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MORALITY BEFORE KNOWLEDGE

Biofilm's double-edged resilience: A critical review of the environmental and clinical paradox in the one health nexus

Muhammad Awaluddin[1,*], Ishmah Sumayyah[2]
[1] *Department of Aquaculture, National Taiwan Ocean University, Keelung City, Taiwan 202, China;*
[2] *Department of Environmental Engineering, Institut Teknologi Bandung, Bandung City, West Java 40132, Indonesia.*
*Correspondence: bussinessuddin@gmail.com

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ABSTRACT
Background: The microbial biofilm, a matrix-encased community, represents a foundational biological entity mediating critical outcomes across both environmental and human health sectors. The purpose of this comprehensive review is to synthesize the disparate literature on biofilm dynamics and its dual impact, thereby establishing the necessary framework for a unified research strategy. A vast body of work confirms the biofilm's role in nutrient cycling and protection, but simultaneously highlights its involvement in major global challenges, including infectious disease and ecological stability loss. **Methods**: This review synthesizes extant literature from environmental microbiology, clinical medicine, and biogeochemistry to construct a cohesive theoretical model. The core theoretical component introduced is the "biofilm health" analogy, which compares a biofilm community to an ecosystem where functional diversity dictates resilience and resistance to invasion. **Findings:** The core finding is the biological paradox inherent in the biofilm's structure: the same resilience mechanism (the Extracellular Polymeric Substance, or EPS) that drives positive ecological processes like bioremediation simultaneously accelerates the global crisis of Antimicrobial Resistance (AMR) in clinical settings, and facilitates the persistence of environmental pathogens and destructive biocorrosion. This paradox mandates a holistic research paradigm shift. **Conclusion**: Addressing this paradox necessitates a unified One Health approach, compelling researchers to develop non-antimicrobial disruption strategies targeting the EPS matrix. **Novelty/Originality of this article:** This work introduces the "biofilm health" analogy as a transdisciplinary conceptual tool and provides the first comprehensive synthesis detailing how the biofilm structure itself acts as the central bottleneck linking ecological stability and clinical health outcomes.

KEYWORDS: antimicrobial resistance; biofilm; ecology; environment; monitoring.

1. Introduction

Biofilms are microorganisms (mostly dominated by bacteria) that develop on a variety of surfaces in various circumstances either in terrestrial or aquatic surface, naturally or man-made. They form when single-celled microbes cling to a surface and secrete a substance known as extracellular polymeric substance (EPS), creating a complex three-dimensional framework that supports various species. Bacteria typically create biofilms as a way of survival when faced with environmental obstacles such as UV radiation, insufficient nutrition, extreme pH levels, high salinity, severe temperatures, increased pressure, and

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exposure to antimicrobial chemicals (Omwenga & Awuor, 2024). The biofilm is a very different life form than planktonic microorganisms: the gel-like matrix allows for the retention of water and nutrients while also protecting its inhabitants from physical, chemical, or biological aggressions and changing environmental conditions (Flemming et al., 2016). It is distinguished by alterations in the irreversible adherence of microbial cells to surfaces, substrates, or each other, encased in EPS, and displaying unique phenotypes in terms of gene transcription and growth rates.

A bacterial biofilm is made up of one or more microorganisms, such as bacteria, fungi, archaea, protozoa, and yeasts, and it features a channel structure that regulates the flow of gases, nutrients, and antimicrobials (Zhao et al., 2023). It is widely recognized that microorganisms inside biofilms, such as prokaryotes and eukaryotes, or unicellular and multicellular creatures, have a symbiotic connection. These symbiotic interactions provide reciprocal benefits. Autotrophic organisms, such as photosynthetic bacteria and algae, produce their own food (organic carbon), while heterotrophs depend on the excess organic matter generated by autotrophs (Bolan et al., 2020). However, in certain conditions, the proliferation of these symbiotic microbes can become uncontrolled, resulting in the production of detrimental biofilm (Sharma et al., 2023).

Almost any bacterium may create biofilms. Biofilms offer a unique opportunity for transdisciplinary and integrative research, particularly in biomonitoring agents and for health-related fields (Grari et al., 2025). According to Muhammad et al. (2020) and Vani et al. (2023), a healthy ecosystem is one that is productive, robust to external stresses, and capable of preserving its structure and human services. In recent decades, integrated health approaches such as One Health and EcoHealth have emerged, recognizing that ecosystem health is strongly influenced by socio-ecological factors, including sustainability and global change (Espunyes et al., 2021).

Biofilms offer several advantages. Biofilms have several useful uses, including their use in microbial fuel cells and the protection of higher species against unwanted microbes. One of the difficulties in future study will be to improve this technology and create strategies for promoting the establishment and durability of desired biofilms (Robertson & Mclean, 2015). Bacterial biofilms have become a significant contributor to global health issues as a result of antibiotic resistance, the host's immunological defense system, and other environmental factors. Biofilms are often found on the surfaces of medical tools and bodily tissue, as well as in industry, food processing facilities, and natural habitats (Schulze et al., 2021). In the food industry, according to Seixas et al. (2016), the positive applications of biofilms are due to their strength, flexibility, water solubility, and permeability to water vapor. In agriculture, biofilms improve plant health and increase agricultural yield. In food processing, they play an important role in wastewater treatment, reducing surplus sludge and increasing water quality. Furthermore, biofilms have been used to make edible films and biodegradable packaging, promoting ecologically friendly packaging options (Omwenga & Awuor, 2024).

Aside from its beneficial features, biofilm resilience against antibiotics is a major factor complicating the treatment of biofilm-associated infections. The rising occurrence of severe biofilm infections, as well as their resistance to antimicrobial therapies, presents significant issues not just in health, but also in agriculture and the environment. Biofilms have an important role in bacterial drug resistance, especially in chronic infections. While antibiotics have been used to manage infections for decades, bacteria have developed resistance mechanisms such as biofilm formation, which protects them from both medicines and the immune system (Banin et al., 2017). Bacterial resistance in biofilms includes both tolerance (survival without growth) and resistance (growth despite antibiotic presence). Nutrient restrictions, metabolic gradients, and adaptive stress responses all contribute to the formation of inactive cell zones within biofilms. These cells are very resistant to antibiotics because they are not impacted by medicines that target active cellular activities (Kester & Fortune, 2014). In the food industry, some pathogens such as Listeria monocytogenes are poor biofilm builders but can adhere to surfaces when they associate with stronger biofilm-forming bacteria, such as *Enterococcus faecium* and *Enterococcus* *faecalis* (da Silva-Fernandes et al., 2015). Similarly, E. coli has been demonstrated to promote biofilm formation when interacting with other bacteria present in food processing facilities, such as Burkholderia caryophylli and Ralstonia insidiosa (Galié et al., 2018).

Despite the vast body of existing work confirming this dual impact, a significant research gap remains: the literature regarding biofilm dynamics is highly disparate and traditionally isolated in fields such as environmental microbiology, clinical medicine, and biogeochemistry. Current approaches often fail to address the fundamental structural paradox linking ecological stability and clinical health outcomes. Consequently, there is a lack of a cohesive theoretical framework capable of uniting these disciplines to advance our understanding of how the EPS structure itself acts as a central bottleneck in both studies. This review highlights the worldwide relevance of biofilm research, bridges existing knowledge gaps, and identifies topics for future study to advance understanding of host and ecosystem health. Moreover, it emphasizes that healthy biofilms are vital for detecting, monitoring, and mitigating the impacts of global and climate change on these communities.

The rationale for this study lies in the urgent need for a transdisciplinary paradigm shift. In recent decades, integrated frameworks such as EcoHealth and One Health have emerged, acknowledging that ecosystem, animal, and human health are deeply interconnected and heavily influenced by socio-ecological factors and global climate change. By synthesizing existing knowledge through this One Health lens, this review introduces a novel "biofilm health" analogy conceptualizing the biofilm community as analogous to an ecosystem where functional diversity dictates resilience and resistance to invasion by either clinical pathogens or environmental destabilizing taxa. The significance of this research rests in its potential to bridge these critical knowledge gaps, highlight the worldwide relevance of biofilm monitoring, and establish a foundational framework that compels researchers to develop novel, non-antimicrobial disruption strategies targeting the EPS matrix.

2. Methods

2.1 Review design and literature search strategy

This study employed a narrative review approach to synthesize literature on microbial biofilms across environmental and clinical domains. A narrative design was selected over a systematic review because the aim was to construct an integrative, transdisciplinary conceptual framework; the "biofilm health" analogy rather than to quantitatively pool or statistically compare study outcomes. A focused literature search was conducted using the PubMed, Scopus, and Web of Science databases, supplemented by relevant academic books and book chapters where appropriate. No date restrictions were applied, allowing inclusion of both historical (foundational) and contemporary (2020–2025) literature relevant to biofilm biology, environmental microbiology, and clinical infection. Search terms included, either individually or in combination using Boolean operators (AND/OR): "biofilm-associated infections", "diagnosis", "device-related infections", "antibiotic resistance", "extracellular polymeric substance", "quorum sensing", and "bacteriophages". Additional terms related to environmental applications (e.g., "bioremediation", "microplastic biofilm", "environmental DNA", "biofouling") were incorporated to capture the ecological dimension of biofilm research.

To achieve this goal, a narrative review technique was employed in conjunction with a focused literature search across the PubMed, Scopus, and Web of Science databases, as well as relevant academic books. No date restrictions were applied, allowing the inclusion of both historical and contemporary literature. The search strategy encompassed keywords such as "biofilm-associated infections", "diagnosis", "device-related infections", "antibiotic resistance", "extracellular polymeric substance", "quorum sensing", and "bacteriophages".

2.2 Inclusion, exclusion criteria and conceptual synthesis approach

Articles were included if they: (1) were peer-reviewed original research, review articles, or academic book chapters; (2) addressed biofilm formation, structure, ecological function, or clinical/industrial impact; (3) were published in English. Articles were excluded if they were non-peer-reviewed sources, conference abstracts without full text, or tangential to the biofilm–health/environment nexus. Findings from environmental microbiology, clinical medicine, and biogeochemistry were thematically synthesized rather than statistically meta-analyzed. To integrate these disparate bodies of literature, the authors developed the "biofilm health" analogy, conceptualizing a biofilm community as analogous to an ecosystem in which functional diversity determines resilience and resistance to invasion (whether by pathogens in a clinical context or by destabilizing taxa in an environmental context). This conceptual lens guided the organization of the review's thematic sections.

3. Results and Discussion

3.1 Eps and biofilm characteristics

Biofilm production is a complicated process that involves planktonic cells adhering to a surface and producing an EPS matrix (Maurya & Raj, 2020). This matrix is a complex combination of polysaccharides, proteins, and DNA that functions as an adhesive, holding the cells together and enabling their attachment to the surface (Figure 1) (Vinod et al., 2020). Microbiota settle on surfaces with low surface tension, hydrophobicity, and microtopography (Boelee et al., 2011; Gbalou et al., 2017; Zhou et al., 2021; Mahto and Das, 2022). As the matrix builds up, the cells in the biofilm start to specialize into several subpopulations, including exopolysaccharide producers, motile cells, and cells that release enzymes and poisons (Ugya et al., 2021). Microbiota are thought to survive via forming biofilms (Saini et al., 2023).

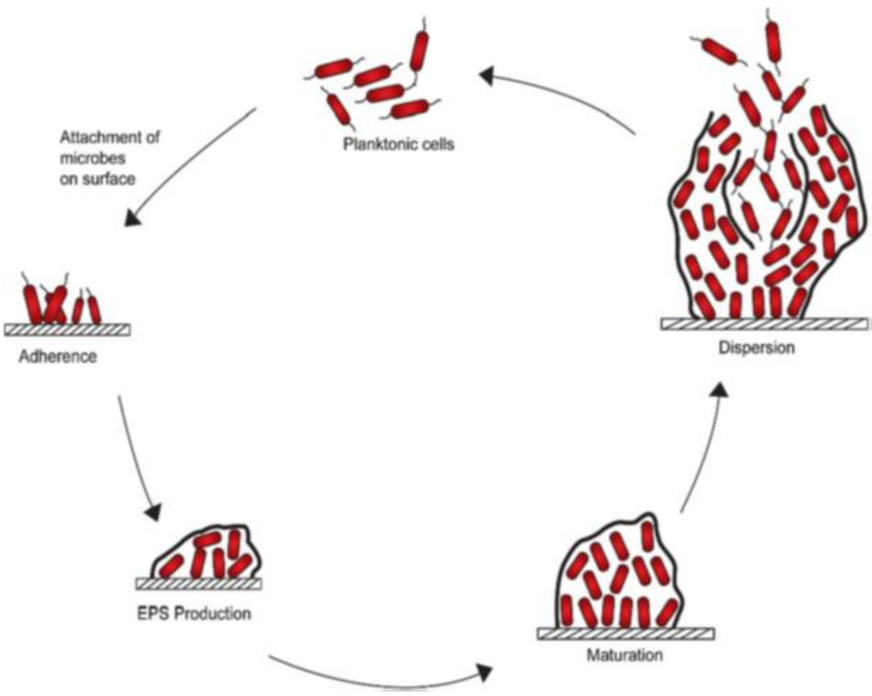

Fig. 1. Mechanism of biofilm formation
(Fani et al., 2023).

The clustering of microorganisms in the EPS matrix protects them against predators, osmotic pressure, and UV radiation (Sing et al., 2021; Mirghani et al., 2022). Biofilm subpopulations collaborate and communicate through various chemical signaling molecules, such as quorum-sensing (QS) compounds, to coordinate their activities and adapt to environmental changes (Azeem et al., 2025). *Acyl homoserine lactones* (AHLs) are a kind of QS molecule that is frequently generated by Gram-negative bacteria (Parga et al., 2023). AHLs activate genes involved in gene expression control and biofilm formation when they diffuse out of microbial cells. Once a threshold concentration is reached, bacteria can synchronize and coordinate collective activities, including biofilm development (Hartmann, 2020).

Biofilm production begins with the establishment of the conditioning layer and bacterial adherence. This process demonstrates biofilm formation, which normally occurs in five phases. The first step is cell attachment to a supporting surface. This is followed by the second stage, which involves cell-to-cell adhesion and microcolony formation. The next phase is characterized by cell proliferation and the production of extracellular polymeric substances (EPS), which drive the transition from early microcolonies to a mature biofilm structure. The mature biofilm creates a habitat that protects resident bacteria from antibiotics and other disruptive elements that enter its perimeter, and it is thought that bacteria survive by forming biofilms (Saini et al., 2023). The fifth step in the biofilm life cycle is bacterial dispersion, which is facilitated by a mature biofilm (Shineh et al., 2023). After attachment, cells reproduce, form microcolonies, and secrete exopolysaccharides (EPS), which results in permanent adherence (Bhegwat et al., 2021).

EPS promotes bacterial growth by maintaining the three-dimensional structure of biofilm. As depicted in Figure 2, EPS encloses biofilm bacteria and microorganisms, acting as a physical framework for cellular structure (Greer et al., 2021). Environmental factors such as temperature and nutritional availability promote bacterial growth through quorum sensing (QS), which is essential for biofilm development and reacts to autoinducers (extracellular signaling molecules) (Piras et al., 2021). Bacteria interact by producing and releasing autoinducers, whose concentration increases as quorum-sensing bacterial populations grow. These autoinducers enable bacteria to detect and monitor local population density (Ma et al., 2022). Once the intracellular concentration of an autoinducer exceeds a critical threshold, the bacterial population alters its gene expression and responds to its environment.

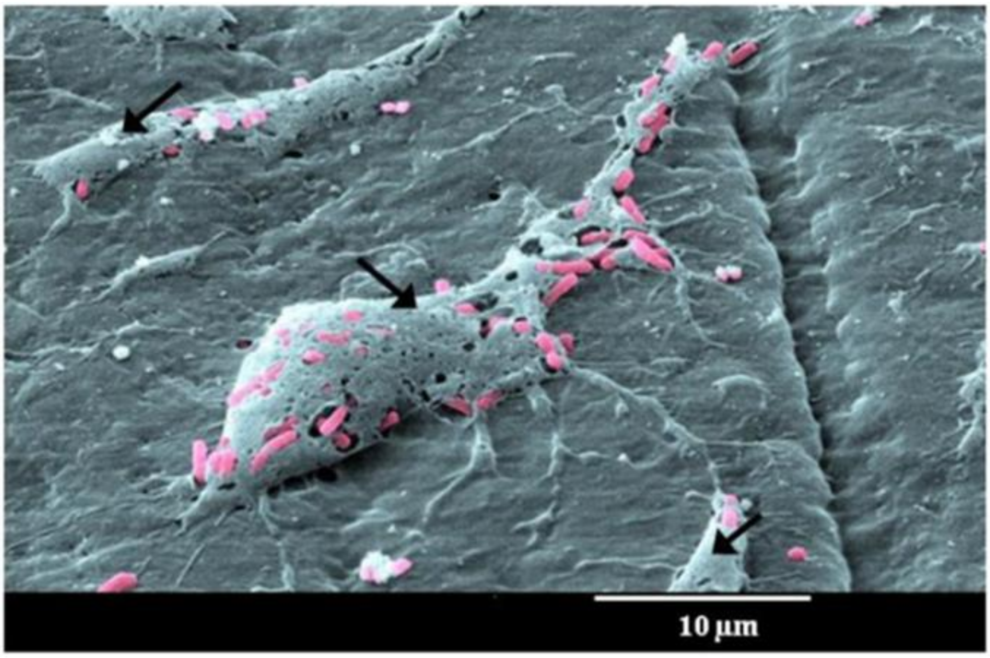

Fig. 2. Scanning electron microscopy image of *E. coli* biofilm on Teflon. The *E. coli* bacteria in the biofilm are coloured in pink, and the extracellular matrix is indicated by arrows (Shineh et al., 2023).

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3.2 Controversial takes: Between harmful presence and beneficial values

Biofilms play a crucial role in numerous biological processes and have both beneficial and harmful impacts on human health and the environment. They can develop on natural and artificial surfaces, including dental enamel, medical implants, industrial systems, and water pipelines. Their ability to withstand antimicrobial agents and evade immune defenses makes biofilm research essential across medicine, environmental science, biotechnology, and industrial operations. In addition, biofilms are increasingly used to detect heavy metal pollution in aquatic systems due to their rapid structural and physiological changes when exposed to toxic compounds. Their capacity to absorb pollutants, grow quickly, and be sampled easily makes them effective early indicators for environmental monitoring (Damyanova & Krasteva, 2025). For instance, biofilms have proven to be highly effective in wastewater treatment through bioremediation, offering a technology that is ecologically friendly and cost-effective compared to traditional chemical and physical procedures (Saini et al*.,* 2023). Within the biofilm matrix, diverse microbial species use enzymes to degrade a wide variety of pollutants, such as xenobiotics, hydrocarbons, pesticides, radionuclides, heavy metals, and explosives. These pollutants are transformed into safer end products, like carbon dioxide and water (Sarkar & Bhattacharjee, 2025). However, this approach has a relatively longer processing time compared to rapid chemical methods. Furthermore, its efficacy decreases in areas of severe pollution and when confronted with persistent, long-lasting man-made chemicals, such as plastics.

The term "productive biofilms" refers to biofilms that synthesize diverse chemicals (Philipp et al*.,* 2022). These biofilms could be in pure cultures or polymicrobial populations. Due to their structure and large number of cells, biofilms are ideal for use as continuous bioreactors (Anju et al*.,* 2022). Biofilms, like suspension culture bioreactors, require careful monitoring of key parameters, including the type of substrate (natural or artificial, which may also serve as an energy source), the flow dynamics of the aqueous phase, system volume, and the kinetics of the conducted activities. A system that allows for fluctuating process conditions can reduce the impact on cells while increasing product yield (Schmeckebier et al*.,* 2022). Liao et al*.* (2021) conducted comparative research on producing menaquinone using a suspension and biofilm reactor with *Bacillus subtilis* subsp. natto, demonstrating a better bioproduct production technique. Using the biofilm reactor resulted in a 58% production increase. Medical research focuses on microorganisms that build biofilms and produce probiotics and prebiotics. Probiotics, defined as "live bacteria that, after moderate intake, have an effective effect on host health", include *Lactobacillus, Bifidobacteria, Enterococcus, Streptococcus, Bacillus, and yeasts* (Yang et al*.,* 2024). To enhance probiotic distribution and stability, packaging should be resistant to environmental influences such as acidic gastric fluids, pancreatic juice, and bile in the small intestine, or utilize a biofilm-resistant form. Biofilm delivery of probiotics will protect bacterial cells from digestive enzymes, improve attachment, and reduce the immune system's ability to eliminate them (Zhao et al*.,* 2025).

Aside from their beneficial roles for the environment and human life, biofilms also pose a substantial public health issue. Biofilm-associated microbes develop resistance to the immune system and medications (Hussaini et al*.,* 2024), and they can be up to 1000 times more tolerant of drugs than planktonic cultures. Bacterial biofilms are known to cause up to 75% of human infectious illnesses (Vestby et al*.,* 2020). Biofilms exhibit intrinsic antimicrobial resistance, and this feature of biofilms to tolerate biocides leads to persistent contamination of environments (Liu et al., 2024). EPS are resistant to antimicrobial treatments due to chemical reactions or limited transport rates (Dutt et al*.,* 2022). This bacterial defense mechanism renders antibiotics ineffective in treating infectious diseases. Biofilm-forming bacteria play a significant role in infectious illnesses. According to Kalia et al*.* (2023), these bacteria are responsible for 65-80% of infectious illnesses, which are often chronic. Bacteria in biofilms are difficult to treat with antibiotics, which can lead to prolonged illness and high morbidity and mortality rates (Cavallo et al*.,* 2024). Biofilm-forming bacteria, including *Escherichia coli, Klebsiella pneumoniae, Pseudomonas* *aeruginosa, Staphylococcus aureus,* and *Streptococcus pneumoniae*, are responsible for 54.9% of these deaths (Vestby et al., 2020). Microbial contamination involves introducing infectious bacteria or their toxins and is a serious issue. It has the potential to cause health issues in medical and nonmedical settings. The aggregation of microbial cells results in biofilm formation, during which bacteria exhibit great resistance to environmental stressors, particularly antibiotics (Liu et al., 2024).

Biofilm development results in significant losses in industrial production and poses human health hazards every year. The warm, damp climate of the knitting and dyeing floor promotes the growth of bacteria such as *Staphylococcus aureus, Escherichia coli, and Pseudomonas sp.* (Sanders et al., 2021). Furthermore, medical biofilm infections can be classified as either device-related or non-device-related. Device-related infections include prosthetic joint infections, cardiac implantable electronic device infections, catheter-associated urinary tract infections, and ventilator-associated pneumonia (Mishra et al., 2024). Non-device-related infections include chronic wound infections, urinary tract infections, infectious kidney stones, periodontitis, dental plaque, infective endocarditis, and cystic fibrosis (Guliy et al., 2024). Microorganisms can invade human teeth, skin, and mucous membranes. Thus, biofilms can be found in the oral, skin, digestive, respiratory, and reproductive systems (Gupta et al., 2025). Furthermore, biofilms can form on pipe walls in water distribution systems, causing issues related to water quality, health, operations, and the environment. For instance, bacteria in these coatings can impact the turbidity, odor, and taste of the water (Waqas et al., 2023). Pathogenic microorganisms in drinking water distribution systems pose a substantial danger to human health. Bacterial growth can also cause corrosion in pipe materials. As a result, the release of pipe debris and impurities into drinking water would further reduce its quality (Taligrot et al., 2025). Bacteria can promote the growth of certain organisms, such as *Asellus spp.*, by forming a trophic food web. However, biofilms in the food industry can lead to economic losses and serious health hazards (Shineh et al., 2023). Bacterial biofilms can grow on food surfaces, packaging, and manufacturing equipment. Their presence on food processing surfaces can cause corrosion and produce unpleasant odors and tastes. In addition to financial losses, biofilm can harm food firms' reputations. Biofilms can harbor pathogenic and toxic bacteria, such as *Escherichia coli, Listeria monocytogenes, Campylobacter jejuni, Salmonella enterica, Pseudomonas aeruginosa, Bacillus cereus,* and *S. aureus* (Liu et al., 2023).

Furthermore, biofilms may cause biofouling due to the deposition of marine organisms on saltwater surfaces (Satasiya et al.,2025). These organisms can be found on ships, buoys, offshore facilities, submarine hulls, and factories (Chang et al., 2024). Fouling species can be divided into two types based on their size: (1) microfouling (biofilm) and (2) macrofouling (Shineh et al., 2023). Biofouling can cause further damage to maritime enterprises and infrastructure (Vishwakarma, 2020). Microbially caused corrosion results in billions of dollars in damages to various industries each year (Rao et al., 2023). Biofouling and microbial growth commonly affect businesses in the power, textile, garment manufacturing, chemical, oil and gas production, transportation, shipping, and construction industries. Textile fibers are susceptible to microbial growth due to their molecular characteristics. In textile manufacturing, biofilms can cause unpleasant odors, color fading, and fabric quality issues (Møllebjerg et al., 2021).

3.3 Biofilm and environmental circumstances

Microbial biofilms are primarily influenced by biogeographic and environmental factors, such as nutrient availability, salinity, and temperature (Oberbeckmann & Labrenz, 2020; Rüthi et al., 2020; Rai et al., 2021). These biofilms serve important functions in ecosystems and influence the life cycles of other macroorganisms, including habitat formers. Biofilms play crucial roles in marine ecosystems and have previously been considered in relation to climate change. Their ability to adsorb heavy metals, antibiotics, and other hazardous compounds enables these contaminants to spread through the aquatic food web (Llorca et al., 2020; Piccardo et al., 2021; He et al., 2022). Furthermore, biofilms can act as bioindicators and early-warning systems for both acute and chronic pollutant exposure, as reflected in functional descriptors such as photosynthetic efficiency and enzyme activity.

Biofilms have a crucial role in sustaining habitat-forming species in marine ecosystems (Dittami et al., 2021). An interdisciplinary research strategy is necessary to incorporate marine biofilm into the greater framework of microbial ecology and, more broadly, aquatic sciences. Shifts in microbial diversity and abundance can influence the chemical compounds released by biofilm communities (Fuggle et al., 2023). Changes in microbial composition have also been linked to altered functioning of propagule settlement–inducing habitat formers (Ritson-William et al., 2009; Rajitha et al., 2020). Biofilms with low biodiversity may be more vulnerable to pathogen invasion (Chen et al., 2022). In macroalgae such as Ulva species, biofilms can influence cell differentiation and division, leading to the formation of persistent holdfasts (Navarrete et al., 2022).

While shifts in microbial composition within a biofilm can indirectly influence signaling (as discussed above), external stressors such as temperature fluctuations can directly affect microbial interactions. Climate change can modify the composition of microbial biofilms and their bioactive compounds, thereby affecting larval settling and survival. Several examples include increased water temperature, reduced salinity, and pH. Exposure can also cause changes in community structure (Carafa et al., 2021). The success of habitat-former propagules, which strongly influences ecological succession, is closely tied to their interactions with microbial biofilms and the bioactive substances these communities release (Cooney et al., 2024). These molecules have unique positive, negative, or neutral associations with pelagic larvae of distinct habitat formers (Hadfield, 2011; Rajitha et al., 2020).

Increased temperatures can cause biofilms to emit chemicals that influence larval behavior and favor tropical migrant habitat formers over resident ones (Roggatz & Parsons, 2022). High temperatures can change gene expression in habitat-forming propagules, potentially disrupting gene regulation by inductive or inhibitory cues (Chui et al., 2023). Temperature and pH can affect the concentration of AHLs, which are important for bacterial signaling in biofilms. Warming may lead to increased AHL concentrations due to slower breakdown (Roggatz & Parsons, 2022). In tropical environments, a decline in coral abundance and destabilized biofilms can lead to the formation of new biofilms, such as turf algae. These new biofilms can replace corals and prevent coral larvae from settling in the future (Lowe et al., 2021). Marine heat waves in subtropical to temperate regions can hinder the recruitment and development of cold-water kelp, enabling warm-affinity benthic organisms, such as tropical corals, to become established (Phelps et al., 2021).

As biofilms change, turf algae can prevent kelp and coral from settling, creating a turf-dominated environment. These examples show how warming waters might affect biological communities at higher latitudes, perhaps leading to divergent tropicalization trajectories (Mizerek et al., 2021). It is crucial to note that the interactions outlined above do not occur separately and are likely to differ among habitat-formers. Indeed, these relationships are extremely complicated and are regulated by a variety of factors, including ambient conditions (Ferguson et al., 2021), accessible surface area (Sims et al., 2021), particular species' physiological tolerances (Abrego et al., 2021), the direct effect of human activities, and microbial and larval supplies (Hadfield, 2011; Hock et al., 2019; Messer et al., 2020). Biofilms have short generation times, are ubiquitous, and are sensitive to toxicants and stressors (e.g., pH, salinity, nutrient pollution). As a result, their initial micro-ecosystem (in terms of community structure, productivity, and resilience) can shift to a more adapted system (e.g., resistance to a pollutant) (Nelson et al., 2020). Biofilms' "ecological memory" has led to the development of structural and functional tools to measure and monitor aquatic ecosystem health (Sunagawa et al., 2020).

3.3.1 Impact of biofilm on the aquatic environment

Biofilms, like other biological systems, can be disrupted and affected by changes in their surroundings. Environmental variations, including pulses, can alter the microbial makeup of biofilms (see Figure 3). Various factors, such as temperature stress (Ferguson et al., 2021), eutrophication (Nelson et al., 2020), acidity (Espinel et al., 2021), and coastal infrastructure, have been linked to changes in biofilm biomass, variety, and community growth. There is growing evidence that biofilms govern carbon fluxes in boreal peatlands and streams (Battin et al., 2016; Wyatt et al., 2021). In these ecosystems, phototrophic biofilms ensure primary production, and heterotrophic biofilms ensure carbon breakdown and cycling (Battin et al., 2003; Bartrons et al.,2012; Besemer, 2015; Keesing & Ostfel, 2021). Biofilms are also an essential trophic level, serving as the sometimes-unique foundation of food webs (Hecky & Hesslein, 1995; Vadeboncoeur & Steinman, 2002; Rott et al., 2006; Weitere et al., 2018; Mahler et al., 2020). Oligotrophic ecosystems, such as alpine or boreal, face extreme conditions that hinder vegetation development due to temperature variability, wind, siltation, and nutrient availability (Geesey et al., 1978; Lock et al., 1984; Rott et al., 2006; Myers et al., 2021).

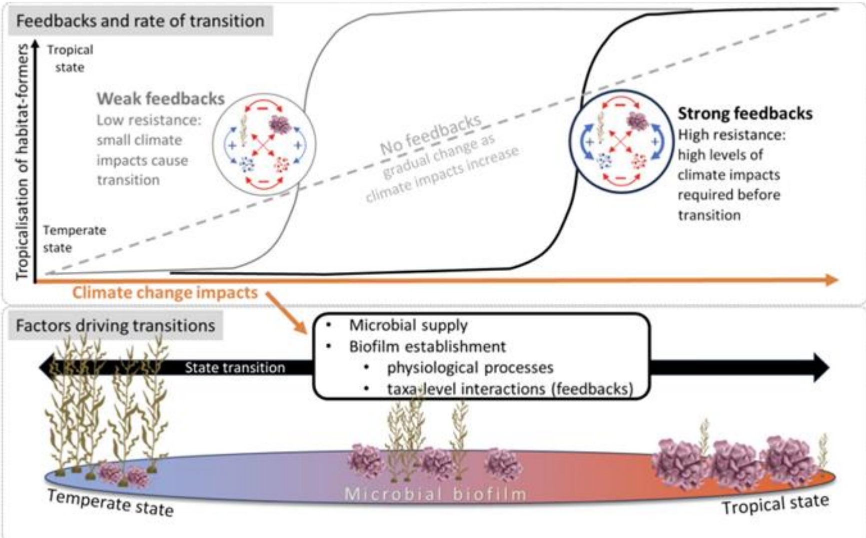

Fig. 3. Climate change alters microbial propagule supply and biofilm composition, driving ecosystem shifts from temperate (kelp-dominated) to tropical (coral-dominated) states. (Cooney et al., 2024).

Biofilms are browsed by invertebrates and vertebrates (Hecky & Hesslein, 1995; Füreder et al., 2003; Rott et al., 2006; Wyatt et al., 2021). The trophic ecology of biofilms and their consumers is crucial for carbon cycling. Lack of top-down control by animals against biofilm herbivory has been linked to increased carbon dioxide emissions in boreal peatlands, highlighting the negative impact of biodiversity loss (Wyatt et al., 2021). Climate change is expected to affect the availability of resources in boreal ecosystems, potentially resulting in the development of heterotrophic biofilm communities under certain conditions (Myers et al., 2021). Periphytic biofilms in fast-flowing waterways also improve water retention in designated areas (Battin et al., 2003). Increasing water retention improves organic particle exchange and nutrient retention, particularly for phosphate and nitrogen. Biofilms in freshwater ecosystems serve as nutrient buffers, influencing biogeochemical and ecological processes (Battin et al., 2003; Lu et al., 2016; Zhu et al., 2020). According to Bonnineau et al. (2020), biofilms stabilize sediments and reduce erosion. Active denitrifying genes were also found in biofilms, suggesting they also have an important role in regulating the global nitrogen cycle, which has been substantially disrupted by the massive use of nitrogen-based fertilizers for food production, emissions from fossil fuel combustion, and the increasing number of humans and livestock (Gruber &

Galloway, 2008; Carthey et al., 2020). These activities have led to the eutrophication and acidification of aquatic ecosystems on a global scale. Nutrient enrichment is considered a type of pollution that affects biofilms, but biofilms can also help mitigate it. For instance, periphyton biofilms have been shown to reduce total nitrogen and phosphorus levels in polluted rivers (Liu et al., 2016; Shabbir et al., 2020).

Nutrient enrichment can cause algal blooms, which affect aquatic ecosystems by producing toxins such as microcystins and depleting water resources, including nutrients, oxygen, and light (Greenspan et al., 2020). Periphyton biofilms, which are composed of diatoms and bacteria, can control cyanobacterial blooms by producing water-soluble allelochemicals that inhibit photosynthesis and compete for nutrients. These biofilms can also reduce algal blooms and restore the health of hyper-eutrophic aquatic ecosystems by providing substrates, such as macrophytes (Wu et al., 2011; Liu et al., 2016; Bernardo-Cravo et al., 2020). Baranov et al. (2020) discovered that periphyton biofilms efficiently eliminate microcystin through adsorption and biodegradation. This finding has significant implications for water quality and the health of animals and humans. Biofilms can minimize pollution in freshwater and soil by absorbing nutrients and contaminants, detoxifying the environment (Cardinale, 2011; Sabater et al., 2002; Pin et al., 2021). Environmental biofilms can capture and metabolize a variety of toxicants, including persistent organic pollutants, pesticides, heavy metals, drugs, industrial effluents, nanomaterials, and plastics (Kohušová et al., 2011; Pérez et al., 2013; Shabbir et al., 2020). Multispecies biofilms may be more effective for remediation than single-species biofilms due to synergistic interactions (Parrilli et al., 2022). Research indicates that biofilms protect natural ecosystems against synthetic chemicals, nutrient pollution, and cyanobacterial blooms (Edwards & Kjellerup, 2013; Joshi et al., 2021).

The diversity of biofilms reflects the diversity of their habitats. Therefore, habitat loss and degradation can significantly impact biofilm functions within ecosystems. However, the magnitude of these modifications varied depending on the original communities, influenced by substrate type (natural rocks or tiles) and biofilm age (Penesyan et al., 2021). In general, the in situ impacts of global warming and other stressors are difficult to forecast because their effects are often synergistic or antagonistic rather than additive (Baulch et al., 2005; Romero et al., 2018; Shu & Huang, 2021). For example, the pesticide triclosan had a considerably harsher effect on river benthic biofilms during drought periods (Vilches et al., 2013; Egipsy et al., 2021). Further research on different stressors is necessary to understand how biofilms and aquatic biota will adapt to global change.

3.3.2 Marine biofilm and microplastic

Microplastics (MPs) are small plastic materials (<5 mm diameter) that have been found in practically every biological niche, making them a growing contaminant concern. Many studies have recently revealed the growing microplastic pollution in oceans and rivers, and this issue has sparked interest in a wide range of areas. Microplastics, due to their tiny size, are easily absorbed into biogeochemical cycles and food webs, having a significant environmental impact. These fragmented plastic particles have a high surface area and can aggregate with natural colloidal substances, pollutants, and suspended solids, such as polychlorinated biphenyls (PCBs) and polyaromatic hydrocarbons, increasing their toxicity (Duan et al., 2021; Atugoda et al., 2021). MPs can easily be ingested by aquatic life, leading to entanglement of digestive systems, physical obstructions, and chronic toxicity from contaminants, such as organic contaminants, antibiotics, and heavy metals (Ali et al., 2024). Biofilms are susceptible to environmental dynamics and the presence of microplastics caused by climate change (Gulizia et al., 2025). It is important to investigate how large-scale climatic changes affect microbial biofilm production and biodiversity from ecological and biochemical perspectives. Plastic production has increased in recent decades, leading to pollution of marine habitats.

MPs from food webs can accumulate in the bodies of higher animals, including humans, posing health risks. Microplastic waste deteriorates the environment and threatens food safety, security, and human health. Proper degradation is necessary to prevent the harmful environmental impacts from MP pollution. Diverse aquatic bacteria play a crucial role in the breakdown of microplastics. Microbial populations produce a biofilm on polymer surfaces, promoting eco-friendly breakdown. Plastic's surface characteristics and density can be influenced by metabolic byproducts. Microplastic particles can aggregate in the water column, float on the surface, or sink to the ocean floor, carrying abiotic and biotic substances (Qaiser et al., 2023). When microplastics are mistaken for food, small marine species consume them. These microscopic animals are then consumed by their predators, who inadvertently swallow the microplastics and move them up the food chain (Bhagwat et al., 2021). Microplastics can spread across the food chain through a mechanism known as 'trophic transfer' (Marcharla et al., 2024).

Microorganisms are essential to the sustainability of aquatic ecosystems and biogeochemical recycling. However, plastic debris, including fragmented polymers, is becoming increasingly prevalent in water and sediment. The degradation of polymers in marine environments is a complex process that depends on factors such as size, concentration, and access to cleavage sites. Larger molecules are more difficult to degrade because they cannot penetrate cells. Bioavailability and chemical stability influence the quality and susceptibility of substrates to microbial colonization and breakdown (Smułek & Kaczorek, 2022). Microplastics at sea are quickly covered by a thin layer of inorganic and organic materials that acts as a substrate for microbial colonization and biofilm development. Biofilms can alter the structure and function of microplastics in various ways. Biofilms use microbial enzymes to change surface characteristics, degrade additives, and release by-products, affecting the fate of microplastics in marine ecosystems (Debroy et al., 2021). Environmental conditions, including temperature and ionic strength, can impact bacterial colonization and biofilm development (Moyal et al., 2023). Microbes use a variety of ways to adhere to surfaces, including changes in cell surface charge and hydrophobicity, as well as EPS formation (see Figure 4). Pioneer colonization can alter the characteristics of a polymer surface, thereby facilitating the assembly of other bacteria. Submerging polyethylenes in seawater for three weeks reduced their hydrophobicity (Affianti et al., 2022). Substrate factors, such as crystallinity, melting temperature, and roughness, might impact microbial community formation during early colonization (Zhang et al., 2025).

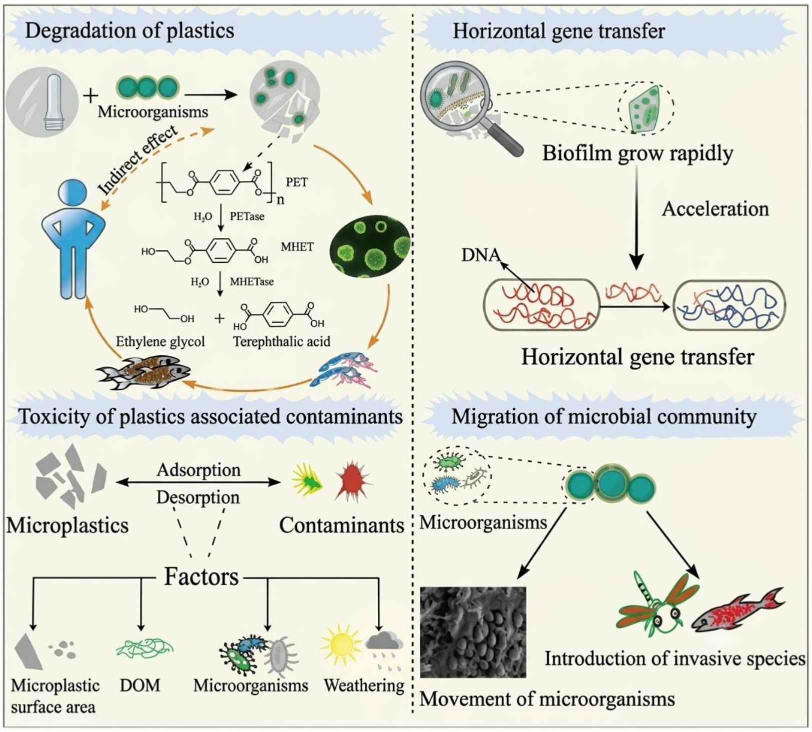

Fig. 4. Implications of biofilm on microplastics
(Sooriyakumar et al., 2022).

Interactions between the microplastic surface and microorganisms result in the colonization of a varied microbial population and biofilm formation. Microplastics serve as colonization platforms for various bacterial populations in aquatic environments. Potential colonizers of plastic include Arcobacter, Vibrio, Colwellia, Escherichia, Pseudomonas, and a few additional species (Zhai et al., 2023). Lear et al. (2022) studied microbial colonization on PET bottles and glass slides submerged in the North Sea. They found seasonal and locational changes in community composition but no difference between populations associated with PET and glass. These results suggest that microbes on polymer surfaces are not substrate-specific. Curren and Leong (2019) collected microplastics from Singapore's coastline and used high-throughput sequencing to identify epiplastic bacterial populations. They discovered a complex bacterial community that included species of Erthrobacter, Vibrio, and Pseudomonas (Curren & Leong, 2023).

Microplastics are connected with a diverse bacterial population, including members of Rhodobacteraceae and Flavobacteriaceae, as well as Albirhodobacter, Methylotenera, and Hydrogenophaga species. Biodegradation of polymers by a single bacterial culture can produce hazardous metabolic byproducts that may hinder growth. Using a bacterial consortium can help eliminate these harmful compounds (Bozkurt et al., 2022). Researchers have proven the effectiveness of using bacterial consortia for microplastic biodegradation. Gao & Sun (2021) found that using a unique strategy to reestablish a bacterial colony on biofilms improved plastic degradation. After screening hundreds of pieces of plastic debris, the researchers identified three bacteria that can degrade plastic. This microbial community, consisting of the chosen bacterial species, has proven its ability to break down polymers such as PET and PE, and the degradation products were analyzed using modern methods. The study concluded that customized biofilms derived from marine bacterial populations could efficiently degrade microplastic debris in aquatic environments. Engineered biofilms can effectively break down microplastics in marine environments. Further investigation of microplastics and biofouling revealed different long-term outcomes in various environments due to variations in biofilm formation. For instance, it can encourage deep-sea settlement or alter the feeding preferences of biota (Gulizia et al., 2025).

3.3.3 Biofilm, environmental pollutants and waste management

Using microbes to remove contaminants is an environmentally favorable practice. Biofilm-based bioremediation reduces contaminants in the environment. However, biofilms, which form the basis of aquatic food webs, absorb toxicants, making them accessible to consumers and posing a health concern. They play a crucial role in monitoring and understanding the fate of toxicants in aquatic environments (Mahler et al., 2020; Bonnineau et al., 2021). Biofilms are natural environments in which bacteria exchange genetic material, signaling chemicals, and metabolites. The use of microbiome-based bioremediation is becoming increasingly important globally. Genetically modified biofilms enhance the potential for contaminant bioremediation. Biofilms have more biomass than suspended cells, which protects them from chemical degradation. The physicochemical structure of biofilms enhances compound availability and sorption, allowing for the coexistence of aerobic and anaerobic bacteria and making biofilms more effective at toxicant degradation (Saini et al., 2023). Anaerobic denitrifiers and oxygen-dependent nitrifiers coexist in biofilms at different levels, effectively removing nitrogen from wastewater (Zhong et al., 2024). Anaerobic ammonium-oxidizing (anammox) microorganisms generate microenvironments comparable to those of aerobic ammonia-oxidizing bacteria. Gradient niches allow reactions with varying oxygen and substrate demands to occur within the same biofilm.

Biofilms are used to clean wastewater by addressing biological oxygen demand (BOD), ammonia, nitrogen, nitrates, and dissolved oxygen (DO). Wastewater nutrients promote the growth of microorganisms and their metabolites, which effectively eliminate pollutants. Microbial exopolymers in biofilms can absorb chlorinated herbicides, such as diclofop-methyl and methyl 2-[4-(2,4-dichlorophenoxy)phenoxy]pyruvate, and bioremediate aromatic chemicals in marine environments into catechols (Chattopadhyay et al., 2022). Aerobic degradation of naphthalene, phenanthrene, and pyrene in saltwater and marine sediments eliminates chlorinated ethenes and degrades chlorinated benzoates, as well as bioremediating naphthalene (Gao et al., 2024). Generation of biofilms on low-density polyethylene (LDPE) surfaces has been shown to degrade phenanthrene, diazinon, and catechol (Saini et al., 2023).

Bacterial species were used for bioremediation, trapping pollutants in EPS and providing nourishment to promote biofilm formation and prevent PAH toxicity. Pseudomonas sp. strain ADP bacteria can use atrazine as a nitrogen source (Davletgildeeva & Kuznetsov, 2024). Another study found that P. aeruginosa N6P6 marine bacteria can form biofilms in the presence of certain PAH concentrations (Kumari et al., 2021). Biofilm-mediated reactors have successfully remediated various insecticides and herbicides, such as 2-propionic acid, diclofop, 2,4-dichlorophenoxyacetic acid, carbendazim, choropropham, methomyl, diazinon, carbamate compounds, carbaryl, and carbofuran (Maurya & Raj, 2020). Biofilm-forming bacteria have been studied for their role in bioremediation, including feeding, oxygen competition, and pollutant tolerance. Bioremediation research has focused on these bacteria because they contribute to feeding, oxygen competition, and pollutant resistance. Biofilms in heavily contaminated areas can endure harsh environmental conditions, such as predation, nutrition levels, salinity, pH levels, temperature fluctuations, UV exposure, and pollutant concentrations (Saini et al., 2023). At pH 7.5, positively charged micro-contaminants have a higher sorption potential in activated sludge than neutrally or negatively charged micropollutants (more than 90%).

Table 1. Biofilm-mediated bioremediation of inorganic micropollutant and organic contamination from wastewater

Biofilm	Wastewater type	Contaminants
Gordonia sp. H19 and P. *monteilii P26*	Artificially created seawater	Crude oil
Batch-dispersed biofilm reactor Sequencing based on algal-bacterial interaction	Sewage from the home	Total phosphorus and total nitrogen
Membrane bioreactor	Sewage from the hospitals	Trimethoprim, propranolol, diatrizoic acid, clarithromycin, and azithromycin
Sphingobacteriales, Flavobacteriales, Xanthomonadales, and *Burkholderiales*	Sewage from the hospitals	Drugs
Batch biofilm reactor using algae as a sequence agent	Sewage from the home	Phosphorus and Nitrogen
MBR	Wastewater with saline conditions	Ammonium and COD
Membrane bioreactor	Sewage from the city	Medicines
Membrane bioreactor	Sewage from the city	4-Nonylphenol, 17b-estradiol, naproxen, and diclofenac
B. mojavensis M1+ R. rhodochrous BX2	Groundwater	Organic cyanide
Arthrobacter sp.	-	Chromium(VI)
B. atrophaeus CN4	-	Naphthalene
Trichosporonasahii B1, Candida tropicalis TH4, and *Candida viswanathii TH1*	-	Pyrene, anthracene, naphthalene, and phenol
Microalgal biofilm	Sewage from the city	Phosphorus and NO^2
Chlorella vulgaris	Wastewater contaminated with aromatic hydrocarbons collected from petroleum storage	Removing nutrients

(Saini et al., 2023).

3.3.4 Climate change dynamic influence

Climate change is altering environmental conditions and connectivity patterns, potentially expanding the range of biofilms (Cooney et al., 2024). Extreme weather events are becoming increasingly common, particularly in desert or continental climates, such as those in the Mediterranean region. This event may lead to stronger, longer, and more frequent temperature maxima in different regions (Kekkou et al., 2025). Several species have been shown to thrive in harsh environments due to their distinct morphology and biochemistry, as well as their capacity to build biofilms. Bacterial biofilms have been found in numerous thermal sources, including the "Bison Basin" in Yellowstone National Park and Loki's Castle in Atlantic Ocean (Malygina et al., 2023). Research indicates that there is a link between biofilm growth and ambient temperature (Alabdullatif, 2024).

Biofilms' multi-layered structure creates an inner zone conducive to growth and development, enabling survival at severe temperatures (Agarwal et al., 2024). Biofilms that form at severe pH levels are also of great interest, with the chemoautotrophic biofilm in the Richmond Mine in the United States being the most researched. Studies from this and other similar zones, such as Rio Tinto in Spain, have demonstrated how biofilm architecture and variety affect tolerance to harsh environmental conditions (Egipsy et al., 2021). Bacterial biofilms have also been found in hypersaline environments, such as salt lakes and evaporation ponds. Aqeel and Liss (2022) proved that biofilms can grow at pH levels exceeding 11.0, focusing on Dietzia and Anaerobranca taxa. The matrix's composition creates a buffer environment that is 1.0–1.5 pH units lower than the ambient environment, which promotes cell growth. Other research has found that higher temperatures during the season promote microbial growth and biofilm development (Preciado et al., 2021). Higher biofilm activity has been linked to greater accumulation of iron (Fe) and manganese (Mn), suggesting that metal accumulation accelerates at higher temperatures (McCormick et al., 2021). Temperature significantly impacts the structure of biofilm bacterial communities. Several studies have found that compositional changes are also influenced by the management method used. At 24°C, Pseudomonas dominated the biofilm bacterial community structure, and genera such as Flavobacterium, Sphingobium, and Methyloversatilis were shown to increase with temperature (Samrot et al., 2021). According to research, bacterial biofilms play a crucial role in microbial survival and evolution in harsh environments. Bacteria adapt to diverse stresses, such as high temperatures, extreme pH levels, and hypersalinity, which provides them with evolutionary benefits (Shree et al., 2023).

3.3.5 Marine ecology monitoring compass and biofilm: E-DNA presence

There is a growing use of environmental DNA in research, particularly for freshwater and marine environments. Anthropogenic stressors significantly impact the biodiversity, ecological processes, and services of these ecosystems (Chen et al., 2025). The eDNA metabarcoding technique has been effectively used to monitor historical and current biodiversity in a variety of environments. Its practical applications include detecting invasive and endangered species (Ahmed et al., 2022), conducting biodiversity surveys to evaluate ecosystem health, and assessing industrial impacts (Djurhuus et al., 2020). Initially, environmental DNA (e-DNA) was used primarily to study microbial diversity. The revolutionary idea underlying this technique is that RNA or DNA extracted from environmental samples can be used to evaluate the natural diversity of microorganisms. Molecular techniques, particularly eDNA studies, have been proposed as a game changer for bioassessment and biodiversity monitoring (Altermatt et al., 2020). PCR amplification has accelerated research on microbial diversity in environmental samples. The discovery of uncultivable bacterial species in the ocean and soil transformed understandings of bacterial diversity (Musiałowski et al., 2025). However, the term "environmental DNA" was not commonly used in the early days of bacterial diversity research. It was coined to refer to the "free" DNA produced by dying cells, also known as "extracellular DNA" (Sahu et al., 2025).

Biofilm communities play a crucial role in stream ecosystems by absorbing, storing, and transforming dissolved organic matter and nutrients (Lines et al., 2025). Biofilms exhibit higher rates of e-DNA transformation, which is concentrated in the biofilm EPS (Bergsveinson et al., 2021). e-DNA may be extracted from a variety of materials, most notably soil, sediment, and water, but also from air, biofilm, and organic remnants such as feces, which may include DNA of numerous sources (Figure 5). This definition prevents size-based physical selection of target taxa, such as sieving or kicknet sampling. Using environmental DNA metabarcoding for high-throughput sequencing of diatom species present in biofilm samples may overcome the constraints of classic microscopic techniques.

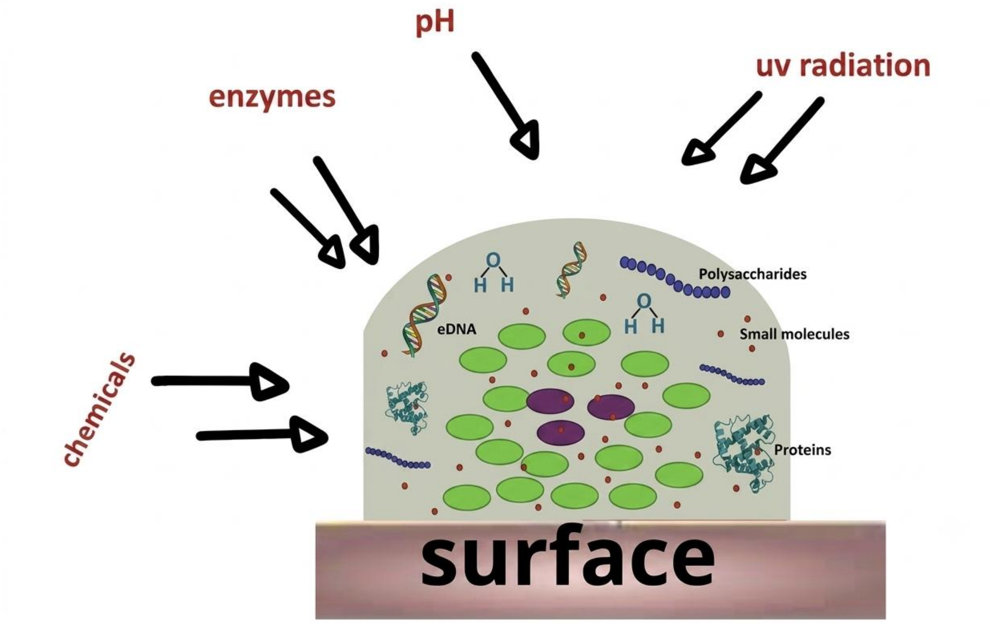

Fig. 5. The biofilm matrix protects eDNA and other biomolecules from environmental stressors by acting as a physicochemical barrier.
(Joseph et al., 2022).

First, determining the ecological state may be much quicker and less costly. Furthermore, the outcomes would be easier to monitor and compare because they are based on comparable sequencing data. Effective monitoring requires accurate, timely, and reliable assessments of the state and changes in organismal communities. This can be achieved by describing biodiversity or calculating indices as proxies for the environmental state (Chang et al., 2025). High-resolution biomonitoring data is essential and may require innovative technology. Indeed, the classical morphotaxonomic method has its own set of biases and limitations, including inadequate taxonomic resolution and information gaps in morphospecies ecology (Gentil et al., 2021). Finally, numerous samples might be processed in parallel using molecular technologies, which is a significant benefit for large-scale and continuous surveys. eDNA/eRNA is more stable when attached to inorganic or organic particles, as long as there is enough time to settle and integrate into the matrix. Contrary to expectations, biological activity in biofilms may help preserve eDNA.

An early study revealed that biofilms' polysaccharide matrix might absorb eDNA (Secchi et al., 2022). The matrix, which includes eDNA, can be destroyed enzymatically, but it is often retained due to its structural, protective, nutritional absorption, cell communication, and horizontal gene transfer functions (Panlilio & Rice, 2021). Moreover, Sakata et al. (2020) discovered that eDNA from soil samples had substantial geographic variability in species composition within a survey site, whereas eDNA in water was more evenly distributed. As a result, sediment samples should be replicated many times within a site to reflect the local biotic communities accurately. Rivera et al. (2021) offer a method for capturing macroinvertebrate eDNA in rivers using biofilms, indicating that biofilms can store this information and need further investigation.

3.4 Biofilm and human nexus: Antibiotics and biofilm

Antibiotic resistance among microorganisms is a global concern for people, veterinary, food, and other industries (Rather et al., 2021). Biofilms are immune to antibiotics, disinfectants, and phagocytosis. They rely on self-produced EPS to survive (Glatthardt et al., 2020). They have a multilayered defense system that includes persister cells, adaptive stress responses, limited antibiotic penetration, limited nutrition, decreased growth and metabolic activity, and antimicrobial inactivation within the EPS matrix (Almatroudi, 2025). The biofilm phenotype of the Salmonella serovar exhibits antibiotic resistance that is a thousand times greater than that of planktonic bacteria (Sung et al., 2023). Milton et al. (2022) found that the foodborne bacterium *Clostridium perfringens* can form biofilms, becoming more resistant to antibiotics in the process. Gu et al. (2022) found a significantly higher frequency of antibiotic-resistant genes (ARGs) in biofilms than in soil and water samples. These genes included sul1, sul2, tetA, and tetW. According to Uruén et al. (2020), *S. aureus* isolates from subclinical bovine mastitis cases overexpressed adhesion and biofilm-related genes and were resistant to β-lactam drugs.

Biofilm resistance to antibiotics can be attributed to various factors, including limited antimicrobial penetration, the presence of antibiotic-modifying enzymes in the matrix, eDNA, hypoxia, reduced growth, variability in physiology, oxidative stress, amino acid starvation responses, efflux pumps, quorum sensing, persister cells, horizontal gene transfer (HGT), a high mutation rate, and colony variants (Almatroudi, 2025). The next section delves deeper into certain aspects.

3.4.1 Biofilm resistance: Medical perspective and spreading of infection

Antibiotic-resistant bacterial infections pose significant healthcare issues, including increased morbidity and death. Treating these infections with currently available medications is more challenging and, in some cases, impossible (Khan et al., 2021). Designing antibacterial, antifouling, anti-biofilm, and anti-infection surfaces and materials reduces the need for systemic antibiotic therapy and the risk of antibiotic resistance in many bacterial species. Antibiotics are antibacterial agents that can be used in coatings to create bactericidal surfaces.

Biofilm engagement in medical domains was first demonstrated in 1982 through electron microscopy investigations (Grari et al., 2025). Infections can arise on numerous devices, such as central venous catheters, urinary catheters, mechanical heart valves, pacemakers, joint implants, and endotracheal tubes. They are particularly problematic in intensive care units, where medical equipment is commonly used (Assefa & Amare, 2022). Biofilm development on medical devices is influenced by factors such as the device surface's physicochemical properties, the flow rate of liquids through the device, and the presence of host-derived proteins such as fibrinogen and fibronectin, which promote microbial adherence. Infectious isolates from healthcare professionals or patients can cause biofilm growth on medical equipment. Pathogens commonly implicated include *S. aureus, S. epidermidis, P. aeruginosa, E. coli, E. faecalis,* and several *Candida* species. Biofilms containing many bacterial and fungal species exacerbate diseases. For example, *Candida albicans* can function as a scaffold for *S. aureus*, leading to increased antibiotic resistance (Karishma & Sophie, 2023).

Antimicrobial resistance in biofilms is a complicated phenomenon comprising several mechanisms that collectively contribute to the greater resilience of biofilm-embedded bacteria compared to planktonic counterparts. These bacteria are 10–1000 times more resistant to antimicrobial treatments than planktonic cells. This increased resistance results from a variety of causes. The EPS works as a physical barrier, slowing down the diffusion of antibiotics and other antimicrobial compounds, which can be further deactivated in the outer layers of the biofilm before reaching the inner cells (Olaimat et al., 2024). One key factor is the limited dispersion of antibacterial drugs within the biofilm matrix. Furthermore, certain EPS components, including polysaccharides and eDNA, might chemically interact with antibiotics, reducing their effectiveness (Figure 6). EPS-associated enzymes can enzymatically inactivate antibiotics by degrading or modifying them into less active forms (Bielecka et al., 2021).

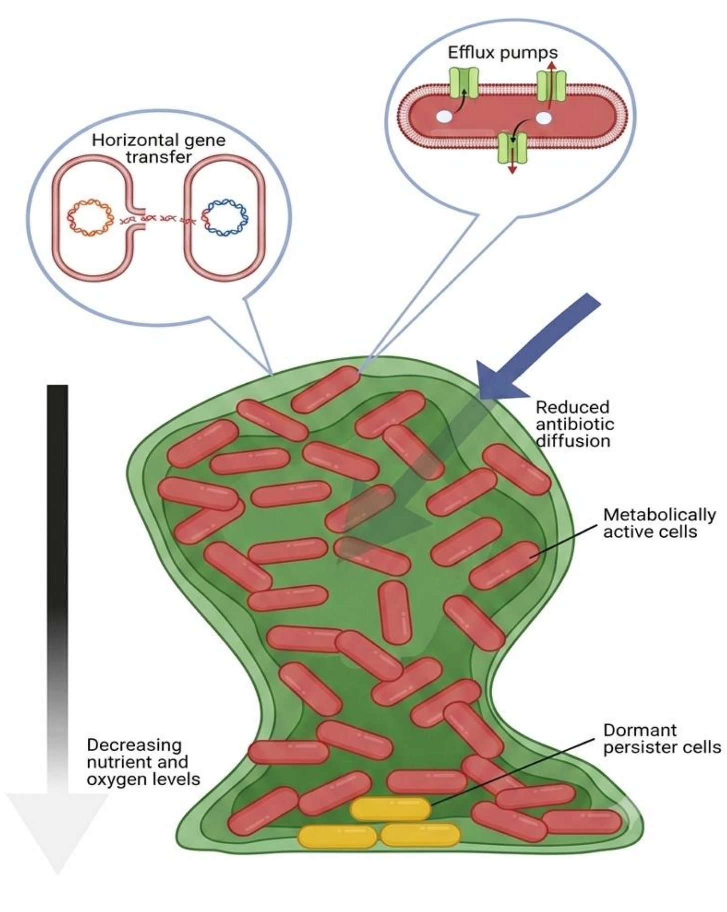

Fig. 6. Mechanism of antimicrobial resistance of biofilm including limited antibiotic diffusion due to EPS matrix that facilitates the dissemination of antibiotic resistance genes.
(Grari et al., 2025).

3.4.2 Biofilm infections, plant disease, food industry and beyond

Biofilms can cause chronic illnesses, nosocomial infections, industrial pipe fouling, food deterioration, dairy product contamination, and ship hull fouling (Muhammad et al., 2020). Biofilms have a wide range of negative impacts on society. They can form on medical device surfaces, as well as on dead (e.g., bone sequestra) and live (e.g., lung tissue and tooth surfaces) tissues (Zhao et al., 2023). These bacteria can grow on biomedical equipment, including catheters, prosthetic heart valves, pacemakers, breast implants, contact lenses, and cerebrospinal fluid shunts (Ciarolla et al., 2022). *P. aeruginosa* biofilms are commonly seen on chronic wounds (Cendra & Torrents, 2021), and chronic osteomyelitis is a biofilm infection where germs stick to dead bone tissue (Wu et al.,2022). Biofilm-related organisms are responsible for approximately 65% of microbial infections and are highly resistant to antimicrobial drugs and human defenses (Bhuiyan, 2025).

Biofilms have a significant influence on human health. Biofilm-forming bacteria can cause life-threatening infections and diseases in humans, including cystic fibrosis, otitis media, periodontitis, infective endocarditis, chronic wounds, and osteomyelitis (Perry & Tan, 2023). *P. aeruginosa* biofilms can cause severe pulmonary infections (Rosa et al., 2025).

Haemophilus influenza biofilm can cause otitis media (Xiao et al*.,* 2023). Pseudomonas biofilms can cause periodontitis; an infection of the gums that damages soft tissues and bones supporting the teeth. According to Mirzaei et al*.* (2020), up to 80% of bacterial infections in people are caused by biofilms, regardless of device type. They are generally caused by the production of biofilms on biotic surfaces of the body, such as mucosal surfaces, teeth, or tissues. Despite their prevalence, they are frequently overlooked when compared to biofilm direct infections. Nevertheless, they account for a significant proportion of persistent bacterial infections. Comparative investigations have found that biofilm development and persistence processes are comparable in device and non-device related illnesses (Mirzaei et al*.,* 2020; Domenico et al*.,* 2022). Chronic wound infections typically involve polymicrobial biofilms that include both skin-related and anaerobic bacteria, which hinders the healing process. Furthermore, non-device-related osteomyelitis, a serious bone infection, has been shown to entail large biofilms that require surgical intervention to resolve. Biofilms can cause chronic sinusitis, native valve endocarditis, prostatitis, infectious kidney stones, and biliary tract infections (Mirzaei et al*.,* 2020).

Biofilm-related disorders have been seen in agricultural contexts. *Xanthomonas citri* biofilm can cause plant diseases, including Pierce's disease in grapevines and citrus canker (He et al*.,* 2022). *Xylella fastidiosa*, a strong biofilm producer, can induce Pierce's disease in grapevines by restricting the plant's vascular system (Castro et al*.,* 2021). *P. syringae* causes brown spot disease in bean plants, which has been linked to biofilms (Chaudhry et al*.,* 2021). Similarly, *P. aeruginosa* biofilm infections on Arabidopsis and *Ocimum basilicum* (sweet basil) roots can quickly lead to death. *Ralstonia solanacearum*, a plant pathogenic bacterium that forms biofilms on xylem arteries, can cause bacterial wilt disease in plants (An & Zhang, 2024).

Biofilms pose significant technical challenges in the food industry by preventing heat flow, increasing friction, and promoting corrosion, resulting in decreased production efficiency (Liu et al*.,* 2023). Biofilms pose a risk of contamination with dangerous microorganisms in the food industry, including instruments and equipment. They can form on surfaces that come into contact with food (Shahid et al*.,* 2025). Biofilms cause around 60% of foodborne outbreaks (Liu et al*.,* 2023). In food processing settings, biofilms can pose a significant threat to food safety and the industry (Olanbiwoninu & Popoola, 2023). Contaminants in food processing facilities often originate from the surrounding air, equipment, or food surfaces (Pakdel et al*.,* 2023). Biofilms in food processing settings can ruin food, posing significant public health risks and economic consequences (Sharan et al*.,* 2022). Shuvra (2023) identified *Escherichia coli* 0157:H7 as a strain that causes hemorrhagic colitis. *Pseudomonas spp.*, *Vibrio parahaemolyticus*, and *Clostridium perfringens* are common spoilage organisms with negative effects on food (Salama & Chennaoui, 2024).

3.4.3 Antibiofilm agents

Antibiotic resistance is the ability of bacteria to proliferate despite antimicrobial treatments or medications. These organisms cause serious infections in humans. Microorganisms gain antibiotic resistance by acquiring plasmids containing the resistance gene. These plasmids can be passed between bacteria through a process called conjugation. These genes can spread rapidly across bacterial cells. These pathways involve several components, including conjugative plasmids, transposons, efflux pumps, and chromosomal alterations (Jacobowski et al*.,* 2025). Biofilms prevent antibiotics and antimicrobial agents from reaching the cell surface, contributing to antibiotic resistance (Muteeb et al*.,* 2023). Sharif *and* Yadav (2025) proposed three hypotheses for biofilm resistance: slow or incomplete antibiotic penetration, an altered chemical microenvironment, and a subpopulation of microorganisms forming a unique, highly protected phenotype similar to spore formation.

Biofilm antimicrobial resistance is a complicated phenomenon involving multiple processes that make biofilm-embedded bacteria more resilient than their planktonic counterparts. These bacteria are more resistant to antimicrobial drugs than planktonic cells (Singh et al., 2025). This increased resistance has a variety of causes. One key factor is the limited dispersion of antibacterial drugs within the biofilm matrix. The EPS acts as a physical barrier that slows the diffusion of antibiotics and other antimicrobial compounds. These compounds may be further deactivated in the outer layers of the biofilm before reaching the inner cells (Pinto et al., 2020). Biofilms exhibit physiological heterogeneity with nutrient and oxygen level gradients, leading to a population of slow-growing or dormant persister cells that are more resistant to antibiotics that target active cellular functions (Jo et al., 2022).

Persistence, unlike genetic resistance, is a temporary characteristic that cannot be passed down across generations. Antibiotics do not promote the proliferation of persister cells. Nonetheless, they can repopulate the biofilm when the antibiotic concentration drops, leading to the recurring and chronic character of biofilm-associated illnesses. Phenotypic tolerance is a significant therapeutic problem as it allows germs to survive even when tested for antibiotic sensitivity (Chang et al., 2019; Galdiero et al., 2020). Therapeutic drugs often get embedded in bacterial biofilms, limiting their penetration (Ali et al., 2023). Natural sources, including plant extracts, marine microorganisms, exopolysaccharides, cyanobacteria, and microalgal chemicals, are being investigated for their biofilm inhibitory potential. In addition to inhibiting biofilm, some chemicals can also inhibit *Vibrio spp*.'s QS (Quorum sensing) mechanism (Behzadnia et al., 2024). Recent research suggests that natural sources, microbial extracts, nanoparticles, and bacteriophages can effectively combat Vibrio spp.'s biofilm development.

3.5 Future studies: Unfolding potential

Biofilms are unusual, multidimensional life forms that challenge traditional understanding of living creatures. For decades, the understanding of biofilms has relied on in vitro models. In vivo biofilms are typically patches or cell aggregates that are less than 200 µm in size (Beloin & McDougald, 2021). This has implications for how biofilms interact with the host (Alhede et al., 2020). There are three perspectives on biofilms: (1) as a lifestyle for individual microbial cells (the single-cell-centric view); (2) as a multicellular organism (the biofilm-centric view); and (3) as structures that foster microbial diversity (a current novel perspective). Though these views seem conflicting, they are actually complementary. Together, they provide a comprehensive understanding of biofilms as a unique microbial lifestyle with multicellular characteristics. Biofilms also serve as protective environments for microbes, generating genotypic and phenotypic diversity prior to release. A broader perspective on biofilms allows us to discover new occurrences and uncover previously overlooked characteristics.

The significance of understanding biofilm biology is often underestimated, and the distinctions between planktonic cultures and biofilms may appear abstract to a larger audience. This might be because the microbial world is invisible to the naked eye. Integrating fields including biology, chemistry, genetics, and environmental sciences can further biofilm research. Future research on biofilms should focus on their biodiversity (Li et al., 2025) and ecological importance in ecosystems. Microbes can also act as catalysts for biogeochemical nutrient cycling and serve as indicators of water quality, similar to how microbes can detect environmental changes in temperate and polar environments (Crowther et al., 2024). QS, which plays a direct role in biofilm development, is an important research topic.

Understanding biofilm dynamics and chemical signals might lead to environmentally friendly antifouling devices using enzymes and QS inhibitors (Liu et al., 2024). Understanding the antibiotic susceptibility and biocide tolerance of bacterial biofilm components can lead to novel biofilm control strategies. Future studies on biofilms and their interactions with higher species present significant hurdles: First, developing superior multi-species biofilm models will help study interactions in multi-species biofilms, which are common in vivo. The next step in studying biofilms in nature and disease contexts is to apply technology to investigate their infectious microenvironment. Second, develop applicable models (in vitro, ex vivo, or in vivo) to better understand how biofilms interact with hosts. These models are crucial for studying biofilm-host interactions and evaluating eradication techniques in real-world scenarios; and finally, develop imaging and detection tools to observe and identify biofilms in situ, including the ability to access individual cell physiology as needed. Using innovative methodologies in biofilm research can provide new information and open up new areas of study.

4. Conclusions

This review emphasizes the critical role of microbial biofilms as complex, heterogeneous systems that influence the interconnected dynamics of environmental and human health. Biofilms function as structured microbial communities exhibiting functional diversity and resilience, which underpin essential ecological processes such as nutrient cycling, biomonitoring, and bioremediation. However, the inherent stability of the extracellular polymeric substance (EPS) matrix also facilitates the persistence of pathogenic organisms, contributing significantly to the global challenge of antimicrobial resistance and environmental biopathologies such as biocorrosion. These dualistic characteristics necessitate a paradigm shift toward a One Health framework, integrating medical and environmental research to address biofilm-associated risks comprehensively. Future research should prioritize the development of targeted, non-antimicrobial strategies disrupting EPS integrity to mitigate pathogenic biofilm establishment, while further investigating the diversity-stability relationship in biofilms across diverse habitats. Conceptualizing biofilm health as a measurable and transdisciplinary parameter will enhance scientific communication, foster collaborative research, and support sustainable interventions at the interface of human and ecosystem health.

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Author Contribution

M.A.: Conceptualization, Methodology, Validation, Formal Analysis, Investigation, Resources, Data Curation, Writing – Original Draft Preparation, Writing – Review *and* Editing, Visualization, Supervision. Ishmah Sumayyah: Writing – Original Draft, Writing – Review *and* Editing,

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The authors confirm that the data supporting the findings of this study are available within the raw data that support the findings of this study are available from the main author and corresponding author, upon reasonable request.

Conflicts of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Declaration of Generative AI Use

During the preparation of this work, the authors used Grammarly and Quillbot to assist in improving the grammar, clarity, and academic tone of the manuscript, and Google Translate to interpret and translate specific passages from reference materials. After using these tools, the authors reviewed and edited the content as needed and took full responsibility for the content of the publication.

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Biographies of Authors

Muhammad Awaluddin, Department of Aquaculture, National Taiwan Ocean University, Keelung City, Taiwan 202, Taiwan, Republic of China.

- Email: bussinessuddin@gmail.com
- ORCID: 0000-0003-1731-1692
- Web of Science ResearcherID: N/A
- Scopus Author ID: N//A
- Homepage: https://scholar.google.com/citations?user=jl6sYTkAAAAJ*and*hl=en

Ishmah Sumayyah, Department of Environmental Engineering, Institut Teknologi Bandung, Bandung City, West Java 40132, Indonesia .

- Email: ishmahsumayyah08@gmail.com
- ORCID: 0009-0009-7005-8519
- Web of Science ResearcherID: N/A
- Scopus Author ID: N/A
- Homepage: N/A