Lakes and ponds

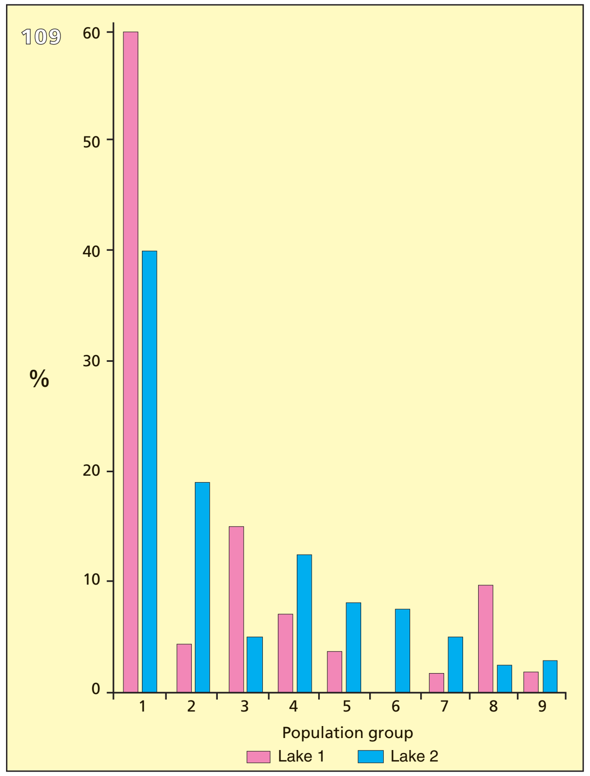
Lakes and ponds are environments that have much in common with running water and, indeed, are often a part of the same water system. A feature of many lakes and ponds is the higher nutrient concentration, stemming from a build-up of deposits and nutrients. This is usually most marked where there is little or no connection with running water systems. Even where strong water flows are present there are likely to be significant areas where water is effectively still and considerable build-up of deposits occurs.

Although the underlying microflora is similar to that of running water, the higher nutrient level of ponds and lakes supports higher numbers of micro-organisms and in some cases a greater variety of types. As the nutrient level increases towards eutrophication the number of types may be reduced by competition. There can be considerable variation between lakes in close geographical proximity (**109**) for which no immediate explanation is available.

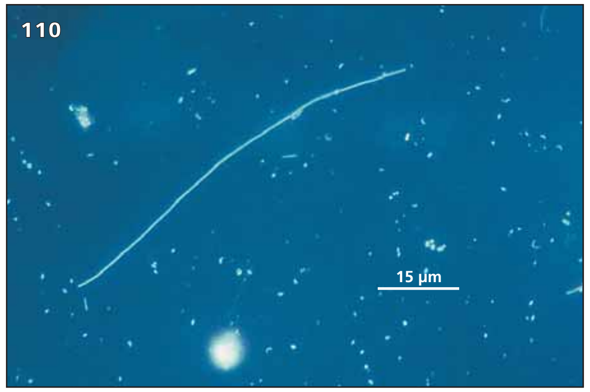
As in many other ecosystems, including both fresh and marine waters, the total bacterial population is limited by predation. Studies of a hypertrophic lake (Sommaruga and Psenner, 1995) revealed the permanent presence of grazing-resistant bacteria. The cells of these comprised filaments, the length of which sometimes exceeded 200 µm (**110**, **111**), the accompanying micro-organisms being small cocci. Such bacteria are larger than nanoflagellates and even metazoans, accounting for 45–81% of bacterial biovolume although only 4–16% of bacterial abundance. The large size appears effective as a defence mechanism against predatory ciliates, such as *Cyclidium* spp., but equally may predispose the cells to attack by larger predators such as *Daphnia* spp. Under these circumstances small cocci persist and it seems likely that the cell size of the dominant bacteria varies in response to the physical size of the major predator. In the case of large bacterial cells, it is likely that persistence results from the selective pressure of a high level of predation by nanoflagellates, but absence of larger predators. It is possible, indeed probable, that large cell size is a defence against predation in other habitats.

As with marine environments, bacteriophages play an important role both in limiting bacterial production and short-circuiting carbon flow. A study of a backwater of the Danube River (Mathias and Kirschner, 1995) showed a difference in susceptibility between rod-shaped and 'vibrioid' bacteria and coccoid bacteria, in that the former two morphological types were much more susceptible to bacteriophage infection. Virus induced mortality was probably at least 15–30% suggesting that up to 33% of carbon cycles within a microbial loop rather than moving up the food chain (see also page 30).

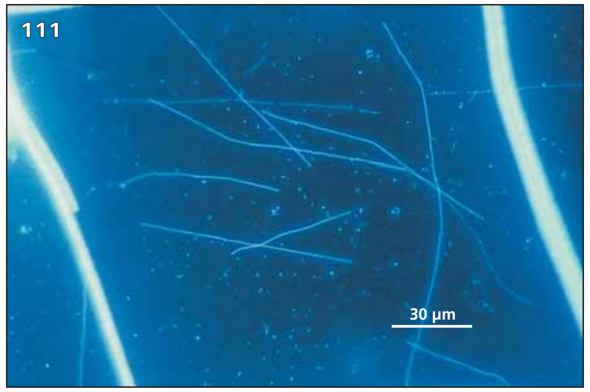
As well as in sea ice, microbial communities can thrive in the ice and snow cover of high mountain lakes as detailed in *Feature 7*, page 70.



109 A series of small, shallow lakes (maximum depth about 2.0 m) were created as part of a riverside nature reserve designed to reduce the environmental impact of nearby commercial development. Some of the lakes are unconnected but share a small stream as water supply and are separated only by narrow (approx. 2.5 m) embankments bearing hoggin paths. Although total microbial numbers are similar in each lake, the distribution of types of bacteria varies widely.

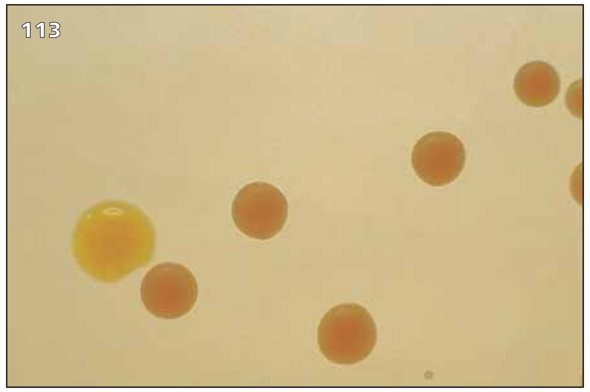


110 A grazing-resistant filamentous bacterium, 75 µm in length, together with a heterotrophic flagellate.



111 An overview of micro-organisms in hypertrophic lake water showing both large filamentous bacteria and small coccal-shaped cells. The filaments in the foreground are of the cyanobacterium *Planktothrix agardhii*. **110** and **111** reproduced with permission from Sommaruga and Psenner, 1995, © 1995 American Society for Microbiology.

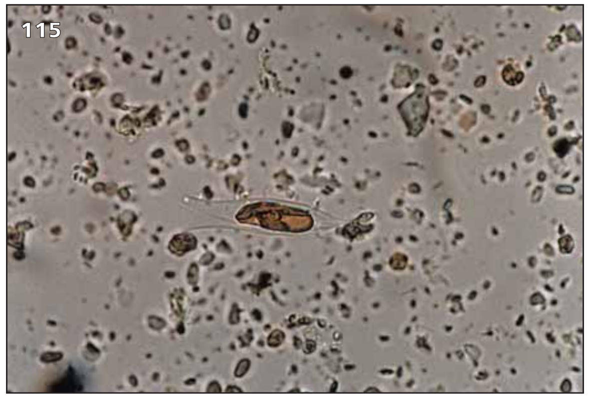
112 A species of *Flectobacillus*. In contrast to *Spirosoma* and *Runella*, a marine species has been isolated having an obligate requirement for the Na⁺ ion. (Source: pond; Medium: MS medium – the addition of 3% NaCl is required for marine species; Incubation: room temperature, 10 days.)



113 *Runella slithiformis*. Distribution was highly variable even over a small sampling area. The yellow colony is of an oxidase-negative, Gram-negative rod. (Source: flooded sand pit – greensand; Medium: MS medium; Incubation: room temperature, 12 days.)

The water column of shallow ponds or shallow areas of larger lakes can contain a particularly wide range of micro-organisms. Such ecosystems appear to be particularly favourable for 'non-motile, Gram-negative, curved bacteria', including *Flectobacillus* (**112**) and *Runella* (**113**). The extent to which Actinomycetes colonize lakes is frequently underestimated, especially in waters which are relatively

114 Snow cover at Estany Rado during winter.



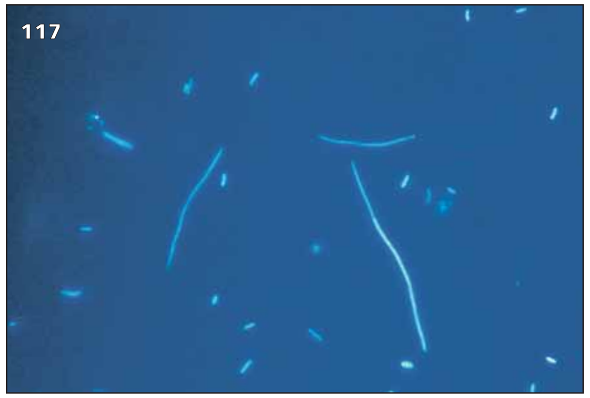
115 The flagellate *Dinobryon cylindricum*, a photosynthetic eukaryote isolated from the Estany Rado snow cover.

116 The ciliate *Perigostrombidium fallax*, a predator on bacteria in the deeper layers of snow cover during the spring melt.

Feature 7. Rocky mountain high

Colonization of sea ice is a well-known phenomenon (see page 55) but it is often not appreciated that active microbial communities can develop in the ice and snow cover of high mountain lakes. Studies in Estany Rado, Spain (**114**), showed that the winter cover provides a highly dynamic environment, the physical structure and chemical characteristics of which change dramatically as a result of snowfall, periodic melting, freezing and flooding. The main groups of micro-organisms, autotrophic and heterotrophic flagellates (**115**), ciliates (**116**) and bacteria (**117**), respond directly to these physicochemical changes with resulting effects on biomass and species composition. There appear to be two distinct phases which have been attributed to different mechanisms of colonization and growth. From January to mid-April (the phase of growth of the cover), microbial assemblages are related to their planktonic counterparts. During this period, colonization is primarily due to plankton derived from the lake water which floods the cover. As light fails to reach the slush layers, the plankton is primarily dependent on bacterivory for growth.

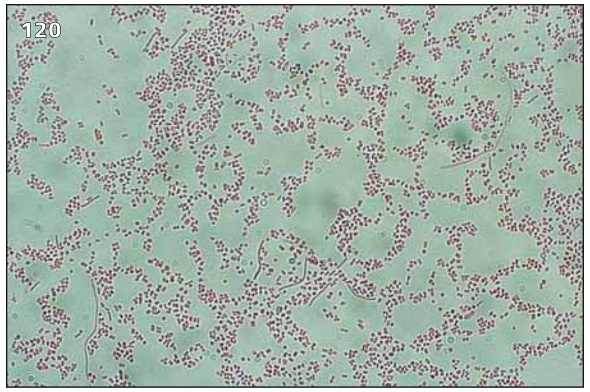
From mid-April to the spring melt in June (ablation), the physicochemical nature of the environment is strongly influenced by the large amount of water (**118**) from the melting of the snowpack. The water is a source of both nutrients and micro-organisms, while the light availability permits the growth of algae and an associated food web. In deeper layers the food web is based on bacteria, providing prey for large ciliates, which are probably derived from littoral or watershed soils. **114–116** and **118** reproduced by courtesy of Dr M. Felip, University of Barcelona, Spain; **117** reproduced with permission from Felip *et al*., 1995, ©1995 American Society for Microbiology.



117 Bacteria, viewed by epifluorescent microscopy after staining with DAPI, from the deeper layers of snow cover during the spring melt. Note the presence of elongated cells; cf. **110**, **111**.

118 Water from melting snow cover at Estany Rado during spring (May). The scale of the melt is apparent from the quantity of water in the foreground.

119 *Legionella pneumophila*. Methods for isolation from natural environments are often time consuming, the most suitable method varying according to the source of the sample (Kusnetsov *et al*., 1994). Colonies on the non-selective medium illustrated may also have a ground glass appearance. (Source: thermally polluted, disused canal; Medium: buffered charcoal–yeast extract agar supplemented with ∝-ketoglutarate – Edelstein, 1981; Incubation: 35°C (95°F), 7 days.)



120 *Legionella pneumophila*. Cells are highly pleomorphic, varying from short cocco-bacilli to long filaments, though filaments are not present during growth in animal tissue. Staining is often weak and may be irregular. (Source: as for **119**; Microscopy: Gram stain, × 1,200.)

nutrient rich. Actinomycetes are important in the decomposition of organic compounds such as chitin and cellulose. The dominant genus is often *Micromonospora* with *Streptomyces* second in importance (Jiang and Xu, 1996).

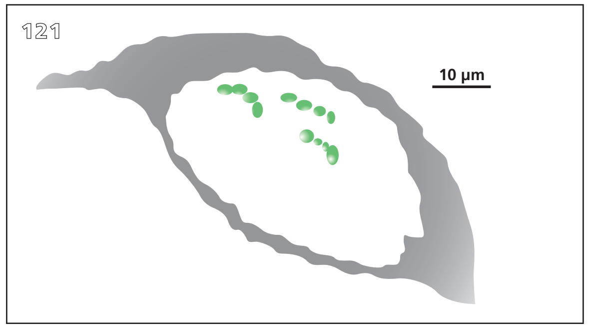
Species of the human pathogen *Legionella* (119, 120) are common in natural waters, especially those of high nutrient content and thermally polluted. Waters of this type appear to serve as a reservoir of *Legionella* from which the bacterium enters and colonizes water cooling systems, humidifiers, and the like. Some of these provide a very favourable environment for *Legionella* in terms of temperature, level of aeration, organic nutrients and iron concentration. In either natural or artificial systems the organism may develop as a member of a biofilm but there is also an association between *L. pneumophila* and the free-living amoeba

Acanthamoeba polyphaga (**121**) (Kilvington and Price, 1990). This involves the intracellular growth of *L. pneumophila* inside amoebic trophozoites. From a public health viewpoint this association is of concern because it enables *L. pneumophila* to survive chlorination of cooling water and similar systems. Aerial distribution inside amoebae, along with fragments of biofilm and large droplets, may also represent a means by which the bacterium bypasses defence mechanisms in the lung. From a broader environmental viewpoint, intracellular growth is probably beneficial to *L. pneumophila* in preventing predation by other protozoa, ensuring a supply of nutrients, including iron, and protecting against UV irradiation to which *Legionella* is highly sensitive.

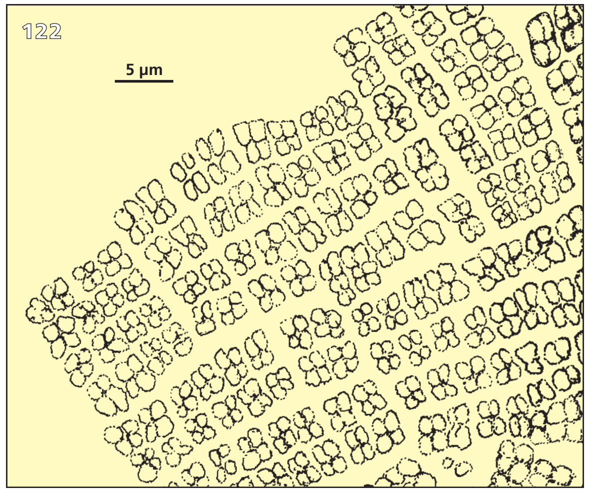
The reduced O_2 concentration in the upper layer of sediments is favourable for growth of *Serpens* and also for true micro-aerophiles such as *Spirillum*. *Spirillum* and similar bacteria may also be found below the surface scum of stagnant water where favourable micro-aerophilic conditions are created by the surface growth of aerobic bacteria. In heavily polluted waters, rich in organic matter, the surface film may include the enigmatic *Lampropedia hyalina* (**122**) although the real habitat of this bacterium is not known.

A number of the micro-aerophilic and anaerobic bacteria of the sediment of lakes and ponds have been shown to exhibit magnetotactic behaviour. These bacteria, which are motile and morphologically diverse, contain high quantities of iron (*c.*0.4% of the dry weight of the cell) as magnetite (Fe_3O_4), the iron sulphide greisite (Fe_3S_4) or a combination of greisite with iron pyrites (FeS_2) which is incorporated as specialist sensing organelles, the magnetosomes (**123**). These function by orientating cells in the unfavourable O_2-containing water column so that their motility results in a return to the favourable conditions of the sediment. There has been a recent description of an organism which contains both magnetite and greisite magnetosomes (**124–126**).

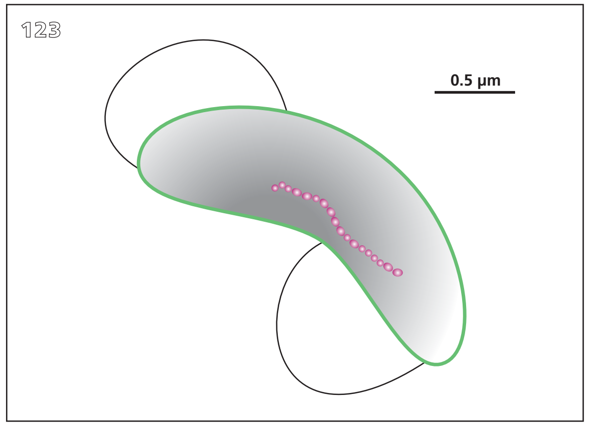
Considerable anaerobic activity occurs in the sediment and can involve both sulphate-reduction and methanogenesis (see page 75). In shallow lakes of high organic matter content, anaerobic conditions can extend throughout the water column with the exception of the water just below the air interface which is occupied by cyanobacteria and eukaryotic algae. A neuston may be formed by *Euglena sanguinea* which contains photoprotective pigments. These migrate within the cell according to light intensity and the water colour may also change. Where significant quantities of H_2S are produced in the underlying sediment (cf. marine sediments), purple and green sulphur bacteria (**127**)



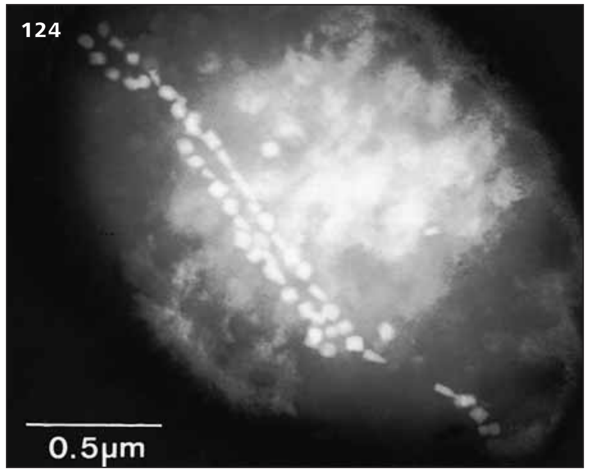
121 Diagrammatic representation of *L. pneumophila* inside an *Acanthamoeba* cell.



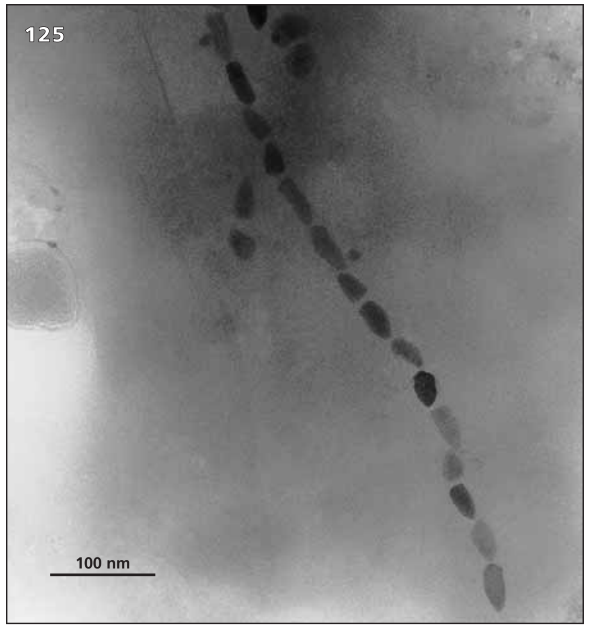
122 *Lampropedia hyalina* showing the characteristic 'window pane' morphology.



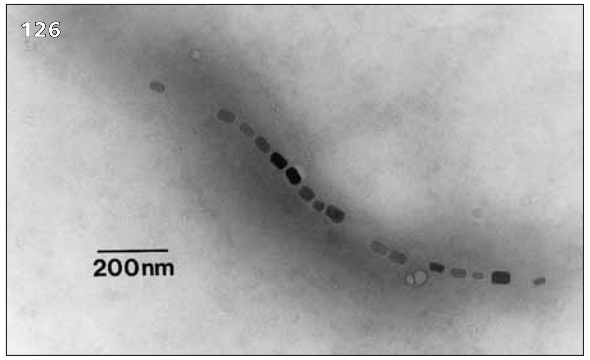
123 A general view of a cell of a magnetotactic bacterium showing the position of magnetosomes. Magnetotactic bacteria in the northern hemisphere are almost entirely north seeking while those in the southern hemisphere are almost entirely south-seeking. Populations collected at the geomagnetic equator, where the magnetic field has no vertical component, are an equal mixture of north- and south-seeking cells.



124 Dark field STEM of rod-shaped, magnetic bacteria showing two chains of magnetosomes. Magnetite magnetosomes are arrow shaped while greisite magnetosomes are rectangular. Reproduced by courtesy of Dr D. Bazylinski, Iowa State University, Ames.



125 TEM showing arrangement of magnetosomes Reproduced by courtesy of Drs D. Bazylinski and B.R. Hayward, Iowa State University, Ames.



126 A magnetotactic micro-organism. Despite the large gap between some magnetosomes, the consistent orientation suggests co-organization by a chain assembly process. Reproduced by courtesy of Drs D. Bazylinski and F.C. Meldrum, from Bazylinski *et al.*, 1995, © 1995 American Society for Microbiology.

127 Green sulphur bacteria. Growth below the oxic surface layer of a predominantly anoxic lake consisted of flakes containing green sulphur bacteria and an unknown non-photosynthetic, Gram-negative bacterium. The bacteria were embedded in a waxy matrix.

develop in a layer directly below the oxic upper zone. Development of anaerobic conditions in shallow lakes can be a particular problem in hot weather when oxygen is less soluble in water and microbial activity high. Aeration of water is often required to prevent the death of fish and recreational lakes, marinas, and the like, can easily lose all amenity value. Anaerobic activity in the sediment extends into the water column and includes outgrowth of clostridial spores (**128**). Under these conditions toxin production by *Cl. botulinum* has caused the death of aquatic birds, including swans and, allegedly, bathing dogs (**129**, **130**).

Deep lakes can exhibit stratification of the water column into aerobic and anaerobic zones (**131**). This results from the differing density of water at different temperatures, density being a maximum at 4°C (39.2°F). Solar heating produces a layer of warm water of low density, the epilimnion, which floats on the colder, denser layer, the hypolimnion. The two layers are separated by the thermocline (chemocline) at a depth of 20–30 m. The upper layer of water is aerobic although where geographical conditions limit mixing an oxygen gradient may be created extending to the thermocline. Conditions are anaerobic below the thermocline with considerable anaerobic activity occurring in the sediment.

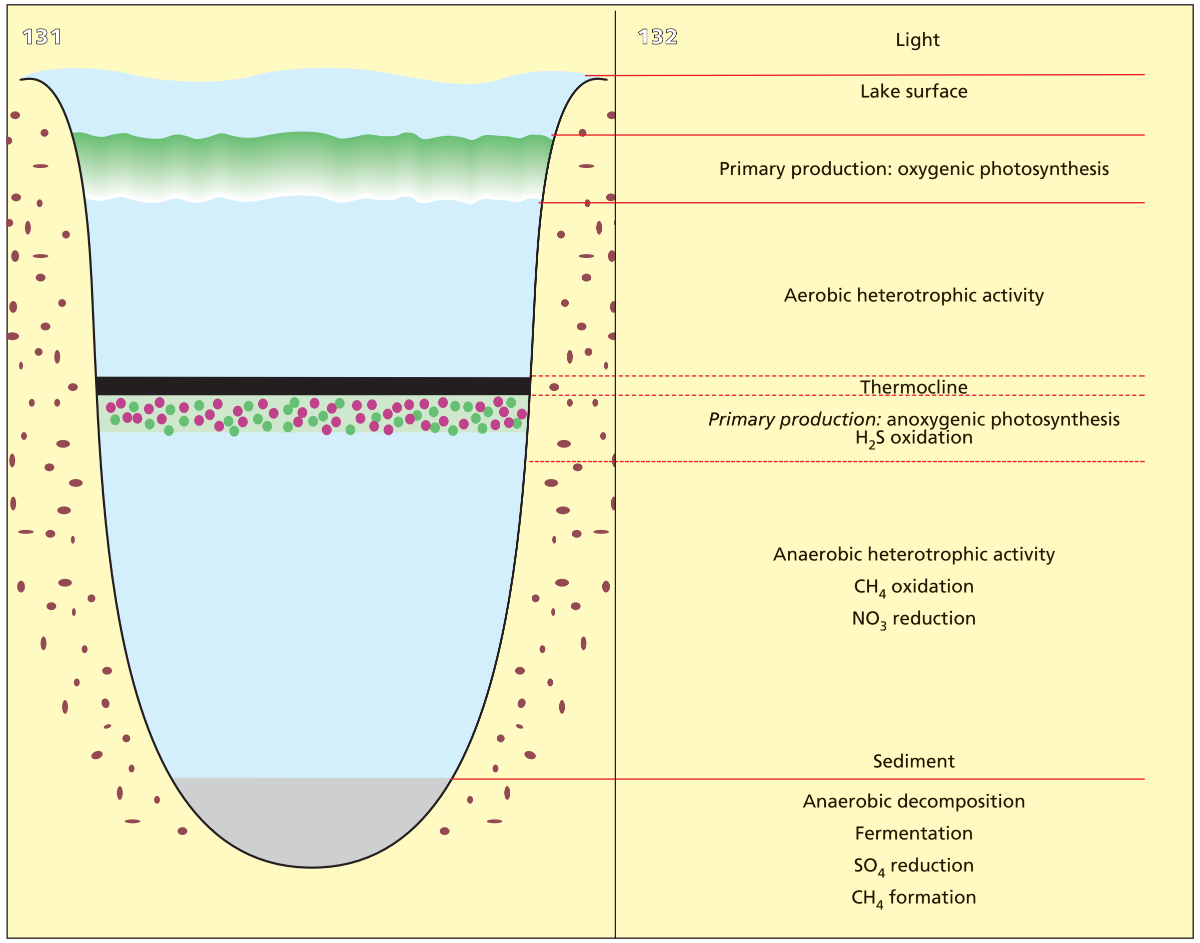
Stratified lakes are of two types. In meromictic lakes, stratification is permanent and persists independent of seasonal changes. Meromictic lakes occur most commonly in tropical and subtropical zones but can also occur in temperate climates. Stratified marine basins (see page 34) are of this type. In holomictic lakes, stratification is seasonal. The epilimnion is established when ambient temperatures rise in the spring and early summer. An algal and cyanobacterial bloom may occur at this stage due to a combination of higher temperature and an elevated nutrient concentration following mixing of the water layers the previous winter. Stratification usually remains stable over the summer, until the temperature of the epilimnion falls during the autumn. At some stage, usually during early winter, the temperature of the epilimnion falls below that of the hypolimnion and stratification becomes unstable. Mixing of the layers occurs, often initially promoted by strong winds. This results in some oxygenation of lower layers and distribution of nutrients from the lower layers throughout the water column. Inverse stratification can occur in the winter when deep, high density water at 4°C (39.2°F) is overlaid by colder, less dense water and ice. Mixing of the layers when surface water temperatures rise in late winter leads to nutrient distribution and contributes to the spring bloom.

Stratification and resulting chemical gradients leads to intense microbiological activity at different depths in the lake (**132**). Organic matter tends to accumulate in the sediment, where anaerobic degradation occurs. The ultimate fate of organic compounds largely depends on the relative importance of sulphate-reducing and methanogenic bacteria. Sulphate-reducing bacteria normally dominate, even where the quantity of organic deposition is such that adequate H_2 is available, permitting a degree of coexistence between the two groups. Sulphate levels in freshwater tend to be much lower than in seawater and can become limiting. Under these conditions production of both H_2S and CH_4 occurs in the sediment, CH_4 being the major end-product. These gases, together with CO_2, enter the water column where oxidation of CH_4 by methophils leads to anaerobic conditions. Methane is only sparingly soluble in water and significant quantities escape as gas bubbles.

128 A putrefactive anaerobe (*Clostridium* sp.). The isolate was phenotypically similar to proteolytic strains of *Cl. botulinum*. (Source: anoxic lake sediment; Medium: blood agar; Incubation: 37°C (98.6°F), 24 hours, 95% H_2:5% CO_2.)

129 *Clostridium botulinum* grows rapidly under suitable conditions. In lakes, corpses of dead birds and animals may be a focal point for growth and toxigenesis.

130 The environment may become sufficiently contaminated by *Clostridium botulinum* to present a severe risk to aquatic birds, including swans.



131, 132 Stratification of, and activity of micro-organisms in, a deep lake. Primary production involves both oxygenic and anoxygenic photosynthesis in distinct zones. This situation occurs only in lakes of high nutrient content.