The Red Queen said: It takes all the running
you can do to keep in the same place

Lewis Carroll, Through the Looking Glass

Chapter 27
From life as a nomad to life as an endosymbiont

In the previous chapter we saw that bacteria can live as single organisms in various habitats but may also tend to settle together on surfaces, creating biofilms. In addition, bacteria are even able to coexist with higher organisms by forming symbiotic relationships.

Symbiosis in a strict sense is an association between two different organisms for the benefit of both partners. Symbiotic nitrogen fixation is a somewhat borderline case because the rhizobial bacteria entering the roots of a plant are subsequently held captive, irreversibly transformed into nitrogen-fixing bacteroids. As individuals they don't gain a thing from this joint venture, but in the end they contribute bound nitrogen to the soil when the plant decays. This in turn benefits their free-living rhizobial colleagues and other microorganisms in the soil. Associations of the bacterial species *Frankia*, *Azospirillum*, and *Azoarcus* with plants, mentioned in Chapter 11, much better fit our definition of symbiosis.

At this point, we will take a short detour to the corals because of their beauty and importance and the numerous dangers they face. Corals live in clear, shallow waters that are low in nutrients, and they require photosynthesis for growth and reproduction. Corals are only able to perform photosynthesis with the help of other organisms, single-cell algae called *Zooxanthellae* that live inside the coral cells. This symbiosis is one of the most delicate relationships found in nature. Slight changes such as a rise in temperature may lead to coral bleaching and even death. For this reason it is feared that global warming, in addition to other severe environmental changes, will cause coral extermination. Bleaching of coral signals the loss of the symbiotic partner, and bacteria could even be the cause. Eugene Rosenberg (Tel Aviv, Israel) and his research team found that the *Zooxanthellae* in Mediterranean corals are attacked by a bacterium called *Vibrio shiloi* whenever the water temperature rises by a few degrees. We asked him to tell us about it:

> "For the last several decades coral reefs have been in a decline, largely due to emerging and re-emerging diseases. The largest environmental factors contributing to these diseases are pollution, over-fishing and rising seawater temperatures. On the global scale, coral bleaching is the most severe disease. Coral bleaching is the disruption of the symbiosis between the coral animal and its endosymbiotic algae, commonly referred to as

Discover the World of Microbes: Bacteria, Archaea, and Viruses, First Edition. Gerhard Gottschalk.

Zooxanthellae. As a result of the loss of the algae, the tissue becomes transparent and appears white because of the calcium carbonate backbone.

In two cases it has been demonstrated by applying Koch's postulates that coral bleaching is a result of bacterial infection: the coral *Oculina patagonica* in the Mediterranean Sea by *Vibrio shiloi* and *Pocillopora damicornis* in the Indian Ocean and Red Sea by *Vibrio corallilyticus*.

In both cases, increased temperature caused the pathogen to express virulence genes. The *V. shiloi* infection cycle has been studied extensively and it has been shown that a peptide toxin is produced which blocks photosynthesis of the algae. Recently, David Bourne and colleagues in Australia have reported that *Vibrio* appeared in large numbers in coral tissue just prior to a mass bleaching event on the Great Barrier Reef. The recovery of the corals when the temperature decreased correlated with the loss of the *Vibrio*.

At present there is a debate on whether mass coral bleaching is the result of bacterial activity or photoinhibition of the algae at high temperature and light intensity."

These findings, shocking enough in themselves, are documented in a photograph (Figure 56) that requires no further comment.

So bacteria were first nomads, then symbionts, and now endosymbionts?

On our way to the endosymbionts, let's first take a look at a few more host–microbe interactions. Pathogenic bacteria such as *Bacillus anthracis* or *Mycobacterium tuberculosis* are parasites (see Chapter 29). A number of bacteria have given up growing and proliferating as free-living organisms. They have become obligate parasites; they are able to divert preformed building blocks, amino acids, coenzymes, and nucleotides from their host cells – in some cases, even ATP. *Rickettsia prowazekii*

Figure 56 Partially bleached coral *Oculina patagonica*. (Photograph: Fine and Loya, 1994, provided by Eugene Rosenberg, Tel Aviv, Israel.)

is such a bacterium: It lives intracellularly and is the cause of typhus, also called typhus fever (not to be confused with typhoid fever). The genus Rickettsia is named after Howard Taylor Rickett (1871–1910), who discovered that Rocky Mountain spotted fever is caused by a species of this genus. During evolution, there has been a transition from certain obligate parasites to endosymbionts. A good example is *Buchnera aphidicola*, which lives in plant aphids and provides its host with certain aromatic amino acids (e.g., tryptophan). Bacterial endosymbionts of the *Buchnera* or *Carsonella* species contain very small genomes because they have given up many of the functions that normally make up a bacterial cell.

Could mitochondria and chloroplasts be the remnants of endosymbionts?

Yes. These ideas were first put forward by the German botanist Andreas F. W. Schimper (1856–1901) in the 1880s, then elaborated upon by the Russian biologist Constantin Mereschkowsky (1855–1921) and later taken up and further developed by Lynn Margulis (Amhurst, Massachusetts, USA).

Mitochondria (let's call them mitos for short) are, as mentioned earlier, the power plants of the cells. In their interior, reducing equivalents are generated that subsequently react with oxygen in the membrane. A proton motive force is generated, and the F_1F_O-ATPase as discussed in Chapter 8 takes advantage of this force to synthesize ATP.

But this surely is not enough to support the theory that mitos originate from endosymbionts.

When we look at other components of the mitos, two features are quite striking. They contain circular DNA, which can be thought of as a minichromosome, and their ribosomes are similar in size (70S) and structure to those from bacteria. In contrast, eukaryotic ribosomes are larger (80S) in general. Human mitochondrial DNA (mtDNA is the short form) contains only 17 000 base pairs and codes for 35 genes, including genes for ribosomal RNAs, some transfer RNAs, and components of the ATPase and the respiratory chain (Figure 57). By all indications, a lot of information has either been lost or else transferred to the nucleus. Despite this, the mitos have retained a certain degree of autonomy. Mitos are special: they carry genetic information that is not present in the DNA of human cells and they cannot be synthesized, so to speak, from scratch. Instead, they reproduce themselves by dividing, just like the bacteria do. Mitos are also in the egg cells of mothers, and that's how they are passed on to the next generation. It's funny that men, whose cells also contain mitos, don't pass them on to the next generation because sperm heads contain no mitos. Worldwide comparisons of mtDNA sequences in humans have made it possible to develop a migrational tree of mankind (or better, womankind). Researchers compared the base sequences of a hypervariable region of the mtDNA that lies between the base pairs 16 001 and 16 559 (Figure 57). An analysis of these sequences in humans from Australia, Europe, America, and other continents revealed not only some differences but also

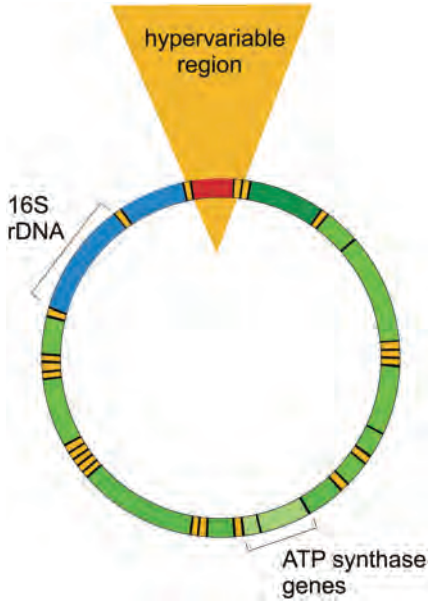

Figure 57 Human mitochondrial DNA. Indicated are: gene encoding 16S-rRNA, the hypervariable region, the transfer-RNA genes (yellow) and the genes for ATP synthase. (Diagram: Anne Kemmling, Goettingen, Germany.)

a basic pattern that is apparently the basis for all the others. This basic pattern is only found in the mitos of African women. That's why we speak of the "mitochondrial Eve," the common mother of all mankind who must have lived in Africa. Sequence analyses indicate that the migration of humans out of Africa and into other continents began less than 100 000 years ago.

As all cells are subject to aging, the mitos also grow "old" and become less efficient. After all, they are constantly exposed to radicals, the toxic reduction products of oxygen (see Chapter 5) generated in their close proximity. Since mitos are both the source and the target of such toxins, the mtDNA obviously is more liable to oxidative damage than the DNA enclosed and protected by the cell nucleus. As the mitos grow older, cumulative DNA damage leads to formation of nonfunctional proteins and to disturbances in the coupling of respiration and ATP synthesis. The resulting loss of efficiency and decline in cellular energy supply accelerates the aging processes even more.

Does anyone have an idea which bacteria could have migrated into the eukaryotic cells?

In 1998, evolutionary biologist Siv Andersson (Uppsala, Sweden) and her colleagues published a paper entitled "The genome sequence of *Rickettsia prowazekii* and the origin of mitochondria". *R. prowazekii* has already been mentioned as the microorganism causing typhus, which has been responsible for devastating epidemics in the past. We may recall that the decimation of Napoleon's army in Russia in 1912 was primarily due to typhus. Siv Andersson's team sequenced the

genome of *R. prowazekii*, which consists of around 1.1 million base pairs. A comparison with known sequences revealed that the closest relatives of *R. prowazekii* are plant mitos as well as those of a protozoan called *Reclinomonas americana*. The DNA of human mitos is not suitable for such comparisons because it is much smaller and only contains a few genes, as we have seen. To a greater extent than the protozoans, human mitos have adapted to the task of energy conservation by reducing and simplifying their genetic information. The DNA not needed was simply lost, or it migrated into the nucleus. Especially the mt genes of said protozoans and those of *R. prowazekii* correspond to a high degree, leading to the conclusion that the precursor of *Rickettsia* once migrated into cells that were larger than bacteria. Such cells had a nucleus but still lived, more or less, like bacteria. Once inside the cell, the immigrants lived on waste materials in their hosts and respired with the oxygen present. They became more and more specialized in energy conservation and eventually evolved into mitos. In plants, the immigrants obviously could not have been *Rickettsia*, which lack a photosynthetic apparatus. Therefore, cyanobacteria must have been the ones to be engulfed by eukaryotic cells.

Again, bacteria have given us quite a lot. All higher organisms owe them their organelles for energy production: the mitochondria for respiration, the chloroplasts for photosynthesis, and the enzyme systems to combat ROS (reactive oxygen species).