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MICROALGAL BIOFILMS: ECOLOGICAL ROLES, BIOTECHNOLOGICAL APPLICATIONS, AND FUTURE PROSPECTS

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ABSTRACT

Microalgal biofilms represent a promising alternative to suspended algal cultures, offering advantages in biomass productivity, stability, and ease of harvesting. These biofilms consist of algae, bacteria, and other microorganisms embedded in extracellular polymeric substances (EPS), which provide structural support and facilitate complex ecological interactions. Algae also plays a crucial role in ecosystem as they are present naturally associated with heterotrophic bacteria and gives an interacting relationship. Algae supply fixed carbon, while bacteria contribute vitamins, siderophores, and growth-promoting factors, creating mutualistic networks that enhance growth and resilience. Such synergistic interactions not only improve nutrient utilization but also support biofilm applications in wastewater treatment, CO_2 capture, and pollutant degradation. Additionally, microalgal biofilms are valuable platforms for the production of high-value bioproducts, including pigments, polyunsaturated fatty acids, and polysaccharides, and can be engineered into biosensors for environmental monitoring. Recent studies highlight the potential of biofilms as cost-effective systems for industrial bioprocessing, though challenges remain in scale-up, axenic culture, and biofilm heterogeneity. Advances in bioreactor design, strain modification, and EPS manipulation are expected to optimize biofilm performance and broaden their applications. This review summarizes the ecological basis, structural features, and biotechnological potential of microalgal biofilms, emphasizing their role as sustainable systems for environmental and industrial biotechnology.

KEYWORDS: Microalgal biofilms; Algae–bacteria interactions; Extracellular polymeric substances (EPS); CO_2 capture; Biosensors; High-value bioproducts; Bioprocessing.

INTRODUCTION

Algae are a diverse group of photosynthetic organisms that use autotrophic, mixotrophic, or heterotrophic metabolic methods to produce oxygen. Since the 1950s, many microalgae species have been exploited to produce useful bioactives (Goodwin and Jamikorn, 1954). The need for environmental preservation and renewable energy substitution has prompted importance in microalgae research in recent decades (Mata et al., 2010; Wang et al., 2017). Natural photoautotrophic conditions for biomass production with CO_2 supplementation have been implemented in the last decade (Gupta et al., 2016), and much research is being done to improve cultivation of microalgae in suspension, both through increasing biomass concentration and through improved process and bioreactor designs, particularly more efficient biomass-recovery technologies (Acien et al., 2012; Berner et al., 2016). Aside from that, there's a lot of interest in looking into a completely new approach: the potential of culture of microalgae as biofilms (Irving and Allen, 2011). Microalgal biofilm culture is still in its early stages of development. In comparison to suspended microalgae cultivation, the experimental results are favourable (Table 1). Bacteria and algae have also been to combat various organic pollutants such as azo compounds, dibenzofuran, black oil, and acetonitrile. Since the interaction between algae and bacteria is based on the development of biofilms between algae and bacteria, it is natural that it has been widely used in the treatment of nutrient rich waste water including sewage and waste water treatment. Biofilm formation is a significant virulence mechanism in the pathogenesis of many medically important bacterial pathogens, such as *Pseudomonas aeruginosa* (Gellatly and Hancock, 2013), *Staphylococcus aureus* (Gordon and Lowy, 2008), and *Escherichia coli* (Beloin et al., 2008).

Biofilm formation is not only relevant to algae but is also a survival strategy of many pathogenic bacteria, highlighting its ecological and medical importance. Understanding bacterial biofilm strategies also provides insights into algal–bacterial biofilms, as both rely on EPS-mediated adhesion and community-level interaction.

1. Physiology of algal-bacterial interaction

The physiology of interaction of algae and bacteria cannot be understood properly individually. They influence the ecosystem together by giving mutualistic to parasitic relationship. Although bacteria are considered as contamination of algae cultures, it was studied that bacteria affect algal growth and physiology both synergistically and antagonistically. Nowadays, algae-bacteria interactions are known as promising in biotechnology, as some recent studies have shown a synergetic effect of algae-bacteria interaction on algal growth and flocculation processes, which have emerging application in algal biotechnology (Ramanan et al., 2015). One of the main roles of the mutualistic interaction between algae and bacteria is the exchange of fixed carbon by algae and vitamins produced by the bacteria, which act as cofactors for enzymes in key metabolic pathways in the alga.

2. Biofilm

An immobilised cell, according to Tampion and Tampion (1987), is a live cell that is stopped from freely moving from its original position to all areas of an aqueous phase of a system by natural or artificial means. The word "biofilm," on the other hand, refers to both organisms that grow adhering to a surface and organisms that grow as aggregates, with the cells held together by extracellular polymeric substances (EPS) (Strieth et al., 2018). In response to physiological challenges in the natural environment, algae and other microbes create these compounds (Roman et al., 2008). The function of EPS is to protect the cell from the environment and to bind the cell to a surface. EPS are composed of polysaccharides, proteins, nucleic acids, and phospholipids (Sheng et al., 2010).

2.1.Algal Biofilms

Microalgal biofilms, which are made up of cyanobacteria and/or green microalgae, can be found in practically every photic aquatic environment. Biofilms have the property of creating and being functionally controlled by gradients of energy sources and chemical products (Bernstein et al., 2014). The requirement to identify two main sub-types of "algal biofilm," as previously established, is a determining point involved in connected cultivation (Wang et al., 2017). The first type is a biofilm made up of only one species of microalga (axenic culture). It can also begin with axenic microalgal biomass, but as the biofilm grows, allochthonous bacteria, fungus, and cyanobacteria settle inside, resulting in a complex microbial community (Wang et al., 2017).Therefore, Mantzorou and Ververidis (2019) defined that a biofilm could be characterized as a consortium of microorganisms, embedded in EPS, forming a complex structure which is developed on solid surfaces.

2.2.Bacterial Biofilms

Bacterial biofilms are structured microbial communities in which cells adhere to surfaces and are encased within a self-produced extracellular polymeric substance (EPS) matrix. This organization provides bacteria with enhanced resistance to environmental stressors, antimicrobial agents, and host immune responses compared to their planktonic counterparts. Biofilm development occurs through distinct stages, including initial adhesion, EPS secretion, microcolony formation, maturation, and eventual dispersal of cells to colonize new surfaces. Within biofilms, bacteria engage in quorum sensing, facilitating coordinated behavior and horizontal gene transfer, which further contributes to adaptability and persistence. While bacterial biofilms are often studied in the context of pathogenicity and medical device infections, they also play critical roles in natural ecosystems and engineered systems. In wastewater treatment and bioremediation, bacterial biofilms are exploited for their ability to degrade organic pollutants and cycle nutrients. Understanding bacterial biofilm physiology and dynamics is therefore essential not only for combating biofilm-associated infections but also for harnessing their potential in industrial and environmental biotechnology.

2.3. Algal–Bacterial associated Biofilms

Algal–bacterial biofilms represent a synergistic form of microbial consortia where algae and bacteria coexist within a matrix of extracellular polymeric substances (EPS). In these systems, algae contribute organic carbon and oxygen through photosynthesis, while bacteria supply essential metabolites such as vitamin B_{12}, siderophores, and phytohormones that stimulate algal growth. This mutualistic exchange enhances nutrient cycling, promotes resilience against environmental stress, and accelerates biofilm development. Furthermore, bacteria assist in biofilm structural stability by contributing to EPS composition, whereas algae provide photosynthetically fixed carbon that sustains bacterial populations. Such interactions are particularly advantageous in applied settings: for example, algal–bacterial biofilms demonstrate superior performance in wastewater treatment, as bacteria degrade organic pollutants while algae assimilate nutrients and sequester CO_2. These dual contributions highlight algal–bacterial biofilms as efficient, low-cost, and robust systems for environmental biotechnology and industrial applications (Fig.1).

2.4. Formation of Biofilm

Biofilms are three-dimensional colonies of microorganisms that adhere to a surface and are wrapped in an exopolymeric substance that protects them. The production of a biofilm occurs in five stages. Individual planktonic cells migrate and adhere to a surface in stage one. These adherent cells commence biofilm development on the surface and become enclosed in modest amounts of exopolymeric material if the right conditions are available. Cell aggregation and matrix formation occur in stage two, when adherent cells secrete an extracellular polymeric substance (EPS) and become irrevocably attached to the surface. The biofilm matures in stage three by forming microcolonies and water channel architecture, as well as becoming much more layered. The fully grown biofilm reaches its maximal cell density in stage four and is now called complete (Fig.2).

2.5.The Extracellular Polymeric Substance (EPS)

The extracellular matrix (EPS) that surrounds the cells in a biofilm is made up of a complex blend of proteins, lipids, nucleic acids (extracellular-DNA), and polysaccharides (Annous et al., 2009). These components not only help the biofilm stick to the surface, but they also trap nutrients, offer structural support, and protect the biofilm against host immune responses and antimicrobial therapies (Flemming et al., 2007). In addition to the tasks listed above, the EPS is responsible for keeping the community of biofilm cells close together, allowing cell-to-cell communication (quorum sensing), and facilitating the interchange of genetic material via horizontal gene transfer (Hausner and Wuertz, 1999).

3. Applications of Biofilm

Microalgae biofilms have potential applications in toxicity measurements (biosensors) (Brayner et al., 2011; Lam and Lee, 2012; Nandimandalam and Gude, 2019), CO2 capture (Hamano et al., 2017), and polycyclic aromatic hydrocarbons accumulation and degradation (Brayner et al., 2011; Lam and Lee, 2012; Nandimandalam and Gude, 2019). (Zhang et al., 2019).

Biofilms have primarily been used in industrial applications, such as wastewater treatment, where no support material is required and cells generate biomass granules and flocs that grow in size over time (Moreno-Garrido, 2008).

Biofilms of a single species are utilised to make industrially important compounds (Abdel-Hameed and Hammouda, 2007). Controlling and maximising the production of desired target products is preferred in chemical production (Qureshi et al., 2005), a scenario that can be achieved by inoculating a single species into a sterile environment and allowing it to form a biofilm before being used to produce a specific product (Zhang et al., 2015; Cheng et al., 2017).

Production of high added value molecules by microalgae allows that forms of valorization, i.e., health food and quality feed, carbohydrates, polyunsaturated fatty acids, pigments (Dixon and Wilken, 2018), are currently penetrating their markets and reach economic competitiveness (Levasseur et al., 2020). Beyond its rich macronutrient composition, microalgae are able accumulate and to express secondary metabolites under stressful conditions (Mukherjee et al., 2015).

Despite the importance of microalgal biofilms in nature, their potential as biological indicators in wastewater systems, and the detrimental role they can play in fouling of surfaces, further research is still needed to fully understand the factors influencing microalgae biofilm development (Burns and Ryder, 2001; Ljaljevic-Grbic et al., 2010; Nováková and Neustupa, 2015; Novoa et al., 2021). The application and limitation are described in Table 2.

Table 1. Comparison of Suspended vs. Biofilm-Based Algal Cultivation.

Parameter	**Suspended Cultivation**	**Biofilm-Based Cultivation**	**Applications**	**Limitations**
Biomass productivity	Moderate, depends on mixing & light	High surface area utilization, dense biomass	Biofuels, pigments, general biomass	Dilute cultures, high water requirement
Harvesting cost	High (centrifugation, flocculation required)	Lower (easy scraping, immobilized cells)	Wastewater treatment, biosensors, high-value molecules	Scale-up challenges, detachment issues
Contamination risk	Higher (open ponds)	Lower (surface-attached communities)	Controlled production of metabolites	Biofilm heterogeneity

Table 2. Applications of Microalgal Biofilms with Literature Support.

Application Area	Description / Role	References
Wastewater treatment	Biofilms immobilize microalgae for efficient nutrient (N, P) removal and pollutant degradation.	de-Bashan & Bashan, 2010; Mallick, 2002
CO_2 capture	Biofilms enhance CO_2 sequestration due to dense biomass and EPS protection.	Hamano et al., 2017
Biosensors (toxicity tests)	Biofilms can be engineered as biosensors to detect toxic compounds and environmental pollutants.	Brayner et al., 2011; Lam & Lee, 2012; Nandimandalam & Gude, 2019
Bioproducts (pigments, PUFA, polysaccharides)	Stress-induced biofilms accumulate high-value molecules with nutraceutical potential.	Dixon & Wilken, 2018; Levasseur et al., 2020
Polycyclic aromatic hydrocarbons degradation	Microalgal biofilms degrade hydrocarbons, showing promise for bioremediation.	Zhang et al., 2019
Industrial bioprocessing	Single-species biofilms can be harnessed for controlled production of specific metabolites.	Qureshi et al., 2005; Zhang et al., 2015; Cheng et al., 2017

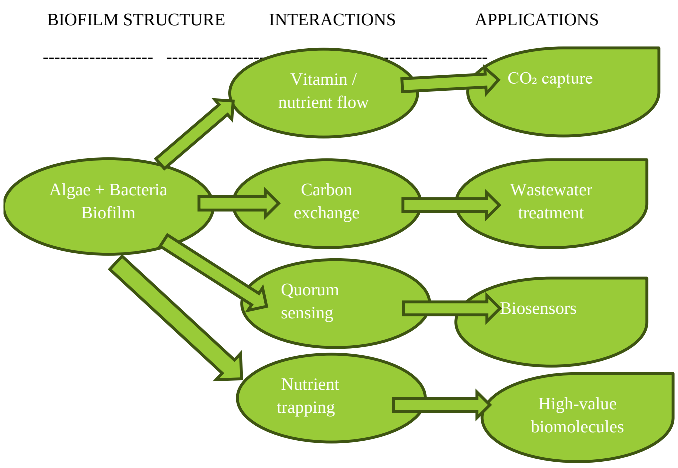

Fig.1. Schematic diagram of algal-bacterial biofilm and their application.

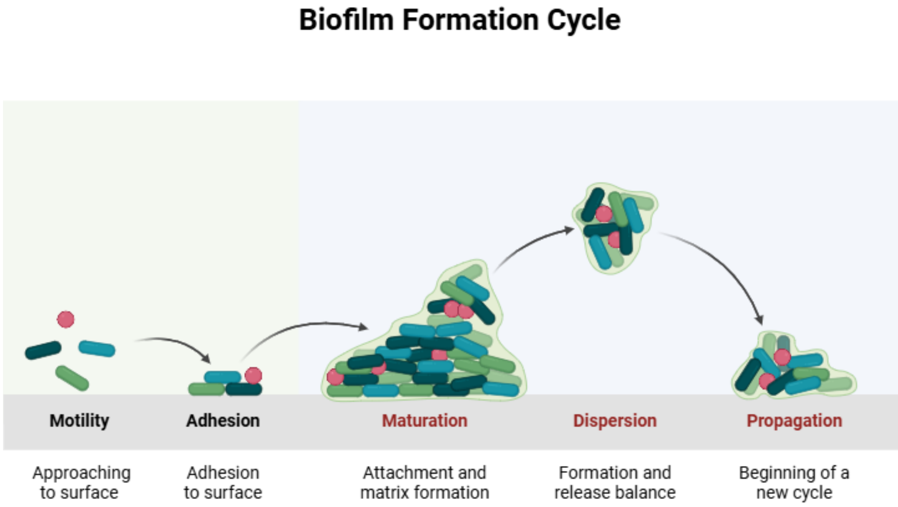

Fig.2. Stages of Biofilm formation.

Critical Research gap

While microalgal biofilms exhibit considerable ecological and biotechnological promise, various challenges and limitations need to be overcome before their broad application can be achieved. A primary limitation is the heterogeneity of biofilms, as structural and metabolic differences within the matrix result in uneven productivity and inconsistent yields of metabolites. Furthermore, issues related to biofilm detachment and stability present risks for continuous bioprocessing, often leading to biomass loss and diminished efficiency in large-scale operations. Another issue pertains to the scaling up of biofilm-based cultivation. Although promising results have been reported in laboratory and pilot-scale studies, transitioning these systems to industrial scales poses challenges due to reactor design complexities, mass transfer constraints, and the need to maintain consistent light penetration through dense biofilms. The economic viability of biofilm cultivation compared to suspension systems also necessitates thorough assessment, especially regarding biomass harvesting, nutrient recycling, and long-term upkeep. The algal-bacterial interactions that facilitate biofilm formation are not yet fully understood at the molecular level. While the mutualistic exchange of vitamins, siderophores, and fixed carbon is well-established, there is a significant gap in mechanistic understanding concerning quorum sensing, EPS regulation, and cross-kingdom signaling. In the absence of this knowledge, attempts to engineer stable, high-performing consortia are limited. Additionally, current applications in wastewater treatment, CO_2 capture, and biosensors have shown proof-of-concept success but encounter technical challenges related to scalability, robustness under varying environmental conditions, and regulatory approval for use in sensitive settings. Likewise, the commercialization of high-value products derived from biofilms is impeded by low yields, inconsistent quality, and a lack of downstream processing strategies specifically designed for immobilized systems. Future progress will rely on integrating omics approaches (genomics, transcriptomics, metabolomics) to better characterize biofilm communities, genetic engineering of both algal and bacterial partners to enhance metabolite exchange, and innovations in bioreactor design to improve light distribution, nutrient delivery, and harvesting. Addressing these bottlenecks is essential to unlock the full industrial potential of microalgal biofilms.

CONCLUSION

Microalgal biofilms represent a dynamic and sustainable platform that integrates ecological complexity with biotechnological utility. Their unique structure, consisting of algae, bacteria, and other microorganisms embedded in extracellular polymeric substances (EPS), provides resilience and functional versatility compared to suspended cultures. These biofilms enable efficient nutrient recycling, enhanced biomass production, and simplified harvesting, making them particularly attractive for applications such as wastewater treatment, CO_2 sequestration, biosensing, and the production of high-value metabolites. Despite their promise, challenges including biofilm detachment, heterogeneity, and scalability must be addressed to fully realize their industrial potential. Advances in bioreactor engineering, genetic and metabolic optimization, and controlled manipulation of microbial consortia are opening new avenues for improving biofilm stability and productivity. By bridging ecological interactions and applied biotechnology, microalgal biofilms offer a sustainable pathway toward environmental remediation, renewable energy production, and the development of novel bio-based products.

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CONFLICT OF INTEREST

There is no Conflict of interest

Declaration of generative AI and AI-assisted technologies in the writing process

During the preparation of this manuscript the authors used ChatGPT in order to improve language and readability. After using this tool, the authors reviewed and edited the content as needed and take full responsibility for the content of the publication.

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