EEET ECOLOGICAL ENGINEERING & ENVIRONMENTAL TECHNOLOGY

Ecological Engineering & Environmental Technology, 2025, 26(11), 101–117
https://doi.org/10.12912/27197050/211755
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Received: 2025.09.22
Accepted: 2025.10.20
Published: 2025.11.01

Environmental and biotechnological potential of *Dunaliella salina* for sustainable environmental applications

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ABSTRACT

This review critically assesses the environmental and biotechnological potential of *Dunaliella salina*, emphasizing how abiotic stress factors shape its physiology, carotenoid and lipid biosynthesis, and its prospects for sustainable applications in industry and ecology. A synthesis of peer-reviewed studies published between 2015 and 2025 highlights the effects of salinity, light intensity, nitrate limitation, temperature, and pH on the species' metabolic responses, while also comparing cultivation systems such as open ponds and photobioreactors in terms of productivity, efficiency, and scalability. Particular attention is given to advances in omics technologies and genetic engineering that enhance metabolite yields. Results show that high salinity (3–4 M NaCl) and strong light exposure (> 300 µmol photons m^{-2} s^{-1}) significantly promote β-carotene accumulation, reaching levels of 10–14% of dry biomass, while nitrogen limitation can double lipid content. Photobioreactors demonstrate β-carotene productivities exceeding 600 $mg \cdot m^{-2} \cdot day^{-1}$, outperforming traditional open pond systems. Moreover, *D. salina* exhibits strong bioremediation capabilities, removing up to 98% of heavy metals in saline effluents and degrading approximately 70% of organophosphate pesticides. Despite these promising results, large-scale industrial deployment remains limited by high production costs, strain variability, and energy requirements for controlled cultivation systems. Nevertheless, the findings position *D. salina* as a valuable resource for sustainable bioindustries, particularly in the context of circular bioeconomy models and environmental remediation strategies in hypersaline and arid environments. By integrating insights from environmental biotechnology, omics-based optimization, and ecological valorization, this work underscores the strategic importance of *D. salina* for the development of the green bioeconomy and as a model organism for climate adaptation approaches.

Keywords: *Dunaliella salina*, β-carotene, halotolerance, microalgae, abiotic stress, photobioreactor, biotechnology, bioeconomy.

INTRODUCTION

Dunaliella salina is a remarkable halophilic unicellular microalga that can grow in extreme conditions not normally occupied by other life (Davidi et al., 2023). Extreme conditions include hypersaline lakes, salt marshes, saline lagoons, and some desert environments. These environmental conditions are defined by extreme salinity, fluctuations in temperature, high ultraviolet radiation (UV), and sometimes low oxygen levels (Oren, 2014). These are uninhabitable conditions, however, *D. salina* is an integral contributor to ecological balance in these environments (Guermazi et al., 2024). *D. salina* is a major primary producer in the hypersaline environment (He et al., 2020), as it can perform photosynthesis in extreme conditions. Often, it is described at the base of the food web, providing biomass and energy to microorganisms, invertebrates, and other life forms adapted to the hypersaline conditions. Further, this microalga is contributing to biogeochemical cycles, such as the carbon cycle, in which it is fixing atmospheric CO_2 to organic matter (Rodriguez-Valera, 2020). Thus, *D. salina* also adds to the stability and resilience of hypersaline aquatic systems during environmental stresses (He et al., 2020). In fact, the presence, and population dynamics, of *Dunaliella salina* are often bioindicators for the detection of environmental changes, including climate change (Zarkami et al., 2020). Their sensitivity to altering abiotic parameters, such as salinity and temperature, makes them important natural sentinels for monitoring and managing extreme ecosystems (Gutierrez-Canovas et al., 2019).

Besides having a possible ecological role, there are a variety of biotechnological applications with environmental implications that may be considered to be a strategic resource to help engineers address pollution, climate change, and sustainability. (Ramos et al., 2011). There are two applications that are especially noteworthy. First, *Dunaliella salina* has excellent potential for use in bioremediation. In addition to having the ability and possible potential to tolerate and accumulate many heavy metals, and a number of pollutants, it would be a strong candidate for remediation of treated waters, especially in industrial and mining areas that create saline effluents (Barbosa et al., 2023). In this manner, *D. salina* could contribute to ecological restoration of degraded and polluted habitats and environments by transforming or sequestering hazardous toxic substances (Sarker and Kaparaju, 2024). Second, *Dunaliella salina* is also a very effective photosynthetic organism that can sustainably grow in some extreme conditions and therefore performs the role of carbon dioxide (CO_2) sequestration, which is a greenhouse gas that greatly contributes to heating the planet (Ambika and Vijayan, 2025). The high biomass yield, and Rapid Growth rates of *Dunaliella salina* indicate that CO_2 could be naturally and sustainably removed from the atmosphere especially for areas that are not suitable for agricultural crop production (Benedetti et al., 2018).

Dunaliella salina also presents an opportunity to optimize the environmental resources considered marginal within a circular economy framework (Santos et al., 2025). The cell can catalyze the organic waste (e.g., crab shells, by-products of mollusks) into biomass when associated with minerals in a hypersaline system that is rich in bioactive materials (e.g., carotenoids, lipids, proteins) in demand by the food, cosmetic, pharmaceutical, and energy sectors (Martins et al., 2023). The biovalorization of this type of local biomass helps to reduce reliance on non-renewable resources and contaminants to waste while potentially providing a framework for a sustainable and responsible natural environment (Haregu et al., 2023). In fact, *Dunaliella salina* has sophisticated multiple levels of adaptive strategies such as huge quantities of antioxidants and osmotic adjustments which create a biological model that may produce innovative ecological applications to assist humans and ecosystems adversely affected by climate change (Castellanos-Huerta et al., 2022). The former strategies could promote the paradigm shift in biotechnologies for managing fragile systems, advancing resistance crops, or generating sustainably bioactive compounds for human health and/or improvements to the environment (Sharma et al., 2024).

The study of sustainable and environmentally responsible biological resources to produce high value-added molecules has generated new interest in microalgae (Ezhumalai et al., 2024). These unicellular photosynthetic microorganisms provide considerable biotechnological options by virtue of their rapid growth, the ability to accumulate numerous secondary metabolites, their ability to survive in extreme environments, and not competing with traditional agriculture (Barone et al., 2023). Within this group of organisms, *Dunaliella salina*, a halotolerant chlorophyte, is no exception; this microalga is recognized for its ability to synthesize substantial amounts of β-carotene, a naturally occurring carotenoid pigment, whose important properties are as an antioxidant, provitamin, and photoprotectant, along with several important fatty acids, which are in demand for food, cosmetics, and biofuels (Sun et al., 2023). *Dunaliella salina* was discovered at the beginning of the 20th century in hypersaline ecosystems, and in particular in salt marshes, salt lakes and artificial solar salterns (Verdev and Dolinar, 2025). Different from many other microalgae, it has no rigid cell wall and therefore possesses extraordinary morphological and osmotic plasticity, to adapt with extreme environmental conditions, particularly high salt concentrations (> 300 g/L) (Simansky et al., 2024). The extraordinary adaptive capacity is paired with significant modulation of metabolism; *D. salina* under abiotic stressors such as salinity, nitrogen deficiency or light stress will activate secondary metabolic pathways that lead to the intracellular accumulation of β-carotene and lipids (Bhardwaj et al., 2025).

In an energy transition context, chemical input reduction and a search for natural sources for industry, *Dunaliella salina* appears to provide a sustainable solution. Its use for the production of natural β-carotene could be an attractive alternative to chemical synthesis, which currently holds the position of being primary but dependent on petroleum and energy-consuming (Harvey and Ben-Amotz, 2020). Besides, its fatty acids can represent a potential source of third-generation biofuels and nutritional supplements (Gajraj et al., 2018). A number of experimental studies have also shown technical feasibility for the cultivation of this microalga in open systems with controlled salinity but lower technological costs (Sun et al., 2022).

Although extensive studies have been conducted on *Dunaliella salina* as a model organism for understanding salt tolerance and carotenoid biosynthesis, clear gaps remain in understanding Dunaliella's potential from the perspective of ecological engineering. Most previous studies examining Dunaliella's potential produced value through optimization of β-carotene or lipid production in laboratory studies, while findings were not examined for sustainable environmental or industrial use. Further, studies on *Dunaliella salina* in circular bioeconomy models, and saline wastewater remediation and nutrient recovery in excessive salinity systems have been limited in publication. Again, to the authors knowledge, limited systematic analysis linking abiotic stress responses such as salinity, light intensity, and limited nitrate concentration to physiologically-driven regulation and bioprocess development at bioprocess scale has yet to be reviewed. Therefore, the novelty of the present study involves linking an important knowledge gap to a coherent synthesis of physiological, biotechnological, and environmental consequences of *Dunaliella salina*. Further, the synthesis will synthesize the integration of recent findings through omics-based literature, cultivated state, and eco-engineering applications. Aditionally, this study provides a framework for *Dunaliella salina* as a sustainable biological platform addressing biological problems related to remediation, extraction of resources, and resisting environmental change across extreme saline habitats. The objective of this review is twofold: (i) to synthesize current knowledge on the impact of abiotic factors on the production of β-carotene and fatty acids in *Dunaliella salina*, based on both classical studies and recent research; and (ii) to explore the prospects offered by modern biotechnologies, such as genetic selection, photobioreactor optimization, and environmental modeling, to improve the yields and industrial applications of this exceptional microalga.

BIOLOGY AND PHYSIOLOGY OF *DUNALIELLA SALINA*

D. salina is a unicellular microalga in the Dunaliellaceae family of Chlorophyceae. It is incredibly tolerant of very saline environments that exceed 30% (w/v) salt concentrations, which is a niche that few organisms can tolerate (Wang et al., 2021). These features offer *D. salina* exclusive attributes as a model organism to investigate osmoregulatory and oxidative stress. Morphologically, *D. salina* does not present a rigid membrane to provide volumetric plasticity. *D. salina* exhibits the ability to modulate its size through tight regulation of intracellular glycerol, an osmoprotectant used to offset osmotic pressure (Lv et al., 2021). Furthermore, *D. salina* regulates secondary metabolites that tie into carotenoid accumulation, such as β-carotene (Chen et al., 2024). This fat-soluble pigment accumulates in lipid globules within the chloroplasts, and can perform a photoprotective role to neutralize excess light and reactive oxygen species. In addition to that, *D. salina* can produce a number of different saturated and unsaturated fatty acids (e.g., palmitic (C16:0) linoleic (C18:2), and linolenic (C18:3) acids) that are key to membrane fluidity and resistance to environmental stressors (Xi et al., 2021).

Recent advancements in genomics and transcriptomics have generated a plethora of genes and regulatory elements associated with carotenoid and lipid biosynthesis, providing opportunities to leverage biotechnology for optimizing production of these metabolites (Xiang et al. 2025). The cultivation potential of *D. salina* given their simple, vegetative life cycle, quick cell cycle and easier growth under controlled conditions, underscores its robust potential for the production of commercially important metabolites (Song et al. 2024). In utilizing data from a variety of environmental conditions to demonstrate the impact on D. salina, we will develop a comparative summary of the most relevant abiotic stressors (salinity, temperature, light intensity, and/or UV light), alongside some indication of how they can impact physiology, carotenoid and lipid accumulation and biomass yield of this microalga; as well as some indication of the strategies provided by the microalga compromising the abiotic stress and environmental class of the productivity (Table 1).

EFFECTS OF ABIOTIC FACTORS ON THE PRODUCTION OF Β-CAROTENE AND FATTY ACIDS IN *DUNALIELLA SALINA*

Abiotic factors play a decisive role in regulating the metabolic pathways of *D. salina* and directly influence growth, biomass production, and the accumulation of high value-added compounds such as β-carotene and fatty acids. Among these factors, salinity, nutrient availability, light intensity, and temperature are the most studied, each inducing specific physiological and biochemical responses (Wu et al., 2020).

Salinity

Salinity is a major environmental variable in the natural environments of the halophyte *D. salina*, which has the ability to tolerate extreme salinities varying from 0.5 M and exceeding 5 M NaCl. The high salinity tolerance is made feasible due to extremely effective osmoregulation mechanisms, most importantly, intracellular glycerol accumulation, allowing osmotic pressures inside and outside the cell to be balanced. In addition to survival, salinity also affects secondary metabolite production. Multiple studies have reported that high salinity stress (e.g., 3–4 M NaCl, approximately 17–23%) can substantially increase β-carotene biosynthesis (Capa-Robles et al., 2021). This has been interpreted as an adaptation to help protect the cell against oxidative stress that might arise from excess chloride and sodium ion influx and any osmotic imbalance. As with other carotenes, the increase in β-carotene provides substantial absorbance of excess light energy, and it has been reported that β-carotene acts as a free radical scavenger (Lamers et al., 2010). Furthermore, salinity also affects lipids. There are multiple studies suggesting that high salinity increases the percentage of saturated fatty acids, including palmitic, which contribute to plasma membrane rigidity and limit lipid peroxidation. However, depending on the salinity levels, high salinity could impede cell growth, so there is an inherent trade-off between the stress producing interesting compounds and sufficient biomass for the future (Hashemi et al., 2021).

Nitrate concentration and nutrient limitation

Nitrates, the source of nitrogen that can be assimilated, have a strong effect on the physiology of *D. salina*. Also, in conditions of high nitrate availability, the microalgae prioritize cell growth and division but rarely accumulate (beta)-carotene. In conditions of nitrogen limitation (low

Table 1. Summary of abiotic stress effects on *Dunaliella salina* physiology and metabolite production

Abiotic factor	Physiological response	Effect on β-carotene production	Effect on lipid production	Effect on growth / biomass	References
High salinity	↑ Intracellular glycerol (osmoprotection), ↑ cell volume regulation	↑↑ Strong stimulation	↑ Moderate stimulation	↓ Slight to moderate reduction	Wang et al., 2021
High light intensity	↑ ROS formation, ↑ photoprotection mechanisms	↑↑ Major stimulation (photoprotection role)	↑ Slight increase	↓ At excessive intensities	Zhang et al., 2017
Nitrogen limitation	↑ Carbon redirection to secondary metabolism	↑ Significant stimulation	↑↑ Major lipid accumulation	↓ Biomass decrease	Fachet et al., 2020
Temperature stress	Altered enzyme activity, membrane fluidity	↑/↓ Variable depending on temperature	↑/↓ Variable	↓ If above/below optimal	Lynch & Thompson, 1982
pH variation	Altered nutrient availability and membrane transport	↑ Slight stimulation (at alkaline pH)	↑ Slight variation	↓ At acidic or highly alkaline pH	Sui & Vlaeminck, 2019
Combined stresses	Synergistic or antagonistic effects	↑↑ Often highest under combined light + salinity	↑↑ Synergy possible	↓ Often reduced due to cumulative stress	Capa-Robles et al., 2021

Note: ↑ : moderate increase ; ↑↑ : strong increase ; ↓ : decrease.

nitrate concentration, low mM KNO_3, e.g. 1–2 mM KNO_3), the accumulation of (beta)-carotene is well promoted (Wu et al., 2015). This accumulation is attributed to a reduction of the metabolic activity in the microalgae, in particular in protein and photosynthetic apparatus synthesis, while the secondary carotenoid pathways become activated, which was induced partly by the metabolic stress imposed on the microalgae. In addition to promoting carotenoid accumulation, nitrogen limitation is also correlated with increases in neutral lipids, mainly in the form of triglycerides, which are implicated in energy storage and cellular protection (Almutairi, 2020). Fatty acids produced under nitrogen-limiting conditions also contain a characteristic range of primarily polyunsaturated fatty acids, which have known positive health benefits for humans (including linolenic acid). Interaction between salinity and nitrate availability is particularly important; a study indicate maximum (beta)-carotenoids can be derived from growing *D. salina* at a salinity (3.5 M NaCl) with moderate available nitrate, indicating the balance of osmotic stress and nutritional stress activates carotenoid accumulation (Najafabadi and Naeimpoor, 2023).

Light intensity and photo-oxidative stress

While light is crucial for photosynthesis, too strong of a light can create oxidative stress through the production of reactive oxygen species and free radicals. *D. salina* responds to oxidative stress by increasing the biosynthesis of-products such as carotenoids like β-carotene, which is an example of both a filter and an antioxidant. Experimental findings have shown an increase in β-carotene levels due to enhanced light intensity, especially from the red and blue wavelengths (Kim et al., 2024). The impact of stress can be dependent on the duration of stress applied. Nonetheless, other studies showed that the duration of stress has little impact on the fatty acid profile. Light management is a critical factor of importance in cultivation systems, especially in photobioreactors, where light can be manipulated. Intermittent light, or modulated light, can promote carotenoids under light stress and mitigate photoinhibition (Aditi et al., 2025).

Temperature, pH, and other factors

Temperature influences the speed of enzyme reaction and membrane fluidity and thus influences growth and metabolism; the ideal temperature for *D. salina* is 25–35 °C, after which productivity decreases sharply after 40 °C. pH of the medium influence's nutrient availability and enzyme stability (Xi et al., 2020). *D. salina* in general can tolerate slightly alkaline pH (7.5–8.5), but variability will affect photosynthetic rate. Other parameters like availability of CO_2 and aeration are also of interest, particularly in intensive cultures, since CO_2 concentration limits photosynthesis (Morowvat and Ghasemi, 2016).

Interactions between abiotic factors

In the natural environment, as in the cultural environment, abiotic factors can be acting at the same time, which creates either synergistic or antagonistic interactions. For example, high salinity with high light stress may produce maximized β-carotene synthesis but reduced growth. Industrial optimization consists of a trade-off for maximum yield of the target product while maintaining adequate biomass (Wu et al., 2020).

CULTIVATION STRATEGIES AND YIELDS OF *DUNALIELLA SALINA*

The cultivation of *Dunaliella salina* for the commercial production of β-carotene and fatty acids requires precise control of environmental conditions and adaptation of cultivation systems to economic and technical constraints. Two main categories of systems are used: open (or semi-intensive) ponds and closed photobioreactors. Each has specific advantages and limitations in terms of yield, costs, and operational complexity (Borovkov et al., 2020).

Open basin cultivation

Open ponds, which are shallow (20 to 30 cm) are the simplest and least expensive systems and can be used for mass cultivation in warm, sunny climates. These ponds can be established in natural salt lakes or artificial salt pans, which can be naturally saline and limit chances of contamination by other microorganisms. As an example, the Tissa continental salt marsh, in Morocco, has a semi-intensive pilot-scale pond system of five ponds with a total of 1.800 L, achieving an average cell density of approximately 3.1×10^6 $cells \cdot mL^{-1}$, with β-carotene production of up to

39 mg·L^{-1}, or approximately 484 mg·m^{-2}·day^{-1}. with minimal investment and a system easily manageable by local onsite operators, demonstrates the potential industrial-scale feasibility of *D. salina* under outdoor open pond conditions (Borovkov et al., 2019). However, open ponds have limitations as systems that can be subject to climate fluctuations, biological contamination (bacteria, other algae), as well as evaporation, which can change, salinity, and pH among other important influencing cultivation parameters. and more challenging light and nutrient control impact on yield and consistency (Al-Mhanna et al., 2023).

Closed photobioreactors

Enclosed photobioreactors (PBRs) are a high-level technological solution that enables effective control of culture parameters (temperature, light, pH, carbon dioxide and nutrients) and more protection from contamination (García-González et al., 2005). PBRs may be tubular, flat, or columnar and tend to be designed to maximize the light exchange surface area per volume of culture. The PBR culture yields higher cell densities, higher β-carotene production and better reproducibility (Zhu and Jiang, 2008). The investment and operational costs of PBRs are substantially higher than for open ponds. They are difficult technology to optimize, as well as requiring substantial energy inputs and consistent specialized maintenance, which limits their adoption in constrained economies or for low-cost production (Prieto et al., 2011).

Optimal cultivation parameters

To maximize the production of β-carotene and fatty acids, several parameters (Al-Mhanna et al., 2023) must be jointly optimized:

- Pond depth: A shallow depth (20–30 cm) allows for good light penetration and efficient aeration,
- Salinity: Between 15 and 30% NaCl, depending on the strain and type of culture,
- Nitrate concentration: Moderate (1–5 mM) to balance growth and metabolite accumulation,
- Light intensity: High but regulated, with alternating cycles to avoid photoinhibition,
- Temperature: Optimal between 25 and 35 °C,
- Aeration and CO_2: Essential for pH stabilization and carbon supply.

Complementary techniques

Stimulation through controlled stress: Cycles of nutrient limitation or light stress can be applied to induce β-carotene accumulation without compromising biomass production.

Co-culture or microalgae/bacteria association: Certain bacteria can stimulate the growth or metabolite synthesis of *Dunaliella salina* (Gonabadi et al., 2022). Automation and sensors: Real-time monitoring and control of physicochemical parameters improve yields and product quality (Dębowski et al., 2025)

Yields and productivity

Productivity varies widely depending on the system, growing conditions, and strain used. Open ponds generally achieve between 300 and 500 mg·m^{-2}·day^{-1} of β-carotene, while PBRs can exceed 600 mg·m^{-2}·day^{-1} under optimal conditions. These values should be compared with biomass productivity, which is often 20 to 40 g·m^{-2}·day^{-1} (Colusse et al., 2020).

INDUSTRIAL APPLICATIONS OF *DUNALIELLA SALINA*

D. salina can produce large quantities of natural β-carotene, essential fatty acids, and other bioactive compounds, which makes it a microalga that is very useful in several industrial sectors: agri-food, cosmetic, pharmaceutical and energy. It is versatile because of its biochemical characteristics, ease of cultivation, and the increasing interest in natural and sustainable products (Francavilla et al., 2010). While *D. salina* presents unique advantages, its biotechnological relevance is better understood when compared with other microalgae widely exploited in industry, such as Chlorella and Nannochloropsis (Table 2). This comparative perspective highlights that although *D. salina* is particularly suited for carotenoid production under extreme conditions, Chlorella and Nannochloropsis dominate other niches such as protein-rich biomass and omega-3 fatty acid production, respectively (Jo et al., 2024; Dinesh et al., 2025).

Production of natural β-carotene

D. salina's main carotenoid pigment is β-carotene. β-carotene is a well-known natural

Table 2. Comparative features of *Dunaliella salina*, *Chlorella* and *Nannochloropsis*

Characteristics	*Dunaliella salina*	*Chlorella* sp.	*Nannochloropsis* sp.
Optimal environment	Hypersaline (>30% NaCl), high stress tolerance	Freshwater mainly, some strains tolerate moderate salinity	Marine and brackish environments
Cell wall	Absent (high osmotic plasticity)	Present, cellulose-rich (more difficult extraction)	Present, resistant (lipids entrapped)
Major metabolites	β-carotene (up to 10–14% dry biomass), glycerol, unsaturated fatty acids	Proteins (up to 50–60% dry biomass), chlorophyll, polysaccharides	Lipids (20–60% dry biomass), EPA (eicosapentaenoic acid, ω-3)
Industrial applications	Natural colorants (β-carotene), cosmetics, biofuels, saline bioremediation	Protein supplements, human and animal nutrition, biofertilizers	Nutraceuticals rich in omega-3, aquaculture, biodiesel
Advantages	Growth under extreme (hypersaline) conditions, high carotenoid productivity	High protein productivity, easy large-scale cultivation	Very rich in lipids/ω-3, high cell density
Limitations	Limited biomass yield, costly extraction	Difficult metabolite extraction (thick cell wall)	Slower growth, costly extraction

antioxidant and provitamin A that is used extensively as a natural food colourant in dairy products, juices, confectionery, and dietary supplements. The natural β-carotene that *D. salina* produces is more bioavailable than petrochemical β-carotene, as well as being in a better isomer configuration making it very useful in the cosmetic and pharmaceutical industries. Applications include anti-aging products, sunscreens, and dietary supplements for vision and immune health. The Moroccan model of open-basin cultivation will provide semi-industrial production, such as in the Tissa saltworks under suitable climate and saline conditions (Xu et al., 2018).

Production of fatty acids and lipids

D. salina produces polyunsaturated fatty acids (ω-3 and ω-6) along with carotenoids, which are essential for human and animal nutrition, given their anti-inflammatory and cardiovascular effects. These lipids could be used for dietary supplements, functional foods, and cosmetics. Their use as feedstocks for third-generation biofuels is also being considered, potentially providing a sustainable energy source (Madkour et al., 2025).

Cosmetics and dermatology

D. salina extracts are being used more often in cosmetic products because of their antioxidant, moisturizing, and photoprotective properties. Carotenoids offer skin protection from environmental stressors, help with cell inflammation, and contribute to elasticity. Their natural origin and safety make them invitational candidates for creams, lotions, and serums in the expanding “green cosmetics” market (Dussably et al., 2022)

Agri-food and animal feed

In the agrifood industry, *D. salina* provides carotenoids and essential fatty acids to add nutritional value. It can be used as a natural pigment in aquaculture to improve the color of shrimp and salmon while adding value to the products. When added to livestock feeds, the carotenoids in *D. salina* will support animal health and animal productivity due to their immunostimulant and antioxidant effects (Sánchez-Martínez et al., 2022).

Energy prospects

Due to its lipid rich biomass, *D. salina* is being assessed as a potential biodiesel and biofuel feedstock. The hypersaline cultivation methods would negate competition with arable land and freshwater resources, and the post-extraction carriers from carotenoid extraction could be a post-extraction bioenergy feedstock. Samples on a larger scale may not yet have converted technologies available but developments in bioprocessing and reasoned macroalgal techno-economic factors could make this bioprocess more economically sustainable in the future (Singh et al., 2024).

BIOTECHNOLOGICAL ADVANCES AND PROSPECTS FOR *DUNALIELLA SALINA*

D. salina a halophilic microalga within the taxonomic group Chlorophyta, is characterized by metabolic flexibility, tolerance to stress, and ability to produce high-value biomolecules, all of which are valuable traits for developing multipurpose biotechnological platforms. Importantly, recent insights from new omics technologies, heightened metabolic engineering, and better cultivation methods are further establishing its capacity for exploitation in industrial, pharmaceutical, and ecological applications (Figure 1) (Castellanos-Huerta et al., 2022).

Integration of omics data and functional engineering

Genomic and transcriptomic studies have identified genes that encode for the components of pathways involved in β-carotene biosynthesis, osmotic stress response, and the regulation of primary and secondary metabolism. Furthermore, comparative studies conducted under different environmental conditions suggest the coordinated activation of pathways involved in photoprotection, carotenoid biosynthesis, and osmotic regulation. Knowledge of these pathways provides the basis for targeted engineering to reinforce metabolic structure during conditions of stress to maintain cell viability (Chen et al., 2024).

Genetic transformation tools and molecular selection

In the past, genetic transformation of *D. salina* has shown their complexity with respect to the lack of a rigid cell wall and variable transgene expression. However, methods for inducing transformation, e.g., electroporation, biolistics, glass bead transfer, Agrobacterium-mediated infiltration and transient expression of heterologous proteins are well-supported by experiments using viral and other natural promoters, such as CaMV35S, actin, GAPDH and regulatory elements like MARs (Quezada et al., 2024). Functionally novel selection markers, such as zeocin resistance and phosphinothricin, are additional tools available for this alga (Castellanos-Huerta et al., 2022).

Metabolic engineering for the optimization of biosynthetic pathways

Metabolic engineering is focused on rerouting carbon flux toward the increased accumulation of β-carotene, lipids, and recombinant proteins. With regard to above-ground-count methods, work is devoted to overexpressing the crucial enzymes in β-carotene synthesis, i.e., phytoene synthase, lycopene cyclase, etc., through chloroplast-targeted transformation through homologous recombination, as nuclear transformation has limitations (Saini et al., 2020). As previously stated, stress condition are known to drive accumulation of metabolites (Section 3). Moreover, using these physiological responses, with genetic engineering, aims to maximal yield.

Advanced cultivation technologies and smart bioreactors

Advances in next-generation photobioreactors (PBRs) with real-time sensors, automated control, and AI-based models allow for precise adjustments to environmental parameters (light, pH, temperature, CO_2) (Shiri et al., 2023). The addition of automation and AI improves reproducibility, optimizes biomolecule production, and allows for continuous extraction and valorisation of co-products, creating complete models for integrated, sustainable industrial uses in biofuels, cosmetics, and nutraceuticals (Chang et al., 2017).

New applications: vaccine biofactories and mucosal administration

In addition to metabolites, *D. salina* is also developing into a potential candidate for recombinant protein production, specifically with a focus on vaccines. The advantages of it being recognized as GRAS (Generally Recognized As Safe), as well as its natural ability to encapsulate antigens, and human-compatible glycosylation pathways, makes *D. salina* a unique opportunity for oral or nasal delivery of vaccines. Prior investigation has identified and demonstrated the expression and stability of viral antigens (e.g., HBsAg, VP28) in edible algae, where edible algae have significant potential as bioencapsulated subunit vaccines, lowering the purification requirement, improving storage stability, and eliciting a mucosal immune response (Castellanos-Huerta et al., 2022).

Future prospects and challenges

Although notable progress has been made, obstacles still exist such as low transformation efficiency, low yields of recombinant proteins, and the unclear regulatory expectations of GMOs. Future research priorities include the integration of multi-omics research, the design of improved expression vectors, and chloroplast-specific engineering. Overcoming these challenges could allow *D. salina* to become a viable, competitive platform for the circular bioeconomy contributing as (1) a platform for high-value metabolites, (2) a host for biopharmaceuticals, and (3) an excellent culture model for sustainable and secure biotechnologies.

D. salina represents a strategic resource for the bioeconomy due to its ability to transform marginal environments (salt lakes, salt marshes, hypersaline effluents) into biomass rich in high value-added molecules (β-carotene, essential fatty acids, proteins). This transformation of areas unsuitable for agriculture into sustainable industrial production sites illustrates its direct role in diversifying raw material sources. In addition, the integrated use of its co-products (pigments, lipids, proteins) in the agri-food, cosmetics, pharmaceutical, and bioenergy sectors helps to fuel local value chains and reduce dependence on fossil resources. Thus, *D. salina* contributes not only to the sustainable production of biomolecules, but also to the transition to a circular bioeconomy by combining bioremediation, salt waste recovery, and the production of bioproducts with a low environmental footprint (Yildirim et al., 2022).

MECHANISMS OF ACTION AND HEALTH BENEFITS ILLUSTRATED

Figure 2 shows the main bioactive products of *D. salina*, β-carotene, polyunsaturated fatty acids (PUFAs), polysaccharides, proteins, antioxidants, and minerals, as well as their biological functions. β-carotene is a powerful antioxidant that regulates the immune system by enhancing the IgG and IgM, and cytokine levels, and helps maintain immune homeostasis while reducing chronic inflammation (El-Baz et al., 2022). PUFAs and other lipids show antimicrobial properties by inhibiting pathogenic bacteria, and polysaccharides are shown to have hypolipidemic activity by lowering LDL cholesterol, while elevating HDL cholesterol (Alghamdi

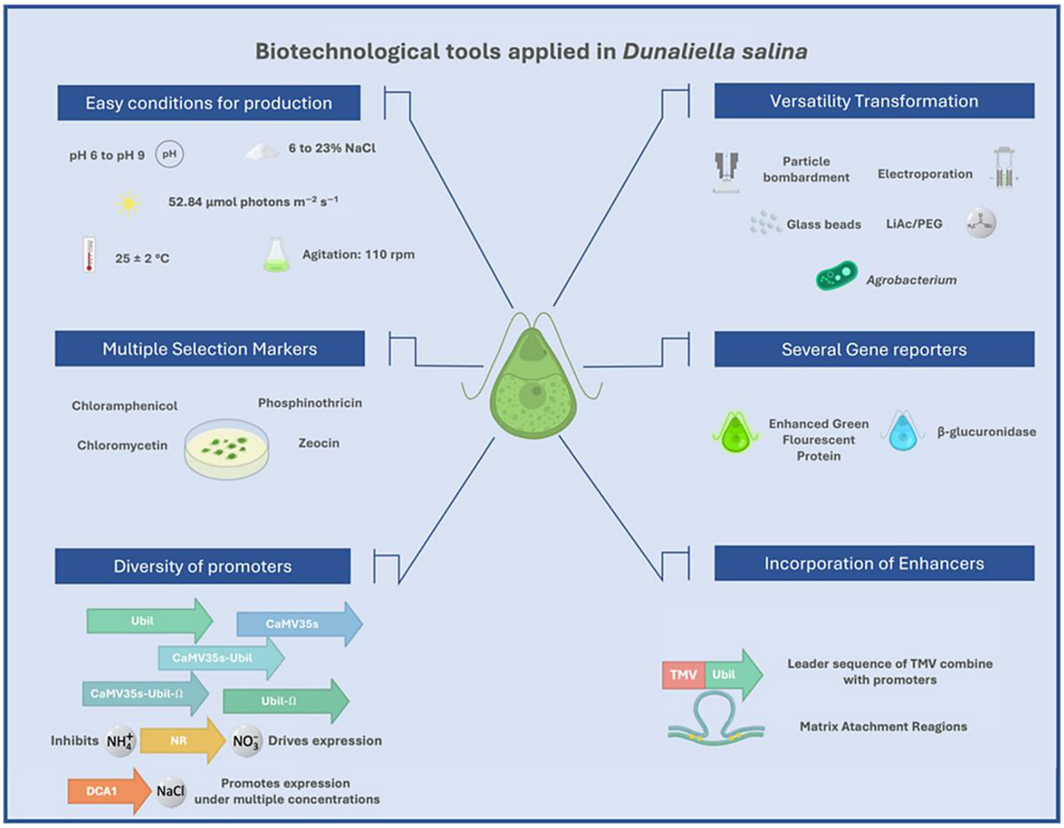

Figure 1. Biotechnological applications using *D. salina* (Castellanos-Huerta et al., 2022)

et al., 2024). The proteins found in *D. salina* have been suggested to enhance gut health by improving the villus structure and shifting the microbiota in the intestine to improve nutrient absorption and gastrointestinal functioning (Fernandes et al., 2020). The antioxidant capacity, which comes from carotenoids and other bioactive metabolites, prevent the interaction of free radicals in the body, thus decreasing oxidative stress and reducing cellular damage (Roy et al., 2021). The minerals in the microalgae would also affect metabolic and physiological functions that are necessary for the health of animals and humans.

These mechanisms are grouped together to allow for a variety of pragmatic functions. *D. salina* can be used in functional foods, nutraceuticals, cosmetics, and pharmaceuticals. In the context of animal nutrition, *D. salina* has shown improvement in growth performance, feed conversion ratio (FCR), egg production, and yolk carotenoid content in poultry. The diverse health benefits provided by *D. salina* expand its opportunity for a multi-functional bioactive supplement in human and animal nutrition.

BIOREMEDIATION POTENTIAL AND ENVIRONMENTAL APPLICATIONS

In bioremediation operations, the use of the microalga *D. salina* provides a viable, sustainable, and cost-effective alternative to traditional wastewater treatment and ecological recovery technologies (Table 3) (El-Sheekh et al., 2025). This microalga is finding increasingly broader recognition as an ecological 'agent' with the ability to manage industrial, agricultural, and urban waste because of its extreme-conditions tolerance and physicochemical properties for absorbing a diverse array of pollutants. *D. salina* and others can also effectively remove heavy metals, excessive nutrients, and dangerous organophosphate toxins. As a result, the valorization of saline wastewater is also emphasized in this matter of bioremediation microalgae, e.g., particularly with *D. salina* in circular bioeconomy approaches (Diankristanti and Ng, 2024).

Heavy metal removal and industrial effluent treatment

D. salina has shown great potential to withstand and absorb multiple pollutants, *positioning* it as an advantageous organism for bioremediation, particularly in hypersaline environments or industrial effluents that will not be helped by typical remediate systems. These microalgae grow in extreme salt environments and are capable of removing heavy metals (cadmium (Cd), lead (Pb), chromium (Cr)) as well as excess nutrients (NO_3^-, PO_4^{3-}) that lead to eutrophication.

Numerous studies have confirmed the potential of strains of Dunaliella, especially *D. salina*

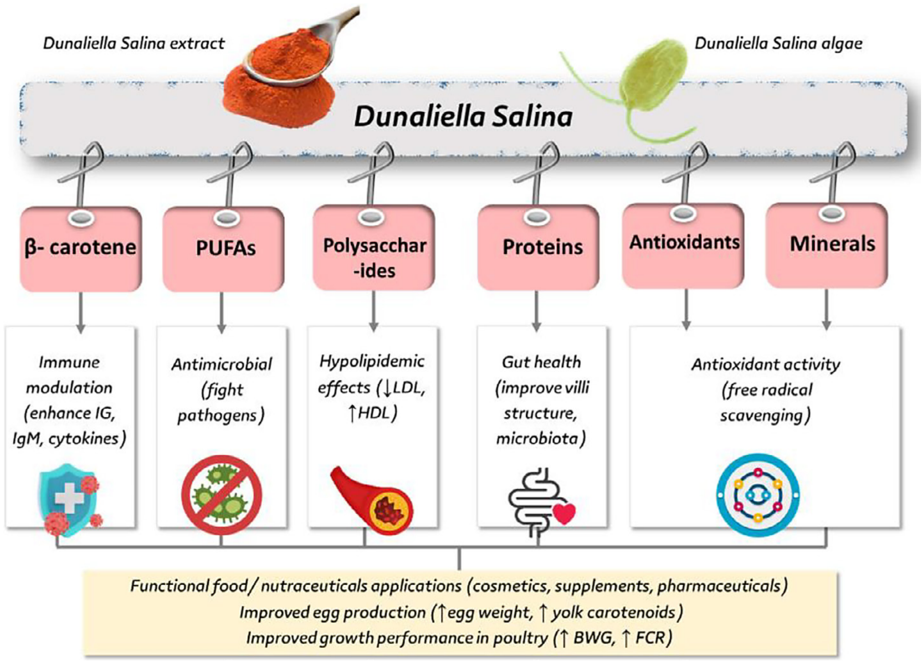

Figure 2. Biological mechanisms and nutritional benefits of *Dunaliella salina* (Alagawany et al., 2025)

and *D. tertiolecta*, to remove heavy metals such as chromium, copper, and arsenic. These microalgae produce metal-binding peptides like phytochelatins that allow them to biosorb and bioaccumulate metal ions (Tsuji et al., 2002). Dahmen-Ben Moussa et al. (2018) observed that Dunaliella strains could remove up to 98% of metals from a closed system after one week. Based on isolation and optimization, a strain of Dunaliella from the Sambhar salt lake (India) demonstrated considerably higher adsorption of hexavalent chromium (Cr(VI)) at conditions of pH 8.6, 10% inoculum, and for 120 hours, reaching 66.4% removal (Vidyalaxmi et al., 2019). This process has also been confirmed using FTIR, SEM-EDS and XRD.

Degradation of organophosphate pesticides

D. salina have capacity to grow in unacceptable hypersaline and nutrient-poor conditions creates possibilities to valorize marginal environments that are unsuitable for terrestrial agriculture or aquaculture. Environments like salt flats, saline deserts, and coastal lagoons – often seen as ecological wastelands – can be transformed into productive areas through proper cultivation of this microalga (Borovkov et al., 2020).

Organophosphate pesticides are widely utilized in the agricultural sector and pose a substantial risk to surface and groundwater. A more recent study by Nasiri et al. (2023) illustrated that the microalga *D. salina* is capable of degrading diazinon, chlorpyrifos, ethion, and profenofos (common organophosphate pesticides) from well water. At a concentration of 5 mg/L, *D. salina* was able to degrade anywhere from 41.84% to 70.25% of the target compounds while also producing identifiable intermediates and demonstrated good degradation kinetics (half-lives between 2.76 and 29.49 days). This allows us to consider *D. salina*'s physiological capacity for degradation of organic pollutants but also highlights its potential use in an integrated wastewater treatment strategy (Nasiri et al., 2023).

Treatment of hypersaline wastewater and circular valorization

The management of hypersaline wastewater represents an environmental issue in industries like agro-food, pharmaceuticals, or textiles. For instance, Elangovan et al. (2025) showed that *D. salina* grown in 5000 L open ponds was able to remove nutrients (NO_3^- (95.4%)); PO_4^{3-} (93.4%); SO_4^{2-} (67.6%)) and 80% COD while simultaneously producing value-added products such as β-carotene (5.2% wet biomass). Thus, the process of phycoremediation, in a waste-to-resource model, demonstrates how we can create a circular system where pollution and the production of industrial biomass can go hand-in-hand.

Nutrient recovery and agroecological applications

In addition to detoxification, *D. salina* also recovers/ recycles vital nutrients such as nitrogen (N), phosphorus (P) and sulfur (S). Sui et al. (2025) demonstrated that *D. salina* could recover close to 100% phosphorus, over 80% nitrogen and a substantial portion of sulfur from enriched media. In addition to halting nutrient-driven eutrophication within aquatic ecosystems, the recovery of these nutrients allows the conversion of recovered protein into bioavailable human nutrition containing high-quality protein biomass with essential amino acid index (EAAI) > 1.6.

CHALLENGES AND FUTURE PERSPECTIVES

Validated and documented applications

Although *D. salina* holds great promise for biotechnological and environmental applications, its large-scale cultivation still faces several well-documented technical and environmental challenges. Open culture systems such as raceways and saline ponds remain highly vulnerable to environmental fluctuations, including temperature shifts, insufficient sunlight, evaporation, and biological contamination by invasive algae, bacteria, or rotifers (Rojas-Villalta et al., 2024). Fluctuations in salinity can lead to osmotic imbalances that negatively affect growth and productivity, while evaporation can increase salt concentration in arid areas or facilitate the introduction of invasive species in humid regions (Hassani et al., 2021). Even with a strategy that leads to growth, optimal salinity is still a technical and economic obstacle (Tanoeiro et al., 2024). As closed systems, photobioreactors offer superior environmental control; but they are costly and may require energy input, which can hinder incorporation into sustainable production schemes (Harvey & Ben-Amotz, 2020). In addition to these technical limits, nutrient availability, adding carbon dioxide, and harvesting on a bulk

Table 3. Recent studies highlighting the bioremediation potential of *Dunaliella salina*

Study	Target pollutants	Removal efficiency / Results	Methodology	Reference
Dahmen-Ben Moussa et al. (2018)	Heavy metals (Cu, As)	98% in closed system, 1 week	Closed-system culture	Dahmen-Ben Moussa et al., 2018
Vidyalaxmi et al. (2019)	Hexavalent chromium (Cr(VI))	66.4% at pH 8.6, 120 h	Box-Behnken, SEM-EDS, FTIR	Vidyalaxmi et al., 2019
Nasiri et al. (2023)	Pesticides (Diazinon, Chlorpyrifos, etc.)	41.84–70.25%	GC-MS, growth, kinetics	Nasiri et al., 2023
Elangovan et al. (2025)	NO_3^-, PO_4^{3-}, SO_4^{2-}, COD	95.4%, 93.4%, 67.6%, 80%	HRAP (5000 L), 45 days	Elangovan et al., 2025
Sui et al. (2025)	N, P, S	>80–100% recovered	N/P/S ratio testing, EAAI	Sui et al., 2025

scale imply significant research and automation. Furthermore, genetic variability among *D. salina* strains plays an important role in biomass yield and carotenoid accumulation. It will be critical to develop resilient *D. salina* strains that produce high yields in variable environmental conditions (Papapostolou et al., 2023).

Prospective and emerging applications

Beyond established barriers, much of the potential research for *D. salina* is still speculative and requires experimental evidence (Fachet et al., 2014). The potential for optimization towards climate adaptation strategies is not fully explored. For example, D. salina's interactions with other microorganisms and microbiota within hypersaline ecosystems may enhance resilience and purification capacity (Dildar et al., 2025). Similarly, while there is some evidence that *D. salina* may be contributing to carbon sequestration, the role of *D. salina* in carbon sequestration has not yet been quantified fully through different biogeographic contexts thus remains a potential application versus a case proven use. from an ecological perspective, *D. salina* could be integrated as part of green infrastructure projects to rehabilitate degraded lands impacted by salinization or seawater intrusions especially on the coast (Araújo et al.,2009). Integration of *D. salina* might reduce erosion, limit evaporative water loss, and maintain soil fertility. In conclusion, possible future developments may rely on functional genomics, and emerging gene-editing technologies (e.g. CRISPR/Cas9) (mandated to biosafety and social acceptance) (Hu et al. 2021). The idea to integrate *D. salina* into North American hybrid agroecological systems for pollution management and/or ecosystem service provision is novel and holds much promise, but more systematic research is not available.

CONCLUSIONS

Dunaliella salina stands out as an ecological and biotechnological model capable of transforming extreme saline environments into productive bioresource systems. The synthesis of available data clearly demonstrates how abiotic stressors, particularly salinity, light, and nitrogen limitation – drive the metabolic reprogramming that leads to high β-carotene and lipid accumulation. These mechanisms, combined with the species' tolerance to pollutants and ability to recover nutrients, make *D. salina* an effective agent for bioremediation and a cornerstone for sustainable biorefineries. Recent advances in genomics, metabolic engineering, and automated photobioreactor technologies now enable targeted enhancement of these pathways. However, future work must address the optimization of large-scale processes, the selection of robust strains, and the integration of renewable energy sources to achieve cost-effective production. The originality of this study lies in bridging biological mechanisms with environmental and industrial applications, underscoring *D. salina*'s potential contribution to a circular bioeconomy and its role in climate resilience and sustainable resource management.

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