5 Ecology of Microorganisms in Freshwater

Abstract

Freshwaters are defined as natural waters containing less than 1,000 mg per liter of dissolved solids, most often salt. Globally, freshwaters are scarce commodities and make up only 0.009% of the earth's total water. Although they generate only about 3% of the earth's total primary biological productivity, they contain about 40% of the world's known fish species. Natural freshwaters are classifiable into atmospheric, surface, and underground waters each type having a unique microbial ecology. Atmospheric waters lose their microorganisms as they fall as rain or snow. Surface freshwaters are found in rivers and lakes, and contain large and diverse groups of microorganisms. Using molecular methods such as 16S rRNA analysis, which enables the study of unculturable microorganisms, new information regarding freshwater microbial ecology has emerged in recent times: There are more phylogenetic groups of bacteria than are observed by cultural methods; there is a unique and distinct bacterial group, which can be termed "typical freshwater bacteria"; contrary to previous knowledge when aquatic bacteria were thought to be mostly Gram-negative bacteria, Gram-positive bacteria are in fact abundant in freshwaters; finally, marine-freshwater transitional populations exist in coastal waters. Ground waters suffer contamination from chemicals and less from microorganisms; the deeper the groundwater, the less likely it is to contain microorganisms, which are filtered away by soil.

Keywords

Underground waters • Pollution of underground waters • Typical freshwater bacteria • Ingoldian fungi • Abundance of Gram-positive bacteria • Transitional microbial population in estuaries

5.1 Microbial Ecology of Atmospheric Waters

Atmospheric water occurs in the form of rain, snow, or hail; all three forms occur in temperate countries. In tropical countries, however, snow is unknown, except perhaps at the peaks of the high mountains which are found, in East Africa, the Cameroon, etc. Hail only falls occasionally. The major source of atmospheric water in tropical countries is therefore rain.

As it falls from clouds (which are themselves condensates of water vapor), rain water collects with it dissolved gases, dust particles, and microorganisms.

N. Okafor, *Environmental Microbiology of Aquatic and Waste Systems*,
DOI 10.1007/978-94-007-1460-1_5,

The result is that if rain falls for a sufficiently long period, rain water would have brought down dust particles and any microorganisms circulating in the atmosphere. Water resulting from such heavy and continuous rain in a rural area free of industrial gases, should be expected to be virtually sterile and almost of the quality of the distilled water prepared in the laboratory. It should be possible to collect such water when heavy rain has continued uninterrupted for upwards of 90 min, if proper aseptic conditions are followed. Indeed in parts of the rural areas of the developing world where water is scarce, harvested rain water often directly forms the only source of drinking water.

It should not be taken for granted, however, that microorganisms from the atmosphere make significant contributions (Jones et al. 2008) to aquatic systems as can be seen from Fig. 5.1.

5.2 Microbial Ecology of Surface Waters

Surface waters include rivers, streams, lakes, ponds, and wetlands. Surface waters may consist of either freshwater or saline water. Freshwater is water with a salt content (salinity) of less than 1 g L^{-1}, while saline waters (seas and oceans) are characterized by salinities >1 g L^{-1}. This chapter will examine the ecology of microorganisms in fresh water (Hahn 2006). The next chapter will look at marine microbiology.

5.2.1 Rivers and Streams

Fresh water systems can be divided into lotic systems composed of running water (rivers, streams, creeks, springs) and lentic systems which comprise still water (ponds and lakes).

5.2.2 Lakes and Ponds

Lakes have been defined as large ponds, the defining size varying according to authors. Thus, the size of defining lakes as opposed to ponds have varied from 5 acres (2 ha) through 12 acres (5 ha) to 20 acres (8 ha). Most lakes have a natural outflow in the form of a river or stream, but some do not, and lose water solely by evaporation or underground seepage or both. Many lakes are artificial and are constructed for hydro-electric power generation, recreational purposes, industrial use, agricultural use, or domestic water supply. Table 1.4 shows the world's largest lakes. Most of them are fresh water.

5.2.3 Wetlands

Wetlands are land pieces either temporarily or permanently submerged or permeated by water. They are characterized by plants adapted to soil conditions saturated by water. Wetlands include fresh and salt water marshes, wooded swamps, bogs, seasonally flooded forest, and sloughs.

Wetland vegetation is typically found in distinct zones that are related mainly to water depth and salinity. Since wetlands are partly defined by the vegetation found in them, the typical vegetation found in wetlands are described briefly below:

1. *Shoreline*: Plants that grow in wet soil on raised hummocks or along the shorelines of streams, ponds, bogs, marshes, and lakes. These plants grow at or above the level of standing water; some may be rooted in shallow water. Examples of plants are temperate climate shoreline plants, e.g., western coneflower (*Rudbeckia occidentalis* Nutt.) and buttonbush (*Cephalanthus occidentalis*). In tropical and semi-tropca climates, for example, the Florida everglades mangroves are common. These are salt tolerant plants climates, two examples of which are red mangrove (*Rhizophora mangle*) and black mangrove (*Avicennia germinans*).
2. *Emergent*: Plants that are rooted in soil that is underwater most of the time. These plants grow up through the water, so that stems, leaves and flowers emerge in air above water level. Some of the commonest plants in the emergent zone are arrowheads (*Sagittaria* spp.). Nearly all parts of the plants, including the roots are used by wild life as food. In Japan and China, they are used as human food. Among the most common wetland plants worldwide are cattails (*Typha* spp.).
3. *Floating*: Plants whose leaves mainly float on the water surface. Much of the plant body is underwater and may or may not be rooted in the substrate. Only small portions, namely flowers, rise above water level. Examples are duckweeds (family Lemnaceae), which are used for animal and human food and pondweeds (*Potamogeton* spp.).

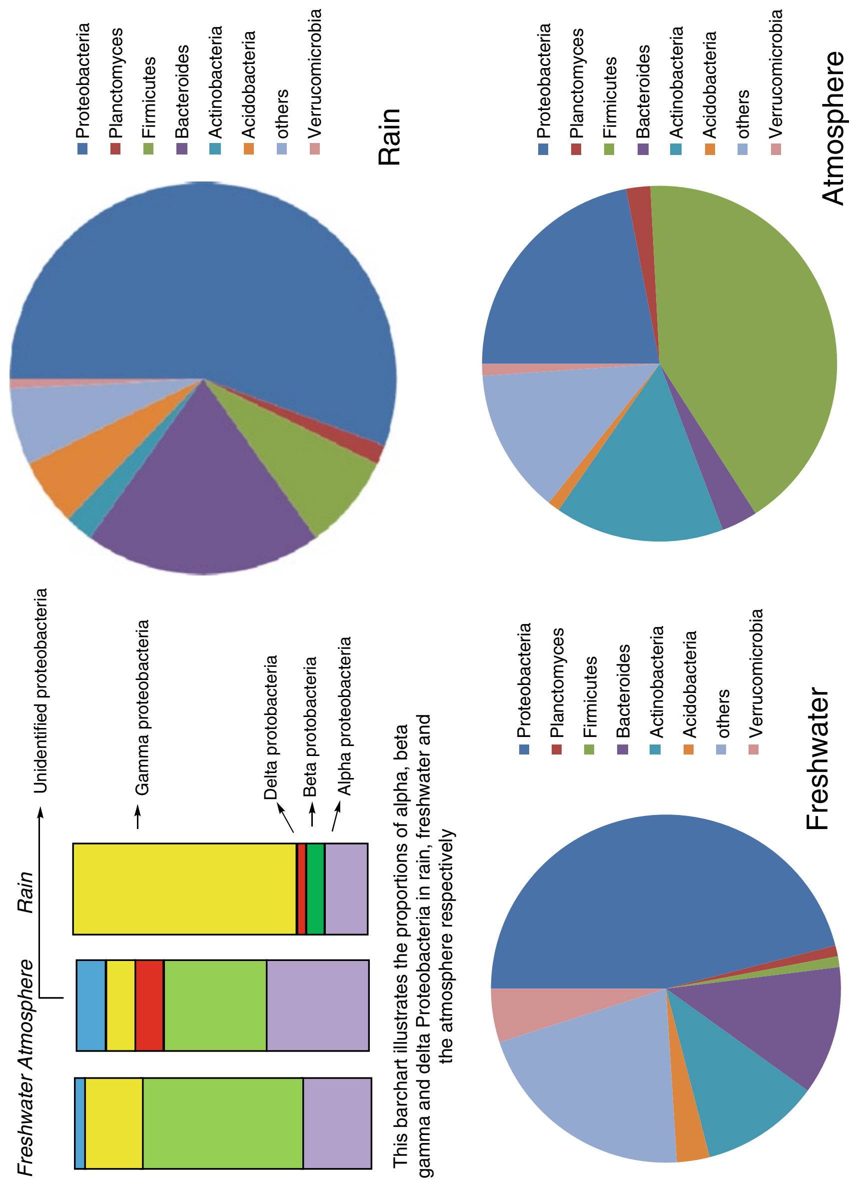

Fig. 5.1 Proportions of taxonomic groups of bacteria in the atmosphere, in rain water and in freshwater (From Jones et al. 2008. With permission)

4. *Submerged*: Plants that are largely underwater with few floating or emergent leaves. Flowers may emerge briefly in some cases for pollination. Examples are water milfoil (*Myriophyllum heterophyllum*) and watercress (*Rorippa nasturium-aquaticum*), a member of the mustard family (Cruciferae), used for food and medicinal purposes.

Wetlands are the only ecosystem designated for conservation by international convention. They have been recognized as particularly useful for the following reasons:

(a) They act as buffers and absorb the impact of water-related traumatic occurrences such as large waves or floods.
(b) They filter off sediments and toxic substances.
(c) They supply food and essential habitat for many species of fish, shellfish, shorebirds, waterfowl, and furbearing mammals.
(d) They provide products for food (wild rice, cranberries, fish, wildfowl), energy (peat, wood, charcoal), and building material (lumber).
(e) They provide avenues for recreation such as hunting, fishing, and bird watching.

Previously wetlands were considered wasteland, and in some countries, they were drained or filled in, so that they could be farmed or built upon. In recent times, however, the value of wetlands has been recognized and efforts have been made to protect them. However, they are still disappearing under the pressure of human activity, and are being threatened by air pollution and climate change.

Some of the steps taken in recent times to preserve wetlands are as follows:

1. Adding sediment to coastal wetlands to keep up with rising sea levels
2. Planting grass to protect coastal sands from erosion
3. Building dikes or barrier islands to protect the wetlands
4. Controlling water levels artificially to ensure the wetland is flooded

5.3 Ground Waters

Groundwater flows slowly through water-bearing formations (aquifers) at different rates. In some places, where groundwater has dissolved limestone to form caverns and large openings, its rate of flow can be relatively fast but this is exceptional.

Many terms are used to describe the nature and extent of the groundwater resource. The level below which all the spaces are filled with water is called the *water table*. Above the water table lies the *unsaturated zone*. Here, the spaces in the rock and soil contain both air and water. Water in this zone is called *soil moisture*. The entire region below the water table is called the *saturated zone*, and water in this saturated zone is called *groundwater*.

Groundwater is not confined to only a few channels or depressions in the same way that surface water is concentrated in streams and lakes. Rather, it exists almost everywhere underground. It is found underground in the spaces between particles of rock and soil, or in crevices and cracks in rock.

The water filling these openings is usually within 100 m of the surface. Much of the earth's fresh water is found in these spaces. At greater depths, because of the weight of overlying rock, these openings are much smaller, and therefore hold considerably smaller quantities of water.

Although groundwater exists everywhere under the ground, some parts of the saturated zone contain more water than others. An *aquifer* is an underground formation of permeable rock or loose material which can produce useful quantities of water when tapped by a well. Aquifer sizes vary. They may be small, only a few hectares in area, or very large, underlying thousands of square kilometers of the earth's surface. They may be only a few meters thick, or they may measure hundreds of meters from top to bottom.

Groundwater circulates as part of the hydrologic cycle. As precipitation and other surface water sources recharge the groundwater, it drains steadily, and sometimes very slowly, toward its discharge point.

When precipitation falls on the land surface, part of the water runs off into the lakes and rivers. Some of the water from melting snow and from rainfall seeps into the soil and percolates into the saturated zone. This process is called *recharge*. Places where recharge occurs are referred to as *recharge areas*.

Eventually, this water reappears above the ground. This is called *discharge*. Groundwater may flow into streams, rivers, marshes, lakes, and oceans, or it may discharge in the form of springs and, when tapped, wells.

The *residence time* of groundwater, i.e., the length of time water spends in the groundwater portion of the

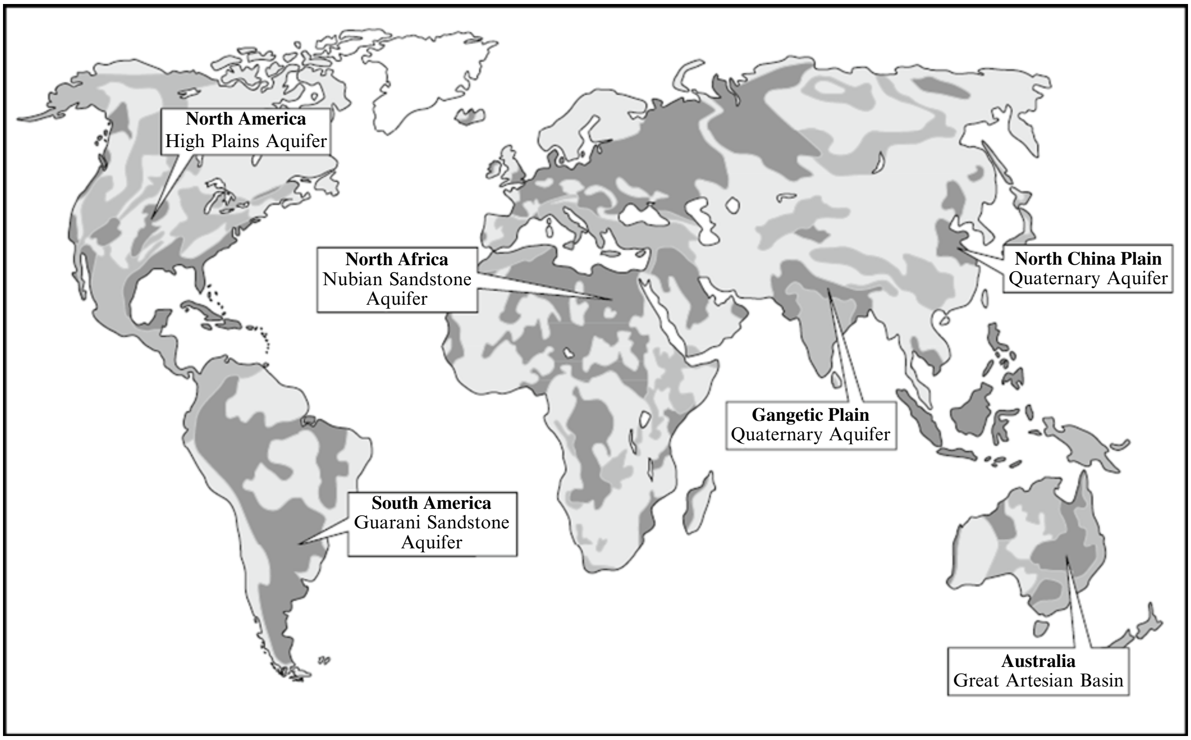

Fig. 5.2 Distribution of underground water world-wide (From Foster and Chilton 2003, with permission). Note: Hydrogeological map of the world showing widespread occurrence of geological formations containing useful ground water and the locations of some of the world's largest aquifers with vast storage reserves. The *dark gray* areas are major regional aquifers, while the midgrey areas contain some important but complex aquifers. The *light gray* areas contain only small aquifers of local importance

hydrologic cycle, is highly variable. Water may spend as little as days or weeks underground, or as much as 10,000 or more years.

Groundwater systems constitute the predominant reservoir and strategic reserve of freshwater storage on earth; about 30% of the global total and as much as 98% if that bound up in the polar ice caps and glaciers is discounted. As seen in Fig. 5.2, some aquifers extend quite uniformly over very large land areas and have much more storage than all of the world's surface reservoirs and lakes. In sharp contrast to surface water bodies, they lose very little of their stored water by direct evaporation.

Although there are country to country differences and local variations, globally, groundwater is estimated to provide at least 50% of current potable water supplies; 40% of the demand from those industries that do not use mains water, and 20% for water use in irrigated agriculture. Compared with surface water, groundwater use often brings large economic benefits per unit volume, because of ready local availability, high drought reliability and generally good quality requiring minimal treatment.

Groundwater contaminants come from two categories of sources: Point sources and distributed or non-point sources. Landfills, leaking gasoline storage tanks, leaking septic tanks, and accidental spills are examples of point sources. Infiltration from farm land treated with pesticides and fertilizers is an example of a non-point source.

Microorganisms are trapped by the particles of soil which act as components of a vast filter. The result of this is that the deeper the ground water, the less likely it is to have microorganisms. Waters collected from deep wells are therefore usually found to contain few bacteria. Artesian springs which erupt from the ground under great pressure have been found to contain less than 300 bacteria per ml. and it is doubtful whether such bacteria have not arisen from the side of the well or introduced during sampling. In very deep bores dug for the exploration of oil, sulfate-reducing bacteria e.g., *Desulphovibrio* have been reported, especially in

Table 5.1 Sources of deterioration of underground waters (Modified from Foster and Chilton 2003. With permission)

Type of problem	Cause	Parameters of concern
Salinization processes	Mobilization and/or fractionation of salinity due to inadequate management of ground water irrigation, mine reservoir exploitation drainage or petroleum; extensive and prolonged surface water irrigation without adequate drainage	Na, Cl and sometimes F, Br SO4
Anthropogenic pollution	Inadequate protection of vulnerable aquifers against man-made discharges/leachates from urban activities	Pathogens, NO_3, NH_4, Cl, SO_4, B, heavy metals, DOC, Aromatic and halogenated hydrocarbons, etc. NO3, Cl2, some pesticides and derivatives
Well head contamination	Inadequate well construction and completion, allowing direct ingress of polluted surface water or shallow ground water	Mainly pathogens, NO3, Cl
Naturally occurring contamination	Related to pH-Eh evolution of ground water and dissolution of minerals from aquifer matrix (can be aggravated by anthropogenic pollution and/or uncontrolled exploitation)	Mainly Fe, F, and sometimes As, I, Mn, Al, Mg, SO_4, Se and NO_3 (from paleo recharge): F and As in particular represent a serious public health hazard for potable supplies

the zones with oil-water mixtures. Some of such findings were made in bores as deep as 2,000 m.

In many parts of the world, drinking water is obtained from underground water. In the US, for example, over 50% of the United States population utilizes groundwater as its drinking water source. Approximately 96% of the groundwater users live in small rural areas often utilizing small individual wells, where the resources for treatment and monitoring are limited. While microorganisms can be filtered out by soil, chemicals are not; ground water is therefore frequently contaminated by chemicals.

Although ground waters are generally free of contaminants, the quality of ground waters can be compromised by both man-made and other sources (see Table 5.1).

5.4 Some Microorganisms Usually Encountered in Fresh Water

5.4.1 Bacteria

Early work (in the 1980s and earlier) on freshwater bacteria was based on bacteria which were culturable. Freshwater bacteria were merely grouped fresh water into:

(a) The flourescent group
(b) The chromogenic bacteria including violet, red and yellow forms
(c) The coliform group
(d) The Proteus group
(e) Non-gas-forming, non-chromogenic, non-spore-forming rods which do not produce Proteus-like colonies and may not acidify milk and liquify gelatin
(f) Aerobic spore-forming rods
(g) White, yellow, and pink cocci

It was believed that there was no clear separation between soil bacteria and aquatic bacteria and that fresh water bacteria did not have a unique population of its own. However, it soon became known that bacteria cultivated from the environment including the soil, and fresh and marine waters represented only a fraction of the actual total, as the bulk of such bacteria were not culturable (Oren 2004).

The advent of molecular techniques and especially the polymerase chain reaction (PCR) has made it possible to obtain information on microbial community composition directly, without cultivation. With new molecular techniques of the 1990s, it became possible to assess the microbial population of a natural environment through culture-independent techniques by isolating the nucleic acid in it, followed by the amplification and sequencing of bacterial 16S rRNA genes (Miskin et al. 1999).

5.4.1.1 New Data Regarding Freshwater Bacteria

Assessing the freshwater environment as described above through the work of Zwart and others (Zwart et al. 2002) led to new conclusions regarding the bacterial population of that environment. After analyzing available database of 16S rRNA sequences from freshwater plankton, including rivers and lakes in the USA, South Korea, and the Netherlands the following conclusions were reached (Table 5.2):

1. *More phylogenetic groups of bacteria observed by molecular methods*
   Most bacterial sequences retrieved from freshwater habitats were neither affiliated with known bacterial species nor represented those phylogenetic groups previously obtained from freshwater habitats using cultivation methods. Bacteria in the freshwater environment which could not be cultivated were observed by direct nucleic acid assessment.
2. *Existence of a unique and distinct bacterial group, "typical freshwater bacteria"*
   Sequences retrieved from freshwater habitats were most closely related to other freshwater clones, whereas relatively few were most closely related to sequences recovered from soil or marine habitats. Thus, there appears to be a specific group of bacteria which are typical of, and thus indigenous in, freshwater. Thirty-four phylogenetic clusters of bacteria representing typical inhabitants of freshwater systems were initially recorded but more have been since added.
3. *The abundance of Gram-positive bacteria in freshwater*
   Prior to the advent of molecular methods, it was shown, because of the limitations of the cultural methods, that most freshwater bacteria were Gram negative and only a few Gram positive bacteria were found therein. With the new techniques, it is now known that Gram positive bacteria, especially *Actinobacteria* are plentiful in freshwater.
4. *A marine-freshwater transitional population exists in coastal waters*
   Distinct bacterial populations exist in both marine and the freshwater environments. However in coastal areas, while there is a preponderance of marine bacteria, there are also some bacteria regarded as typically of the freshwater niche. This shows that waters in the coastal areas of the seas and oceans can be regarded as being transitional (Rappe et al. 2000).
5. *Some factors affecting the distribution of bacteria in freshwater*
   The factors which affect the distribution of bacteria in freshwater are complex. At present, not much information is available, but some factors affecting the nature of the bacterial community in a body of fresh water include the following: Water chemistry (including pH), water temperature, metazooplankton predation, protistan predation, phytoplankton composition, organic matter supply, intensity of ultraviolet radiation, habitat size, and water retention time (Lindstrom et al. 2005).

Table 5.2 Examples of "typical freshwater bacteria" (Modified from Zwart 2002. With permission)

S/No	Bacterial cluster name	Division
1	LD12	α-*Proteobacteria*
2	*Brevundimonas intermedia*	α-*Proteobacteria*
3	CR-FL11	α-*Proteobacteria*
4	GOBB3-C201	α-*Proteobacteria*
5	*Novosphingobium subarctica*	β-*Proteobacteria*
6	*Polynucleobacter necessaries*	β-*Proteobacteria*
7	LD28	β-*Proteobacteria*
8	GKS98	β-*Proteobacteria*
9	*Ralstonia picketti*	β-*Proteobacteria*
10	*Rhodoferax* sp. Bal47	β-*Proteobacteria*
11	GKS16	γ-*Proteobacteria*
12	*Methylobacter psychrophilus*	Verrucomicrobia
13	CL120-10	*Verrucomicrobia*
14	CL0-14	*Verrucomicrobia*
15	FukuN18	*Verrucomicrobia*
16	Sta2-35	*Verrucomicrobia*
17	LD19	*Actinobacteria*
18	ACK-m1	*Actinobacteria*
19	STA2-30	*Actinobacteria*
20	MED0-06	*Actinobacteria*
21	URK0-14	*Actinobacteria*
22	CL500-29	*Cytophaga-Flavobacterium-Bacteroides*
23	LD2	*Cytophaga-Flavobacterium-Bacteroides*
24	FukuN47	*Cytophaga-Flavobacterium-Bacteroides*
25	PRD01a001B	*Cytophaga-Flavobacterium-Bacteroides*
26	CL500-6	*Cytophaga-Flavobacterium-Bacteroides*
27	*Synechococcus* 6b	*Cyanobacteria*
28	*Planktothrix agardhii*	*Cyanobacteria*
29	*Aphanizomenon flos aquae*	*Cyanobacteria*
30	*Microcystis*	*Cyanobacteria*
31	CL500-11	Green non-sulfur bacteria
32	CLO-84	OP10
33	CL500-15	*Planctomyces*

5.4.1.2 Some Bacteria in Freshwater

More recent publications list the organisms, but it is clear from these that much work remains to be done in fresh waters of various categories, especially in tropical countries for a rounded picture to be built.

The nature of the bacteria flora of a fresh water varies. The bulk of such bacteria are heterotrophic while a small proportion are photo- or chemo-autotrophic.

In oligotrophic surface waters, e. g., springs recently emerging from their source, the bacterial population consists among the nonbenthic population largely of Gram-negative, non-spore- forming rods, especially *Achromobacter* and *Flavobacterium.* With increasing eutrophication, *Pseudomonas, Proteus, Bacillus,* and *Enterobactetriaceae* become more important. Other bacteria usually encountered include *Vibrio* and *Actinomycetales.* When eutrophication is heavy, and due to organic material. *Zooglea* tends to occur; if the water is lotic or fast flowing, then the "sewage fungus" *Sphaerotilus natans* may also develop. Most of the above-mentioned bacteria are Gram-negative; in general, Gram-negative bacteria are planktonic or tectonic. Among the "Aufwuchs" are actinomycetes, which are usually epiphytic. Most benthic bacteria are generally Gram-positive and include spore-formers, and Gram-positive cocci, *Clostridium,* and pleomorphic forms (e.g., *Arthrobacter, Nocardia*).

Chemo-autotrophic bacteria, e. g., *Nitrosomonas* and *Nitrobacter* are found in some lake waters while photosynthetic bacteria are found in some rivers. Certain aquatic bacteria are considered as nuisance bacteria in drinking water. These include iron, sulfur, and sulfate-reducing bacteria. These nuisance bacteria may cause odor, taste or turbidity in water as well as destroy water pipes. "Iron" bacteria withdraw iron which is present in the environment and deposit it in the form of hydrated ferric hydroxide in mucilaginous secretions· and this imparts a reddish tinge to water and may stain clothes. Some well-known iron bacteria genera include *Sphaerotilus, Leptothrix, Toxothrix, Crenothrix, Callionella, Siderobacter,* and *Ferrobacillus.*

The sulfur bacteria include the green photosynthetic and nonphotosynthetic members. Many of the photosynthetic bacteria produce H_2S which may impart odor to drinking water. Among some well-known colorless (non-photosynthetic) sulfur bacteria are *Thiobacterium* (short rods which deposit sulfur within or outside the cells), *Macromonas* (large slow-moving organisms which contain $CaCO_J$ as, well as sulfur), and *Thiovulum* (spherical cells up to 20 μ). A well-known color-less sulpur bacterium is *Thiobacillus,* an autotrophic organism, the best known of which is *T. thioxidans.* It oxidizes thiosulphate first to sulfur and then to sulphuric acid. The H_2SO_4 may then corrode pipes and concrete sewers. All the above usually occur singly.

Beggiatoa and *Thiothrix* are usually filamentous. *Beggiatoa* move by gliding and contain sulfur granules. Some species may be up to 50 μ in diameter. *Thiothrix* is non-motile.

5.4.2 Fungi

Fungi are primarily terrestrial, but some are aquatic. Most of the water-dwelling fungi are *Phycomycetes,* although representatives of the other groups (*Ascomycetes, Basidiomycetes*, and especially Fungi imperfecti (*Hyphomycetes*)) contain some aquatic counterparts. Aquatic *Hyphomycetes* have been well described.

It is perhaps not surprising that it is only among the *Phycomycetes* that fungi with motile zoospores are to be found. Among the Phycomycetes, the following orders are aquatic: *Chytriditlles, Blastocladiales, Monoblepharidales, Hyphochytriales, Leptomitales* and *Lagenidiales.* These fungi are saprophytes or parasites on various plants and animals or their parts in water. In other words, they are mostly, "Aufwuchs," that is attached and may be found on any aquatic plant or animal, algae, fish or even other fungi.

Among the more common genera of fungi encountered in water are: *Allomyces*, *Achyla*, *Sapromyces*, and various chytrids. *Fusarium* (not a phycomycete but in the Fungi Imperfecti) is also common in water. Aquatic yeasts have been discovered in large numbers in recent times particularly in waters with high organic matter contents and many *Hyphomycetes* (Fungi Imperfecti) have also been recorded. There are more than 600 species of freshwater fungi with a greater number known from temperate, as compared to tropical, regions.

The Fungi Imperfecti (called mitosporic fungi by some authors) are classified into two main classes, namely hyphomycetes, and coelomycetes. The hyphomycetes produce conidia directly from vegetative structures (hyphae) or on distinct conidiophores (a specialized hypha that bears conidiogenous cells and conidia, for example in *Aspergillus* or *Penicillium*) whereas, the coelomycetes produce conidia within asexual fruit bodies called pycnidia.

The freshwater Fungi Imperfecti (mitosporic fungi) are classified into three groups: The Ingoldian hyphomycetes (also called the aquatic hyphomycetes), the aeroaquatic hyphomycetes, and the dematiaceous (dark colored) and hyaline (light colored) hyphomycetes and coelomycetes (see Fig. 2.2).

(a) *The Ingoldian hyphomycetes*
The aquatic or fresh water hyphomycetes are also known as the Ingoldian hyphomycetes, after Ingold who first described them in 1942. They are Fungi Imperfecti (i.e., Ascomycetes whose perfect or sexual stages have not been described) as well as some Basidiomycetes. Ingoldian hyphomycetes produce conidia that are mostly unpigmented and branched or long and narrow, and are adapted for life in running water. The Ingoldian hyphomycetes most commonly occur on dead leaves, and wood immersed in water.

(b) *Aeroaquatic hyphomycetes*
The aeroaquatic hyphomycetes produce purely vegetative mycelium in substrates under water, but produce conidia with special flotation devices, only when the substrates on which the fungus is growing are exposed to a moist aerial environment. They are found in stagnant ponds, ditches, or slow flowing freshwater. Their vegetative hyphae grow on submerged leaves and woody substrates under semi-anaerobic freshwater conditions and are found around the world.

(c) *Dematiaceous and hyaline fungi imperfecti*
The dematiaceous and hyaline hyphomycetes and coelomycetes are distinct from Ingoldian hyphomycetes, because the conidia is not specifically adapted for aquatic existence. The fungi occur mainly on decaying herbaceous plant material and woody debris in aquatic and semi aquatic habitats worldwide.

They are classified into two main groups: indwellers and immigrants. Indwellers have been reported only from freshwater habitats, whilst immigrants have been reported from terrestrial as well as freshwater habitats.

Freshwater fungi are thought to have evolved from terrestrial ancestors. Many species are clearly adapted to life in freshwater as their propagules have specialized aquatic dispersal abilities. Freshwater fungi are involved in the decay of wood and leafy material and also cause diseases of plants and animals (see Fig. 4.21).

5.4.3 Algae

Algae are primarily aquatic organisms and hence are to be found in large numbers in water, including freshwater. Some algae commonly encountered in drinking water include the blue-green algae: *Microcystis aeroginesa* (which yields a material toxic to man and animals), *Aphanozomenon flos-aquae,* and *Anabaena circinalis,* all of which hamper filteration processes in water purification. The chrysophyte *Sunura uvella* imparts to drinking water a cucumber taste.

The following diatoms also cause filteration problems:

Asterionella formosa, *Nitzschia acicularis*, *Stephanodisus astrea*, *Melosira* spp. as well as the Xanthophyte, *Tribonema bombycinum.*

5.4.4 Protozoa

The Protozoa also show a pattern of succession in their use of bacteria as food. The earliest occurring Protozoa during the eutrophication of a fresh body of water are Sarcodina and Mastigophora which are found in large numbers sonly in freshly contaminated waters.

Like the bacteria which are induced to develop by eutrophication, they also absorb soluble nutrients (Fig. 5.3).

5.5 Succession of Organisms in the Breakdown Materials Added to Aquatic Systems

5.5.1 The Physiological Basis of Aquatic Microbial Ecology

In water, as in most other natural environments, microorganisms do not usually exist as pure cultures, rather they exist as mixed cultures. The abundance of any particular organism or groups of organisms depends on the conditions operating at a given time in the given environment. Some of such conditions are discussed below, although it ought to be noted that the situation is complex in that several of these conditions may operate at the same time.

1. *Nutrient availability*: The quality as well as the quantity of nutrients entering water is important in the microbial ecology of aquatic systems. Any

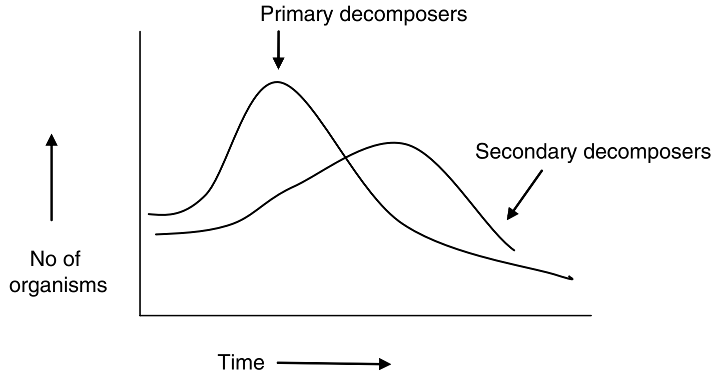

Fig. 5.3 Succession of organisms in the decomposition of materials added to aquatic systems

increase in the concentration of available nutrients in water is known as *eutrophication,* which may be man-made as in sewage discharge into a stream, or natural as with rain water washings.

In the mixed populations of freshwater systems, the addition of the easily digested carbohydrates such as starches and sugars ·stimulate the general development of bacteria and fungi. Among the bacteria, the versatile *Pseudomonas* is usually present and proteins when added to water stimulate the development of bacteria such as *Alcaligines* and *Flavobacterium.*

As a consequence of eutrophication, the organisms which can utilize any particular added materials grow rapidly. On the exhaustion of the substrate, these primary invaders die, releasing cellular materials which are predominantly protein. This release of proteins encourages a secondary development of proteinaceous and other bacteria.

Among the Protozoa, the tectonic cliates soon over run their predecessors, Sarcodina and Mastigophora which then die off. The bacterial population, the source of nourishment for the tectonic ciliates, soon decline in population to the extent that they no longer satisfy the needs for these active organisms. At this stage, the stalked ciliates *(Suctoria)*, needing less nourishment because of lesser activity suck in the remaining bacteria; but sooner or later, these remaining bacteria may not be enough to sustain even these sessile ciliates. It is at this stage that rotifers and crustaceans move in, to scavenge off any living microorganisms available such as the insoluble remains of the dead ones (Hugenholtz et al. 1998). This relationship can be represented as shown in Fig. 5.4.

2. *pH*: At pH values of 4.0–5.0, the fungi predominate and bacteria are completely excluded. At pH values of 7 and above, the predominant organisms are bacteria. Thus *Vibrio* will predominate at pH values of over 8.0.
3. *The oxygen tension*: When the system is aerated, fungi, bacteria, and protozoa grow rapidly, and organic materials are broken down ultimately into CO_2 and water and new cells are produced.
   Under anaerobic conditions, anaerobic bacteria develop and fungi and protozoa are generally absent. If sulfate is present, the sulfate-reducing bacteria e.g., *Desulfovibrio* (small Gram-negative anaerobic curved rods) occur in large numbers, forming sulfide in the process. Some clostridia, e.g., *C nigrificans* also reduce sulfate to H_2S.
   If sulfate is absent methane-producing bacteria occur. These are able to use CO_2 as electron transport acceptor for anaerobic respiration, resulting in its reduction to the gas methane (CH_4). Three genera are involved: *Methanobacterium, Methanococcus,* and *Methanosarcina* (see Chap. 10).
   Anaerobic bacteria are found in muds where their activities lead, as shown above, to the formation of foul gases such as methane and hydrogen sulfide.
4. *Temperature*: High temperatures (about 30–37°C) encourage enteric organisms, while extremely high temperatures (higher than 45°C) select thermophiles. Thus in hot springs, thermophilic bacteria and bluegreen algae are encountered. Low temperatures such as the ones found in oceans (4–10°C) encourage the growth of psychrophyls.
5. *Depth of the water*: More bacteria are generally found in the upper portion of the water than the lower. Furthermore, the type of organisms developing at the various regions of a stream or lake differ one from the other. Cocci, for example, are generally benthic whereas the Gram-negative mobile rods are generally tectonic.

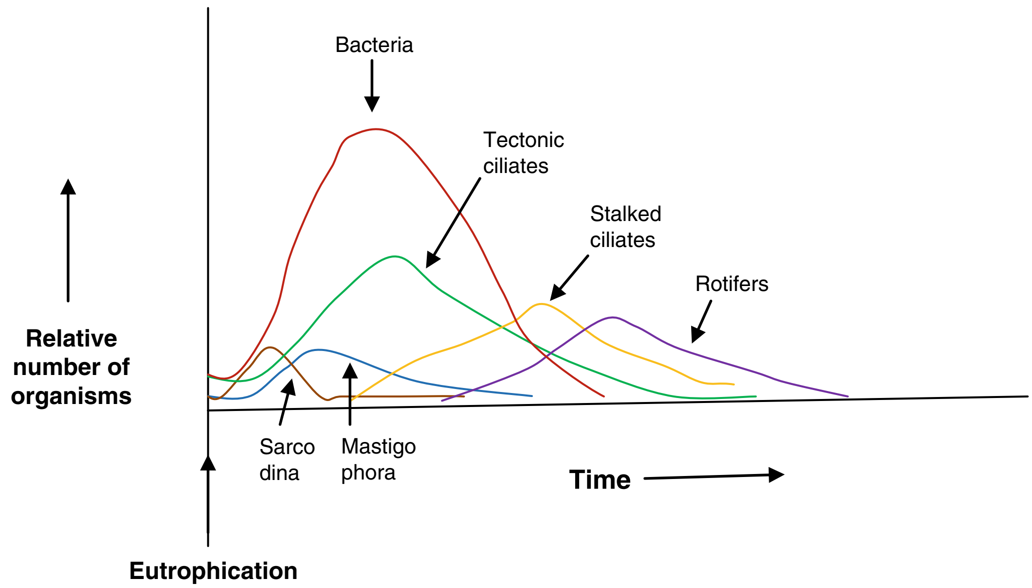

Fig. 5.4 Succession of microorganisms in an aquatic system

6. *Flow velocity in moving waters*: The velocity of the water affects the type of organisms developing in water. It has been established for example that some aquatic algae and bacteria (e.g., *Sphaerotilus natans* and *Leptomitus lacteus)* grow better in flowing waters under natural conditions than in stagnant water. The protozoan *Vorticella* (attached ciliate) and *Nitzschia paleae* (diatoms) also grow better in slow-moving waters.
7. *Light*: Light tends to inhibit the growth of bacteria, but encourages the development of the algae. When nutrients are plentiful under aerobic and light conditions such as in an oxidation pond (see Chap. 10), the bacteria break down the organic matter releasing CO_2 in the process. The CO_2 is then used up, by the algae, for photosynthesis. The latter process releases oxygen which is required by the bacteria.

5.6 Microbial Loop and the Food Web in a Freshwater System

The succession of organisms in an aquatic system is intimately related to the food web (see Fig. 5.5). With better techniques for assessing the load of viruses in aquatic system, we now know that viruses are not only very abundant in aquatic systems, freshwater and saline, but that they also play a major role in the food web of the system. They attack all classes of microorganisms in the system, bacteria, fungi algae, and protozoa, and the carcasses of these organisms contribute to the dissolved organic matter which are absorbed bacteria in the microbial loop shown in Fig. 5.5. The microbial loop is a new concept in the food chain of aquatic systems.

In Fig. 5.5, the regular food chain (shown in unbroken lines) begins with primary produces such as algae which capture the energy of the sun in photosynthesis. These are eaten by the smaller aquatic life such as small fish and krils. The fish are eaten by bigger fish which are eventually eaten by humans. Small fish, big fish, and humans all produce wastes which are broken down by aerobic bacteria in water. When the algae and the aquatic animals die, they are also broken down by aerobic bacteria. The bacteria themselves are eaten by protozoa; the protozoa are eaten by krils and small fish and the cycle goes on. The release of breakdown products from wastes and the carcasses of aquatic organisms contributes to the dissolved organic matter of aquatic systems and provides nourishment for aquatic organisms. The dissolved organic matter of aquatic systems is also increased by eutrophication, i.e., the addition of organic matter.

The new concept is the "microbial loop." The concept has come into being because of the recent recognition of the place of viruses in aquatic microbial ecology. As seen from Fig. 5.5, viruses attack and kill bacteria,

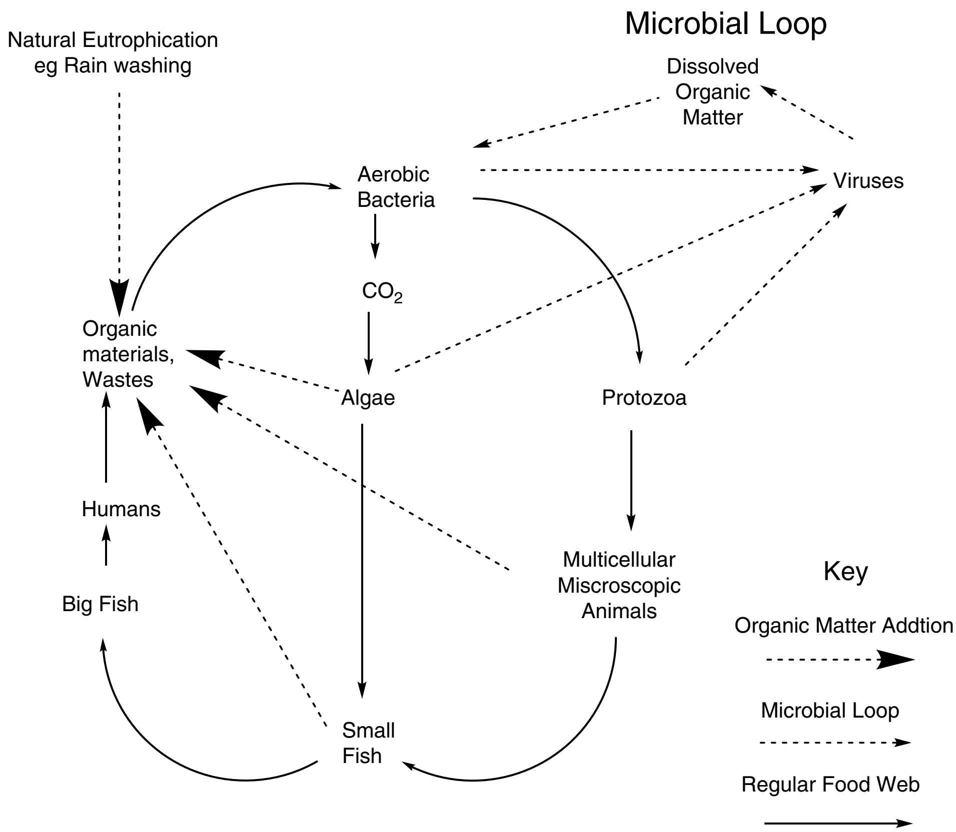

Fig. 5.5 The microbial loop in the food web in a freshwater system

fungi, protozoa, and algae and release materials which contribute to the dissolved organic matter. The microbial loop is that aspect of the food chain which depends on the activities and contribution of microorganisms.

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