and microbial pathogen control but are significantly more expensive and energy-intensive [65]. Therefore, if the primary goal is high-value biomass production (lipids, pigments, feedstock) and controlled, high-efficiency nutrient removal, PBRs are more efficient but costly. If the goal is cost-effective WWW treatment with acceptable nutrient removal and biomass generation, open ponds are generally more practical, especially in regions with favorable climate. To balance cost and performance, hybrid systems that combine features of both open and closed cultivation have been proposed. For example, a PBR may be used for inoculum preparation followed by scale-up in raceway ponds. These systems can enhance process stability while reducing overall costs and have shown promise in wastewater treatment applications, including agro-industrial effluents such as WWW [66].

Typical mass outdoor microalgae cultivation is autotrophic, although heterotrophic growth is also possible depending on system design and light availability. However, mixotrophic growth is often the preferred mode for microalgae cultivated on WWW, because this wastewater contains both inorganic nutrients (like orthophosphate) and a high concentration of organic compounds (such as ethanol, organic acids, and phenolics) that can be used as a carbon and phosphorous sources. Mixotrophy allows cells to use both photosynthesis and organic substrate metabolism, improving growth rates and biomass yields. Several studies report enhanced productivity under mixotrophic conditions when using agro-industrial effluents [67, 68].

In the absence of light (e.g., in closed or shaded systems), microalgae can grow heterotrophically using only organic compounds as energy source. WWW often contains biodegradable organics like ethanol and volatile fatty acids, which support heterotrophic growth. However, heterotrophic cultivation requires strict control to avoid bacterial contamination and is less commonly used at full scale for wastewater treatment [64, 68]. While less efficient on WWW alone (due to limited inorganic carbon and high organic load), autotrophic growth is possible in diluted or pretreated WWW, or in systems with CO_2 supplementation, being commonly used in open raceway ponds exposed to sunlight [59].

Strategies and Approaches for Optimizing Microalgae Cultivation on WWW

Effective application of microalgae in WWW treatment relies on careful selection of cultivation systems, process optimization, and potential integration with other treatment technologies. Given the complex composition of WWW, which often contains high organic loads, variable nutrient ratios, and potentially inhibitory compounds, the success of microalgae-based approaches depends on tailoring the cultivation strategy to the specific characteristics of wastewater and operational goals. Successful microalgal cultivation on WWW also requires optimization of key environmental and operational parameters. Light intensity and photoperiod must be tuned to support photosynthesis while avoiding photoinhibition. Shading due to suspended solids in WWW can limit light penetration, particularly in dense cultures or untreated effluent. Several approaches can be considered to reduce localized shading and increase average light exposure across the culture [69]. Mixing is essential for keeping microalgal cells suspended uniformly in the culture medium. Without proper agitation, cells would settle at the bottom, leading to light limitation for cells in the lower layers and photoinhibition (damage from excessive light) for cells at the surface [70]. By circulating the cells, mixing ensures that all cells are exposed to light intermittently, a process known as the "light–dark cycle". This dynamic exposure optimizes light utilization efficiency and promotes higher overall biomass productivity. Beyond gas exchange, mixing helps maintain the chemical and thermal homogeneity of the culture. It prevents the formation of stagnant zones and nutrient gradients that could limit growth. Crucially, in a wastewater treatment context, constant mixing ensures that the oxygen produced by the microalgae is distributed throughout the system. This prevents the formation of anaerobic pockets, which could lead to a drop in treatment efficiency and the production of foul-smelling gases like hydrogen sulfide (H_2S), a common issue in wastewater with high organic loads [71]. Therefore, mixing intensity must be carefully optimized. While more vigorous mixing enhances all these processes, it also requires more energy, leading to higher operational costs. The ideal mixing strategy balances the biological needs of the microbial consortium with the economic feasibility of the system [72]. Sedimentation, flotation or flocculation can be applied prior to algal cultivation to reduce turbidity. Filtration (e.g., mesh or sand filters) or centrifugation can also help remove fine particles. This step enhances light transmission and creates a more favorable environment for algal growth. Other options include diluting WWW, as above mentioned, to reduce the concentration of suspended particles and organic load, contributing to maintain a cell density which improves light penetration, especially in open ponds. The use of shallow raceway ponds or thin-layer cascade systems improve surface area-to-volume ratios, maximizing light availability, even with some suspended particles present [73]. In closed systems, artificial lighting (e.g., LED panels or fiber optics) can ensure uniform light delivery regardless of external turbidity. Internal light guides can deliver light deeper into dense cultures [74]. Frequent mixing or circulation ensures light–dark cycles by rotating algal cells between illuminated and shaded zones [75].

Temperature plays a crucial role in microalgal physiology, affecting photosynthesis, nutrient uptake, and biomass productivity. Although most microalgal species grow optimally within a temperature range of 20 ºC –30 °C, such conditions are not always maintained in outdoor cultivation systems, which are commonly used for WWW treatment due to their lower operational costs. In these systems, temperature fluctuations caused by daily and seasonal changes can impact algal performance, particularly in temperate climates [76]. While temperature regulation in closed systems (e.g., photobioreactors) can improve performance and stability, it also incurs higher energy costs, which may contradict the sustainability goals of WWW treatment [77]. To address this, most studies on WWW valorization by microalgae have adopted ambient temperature cultivation, favoring cost-efficiency over maximum productivity. Maintaining an optimal pH range during microalgal cultivation on WWW is also critical for maximizing nutrient removal efficiency, sustaining algal growth, and ensuring the stability of co-cultured microbial communities. Although initial pH correction is often necessary—particularly since WWW tends to be acidic—microalgal metabolism can significantly alter the medium pH over time, potentially impairing nutrient uptake, photosynthesis, and biomass productivity [78]. Stable pH conditions (typically between 6.5 and 8.5) enhance nitrogen and phosphorus assimilation, support bacterial activity (e.g., vitamin synthesis), and can influence the accumulation of valuable intracellular products such as lipids and pigments. While continuous pH control may not always be economically feasible in large-scale open pond systems for microalgae cultivation, partial regulation through CO_2 addition, buffering agents, or the use of pH-tolerant strains is often justified to maintain treatment efficiency and biomass quality [79–81].

Strategies for recycling the culture medium in microalgae cultivation on wastewater are also crucial for increasing efficiency and reducing operational costs of WWW treatment. This involves reusing the aqueous phase after the microalgae have been harvested, a process that is a core part of the circular economy model. After the microalgal biomass is harvested from the wastewater, the remaining liquid—now called the effluent—is a treated medium with a lower concentration of pollutants. This effluent can be reused through partial or full recycling, whereby it is either partially or fully mixed with incoming WWW to dilute it and initiate a new treatment cycle. This reduces the need for freshwater dilution and helps maintain a stable microbial environment by inoculating the new batch with existing microbes [82–84]. Recycling can also be used as nutrient supplementation. Indeed, although the treated effluent has lower pollutant levels, it may still contain some residual nutrients. These nutrients can be supplemented with additional nitrogen and phosphorus to bring them to optimal levels for a new cycle of microalgae growth. This approach minimizes the amount of fresh nutrients that need to be added to the system [85].

Microalgal Biomass Harvesting and Bioproduct Recovery After WWW Treatment

Microalgal biomass harvesting after WWW treatment is a critical step that significantly influences the overall process efficiency and economic viability. Common harvesting techniques include centrifugation, flocculation (chemical or bio-flocculation), filtration, sedimentation, and flotation [86]. Centrifugation provides rapid and efficient biomass recovery with high purity but is energy-intensive and costly, often limiting its use to downstream high-value applications. Flocculation, using chemical agents like alum or natural bio-flocculants such as chitosan, offers a cost-effective alternative by aggregating microalgal cells into larger particles that settle more easily; however, residual chemicals may contaminate the biomass, restricting its use in sensitive applications [87]. Filtration and sedimentation are low-energy methods but can be hindered by the small size and low density of microalgal cells, as well as the presence of suspended solids typical in WWW, which may clog filters or slow settling rates [88]. Flotation techniques, including dissolved air flotation, leverage air bubbles to separate biomass but require specialized equipment [64]. A key challenge in harvesting microalgae from WWW is the heterogeneous nature of the biomass and the presence of organic and inorganic contaminants, which complicate separation and increase operational costs [34]. Nevertheless, optimizing harvesting strategies—often by combining methods or using innovative approaches such as magnetic harvesting or membrane filtration—can enhance recovery efficiency, reduce costs, and improve biomass quality, thereby supporting downstream valorization in bioproduct and biofuel production [89]. After harvesting, the algal biomass produced during WWW treatment is often rich in proteins, making it a suitable candidate for animal feed applications. Species such as *Scenedesmus* and *Chlorella* grown in nutrient-rich wastewaters have demonstrated protein contents above 40% of dry weight [37, 90].

Table 4 Bioproducts from microalgal biomass obtained from WWW microalgae treatment

Bioproduct	Main microalgal species	Application	References
Protein and rich biomass	*Chlorella vulgaris, Arthrospira platensis*	Animal feed, aquafeed	[35]
Lipids	*Chlorella vulgaris, Arthrospira platensis*	Biofuels	[34]
Pigments (lutein, β-carotene, tocopherols)	*Chlorella sorokiniana*	Nutraceuticals, cosmetics, animal feed	[10]
Biogas/Biofertilizer from biomass	Mixed microalgae biomass	Anaerobic digestion, soil amendment	[94]
Biofuels, chemicals	*Chlorella vulgaris, Arthrospira platensis*	Pyrolysis	[95]

Certain microalgae produce commercially valuable pigments such as β-carotene, lutein, and phycocyanin which may be produced by cells when growing on WWW. These compounds have broad applications as natural colorants in the food industry, antioxidant ingredients in cosmetics, and nutraceutical agents due to their health-promoting properties [10]. In addition, the residual biomass remaining after lipid extraction and carotenoid extraction (defatted biomass) retains high levels of nutrients and can be used as a biofertilizer, contributing to soil amendment and improved crop productivity, including vineyards [37, 91]. Importantly, safety assessments must be conducted to ensure the absence of heavy metals, pathogens, or residual phytotoxins, especially when the biomass is intended for use in food chains or soil applications [92].

Table 4 summarizes some of the key bioproducts that can be derived from microalgae cultivated on WWW and their potential applications.

These bioproducts not only enhance the economic feasibility of microalgae-based wastewater treatment but also align with circular economy principles by converting waste into valuable commodities. In addition to its nutritional and pigment applications, microalgal biomass derived from WWW treatment also represents a valuable feedstock for bioenergy production. Depending on the biochemical composition of the biomass, different energy routes can be pursued, including biodiesel from lipids, biogas via anaerobic digestion of residual carbohydrates and proteins, or bioethanol from fermentable sugars. Coupling wastewater remediation with bioenergy generation enhances process sustainability and creates an integrated biorefinery concept. Certain microalgal strains cultivated on WWW accumulated lipids which were converted into biodiesel, via transesterification [24]. Since microalgal biomass can contain significant amounts of carbohydrates—Lopes da Silva et al. [59] reported approximately 50% (w/w) in *Chlorella vulgaris* grown on WWW—these carbohydrates can be further hydrolyzed into fermentable sugars, which can subsequently be converted into bioethanol by microorganisms such as yeast [93], although this approach has never been applied to microalgal biomass obtained after WWW treatment.

Avila et al. [94] presented a pilot-scale case study integrating a WWW microalgae-based treatment with anaerobic digestion of secondary sludge and co-digestion with algal biomass to enhance resource and energy recovery. The digestates (from mono- and co-digestion) and the dry algal biomass were tested in plant growth bioassays, where all treatments enhanced plant biomass relative to a commercial amendment. Thermochemical conversion technologies, such as hydrothermal liquefaction (HTL) and hydrodeoxygenation (HDO) allow efficient processing of wet microalgal residues obtained from WWW treatment, into bio-oils and upgraded biofuels, without the need for energy-intensive drying steps [95].

Integrating these bioenergy valorization routes with the production of high-value products from microalgal biomass obtained from WWW treatment will enhance both the economic feasibility and environmental sustainability of microalgae-based systems, by closing resource loops and generating renewable energy alongside effective wastewater remediation. Nevertheless, despite the promising potential of microalgae for WWW remediation, downstream valorization of the resulting biomass remains largely limited (Table 4).

Only a few studies have demonstrated the recovery of high-value compounds—such as proteins and pigments—from microalgal cultures grown on WWW, whereas applications like lipid-based biodiesel production or biopolymer synthesis are still not documented. Further research is needed to optimize bioproduct recovery and to develop integrated biorefinery approaches that maximize both WWW treatment efficiency and biomass utilization. Integrated biorefinery approaches are essential to provide a sustainable and circular strategy for WWW treatment, by maximizing resource recovery from microalgal biomass. Additionally, CO_2 capture and recycling systems—such as those recovering CO_2 from ethanol fermentation of winery by-products (e.g., grape pomace or must) or from anaerobic digestion or HTL/HDO processing of algal residues, can enhance carbon utilization by supplying a continuous, low-cost source of inorganic carbon to sustain algal growth. This integration increases biomass productivity, while reducing the overall carbon footprint of the biorefinery, enhances the overall energy and nutrient balance of the system and contributes to greenhouse gas mitigation.

Techno-Economic Analysis (TEA), Life Cycle Assessment (LCA), and Social Life Cycle Assessment (S-LCA) in Microalgae-Based WWW Treatment

To support scale-up and sustainable implementation, comprehensive techno-economic analyses (TEA) and life-cycle assessments (LCA) are essential for identifying key cost drivers and environmental trade-offs [2]. Evaluating the sustainability and scalability of microalgae-based WWW treatment systems requires robust, integrative assessment tools that holistically assess economic viability, environmental impacts, and social dimensions. The combined use of Techno-Economic Analysis (TEA) and Life Cycle Assessment (LCA) facilitates data-driven decision making, guiding the development of scalable, resource-efficient wastewater treatment and biorefinery systems. TEA provides insight into capital (CAPEX) and operational expenditure (OPEX), profitability, and market competitiveness, while LCA evaluates environmental impacts throughout the entire value chain, from resource extraction to disposal.

Recent benchmarking studies reveal that microalgae-based systems tend to incur higher CAPEX and OPEX than conventional agricultural or chemical processes but offer crucial benefits such as reduced land and water requirements, and the potential for long-term environmental sustainability. Their economic viability strongly depends on factors like process scale, product diversification (single vs. multiproduct biorefineries), and integration with waste valorization strategies [96]. For instance, Ioannidou et al. [2] assessed a WWW and pomace-integrated biorefinery producing succinic acid alongside high-value co-products. They found cultivation infrastructure to be the main capital cost driver, while energy-intensive biomass harvesting and solvent extraction dominate operational costs. Profitability increased when multi-product streams like proteins and bio-based materials were incorporated. Corresponding LCA identified biomass drying and extraction as environmental hotspots, but renewable energy integration and anaerobic digestion coupling slashed impacts by over 70%, underscoring system-level optimizations [96].

Similarly, El Bakraoui et al. [97] corroborated the high efficiency of microalgae biorefineries in pollutant removal and biomolecule production but highlighted the high energy demands in upstream and downstream processes as barriers to industrial deployment. The lack of standardized, conventional process designs complicates consistent TEA and LCA comparisons, emphasizing a need for robust, harmonized data to support scale-up.

Recent LCA case studies, such as Crippa et al. [98], demonstrate that environmentally friendly valorization options like biostimulant production and agricultural application reduce environmental impacts across most categories, replacing chemical fertilizers and lowering pesticide and irrigation needs. However, unresolved challenges like ammonia emissions from open algal ponds threaten long-term sustainability and require mitigation.

Kumar et al. [99] introduced an advanced TEA–LCA framework advocating for multiproduct valorization—including pigments, proteins, and biofertilizers—because single-product operations rarely sustain profitability or environmental benefits. Complementing this, Geng et al. [100] provide a comprehensive systematic review of microalgal biorefineries, with particular emphasis on TEA and LCA-based trade-offs. They highlight that using waste feedstocks (including wastewater) and integrating co-product valorization (e.g. proteins, pigments, biofertilisers, biostimulants, biopesticides) are among the few routes by which physical and economic thresholds (CAPEX, OPEX, energy input) can approach commercially viable levels. However, they also identify major bottlenecks: inefficient downstream processing (harvesting, dewatering, drying), and inconsistencies in LCA boundary choices that lead to ± 30–40% variation in reported environmental outcomes. For WWW treatment, these findings suggest that coupling cultivation with WWW as nutrient source, selecting robust strains adapted to local climates, and designing low-energy harvesting and drying systems could significantly improve both economic feasibility and environmental performance.

Praveen et al. [31] provided one of the first winery-specific TEA-integrated studies, demonstrating that strain selection and system design mitigate cultivation inefficiencies to yield positive economic indicators. Nevertheless, the omitted analysis of biomass downstream valorization reveals a persistent gap, as financial feasibility at the treatment stage does not guarantee overall sustainability.

A critical shortcoming in microalgae-based WWW sustainability assessments is the insufficient consideration of social impacts. According to the United Nations Environment Programme (UNEP), Social Life Cycle Assessment (S-LCA) evaluates project effects on stakeholders including workers, local communities, consumers, and other actors across the value chain. across the value chain. Recent reviews and case studies highlight indicate that most S-LCA studies focus on easily quantifiable metrics (e.g., job creation, wage ratios), with less attention to qualitative aspects (e.g., worker satisfaction, equity, discrimination). Moreover, active stakeholder engagement (e.g., surveys, interviews) is essential for identifying social hotspots and ensuring project legitimacy. There is an urgent call for harmonized indicators and standardized S-LCA methodologies to enable meaningful integration with TEA and LCA in contexts such as WWW treatment [101, 102].

Integrating TEA, LCA and S-LCA—known as Life Cycle Sustainability Assessment (LCSA)—is increasingly accepted as the gold standard for biorefinery evaluation, enabling a balanced assessment of economic, environmental, and social pillars that supports responsible innovation and policy formulation.

In conclusion, TEA and LCA evidence supports that microalgae-based WWW treatment can be both viable and sustainable when designed carefully for co-product valorization, energy efficiency, and integration with larger systems. Absence of these factors risks economic marginality and suboptimal environmental performance.

Policy and Regulatory Framework for Treated WWW

The adoption and upscaling of microalgae-based systems for WWW treatment and biomass valorization are strongly influenced by policy and regulatory frameworks defining effluent discharge standards, treated water reuse quality, and biomass classification (product *vs.* waste). Regulatory landscapes vary globally but converge around principles such as those preserved in the United Nations Sustainable Development Goals (SDGs), particularly SDG 6 (Clean Water), SDG 12 (Responsible Consumption and Production) and SDG 13 (Climate Action). These initiatives promote sustainable water management, pollution control and circular economy practices.

International guidelines like those of the Food and Agriculture Organization (FAO) and the World Health Organization (WHO) encourage safe wastewater reuse in agriculture, supporting alternative water sourcing and growing scarcity. The Organisation for Economic Co-operation and Development (OECD) advocates circular economy principles, emphasizing nutrient recovery and bio-based innovation. Common regulatory parameters include effluent quality limits for chemical oxygen demand (COD), biological oxygen demand (BOD), total nitrogen, and phosphorus in effluent discharge. Increasingly, governments tend to support water reuse, bioproduct recovery, and integration of bioeconomy strategies. However, challenges remain regarding standardization and risk assessment, especially for emerging technologies like microalgal systems.

The EU exemplifies a comprehensive regulatory environment with frameworks such as the EU Green Deal [103], the Circular Economy Action Plan [104], and the Farm to Fork Strategy [105], all aiming to enhance sustainability and resource efficiency in wastewater treatment and bioproduct recovery. Environmental discharge limitations and backing nutrient recycling and low-carbon technologies are regulatory drivers fostering microalgal solutions. However, challenges persist, such as the unclear regulatory status of algal biomass (product *vs*. waste), lack of harmonized quality and safety standards for algal biofertilizers or feed additives, and regulatory gaps concerning treated industrial wastewater reuse beyond agriculture.

Specifically, Garciarena et al. [92] note that microalgae-derived food/feed products must comply with European Food Safety Authority (EFSA) safety assessments. Strains with “Qualified Presumption of Safety” (QPS) benefit from streamlined evaluations, while non-QPS strains require full assessment, including strain identification, compositional, toxicological and allergenicity testing. Such complex regulatory requisites may delay market entry and impede adoption, especially for small-scale producers or regions lacking regulatory harmonization.

Stakeholders and Their Roles in Microalgae-Based WWW Treatment

The successful implementation of large-scale microalgae-based systems for WWW treatment requires the coordinated participation of diverse stakeholders, including winery operators, microalgae and biotech companies, technology and engineering firms, policymakers and regulators, research institutions and academia, investors and funding agencies, consumers and end-users, and water utilities and waste management companies. These actors collectively influence the technical, economic, regulatory, and social feasibility of the systems, playing critical roles throughout the entire value chain—from wastewater generation and treatment to biomass valorization and final product commercialization.

Wineries represent both the primary sources of WWW and the main beneficiaries of its treatment. Their motivations typically include achieving environmental compliance, reducing treatment costs, and valorizing the resulting algal biomass [47]. However, as reported by several studies, adoption barriers persist, including limited technical expertise, spatial constraints, and the need for upfront capital investment [10].

Microalgae and biotechnology companies are fundamental in strain selection, system design, and process optimization, serving as bridges between laboratory research and industrial deployment. They also drive innovation in photobioreactor development, nutrient recycling, and biomass harvesting, which can strengthen process scalability and economic feasibility [64]. Technology providers and engineering firms, in turn, play a crucial role in ensuring modularity, automation, and energy efficiency of the treatment units—key factors for reducing operational costs and minimizing environmental impacts.

Policy makers and regulatory authorities at local, national and European levels define the legal framework that governs wastewater discharge limits, water reuse standards, and biomass valorization pathways [104, 106, 107]. Clear and stable regulation is essential to foster confidence among investors and operators. Researchers and academic institutions contribute by addressing knowledge gaps related to microalgae-WWW interactions, optimizing bioreactor performance, and assessing the environmental, economic, and social sustainability of these systems [10, 108]. Importantly, recent findings by Ortigueira and Lopes [101] highlight that stakeholder participation should be integrated into early-stage project design and assessment, for instance through social acceptance surveys or participatory S-LCA frameworks, to identify potential social hotspots before they escalate into negative impacts.

Investors and funding agencies are key enablers of scale-up, particularly since most microalgae-based WWW systems remain at pilot or pre-commercial stages. Their involvements depend on credible techno-economic data, return-on-investment prospects, and policy stability. European funding mechanisms such as Horizon Europe and the LIFE Programme have been instrumental in advancing early-stage projects by de-risking innovation and promoting circular economy solutions.

Consumers and end-users of products derived from microalgal biomass—such as biofertilizers, biostimulants, biopesticides, animal feed additives, cosmetics, and natural pigments—also play a decisive role in the market success of these systems. As emphasized by Ortigueira & Lopes (2025)[101], social acceptance and perception are not peripheral issues but central to sustainability transitions [101]. Consumer preferences for environmentally friendly and ethically sourced products can create a strong market pull, enhancing the profitability of biomass valorization pathways. Conversely, skepticism regarding products originating from wastewater-grown microalgae may hinder market uptake, underscoring the importance of transparency, quality control, and effective communication of environmental and social benefits [109–111].

Despite increasing research interest, the deployment of full-scale microalgae-based WWW treatment systems remains limited. Most projects are constrained to laboratory or small pilot scales [10, 24, 31, 33], often located in wine-producing regions such as Europe and South America. This limited adoption can be attributed to several persistent challenges: the variability in wastewater composition across seasons, lack of standardized operational and assessment protocols, high initial investment requirements, and regulatory uncertainty.

However, as highlighted by Ortigueira and Lopes (2025) [101] the path forward lies in fostering interdisciplinary collaboration and standardizing social and sustainability metrics. The incorporation of social dimensions—through S-LCA methodologies and stakeholder engagement—ensures that biorefinery-type systems, such as microalgae-based wastewater treatment, evolve in socially responsible and context-sensitive ways. The integration of technical, environmental, and social criteria in decision-making processes can support the transition from pilot to commercial scale, enhancing both societal acceptance and overall sustainability.

In summary, as policy support for circular economy strategies increases and technological advances in algal biotechnology continue, the feasibility and social legitimacy of microalgae-based wastewater treatment systems are expected to improve. Integrating S-LCA frameworks and multi-stakeholder engagement—from wineries and researchers to consumers—can provide a more holistic and equitable pathway toward the large-scale adoption of these sustainable treatment technologies [112].

Challenges, Limitations, and Future Perspectives

Despite notable advances in the application of microalgae for WWW treatment, several challenges remain before these systems can reach industrial-scale implementation. One of the main constraints lies in the variability and complexity of WWW composition, which fluctuates according to grape variety, winemaking techniques, and seasonal production cycles. Such heterogeneity affects nutrient balance, salinity, and organic load, thereby complicating process optimization and standardization across sites.

Operational and environmental limitations further constrain system performance. The high turbidity and dark coloration typical of WWW reduce light penetration, limiting photosynthetic activity in autotrophic cultures [59]. Similarly, high COD and variable organic loads can lead to oxygen depletion, pH instability, and the accumulation of inhibitory compounds, which complicate the management of both open ponds and closed photobioreactors [20]. Maintaining stable operational parameters thus requires robust monitoring and adaptive control strategies.

From a biological perspective, strain selection remains a critical bottleneck. Most studies still rely on model microalgae tested under controlled laboratory conditions, which do not reflect the physicochemical variability of real effluents. Developing or isolating polyextremophilic strains that can tolerate fluctuating temperatures, high salinity, and elevated organic loads while maintaining productivity and pollutant removal capacity is essential [31]. The use of algal–bacterial consortia represent a promising approach to enhance system resilience and biodegradation efficiency, yet further research is required to elucidate the ecological interactions and metabolic exchanges that underpin their stability and performance [26].

Monitoring and process control technologies are also evolving. Reliable, real-time tools to monitor microbial activity, biomass quality, and pollutant removal are not yet fully established for complex wastewater matrices. Advances in biosensing, spectroscopic techniques, and omics-based diagnostics are expected to improve system control, particularly when coupled with artificial intelligence or digital-twin frameworks [63]. These technologies could enable predictive maintenance, early detection of contamination or inhibition events, and improved decision-making in dynamic operational environments.

Regulatory and economic challenges represent another major barrier to deployment. The valorization of biomass derived from WWW in food, feed, or cosmetic applications is subject to stringent and often inconsistent regulations across jurisdictions [92]. In addition, the economic feasibility of microalgae-based treatment remains marginal under current market conditions. Standalone operation is rarely profitable unless integrated with existing winery infrastructures or supported by incentives for pollution abatement, water reuse, or circular resource recovery [113].

Addressing these challenges requires an integrated research and policy effort. Future studies should prioritize:

(i) strain improvement through adaptive evolution and genetic engineering;
(ii) development of hybrid systems that couple autotrophic and heterotrophic cultivation modes;
(iii) integration of renewable energy inputs and waste heat recovery to reduce operational costs; and
(iv) long-term demonstration studies combining techno-economic analysis and life cycle assessment under real operating conditions.

Beyond technical and economic constraints, Ortigueira and Lopes (2025) [101] emphasize that achieving sustainable scalability also depends on the inclusion of social dimensions and stakeholder engagement. The deployment of microalgae-based technology should therefore incorporate participatory approaches and S-LCA to anticipate social risks, evaluate community acceptance, and ensure equitable benefit distribution across actors [102]. Standardization of social and environmental indicators is likewise essential to improve comparability, enhance transparency, and strengthen policy and investment decisions.

Ultimately, translating the concept of microalgae-based WWW treatment into resilient, scalable, and socially accepted solutions requires a systemic approach that harmonizes technical innovation with regulatory frameworks and societal expectations. Establishing harmonized standards across sectors and countries would provide legal clarity, support market access, and promote circular bioeconomy development. Dynamic and adaptive policies—acknowledging the multifunctional role of these systems in wastewater purification, carbon mitigation, and biomass valorization—will be crucial to accelerate their transition from pilot demonstrations to widespread commercial adoption.

Conclusions

Microalgae-based treatment of WWW presents a compelling strategy for advancing sustainability and circular economy objectives. By integrating effective pollutant removal with the generation of value-added biomass, these systems offer dual benefits: environmental remediation and the production of bioproducts such as pigments, biofertilizers, biostimulants, biopesticides and proteins. This approach exemplifies the principles of circular resource management, where waste streams are transformed into economically and environmentally beneficial outputs.

Recent laboratory and pilot-scale research demonstrates that microalgal systems, when optimized for wastewater characteristics, cultivation conditions, and energy inputs, can achieve significant pollutant removal and resource recovery. TEA and LCA provide evidence that these systems have the potential for sustainability, provided that operational efficiencies are maximized and valorization pathways for the produced biomass are clearly defined. However, several operational challenges persist. These include process efficiency limitations, energy-intensive harvesting, and the need to enhance system robustness against wastewater variability. As a result, most implementations remain at the pilot or demonstration stage, highlighting the gap between promising research outcomes and widespread commercial adoption.

Scaling up microalgae-based WWW treatment is impeded by complex regulatory and economic realities. The valorization of biomass for food, feed, or cosmetic applications is subject to stringent and sometimes inconsistent regulations across different jurisdictions, creating uncertainty for industry stakeholders. From an economic perspective, the feasibility of these systems is often marginal under current market conditions, with standalone operations rarely profitable unless integrated with existing winery infrastructure or supported by incentives for pollution abatement, water reuse, or resource recovery. Additionally, the absence of standardized protocols and harmonized regulatory frameworks further complicates commercial deployment and market access.

Beyond technical and economic challenges, social dimensions play a critical role in the successful adoption of microalgae-based WWW treatment. Meaningful stakeholder engagement—including regulators, industry partners, local communities, and end-users—is essential to bridge the gap between laboratory success and practical implementation. Incorporating participatory approaches and S-LCA helps identify potential social risks, gauge community acceptance, and ensure equitable distribution of benefits. Standardizing social and environmental indicators is also vital for improving comparability, transparency, and the robustness of policy and investment decisions.

Achieving resilient, scalable, and socially accepted microalgae-based WWW treatment requires a systemic, integrated approach. Harmonizing regulatory standards across sectors and countries would reduce legal uncertainty, facilitate market access, and foster the development of a circular bioeconomy. Adaptive policies that recognize the multifunctional role of these systems—in wastewater purification, carbon mitigation, and biomass valorization—will be crucial to accelerate their transition from pilot projects to commercial reality. Future research and policy efforts should prioritize strain improvement, the development of hybrid cultivation systems, the integration of renewable energy and waste heat, and long-term demonstration studies combining TEA and LCA in real-world settings.

Microalgae-based WWW treatment holds significant promise for sustainable resource recovery and circular economy advancement. However, realizing its full potential requires overcoming technical, regulatory, economic, and social barriers through coordinated innovation, stakeholder involvement, and policy harmonization. A systemic approach that integrates technical advances with regulatory clarity, economic incentives, and inclusive stakeholder participation will be essential to transition from conceptual promise to practical, efficient, and broadly accepted solutions for WWW management.

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Data Availability No new data were created or analyzed in this study. Data sharing is not applicable.

Declarations

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