



Electrical Stimulation for Microalgal Growth and High-Value Bioproduct Production: Mechanistic Insights and Emerging Opportunities

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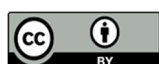
Abstract: The increasing severity of climate change and global energy insecurity has intensified the demand for sustainable and carbon-neutral biofuel production technologies. Microalgae are considered promising bioresources due to their rapid growth, high photosynthetic efficiency, and exceptional CO₂ fixation capability. However, large-scale commercialization remains limited by the metabolic trade-off between biomass growth and metabolite accumulation. Electrical stimulation (ES) has gained wide attention as an emerging strategy for regulating microalgal metabolism through modulation of membrane permeability, electron transport, intracellular redox balance, and reactive oxygen species (ROS)-mediated signaling pathways. This narrative review systematically summarizes recent advances in ES-assisted microalgal systems, focusing on biomass enhancement, lipid accumulation, and astaxanthin biosynthesis. Particular attention is given to the mechanisms by which electrical inputs affect photosynthetic activity, calcium-mediated signaling, intracellular carbon partitioning, and stress-responsive metabolic mechanisms. Overall, ES offers a versatile and environmentally friendly platform to stimulate microalgal growth and metabolite accumulation, supporting the advancement of sustainable microalgae-based biorefineries.

Keywords: sustainable biorefinery; lipid accumulation; astaxanthin biosynthesis; electrophysiology; nanosecond pulsed electric field; reactive oxygen species (ROS)

1. Introduction

The escalating global energy crisis and climate change are fundamentally driven by the continued overreliance on fossil fuels, which still account for more than 85% of global energy consumption according to Energy Institute [1]. This dependence is not only environmentally unsustainable but also geopolitically destabilizing due to the uneven distribution of fossil resources. For instance, disruptions in critical energy chokepoints such as the Strait of Hormuz—which handles approximately one-fifth of global oil and liquefied natural gas trade—can rapidly trigger global price volatility and energy insecurity [2]. Simultaneously, cumulative greenhouse gas emissions have already driven global temperatures to ~1.47 °C above pre-industrial levels in 2025, placing the targets of the Paris Agreement at imminent risk [3]. These converging pressures highlight an urgent need to develop scalable, carbon-neutral energy systems, particularly liquid fuels, which remain indispensable for sectors such as aviation, maritime transport, and heavy industry where electrification is not readily feasible.

Microalgae have gained considerable attention as a next-generation platform for sustainable biofuel production. As photosynthetic microorganisms—including green algae, cyanobacteria, diatoms, and industrially relevant genera such as *Haematococcus*, *Nannochloropsis*, and *Schizochytrium*—microalgae exhibit rapid growth kinetics and high photosynthetic efficiency, with the ability to fix approximately 1.83 kg of CO₂ per kg of biomass [4]. Under optimal conditions, certain species can double within ~3.5 h, enabling carbon fixation rates that are 10–50 times higher than those of terrestrial plants. This superior performance is attributed to their simplified unicellular structure and the presence of a CO₂ concentrating mechanism (CCM), which elevates intracellular CO₂ levels around Rubisco, thereby suppressing photorespiration and enhancing Calvin cycle efficiency [5,6]. Furthermore, microalgae



can be cultivated on non-arable land using wastewater or saline resources, positioning them as a highly versatile “cell factory” capable of simultaneously addressing carbon capture, resource recovery, and biofuel production [7,8].

Despite these advantages, the large-scale deployment of microalgae-based biofuels remains constrained by a fundamental metabolic trade-off between biomass accumulation and lipid synthesis [9]. Conventional strategies for lipid induction such as nitrogen limitation, salinity stress, or high light intensity, typically redirect carbon flux toward storage lipids at the expense of cellular growth, thereby reducing overall lipid productivity [10,11]. This trade-off reflects a shift from primary metabolism (biomass formation) to secondary metabolism (energy storage), often mediated by stress-induced signaling pathways and redox imbalance [12]. As a result, achieving simultaneous enhancement of both biomass and lipid content remains a primary bottleneck, limiting the economic feasibility of microalgal biofuel systems [13].

In this context, electrical stimulation (ES) has been increasingly investigated as a promising non-chemical, physically controllable strategy to regulate microalgal metabolism [14]. Unlike traditional stress-based approaches, ES enables precise modulation of cellular processes through tunable parameters such as voltage, current density, frequency, and exposure duration [15]. Figure 1 summarizes the proposed physiological effects of ES treatment in microalgae. Under an optimized safe-effective operating conditions, ES enhances photosynthetic electron transport efficiency while mitigating photoinhibition. It further regulates intracellular redox balance, reactive oxygen species (ROS) homeostasis, and membrane potential, leading to redirected carbon allocation through the Calvin cycle. These coordinated responses ultimately improve CO₂ fixation, biomass productivity, and the accumulation of valuable metabolites, including lipids and pigments [15].

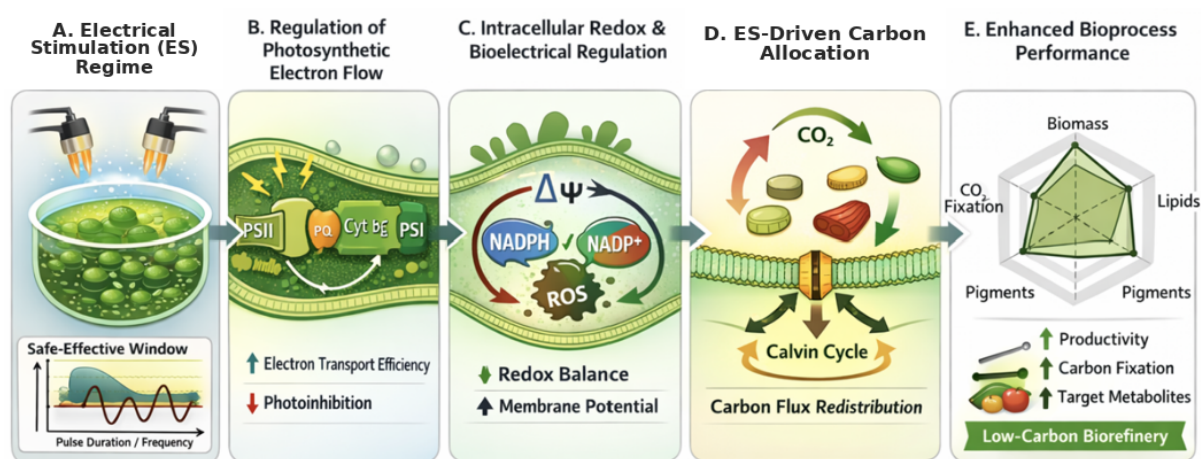


Figure 1. Schematic illustration of induced physiological regulation in microalgae by electrical stimulation (ES).

However, the effects of ES are highly nonlinear and system-dependent. While moderate stimulation can enhance metabolic performance, excessive intensity may lead to ROS overaccumulation, membrane damage, mitochondrial dysfunction, and ultimately growth inhibition or cell death [15,16]. This dual effect indicates that ES should not be regarded as a purely promotive factor, but rather as a precision regulatory tool governed by a delicate balance between beneficial and detrimental responses. Importantly, current studies remain fragmented, with significant variability in experimental conditions, species selection, and reported outcomes, leading to a lack of consensus on optimal operational parameters and mechanistic interpretation [17].

Therefore, a major gap in the field lies in the absence of an integrated framework for defining optimal stimulation regimes that maximize beneficial metabolic responses while minimizing the risk of cellular damage. This concept extends beyond simple parameter optimization, requiring an integrated understanding of the relationships among electrical stimuli, electrophysiological responses, redox dynamics, and metabolic flux regulation. Addressing this gap is essential for transforming ES from an empirical technique into a predictive and scalable engineering strategy.

Therefore, this narrative review systematically synthesizes recent advances in ES-assisted microalgal systems, with particular emphasis on mechanistic understanding and process optimization. Unlike previous reviews, which have primarily focused on the effects of electric-field treatment on microalgal growth [18] or on bioelectrochemical systems and external electrostimulation for lipid accumulation [19], the present review provides a broader and more integrated perspective. Specifically, it summarizes the effects of ES on microalgal growth and on the accumulation of lipids, pigments, and other value-added metabolites. By integrating biological, electrochemical, and process-engineering perspectives, this review highlights ES as a tunable strategy for regulating microalgal

carbon and energy fluxes, thereby supporting the development of economically viable and sustainable microalgal biofuel and bioproduct systems.

2. Electrostimulation as a Bio-Physical Modulation Strategy in Microalgae

Literature has demonstrated that externally applied electric fields can significantly modulate microalgal physiology; however, the underlying mechanisms, optimal operational conditions, and scalability remain insufficiently resolved due to substantial heterogeneity in experimental designs. This variability spans electrical modes (direct current, DC, alternative current, AC, nanosecond pulsed electric fields, nsPEFs), parameter ranges (μA to kV cm^{-1}), and cultivation environments, ultimately limiting cross-study comparability and process translation. Table 1 summarizes the employment of electrostimulation strategies in microalgae cultivation applications.

Electric fields disrupt cellular electrophysiology, including membrane potential, ion transport, and intracellular redox balance. For example, low-voltage AC stimulation at 1.36 kHz and 1.2 V achieved >95% inhibition of diatom adhesion, attributed to ROS overproduction, reduced dehydrogenase activity, mitochondrial dysfunction, and ATP depletion [20]. While this study provides strong mechanistic evidence for antifouling applications, its focus on inhibition rather than growth or metabolic enhancement limits direct applicability to bioprocess optimization. Moreover, reliance on endpoint metrics (e.g., adhesion inhibition) constrains insight into dynamic intracellular responses and dose–response behavior. In contrast, low-voltage DC stimulation under an optimal 0.005 V increased biomass of *Nannochloropsis oceanica* by 32.09%, primarily through enhanced photosystem II efficiency and electron transport, whereas excessive voltage (1 V) induced cellular damage [21].

Controlled electrical inputs can redirect microalgal carbon flux toward specific biochemical products, although the response depends strongly on the applied power, field strength, and treatment duration. Barreiros et al. showed that application of in situ electric field (INEF) of 0.82 W and 0.56 V cm^{-1} for 48 h on *Pavlova gyrans* increased lipid and carbohydrate accumulation by 4.72% and 18.7%, respectively [22]. By contrast, a shorter treatment of 0.21 W and 0.39 V cm^{-1} for 6 h preferentially enhanced chlorophyll and carotenoids by 74.9% and 66.2%, respectively. A similar dependence on system conditions was observed in *Dunaliella salina*, where low-current AC stimulation produced markedly different outcomes in different media. Specifically, 990 μA increased biomass by approximately 80% in synthetic medium, while only 50 μA was optimal in hypersaline produced water, increasing lipid productivity by 69% [23].

The importance of exposure mode is evident from DC-based stimulation studies. Short-term DC treatment was more effective than continuous exposure for *Nitzschia closterium* f. *minutissima* [16]. A 1 V short-term treatment increased cell density by 13.14%, whereas 3 V increased lipid content by 67.66%, suggesting that carbon allocation could be redirected toward storage compounds under appropriately selected conditions. However, continuous DC stimulation was generally less effective and even inhibitory, with only the moderate 0.5 V condition increased biomass yield by 39.47%.

Evidence from wastewater-based cultivation further extends the relevance of electrostimulation beyond defined laboratory media. Under optimized conditions involving light regime, growth phase, and energization time, electric field application increased microalgal biomass dry weight by 47.45% and lipid content by 28.28% [15]. Nutrient removal was also improved, with total nitrogen (TN) and NH_4^+ removal increasing by 18.5% and 10.9%, respectively. These responses were associated with changes in antioxidant enzyme activities, including catalase (CAT) and total superoxide dismutase (T-SOD), indicating that controlled oxidative stress may contribute to both metabolic regulation and nutrient assimilation. Thus, ES may provide value not only as a metabolite-induction strategy but also as a process intensification approach for wastewater-based microalgal biorefineries.

Table 1. Summary of electrostimulation as a bio-physical modulation strategy in microalgae.

Electrical Stimulation Mode	Objective/Mechanism	Key Results	References
Low-intensity in-situ electric field (INEF)	Electric fields act as a metabolic steering tool, more suitable for regulating product composition rather than maximizing biomass.	No significant growth enhancement, but substantial compositional shifts: chlorophyll a increased by 74.9%, carotenoids by 66.2%, lipids by 4.72%, carbohydrates by 18.7%, and proteins by 5.41%.	[22]
Low-voltage AC electric field	Antifouling / anti-adhesion application (not growth promotion). Low-voltage stimulation induces metabolic imbalance in inhibit attachment.	Diatom adhesion was significantly inhibited; ~95% inhibition by impedance method and ~97.2% by chlorophyll fluorescence. Accompanied by increased ROS, decreased ATP, and mitochondrial damage.	[20]
	Medium properties alter optimal stimulation conditions; high-conductivity media (e.g., produced water, PW) favor ultra-low current stimulation.	In synthetic medium: 990 μ A (twice daily) increased biomass by 80%; 750–990 μ A (once daily) increased total lipids by 68%. In PW: optimal at 50 μ A (once daily), with biomass increased by 23%, intracellular lipid fraction increased by 36%, and total lipid productivity increased by 69%.	[23]
Low-voltage DC electric field	DC stimulation exhibits an optimal operational window: too low is ineffective, too high is damaging.	Optimal at 0.005 V, increasing biomass dry weight by 32.09%. 0.5 V also promoted growth but less effectively. At 1 V, cells showed shrinkage, damage, and death.	[21]
Moderate-strength electrostatic field	Static electric fields can serve as a pretreatment strategy, with effects dependent on exposure time and inoculum density.	Exhibited a hormetic effect: moderate stimulation enhanced growth, while excessive exposure inhibited it. Total chlorophyll increased, but macromolecular composition (protein/lipid/carbohydrate) remained unchanged.	[24]
Nanosecond pulsed electric fields (nsPEFs)	nsPEFs act not only as electroporation tools but also as differentiation and stress signaling triggers.	Short-term: increased membrane permeability, cell swelling, elevated ROS/MDA. Long-term: cessation of cell division, increased cell size, secondary oxidative stress, and enhanced palmella differentiation.	[25]
	nsPEFs may also promote proliferation, but effects are highly dependent on stimulation frequency and timing, differing from differentiation responses reported elsewhere.	Single or one repeated treatment showed no effect; after three repetitions, growth significantly increased along with increased phycocyanin content.	[26]

Dose and duration are key factors determining whether electrical exposure promotes or suppresses microalgal growth. Nezammahalleh et al. reported that a single electrostatic treatment at 2.77 kV cm^{-1} enhanced the growth of *Chlorella vulgaris* [24]. The optimal condition of 0.4 g L^{-1} initial biomass concentration and 50 min treatment duration increased the growth rate by 51%. However, extending the exposure time to 70 min substantially reduced this beneficial effect. This time-dependent decline provides quantitative evidence that the positive effects of ES occur only within a limited exposure range, beyond which excessive electrical input may impair cellular function rather than promote growth.

High-intensity or pulsed electric fields induce significant physical alterations and physiological stress responses. nsPEF treatment at 40 kV cm^{-1} with 25–50 ns pulses induced transient membrane permeabilization, ROS accumulation, and subsequent growth arrest in *Chlamydomonas reinhardtii*, ultimately promoting differentiation into palmella-like states [25]. Repeated nsPEF exposure at 10.23 kV cm^{-1} with 100 ns pulses increased biomass in *Arthrospira platensis* while delaying cell division, suggesting a decoupling between biomass accumulation and proliferation [26]. The biorefinery relevance of nsPEFs was further supported by experiments with *C. vulgaris*, where treatment with 100 ns pulses at 5 Hz and 10 kV cm^{-1} during the early exponential phase increased biomass yield by up to $17.53 \pm 10.46\%$ ($p = 3.18 \times 10^{-5}$), with limited changes in carbon and pigment content but a reduction in protein fraction [27].

Across all studies, a consistent dose-dependent duality emerges: moderate stimulation enhances growth and metabolite accumulation, whereas excessive intensity or duration induces oxidative stress, membrane damage, and metabolic imbalance. This phenomenon is further influenced by system-specific parameters such as medium conductivity and impedance. Overall, these findings indicate that effective ES occurs within a narrow and system-dependent range of conditions, governed by electrical intensity, exposure duration, operational mode, and environmental factors.

3. Electrically Induced Lipid Accumulation in Microalgae: Mechanistic and Quantitative Synthesis

Electrical stimulation (ES) offers a non-chemical alternative to conventional nutrient starvation or chemical induction approaches for enhancing lipid accumulation in microalgae. Across different studies, lipid enhancement ranging from ~28.8% to 144% has been consistently reported under optimized electrical conditions [17,28]. These improvements are not merely empirical but arise from coordinated regulation of electrophysiological responses, redox balance, and intracellular carbon partitioning, collectively forming a mechanistic basis for targeted lipid biosynthesis. Table 2 compares different ES strategies for microalgal lipid accumulation.

Low-voltage DC stimulation (1–3 V) to *Nitzschia closterium* raised lipid content up to 67.66% [16], while a 2.09-fold elevation in triacylglycerol (TAG) was achieved in *Chlorella* sp. as a result of electrochemical treatment at 4 V and 31 mA for 4 h [29]. Meanwhile, AC stimulation at 50 mA with 30 min daily enhanced lipid content by 144%, with performance closely correlated to medium impedance [17]. These increments are also closely associated with elevated ROS levels, which function as regulatory molecules that activate pathways related to fatty acid and TAG formation. Specifically, ROS-mediated regulation has been linked to the upregulation of key enzymes such as acetyl-CoA carboxylase (ACCCase) and diacylglycerol acyltransferase (DGAT), thereby promoting TAG synthesis [13].

In a two-stage cultivation strategy combining microcurrent and salinity stress, lipid content reached 42.11% (53.13% improvement), accompanied by a 61.25% increase in biomass [11]. This is attributed to enhanced availability of acetyl-CoA, derived from the mobilization of carbohydrate reserves, and concurrent suppression of protein synthesis routes [11,29]. This indicates that ES acts as a tunable redox switch, directing metabolic flux toward lipid storage as a protective response to oxidative stress.

At the process scale, ES has demonstrated significant potential to enhance lipid biosynthesis in integrated platforms. In wastewater-based growth system, applying $1.0\text{--}2.0 \text{ A/m}^2$ current density raised lipid accumulation by 30.1% while simultaneously improving nutrient removal [10]. Similarly, electro-assisted bacteria–algae cultures achieved a 13.3% enhancement in lipid level under 1.0 V DC treatment, alongside enhanced phosphorus removal efficiency of 94.4% [30].

Table 2. Comparison of different ES strategies for microalgal lipid accumulation.

Electro-Stimulation Strategies	Primary Mechanism	Advantages	Limitations/Challenges
Alternating Current (AC)	Cell membrane potential regulation, metabolic activity enhancement	Low energy consumption, stable operation	Requires precise micro-ampere level control
Direct Current (DC)	Accelerated electron transfer, environmental electrochemical changes	Can combine with wastewater treatment, improves TN/TP removal	Continuous application may cause electrochemical toxicity or inhibition
Pulsed Electric Field (PEF)	Cell membrane electroporation, temporary increase in membrane permeability	Strong lipid induction effect	Requires high-voltage equipment, complex parameter optimization
Magneto-electric Composite Fields (MECs)	Metabolic activity enhancement, carbon distribution regulation	Multiple stimulation effects, promotes storage metabolite accumulation	Complex equipment, mechanisms require further study

Notably, optimal lipid yield consistently occurs within a moderate operating mode, where oxidative signaling and metabolic activation are maximized without inducing cellular damage [17, 28]. This highlights the importance of defining safe and effective operational conditions for lipid-oriented microalgal bioprocesses, thereby providing a rational basis for the design and optimization of electrically assisted lipid production systems.

4. Electrically Induced Astaxanthin Accumulation in Microalgae: Mechanistic and Quantitative Synthesis

Early work on *Haematococcus pluvialis* provided initial evidence that ES can promote both cell growth and astaxanthin formation, although the beneficial response appears to occur only within a restricted operational range. In the study by Kim et al., treatment at 100 mA and 25 V increased cell density by approximately 120%, while periodic stimulation further improved astaxanthin concentration by approximately 10%, reaching 32.6 mg/L [31]. The greater effectiveness of periodic rather than continuous exposure indicates that the temporal pattern of electrical input is as important as the applied current itself. By introducing recovery intervals between treatments, an intermittent regime may help preserve membrane integrity, redox balance, and cellular metabolism, thereby reducing the cumulative damage associated with prolonged exposure.

The importance of electrical polarity was further demonstrated by Fitriana et al., who compared anodic and cathodic treatment in a dual-chamber bioelectrochemical system using *H. pluvialis* [32]. Under anodic conditions, treatment at 4 V and 30 mA for 2 days enhanced astaxanthin production by 36.9% relative to the control; however, increasing the voltage to 6 V reduced astaxanthin productivity. Thus, higher energy input does not necessarily produce stronger carotenoid induction. The cathodic response was more damaging: treatment at 2 V and 3 mA caused severe cell lysis, membrane rupture, and a 57% decrease in astaxanthin content. Cyclic voltammetry analysis further showed that oxidative and reductive environments elicited distinct cellular responses, even within apparently comparable voltage ranges. These observations indicate that polarity, current intensity, and the resulting redox environment collectively determine whether ROS function as metabolic signals or cytotoxic stressors.

A comparable dose-dependent response was reported by Sathiyavahisan et al. in *Haematococcus lacustris* [33]. Mild anodic stimulation at 10 mA for 4 h increased astaxanthin content by 21.8–34.9% and also promoted the accumulation of other carotenoids. When the current was raised to 20–30 mA, cell viability declined to 84.2% and 65.6%, respectively, accompanied by lower pigment production. This shift from induction to inhibition supports a threshold-based interpretation of ROS activity. At moderate levels, ROS can activate stress-responsive carotenoid biosynthesis; beyond this range, oxidative pressure exceeds cellular antioxidant capacity and leads to injury. ROS-mediated signaling should therefore be regarded as condition-dependent rather than uniformly beneficial.

Beyond conventional ES strategies, nanosecond pulsed electric fields (nsPEFs) provide a more targeted means of perturbing intracellular regulatory pathways. Bai et al. found that nsPEFs altered the expression of key astaxanthin biosynthesis genes, including *psy*, *crtR-b*, and *bkt1* [34]. Among the tested pulse lengths, the 50 ns treatment produced the strongest transcriptional upregulation by the third cultivation day, whereas the 25 ns pulse caused transcript levels to decline after two days. Pulse length therefore influences both the intensity and persistence of metabolic regulation. Evidence involving respiratory burst oxidase homolog (RboH) and Ca²⁺ regulation also suggests that plasma membrane-mediated processes participate in cellular adaptation to nsPEFs. One proposed mechanism involves transient permeabilization of the plasma or chloroplast membrane, followed by plastid-to-nucleus retrograde communication and subsequent modulation of stress-responsive gene expression.

Whether these transcriptional changes can be converted into stable productivity gains under scalable cultivation conditions remains unresolved.

ES may also affect developmental transitions that are directly associated with astaxanthin yield. In the study by Lakshmi Narasimhan et al., ES was applied to induce cyst germination in *H. lacustris*, with optimal conditions of 2 V, 1.4 A, and 60 min increasing the germination rate by 2.2-fold and astaxanthin formation by 73% [35]. The researchers attributed this response to the activation of defense-related ROS signaling, improved germination activity, accelerated nutrient depletion, and intensified astaxanthin production as a stress-protective adaptation. Rather than acting only as a direct metabolic trigger, ES may therefore also serve as a tool for coordinating life-cycle status with secondary metabolite production.

Collectively, these studies show that ES can enhance astaxanthin accumulation, with reported improvements in total astaxanthin yield and productivity of up to 1.7-fold and 1.5-fold, respectively. Direct comparison remains challenging because the reported systems differ substantially in voltage, current intensity, polarity, pulse duration, waveform, frequency, exposure time, medium conductivity, and reactor configuration. Despite this variability, a consistent working model can be proposed: moderate ES induces controlled ROS- and Ca²⁺-related signaling, modifies membrane permeability, and activates pigment biosynthesis, whereas excessive exposure promotes oxidative damage, membrane disruption, and loss of viability.

5. Conclusions and Future Perspectives

Electrical stimulation (ES) shows strong potential as a non-chemical and tunable strategy for regulating microalgal metabolism, with demonstrated benefits for biomass productivity, lipid accumulation, nutrient removal, and astaxanthin biosynthesis. Current evidence indicates that ES modulates cellular electrophysiology, membrane permeability, electron transport, intracellular redox balance, ROS-mediated signaling, and calcium-related responses, thereby redirecting carbon and energy fluxes toward targeted metabolic outcomes. Under optimized conditions, moderate electrical inputs can enhance growth, lipid synthesis, pigment accumulation, and nutrient assimilation across diverse microalgal species and treatment modes. However, these responses are highly nonlinear and strongly influenced by electrical intensity, duration, waveform, operational mode, medium properties, and cultivation environment. Moderate ES generally acts as a beneficial metabolic trigger by promoting controlled ROS signaling, membrane polarization, and electron transport efficiency, whereas excessive exposure may induce oxidative stress, membrane disruption, mitochondrial dysfunction, growth inhibition, or cell death. These findings emphasize the need to define system-specific stimulation regimes that maximize metabolic enhancement while maintaining cellular integrity.

Future research should prioritize standardized and multidimensional ES frameworks that integrate voltage, current density, pulse duration, frequency, waveform, exposure mode, medium conductivity, impedance, ionic strength, and reactor hydrodynamics into predictive operational models. The incorporation of real-time physiological indicators, including ROS dynamics, NAD(P)H balance, photosystem II efficiency, membrane potential, and antioxidant activity, could support adaptive and feedback-controlled ES systems. In parallel, pilot-scale validation in wastewater-based, continuous, and integrated biorefinery platforms is essential to assess long-term stability, energy efficiency, electrode durability, and techno-economic feasibility. With improved mechanistic understanding, process optimization, and scale-up engineering, ES could advance from an empirical stimulation approach to a precision bio-physical regulation strategy for next-generation sustainable microalgae biorefineries and carbon-neutral biofuel production.

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