Revista Brasileira de Recursos Hídricos
Brazilian Journal of Water Resources
Versão On-line ISSN 2318-0331
RBRH, Porto Alegre, v. 30, e11, 2025
Scientific/Technical Article

https://doi.org/10.1590/2318-0331.302520240083

The influence of hydrological variables on river biofilms: a review of relationships and environmental implications

A influência das variáveis hidrológicas nos biofilmes de rios: uma revisão das relações e implicações ambientais

Bárbara Alves de Lima[1], Tatiani Andressa Modkovski[1], Juliane Ribeiro das Chaves[1], Luis Otavio Miranda Peixoto[1], Samanta Teixeira Medeiros[1], Gabriel Helmer Baer[1], Cristóvão Vicente Scapulatempo Fernandes[2] & Júlio César Rodrigues de Azevedo[1]

[1]Universidade Tecnológica Federal do Paraná, Curitiba, PR, Brasil
[2]Universidade Federal do Paraná, Curitiba, PR, Brasil

E-mails: engbarbaradelima@gmail.com (BAL), tatianiandressa@gmail.com (TAM), juliane.rib21@gmail.com (JRC), luisotaviopeixoto@gmail.com (OMP), samantamedeiros@alunos.utfpr.edu.br (STM), gabrielhelmer@alunos.utfpr.edu.br (GHB), cris.dhs@ufpr.br (CVSF), jcrazevedo.utfpr@gmail.com (JCRA)

Received: August 20, 2024 - Revised: October 19, 2024 - Accepted: December 06, 2024

ABSTRACT

Human actions are caused by multiple ecological stresses that affect native biotas. River biofilms are large aggregates composed of a wide range of organisms. In water resources, biofilms are subject to hydrological variations, which can influence colony processes. This study aimed to verify the state of the art on the relationship between hydrological variables and natural river biofilms, through a literature review. The results show that the most recurrent hydrological variable in studies was discharge/fluxes, while for biofilms, biomass formation was the most frequent variable. Because they represent a portion of the beginning of the trophic chain, there is an urgent need to verify the synergistic effects to which biofilms are subjected in natural environments. It is expected that this review will show the approaches used to understand these relationships, and also expand the current research scenario, to elucidate gaps to be filled.

Keywords: Flow; Water level; Environmental monitoring; Biofilm; Biomass.

RESUMO

As ações humanas têm causado múltiplos estresses ecológicos que afetam as biotas nativas. Os biofilmes de rios são grandes agregados compostos por uma ampla gama de organismos. Nos recursos hídricos, os biofilmes estão sujeitos às variações hidrológicas, que podem influenciar os processos das colônias. Este estudo teve como objetivo verificar o estado da arte sobre a relação entre variáveis hidrológicas e biofilmes naturais de rios, por meio de uma revisão de literatura. Os resultados mostram que a variável hidrológica mais recorrente em estudos foi a vazão/fluxos, enquanto para o biofilme a formação de biomassa foi a variável mais frequente. Por representarem uma parcela do início da cadeia trófica, há urgência em verificar os efeitos sinérgicos aos quais os biofilmes estão submetidos em ambientes naturais. Espera-se que esta revisão mostre as abordagens utilizadas para compreender estas relações, e ainda, amplie o cenário atual de pesquisas, para a elucidação de lacunas a serem preenchidas.

Palavras-chave: Fluxo; Nível de água; Monitoramento ambiental; Biofilme; Biomassa.

INTRODUCTION

The advancement of industrial activities and the intensification of urbanization processes have contributed to the degradation of water bodies and water quality indexes and the surrounding ecosystem, bringing risks to future generations, to the native biota of rivers and bringing reflection on mitigation and recovery strategies for these systems (Montagner et al., 2019; Palmer & Ruhi, 2019; Böger et al., 2021; Cui et al., 2023; Shi et al., 2024).

Anthropogenic changes in ecosystems, along with the worsening of extreme hydrological events such as droughts and prolonged rains, caused by the climate change scenario, have affected the hydrology of rivers and streams, altering the frequencies and durations of such events, impacting the environment in countless ways (Palmer & Ruhi, 2019; Smeti et al., 2019; Pereda et al., 2021; B-Béres et al., 2022; Bertrans-Tubau et al., 2023). In the context of water resources, these changes are related to changes in flow dynamics, basal temperatures, water quality, water chemistry, and extreme events (Myrstener et al., 2020; Pereda et al., 2021; Ramoneda et al., 2021; Flemming et al., 2023), causing changes in living organisms and their relationships and functioning (Palmer & Ruhi, 2019; Pan et al., 2020).

Biofilms are complex communities composed of aggregates of various microbial cells together with several other organisms with heterotrophic and autotrophic characteristics inside, surrounded by an external matrix of polysaccharides and proteins (Sabater et al., 2007; Chaumet et al., 2019; Pu et al., 2019).

Due their heterogeneous nature, biofilms have a great capacity to respond to ecosystem conditions, mainly due to the fast growth and development capacity of colonies, in addition to the diversity and variety of species in the composition and consequent physiology of such organisms (Sabater et al., 2007). This internal variety can be significantly influenced by natural factors, as well as by external physical and chemical stressors, often related to anthropogenic activities (Villeneuve et al., 2011; Calapez et al., 2020).

These external factors, individually, act on the internal dynamics of the biofilm. However, it is the synergistic actions of these aspects that can promote an effect different from that expected when considering only the parameters in isolation, depending on the environment in which they are inserted (Calapez et al., 2020). In this sense, hydrological variables and environmental quality act as major stressors, which may or may not be favorable to the formation and maintenance of biofilms, in addition to influencing the characteristics of species richness in aquatic ecosystems (Adame et al., 2018; Romero et al., 2019).

Biofilms have become interesting in the eyes of science because they represent one of the pillars of the food chain in the environment and, therefore, constitute an extremely important part of the trophic levels (McKay & King, 2006; Alam et al., 2020; Bowen, 2022). They also act in compound biodegradation processes (Chen et al., 2022) and participate in the planet's biogeochemical processes, therefore, it is necessary to better understand how they work, the impacts to which they are subjected and the complex relationships between both sciences (Fernandes et al., 2020; Guo et al., 2020; Hou et al., 2022).

In view of the forecast of new environmental challenges arising from the effects of imminent climate change and, based on the scenario of research already carried out on biofilms formed in rivers and the influence of hydrology on these ecosystems (Palmer & Ruhi, 2019), the goal of this study is to verify the state of the art through a literature review, whose central theme was the influence of hydrological variables on river biofilms. This strategy highlights the techniques and contexts used for evaluation of biofilms and hydrological variables, as well as to understand the effects that such hydrological variables have on the dynamics of natural river biofilms and the perception of the literature in qualifying such beings as a possible environmental matrix for ecosystem diagnosis, expanding the range of possibilities for future research.

METHODOLOGY

The published literature on the relationship between hydrological variables and river biofilms, for this narrative review, was retrieved from the Scopus database, with peer-reviewed articles related to the topic being considered for evaluation.

For a better understanding of the information obtained on the subject presented, this review was fragmented into topics of interest, so that the surrounding context of the relationships between biofilms and hydrological variables can be better understood. Figure 1 illustrates, in an integrated manner, the approach and strategy of the review.

This review was then consolidated into topics that address the general characteristics of biofilms, especially from an environmental perspective. The existence of biofilms in natural environments, such as rivers, was then discussed, as well as the different global environments and strategies used by researchers to address hydrological variables in biofilm studies. Furthermore, the types of samplers used to form colonies, sampling strategies, incubation times or exposure of biofilms in rivers, and biofilm variables used for evaluation and comparison in relation to the environment were discussed. Finally, the relationships found between the characteristics of biofilms influenced by hydrological variables and the environmental consequences of these effects and relationships were discussed.

Table 1 presents the references of the most relevant aspects addressed in this narrative review, presented in temporal order of publication.

General characteristics of biofilms

The study of biofilms began in the 20th century and, since then, the ability of marine bacteria to adhere, survive and multiply on different surfaces has been observed, forming colonies as organic matter in the environment became available (Costerton et al., 1995).

With scientific evolution, biofilms were recognized as highly complex colonies, with a diverse internal composition and consisting of numerous classes of organisms in addition to bacteria, such as algae, fungi, protozoa, among others. These organisms produce an external layer of polymers that provides stability and mechanical integrity, facilitating adhesion to substrates and surfaces (Marshall et al., 1971; Huerta et al., 2016; Pu et al., 2019; Barral-Fraga et al., 2020; Fernandes et al., 2020; Valdés et al., 2021).

Figure 1. Integration of the review strategy. Natural river biofilm as the central object, surrounded by the elements with which it can be related: biofilm internal characteristics and heterogeneity (green circle on the right), exposure time of colonies in the environment (purple circle on the right), variety of sampler types such as stones, logs, artificial samplers (pink circle at the bottom), metrics related to biofilm characteristics such as weight, hydrological issues (green circle on the left), the different environments around the world, with different characteristics, in which biofilms can be found and evaluated (purple circle on the left) and the consequences for the environment (pink circle at the top).

Biofilms act in the sphere of major biogeochemical processes, carrying out the cycling of nutrients and carbon (Mai et al., 2020; Valdés et al., 2021; Hou et al., 2022; Zhou et al., 2024), being one of the predominant forms of biomass in rivers and, therefore, has gained notoriety in the scientific community (Fernandes et al., 2020).

The formation process of these colonies begins with the adhesion stage of microbial cells, followed by the increase and continuous growth from structural changes that occur inside (Sabater et al., 2007). Then, the colonies reach the maturation period and, finally, dispersion.

Internally, microorganisms, especially bacteria, release substances through excretion processes, which are the product of cell destruction and hydrolysis of macromolecules (Sheng et al., 2010; Fernandes et al., 2020). These substances make up the extracellular polymeric matrix and can be from a wide range of macromolecules such as polysaccharides, proteins, nucleic acids, lipid compounds and may or may not be in direct contact with the bacterial cells that produce the released substances, also being a source of carbon, nutrients and energy for the colony itself, in the absence of substrates (Sheng et al., 2010; Zhang et al., 2018; Karygianni et al., 2020).

This layer is responsible for adhesion, colony fixation and structural stability on different substrates, and also provides protection against predators, antimicrobial chemical agents and other variables that pose a risk to the integrity of the cluster (Costerton et al., 1995; Karygianni et al., 2020).

The increase in biomass occurs from the colonization by distinct classes of beings such as small animals, fungi, algae, bacteria, in addition to the organic and inorganic availability of the environment, while the losses of formed biomass can occur due to intrinsic and diffusive phenomena such as respiration, excretion or removal of organic matter (Moschini-Carlos et al., 2000). These processes are combined with external factors linked to the ecosystem where biofilms live, such as the quality of the environment, grazing and shear forces resulting from water flows (Casartelli et al., 2016).

Because they are highly resilient and demonstrate the ability of microbial communities to adapt, they are successful in occupying distinct habitats, becoming ubiquitous and establishing colonies with a high variety of beings in the formation of biomass (Costerton et al., 1995; Bowen, 2022). The ease of adaptation is also related to the ability of colonies to break down organic molecules and synthetic compounds, enabling the colonization of natural ecosystems in non-ideal and challenging quality conditions for the colony, also having characteristics of compound sorption as a response to the environment (Sheng et al., 2010; Borges et al., 2016; Valdés et al., 2021).

Table 1. References of the aspects covered in the review

Approach	Description	Related articles
General characteristics of biofilms	Main articles related to general information on biofilms such as composition and formation of the extracellular polymeric matrix, internal characteristics of biofilms and majority composition, formation processes and kinetics	Costerton et al. (1995), Marshall et al. (1971), Sabater et al. (2007), Sheng et al. (2010), Sabater (2017), Chaumet et al. (2019), Karygianni et al. (2020)
Internal diversity of biofilm colonies	Main articles presenting information on the taxonomic composition of colonies in different water resources	Andrus et al. (2013), Villeneuve et al. (2011), Adame et al. (2018), Borges et al. (2016), Zhang et al. (2018), Pu et al. (2019), Qu et al. (2019), Smeti et al. (2019), Alam et al. (2020), B-Béres et al (2022), Pan et al. (2020), Bagnoud et al. (2020), Bighiu et al. (2020), Mai et al. (2020), Pan et al. (2020), Ramoneda et al. (2021), Vlaičević et al. (2021), Bondar-Kunze et al. (2021), Vincent et al. (2022), Vlaičević et al. (2022), Bagra et al. (2023), Gionchetta et al. (2023), Lin et al. (2020), Madge Pimentel et al. (2024), Zhou et al. (2024)
Biomass assessment	Main articles evaluating biofilm biomass of various water resources as a metric	Moschini-Carlos et al. (2000), Robinson et al. (2004), McKay & King. (2006), Leandrini et al. (2008), Villeneuve et al. (2011), Proia et al. (2013), Andrus et al. (2013), Osorio et al. (2015), Arroita et al. (2016), Casartelli et al. (2016), Smeti et al. (2019), Chaumet et al. (2019), Romero et al., (2019), Colls et al. (2019), Salis et al. (2019), Skovsholt et al. (2020), Myrstener et al. (2020), Guo et al. (2020), Pan et al. (2020), Bondar-Kunze et al. (2021), Huang et al. (2021), Pereda et al. (2021), Valdés et al. (2021), Vlaičević et al. (2021), Vlaičević et al. (2022), Weitere et al. (2021), Vincent et al. (2022), Bertrans-Tubau et al. (2023), Gionchetta et al. (2023)
General effects of high river flows on biofilms	Main articles that present as their scope the effects of high flows on the characteristics of natural biofilms, formed and collected in rivers	Moschini-Carlos et al. (2000), Robinson et al. (2004), McKay & King. (2006), Leandrini et al. (2008), Romaní et al. (2008), Villeneuve et al. (2011), Osorio et al. (2015), Andrus et al. (2013), Arroita et al. (2016), Colls et al. (2019), Qu et al. (2019), Salis et al. (2019), Smeti et al. (2019), Alam et al. (2020), Guo et al. (2020), Myrstener et al. (2020), Skovsholt et al. (2020), Bondar-Kunze et al. (2021), Pereda et al. (2021) Valdés et al. (2021), Huang et al. (2021), Ramoneda et al. (2021), Weitere et al. (2021), Coulson et al. (2022), B-Béres et al. (2022); Gionchetta et al. (2023), Bertrans-Tubau et al. (2023), Madge Pimentel et al. (2024), Mai et al. (2020), Zhou et al. (2024)
Effects of water pollution on biofilms	Main articles presenting pollution relationships between polluted water resources and natural biofilms	Romaní et al. (2008), Villeneuve et al. (2011), Andrus et al. (2013), Proia et al. (2013), Osorio et al. (2015), Borges et al. (2016), Huerta et al. (2016), Aubertheau et al. (2016), Chaumet et al. (2019), Pu et al. (2019), Qu et al. (2019), Romero et al. (2019), Salis et al. (2019), Smeti et al. (2019), Alam et al. (2020), Bighiu et al. (2020), Guo et al., (2020), Mai et al. (2020), Pereda et al. (2021), Weitere et al. (2021) Ramoneda et al. (2021), Valdés et al (2021), B-Béres et al. (2022), Coulson et al. (2022), Vincent et al (2022), Cui et al. (2023), Bertrans-Tubau et al. (2023), Gionchetta et al. (2023), Madge-Pimentel et al. (2024), Zhou et al. (2024).

Internal complexity allows biofilms to act not only as a community, but also as a place where several micro-niches of beings can coexist, cooperate or compete simultaneously for survival, given their internal processes, such as the possibility of predominance in the autotrophic or heterotrophic characteristics of colonies (Sabater, 2017; Barral-Fraga et al., 2020). This heterogeneity of beings plays different roles within the collective and allows distinct responses to external stressors (Sabater, 2017).

In this sense, the colonization and maintenance of biofilms on natural or artificial substrates also come down to a relationship with a series of external variables that act together. This is the case of hydrological variables such as ambient temperature, light incidence, current flow and hydrodynamics, which permeate river stretches and interfere with water quality (Moschini-Carlos et al., 2000; Casartelli et al., 2016). This scenario has raised curiosity about the possibility of using biofilms as a biological and chemical marker for diagnostics and monitoring (Cui et al., 2023) of aquatic ecosystems.

Biofilms in natural water bodies

Resilience allows biofilms to adapt to water resources with the most diverse hydrological, climatological and topographic characteristics. Several natural environments have been explored considering the responses of biofilms to hydrological stressors in the environment. In this regard, river dam regions (Robinson et al., 2004), rivers and streams in forested and mountainous basins (McKay & King, 2006; Arroita et al., 2016; Zhou et al., 2024), or subjected to agricultural impacts (Villeneuve et al., 2011; Andrus et al., 2013), as well as temporary or intermittent streams (Colls et al., 2019; Coulson et al., 2022), floodplain regions related to large rivers (Leandrini et al., 2008; Guo et al., 2020) and urban rivers (Osorio et al., 2015; Cui et al., 2023; Madge Pimentel et al., 2024), are some examples of environments that can be evaluated.

Of the total number of articles in this narrative review, 30 specifically addressed the relationships between hydrological variables and biofilms in rivers, of which 57% covered the European region, 17% the Americas and 13% the Asian region. The other 13% were divided between Africa, Antarctica and Oceania. The remaining articles covered lakes, reservoirs, artificial environments and literature reviews.

From this same perspective, the climatic differences across the globe were also not an impediment to the evaluation of biofilms. A diverse range of regions with distinct climatic varieties such as temperate (Robinson et al., 2004; Guo et al., 2020; B-Béres et al., 2022; Coulson et al., 2022), tropical (Zhou et al., 2024); subtropical (Leandrini et al., 2008), arctic (Skovsholt et al., 2020) and Mediterranean (Osorio et al., 2015; Colls et al., 2019) were evaluated and found to be capable of promoting the formation of these individuals in natural environments, regardless of the differences and particularities.

Arctic and subarctic climates, covered in two articles in this collection, mainly addressed issues related to the melting of glacier areas and the changes in hydrological dynamics caused by this, as well as nutrients, seasonality in the ecosystem and primary production (biofilms). Since these are regions where, historically, extreme cold predominates for most of the year, the temperature increases experienced today have generated concern due to predictions associated with imminent climate change. This same alert was given in a polar climate region, assessed in one of the articles considered in this review, whose concern was also shown regarding the patterns of connectivity between hydrological systems, due to the increase in water levels due to the melting of glaciers.

Mediterranean climate regions were the subject of seven articles, whose approaches were divided between evaluating, in addition to perennial rivers, events and locations of water scarcity and intermittency, as well as issues related to pollution levels and hydrological stress in the characteristics of river biodiversity, especially in biofilms.

In three articles conducted in a tropical region, it was noted that the main objectives were to verify seasonal patterns and those related to the characteristics of biofilms and the hydrological patterns characteristic of each environment, that is, to understand the functioning of biofilms in real environments.

Three articles were conducted in a subtropical region, and in this region, the approaches varied between verifying the effects of hydrology and pollution on biofilms, as well as understanding biofilm behavior in real environments, with their structural and physiological characteristics.

Finally, the last region evaluated was the temperate climate, which had fourteen articles, being the largest group in this segregation. This region made efforts to evaluate the responses of biofilms to hydrological stressors such as intermittent and perennial locations, occasionally extreme conditions (droughts and floods) and, in some cases, associated with the presence of pollutants. The most robust literature is concentrated in the European region, however, there are multiple efforts around the world to understand the characteristics and functioning of natural biofilms in water resources, especially in rivers, under hydrological effects. It is also clear that issues associated with hydrology, such as the dispersion of pollutants, are also on the radar of science, especially due to the continuous increase in pollutant levels and the various new pollutants introduced into the environment, an issue that will be addressed later in this article.

In addition, issues related to climate crises involving droughts, floods and glacier melting have gained notoriety in the literature given the extreme events that several areas of the world are facing and will probably face more frequently in the future.

Furthermore, the resilience of the biofilm allows the adhesion and formation of colonies in environments where human action alters the natural conditions of aquatic environments, whether by changing flows such as damming, capture, flooding (Robinson et al., 2004; McKay & King, 2006; Leandrini et al., 2008; Arroita et al., 2016; Adame et al., 2018; Bondar-Kunze et al., 2021), changes in temperature and climate change (Bondar-Kunze et al., 2021; Ramoneda et al., 2021; Vlaičević et al., 2022), or even changes in the physical-chemical characteristics of water (Andrus et al., 2013; Osorio et al., 2015; Pereda et al., 2021), especially linked to effluent discharges and consequent environmental pollution.

Additionally, as these are water bodies and, therefore, highly complex ecosystems, the literature was not limited to considering only biofilms as a comparison in the scope of projects and, therefore, other matrices were incorporated into many evaluations, as a way of expanding the understanding of the environmental system and scientific advancement.

In this scenario, in addition to the biofilm as the central object of research and observation, the condition of the water in which the periphytic community resides has proven to be of great value, being an adjacent object of investigation in many of the studies, considering several parameters as complementary interferences. Physicochemical variables, especially dissolved oxygen, nutrients (mainly nitrogen and phosphorus) and organic carbon, also to microcontaminants were recurrent in several studies (McKay & King, 2006; Osorio et al., 2015; Arroita et al., 2016; Smeti et al., 2019; Pereda et al., 2021; B-Béres et al., 2022; Cui et al., 2023; Zhou et al., 2024).

The literature also points to the joint consideration of other classes of living beings in research contexts, such as macro and microinvertebrates (Robinson et al., 2004; McKay & King, 2006; Andrus et al., 2013; Qu et al., 2019; Cui et al., 2023), vertebrates, such as fish (Qu et al., 2019; Bowen, 2022), as well as solid matrices, such as sediment (Robinson et al., 2004; Bagnoud et al., 2020; Mai et al., 2020).

As with water, the qualification of these matrices acts as a possibility to be analyzed and correlated both for understanding the environment and for understanding the behavior and characteristics of biofilms.

Use of hydrological variables in biofilm studies

Biofilm responses to the ecosystem are evaluated in a variety of ways in the available literature, especially by attempting to evaluate the effect of an isolated hydrological variable on colony functions.

There are several hydrological variables that act synergistically in a water environment, and each one can influence the ecosystem in a particular way, both positively and negatively.

Changes in flow, discharges and water levels, for example, can alter the availability of nutrients, as verified by Adame et al. (2018) in floodplain lakes in Brazil and Alam et al. (2020) in gravel streams in Japan, facilitating the formation and maintenance of biofilms, or hindering them, depending on the concentrations available.

High flows, caused by pulses or extreme flood events, bring with them the transport of materials, which can elevate suspended solids and, therefore, vary the turbidity, luminosity of the water body and the availability of structural elements for biofilms, also verified by Adame et al. (2018) and Alam et al. (2020). Scenarios such as this facilitate drag effects due to shear forces, as observed by Osorio et al. (2015) in rivers in Catalonia, and Gionchetta et al. (2023), in rivers influenced by treatment plants in Switzerland. Such an increased flow context culminates in the generation of turbulence and structural changes in colonies.

On the other hand, low flows, caused by water abstraction or prolonged drought events, can reduce the wetted perimeter of rivers and, therefore, hinder the quantity of habitats favorable to the formation and survival of colonies, as experienced in mountain rivers and streams by Arroita et al. (2016) or by B-Béres et al. (2022), in Spain and Hungary, respectively, or in streams in France, verified by Bertrans-Tubau et al. (2023). Furthermore, reducing flow rates to minimum levels can hinder or interrupt hydrological connectivity between environments, and thus harm nutrient cycling and water quality after dry stretches (Arroita et al., 2016).

Therefore, changes in flow have become an object of interest to understand the behavior of biofilms in real environments, and the way to obtain data or perform measurements of this variable has presented itself in a variety of ways.

From a hydrological perspective, Robinson et al. (2004) investigated the impact of controlled flows in experimental floods. In contrast, other studies have looked at the opposite scenario, analyzing the effects of manipulated droughts on water resources (McKay & King, 2006; Arroita et al., 2016; Coulson et al., 2022).

The actual hydrology of the site, particularly in relation to water levels, flow velocities and discharges, can be used as comparison parameters without the need to manipulate these variables. To this end, these variables are measured in real time, as a result of the natural processes of the environment, and may or may not represent an extreme event (Leandrini et al., 2008; Osorio et al., 2015; Guo et al., 2020).

Furthermore, there is no standard methodological protocol for measuring flow and fluxes, and in this case, numerous methods are available. Robinson et al. (2004), for example, used monthly parameter measurements with subsequent conversion into flow by saline dilution curve for one of the study sites, as well as hydrological data provided by local agencies for the other area. Flow is also measured using parameters such as cross-section, velocities and water level of the studied locations, obtained by calibrated equipment, and may or may not undergo mathematical modeling (McKay & King, 2006; Andrus et al., 2013; Colls et al., 2019; Qu et al., 2019; Skovsholt et al., 2020). In addition, the use of time-conductivity curves adjusted to mass balance has been reported (Arroita et al., 2016), as well as flows obtained from databases of specialized or governmental agencies (Osorio et al., 2015; Alam et al., 2020; Guo et al., 2020).

Other hydrological variables are also considered in the literature when it comes to studies on the response of river biofilms to hydrological factors, although with less emphasis.

Temperature, for example, affects the maintenance characteristics of biofilms, and depending on the variation, it can change, which can cause variations in the characteristics of the organisms inside the biofilms, as well as in the growth and metabolism of the colonies (Romero et al., 2019). It is one of the variables that guarantee similar environmental conditions in comparative studies (Romaní et al., 2008).

Higher temperatures tend to stress aquatic organisms and increase the bacterial and heterotrophic capacity of colonies (Palmer & Ruhi, 2019; Romero et al., 2019), and also generate resistant colonies, becoming a warning factor, especially in shallow areas and with water stagnation that can reach high temperatures depending on the time of year, which tend to be aggravated in the future (Gionchetta et al., 2023). In experiments carried out in real environments, temperature is used more as a variable to characterize the environment, or to inform the reader about the experimental context, rather than as a discussion piece of data. However, in studies involving areas of ice melt, such as that of Myrstener et al. (2020) in Sweden, or that of Skovsholt et al. (2020) in Greenland, or that of Ramoneda et al. (2021) in Antarctica, it is understood that rising temperatures are a high-impact factor, even if the data are not explicit.

Like temperature, precipitation ends up being a variable considered indirectly through its influence on other variables, especially flows (Palmer & Ruhi, 2019).

Dimensions of the water body (Robinson et al., 2004), measurements of the water level and its variation in different locations (McKay & King, 2006; Leandrini et al., 2008; Arroita et al., 2016; Colls et al., 2019), air or water temperature (Robinson et al., 2004; Leandrini et al., 2008; Arroita et al., 2016; Colls et al., 2019), precipitation (Andrus et al., 2013) and measurements specifically related to water parameters as previously explained.

In this context, flow (also referred to as fluxes or discharge) emerges as the most frequently analyzed variable in studies investigating biofilm responses to hydrological pressures. The other variables, for the most part, act as complementary information, refining the intended diagnosis.

In this sense, it is clear that there is a lack of studies in the literature focused on evaluating, in a natural environment, such variables in greater detail and, possibly, in isolation, especially precipitation and water and air temperature.

A critical analysis of the use of these variables will be presented in greater depth in the section: Effects of hydrological variables on river biofilms.

Samplers and sampling

Biofilms develop on surfaces with different internal characteristics and different degrees of rigidity, such as stones, wood, leaf litter, plants or even sediment, as long as the environment is suitable for diffusion processes, favors adhesion and ensures the fixation and maintenance of colonies (Romaní et al., 2008; Bowen, 2022; Bertrans-Tubau et al., 2023).

Biofilm samplers (also known as incubators, substrates or surfaces to which biofilms are adhered and will be sampled) are presented in the literature as natural or artificial surfaces (Figure 2).

Natural samplers occur with the use of an element from the site itself, such as the biofilm formed on the petiole of macrophytes located in rivers (Leandrini et al., 2008; B-Béres et al., 2022), biofilms located in the sediment (Coulson et al., 2022) or branches (B-Béres et al., 2022).

However, the use of more rigid structures with an area that can be estimated from the environment itself, such as rocks and river stones or boulders, containing biofilm adhered to the surface, is common and recurrent (Robinson et al., 2004; McKay & King, 2006; Villeneuve et al., 2011; Arroita et al., 2016; Colls et al., 2019; Qu et al., 2019; Bighiu et al., 2020; Myrstener et al., 2020; Skovsholt et al., 2020; B-Béres et al., 2022; Zhou et al., 2024).

Under previously adapted and prepared structures with the specific purpose of biofilm adhesion, the artificial samplers and the way of fixing these artifacts come in the most diverse shapes, sizes and materials, such as glass tiles fixed to methacrylate supports attached to the banks (Osorio et al., 2015), steel structures with slate tiles fixed to floats (Andrus et al., 2013), ceramic tiles or tiles (Lin et al., 2020; Myrstener et al., 2020), structures of plastic cups with agar wrapped in glass discs (Guo et al., 2020), glass plates fixed to concrete (Bagra et al., 2023; Bertrans-Tubau et al., 2023), plastic support tied to slope structures (Pereda et al., 2021), glass slides (Vlaičević et al., 2022), and others.

The variety of substrates evaluated allows biofilm formation to be viable under any condition, with the choice of substrate being based on the objectives of the work and the structural condition of the groups that will perform the sampling. In general, the difference between artificial and natural samplers mainly involves the known and precise area of biofilm adhesion and the possibility of controlling the zero mark of the biofilm incubation period (Moschini-Carlos et al., 2000).

In this sense, the possibility of measuring surfaces and verifying the known and precise area, which can be related to biomass formation, exposure time and measured hydrological variables, such as flow. From these relationships, it becomes possible to create measurement units that facilitate comparison between different events and studies, becoming an advantage, depending on the diagnostic objectives.

Figure 2. Sampler examples. A and B represent natural samplers, while C represents an artificial sampler. A - Piece of tree trunk with natural biofilms on top (wood). B – Stone full os natural biofilm. C - wooden box with internal glass sheets, to be submerged in the river, for biofilm adhesion.

The removal of biofilms from their respective substrates is also an item without fixed methodological protocols in the scientific literature, however, the most recurrent process is the removal of biofilm biomass by scraping with the most diverse materials or even brushing the place where they are adhered to a new prepared container. Such a process may or may not include washing with water (Robinson et al., 2004; McKay & King, 2006; Qu et al., 2019; Bighiu et al., 2020; Guo et al., 2020; Lin et al., 2020; Myrstener et al., 2020; Skovsholt et al., 2020; B-Béres et al., 2022; Vlaičević et al., 2022; Madge Pimentel et al., 2024), use of ultrasound and solvent extraction (Osorio et al., 2015; Pereda et al., 2021).

Seasonality issues can be observed mainly from the perspective of comparing and diagnosing environments, and therefore it is necessary to evaluate the inclusion of this variable in sampling protocols. Depending on the geographic region, the seasons in the same place are determined by very different climatic conditions in terms of flows, rainfall patterns, and temperatures. As mentioned in the previous sections, biofilms are susceptible to such variations and may have different internal characteristics depending on local conditions, potentially influencing the entire biota, since they start the food chain. Furthermore, it is easier to compare different studies in naturally contrasting regions such as Permafrost regions, explored by Myrstener et al. (2020), which, according to the authors, have average annual temperatures of -1°, while in China, Qu et al. (2019) report average annual temperatures of 9°. In Brazil, even the low temperatures experienced by Moschini-Carlos et al. (2000), in the Alto Paraná region, probably had a much shorter duration, compared to other situations mentioned, and, in other seasons, high temperature levels.

Therefore, the standardization of protocols and the inclusion of seasonal factors would allow the comparison of similar scopes, and, in studies involving the evaluation of seasonal periods in a location, it could be a factor for improving and deepening the understanding of the behavior of biofilms in environments influenced by hydrology.

Incubation time

When using samplers/natural substrates in situ (macrophyte biofilms, bottom stones or similar), it is not always possible to estimate the initial incubation period and the consequent exact time of colony formation. However, it is common for these assessments to report the sampling frequency and the criteria chosen for comparison.

The frequency between biofilm samplings is another variable with diverse methodological spectra, with fortnightly intervals reported (McKay & King, 2006; Arroita et al., 2016), four to five weeks (Robinson et al., 2004), or even considering each hydrological or seasonal period (Leandrini et al., 2008).

Studies that adopted artificial samplers followed a different dynamic. Andrus et al. (2013), for example, kept the biofilms incubated for a month before the first collection and monitored weekly collections, while Osorio et al. (2015) performed the first sampling after 8 and 22 days of incubation and, after transplanting the sampler to another location, sampling occurred approximately at 2 and 9 days of the new incubation. Guo et al. (2020) and Vlaičević et al. (2022) performed sampling with an average incubation time of 4 weeks.

Just like the aspect of biofilm adhesion substrates, incubation and sampling periods vary greatly, depending on the intended objective in each project scope, related not only to the biofilm, but also to what is desired in relation to hydrological variables.

Casartelli et al. (2016) report that colonization time can be an important factor in increasing biomass and forming river biofilm; however, biomass accumulation can be influenced by seasonal variation in terms of internal composition heterogeneity and maximum accumulation rate. It should therefore be considered that, since these are assessments carried out in different regions of the globe, issues of seasonality, climate, biome, economic and cultural factors can guide decisions regarding the temporality of the research and its execution dynamics, possibly justifying the variation, since protocols on the subject are still scarce.

Metrics in biofilms studies

The studies in this review show a wide variety of possible approaches to assess biofilms. Characteristics and responses of biofilms are observed and measured to achieve the specific objectives related to each proposed scope, whether it is to assess the biofilm itself or to use the biofilm as a metric to assess the environment.

The variables evaluated related to the biofilm are closely linked to the specific objectives of each project; however, the biomass formed by the biofilm ends up being the most frequent characteristic. This biomass is usually evaluated based on the analyses and calculations of chlorophyll-a and b (autotrophic and heterotrophic biomass), dry weight and ash-free dry weight (total biomass) (Robinson et al., 2004; McKay & King, 2006; Leandrini et al., 2008; Andrus et al., 2013; Osorio et al., 2015; Arroita et al., 2016; Colls et al., 2019; Guo et al., 2020; Myrstener et al., 2020; Vlaičević et al., 2022; Cui et al., 2023).

Biomass is a quantity related to the process of formation and development of colonies, being a metric that provides information about growth, productivity, quantity of living or dead materials inside them, and the integrity of colony structures (Robinson et al., 2004; Proia et al., 2013; Guo et al., 2020; Bertrans-Tubau et al., 2023). It can also be used to evaluate the photosynthetic activity of the biofilm colony, when estimated by means of chlorophyll-a, indicating the presence and abundance of primary producer and autotrophic organisms, as experienced by Salis et al. (2019), Romero et al. (2019) and Valdés et al. (2021). Several of the studies in this review used this metric as a reference parameter and comparison with hydrological effects, for example, to verify the effects of colony detachment due to strong flows or flooding events, as observed by Robinson et al. (2004) and Andrus et al. (2013).

Or, used as a parameter to understand the inverse effect, of drying effects on the water environment, as observed by McKay & King (2006), Arroita et al. (2016) and Colls et al. (2019) in streams characterized by intermittent flows or by water capture processes from their courses.

Furthermore, biomass can also be used as a parameter to evaluate the effects of different hydrological periods of connected environments, as carried out by Leandrini et al. (2008), who evaluated several aquatic environments in a Brazilian river basin. Or, to evaluate translocation effects between aquatic environments, as seen by Osorio et al. (2015), in rivers with different pollution characteristics.

Additionally, other assessments focused on the internal processes and characteristics of biofilms, such as productivity, cellular and bacterial variability, extracellular enzymatic activity, exoenzymatic activity, taxonomy, assessments focused on the metabolism and functionalities of biofilms, are also usually considered (Villeneuve et al., 2011; Arroita et al., 2016; Colls et al., 2019; Guo et al., 2020; Myrstener et al., 2020; Coulson et al., 2022; Bertrans-Tubau et al., 2023; Madge Pimentel et al., 2024).

In addition to biomass as one of the relevant metrics for biofilm studies, a large number of studies were observed that evaluated species richness, as well as the composition of biofilm communities as metrics of great importance. These metrics allow visualizing the heterogeneity in time and space evaluated, as well as which species exist and predominate in the evaluated biofilm, as performed by Madge Pimentel et al. (2024) and Andrus et al. (2013), as well as which are resistant to hydrological events, as also verified by Andrus et al. (2013). Furthermore, this metric facilitates the understanding of the ecosystem function of the colony and can also be a good mirror for mapping pollutants in the environment, as suggested by Valdés et al. (2021).

As mentioned, to a lesser extent, metrics involving specific biofilm performances were also present in the scope of articles in this review.

Measuring enzymatic activity can help understand the capacity of biofilm bacteria to process organic matter in a given environment, as assessed by Weitere et al. (2021), or phosphorus and cellulose, as seen by Arroita et al. (2016). It can also be used to measure the microbial response to bed drying processes in intermittent regions, as experienced by Coulson et al. (2022).

In this same sense, cellular respiration was assessed by Vincent et al. (2022), Colls et al. (2019), Bertrans-Tubau et al. (2023), and others. This variable allows the evaluation of metabolic characteristics of the biofilm, aiding in the diagnosis of assessing the health of the water resource and colonies, and allowing the comparison between responses after hydrological events, especially flows. Ramoneda et al. (2021) also proposes that techniques involving transcription and evaluation of genetic materials, such as transcriptomics and metagenomics, can help to understand colonies in greater depth, as well as the effects of water pollution on biofilms.

It was found that there is no standardization regarding which metrics related to biofilms should be used in the studies of this review. However, it is clear that because these are highly complex matrices, the greater amount of information seems to offer greater possibilities for diagnostic approaches, although it is common knowledge that the analytical approach depends on several factors to be defined and, eventually, it is not possible to contemplate so many fronts of observation.

Therefore, it is clear that the measurement of biomass through total weight, as well as the evaluation of chlorophyll-a, deserve to be evaluated, including as two distinct metrics, since they can offer additional information when operationalized in this way. Together, the studies demonstrated great value in measuring the richness and composition of species within biofilms, being quite recurrent.

Effects of hydrological variables on river biofilms

As explained in the previous sections, the most common metrics for assessing biofilm response to hydrological stresses are related to the biomass and internal composition of colonies, while hydrological stresses are mostly assessed by fluctuations, intermittency or extreme events related to water flows. The discussions will be guided mainly from this perspective, without, however, considering the importance of other relevant metrics in each study.

The microbiota present in water resources is subject to several sudden variations in discharges and flows that alter their mechanisms depending on the changes caused in the systems (Palmer & Ruhi, 2019).

The response of biofilms to extreme hydrological events, especially floods or water shortages, simulated or real, was evaluated in rivers from different parts of the world with diverse hydrological, climatic, land use and occupation, and cultural conditions.

Robinson et al. (2004) tested flood events of different magnitudes in the border region between Switzerland and Italy and observed that the volume of biomass (chlorophyll-a and b, and ash-free dry mass) of biofilms decreased, making it difficult to recover this biomass after successive flood events. According to the authors, the losses may be a reflection of the shear forces of the flows, caused by changes in water velocity during floods, which lead to an effect of alteration and cleaning of the colonies.

However, the increase in biomass, although brief and subtle, was observed in the opposite hydrological context of water restriction. McKay & King (2006) evaluated the responses of ecosystems to different experimental water extraction events in Australia and found trends in biofilm biomass values (also measured in dry mass and ash-free dry mass and chlorophyll-a content) slightly increased in stretches with reduced flow. For the authors, the slight increase in biomass in the diversion areas in this region is related to the decrease in the quantity and availability of wetlands, the depth and flow of streams that the manipulation of flows generated.

In contrast to McKay & King (2006), Arroita et al. (2016) and Colls et al. (2019) noted that the water extraction process and, consequently, the reduction in river flows, generated a response of reduction in the biomass of biofilms formed in the regions. Drought or intermittent events can alter metabolic issues of biofilms, especially the internal phosphorus utilization capacity of the periphytic community and the rate of nutrient absorption in catchment areas (Arroita et al., 2016), modifying the physiological dynamics that make up the colony growth process.

In places where flows are very low, the increase in biomass may be related to the fact that there is no favoritism to the detachment of colonies in the total biomass of biofilm from the substrates and, depending on the availability of resources in the environment (McKay & King, 2006). On the other hand, in the same hydrological context of scarcity, the reduction in the biomass of biofilm colonies may be a process resulting from the contraction of the wet channel and changes in the morphology of the channel due to the captures made, which can transform the external layers of the cluster, hindering the internal transport of nutrients and, therefore, reducing the density of the biomass (Arroita et al., 2016).

In these cases, both the total and autotrophic biomass of biofilms are more responsive to the duration and severity of the event than to the frequency of the event itself (Colls et al., 2019; Coulson et al., 2022). In other words, the duration of the drought together with the variables of light incidence and temperature can reduce the biomass of biofilms and also alter their metabolic characteristics, such as a decrease in photosynthetic efficiency. This finding is reinforced by Bertrans-Tubau et al. (2023) who found, in addition to the loss of biomass, also a reduction in the density of the extracellular polymeric layer and microalgal density, with the temporal extension of droughts.

Nevertheless, B-Béres et al. (2022), when evaluating diatoms within biofilms of intermittent rivers, and Madge Pimentel et al. (2024), when verifying the functions of biofilms in the face of pulses of flow and salinity, realized that drought alone was not enough to alter the internal biodiversity of the colonies. Part of this may be related to adaptation strategies of the algae themselves, since diatoms tend to be resistant and adaptable, also, in turbulent and polluted environments (Smeti et al., 2019; Pan et al., 2020). In the case of Madge Pimentel et al. (2024), the parasitic community residing in biofilms showed an increase in diversity with the reduction of flows. However, as mentioned above, it is reinforced by the authors that the duration of the event may be a relevant point, since the longer the duration, the lower the self-protection capacity of the diatom assemblages (B-Béres et al., 2022).

In intermittent channels, despite drought events with consequences for the biota, rewetting the environment may represent a possibility for ecological recovery. This process can bring new functionality to the environment, depending on the flow rates, the conditions of deposition of organic material and the species most resistant to extreme events, as well as their functional readaptation capacities (Lin et al., 2020; B-Béres et al., 2022; Coulson et al., 2022; Huang et al., 2022).

Osorio et al. (2015), evaluating flood events, noted that biofilms presented greater autotrophic biomass, as well as greater heterotrophic activity (extracellular enzymatic activity) and a higher rate of biofilm accumulation at lower flow rates, while high flow events act in the opposite way. Likewise, high flows generated lower species richness, uniformity and diversity, corroborating the findings of Robinson et al. (2004), McKay & King (2006), Andrus et al. (2013) and Guo et al. (2020).

In places with dams, for example, during periods when water levels were below the regional average, the likely influence of the upstream dam is the promotion of peaks in water level elevations that favor the increase in nutrients, enriching the ecosystem and influencing the responses of biofilms and, in this case, the increase in flows may culminate in an increase in biomass density due to the increased availability of resources of interest to the colony (Leandrini et al., 2008).

Adame et al. (2018) evaluated lakes and ponds and found that water level modulation, combined with seasonal factors such as seasonal temperatures, can directly affect the internal composition of species in biofilm clusters. In some cases, higher water levels resulted in greater species diversity, but the complexity of the environment allowed isolated peaks in taxonomic variety, even at low water levels, justified by external factors such as the entry of nutrients and propagules in campaigns.

In this sense, biofilms formed in different types of habitats (lentic, lotic, connected or not to the main rivers) also respond in different ways to internal and external stressors, and, in addition to variations in the water level of the channel, there is a relationship with the degree of connection with the main rivers (Leandrini et al., 2008; Vlaičević et al., 2021), which influence the hydrology of these locations. Additionally, the internal heterogeneity of the clusters and biofilm grazing issues are also relevant in the variations of scenarios when observed from the perspective of increasing or reducing biomass (McKay & King, 2006), as well as the distribution of nutrients, distinct classes of chemical compounds in the water and seasonality (Salis et al., 2019; Guo et al., 2020).

Seasonal factors, from the change of season, can alter the degrees of connectivity between environments, often resulting in the loss of periphytic biomass, especially due to the decrease in the incidence of light in the environment and the reduction of nutrients, which are used by surface vegetation, along with the change in the taxonomic diversity of beings, such as ciliates, since hydrological variations modify the food structure of lake communities (Vlaičević et al., 2021).

In this scenario, it is clear that large and prolonged hydrological stresses in water bodies consequently generate major disturbances in the ecosystem, severely altering the natural functions of the environment, even with the great hydrological variability (Guo et al., 2020; Pereda et al., 2021; Coulson et al., 2022).

Along with the more evident relationship between variations in flow and biofilm density reported in the literature, there are also changes in the structure of the water environment, due to the action of water flows, acting as a secondary effect on the dynamics and characteristics of the biofilm.

Changes in channel morphology due to major floods can alter the dynamics of sediment transport, which, through erosion, fills pools and generates the movement of stones, which function as sample substrates for the incubation of biofilms and the biota itself (Robinson et al., 2004). In estuarine regions, for example, peak flows from adjacent rivers can alter the hydrological pattern of the estuary, together with soil dynamics, conductivity and salinity, influencing and modifying the bacterial community of the biofilm (Mai et al., 2020).

In dry regions, the narrowing of the channel, the decrease in the perimeter of the wetland and the depth and water level of the stream, in addition to the reduced flow rate, may be related to the response of the biota, especially with the abundance of biofilm biomass, the interference in the distribution and absorption of nutrients, and the variety of macroinvertebrates and herbivores (McKay & King, 2006; Arroita et al., 2016).

In general, flows, fluxes and water levels were the most recurrent hydrological variables when related to the response of the biofilm community, while biomass was the most recurrent biofilm-related variable. Hydrological variations in natural environments, in this context, generated a spectrum of scenarios, depending on the location, the context of the environment and the approach evaluated in each diagnosis.

Thus, it is noted that there are numerous results obtained in the literature on the effects of hydrological variables on biofilms. Although much of the literature points to inverse relationships between water flows in rivers and biofilm characteristics, such as abundance, biomass density and colony heterogeneity, opposite scenarios have also been verified, which reinforces the findings that the complexity of the environment, above all, drives many of the behaviors and responses of the relationship between biotic and abiotic factors (Palmer & Ruhi, 2019; Qu et al., 2019; Guo et al., 2020).

It was also clear that extreme and prolonged events seem to have more significant and more apparent effects on the periphytic community, at different scales, which integrate structural and functional issues of these colonies.

Therefore, there is a need to develop protocols and standardization in the context of project design, scope, execution of field work and analysis, in addition to deepening and expanding studies with an integrated approach, so that it is possible to understand the effects of variables in natural environments since they act in synergy.

Environmental consequences

Along with the degrading water quality, the pollution pose a risk to the environment, reaching the biological sphere and impacting plants, animals and microbial communities.

Within biofilm colonies, there are differences in the levels of resistance and stability to environmental impacts in benthic algae families, depending on the internal variety of organisms and classes (Qu et al., 2019; Mai et al., 2020). Thus, both pollution levels and hydrological events can interfere with the heterogeneity and predominance of species in communities (Mai et al., 2020; Vlaičević et al., 2022), keeping the beings more adapted to the environmental conditions.

The reduction in river flow can result in community and functional changes in the biofilm, as observed by Madge Pimentel et al. (2024), who evaluated a stream in Germany. They observed a dominance of algae, especially diatoms (Bacillariophyta), and, to a lesser extent, green algae (Ulotrichales) and golden algae (Chrysophyceae). The study also evaluated parasites, which were the most varied community, with the families Oomycota, Amphifilida and Chytridiomycotina being the most diverse groups. In this case, it was observed that the reduction in flows altered the dynamics of eukaryotic algae, with filamentous green algae becoming the dominant ones, as they were less susceptible to shear stress.

A study carried out by Adame et al. (2018), for example, found that seasonal flows in the Alto Paraná region in Brazil, characterized by flood pulses, showed that diatoms present in the biofilm were predominant under different hydrological conditions, as well as different seasonalities, due to their high resilience, ability to adapt to changes in the environment and the ability to produce mucilage and facilitate adhesion to different substrates, under different circumstances.

In contrast, Andrus et al. (2013) found that high flows promoted a washing effect on the biofilm colonies evaluated, causing a decline in the volume of diatoms in the biofilm. However, one group that was resilient to changes in flow was the cyanobacteria, which did not show a significant reduction in volume.

The study by Andrus et al. (2013), however, found this same resilience response of diatoms, but related to pollution of the environment, and not so much to the effect of flows. In this sense, these algae were predominant even in environments with higher levels of pollution caused by high concentrations of phosphorus, nitrogen and solids, and it was also found that biofilms that presented a predominance of these algae were less sensitive to exposure to the pollutant element of greatest concern, atrazine.

The overall characteristics of a site, resulting from hydrological action and surrounding activities, significantly influence the composition of biofilms. Vincent et al. (2022) found that the taxonomic composition of colonies commonly found in mountainous areas differs from that of colonies formed in urban rivers. Biofilms from mountainous areas tend to have more active taxa, while biofilms from urbanized areas were composed mainly of the bacteria Comamonadaceae, Rhodobacteraceae, Sphingomonadaceae, Rhizobiales, and Burkholderiales; the fungi Chytridiomycota, which were more common among the sites evaluated, although the largest quantity in all sites were of fungi that could not be classified; and the algae Bacillariophyt, Bacillariophyceae (diatoms), and eukaryotic algae. Diatom algae predominated in almost all sampling sites, corroborating the studies previously cited.

The biofilms formed in different urban areas presented relatively homogeneous data, suggesting that, despite being different, the hydrological impacts of urbanization, such as increased runoff, together with the effect of high pollution, generate a tendency for the microbial communities of the biofilms to become homogenized, becoming similar, as explained by the authors.

Pu et al. (2019) used metagenomic analysis to evaluate the taxonomic composition of biofilms from three polluted regions in China and found that more polluted regions tend to have greater species richness. Acidobacteria and Armatimonadetes were the most frequently found bacteria, with the former being recognized for its ecological functions, while the latter is usually associated with heavy metals. Chlorobi bacteria showed low abundance and an inverse relationship with the outer polymeric layer of the biofilm, while cyanobacteria showed the opposite behavior. The authors point to the complexity of the system and the understanding of the cause and effect relationship of pollution with taxonomy and also suggest that physical factors (pH, temperature and light), as well as hydrological factors, also contribute to this complex dynamic.

The effect of pollution can interfere with the feeding dynamics of rivers. For example, there is the study by Bowen (2022) who, evaluating the feeding behavior of fish, verified the characteristic of selectivity and predilection in the feeding of these animals for better quality biofilms.

Under such conditions, biofilms can be an impact factor, not only in the feeding of other beings, but also in the entire trophic chain, both when there is an excessive increase in biomass and when there is a severe decrease in its availability. Furthermore, Bowen (2022) found that habitat characteristics affect both the formation and retention of benthic biofilm, this includes hydrological variation, translated by variations in water levels, rainfall levels, lighting, temperature and water quality.

This difference in characteristics of biofilms impacts the feeding dynamics of fish that consume them, since some species reject biofilms with low nutritional content. Polluted environments can present an increase in nutrient concentrations and, therefore, increase the volumes of biofilm, which act as a significant source of carbon for several benthic invertebrates and, in this sense, deregulate the feeding dynamics of local trophic levels, as is the case of the hyporheic zones evaluated by Alam et al. (2020). In contrast, some environments with nutrient scarcity affect the growth capacity of biofilms, decreasing their biomass levels (Weitere et al., 2021).

Climate change can also represent a major environmental problem, as scarcity or flooding events generate turbulence in the natural hydrological characteristics of lakes and streams and, as a result, can alter the structure of natural microbial communities that are already adapted to their habitat, also bringing concern for the maintenance of food chains (Alam et al., 2020; Skovsholt et al., 2020; Zhou et al., 2024). In regions with freezing and thawing characteristics, for example, changes in hydrology may lead to physical and chemical changes in water and in connectivity between locations, compromising the maintenance of microbial communities that are not adapted to the process of mixing and stabilizing new water conditions, as well as the contribution of winds and the environmental filtering capacity of benthic communities (Skovsholt et al., 2020; Ramoneda et al., 2021).

In this sense, punctual or diffuse increases in pollution can be reinforced by contributions and changes in the local hydrology. However, such rates are aggravated by the habit patterns of human activities and by extreme hydrological events that occur due to climate change, and also by anthropogenic pressures that deregulate the environment and alter the natural relationships of ecosystems. This cyclical scenario possibly interferes with the trophic dynamics of rivers and may be aggravated in the future.

Due to the ability of biofilms to respond to pollution, communities may be exposed to unfavorable conditions, with the recurrent presence of microcontaminants in water resources, especially elements that can confer the characteristic of bacterial resistance to biofilms and alter their physiological functions, such as antibiotics and several classes of drugs (Proia et al., 2013; Aubertheau et al., 2016; Smeti et al., 2019; Valdés et al., 2021; Gionchetta et al., 2023).

Osorio et al. (2015), for example, noted that water pollution by microcontaminants, probably originating from the WWTP, followed a dynamic inversely proportional to the river flow, due to the dilution effect, resulting in lower concentrations of these pollutants in the water during flood periods, and this fact may have altered the functional responses of biofilms according to the region sampled, mainly due to the use of antimicrobials and antibiotics. This fact was reinforced by the authors when the biofilm was modified from a less polluted location to a more polluted one, causing the response of biofilms to increased pollution to be an increase in the number of dead cells inside them.

Contrary to Osorio et al. (2015), Pereda et al. (2021) noted an increase in biomass volume in regions with greater pollutant discharge; however, metabolic functions such as phosphorus processing capacity and organic matter degradation were reduced.

On the other hand, Andrus et al. (2013) observed that the presence of Atrazine in the medium had little influence on the biofilm variables. According to the authors, this may have occurred not necessarily because the compound is not harmful to biota, but because the concentration of the herbicide was below the limits reported as causing biologically significant effects on communities, and the exposure, in this case, was not sufficient to generate relevant interference. Similar conclusions were found by Bighiu et al. (2020), evaluating several pesticides in water and river biota in Sweden.

In this sense, although the limit values of several pollutants are not yet known, as well as their synergistic effects on biofilms, their presence in both water bodies and biofilms has been evaluated, given the concerns about bioaccumulation and health of the water environment and trophic levels. In a study carried out by Huerta et al. (2016), seven pharmaceutical products were quantified in biofilms, including psychiatric drugs such as Venlafaxine at concentrations of 0.6 ng g^{-1}, preservatives such as methylparaben (200 ng g^{-1}), among others.

Aubertheau et al. (2016) observed that biofilms evaluated after a wastewater treatment plant presented significant values of pharmaceutical products such as diclofenac and antibiotics present in the biomass. Concentrations of 190 ng g^{-1} and 276 ng g^{-1} were found, respectively. The study found a change in the diversity of cyanobacteria, as well as an increase of up to 31 times in the number of integrons (elements capable of expressing genes, including resistance). Valdés et al (2021) detected antibiotics in biofilms, finding a maximum concentration of 652 μg kg^{-1}, as is the case of ciprofloxacin. As a result, it was noticed that the biofilm community existing after the Wastewater Treatment Plant had distinct characteristics from the communities upstream of it, having a heterotrophic predominance, as well as darker coloration, a more vertical composition surrounded by a very evident gelatinous matrix.

Regions subjected to continuous pollution can generate a class of biofilms classified as undesirable (previously known as sewage fungi) or even proliferate invasive species within them (Huang et al., 2021; Bagra et al., 2023; Gionchetta et al., 2023; Exton et al., 2024). Although naturally occurring, this class of biofilms can present highly adapted, resilient and paradoxical behavior, as it poses a risk to the environment due to the characteristic of, at the same time that it consumes excess dissolved organic carbon from the water, it also reduces the dissolved oxygen levels in the environment and generates more water stress, requiring preventive or remedial actions (Huang et al., 2021; Exton et al., 2024).

The bacterial and fungal diversity of biofilms can be greatly driven by physical and chemical factors in the environment (Zhou et al., 2024), and, in this case, invasive elements can proliferate more easily in situations of water stress, reducing diversity within colonies and, possibly, eliminating species that are fundamental to maintaining the community (Bagra et al., 2023).

Locations influenced by WWTPs and dams, for example, may receive contributions of water flow and increased flow, as well as increased concentration of pollutants, with discharge downstream, which may generate long-term consequences for the biological communities belonging to the ecosystem (Robinson et al., 2004; Osorio et al., 2015; Pereda et al., 2021). This shows that interference in the course, natural flow and physical-chemical characteristics of water can bring irreversible consequences for biofilms, even if indirectly.

In contrast to polluted sites, B-Béres et al. (2022), when evaluating existing diatoms in stream biofilms, found that the nutrient levels of an environment (nitrates, ammonium, phosphates, chlorides) can affect diatom richness, although, as previously exemplified, these are algae that are resilient to environmental stressors. In the case of the study, it was observed that due to low nutrient levels together with stable flows, some diatom taxa prevail, such as stem-forming diatoms.

It is noticeable that, depending on the scenario, the characteristics of the location and the compounds found in the water can interfere as much or more in the characteristics of formation and composition of biofilms than the variations in flow or extreme events of the location, since biofilms can be more sensitive to some compounds present in the water than to the effects associated with the dilution of the flow.

In a study carried out by Bagra et al. (2023), in two rivers without the influence of a Wastewater Treatment Plant, in Germany, and, therefore, with reduced influences from anthropic effects, it was noticed that there were predominant bacteria in both locations (Proteobacteria and Bacteroidetes), with differences in abundance. The intentional submission, carried out in artificial environments, of the communities by copper (a recurrent contaminant in rivers due to the use of fertilizers or effluent discharge), generated a loss in the microbial diversity of the biofilms, favoring the invasion by E. coli inside the colonies. In this sense, the study shows that chemical stresses in the environment can favor invasion processes of invasive communities with high adaptive and resistance potential, bringing a new configuration, transient or permanent, to the composition of biofilms.

Similarly, Gionchetta et al (2023) found that the composition of the heterotrophic biofilm microbial community in a region influenced by wastewater treatment plants diversifies during physical events of water stagnation, being highly influenced by water pH and temperature. The prominent and resistant bacterial groups were Pseudomonadota, Actinomycetota, Firmicutes, Chloroflexi, and Acidobacteria, and transient invasion of E. coli into the biofilms was observed when there was hydrological stagnation. The entry of E. coli into the system was possibly facilitated due to low shear stresses and turbulent events; however, the invasion continued when environmental conditions eventually became limited. In this sense, the hydrological condition of the channel and pollution effects may have facilitated the entry of an invasive species, while, in the hydrological condition evaluated, factors related to temperature and pH were better related to the internal diversity of biofilm colonies.

Furthermore, some compounds have the capacity to persist in the environment regardless of the hydrological scenario, with chemical characteristics that determine their adhesion preferences to the different available matrices (Moschini-Carlos et al., 2000; Osorio et al., 2015; Huerta et al., 2016; Pereda et al., 2021), while others will not bring such prominent effects due to the concentration found during the matrix exposure period (Andrus et al., 2013; Valdés et al., 2021), in which case the other stressors are possibly more significant.

In contrast, Bertrans-Tubau et al. (2023), in bench research, noticed, when simulating different levels and severities of capture periods, that droughts modify the internal structure of biofilms, affecting not only structural issues, but also the capacity to dissipate pesticides.

The distance from the pollution emissary can also be a contributing factor, since the greater the distance, the greater the interference of dilution, sorption effects, competition and biofilm interaction (Aubertheau et al., 2016).

This shows how complex systems need to be assessed as a set of factors that act within an orchestrated synergy. Therefore, understanding the stressors and hydrological dynamics in biofilms cannot be an isolated assessment, requiring a combination of efforts to understand several variables that act in complex ecosystems such as analytical chemistry, hydrology, channel morphology and ecology and that make it difficult to understand the dynamics of such challenging biofilm processes (Sabater et al., 2007; Osorio et al., 2015; Arroita et al., 2016; Fernandes et al., 2020; Weitere et al., 2021).

It is clear that both physical factors such as temperature, pH and hydrological conditions of flows, as well as chemical pollution caused by multiple pollutants such as excess nutrients, toxic compounds or micropollutants can impact the formation and diversity of biofilm. However, due to the synergistic effect between the variables and the complexity of the environment, the predominance of the cause and effect relationship is still unknown and difficult to measure. Furthermore, there is a gap in the literature of studies that evaluate all these physical and chemical variables and address a range of organisms for taxonomic evaluation, both of the environment (water, sediment) and of the biofilm, in a single location or study.

Finally, it is necessary to consider the different characteristics of extension, biomes, climate and environmental culture of each region in which biofilms are formed, which can confer different behaviors to the variables involving growth, permanence, development and heterogeneity for the clusters. In this sense, it reinforces what practically all studies state: the biofilm responds well to hydrological and environmental variables, although not in a unique way, and could act as a good biomarker. However, understanding the scenario and diagnosis depend on integrated assessments that seek to understand the real dynamics of the variables and the responses of the microbiota (Sabater, 2017; Exton et al., 2024).

With the imminence of more climate change events, variations in flow may generate higher concentrations of pollutants due to the lack of dilution in drought events, as well as the dragging of pollutants via surface runoff, including pesticides (Smeti et al., 2019; Bighiu et al., 2020), various microcontaminants (Smeti et al., 2019) and microplastics (Bhatt et al., 2021; Vincent et al., 2022), influencing exchanges in the aquatic environment and impacting biota in a multifactorial manner.

It is therefore suggested that new studies incorporate multifactorial and multidisciplinary approaches to diagnostic assessment for damage prevention and mitigation.

CONCLUSION

The studies considered within the scope of this review show the complexity of the aquatic ecosystem and that it has several stressors acting on the relationship between biofilms and hydrological variables.

The most recurrent variables in the literature were flow/fluxes (related to hydrological stressors) and biofilm biomass (related to biofilm response), and these were also the variables that presented the most responses in terms of effective relationships and, possibly, of great value for future approaches and in-depth studies.

However, metrics related to species richness and biofilm community composition were found to be important and therefore eligible for new scopes. The assessment of metabolic and physiological activities is promising for future studies, and techniques that delve deeper into the unraveling of genetic materials, such as metabarcoding, metagenomics, and transcriptomics, are recommended.

As for hydrological variables, it is necessary to consider variables that are currently used as supporting data, such as temperature, precipitation levels, and seasonal assessment, so that it is possible to eventually understand effects that flow changes, when assessed in isolation, are not capable of explaining.

It is also recommended that methodological protocols for biofilm collection be developed especially in relation to: samplers/substrates, and in this case, the possibility of developing artificial samplers is highlighted as a comprehensive and possibly effective methodological alternative; incubation/exposure time; and the process of removing biofilm from substrates. The need to include seasonal assessment in sampling projects is reinforced, although the variable itself is classified as hydrological, as it can produce different responses in biofilms that become interesting in the context of their assessment.

The literature shows that hydrological variables act significantly in periphytic communities, however, in a distinct and variable manner, making it necessary to understand the scenario evaluated in its particularities so that the relationships can be adjusted and the effects diagnosed or predicted.

Furthermore, the literature also points to secondary effects of hydrological variations that culminate in pollution processes and affect the functioning of microbiological communities, altering the joint action of hydrological and chemical variations in the environment as major drivers of stress in biofilms.

There is a clear need to expand, deepen and integrate multifactors in studies that verify the relationships of hydrological response in natural biofilms, since the relationships in real environments still present great distinctions and gaps, probably due to the complexity of the water systems, the surroundings, and the biofilm community itself.

The study of river biofilms has gained notoriety over the years and, given their ecological importance, and in view of the hydrological events arising from imminent climate change, the need to understand the functioning of biofilms in real environments and the direct responses to environmental stressors and other interferents that act in synergy has become urgent, so that, in the future, they can be used as environmental monitoring matrices, regulated by the competent authorities, as well as their predicted behaviors to safeguard water environments.

Finally, the studies showed that the relationship between natural biofilms in rivers and the interference of hydrological variables is a large field to be unraveled and, therefore, a good opportunity for the scientific community to deepen its knowledge.

ACKNOWLEDGEMENTS

The authors would like to thank the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) - Grant Number 88882.382701/2019-01, the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), the Research Productivity Grant (Process 312478/2022-4) and Public Call FA 07/2023 - Senior Grant Program of the Araucária Foundation and the Retention Program for Senior Professors and Researchers of UTFPR, for funding this project.

REFERENCES

Adame, K. L., Dunck, B., & Rodrigues, L. (2018). Periphytic algal community in lentic environments of the Upper Paraná River floodplain: seasonal and spatial variation. *Acta Limnologica Brasiliensia*, *30*, e205. http://doi.org/10.1590/s2179-975x5017.

Alam, M. K., Negishi, J. N., Rahman, M. A. T. M. T., & Rodulfo Tolod, J. (2020). Stable isotope ratios of emergent adult aquatic insects can be used as indicators of water pollution in the hyporheic food web. *Ecological Indicators*, *118*, 106738. http://doi.org/10.1016/j.ecolind.2020.106738.

Andrus, J. M., Winter, D., Scanlan, M., Sullivan, S., Bollman, W., Waggoner, J. B., Hosmer, A. J., & Brain, R. A. (2013). Seasonal synchronicity of algal assemblages in three Midwestern agricultural streams having varying concentrations of atrazine, nutrients, and sediment. *The Science of the Total Environment*, *458*(460), 125-139. http://doi.org/10.1016/j.scitotenv.2013.03.070.

Arroita, M., Flores, L., Larranaga, A., Martínez, A., Martínez-Santos, M., Pereda, O., Ruiz-Romera, E., Solagaistua, L., & Elosegi, A. (2016). Water abstraction impacts stream ecosystem functioning via wetted-channel contraction. *Freshwater Biology*, *62*(2), 243-257. http://doi.org/10.1111/fwb.12864.

Aubertheau, E., Stalder, T., Mondamert, L., Ploy, M., Dagot, C., & Labanowski, J. (2016). Impact of wastewater treatment plant discharge on the contamination of river bio fi lms by pharmaceuticals and antibiotic resistance. *Science of the Total Environment*, *579*, 1387-1398. http://dx.doi.org/10.1016/j.scitotenv.2016.11.136.

Bagnoud, A., Pramateftaki, P., Bogard, M. J., Battin, T. J., & Peter, H. (2020). Microbial Ecology of Methanotrophy in Streams Along a Gradient of CH4 Availability. *Frontiers in Microbiology*, *11*, 771. http://doi.org/10.3389/fmicb.2020.00771.

Bagra, K., Bellanger, X., Merlin, C., Singh, G., Berendonk, T., & Klümper, U. (2023). Environmental stress increases the invasion success of antimicrobial resistant bacteria in river microbial communities. *The Science of the Total Environment*, *904*, 166661. http://doi.org/10.1016/j.scitotenv.2023.166661.

Barral-Fraga, L., Barral, M. T., MacNeill, K. L., Martiñá-Prieto, D., Morin, S., Rodríguez-Castro, M. C., Tuulaikhuu, B. A., & Guasch, H. (2020). Biotic and abiotic factors influencing arsenic biogeochemistry and toxicity in fluvial ecosystems: a review. *International Journal of Environmental Research and Public Health*, *17*(7), 2331. http://doi.org/10.3390/ijerph17072331.

B-Béres, V., Kókai, Z., Varbiro, G., Mustazhapova, G., Csabai, Z., Pernecker, B., Borics, G., Bácsi, I., & Boda, P. (2022). Flow intermittence drives the benthic algal composition, biodiversity and diatom-based quality of small hilly streams in the pannonian ecoregion, Hungary. *Frontiers in Ecology and Evolution*, *10*, 834548. http://dx.doi.org/10.3389/fevo.2022.834548.

Bertrans-Tubau, L., Menard, Y., Batisson, I., Creusot, N., Mazzella, N., Millan-Navarro, D., Moreira, A., Morin, S., Ponsá, S., Abril, M., Proia, L., Romaní, A., & Artigas, J. (2023). Dissipation of pesticides by stream biofilms is influenced by hydrological histories. *FEMS Microbiology Ecology*, *99*(9), fiad083. http://dx.doi.org/10.1093/femsec/fiad083.

Bhatt, P., Pathak, V. M., Bagheri, A. R., & Bilal, M. (2021). Microplastic contaminants in the aqueous environment, fate, toxicity consequences, and remediation strategies. *Environmental Research*, *200*, 111762. http://doi.org/10.1016/j.envres.2021.111762.

Bighiu, M. A., Höss, S., Traunspurger, W., Kahlert, M., & Goedkoop, W. (2020). Limited effects of pesticides on stream macroinvertebrates, biofilm nematodes, and algae in intensive agricultural landscapes in Sweden. *Water Research*, *174*, 115640. http://doi.org/10.1016/j.watres.2020.115640.

Böger, B., Surek, M., Vilhena, R., Fachi, M., Junkert, A., Santos, J., Domingos, E., de Fátima, A., Momade, D., & Pontarolo, R. (2021). Occurrence of antibiotics and antibiotic resistant bacteria in subtropical urban rivers in Brazil. *Journal of Hazardous Materials*, *402*, 123448. http://doi.org/10.1016/j.jhazmat.2020.123448.

Bondar-Kunze, E., Kasper, V., & Hein, T. (2021). Responses of periphyton communities to abrupt changes in water temperature and velocity, and the relevance of morphology: A mesocosm approach. *The Science of the Total Environment*, *768*, 145200. http://doi.org/10.1016/j.scitotenv.2021.145200.

Borges, R. M., Minillo, A., Lemos, E. G. M., do Prado, H. F. A., & Tangerino, E. P. (2016). Uso de filtros de carvão ativado granular associado a microrganismos para remoção de fármacos no tratamento de água de abastecimento. *Engenharia Sanitaria e Ambiental*, *21*(4), 709-720. http://doi.org/10.1590/s1413-41522016118787.

Bowen, S. (2022). The river flood pulse, benthic biofilm, and the nutrition of Prochilodus lineatus. *Environmental Biology of Fishes*, *105*, 213-230. http://dx.doi.org/10.1007/s10641-022-01211-1.

Calapez, A. R., Elias, C. L., Alves, A., Almeida, S. F. P., Brito, A. G., & João, M. (2020). Shifts in bio films' composition induced by flow stagnation, sewage contamination and grazing. *Ecological Indicators*, *111*, 106006. http://doi.org/10.1016/j.ecolind.2019.106006.

Casartelli, M. R., Lavagnolli, G. J., & Ferragut, C. (2016). Periphyton biomass accrual rate changes over the colonization process in a shallow mesotrophic reservoir. *Acta Limnologica Brasiliensia*, *28*, e9. http://doi.org/10.1590/S2179-975X0116.

Chaumet, B., Morin, S., Hourtané, O., Artigas, J., Delest, B., Eon, M., & Mazzella, N. (2019). Flow conditions influence diuron toxicokinetics and toxicodynamics in freshwater biofilms. *The Science of the Total Environment*, *652*, 1242-1251. http://doi.org/10.1016/j.scitotenv.2018.10.265.

Chen, B., Chau, S. N., Zhang, N., Liang, H., Sun, L., Zhao, X., Guo, J., & Lu, H. (2022). Micro and nano bubbles promoted biofilm formation with strengthen of COD and TN removal synchronously in a blackened and odorous water. *The Science of the Total Environment*, *837*, 155578. http://doi.org/10.1016/j.scitotenv.2022.155578.

Colls, M., Timoner, X., Font, C., Sabater, S., & Acuña, V. (2019). Effects of duration, frequency, and severity of the non-flow period on stream biofilm metabolism. *Ecosystems (New York, N.Y.)*, *22*(6), 1393-1405. http://doi.org/10.1007/s10021-019-00345-1.

Costerton, J. W., Lewandowski, Z., Caldwell, D., Korber, D. R., & Lappin-scott, H. M. (1995). Microbial biofilms. *Annual Review of Microbiology*, *49*(1), 711-745. http://dx.doi.org/10.1146/annurev.mi.49.100195.003431.

Coulson, L., Feldbacher, E., Pitzl, B., & Weigelhofer, G. (2022). Effects of intermittent flow on biofilms are driven by stream characteristics rather than history of intermittency. *The Science of the Total Environment*, *849*, 157809. http://doi.org/10.1016/j.scitotenv.2022.157809.

Cui, Z., Fan, W., Chen, C., Mo, K., Chen, Q., Zhang, Q., & He, R. (2023). Ecosystem health evaluation of urban rivers based on multitrophic aquatic organisms. *Journal of Environmental Management*, *349*, 119476. http://doi.org/10.1016/j.jenvman.2023.119476.

Exton, B., Hassard, F., Medina-Vaya, A., & Grabowski, R. (2024). Undesirable river biofilms: the composition, environmental drivers, and occurrence of sewage fungus. *Ecological Indicators*, *161*, 111949. http://doi.org/10.1016/j.ecolind.2024.111949.

Fernandes, G., Bastos, M., Vargas, J., Le Guet, T., Clasen, B., & dos Santos, D. (2020). The use of epilithic biofilms as bioaccumulators of pesticides and pharmaceuticals in aquatic environments. *Ecotoxicology (London, England)*, *29*(9), 1293-1305. http://doi.org/10.1007/s10646-020-02259-4.

Flemming, H.-C., van Hullebusch, E., Neu, T., Nielsen, P., Seviour, T., Stoodley, P., Wingender, J., & Wuertz, S. (2023). The biofilm matrix: multitasking in a shared space. *Nature Reviews Microbiology*, *21*, 70-86. http://dx.doi.org/10.1038/s41579-022-00791-0.

Gionchetta, G., Snead, D., Semerad, S., Beck, K., Pruden, A., & Bürgmann, H. (2023). Dynamics of antibiotic resistance markers and Escherichia coli invasion in riverine heterotrophic biofilms facing increasing heat and flow stagnation. *The Science of the Total Environment*, *893*, 164658. http://doi.org/10.1016/j.scitotenv.2023.164658.

Guo, K., Wu, N., Manolaki, P., Baattrup-Pedersen, A., & Riis, T. (2020). Short-period hydrological regimes override physico-chemical variables in shaping stream diatom traits, biomass and biofilm community functions. *The Science of the Total Environment*, *743*, 140720. http://doi.org/10.1016/j.scitotenv.2020.140720.

Hou, J., Shao, G., Adyel, T. M., Li, C., Liu, Z., Liu, S., & Miao, Li. (2022). Can the carbon metabolic activity of biofilm be regulated by the hydrodynamic conditions in urban rivers? *Science of The Total Environment*, *832*, 155082. http://dx.doi.org/10.1016/j.scitotenv.2022.155082.

Huang, W. L., Wang, Z., Yano, S., Li, J., Hao, G., & Zhang, J. (2021). Modeling periphyton biomass in a flow-reduced river based on a least squares support vector machines model: implications for managing the risk of nuisance periphyton. *Journal of Cleaner Production*, *286*, 124884. http://doi.org/10.1016/j.jclepro.2020.124884.

Huang, W., Wang, Z., Liu, X., Zhu, D., Wang, Y., & Wu, L. (2022). The microbial community and functional indicators response to flow restoration in gradient in a simulated water flume. *Frontiers in Microbiology. 13*, http://dx.doi.org/10.3389/fmicb.2022.1051375.

Huerta, B., Rodriguez-Mozaz, S., Nannou, C., Nakis, L., Ruhí, A., Acuña, V., Sabater, S., & Barcelo, D. (2016). Determination of a broad spectrum of pharmaceuticals and endocrine disruptors in bio fi lm from a waste water treatment plant-impacted river. *The Science of the Total Environment*, *540*, 241-249. http://doi.org/10.1016/j.scitotenv.2015.05.049.

Karygianni, L., Ren, Z., Koo, H., & Thurnheer, T. (2020). Biofilm matrixome: extracellular components in structured microbial communities. *Trends in Microbiology*, *28*(8), 668-681. http://doi.org/10.1016/j.tim.2020.03.016.

Leandrini, J. A., Fonseca, I. A., & Rodrigues, L. (2008). Characterization of habitats based on algal periphyton biomass in the upper Paraná River floodplain, Brazil. *Brazilian Journal of Biology = Revista Brasileira de Biologia*, *68*(3), 503-509. http://doi.org/10.1590/S1519-69842008000300006.

Lin, Q., Zhang, Y., Marrs, R., Sekar, R., Luo, X., & Wu, N. (2020). Evaluating ecosystem functioning following river restoration: the role of hydromorphology, bacteria, and macroinvertebrates. *The Science of the Total Environment*, *743*, 140583. http://doi.org/10.1016/j.scitotenv.2020.140583.

Madge Pimentel, I., Baikova, D., Buchner, D., Burfeid Castellanos, A., David, G., Doliwa, A., Hadžiomerović, U., Mayombo, N. A. S., Prati, S., Spyra, M., Vermiert, A. M., Beisser, D., Dunthorn, M., Piggott, J., Sures, B., Tiegs, S., Leese, F., & Beermann, A. (2024). Assessing the response of an urban stream ecosystem to salinization under different flow regimes. *The Science of the Total Environment*, *926*, 171849. http://doi.org/10.1016/j.scitotenv.2024.171849.

Mai, Y. Z., Peng, S. Y., & Lai, Z. N. (2020). Structural and functional diversity of biofilm bacterial communities along the Pearl River Estuary, South China. *Regional Studies in Marine Science*, *33*, 100926. http://doi.org/10.1016/j.rsma.2019.100926.

Marshall, K. C., Stout, R., & Mitchell, R. (1971). Mechanism of the initial events in the sorption of marine bacteria to surfaces. *Journal of General Microbiology*, *68*(3), 337-348. http://doi.org/10.1099/00221287-68-3-337.

McKay, S. F., & King, A. J. (2006). Potential ecological effects of water extraction in small, unregulated streams. *River Research and Applications*, *22*(9), 1023-1037. http://doi.org/10.1002/rra.958.

Montagner, C., Sodré, F., Acayaba, R., Vidal, C., Campestrini, I., Locatelli, M., Pescara, I. C., Albuquerque, A. F., Umbuzeiro, G. A., & Jardim, W. (2019). Ten years-snapshot of the occurrence of emerging contaminants in drinking, surface and ground waters and wastewaters from São Paulo State, Brazil. *Journal of the Brazilian Chemical Society*, http://dx.doi.org/10.21577/0103-5053.20180232.

Moschini-Carlos, V., Henry, R., & Pompêo, M. L. M. (2000). Seasonal variation of biomass and productivity of the periphytic community on artificial substrata in the Jurumirim Reservoir. *Hydrobiologia*, *434*(1/3), 35-40. http://doi.org/10.1023/A:1004086623922.

Myrstener, M., Gómez-Gener, L., Rocher-Ros, G., Giesler, R., & Sponseller, R. A. (2020). Nutrients influence seasonal metabolic patterns and total productivity of Arctic streams. *Limnology and Oceanography*, http://dx.doi.org/10.1002/lno.11614.

Osorio, V., Proia, L., Ricart, M., Pérez, S., Ginebreda, A., Luís, J., Sabater, S., & Barceló, D. (2015). Hydrological variation modulates pharmaceutical levels and bio film responses in a Mediterranean river. *The Science of the Total Environment*, *472*, 1052-1061. http://doi.org/10.1016/j.scitotenv.2013.11.069.

Palmer, M., & Ruhi, A. (2019). Linkages between flow regime, biota, and ecosystem processes: implications for river restoration. *Science*, *365*(6459), eaaw2087. http://doi.org/10.1126/science.aaw2087.

Pan, M., Liu, X., Ma, W., Li, X., Li, H., Ding, C., Chen, Y., & Chen, R. (2020). The effect of hydrodynamics on the succession of autotrophic and heterotrophic organisms of biofilms in river ecosystems. *Water Science and Technology*, *83*(1), 63-76. http://dx.doi.org/10.2166/wst.2020.536.

Pereda, O., Schiller, D., & Von, (2021). Combined effects of urban pollution and hydrological stress on ecosystem functions of Mediterranean streams. *The Science of the Total Environment*, *753*, 141971. http://doi.org/10.1016/j.scitotenv.2020.141971.

Proia, L., Osorio, V., Soley, S., Köck-schulmeyer, M., Pérez, S., Barceló, D., Romaní, A. M., & Sabater, S. (2013). Effects of pesticides and pharmaceuticals on bio fi lms in a highly impacted river. *Environmental Pollution*, *178*, 220-228. http://doi.org/10.1016/j.envpol.2013.02.022.

Pu, Y., Ngan, W. Y., Yao, Y., & Habimana, O. (2019). Could benthic biofilm analyses be used as a reliable proxy for freshwater environmental health? *Environmental Pollution*, *252*, 440-449. http://doi.org/10.1016/j.envpol.2019.05.111.

Qu, X., Peng, W., Liu, Y., Zhang, M., Ren, Z., Wu, N., & Liu, X. (2019). Networks and ordination analyses reveal the stream community structures of fish, macroinvertebrate and benthic algae, and their responses to nutrient enrichment. *Ecological Indicators*, *101*, 501-511. http://doi.org/10.1016/j.ecolind.2019.01.030.

Ramoneda, J., Hawes, I., Pascual-García, A. J., Mackey, T., Sumner, D., & Jungblut, A. (2021). Importance of environmental factors over habitat connectivity in shaping bacterial communities in microbial mats and bacterioplankton in an Antarctic freshwater system. *FEMS Microbiology Ecology*, *97*(4), fiab044. http://doi.org/10.1093/femsec/fiab044.

Robinson, C. T., Uehlinger, U. R. S., & Monaghan, M. T. (2004). Stream ecosystem response to multiple experimental floods from a reservoir. *River Research and Applications*, *20*(4), 359-377. http://doi.org/10.1002/rra.743.

Romaní, A. M., Fund, K., Artigas, J., Schwartz, T., Sabater, S., & Obst, U. (2008). Relevance of polymeric matrix enzymes during biofilm formation. *Microbial Ecology*, *56*(3), 427-436. http://doi.org/10.1007/s00248-007-9361-8.

Romero, F., Acuña, V., Font, C., Freixa, A., & Sabater, S. (2019). Effects of multiple stressors on river biofilms depend on the time scale. *Scientific Reports*, *9*(1), 15810. http://doi.org/10.1038/s41598-019-52320-4.

Sabater, S. (2017). Microbial ecotoxicology: looking to the future. In C. Cravo-Laureau, C. Cagnon, B. Lauga & R. Duran (Eds.), *Microbial ecotoxicology* (pp. 339-352). Cham: Springer International Publishing. http://doi.org/10.1007/978-3-319-61795-4_14.

Sabater, S., Guasch, H., Ricart, M., Romaní, A., Vidal, G., Klünder, C., & Schmitt-Jansen, M. (2007). Monitoring the effect of chemicals on biological communities. The biofilm as an interface. *Analytical and Bioanalytical Chemistry*, *387*(4), 1425-1434. http://doi.org/10.1007/s00216-006-1051-8.

Salis, R. K., Bruder, A., Piggott, J. J., Summerfield, T. C., & Matthaei, C. D. (2019). Multiple-stressor effects of dicyandiamide (DCD) and agricultural stressors on trait-based responses of stream benthic algal communities. *The Science of the Total Environment*, *693*, 133305. http://doi.org/10.1016/j.scitotenv.2019.07.111.

Sheng, G.-P., Yu, H.-Q., & Li, X.-Y. (2010). Extracellular polymeric substances (EPS) of microbial aggregates in biological wastewater treatment systems: a review. *Biotechnology Advances*, *28*(6), 882-894. http://doi.org/10.1016/j.biotechadv.2010.08.001.

Shi, X., Wang, L., Chen, A., Yu, W., Liu, Y., Huang, X., Long, X., Du, Y., & Qu, D. (2024). Enhancing water quality and ecosystems of reclaimed water-replenished river: a case study of Dongsha River, Beijing, China. *The Science of the Total Environment*, *926*, 172024. http://doi.org/10.1016/j.scitotenv.2024.172024.

Skovsholt, L. J., Pastor, A., Docherty, C. L., Milner, A. M., & Riis, T. (2020). Changes in hydrology affects stream nutrient uptake and primary production in a high-Arctic stream. *Biogeochemistry*, *151*(2-3), 187-201. http://doi.org/10.1007/s10533-020-00719-x.

Smeti, E., von Schiller, D., Karaouzas, I., Laschou, S., Vardakas, L., Sabater, S., Tornés, E., Monlor-Alcaraz, L. S., Guillem-Argiles, N., Martinez, E., Barcelò, D., Alda, M. L., Kalogianni, E., Elosegi, A., & Skoulikidis, N. (2019). Multiple stressor effects on biodiversity and ecosystem functioning in a Mediterranean temporary river. *The Science of the Total Environment*, *647*, 1179-1187. http://doi.org/10.1016/j.scitotenv.2018.08.105.

Valdés, M. E., Santos, L. H. M. L. M., Rodríguez Castro, M. C., Giorgi, A., Barceló, D., Rodríguez-Mozaz, S., & Amé, M. V. (2021). Distribution of antibiotics in water, sediments and biofilm in an urban river (Córdoba, Argentina, LA). *Environmental Pollution*, *269*, 116133. http://doi.org/10.1016/j.envpol.2020.116133.

Villeneuve, A., Bouchez, A., & Montuelle, B. (2011). In situ interactions between the effects of season, current velocity and pollution on a river biofilm. *Freshwater Biology*, *56*(11), 2245-2259. http://doi.org/10.1111/j.1365-2427.2011.02649.x.

Vincent, A. E. S., Chaudhary, A., Kelly, J. J., & Hoellein, T. J. (2022). Biofilm assemblage and activity on plastic in urban streams at a continental scale: site characteristics are more important than substrate type. *The Science of the Total Environment*, *835*, 155398. http://doi.org/10.1016/j.scitotenv.2022.155398.

Vlaičević, B., Gulin Beljak, V., Matoničkin Kepčija, R., & Turković Čakalić, I. (2022). Periphytic ciliate communities in lake ecosystem of temperate riverine floodplain: variability in taxonomic and functional composition and diversity with seasons and hydrological changes. *Water (Basel)*, *14*(4), 551. http://doi.org/10.3390/w14040551.

Vlaičević, B., Kepčija, R. M., & Čerba, D. (2021). Structure and dynamics of the periphytic ciliate community under different hydrological conditions in a Danubian floodplain lake. *Limnologica*, *87*, 125847. http://doi.org/10.1016/j.limno.2020.125847.

Weitere, M., Altenburger, R., Anlanger, C., Baborowski, M., Bärlund, I., Beckers, L.-M., & Brauns, M. (2021). Disentangling multiple chemical and non-chemical stressors in a lotic ecosystem using a longitudinal approach. *The Science of the Total Environment*, *769*, 144324. http://doi.org/10.1016/j.scitotenv.2020.144324.

Zhang, L., Dong, D., Hua, X., & Guo, Z. (2018). Inhibitory effects of extracellular polymeric substances on ofloxacin sorption by natural biofilms. *Science of The Total Environment*, 625, 178-184. http://doi.org/10.1016/j.scitotenv.2017.12.271.

Zhou, Y., Meng, F., Ochieng, B., Xu, J., Zhang, L., Kimirei, I. A., Feng, M., Zhu, L., & Wang, J. (2024). Climate and environmental variables drive stream biofilm bacterial and fungal diversity on

Tropical Mountainsides. *Microbial Ecology*, *87*(1), 28. http://doi.org/10.1007/s00248-023-02335-2.

AUTHOR CONTRIBUTIONS

Bárbara Alves de Lima: Collaboreted with the idea for the article, the conception and design of the study, the bibliographic search, data analysis and the first version of the manuscript.

Tatiani Andressa Modkovski: Collaboreted with the conception and design of the study, the bibliographic search, data analysis and the first version of the manuscript.

Juliane Ribeiro das Chaves: Collaboreted with the conception and design of the study, the bibliographic search, data analysis and the first version of the manuscript.

Luis Otavio Miranda Peixoto: Collaboreted with the conception and design of the study, and critical review of the work.

Samanta Teixeira Medeiros: Collaboreted with the conception and design of the study, the bibliographic search, data analysis and the first version of the manuscript.

Gabriel Helmer Baer: Collaboreted with the conception and design of the study, the bibliographic search, data analysis and the first version of the manuscript.

Cristóvão Vicente Scapulatempo Fernandes: Collaboreted with the conception and design of the study, and critical review of the work.

Júlio César Rodrigues de Azevedo: Collaboreted with the conception and design of the study, and critical review of the work.

Editor in-Chief: Adilson Pinheiro

Associated Editor: Fernando Mainardi Fan