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Review

Microbial mat ecosystems: Structure types, functional diversity, and biotechnological application

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ABSTRACT

Microbial mats are horizontally stratified microbial communities, exhibiting a structure defined by physiochemical gradients, which models microbial diversity, physiological activities, and their dynamics as a whole system. These ecosystems are commonly associated with aquatic habitats, including hot springs, hypersaline ponds, and intertidal coastal zones and oligotrophic environments, all of them harbour phototrophic mats and other environments such as acidic hot springs or acid mine drainage harbour non-photosynthetic mats. This review analyses the complex structure, diversity, and interactions between the microorganisms that form the framework of different types of microbial mats located around the globe. Furthermore, the many tools that allow studying microbial mats in depth and their potential biotechnological applications are discussed.

Contents

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1. Introduction

In nature, microorganisms often form communities adhering to a solid surface to form complex ecological assemblages in different habitats around the world [1,2]. Adherence to a surface is a strategy used for millions of years by microorganisms to survive and evolve in community and allows microorganisms to cope with the various abiotic factors that surround them, with some of them being stressful. These types of biological organizations range from simple monospecific biofilms to complex microbial mats formed by a wide variety of microorganisms, wherein a wide variety of ecological interactions are observed [3].

Microbial mats are benthic, vertically layered, and self-sustaining communities that develop in the liquid–solid interface of various environments [4]. Furthermore, they comprise millions of microorganisms belonging to different species, which interact and exchange signals, embedded in a matrix of exopolysaccharides, and nutrients to enable a greater flow of resources and energy for the survival of the community [5]. The associations observed are restricted, with some of them being symbiotic, which confers them a selective advantage [6,7].

Microbial mats have been present on Earth for millions of years, the oldest of which are found in sedimentary rocks of 3.7 Ga and 3.4 Ga west of Australia [7,8,9,10,11] and South Africa [12], respectively, from the Archaean era. However, it was present in a greater abundance in the Proterozoic (2.5–0.57 Ga) era, with worldwide distribution [13]. The extensive fossil record suggests that these communities are highly stable and flexible in adapting to continuous environmental changes [13]; these ecological assemblages today persist in extreme environments such as hypersaline ponds, hot springs, and sulfur springs, where environmental conditions restrict and limit the growth of some multicellular and eukaryotic organisms [14,15].

The role of microbial mats has been crucial throughout the history of the Earth for the composition and modification of the atmosphere, producing O_2, H_2, and CH_4 [16] and also represents the first ecosystems together with stromatolites. Thus, microbial mats are, undoubtedly, a natural laboratory where microbial diversity (patterns and community structure), evolutionary processes, and their adaptation to extreme environments can be studied [17,18,19]. In this review, we analyze in detail the complex structures that comprise a microbial mat, the different types of microbial mats, and their microbial diversity. Furthermore, we have analyzed the main tools, including a perspective on its potential application in areas such as medicine, different industries, and bioremediation of contamination due to luminaires used for studying microbial mats in the last decade.

2. Structure, functionality and ecological dynamics of microbial mats

Microbial mats are structures visible to the naked eye, with the thickness ranging from millimeters to several centimeters, and are formed by multiple biofilms of microorganisms embedded in a matrix of exopolysaccharides [20] in a vertical fashion due to the physical gradients (Fig. 1) [21]. One of the main factors of biological diversity in microbial mats is attributed to its dynamic physiochemical gradients, which are largely modified by the biological processes of the inhabiting microorganisms. These biological processes and physical gradients provide the required microenvironments and ecological niches for microorganisms with specific needs [4,7,22]. These communities are essentially formed by organisms of the domain 'Bacteria'; however, the domains 'Archaea' and 'Eukarya' are also involved in forming microbial mats, although less diverse and abundant in nature [23].

The chemical parameters to be considered for studying microbial mats are the presence of oxygen, pH, redox potential, saline concentration, presence of electron donor and acceptor compounds, and the diversity of chemical species, whereas the important physical parameters to be considered include light, temperature, and pressure. The study of biological interactions (symbiotic, neutralism and amensalism) in the mats is another relevant aspect that we have considered in this review [24]. Relevant processes such as photosynthesis, nitrogen fixation, denitrification, metal reduction, sulfate reduction, and methanogenesis are vital to the performance of mats [25,26].

Microbial mats consist of various basic biofunctional groups such as Cyanobacteria, anoxygenic photosynthetic bacteria (generally represented by non-sulfur green bacteria of the Chloroflexi division), green sulfur bacteria (Chlorobi) and purple bacteria (Proteobacteria division), aerobic heterotrophs and anaerobes, sulfate-reducing bacteria (SRB), sulfur oxidizing bacteria and methanogenic archaea [22,27].

The main source of energy and nutrition of microbial mats is through photosynthesis [7], although non-photosynthetic mats exist. In a typical mat, the first step for survival of this trophic network is photosynthesis, a process in which light energy is utilized to fix inorganic carbon (CO_2) to organic carbon ($(CH_2O)_n$), thereby releasing oxygen (Fig. 2), performed by the primary producers Cyanobacteria [28,29]. Microbial mats function as a consortium where biogeochemical cycles and biochemical processes are coupled [30], and this close interaction allows the products of the metabolism of one group to be available and used by other microorganisms.

Nitrogen fixation is primarily performed by unicellular and filamentous Cyanobacteria; however, SRB have been found to play a key role in this biological process [31]. SRB are an important group of bacteria capable of reducing sulfates to sulfur, oxidizing organic matter, and obtaining energy in the process. In addition, SRB are essential for calcium precipitation and lithification of mats, and therefore, are responsible for mat preservation in fossil record [28].

The formation of these complex communities is performed by a process of ecological succession, wherein the Cyanobacteria are the colonizing organisms and microenvironment modifiers for the later colonization of more specialized bacteria and with higher and specific environmental requirements [32]. In addition, a microbial mat is a dynamic community in which microorganisms are capable of motility

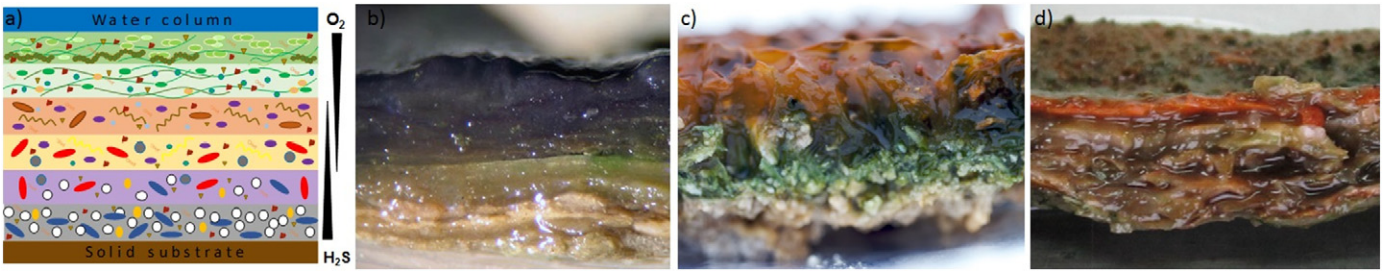

Fig. 1. General structures of microbial mats. The thickness can range from millimeters to several centimeters, and are formed by multiple biofilms of microorganisms embedded in a matrix of exopolysaccharides, in a vertical fashion due to the physical gradients.

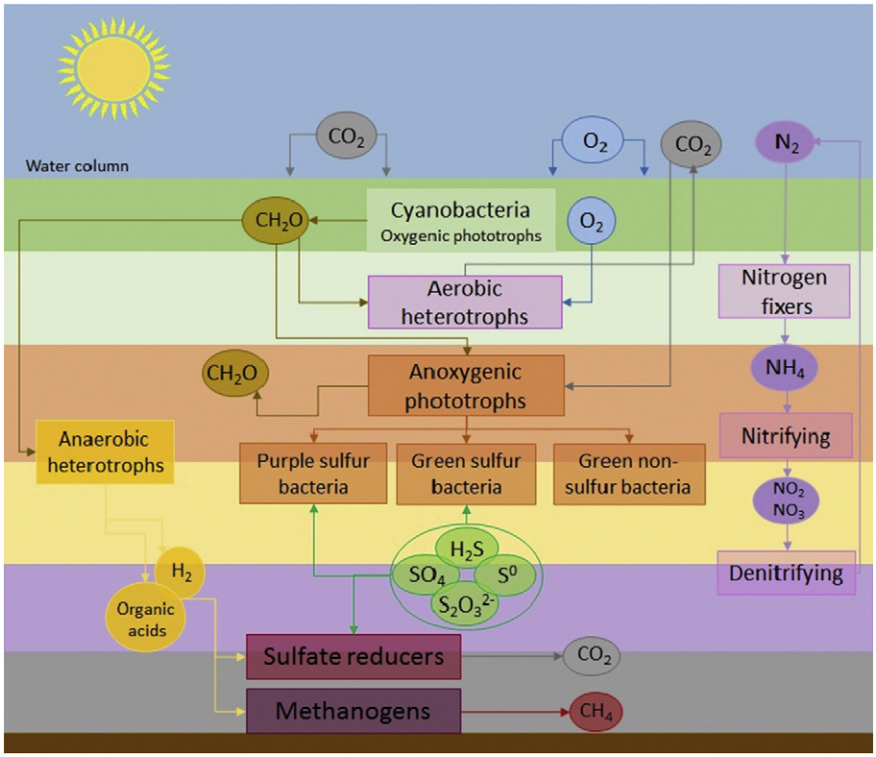

Fig. 2. Structure and metabolism overview of photosynthetic mats. See for details: Section 2. Structure, Functionality and Ecological dynamics of microbial mats.

and, thus, modifying their position in the mat in search of favorable environmental conditions such as luminous intensity and redox potential [33].

3. Types of microbial mats around the globe

Microbial mats prosper under extreme environments; however, they are widely distributed in the biosphere (Table 1). The microbial diversity associated with different types of mats is described in the sections below. A vast majority are phototrophic mats, with a significant photosynthetic component, so they are highly dependent on the presence of light.

3.1. Hypersaline mats

Hypersaline mats, generally associated with saline lakes, are among the best studied along with thermophiles and coastal mats. Moreover, these mats thrive on extreme conditions such as a high degree of water salinity, high temperatures and/or high levels of radiation, without these factors obstructing the formation of complex microbial communities [5,34].

Microbial mats in Guerrero Negro, a town located in the state Baja California Sur in the north of Mexico, project as saline mats. The geochemical complexity of these saline water sources is only matched by the complexity of its microbial communities, and despite high salt concentrations, mats are formed in its water sources [35]. It is one of the most diverse microbial ecosystems. Microbial culture, amplification, cloning and sequencing of 16S rRNA as well as metagenomics have provided sufficient information for the description of this community. This community is primarily dominated by bacteria in a proportion of 90%:9%:1% of bacteria, archaea, and eukaryotes, respectively [7]. The vertical distribution of the major bacterial divisions is intimately associated with the presence of light, oxygen and H_2S, and the formation of spatiotemporal chemical gradients has a strong effect on the structure of the microbial community [36].

Cyanobacteria have been considered as the primary producers of all phototrophic mats; classical microbiological analysis has shown abundant predominant filamentous forms such as Oscillatoriales (*Microcoleus*, *Oscillatoria*, *Phormidium*, and *Lyngbya*), Nostocales (*Calothrix*), and Chroococcales (*Gloeocapsa*) in the upper layer. Recent studies have shown that Cyanobacteria are primarily distributed in the aerobic zone of the mat (2–3 mm) [7].

In Guerrero Negro, 42 phyla and 752 species [7], which were recovered from the 16S rRNA libraries, were found, in which Chloroflexi were the most abundant division with the presence of Bacteroidetes, Proteobacteria, Planctomycetes, Cyanobacteria, Spirochaetes, and Verrucomicrobia [7,35]. In addition, it also harbored archaea and eukaryotes to a lesser extent, with archaea constituting 9% of the total recovered sequences, with Crenarchaeota (6%) and Euryarchaeota (3%) as the present divisions [37]. Also, nematodes, arthropods, stramenipiles, alveolates, fungi, and chlorophytes were also found in the mats, constituting the 1% of the total biological community [38].

The Atacama Desert of Chile is one of the driest places on earth. The Llamará Salt Flat is located in this desert, forming a saltine crust comes to be flooded by rainfall some months of the year, and microbial mats are formed in this extreme environmental conditions with high concentrations of salt. The study of the flat laminated communities has shown a predominance of both unicellular cyanobacteria such as *Synechococcus* and *Cyanothece*, as well as filament forms such as *Microccoleus*, *Oscillatoria* and also *Gloeocapsa* and *Gloeobacter*. In addition, important members such as anoxygenic photosynthetic bacteria and the sulfate reducing bacteria were detected, as well as unidentified cocci and bacilli [39].

In the hypersaline mats of Shark Bay, located in Australia, 58 bacterial phyla have been detected. Cyanobacteria (38%) are predominant in the superficial part (2 mm) of the mat, the genera *Microcoleus*, *Halomicronema*, and *Leptolyngbya* dominate the photic zone, the class Anaerolineae of the Chloroflexi division dominates the rest of the mat and finally at the bottom of the mat are Firmicutes

Table 1
Published relevant works portraying the microbial diversity of different types of mat ecosystems around the globe. The cultivable, microscopic or molecular tools employed are also described (analyzed in this review).

Microbial mat name/location	Mat structure type/ Physicochemical traits	Dominant microbial diversity	Technique employed	Reference
La Salada de Chiprana/ (Northeastern Spain)	Benthic microbial mat community, phototrophic/Hypersaline	Cyanobacteria-dominated, anoxygenic phototrophic, aerobic heterotrophic, colorless sulfur-, and sulfate-reducing bacteria	DGGE, Microscopic analysis and serial dilution	[40]
Guerrero Negro/Baja California Sur, México	Phototrophic microbial mat/Hypersaline	Chloroflexi, Proteobacteria, Bacteroidetes, Planctomycetes, Spirochaetas, Verrucomicrobia, Cyanobacteria	SSU rRNA libraries and sequenced/Metagenomics (Sanger and Pyrosequencing 454)	[7,35]
Shark Bay/Nimelah, Hamelin Pool, Australia	Photosynthetic microbial mat (smooth SM/pustular PM), Hypersaline	SM: Proteobacteria, Chloroflexi, Planctomycetes, Cyanobacteria, Bacteroidetes, Spirochaetes, Caldithrix, Firmicutes, GN04, OP8 PM: Proteobacteria, Bacteroidetes, Planctomycetes, Chloroflexi, Cyanobacteria, Acidobacteria, Spirochaetes, GN04, Verrucomicrobia, Gemmatimonadetes, Actinobacteria	Metagenomics, Illumina Miseq	[34]
Schiermonnikoog Island/North Sea beach of the Dutch Barrier/Netherlands	Photosynthetic microbial mat/Coastal	ST1 (freshwater zone): Proteobacterias (α, γ, β, δ), Bacteroidetes, Cyanobacteria, Actinobacterias, Firmicutes, Chloroflexi ST2 (Intermediate zone): Proteobacterias (α, β, δ), Bacteroidetes, Cyanobacteria, Actinobacterias, Verrucomicrobia ST3 (Intertidal zone): Proteobacteria (α, γ, δ), Bacteroidetes, Actinobacterias, Verrucomicrobia, Planctomycetes	Massive sequencing of 16S rRNA (V6 region)	[20]
Great Sippewissett salt marsh/Buzzards Bay/Massachussets/USA	Photosynthetic intertidal mat/Coastal	Cyanobacteria, Proteobacteria (particularly Chromatiales), Chloroflexi, Spirochaetes, Acidobacteria, Verrucomicrobia, Caldithrix, Actinobacteria	SSU-rRNA amplicon libraries-454 sequencing and metagenomic direct sequencing	[21]
Cuatro Cienegas/Chihuahuan desert, Coahuila, México	Photosynthetic stable microbial mat SM, Disturbed mat DM/Oligotrophic environment	SM: Cyanobacteria, Proteobacteria (γ, ε, δ), Bacteroidetes, Chloroflexi, Cholrobi, Acidobacteria, Firmicutes (Clostridia), Planctomycetes, Nitrospira DM: Proteobacteria (γ, β, α, ε), Fimircutes (Bacilli), Actinobacteria	16S rRNA gene clone libraries, Metagenomic protein gene analyses	[3]
Ward Hunt Lake/Northern Ellesmere Island region/Canadian High Arctic	Benthic phototrophic microbial mat/Psychrophilic	Cyanobacteria Chlorophytes, Heterotrophic bacteria.	CLSM, SEM, EDS and LTSEM	[48,49]
Continental Antarctica and the Antarctic Peninsula	Aquatic microbial mat/Psychrophilic	Actinobacteria, Bacteroidetes, Proteobacteria, Firmicutes, *Deinococcus-Thermus*	Microbial culture, Rep-PCR, 16rRNA gene sequencing	[47]
Ady Entre and Roşiori/Romania	Phototrophic microbial mat/thermophilic	Cyanobacteria, Chloroflexi, Proteobacteria	SEM, 16S rDNA clone library construction, sequencing and phylogenetic analysis	[59]
Central and Central-Eastern Tibet	Phototrophic microbial mat/thermophilic	Firmicutes, Proteobacteria, Cyanobacteria, Chloroflexi, Bacteroidetes, Acidobacteria, Nitrospirae, Planctomycetes, Thermodesulfobacteria, Aquificae	Powder X-ray diffraction, 16S rDNA Clone library construction	[58]
Porcelana, Cahuelmó/Northern Patagonia, Chile	Phototrophic microbial mat/thermophilic	Cyanobacteria, Bacteroidetes, *Deinococcus-Thermus*, Proteobacteria (β, γ, ε), Acidobacteria	DGGE	[57]
Boekleung (Western Thailand)	Phototrophic microbial mat/thermophilic	Cyanobacteria, Chloroflexi, Bacteroidetes, OP10, Actinobacteria, Planctomycetes	DGGE	[56]
Wonder Lake/Luzon Island, Philippines	Phototrophic microbial mat/thermophilic	Cyanobacteria, Bacteriodetes, Chlorobi, Chloroflexi, Firmicutes, Proteobacteria (γ)	Microscopy, Pigment determination, DGGE	[60]
Araró/Michoacán, México	Phototrophic microbial mat/thermophilic	Firmicutes, Proteobacteria, Actinobacteria	Microbial culture	[64]
Acid Mine Drainage/Iron Mountain, California, USA	Acidic non-Phototrophic microbial mat	Nitrospirae (*Leptospirillum*), Euryarchaeota (Thermoplasmales), Actinobacteria (*Acidimicrobium*, *Ferrimicrobium*),	Microscopy, RFLP, 16S rRNA clone library	[66]
One Hundred Spring Plain, Beowulf/Norris Geyser Basin, Yellowstone National Park, USA	Acidic thermophilic Non-Phototrophic microbial mat	Aquificae (*Hydrogenobaculum*), Crenarchaeota (*Metallosphaera yellowstonensis*), Geoarchaeota	Iron Accretion Rates, SEM, FISH, 16S rRNA Gene sequencing	[67]
Atacama Desert, Chile/Llamará Salt Flat	Hypersaline mat	*Synechococcus, Cyanothece, Microccoleus, Oscillatoria, Gloeocapsa, Gloeobacter*. Anoxygenic photosynthetic bacteria and unidentified cocci and bacilli	Macro and Microscopy description	[39]

Abbreviations: DGGE (Denaturing Gradient Gel Electrophoresis); FISH (Fluorescence in situ Hybridization); RFLP (Terminal-Restriction Fragment Length Polymorphism); Rep-PCR (Repetitive element sequence-based PCR); SSU-rRNA (Small subunit ribosomal RNA); CLSM (Confocal Laser Scanning Microscopy), SEM (Scanning Electron Microscopy); EDS (X-ray Energy, Dispersive, Spectroscopy); LTSEM (Low Temperature Scanning Electron Microscopy).

and Planctomycetes (Brocadiae). Bacteroidetes and Proteobacteria are also found in the mat. Furthermore, α-proteobacteria (*Dichotomicrobium thermohalophilum*), γ-proteobacteria (Class Anaerolineae), δ-proteobacteria (Desulfobacterales and Desulfovibrionales) are also found in abundance in the mat [34].

Another example of hypersaline mats is La Salada de Chiprana found in Spain, which also has high levels of magnesium (seven times more than seawater) and sulfates. Of interest, microbial mats have developed even in such conditions. Several techniques are used for studying this community such as microbial culture, microscopy and denaturing gradient gel electrophoresis (DGGE). Cyanobacteria (*Halothece*-like, *Microcoleus*-like, *Pseudoanabaena*-like and *Gloeocapsa*-like) and Chloroflexi (*Chloroflexus*-like) are the major component of this mat. In addition, purple sulfur bacteria, aerobic heterotrophic bacteria, colorless sulfur bacteria and SRB were detected by DGGE. This community has a high availability of organic substrates,

during day and night, and dissolved organic carbon in the form of fatty acids may be the reason for an unusual top layer of *Chloroflexus*, a photoheterotrophic bacterium [40].

3.2. Coastal mats

Coastal and hypersaline mats are the most biologically diverse and have extensive coastal distribution [4]. The intertidal coastal zones present irregular floods, high saline concentration fluctuations and intense temperature changes and are primarily inhabited by Cyanobacteria [41], although recent studies have discovered the importance of other bacterial groups, viz. Proteobacteria and Bacteroidetes [20].

The Schiermonnikoog Island of the Netherlands contains a 'green beach', a huge strip with microbial mats measuring 300 m wide to 5 km long. Substantial analysis of the 16S rRNA genes has shown that Proteobacteria (Subdivision α-proteobacteria of the Rhodobacteriales and Sphingomonadales orders, γ-proteobacteria order Chromatiales and δ-proteobacteria of the Desulfobacteriales and Desulfovibrionales orders), Bacteroidetes (Flavobacteriales and Sphingobacteriales), Cyanobacteria, and Actinobacterias are the dominant bacterial divisions. Although, Euryarchaeota (particularly Methanogens) and Crenarchaeota are present to a lesser extent, they are important Archean elements [20].

Armitage and collaborators [21] found that Proteobacteria, Cyanobacteria and Chloroflexi are the most abundant divisions in the Great Swamp of Sippewisset (Massachussets, USA), with the presence of Spirochaetes, Acidobacteria, Verrucomicrobia, Caldithrix, and Actinobacteria in a smaller proportion. Most of the microbial mats presented with structural and organizational similarities; however, coastal mats have a large number of eukaryotic representatives, primarily diatoms (*Navicula* sp., *Diploneis* sp., *Amphora* sp. and *Cylindrotheca*) and algae (*Chlorophyta* and *Enteromorpha* sp.) [42,43].

3.3. Microbial mats in oligotrophic environments

The oligotrophic mats of Cuatro Ciénegas in the desert of Coahuila, to the north of Mexico, are best studied. These mats are rare, but the importance of their study lies in the search for life outside the planet with similar atmosphere. Cuatro Ciénegas is distinguished by its extremely low phosphorus content, which is an important limiting factor for the existence of life because phosphorus in the form of phosphates is a vital constituent of DNA, proteins and energy molecules.

However, it has been observed that the geographic isolation has affected the speciation at the microbial level, with some exclusive microorganisms, *Bacillus coahuilensis* [44], a Firmicute found in this portion has shown specific adaptations such as the high presence of sphingolipids in their membrane to survive in a low phosphorus environment [45].

Bonilla-Rosso et al. [3] analyzed two mats under an independent cultivation approach, one mat in stable conditions and the other with constant disturbances, and revealed the following interpretations: first, these constant disturbances have a strong effect on the communities, thereby preventing an increase in diversity, and second, even at low concentrations of phosphorus the stable community can develop a high biological diversity.

Mats that are not exposed to constant disturbances show a diverse community with no dominant groups, with Proteobacteria, Cyanobacteria and Bacteroidetes as the most diverse groups, as well as 16 other divisions and 28 bacterial orders.

3.4. Psychrophile microbial mats

The largest proportion of the planet has low temperatures (below 5°C), with a vast array of cold environments from the oceans, alpine areas, caves and polar regions [46]. Antarctica and the Arctic shelter microbial mats (polar region mats), represent hot spots of biological diversity and primary production [47].

The photosynthetic mats of the poles are dominated by filamentous Cyanobacteria (orders: *Dichothrix*, Nostocales-*Tolypothrix*, and Oscillatoriales-*Tychonema*), which produce a polysaccharide matrix that provides protection to organisms with lower tolerance; diatoms, algae, flagellates, ciliates, nematodes, rotifers and microinvertebrates are part of this community [48,49].

Extreme polar conditions impose strong selective pressure, low temperatures, high solar radiation, prolonged winter darkness, drought, nutrient deficiency, and freezing and thawing cycles [50,51]. In Antarctica, it has been observed that heterotrophic bacteria play a major role in nutrient cycling, where Actinobacteria, Proteobacteria, Bacteroidetes, Firmicutes and Deinococcus–Thermus are the major divisions [47]. The Cyanobacteria constitute the dominant group of photosynthetic bacteria, with the filamentous forms of the Nostocales and Oscillatoriales order being the most common [52]. Other photosynthetic groups (Chloroflexi and Chlorobi) are present in a smaller proportion [50].

3.5. Hot springs microbial mats

High temperature environments such as hot springs, geysers and currents from them represent an extreme environment for life. pH, sulfur concentration and temperature are the main limiting characteristics for the development of life, with temperature being essential in the modeling of these peculiar communities [53]. These mats are the least diverse within phototrophs; however, they are key to understanding the oldest communities on Earth [4].

Thermophilic communities may be associated with sediments, streams, water columns and microbial mats [17]. In microbial mats, photosynthetic organisms play an important role in the metabolic dynamics of the community. Photosynthesis in thermal waters presents two major obstacles, high temperatures that decrease dissolved gases such as O_2 and CO_2 and denaturation of proteins and other biomolecules due to high temperature; therefore, the temperature limit for the growth of photosynthetic bacteria is 75°C, the temperature at which chlorophyll degrades [54].

Among the most prolific members of this category of mats are Cyanobacteria. Cyanobacteria play two key roles in the community, carbon and nitrogen fixation. Unicellular Cyanobacteria, particularly *Synechococcus*, are predominant in springs wherein the temperature of the thermal water exceeds 55°C [19,55]. Thermophilic microbial mats are located in different parts of the world, viz. Thailand [56], Patagonia [57], Tibet [58], Romania [59] and the Philippines [60], and their geographical distribution is directly linked to geothermal zones. However, to date, the most studied are those of the Yellowstone National Park (YNP) in the USA [61,62].

Cyanobacteria constitute the bacterial group that is common to all phototrophic mats, and together with Proteobacteria, Chloroflexi, Bacteroidetes and Deinococcus–Thermus they represent the most abundant groups in these mats; other divisions that are present but in minute abundance are Planctomycetes, Firmicutes, Acidobacteria, Verrucomicrobia, Nitrospirae, Actinobacteria, Synergistetes, and Armatimonadetes [56,57,58,59]. In addition, when the temperature is around 40–55°C, the presence of filamentous Cyanobacteria and a marked lack of unicellular forms are observed [57,60].

The ecological analysis of these communities has shown a slight effect of season changes on populations of *Synechococcus* and Chloroflexi in the mats obtained from Octopus at the YNP [63]; however, Lacap et al. [60] found a significant change in the community when analyzed during the rainy season and drought, suggesting the importance of precipitation in the structure of the mats. These communities are stable against abrupt environmental changes, and biological diversity may be one of the responses to external shocks.

The geothermal zone of Araró is located within the Trans-Mexican Volcanic belt, north of Michoacán in Mexico. This locality harbors many thermal springs with high levels of arsenic and salts, but only some of these springs enable the growth of thermophilic microbial mats. We were handed with the task of cultivating aerobic heterotrophs, given their important ecological role in cycling carbon and regulating oxygen levels in these communities. The isolated bacteria mostly belonged to members of the Firmicutes Division (*Bacillus*, *Paenibacillus* and *Exiguobacterium*), followed by Proteobacteria (*Pseudomonas* and *Aeromonas*) and Actinobacterias (*Microbacterium*) [64].

3.6. Acid microbial mats

A wide variety of microbial mats develops primarily at alkaline pH, and some can form in acidic environments. Mats that do not present photosynthetic microbial groups are shown, and these communities present oxidation of iron and sulfur as the predominant metabolism (Fig. 3). Acid mine drainage with sulfur minerals (e.g. Pyrite, FeS_2; arsenopyrite, FeAsS and Chalcopyrite, $CuFeS_2$) has acidic pH values between 0.77 and 1.21 and a high amount of toxic metals [65,66]. These conditions limit the propagation of the microbial diversity to very low values. The inhabitants are usually bacteria and archaea because their metabolisms are directly linked to the reduction and oxidation of iron and sulfate reduction. The phyla that are present in the mats are Actinobacteria, Firmicutes, δ-Proteobacteria, *Nitrospira*, *Leptospirillum*, *Acidomicrobium*, *Ferromicrobium acidophilum*, and Thermoplasmales [65,66].

Moreover, acidic springs have a pH range of approximately 3–3.5, and among the most studied mats are those obtained from the YNP. In acid mines, the metabolism of iron and sulfate are essential for the dynamics of the community. The springs One Hundred Spring Plain and Beowulf presented with *Hydrogenobaculum* spp., *Metallosphaera yellowstonensis*, heterotrophic archaea (unidentified) and even members of a new Geoarchaeota archaea division [67,68,69].

4. Tools for the study of microbial mats

Microbiology and microbial culture were the first tools to assess the unknown microscopic world [70]. Since then, microbial mats have been studied with different approaches, and the information that has been collected has allowed knowing and understanding of some of these intriguing communities. In the study of microbial mats, scanning electron microscopy (SEM) has been fundamental to study microstructure and the morphology of the bacteria that comprise the mats [19,59,68,71,72]. SRB are distributed in different sheets throughout the mat, contrary to the idea that they inhabit just the anaerobic zone [73].

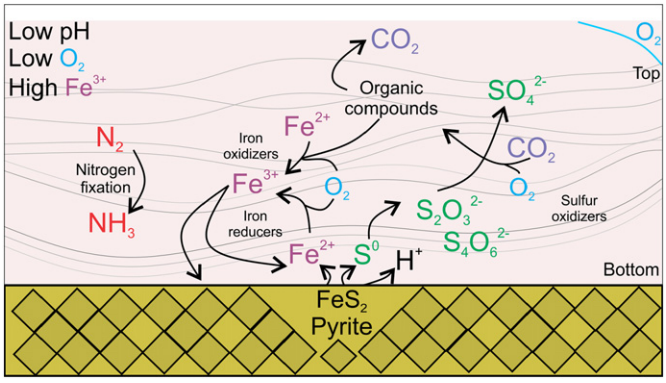

Fig. 3. Scheme of general acid microbial, where are usually developed in the absence of light. Oxidation–reduction of iron and sulfur are the predominant metabolisms. Environmental conditions are low pH and low oxygen concentration. The figure shows the main compounds that are cyclized in non-photosynthetic mats. See for details: Section 3.6. Acid microbial mats.

Furthermore, microbial culture has proved to be a technique par excellence in the discovery, description and exploration of the microbial diversity and has allowed studying the basic aspects of the biology of microorganisms. Since 1969, Castenholz (Professor Emeritus) and associates have been cultivating microorganisms of microbial mats from the thermal pools of the YNP, allowing a detailed description, likewise in Guerrero Negro, Mexico analyzed the microbial diversity in Hypersaline carpets [74], primarily of the thermophilic Cyanobacteria present. Brock et al. [88] noted that the physiology of bacteria changed drastically when grown in vitro. Therefore, they observed the behavior of photosynthetic bacteria in their natural habitat by cultivating the bacteria in an in situ culture system, a simple and effective method [70]. In addition, some bacteria are favored by culture, whereas others cannot be detected [75]. In another approach, some researchers have cultivated mats (mesocosmos) and have tested the entire community's response to different environmental conditions of temperature and UV radiation and have observed a drastic change in the community structure [76].

Currently, microbial cultures present serious problems to reflect microbial diversity. Estimates of diversity range from 1% to 10% cultivable microorganisms, with 90–99% of the diversity having low chances of being discovered with this procedure. Therefore, with limitations in microscopy techniques and the advent of molecular techniques and the refinement of PCR, new technologies such as DGGE [77], temperature gradient gel electrophoresis (TGGE) [78], rRNA intergenic spacer analysis (RISA) [79] and terminal-restriction fragment length polymorphism (T-RFLP) [80] were developed. DNA fingerprinting is another technique that has been vital for analyzing microbial mats; however, DGGE, wherein each band represents a different species, population or operational taxonomic unit (OTU) in the banding pattern, is the most routinely employed technique. The distribution of bacterial population in the different layers of the mat, the effect of different seasons, adaptation to different temperatures [57,63], present diversity and adaptive radiation have also been studied [81].

With the onset and momentum of metagenomic approaches, analysis of the 16S rRNA through cloning, transformation and sequencing has been gaining popularity, and large 16S rRNA libraries are sequenced and classified to study biodiversity. One of the most impressive study of microbial mats was performed in Guerrero Negro; the hypersaline mats in this region were one of the most diverse microbial ecosystems, with more than 750 species detected [7]. Of today, second and third generation mass sequencing technologies have resulted in the generation of large numbers of metagenomes from very diverse environments [82,83] and microbial mats [17] are no exception to these analyses.

Researchers from Guerrero Negro and Cuatro Ciénegas have studied the taxonomic and functional diversity of these mats by sequencing and analyzing metagenomes [3,35]. In the YNP, a very ambitious and complete project was undertaken where the metagenomes of 20 geothermal sites with distinctive chemical characteristics were sequenced, including two with phototrophic mats, and the results revealed the diversity and distribution of the main microbial groups in each geothermal environment [17].

Analyzing the diversity of these environments is a challenge but assessing the functionality is even more complicated. Klatt et al. [84] have used metatranscriptomics to identify and study the genes expressed by the communities observed at the site. Studies of the mats in springs Octopus and Mushroom in the YNP using metagenomics revealed that Cyanobacteria and Chloroflexi were the dominant group, and further analyses of the metagenome enabled to establish the networks of interaction between the microbial groups and helped in revealing proof for horizontal gene transfer [84]. Moreover, genes involved in nitrogen fixation and diel cycle [85] and

the expression of genes involved in photosynthesis, such as the production of bacteriochlorophyll, were also studied.

In a recent study by Drewniak et al. [86], they used a combination of molecular and biochemical tools as well as metagenomics and electron microscopy to study the diversity, structure and ecological role of two microbial mats located in two mines, one gold and one uranium, in Poland. Of relevance, the authors observed that the microbial mats were capable of decontaminating and purifying the water containing high levels of heavy metals that runs through drainage systems. Metagenomic analysis revealed that the community harbored the families Methylococcaceae and Methylophilaceae in abundance, along with the filamentous bacteria *Leptothrix*, *Thiothrix,* and *Beggiatoa*, which were a central part of the community. It is interesting to note that the authors suggest that microbial mats form a natural barrier to purify water as a result of its biofilm formation capability and because they employ heavy metals in the respiration processes (oxide reduction).

Multiple data entries are submitted to the databases routinely; however, the study, analysis, and interpretation of the data are an arduous task that requires a lot of work. Much is still unknown of these communities, and the close relationship that bacteria form with their environment and with other microbial groups is attributable to their genetic coding, which is a challenging study.

5. Biotechnological applications of microbial mats

Microbial mats can be formed, as discussed above, under conditions that may be considered extreme. In other words, high temperatures outside the mesophilic range (>40°C) or temperatures that slightly exceed the water freezing point are important limiting factors for cell growth and reproduction [87]. A classic example of enzyme that has been found in thermophilic organisms is Taq polymerase, a DNA polymerase isolated from *Thermophilus aquaticus* or *Thermus aquaticus*, a bacterium isolated from the Lower Geyser Basin in the YNP by the microbiologist Thomas Brock in 1969 [88]. Taq polymerase has many applications in molecular biology, particularly in PCR.

Apart from Taq polymerase, enzymes exhibiting activities such as degrading cellulose, lignin or chitin as well as various polysaccharides, lipids or proteins have also been discovered. Many of these enzymes are used in industries such as paper, detergent, leather processing and shoe production [89]. Therefore, microbial mats are excellent candidates for searching and studying such enzymes and their functions.

Other metabolites, compounds and by-products of metabolism have been isolated from microorganisms that are part of the microbial mats in thermophilic environments. For example, antimicrobial compounds and inhibitors of quorum sensing have been described in Cyanobacteria mats located near thermal springs. Some compounds showed excellent antibacterial activity against *Bacillus* sp., *Micrococcus luteus*, *Shigella sonnei*, *Salmonella enterica,* and *Klebsiella pneumoniae*. Some of the quorum inhibitor compounds exhibited activity against many model strains such as *Chromobacterium violaceum* CV017 and *Agrobacterium tumefaciens* NTL4 [90].

In another study, Putri et al. [91] reported the isolation and characterization of a new antibiotic, the tetramic compound ophiosetin, which showed a wide range of activity against bacteria (*Staphylococcus aureus* and *Enterococcus faecalis*), tumor cell lines type P388 and viruses (HIV). In this sense, thermal springs and microbial mats formed in thermal springs have been described as important sources for discovering new antibiotic compounds with various applications in medicine [92].

However, biotechnological applications, relative to compounds or enzymes derived from microbial mats, are not limited to thermophilic organisms because psychrophiles can form microbial mats in polar regions, where the temperatures are commonly below zero. In fact, various strains of the *Pseudomonas* genus have multiple metabolic capacities, including the degradation of various contaminants and ability to grow in environments with high concentrations of heavy metals. Therefore, some psychrophilic strains of the *Pseudomonas* species are being evaluated for their bioremediation capabilities in places such as Antarctica [93]. Psychrophiles are also desirable in the detergent and food processing industries because in some cases enzymes with high stability at alkaline pH and low temperatures are required. One example is proteases, including subtilisin, which was initially isolated from multiple strains of *Bacillus* and used in the detergent and food processing industry [94].

Microbial mats can be found in diverse regions and environments around the world, with equal diversity in environmental conditions that are considered extreme and challenging for many organisms. For example, bacteria growing in high saline concentration (halophiles), radiation (radiophiles), low pH (acidophiles) and high pressure (barophiles) [87]. However, microorganisms have developed and evolved with strategies that allow them to colonize such environments (e.g. biofilm formation), allowing them to proliferate since millions of years. Therefore, these extreme capabilities of microorganisms in microbial mats can be further exploited for biotechnological application [95,96]. Likewise, it is desirable to find certain activities that allow bioremediation of contaminated soils, wherein some microbial mats develop under high concentrations of heavy metals. The bacterium *Lampropedia cohaerens* strain CT6 was recently isolated and sequenced from a microbial mat in thermal pools of the Himalayas that contained high concentrations of arsenic [97]. Other works have also shown the potential of bioremediation of oil-contaminated sites through the use of marine microbial mats [98].

Microbial mats are natural ecosystems that produce gases such as methane, CO_2 or hydrogen, thereby promoting the potent use of these gases, primarily produced by Cyanobacteria, as biofuels [99]. Therefore, microbial genome isolated from microbial mats presents enormous biotechnological potential that is eco-friendly and sustainable with no harm to human health.

6. Conclusions

Knowledge about the functioning of microbial communities mostly comes from eukaryotic macroscopic communities; therefore, an endless number of relationships, interactions and functioning of bacterial and archaea communities are unknown [21]. Microbial mats as a study model is an easily accessible viable laboratory, and these communities present with varying degrees of complexity from simple non-phototrophic mats, YNP mats with a low diversity of phototrophs, to the complex hypersaline and mats obtained from Guerrero Negro, México and Llamará, Chile. The various molecular tools such as metagenomics and prediction of functions based on 16S rRNA profiles have opened up endless possibilities for studying the microbial communities and interrelations of mats without the need to cultivate the individual microorganisms that comprise the mats [100]. However, microbial culture should not be ignored as it helps in detailing the metabolic functions of individual microorganisms. Countless examples exist wherein the efforts to cultivate microorganisms requiring ultra-specific culture media have been successful [101]. Moreover, this information needs to be studied further to determine the functions of novel and unknown enzymes that have an application in resolving the persisting environmental and health problems.

Conflict of interest

None.

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References

[1] Davey ME, O'Toole GA. Microbial biofilms: from ecology to molecular genetics. Microbiol Mol Biol Rev 2000;64(4):847–67. https://doi.org/10.1128/MMBR.64.4.847-867.2000.

[2] Neelakanta G, Sultana H. The use of metagenomic approaches to analyze changes in microbial communities. Microbiol Insights 2013;6:37–48. https://doi.org/10.4137/MBI.S10819.

[3] Bonilla-Rosso G, Peimbert M, Alcaraz LD, et al. Comparative metagenomics of two microbial mats at Cuatro Ciénegas basin ll: Community structure and composition in oligotrophic environments. Astrobiology 2012;12(7):659–73. https://doi.org/10.1089/ast.2011.0724.

[4] Bolhuis H, Cretoiu MS, Stal LJ. Molecular ecology of microbial mats. FEMS Microbiol Ecol 2014;90:335–50. https://doi.org/10.1111/1574-6941.12408.

[5] Ruvindy R, White III RA, Neilan BA, et al. Unravelling core microbial metabolisms in the hypersaline microbial mats of Shark Bay using high-throughput metagenomics. ISME J 2016;10:183–96. https://doi.org/10.1038/ismej.2015.87.

[6] Al-Thani R, Al Najjar MAA, Al Raei AM, et al. Community structure and activity of a highly dynamic and nutrient-limited hypersaline microbial mat in Um Alhool Sabkha, Qatar. PLoS One 2014;9(3):e92405. https://doi.org/10.1371/journal.pone.0092405.

[7] Ley RE, Harris K, Wilcox J, et al. Unexpected diversity and complexity of the Guerrero Negro hypersaline microbial mat. Appl Environ Microbiol 2006;72(5): 3685–95. Doi: https://doi.org/10.1128/AEM.72.5.3685-3695.2006.

[8] Allwood AC, Walter MR, Kamber BS, et al. Stromatolite reef from the Early Archaean era of Australia. Nature 2006;441:714–8. https://doi.org/10.1038/nature04764.

[9] Noffke N, Christian D, Wacey D, et al. Microbially induced sedimentary structures recording an ancient ecosystem in the ca. 3.48 billion-year-old dresser formation, Pilbara, Western Australia. Astrobiology 2013;13(12):1–21. https://doi.org/10.1089/ast.2013.1030.

[10] Nutman AP, Bennett VC, Friend CRL, et al. Rapid emergence of life shown by discovery of 3,700-million-year-old microbial structure. Nature 2016;537:535–8. https://doi.org/10.1038/nature19355.

[11] Westall F, de Vries ST, Nijman W, et al. The 3.466 Ga "Kitty's gap Chert," an early Archean microbial ecosystem. In: Reimod WU, Gibson RL, editors. Processes on the early earth, 405. Geol Soc am spec pap; 2006. p. 105–31. https://doi.org/10.1130/2006.2405(07).

[12] Tice MM, Lowe DR. Photosynthetic microbial mats in the 3.416-Myr-old ocean. Nature 2004;431:549–52. https://doi.org/10.1038/nature02888.

[13] Green SJ, Jahnke LL. Molecular investigations and experimental manipulations of microbial mats: A view to paleomicrobial ecosystems. In: Seckbach J, Oren A, editors. Microbial Mats: Modern and ancient microorganisms in stratified systems, cellular origin, life in extreme habitats and astrobiology. Dordrecht: Springer; 2010. p. 183–206. https://doi.org/10.1007/978-90-481-3799-2_9.

[14] Seckbach J, Oren A, editors. Microbial Mats: Modern and ancient microorganisms in stratified systems, cellular origin, life in extreme habitats and astrobiology. Dordrecht: Springer; 2010. https://doi.org/10.1007/978-90-481-3799-2.

[15] Revsbech NP, Trampe E, Lichtenberg M, et al. *In situ* hydrogen dynamics in a spring microbial mat during a diel cycle. Appl Environ Microbiol 2016;82(14):4209–17. https://doi.org/10.1128/AEM.00710-16.

[16] Hoehler TM, Bebout BM, Des Marais DJ. The role of microbial mats in the production of reduced gases on the early earth. Nature 2001;412:324–7. https://doi.org/10.1038/35085554.

[17] Inskeep WP, Jay ZJ, Tringe SG, et al. Metagenome project steering committee and working group members. The YNP metagenome project: environmental parameters responsible for microbial distribution in the Yellowstone geothermal ecosystem. Front Microbiol 2013;4(67):1–15. https://doi.org/10.3389/fmicb.2013.00067.

[18] Villanueva L, Navarrete A, Urmeneta J, et al. Analysis of diurnal and vertical microbial diversity of a hypersaline microbial mat. Arch Microbiol 2007; 188:137–46. https://doi.org/10.1007/s00203-007-0229-6.

[19] Ward DM, Ferris MJ, Nold SC, et al. A natural view of microbial biodiversity within hot spring cyanobacterial mat communities. Microbiol Mol Biol Rev 1998;62(4): 1353–70.

[20] Bolhuis H, Stal LJ. Analysis of bacterial and archaeal diversity in coastal microbial mats using massive parallel 16S rRNA gene tag sequencing. ISME J 2011;5: 1701–12. https://doi.org/10.1038/ismej.2011.52.

[21] Armitage DW, Gallangher KL, Youngblut ND, et al. Millimeter-scale patterns of phylogenetic trait and diversity in a salt marsh microbial mat. Front Microbiol 2012; 3(293):1–16. https://doi.org/10.3389/fmicb.2012.00293.

[22] van Gemerden H. Microbial mats: A joint venture. Mar Geol 1993;113:3–25. https://doi.org/10.1016/0025-3227(93)90146-M.

[23] Casamayor EO, Massana R, Benlloch S, et al. Changes in archaeal, bacterial and eukaryal assemblages along a salinity gradient by comparison of genetic fingerprinting methods in a multipond solar saltern. Environ Microbiol 2002;4(6): 338–48. https://doi.org/10.1046/j.1462-2920.2002.00297.x.

[24] Franks J, Stolz J. Flat laminated microbial mat communities. Earth Sci Rev 2009;96: 163–72. https://doi.org/10.1016/j.earscirev.2008.10.004.

[25] Paerl HW, Pinckney JL. A mini-review of microbial consortia: their roles in aquatic production and biogeochemical cycling. Microb Ecol 1996;31:225–47. https://doi.org/10.1007/BF00171569.

[26] Woebken D, Burow LC, Behnam F, et al. Revisiting N_2 fixation in Guerrero Negro intertidal microbial mats with a functional single-cell approach. ISME J 2015;9: 485–96. https://doi.org/10.1038/ismej.2014.144.

[27] Klatt JM, Meyer S, Hausler S, et al. Structure and function of natural sulphide-oxidizing microbial mats under dynamic input of light and chemical energy. ISME J 2016;10:921–33. https://doi.org/10.1038/ismej.2015.167.

[28] Baumgartner LK, Reid RP, Dupraz C, et al. Sulfate reducing bacteria in microbial mats: changing paradigms, new discoveries. Sediment Geol 2006;185:131–45. https://doi.org/10.1016/j.sedgeo.2005.12.008.

[29] Severin I, Acinas SG, Stal LJ. Diversity of nitrogen-fixing bacteria in cyanobacterial mats. FEMS Microbiol Ecol 2010;73(3):514–25. https://doi.org/10.1111/j.1574-6941.2010.00925.x.

[30] Paerl HW, Pinckney JL, Steppe TF. Cyanobacterial–bacterial mat consortia: examining the functional unit of microbial survival and growth in extreme environments. Environ Microbiol 2000;2(1):11–26. https://doi.org/10.1046/j.1462-2920.2000.00071.x.

[31] Falcón LI, Cerritos R, Eguiarte LE, et al. Nitrogen fixation in microbial mat and stromatolite communities from Cuatro Cienegas, México. Microb Ecol 2007;54:363–73. https://doi.org/10.1007/s00248-007-9240-3.

[32] Boomer SM, Noll KL, Geesey GG, et al. Formation of multilayered photosynthetic biofilms in an alkaline thermal spring in Yellowstone National Park, Wyoming. Appl Environ Microbiol 2009;75(8):2464–75. https://doi.org/10.1128/AEM.01802-08.

[33] Quesada A, Sanchez-Contreras M, Fernández-Valiente E. Tolerance of Antarctic cyanobacterial microbial mats to natural UV radiation. Nova Hedwigia 2001;123: 275–90.

[34] Lun Wong H, Smith D-L, Visscher PT, et al. Niche differentiation of bacterial communities at a millimeter scale in Shark bay microbial mats. Nature 2015;5:15607. https://doi.org/10.1038/srep15607.

[35] Harris JK, Caporaso JG, Walker JJ, et al. Phylogenetic stratigraphy in the Guerrero Negro hypersaline microbial mat. ISME J 2013;7:50–60. https://doi.org/10.1038/ismej.2012.79.

[36] Kunin V, Raes J, Harris JK, et al. Millimeter-scale genetic gradients and community-level molecular convergence in a hypersaline microbial mat. Mol Syst Biol 2008;4: 198. Doi: https://doi.org/10.1038/msb.2008.35.

[37] Robertson CE, Spear JR, Harris JK, et al. Diversity and stratification of archaea in a hypersaline microbial mat. Appl Environ Microbiol 2009;75(7):1801–10. https://doi.org/10.1128/AEM.01811-08.

[38] Feazel LM, Spear JR, Berger AB, et al. Eucaryotic diversity in a hypersaline microbial mat. Appl Environ Microbiol 2008;74(1):329–32. https://doi.org/10.1128/AEM.01448-07.

[39] Demergasso C, Chong G, Galleguillos P, et al. Tapetes microbianos del Salar de Llamará, Norte de Chile. Rev Chil Hist Nat 2003;76(3):485–99. https://doi.org/10.4067/S0716-078X2003000300012.

[40] Jonkers HM, Ludwig R, De Wit R, et al. Structural and functional analysis of a microbial mat ecosystem from a unique permanent hypersaline inland lake: 'La Salada de Chiprana' (NE Spain). FEMS Microbiol Ecol 2003;44:175–89. https://doi.org/10.1016/S0168-6496(02)00464-6.

[41] Cardoso DC, Sandionigi A, Cretoiu MS, et al. Comparison of the active and resident community of a coastal microbial mat. Sci Rep 2017;7:2969. https://doi.org/10.1038/s41598-017-03095-z.

[42] Bolhuis H, Fillinger L, Stal LJ. Coastal microbial mat diversity along a natural salinity gradient. PLoS One 2013;8(5):e63166. https://doi.org/10.1371/journal.pone.0063166.

[43] Dijkman NA, Boschker HTS, Stal LJ, et al. Composition and heterogeneity of the microbial community in a coastal microbial mat as revealed by the analysis of pigments and phospholid- derived fatty acids. J Sea Res 2010;63:62–70. https://doi.org/10.1016/j.seares.2009.10.002.

[44] Cerritos R, Vinuesa P, Eguiarte LE, et al. *Bacillus coahuilensis* sp. nov., a moderately halophilic species from a dessication lagoon in Cuatro Cienegas valley in Coahuila, Mexico. Int J Syst Evol Microbiol 2008;58:919–23. https://doi.org/10.1099/ijs.0.64959-0.

[45] Alcaraz LD, Olmedo G, Bonilla G, et al. The genome of *Bacillus coahuilensis* reveals adaptations essential for survival in the relic of an ancient marine environment. Proc Natl Acad Sci U S A 2008;105(15):5803–8. https://doi.org/10.1073/pnas.0800981105.

[46] Siddiqui KS, Cavichioli R. Cold-adapted enzymes. Annu Rev Biochem 2006;75: 403–33. https://doi.org/10.1146/annurev.biochem.75.103004.142723.

[47] Peeters K, Verleyen E, Hudgson DA, et al. Heterotrophic bacterial diversity in aquatic microbial mat communities from Antarctica. Polar Biol 2012;35:543–54. https://doi.org/10.1007/s00300-011-1100-4.

[48] De Los Ríos A, Ascaso C, Wierzchos J, et al. Microstructure and cyanobacterial composition of microbial mats from the High Arctic. Biodivers Conserv 2015;24: 841–63. https://doi.org/10.1007/s10531-015-0907-7.

[49] Vincent WF, Gibson JAE, Pienitz R, et al. Ice shelf microbial ecosystem in the high Arctic and implications for life on snowball earth. Naturwissenschaften 2000;87: 137–41. https://doi.org/10.1007/s001140050692.

[50] Tytgat B, Verleyen E, Obbels K, et al. Bacterial diversity assessment in Antarctic terrestrial and aquatic microbial mats: a comparison between bidirectional pyrosequencing and cultivation. PLoS One 2014;9(6):1–11. https://doi.org/10.1371/journal.pone.0097564.

[51] Varin T, Lovejoy C, Jungblut AD, et al. Metagenomic analysis of stress genes in microbial mat communities from Antarctica and the high Arctic. Appl Environ Microbiol 2011:549–59. https://doi.org/10.1128/AEM.06354-11.

[52] Taton A, Grubisic S, Brambilla E, et al. Cyanobacterial diversity in natural and artificial microbial mats of Lake Fryxell (McMurdo dry valleys, Antarctica): a

morphological and molecular approach. Appl Environ Microbiol 2003:5157–69. https://doi.org/10.1128/AEM.69.9.5157-5169.2003.
[53] López-López O, Cerdán ME, González-Siso MI. Hot spring metagenomics. Life 2013; 3(2):308–20. https://doi.org/10.3390/life3020308.
[54] Rothschild LJ, Mancinelli R. Life in extreme environments. Nature 2001;409: 1092–101. https://doi.org/10.1038/35059215.
[55] Hongmei J, Aitchison JC, Lacap DC, et al. Community phylogenetic analysis of moderately thermophilic cyanobacterial mats from China, the Philippines and Thailand. Extremophiles 2005;9:325–32. https://doi.org/10.1007/s00792-005-0456-1.
[56] Portillo MC, Sririn V, Kanoksilapatham W, et al. Differential microbial communities in hot spring mats from Western Thailand. Extremophiles 2009;13:321–31. doi: https://doi.org/10.1007/s00792-008-0219-x.
[57] Mackenzie R, Pedrós-Alió C, Díez B. Bacterial composition of microbial mats in hot springs in Northern Patagonia: variations with seasons and temperature. Extremophiles 2013;17:123–36. https://doi.org/10.1007/s00792-012-0499-z.
[58] Huang Q, Dong CZ, Dong RM, et al. Archaeal and bacterial diversity in hot springs on the Tibetan Plateau, China. Extremophiles 2011;15(5):549–63. https://doi.org/10.1007/s00792-011-0386-z.
[59] Coman C, Druga B, Hegedus A, et al. Archaeal and bacterial diversity in two hot spring microbial mats from a geothermal region in Romania. Extremophiles 2013;17:523–34. https://doi.org/10.1007/s00792-013-0537-5.
[60] Lacap DC, Barraquio W, Pointing SB. Thermophilic microbial mats in a tropical geothermal location display pronounced seasonal changes but appear resilient to stochastic disturbance. Environ Microbiol 2007;9(12):3065–76. https://doi.org/10.1111/j.1462-2920.2007.01417.x.
[61] Thiel V, Wood JM, Olsen MT, et al. The dark side of the mushroom spring microbial mat: life in the shadow of Chlorophototrophs I microbial diversity based on 16S rRNA gene amplicons and metagenome sequencing. Front Microbiol 2016;7:919. https://doi.org/10.3389/fmicb.2016.00919.
[62] Thiel V, Hugler M, Ward DM, et al. The dark side of the mushroom spring microbial mat: life in the shadows if Chlorophototrophs II. Metabolic functions of abundant community members predicted from metagenomic analyses. Front Microbiol 2017;8:943. https://doi.org/10.3389/fmicb.2017.00943.
[63] Ferris MJ, Ward DM. Seasonal distributions of dominant 16S rRNA-defined populations in hot spring microbial mat examined by denaturing gradient gel electrophoresis. Appl Environ Microbiol 1997;63(4):1375–81.
[64] Prieto-Barajas CM, Alfaro-Cuevas R, Valencia-Cantero E, et al. Effect of seasonality and physicochemical parameters on bacterial communities in two hot spring microbial mats from Araró, México. Rev Mex Biodivers 2017;88(3):616–24. doi.org/https://doi.org/10.1016/j.rmb.2017.07.010.
[65] Baker BJ, Banfield JF. Microbial communities in acid mine drainage. FEMS Microbiol Ecol 2003;44:139–52. https://doi.org/10.1016/S0168-6496(03)00028-X.
[66] Bond PI, Smriga SP, Banfield JF. Phylogeny of microorganisms populating a thick, subaerial, predominantly lithotrophic biofilm at an extreme acid mine drainage site. Appl Environ Microbiol 2000;66(9):3842–9. https://doi.org/10.1128/AEM.66.9.3842-3849.2000.
[67] Beam JP, Bernstein HC, Jay ZJ, et al. Assembly and succession of iron oxide microbial mat communities in acidic geothermal springs. Front Microbiol 2016;7:25. https://doi.org/10.3389/fmicb.2016.00025.
[68] Kozubal MA, Macur RE, Jay ZJ, et al. Microbial iron cycling in acidic geothermal springs of Yellowstone National Park: integrating molecular surveys, geochemical processes, and isolation of novel Fe-active microorganisms. Front Microbiol 2012; 3(109):1–16. https://doi.org/10.3389/fmicb.2012.00109.
[69] Kozubal MA, Romine M, deM Jennings R, et al. Geoarchaeota: a new candidate phylum in the archaea from high-temperature acidic iron mats in Yellowstone National Park. ISME J 2013;7:622–34. https://doi.org/10.1038/ismej.2012.132.
[70] Handelsman J. Metagenomics: application of genomics to uncultured microorganisms. Microbiol Mol Biol Rev 2004;68:669–85. https://doi.org/10.1128/MMBR.68.4.669-685.2004.
[71] Donlan RM. Biofilms: microbial life on surfaces. Emerg Infect Dis 2002;8(9): 881–90. Doi: https://doi.org/10.3201/eid0809.020063.
[72] Visscher PT, Stolz JF. Microbial mats as bioreactors: populations, processes, and products. Palaeogeogr Palaeoclimatol Palaeoecol 2005;219:87–100. https://doi.org/10.1016/j.palaeo.2004.10.016.
[73] Fike DA, Gammon CL, Ziebis W, et al. Micron-scale mapping of sulfur cycling across the oxycline of a cyanobacterial mat: a paired nanoSIMS and CARD-FISH approach. ISME J 2008;2:749–59. https://doi.org/10.1038/ismej.2008.39.
[74] Javor BJ, Castenholtz RW. Laminated microbial mats, Laguna Guerrero Negro, Mexico. Geomicrobiol J 1981;2(3):237–73. https://doi.org/10.1080/01490458109377766.
[75] Ferris MJ, Ruff-Roberts AL, Kopczynski ED, et al. Enrichment culture and microscopy conceal diverse thermophilic *Synechococcus* populations in a single hot spring microbial mat habitat. Appl Environ Microbiol 1996;62(3):1045–50.
[76] Pajares S, Bonilla-Rosso G, Travisano M, et al. Mesocosms of aquatic bacterial communities from Cuatro Cienegas basin (Mexico): a tool to test bacterial community response to environmental stress. Microb Ecol 2012;64:346–58. https://doi.org/10.1007/s00248-012-0045-7.
[77] Ramos VMC, Castelo-Branco R, Leao PN, et al. Cyanobacterial diversity in microbial mats from the hypersaline lagoon system of Araruama, Brazil: an in-depth polyphasic study. Front Microbiol 2017;8:1233. https://doi.org/10.3389/fmicb.2017.01233.
[78] Dillewijn P, Villadas PJ, Toro N. Effect of a Sinorhizobium meliloti strain with a modified putA gene on the rhizosphere microbial community of alfalfa. Appl Environ Microbiol 2002;68(9):4201–8. https://doi.org/10.1128/AEM.68.9.4201-4208.2002.
[79] Benlloch S, Acinas SG, Antón J, et al. Archaeal biodiversity in crystallizer ponds from a solar saltern: culture versus PCR. Microb Ecol 2001;41:12–9. https://doi.org/10.1007/s00248000006.
[80] Brito EMS, Villegas-Negret N, Sotelo-González IA, et al. Microbial diversity in Los Azufres geothermal field (Michoacán, Mexico) and isolation of representative sulfate and sulfur reducers. Extremophiles 2014;18(2):383–98. https://doi.org/10.1007/s00792-013-0624-7.
[81] Ward DM, Bateson MM, Ferris MJ, et al. Cyanobacterial ecotypes in the microbial mat community of Mushroom Spring (Yellowstone National Park, Wyoming) as species-like units linking microbial community composition structure and function. Philos Trans R Soc B 2006;361:1997–2008. https://doi.org/10.1098/rstb.2006.1919.
[82] Gomez-Alvarez V, Teal TK, Schmidt TM. Systematic artifacts in metagenomes from complex microbial communities. ISME J 2009;3:1314–7. https://doi.org/10.1038/ismej.2009.72.
[83] Herlemann DPR, Lundin D, Labrenz M, et al. Metagenomic de novo assembly of an aquatic representative of verrucomicrobial class Spartobacteria. MBio 2013;4(3): e00569-2. https://doi.org/10.1128/mBio.00569-12.
[84] Klatt CG, Wood JM, Rusch DB, et al. Community ecology of hot spring cyanobacterial mats: predominant populations and their functional potential. ISME J 2011;5:1262–78. https://doi.org/10.1038/ismej.2011.73.
[85] Klatt CG, Liu Z, Ludwig M, et al. Temporal metatranscriptomic patterning in phototrophic Chloroflexi inhabiting a microbial mat in a geothermal spring. ISME J 2013;7:1775–89. https://doi.org/10.1038/ismej.2013.52.
[86] Drewniak L, Krawczyk PS, Mielnicki S, et al. Physiological and metagenomic analyses of microbial mats involved in self-purification of mine waters contamined with heavy metals. Front Microbiol 2016;7:1252. https://doi.org/10.3389/fmicb.2016.01252.
[87] Charlesworth J, Burns BP. Extremophilic adaptations and biotechnological applications in diverse environments. AIMS Microbiol 2016;2(3):251–61. https://doi.org/10.3934/microbiol.2016.3.251.
[88] Brock TD, Freeze H. *Thermus aquaticus* gen. n. and sp. n., a nonsporulating extreme thermophile. J Bacteriol 1969;98(1):289–97.
[89] Elleuche S, Schäfers C, Blank S, et al. Exploration of extremophiles for high temperature biotechnological processes. Curr Opin Microbiol 2015;25:113–9. https://doi.org/10.1016/j.mib.2015.05.011.
[90] Dobretsov S, Abed RMM, Al Maskari SMS, et al. Cyanobacterial mats from hot springs produce antimicrobial compounds and quorum-sensing inhibitors under natural conditions. J Appl Phycol 2010;23:983–93. https://doi.org/10.1007/s10811-010-9627-2.
[91] Putri SP, Kinoshita H, Ihara F, et al. Ophiosetin, a new tetramic acid derivative from the mycopathogenic fungus, *Elahocordyceps ophioglossoides*. J Antibiot 2010;63: 195–8. https://doi.org/10.1038/ja.2010.8.
[92] Mahajan GB, Balachandran LB. Sources of antibiotics: hot springs. Biochem Pharmacol 2017;134:35–41. https://doi.org/10.1016/j.bcp.2016.11.021.
[93] Stallwood B, Shears J, Williams PA, et al. Low temperature bioremediation of oil-contaminated soil using biostimulation and bioaugmentation with a *Pseudomonas* sp. from maritime Antarctica. J Appl Microbiol 2005;99:794–802. https://doi.org/10.1111/j.1365-2672.2005.02678.x.
[94] Feller G. Psychrophilic enzymes: From folding to function and biotechnology. Forensic Sci 2013;2013:512840. https://doi.org/10.1155/2013/512840.
[95] Roeselers G, van Loosdrecht MCM, Muyzer G. Phototrophic biofilms and their potential applications. J Appl Phycol 2008;20:227–35. https://doi.org/10.1007/s18011-007-9223-2.
[96] Adessi A, Corneli E, De Philippis R. Photosynthetic purple non sulfur bacteria in hydrogen producing systems: New approaches in the use of well known and innovative substrates. In: Hallenbeck PC, editor. Modern topics in the phototrophic prokaryotes. Quebec, Canada: Springer; 2017. p. 321–50. https://doi.org/10.1007/978-3-319-46261-5.
[97] Tripathi C, Mahato NK, Rani P, et al. Draft genome sequence of *Lampropedia coharens* strain CT6^T isolated from arsenic rich microbial mats of a Himalayan hot water spring. Stand Genomic Sci 2016;11:64. https://doi.org/10.1186/s40793-016-0179-1.
[98] Cohen Y. Bioremediation of oil by marine microbial mats. Int Microbiol 2002;5(4): 189–93. https://doi.org/10.1007/s10123-002-0089-5.
[99] Nielsen M, Revsbech NP, Kühl M. Microsensor measurements of hydrogen gas dynamics in cyanobacterial microbial mats. Front Microbiol 2015;6:726. https://doi.org/10.3389/fmicb.2015.00726.
[100] Langille MG, Zaneveld J, Caporaso JG, et al. Predictive functional profiling of microbial communities using 16S rRNA marker gene sequences. Nat Biotechnol 2013; 31(9):814–21. https://doi.org/10.1038/nbt.2676.
[101] Sauder LA, Albertsen M, Engel K, et al. Cultivation and characterization of Candidatus *Nitrosocosmicus exaquare*, an ammonia-oxidizing archaeon from a municipal wastewater treatment system. ISME J 2017:1–16. https://doi.org/10.1038/ismej.2016.192.