Advances in Environmental Microbiology 7

Christon J. Hurst Editor

The Structure and Function of Aquatic Microbial Communities



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Volume 7

Series Editor

Christon J. Hurst Cincinnati, Ohio USA

and

Universidad del Valle Santiago de Cali, Valle Colombia



Periodic spring located adjacent to the Miller-Leuser Log House in Anderson Township, Hamilton County, Ohio, United States of America. Photo by Christon J. Hurst, used with permission

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The Structure and Function of Aquatic Microbial Communities



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Universidad del Valle Santiago de Cali, Valle, Colombia

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Can there be a combination of place and time which leaves a perfect memory, a place to which you do not return because you know that it will have changed? I believe that to be true. During the months of July, August and September of 1998 I was a Fulbright Senior Scholar in Cali, Colombia, staying in what was then named the Hotel Pacifico Royal. Because I was to be living there for three months, the hotel gave me a free upgrade to a two room suite complete with a small dining table. The university was in fiscal crisis, and so I agreed ahead of time to use my salary from the Fulbright Commission to pay for my own lodging and meals. The financial arrangement meant that my meals had to be inexpensive. My evening meal usually was only what I could heat in my rooms' microwave oven, and generally all that I could afford were the corn meal cakes called arepas plus some marmalade spread on top of them. Therefore, I initially had little need for a dining table in my hotel room.

The hotel employees were very kind to me and quickly became my friends. Breakfast

fortunately was provided by the hotel and cooked by the breakfast buffet chef, Rocío. She smilingly reminded me that she could prepare whatever kind of omelet I might want. But, Rocío knew that for breakfast I only ate croissants with of course marmalade, and that I drank hot chocolate. On days when I seemed to arrive late for breakfast, Rocío would have saved for me a plateful of fresh croissants on a shelf under the buffet table and hot chocolate would be awaiting me. Once each two weeks I washed my laundry in the hotel room bathtub and then hung up the wet laundry all around my room for drying. The chambermaid, Blanca, told me that the hotel offered laundry service, but I said that I had no money to afford it. Blanca eventually told me that the hotel would dry all of my clothes for free! That was very kind, but I felt too guilty to ask the assistance. Two weeks later, when my suite was again filled with wet laundry, Blanca once more offered to have the clothes dried for me. I thought about it, and asked if they could dry just my jeans because those took three days to air dry in the summer humidity. *Following that, each morning after I had done* my laundry the wet jeans would disappear only to magically reappear a few hours later dried and carefully folded. The hotel men's soccer team adopted me as their unofficial mascot and made sure that I got to each of their practice sessions and games. I eventually learned that one time their team even had hired the car that transported me to the game and back. I remember José the doorman. who always was kind and never would accept a tip from me because I was his friend. Plus, of course, there was the concierge María

Fernanda whose kindness made everyone around her feel touched by her presence.

I eventually did find a good usage for that table. Each workday I walked through a shopping mall during trips between my hotel and the university. After a couple of weeks, I noticed that a stationerv store in the shopping mall sold wet clay. My hobbies included pottery and ceramic sculpture, and with that I found a solution for occupying my spare time at the hotel. I covered the top of my dining table with plastic, paper, and on top of that I placed some heavy canvas purchased from a fabric store in the shopping mall. The clay contained sticks and pebbles that needed to be removed before the clay could be worked, and with a few simple tools plus a ceramic ashtray used as a rolling pin I made presents for the hotel staff. Almost all of those things that I made at the dining table can be seen in this picture.



Sculpture made in Cali Rm 902 Hotel Pacifico Royal 1998

I had no means of firing those objects, and so they were given away air dried. When my

birthday came around, María Fernanda quietly collected enough money as donations from the hotel staff to buy a glass of wine for me from the hotel's bar. At her request, the hotel kitchen volunteered a slice a cake. I was out to a museum on that day, but everyone who could wait did stav until I returned to the hotel. Together, my friends at the hotel then presented to me the cake and wine, and they sang Happy Birthday to me. I again spent some time at the hotel in 2000, and although staying there was similar and very nice, something of what had seemed magical during that summer in 1998 was a bit changed. I have not returned to that hotel because, although I would imagine it currently is a very nice place, I want my memories to remain as perfect as it all seemed in 1998. To whom among those people would I dedicate my work on this book? Most certainly it should be María Fernanda, for that kind specialness in her soul which I am sure leaves everyone believing the world is a *better place.*



María Fernanda Gutiérrez Herrera in Cali 2000

Series Preface

The light of natural philosophy illuminates many subject areas including an understanding that microorganisms represent the foundation stone of our biosphere by having been the origin of life on Earth. Microbes therefore comprise the basis of our biological legacy. Comprehending the role of microbes in this world which together all species must share, studying not only the survival of microorganisms but as well their involvement in environmental processes, and defining their role in the ecology of other species, does represent for many of us the Mount Everest of science. Research in this area of biology dates to the original discovery of microorganisms by Antonie van Leeuwenhoek, when in 1675 and 1676 he used a microscope of his own creation to view what he termed "animalcula," or the "little animals" which lived and replicated in environmental samples of rainwater, well water, seawater, and water from snow melt. van Leeuwenhoek maintained those environmental samples in his house and observed that the types and relative concentrations of organisms present in his samples changed and fluctuated with respect to time. During the intervening centuries we have expanded our collective knowledge of these subjects which we now term to be environmental microbiology, but easily still recognize that many of the individual topics we have come to better understand and characterize initially were described by van Leeuwenhoek. van Leeuwenhoek was a draper by profession and fortunately for us his academic interests as a hobbyist went far beyond his professional challenges.

It is the goal of this series to present a broadly encompassing perspective regarding the principles of environmental microbiology and general microbial ecology. I am not sure whether Antonie van Leeuwenhoek could have foreseen where his discoveries have led, to the diversity of environmental microbiology subjects that we now study and the wealth of knowledge that we have accumulated. However, just as I always have enjoyed reading his account of environmental microorganisms, I feel that he would enjoy our efforts through this series to summarize what we have learned. I wonder, too, what the microbiologists of still future centuries would think of our efforts in comparison with those now unimaginable discoveries which they will have achieved. While we study the many wonders of microbiology, we also

further our recognition that the microbes are our biological critics, and in the end they undoubtedly will have the final word regarding life on this planet.



Christon J. Hurst in Heidelberg

Indebted with gratitude, I wish to thank the numerous scientists whose collaborative efforts will be creating this series and those giants in microbiology upon whose shoulders we have stood, for we could not accomplish this goal without the advantage that those giants have afforded us. The confidence and very positive encouragement of the editorial staff at Springer DE has been appreciated tremendously and it is through their help that my colleagues and I are able to present this book series to you, our audience.

Cincinnati, OH

Christon J. Hurst

Volume Preface

Microbial communities exist in all potential aquatic habitats including surface water, aquifers, and discarded materials on which precipitation collects. Some members of those microbial communities will naturally be floating on the surface, many others will live suspended at particular depths in the water column, a broad range of microorganisms make their existence in sediments, and still there are other communities that attach to solid matrices and contribute to the formation of biofilms.

The authors of this book describe how aquatic microbial communities are structured in ways that optimize the community's functioning and the authors further explain that community structuration often includes functional stratification. Structuration can be visibly obvious in biofilms including the presence of layers that typically extend from a community's surface of attachment outward into the aqueous surroundings. Vertical stratification often occurs within photic zones, and the photosynthetic pigments produced by microbes in photic biofilms may be evident as color banding, with the banding representing a community structured on the basis of optimal wavelength usage. In areas of low disturbance by metazoans, that vertical banding of photic communities can include seasonal laminations. The authors also explain that aquatic utilization of organic detritus as an energy source often begins with fungal colonization which starts a food chain. Further processing of sedimented organic material occurs by microbial communities that may be stratified on the basis of reduction potential, and their activity can include methanogenesis.

Preparing microbiologically safe drinking water requires eliminating hazardous components of the aquatic microbial community and often the processed water then needs to be safely distributed. *Vibrio cholerae* represents one example of pathogenic aquatic microorganisms for which consideration must be given whenever microbial ecology favors presence of that species in water that would be used for drinking. This book explains the techniques used for accomplishing microbial removal from water and also addresses the fact that drinking water distribution relies upon understanding and controlling aquatic environments which are designed to be isolated from their surroundings. The isolation is intended both to contain the product water and to reduce contamination caused by external impacts. Most

often, municipal communities will have organized utility services which supply drinking water via enclosed piping distribution systems. Some human communities, particularly those which may be remote and isolated, are not serviced by piping networks and instead reply upon tanker trucks for the transportation of drinking water. Even with those options, many individuals choose to purchase bottled drinking water and very often emergency situations temporarily may necessitate the supplying of bottled water. It must be understood that drinking water provided by any of those means will not be sterile. The authors provide an understanding of those microbial communities that are suspended in drinking water and also explain the microbial biofilms that develop on the inner walls of water distribution systems. We need to understand those microbial communities and control the health risk which they may present, both because some of the microbes in drinking water are pathogenic and because drinking water biofilms can interfere with water disinfection practices.

Studying aquatic microorganisms often entails identifying them, and for that reason this book additionally provides knowledge of the techniques that have been developed for successful isolation and cultivation of bacteria.

I am tremendously grateful to Andrea Schlitzberger, Markus Späth, and Isabel Ullmann at Springer DE, for their help and constant encouragement which has enabled myself and the other authors to achieve publication of this collaborative project.

Cincinnati, OH

Christon J. Hurst

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Chapter 1 Understanding Aquatic Microbial Communities



Christon J. Hurst

Abstract Aquatic environments are divided both physically and functionally into ecosystems whose community organizations include competitions as well as cooperations. Bodies of water that are small and shallow are more likely to be energetically dependent upon allochthonous nutrient inputs from the land. And, at the same time, small and shallow bodies of water will have less buffering capacity against the potential abruptness of fluctuations in allochthonous inputs. Larger bodies of water will by nature of their size have more buffering capacity against allochthonous impacts, and larger bodies of water also will be more reliant upon their autochthonous energy resources. All of the surfaces within aquatic systems contain biofilms, and in a sense it often takes a biofilm to nurture a microbe. Being part of a biofilm has both its blessings and curses, its benefits as well as limitations. Our task of understanding the nature of aquatic microbial communities requires recognizing interrelationships between the good, the bad, and the ugly, with slimy and smelly being part for the course.

1.1 Introduction

The life that we know on this planet has a suggested starting point of perhaps 4.5 billion years ago. That beginning likely occurred sometime after the presumed collision of earth with either a very large asteroid or a small planet, and that object has since been named Theia. The collision eventually resulted in creation of our moon. It is understood that the heat arising from such an impact event almost certainly would have sterilized the earth, eliminating any previously existing life. Following that impact the earth's surface eventually would have cooled sufficiently for liquid water to appear. We presume that the life now recognized on this planet

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began shortly after liquid water recollected, with microorganisms initially evolving and growing in that water. And so, in a sense, aquatic microbial life must be considered to represent the start of our biological heritage. Aquatic microbial life now exists from the very surface in all fresh, estuarine and oceanic waters, to the abyssal depths of the oceans. It has been suggested that we might even give a name to the common ancestor of all these life forms, with Glansdorff et al. (2008) having proposed that to be "Luca".

1.2 The Water Has Depth and Divides

In ponds and lakes, we describe vertical stratification of the water column from the surface to the bottom, according naming its three main layers the epilimnion, metalimnion and hypolimnion. The epilimnion is the uppermost layer and usually it will be the warmest because of sunlight. The epilimnion layer also will be oxic because water circulation within the epilimnion distributes atmospheric oxygen throughout that zone. The hypolimnion is the lowermost and generally coldest area of the water column, which also has a relatively low dissolved oxygen level because water circulation within that layer usually is limited to occurring between the bottom of the waterbody and the position of the thermocline. The metalimnion marks the thermocline, which is intermediate both with respect to its vertical position as well as its temperature and level of dissolved oxygen. Each of those zones has its microbial communities and their ecological activities. The thermocline can rise during the daylight hours and fall during the nighttime hours on a daily cycle during much of the year, depending upon the extent to which the surface water is heated by the sun. Very shallow water bodies may not have a thermocline at all. Moderately deep waterbodies, of perhaps a meter of so in depth, may have their thermocline sink to the bottom and disappear during the night and then the thermocline can subsequently reform and rise with the next days sunlight. Disappearance of the thermocline is important because it allows more oxygen to reach the otherwise relatively oxygen deprived bottom of the water column and sediments. I once received a teaching grant from El Instituto para el Control y la Conservación de la Cuenca Hidrográfica del Lago de Maracaibo (ICLAM), in Venezuela, where they interestingly designed the depth for a set of wastewater stabilization canals such that a thermocline would develop in those canals during the day, then the thermocline would sink and disappear each night. That daily thermocline pattern meant the sedimented solids could undergo alternating cycles of aerobic and anaerobic microbial activity. Moderately shallow lakes of perhaps a few meters may have their thermoclines disappear not daily but seasonally, completely dissipating during the winter and reforming during the spring. Quickly flowing bodies of water, such as streams, may experience sufficient mixing that thermoclines do not establish. The oceanic water column generally is divided into five layers called pelagic zones, and from top to bottom those are the epipelagic, mesopelagic, bathypelagic, abyssopelagic, and hadopelagic. Beneath the water column is the sediment, which has a low oxygen level at its surface and becomes anoxic not far beneath the sediment surface.

In a shallow lake with low turbidity, all of the water column may be photic zone meaning that sunlight penetrates even to the bottom such that photosynthesis can occur as a primary energy source throughout the system. In deeper lakes, and in the ocean, there will be a vast aphotic zone which underlies the photic zone. The amount of sunlight penetration into the aphotic zone is either too limited, or indeed nonexistent, such that other energy sources sometimes drive the deeper water lifecycles. However, with exception of locations such as deep sea hydrothermal vent plumes, most life in the column water and sediment is affected by what happens energetically higher in the water column, and most aquatic life utilizes food chains that are based upon photosynthesis (Cavan et al. 2018; Salter et al. 2012; Wolff et al. 2011).

There are aquatic microbial mechanisms, including those occurring among the communities in aquifers, which metabolize metals (Legg et al. 2012). Microbial mechanisms also help to cycle other natural elements including carbon (Painter et al. 2017; Poulton et al. 2017; Sanders et al. 2016), nitrogen (Painter et al. 2017), phosphorus (Davis et al. 2014; Painter et al. 2017; Poulton et al. 2017) and sulfur (Wasmund et al. 2017). Microbial life often converts those from elemental forms into organically useable forms, and sometimes back into elemental forms, done as enzymatic processes for deriving operating energy. Microbial processing of the minerals emitted from hydrothermal sources such as vent plumes have produced some amazing lifestyles (Dick et al. 2013). Aquatic microbial communities also have some capacity for degrading compounds such as spilled natural gas and oil (Redmond and Valentine 2012).

1.3 And Its Ecosystems Are Filled with Amazing Life Forms

Aquatic environments are divided into ecosystems. When we view from a large perspective some small body of water, such as a pond or stream which is narrow and shallow, we typically see that the water has an upper fluid surface exposed to the atmosphere plus there will be inanimate solid surfaces including the bottom and sides. Some of those solid surfaces are completely and permanently submerged, but other surfaces may not be constantly covered with water. We visibly may notice some living aquatic plants and macroalgae, plus some vertebrates and invertebrates, all of which also represent aquatic surfaces contained within that body of water. There additionally will be debris including detritus that once was living. When we view the ocean from a boat that is distant from shore, we mostly see only the surface of the water and floating debris, but we presume that the other types of solid surfaces are present within that ocean even though we cannot see them. When we touch any of those solid surfaces, even the debris, we will notice that none of it feels clean, and instead all of it feels slimy because in fact something is growing on it. Even some of the smallest aquatic organisms have other life forms growing on them (Huq et al. 1983).

Chapter 2 of this book, "Relationship Between Lifestyle and Structure of Bacterial Communities and Their Functionality in Aquatic Systems" by Luca Zoccarato and Hans-Peter Grossart, pp. 13–52, helps us to understand that when we view the same body of water from a more narrow perspective we can perceive it consists of numerous microhabitats filled with microscopic life. Broadly grouped, those microhabitats include: the diffusion controlled water phase; a colloidal phase of nanogels and microgels; particles which may be either exudates, or carcasses, or aggregates; and the living biosphere which has among its components such things as algae, zooplankton, and fish. Each of those microhabitats represents a different range of chemical and physical characteristics. Among those characteristics are the associated abiotic factors such as light, temperature, and oxygen. There also are differences in the concentration and composition of organic matter. Some of the organic material will be particulate in nature, and some will be dissolved, although there often is not an absolute distinction between those two broad categories of organic matter. Much of the organic matter still is living! A major goal always is to eat and not be eaten.

The variables of a water body's size and land proximity, or volume and distance from the center of the waterbody to the shoreline, can be grouped and described as a factor which contributes to an aquatic ecosystems buffering capacity, with the areas in smaller bodies of water experiencing less buffering against terrestrial inputs. The ecosystems within smaller bodies of water, and the near shore areas of larger bodies of water, will be more subject to short term disruptions effected by allochthonous materials that arrive from terrestrial environments and also will be more susceptible to the short term changes in terrestrial inputs caused by climate fluctuations associated with local and regional weather patterns (Davis et al. 2018) including precipitation. It is important to remember that precipitation drives both land surface runoff and groundwater discharge. Loading received from terrestrial inputs will change both the microbial community composition and the community functions. Those allochthonous materials include plant and animal debris which may drive the aquatic carbon cycle, along with both natural and anthropogenic chemicals that wash in from the land surfaces.

The flow of air also brings both nutrients and often contaminants to the water. Windborne solids can travel huge distances, although their impact may be most noticed near to the shore (Griffin and Kellogg 2004). Those inputs are factors which can vary daily, they will include larger scale seasonality patterns, and also encompass very long scale climate change patterns. There even are vast swathes of the earth that have cycled between aquatic environments and dry land in relationship to water and land usage patterns, with examples being the Mesopotamian Marshes in Iraq, Iran and Kuwait. Kenya's Lake Turkana Basin additionally has experienced very long scale climate change patterns (Bloszies et al. 2015; Goldstein et al. 2017).

Larger bodies of water and also the distantly offshore areas of oceans will have their ecologies and carbon cycles more driven by sunlight that directly reaches the water's surface. In some places oceanic deep sea thermal vents additionally will make their contribution as a local basic energy source. And thus, the carbon cycles in those aquatic areas will have an autochthonous base.

Oceanic currents bring changes including a supply of nutrients to aquatic areas that are far distant from the shore, acting in much the same way as do rivers carrying inputs from the land surface to the near shore areas. Some of those nutrient changes which are due to oceanic currents indeed occur very far distant from land while other changes due to currents can occur relatively near to the shore. Those oceanic flow patterns often carry contaminants in addition to bringing inputs of nutrients. Fluctuations associated with oceanic currents generally are less erratic than are fluctuations associated with terrestrial rivers, although the fluctuations associated with oceanic currents which bring upwellings of cold, nutrient laden water (Hosegood et al. 2017) often result in plankton blooms that feed and increase fish populations. When those water flow patterns change they can disrupt planktonic growth, and have an opposite effect by causing fish populations to decline.

1.4 It Often Takes a Biofilm to Nurture a Microbe

Living organisms interact with their surrounding environment, being both affected by the environment and in return modifying it. Those actions involve the organisms collecting and taking in what they need from their surroundings and nearly simultaneously leaving behind what is, for them, unneeded refuse. The selection processes as to what is taken in versus left out are not random, they are in fact quite purposeful and for each species those processes both have driven evolution and been modified by evolution. And, generally, an organism never exists by itself. It is important to recognize the fact that life typically is a communal process and we must understand the key significance of microbial existence within a structured community.

When microorganisms exist in communities, the metabolic functions of individual organisms become interdependent and often competitive, and the community develops its communal characteristics. Interdependence within a community can mean selectively sharing metabolic products with other microorganisms and macroorganisms in ways that are mutually beneficial. One organism's requirements often are different from those of another, and what one microbe either intentionally overlooks or leaves as waste can represent the essential needs of another organism. That type of specialization leads to a community organization which efficiently allows one microbes trash to become another microbes treasure. The resulting community structuration may include metabolic zonation and also metabolic coupling. Competition often is only unilaterally beneficial and includes cheating, which can extend to the point of stealing needed resources from your competitors so as to energetically starve those competitors out of existence, or even just simply and outright eating your competitors.

There is an African proverb "It takes a village to raise a child", and by analogy it often takes a biofilm to nurture a microbe. Microbial communities very often develop on surfaces which form collection points. The resulting biofilms have both temporal and spatial heterogeneity as a consequence of the groups organizational development. That development is a process which strives to best utilize available nutrient and energy sources. Sometimes, the surface on which the biofilm exists has been chosen because the surface provides a basic energy source, such as the wall of a metal drinking water distribution pipe, a piece of organic detritus, or a living being that may become detritus due to actions of the accumulating biofilm. At other times, the surface may provide an anchorage point which facilitates communal exposure to an energy source such as sunlight. Energetically, there also may be organizational aspects which rely upon the availability of electron donors and acceptors, represented by reduction potential gradients within a biofilm. We humans are indeed a part of the biofilm which has formed on this rocky planet that we call our home. Our evolutionary path has been directed in part by the environment imposing requirements, and we undoubtedly have modified the environment by leaving a tale of changes in our wake. Through it all we must acknowledge and remember that microbes were our biological origin, biologically the microbes sustain us, and biologically the microbes will survive our lifetimes endpoint.

The characteristics and composition of microbial biofilms differ based upon where they form, the conditions under which they form, and the conditions under which they either continue to exist healthily or senesce. Chapter 3 of this book, "Biofilms: Besieged Cities or Thriving Ports?" by Otini Kroukamp, Elanna Bester and Gideon M. Wolfaardt, pp. 53–90 asks the question of whether biofilms which exist in flowing conditions are either more like thriving ports where there is organization and good to be found associated with everything that comes and goes, or if biofilms like besieged cities are potentially challenged by everything that comes near.

1.5 When Life Hits the Mat

Photosynthetic mats have a biofilm structure that is uniquely interwoven. Chapter 4 of this book, "Complex Structure but Simple Function in Microbial Mats from Antarctic Lakes" by Ian Hawes, Dawn Sumner and Anne Jungblut, pp. 91–120, describes and discusses the physical characteristics and cooperation found within the photosynthetic microbial mats which grow as benthic communities under the continuous ice cover of Antarctic lakes. Those authors explain that growth in such microbial communities shows a very high level of evolved structural organization. The community structure can include seasonal lamination because there is limited environmental disturbance and also because the habitats lack large metazoans that might disrupt or potentially even consume portions of the microbial community. Zonation in those communities is energetically driven and the authors examine the interacting dynamic forces which produce those mat structures.

1.6 The Fungi Will Get You If You Land in the Water

Fungi play a large role in aquatic recycling of organic carbon through their decomposition of detritus from plants and animals. That detritus represents a major source of nitrogen and phosphorus in addition to the obviousness of its carbon content. In chapter 5 of this book, "Fungal Decomposers in Freshwater Environments" by Vladislav Gulis, Rong Su and Kevin A. Kuehn, pp. 121–155, its authors explain that in aquatic environments the fungal biomass tends to have a dominate association with submerged leaf litter and wood. There, fungi valuably act by enzymatically breaking down the large plant polymers such as cellulose, hemicelluloses, lignins, and pectin. The kinetics of those enzymatic activities are affected by the chemical characteristics and oxygen levels of the surrounding water. Some of the fungal activity represents a direct recycling of nutrients to produce not only fungal elements which will be contained in the detritus but also fungal spores that will be released into the water. Much of the nutrients in that detritus, including the fungal elements themselves, will then be consumed by other aquatic microorganisms and macroorganisms. Photosynthetically active algae also play a role in the degradation of detritus, as do bacteria. Some algae are surface associated while others are suspended in the water column. The predominance of aquatic bacterial biomass is associated with fine particulate material.

1.7 Researching Microbiology Even When You Are Up to Your Waist in Alligators While Studying Respiration Without Oxygen, in a Swamp

Wetlands are a connecting point between aquatic and terrestrial life. They contain both aquatic and terrestrial characteristics and in a sense have their own combination of microbial activities. Seasonal wetlands can be aquatic environments during some time periods of the year, and terrestrial environments during other times, depending upon whether the water level is above versus below the ground surface. And, wetlands merge with one another and do as well merge with stream environments (Vanderhoof et al. 2016).

The Everglades is a wide and generally quite shallow river in Southern Florida of the United States, it now is a patterned peat land and serves as an example of a complex wetland with interdependent ecosystems. Peat is a product of partial plant material decomposition which occurs in wetlands (Hohner and Dreschel 2015) and represents long term carbon storage. Conversion of that plant material into coal would represent longer, geologic scale storage of its carbon. Chapter 6 of this book, "The Ecology of Methanogenic Archaea in a Nutrient Impacted Wetland" by Andrew Ogram, Hee-Sung Bae, and Ashvini Chauhan, pp. 157–172, explains that methane production is a key microbial characteristic of wetlands. The anoxic environment of peat systems results in slow decomposition of organic carbon. Therein, the methanogenesis that occurs is a form of anaerobic respiration which uses as its terminal electron acceptor any available atoms of carbon contained in either low molecular weight organic compounds or carbon dioxide, and the result is production of methane. Sulfate reduction also occurs in wetlands, where it is done by microorganisms that perform anaerobic respiration using sulfate as a terminal electron receptor and reduce that to hydrogen sulfide. Although it often is thought that methanogenesis generally occurs only when sulfates are depleted, both processes can occur in the same zonal area (Sela-Adler et al. 2017).

In a sense, methane production is an indicator of the microbial health of wetlands. Methane production is an anaerobic activity associated with archaea and that activity also takes place in the intestines of ruminants (Patra et al. 2017) and many other mammals. Methane production also occurs in anaerobic wastewater treatment processes (Qiao et al. 2015; Świątczak et al. 2017). Methanogenesis additionally occurs in landfills (Staley et al. 2012). The microbes which generate methane by those process are termed methanogens.

1.8 And Microbes Are even in the Water that We Would Want to Drink

Some of the microorganisms present in water cause disease in humans (Hurst 2018) including the notorious Vibrio cholerae (Ali et al. 2015; Azman et al. 2013) which is a commensal to crustaceans including those which are considered zooplankton, notably copepods (de Magny et al. 2011). Vibrio cholerae causes the disease cholera as explained in chapter 7 of this book, "Briefly Summarizing our Understanding of Vibrio cholerae and the Disease Cholera" by Hurst, pp. 173-184. One of our public health goals is removing hazardous microorganisms from drinking water, and for that purpose we use methods which vary in complexity and effectiveness. Water treatment processes are designed differently with regard to the volume of water that can be treated simultaneously (Hug et al. 2010; Hurst 2018) and the microbial community changes as a result of drinking water treatment processes (Liao et al. 2015). The procedures used for processing water to either remove hazardous microorganisms or destroy their infectiousness and thereby render the water safe for ingestion will differ depending upon the volume of water being treated. Sometimes the general treatment methodology is similar at large and small size scales but performed in different ways. The subject of supplying microbially safe drinking water is presented in chapter 8 of this book, "Options for Providing Microbiologically Safe Drinking Water" by Hurst, pp. 185-260.

Water treatment for large communities often is followed by efforts to safely distribute the treated water using community drinking water distribution networks that involving piping. Those drinking water distribution systems are enclosed man made aquatic environments colonized by a wide range of both microorganisms and small macroorganisms that arrive from several sources, as explained in chapter 9 of this book, "Microbiome of Drinking Water Distribution Systems" by Laurence Mathieu, Tony Paris and Jean-Claude Block, pp. 261–311. Organisms invariably will enter into the distribution system as constituents of the source water that is being distributed, even if that source water was processed by physical and chemical treatment processes. Those organisms will be joined by other biological

contaminants which represent accidental arrivals that have entered the distribution system in association with infiltrations related to piping engineering failures. Such failures include inadequate sealing of the pipe connections, as well as breakage of piping and pipe connections. Accidental cross contamination of the drinking water and septage collection systems represents yet another source of microorganisms. Consequently, the plumbing of drinking water networks are aquatic ecosystems which contain archaea, bacteria, fungi, viruses, protozoa, and small invertebrate metazoa, many of which certainly can cause disease in humans who ingest the water from distribution systems. That accumulation of biomass and its ecosystem structure must be understood and controlled, because the biomass in drinking water interferes with chemical disinfectant processes upon which we rely for delivering safe product water to consumers.

Normally we think of removing phosphorus as something done for treating domestic wastewater to help protect the natural ecosystem from eutrophication (Fig. 1.1). However, there also is some consideration that removing phosphorus during drinking water treatment beneficially may reduce the microbial growth that occurs within water distribution systems (Wang et al. 2014).

1.9 They Are more than Just Nameless Faces: There Are Ways to Culture and Identify even the Seemingly Ungrowable

Aquatic microbial communities contain a vast range of organisms, each with its own activities and necessary requirements. Often, all but some viruses may be culturable if the appropriate conditions can be met. The subject of "Isolation and Cultivation of Bacteria" is addressed in chapter 10 of this book by Martin W. Hahn, Ulrike Koll and Johanna Schmidt, pp. 313–351. The information which they summarize helps us to focus upon and understand an important objective. Its successful achievement often requires complicated technical processes because those microorganisms which represent the study goal have evolved to naturally exist and thrive in their own environmental milieu, under conditions that perhaps require very specific physical and chemical parameter ranges.

Selective physical isolation procedures, including particle size class separations, often can be developed as a helpful early step to aid with identifying the microbes present in an environmental sample. The next task is a requirement to replicate the conditions under which the microorganisms would have been growing in their natural environment. If existing cultivation media formulations prove inadequate, then it may be necessary to design new cultivation media and include some provision of appropriate energy sources such as light of particular wavelength ranges. Flow cytometry is one of the techniques that can be used to assess cultivation efficiency. Gene sequencing is the most accurate testing approach for determining if the resultingly isolated and cultivated organisms either are newly discovered or



Fig. 1.1 The image shows eutrophication at a waste water outlet in the Potomac River, Washington, D.C. It is titled "Potomac green water" by Alexandr Trubetskoy and used under the Creative Commons Attribution-Share Alike 3.0 license

previously have been described, and for learning if perhaps those organisms have previously even been named by other researchers.

Compliance with Ethical Standards

Conflict of Interest Christon J. Hurst declares that he has no conflict of interest.

Ethical Approval This article does not contain any studies with human participants or animals.

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