

Lynn Margulis & the Question of How Cells Evolved

JOEL B. HAGEN

□ INTRODUCTION

Modern biology inherited two great theories from the nineteenth century: evolutionary theory and cell theory. Surprisingly, these theories, so central to our understanding of the living world, have had a rather uneasy relationship. Until quite recently, most cell biologists ignored evolution, and most evolutionary biologists ignored cells. The exception to this historical generalization was the chromosomes, which both evolutionary biologists and cell biologists studied. But what about the cytoplasm, the contents of the cell outside the nucleus? Could knowing about other cellular structures (organelles) add anything to evolutionary theory? Could evolutionary theory suggest interesting questions about the structure or function of organelles? For most biologists, the answer to these questions was no. The cytoplasm added little to understanding evolutionary theory, and vice versa.

Occasionally, some biologists tried to bridge the theoretical gap, but they usually met with derision. For example, during the 1920s the microbiologist Ivan Wallin made the remarkable claim that mitochondria had originated as free-living bacteria. According to Wallin, the former bacteria and their host cells evolved together to establish an inseparable symbiotic partnership. He even claimed to have removed mitochondria from cells and grown them in isolation.

Wallin's idea was almost universally rejected, and he was often ridiculed for his wild speculations. According to his critics, evolution by symbiosis was as improbable as that other great pseudoscientific idea of the 1920s: continental drift (see Chapter 16). Although intrigued by the possibility that mitochondria evolved from bacteria, America's leading cell biologist, E. B. Wilson, remarked that Wallin's ideas were "too fantastic for present mention in polite biological society."

With the benefit of hindsight it is easy to smile at the comparison between continental drift and endosymbiosis, two great scientific heresies that later revolutionized the way we look at the natural world. The criticisms were, however, justified. Wallin's theory was quite speculative. No one, then or now, has verified his claim that mitochondria can be grown outside of cells.

PROBLEM

Assuming that mitochondria really did evolve from free-living bacteria, why might it be difficult or impossible to experimentally grow them outside of the host cell? How can you explain Wallin's unverified claim that he had isolated and grown mitochondria outside of cells?

Both the structure and the function of mitochondria were mysteries in 1920. The internal anatomy of bacteria was also almost totally unknown. The evidence Wallin needed to support his theory required the electron microscope and other sophisticated laboratory techniques developed only after World War II. As in the case of continental drift, the theory of symbiosis in cellular evolution that was finally accepted during the 1970s was very different from the one suggested by Wallin in the 1920s.

LYNN MARGULIS: A REVOLUTIONARY SCIENTIST

Like the eventual acceptance of continental drift, acceptance of a symbiotic theory of cell evolution has often been hailed as a scientific revolution. The woman most responsible for bringing the idea to scientific respectability is Lynn Margulis (Figure 3.1). A prolific writer and dynamic speaker, Margulis captivates audiences and often irritates more traditional biologists with her unorthodox ideas. A profile in *Science* described her as an unruly provocateur, but as one of the world's leading authorities on cellular evolution, she supports her claims with abundant evidence. Although many biologists continue to disagree with some of her ideas, everyone takes endosymbiosis seriously.



FIGURE 3.1 Lynn Margulis. *Source:* Courtesy of Lynn Margulis and the University of Massachusetts Photo Service.

Margulis entered biology during a particularly exciting period. James Watson and Francis Crick were just discovering the structure of DNA when Margulis was in college. A few years later, when she was a graduate student, two of her professors discovered DNA in chloroplasts. Other scientists reported finding DNA in mitochondria (Figure 3.2). Because these early reports were hotly disputed, searching for DNA outside the nucleus was not the sort of research project that most graduate students would have chosen. Despite warnings, Margulis plunged into the controversial problem for her Ph.D. dissertation. Using radioactively labeled nucleotides, she convincingly demonstrated the presence of DNA in the chloroplasts of *Euglena gracilis*, one of the curious unicellular organisms that shares both plant and animal characteristics.

Margulis wrote her first article on the endosymbiotic theory in 1967, two years after she completed her Ph.D. At the time, she was a single mother without a permanent teaching position. She was also writing her first book on endosymbiosis, which sparked a lively controversy when it was published in 1970. Although it initially brought Margulis notoriety, the controversy over cellular evolution was rather short lived. By the time she published a second book on endosymbiosis in 1981, most biologists accepted important parts of her theory. As a result, Margulis became a scientific celebrity whose success was publicized in both popular and professional magazines.

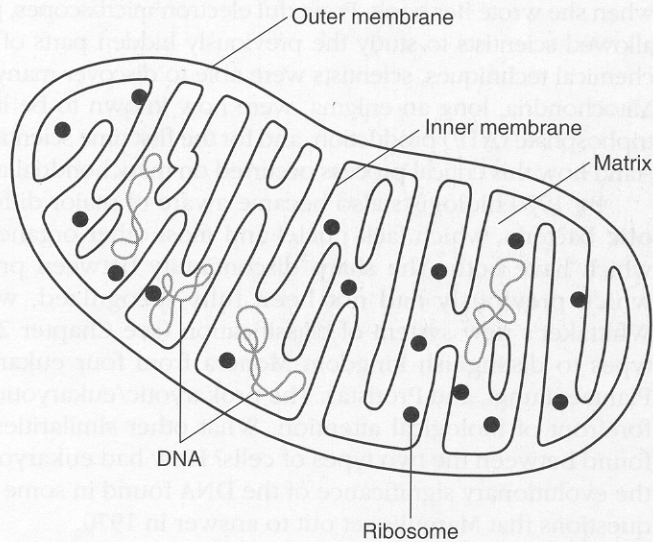


FIGURE 3.2 Cross section of a mitochondrion containing DNA molecules and protein-building ribosomes similar to those found in bacteria. The liquid matrix contains the enzymes responsible for the early steps of respiration (Krebs cycle). Most of the ATP produced during respiration is generated by the oxygen-consuming reactions (electron transport system) that occur on the highly folded inner membrane.

BACKGROUND TO A CONTROVERSY

In 1970, when Margulis's first book was published, most biologists had never heard of endosymbiosis. Those who knew about it usually dismissed it. In order to succeed, Margulis had to carefully distinguish her ideas from the discredited theory proposed by Ivan Wallin half a century earlier. She also had to overcome a basic assumption about evolution held by nearly all biologists at the time. According to the traditional view, evolution usually occurs gradually; endosymbiosis, however, is based on the idea of rather sudden evolutionary changes. Finally, Margulis had to convince biologists to take DNA in the cytoplasm seriously. Although evidence for DNA in chloroplasts and mitochondria was growing stronger, the idea that some genes reside outside the nucleus remained unorthodox.

Despite these biases against endosymbiosis, Margulis's book was widely read. Even those who strongly disagreed with her did not ridicule her theory the way biologists had belittled Ivan Wallin's theory about the evolution of mitochondria. Indeed, the book convinced many biologists that cellular evolution was an exciting, if controversial, field. How had cell biology changed during the 50 years after Wallin proposed his unsuccessful theory?

Much more was known about the internal structure of cells in 1970 than in 1920. Unlike Wallin, who knew little about the internal structure or function of mitochondria, Margulis had access to a great deal of information about the internal structure of cells when she wrote her book. Powerful electron microscopes, perfected after World War II, allowed scientists to study the previously hidden parts of organelles. Using new biochemical techniques, scientists were able to discover many details of cellular activities. Mitochondria, long an enigma, were now known to be important sites of adenosine triphosphate (ATP) production, and for the first time scientists were beginning to understand how this critical process occurred on mitochondrial membranes (see Chapter 8).

By 1970 biologists also became aware of major differences between **prokaryotic** bacteria, which lack nuclei and most other organelles, and **eukaryotic** cells, which have both. The sharp discontinuity between prokaryotes and eukaryotes, which previously had not been fully recognized, was highlighted by Robert Whittaker's new system of classification (see Chapter 2), which used the two cell types to distinguish kingdom Monera from four eukaryotic kingdoms (Animalia, Plantae, Fungi, and Protista). The prokaryotic/eukaryotic distinction was now at the forefront of biological attention. What other similarities and differences might be found between the two types of cells? How had eukaryotic cells evolved? What was the evolutionary significance of the DNA found in some organelles? These were the questions that Margulis set out to answer in 1970.

THE SERIAL ENDOSYMBIOTIC THEORY (SET)

According to Margulis, eukaryotic cells evolved through a series of symbiotic partnerships involving several different kinds of prokaryotic cells. The smaller partners invaded larger host cells and eventually evolved into three different kinds of

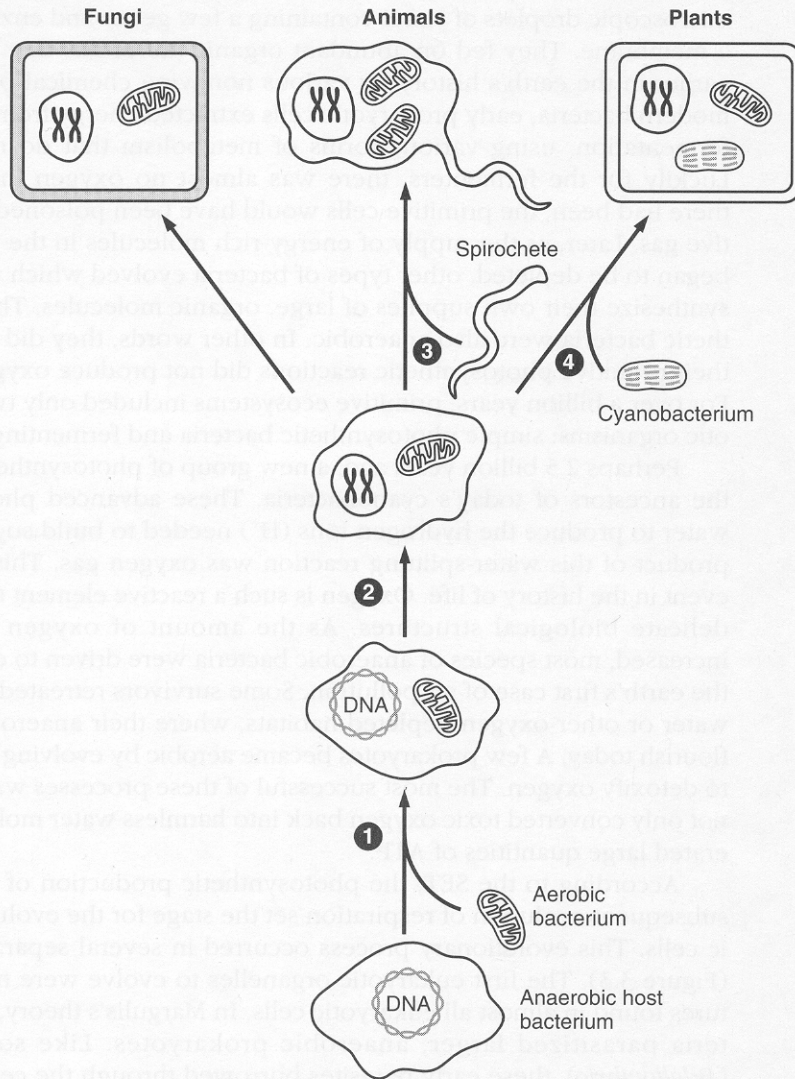


FIGURE 3.3 Evolution of eukaryotic cells by a series of endosymbiotic events: (1) mitochondria evolve from small, free-living, respiring bacteria; (2) the nucleus evolves from the simpler prokaryotic DNA molecule; (3) flagella (undulipodia) evolve from symbiotic spirochetes; (4) chloroplasts arise from free-living cyanobacteria. Cell walls in plants and fungi, which are structurally quite different, evolve independently.

organelles: mitochondria, chloroplasts, and flagella (Figure 3.3). Because these evolutionary steps supposedly occurred as a series of discrete events, Margulis's theory is often referred to as the **SET: serial endosymbiotic theory**.

Like other evolutionary biologists, Margulis believes that life first appeared on the earth about four billion years ago. The first organisms were extremely simple—

microscopic droplets of water containing a few genes and enzymes surrounded by a membrane. They fed on abundant organic molecules that had been produced earlier in the earth's history by various nonliving chemical processes. Like some modern bacteria, early prokaryotic cells extracted energy from these molecules by fermentation, using various forms of metabolism that do not require oxygen. Luckily for the fermenters, there was almost no oxygen in the atmosphere. If there had been, the primitive cells would have been poisoned by this highly reactive gas. Later, as the supply of energy-rich molecules in the watery environment began to be depleted, other types of bacteria evolved which used solar energy to synthesize their own supplies of large, organic molecules. These early photosynthetic bacteria were also anaerobic. In other words, they did not use oxygen and their primitive photosynthetic reactions did not produce oxygen as a by-product. For over a billion years, primitive ecosystems included only two types of prokaryotic organisms: simple photosynthetic bacteria and fermenting bacteria.

Perhaps 2.5 billion years ago, a new group of photosynthetic bacteria evolved, the ancestors of today's cyanobacteria. These advanced photosynthesizers split water to produce the hydrogen ions (H^+) needed to build sugar molecules. A by-product of this water-splitting reaction was oxygen gas. This was a catastrophic event in the history of life. Oxygen is such a reactive element that it easily destroys delicate biological structures. As the amount of oxygen in the atmosphere increased, most species of anaerobic bacteria were driven to extinction, victims of the earth's first case of air pollution. Some survivors retreated to areas of brackish water or other oxygen-depleted habitats, where their anaerobic descendants still flourish today. A few prokaryotes became aerobic by evolving various mechanisms to detoxify oxygen. The most successful of these processes was respiration, which not only converted toxic oxygen back into harmless water molecules, but also generated large quantities of ATP.

According to the SET, the photosynthetic production of oxygen gas and the subsequent evolution of respiration set the stage for the evolution of all eukaryotic cells. This evolutionary process occurred in several separate symbiotic events (Figure 3.3). The first eukaryotic organelles to evolve were mitochondria—structures found in almost all eukaryotic cells. In Margulis's theory, small respiring bacteria parasitized larger, anaerobic prokaryotes. Like some bacteria today (*Bdellovibrio*), these early parasites burrowed through the cell walls of their prey and invaded their cytoplasm. Either the host or the parasite was often killed in the process, but in a few cases the two cells established an uneasy coexistence.

The mutual benefits to the partners are obvious. The respiring parasite, which actually required oxygen, would allow its host to survive in previously uninhabitable, oxygen-rich environments. Perhaps the parasite also shared with its host some of the ATP that it produced using oxygen. In exchange, the host provided sugar or other organic molecules to serve as fuel for aerobic respiration. Eventually, as often occurs with parasites, the protomitochondria lost many metabolic functions provided by the host cell. Similarly, as oxygen in the atmosphere continued to increase, the host became more and more dependent upon its pro-

tomitochondria to detoxify the gas. What began as a case of opportunistic parasitism evolved into an obligatory partnership. The small respiratory bacteria eventually evolved into the mitochondria of eukaryotic cells.

Although virtually all eukaryotic cells contain mitochondria, only those of plants and certain protists contain chloroplasts. Therefore, it seems likely that chloroplasts evolved in only a few lines of eukaryotic cells, and this event occurred after mitochondria were already well established. How did this new evolutionary partnership evolve? With higher metabolic rates, cells containing mitochondria were more efficient than anaerobic cells. Some of these newer, unicellular organisms grew larger and evolved into predators capable of eating smaller cells. Their prey undoubtedly included cyanobacteria. In rare cases, these small photosynthetic cells may have resisted digestion after being engulfed. Inside the predator, they set up a semi-independent existence and eventually evolved into chloroplasts.

Although such a scenario may seem far-fetched, we know that similar partnerships exist today. For example, the unusual ciliate *Paramecium bursaria* is host to many unicellular green algae in the genus *Chlorella*. These "pseudochloroplasts" produce sugar molecules that are shared with the host. If the *Chlorella* are experimentally removed, both partners continue to exist independently. Without its photosynthetic partners, however, the *Paramecium* becomes totally dependent upon external sources of food. Provided the opportunity, the *Paramecium* will eat *Chlorella* but will not digest them, thus reestablishing the symbiotic partnership. *Paramecium bursaria* is not a unique case of modern endosymbiosis. Many other organisms, including several multicellular animals, also play host to photosynthetic algae or cyanobacteria.

The most controversial claim made by Margulis is that eukaryotic flagella evolved from small, corkscrew-shaped bacteria called **spirochetes**. Many spirochetes are parasites (the best known, *Treponema pallidum*, causes syphilis). Others are free-living, found in such exotic environments as the intestines of termites. Regardless of how they live, these unusual bacteria swim with an undulating motion reminiscent of the whiplike movement of eukaryotic flagella. Is this similarity evidence for Margulis's evolutionary claim, or is it simply a coincidence? Why not accept the more orthodox explanation that eukaryotic flagella gradually evolved from the simpler flagella found on many bacteria?

Margulis points out that although both types of flagella are used for locomotion, prokaryotic and eukaryotic structures are very different (Figures 3.4(A) and 3.4(B)). Prokaryotic flagella consist of a single, hollow filament of protein that spins on its axis like a tiny propeller. Eukaryotic flagella are much larger; they contain a complex arrangement of 11 microtubules, and the entire structure is surrounded by an extension of the cell membrane. In contrast to the spinning prokaryotic flagellum, the eukaryotic structure propels the cell by lashing back and forth in a whiplike fashion. Because they are so different in structure, function, and perhaps evolutionary origin, Margulis proposes that the eukaryotic flagellum should be referred to by a different term: **undulipodium**.

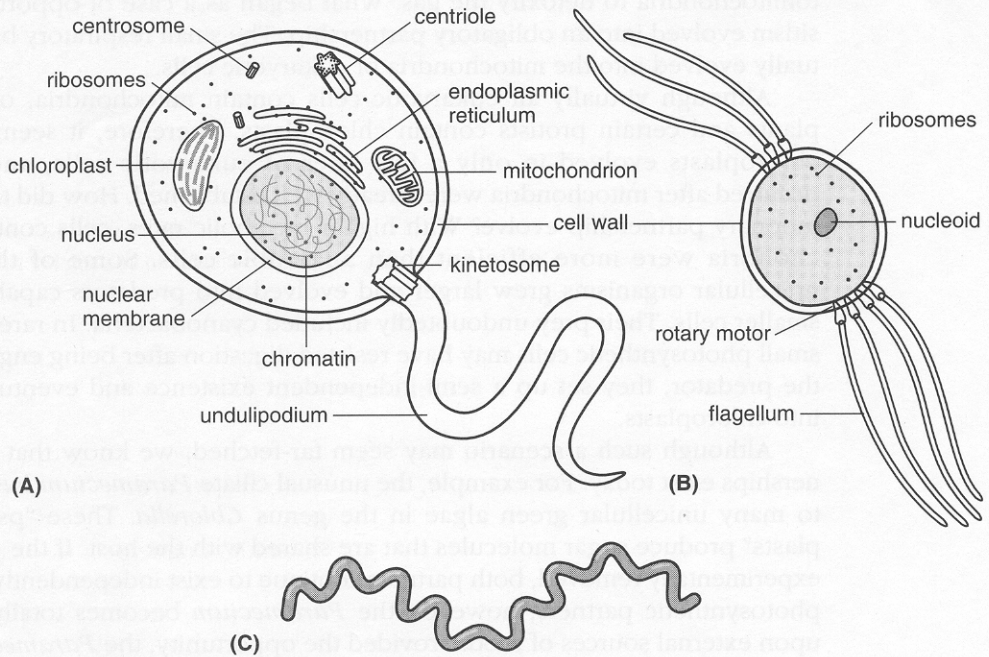


FIGURE 3.4 (A) Eukaryotic cell with undulipodium that moves in a whiplike manner. (B) Nonnucleated, prokaryotic cell with several simple flagella that move by rotation. (C) Spirochete that is propelled in an undulating motion by axial filaments and flagella inside the bacterial cell.

Could undulipodia really have evolved from spirochetes? Margulis claims that the complex arrangement of microtubules in the undulipodium is somewhat similar to long protein filaments (axial filaments) located between the cell wall and an outer sheath membrane in spirochetes (Figure 3.4(C)). Intriguing evidence further supporting her hypothesis comes from cases of “motility symbiosis” described by Margulis. In the hindguts of many termites live a wide variety of protozoans and bacteria, including several types of spirochetes. Biologists had long believed that one of the protozoans (*Mixotricha paradoxa*) was covered with thousands of synchronously beating flagella. Closer examination, however, showed that most of these “flagella” were actually spirochetes regularly arranged in rows on the surface of the protozoan. The rhythmic beating of the symbiotic spirochetes propels the host protozoan through the intestines of the termite.

Mixotricha and other cases of motility symbiosis are intriguing, but the spirochete-undulipodium hypothesis remains far more controversial than the other parts of the SET. Part of the problem is lack of strong supporting evidence. Undulipodia do not contain DNA, RNA, or ribosomes—the remnants of an independent protein-building machinery, which are always found in chloroplasts and mitochondria. If they evolved from free-living bacteria, shouldn’t undulipodia also show this evidence of their ancestry? Pointing out this missing evidence, many critics dismiss the spiro-

chete-undulipodium hypothesis as speculation. Margulis claims that critics are too conservative and further research will ultimately confirm her hypothesis. Most biologists remain skeptical, although many admit that some of Margulis's previous "far-fetched ideas" later turned out to be correct.

PROBLEM

Nobel laureate Joshua Lederberg claims that it is impossible to demonstrate convincingly that eukaryotic flagella evolved from spirochetes. Nonetheless, he believes that Margulis's hypothesis is very important. How might an unprovable speculation be useful to scientists?

AUTOGENY: AN ALTERNATIVE THEORY OF CELLULAR EVOLUTION

When Margulis's book appeared in 1970, most biologists accepted the general belief that eukaryotic cells gradually evolved from prokaryotic ancestors. Because cellular evolution was not a major concern of most evolutionary biologists, however, there were few detailed theories to explain how this might have occurred. This situation quickly changed during the 1970s, when Margulis's critics proposed alternative theories of gradual cellular evolution (**autogeny**). Perhaps the best-known of these competing theories was one presented by F. J. R. Taylor, a Canadian botanist.

According to Taylor, all eukaryotic cells evolved through a process of slow, branching evolution. He believed that the original ancestor must have been a photosynthetic bacterium, somewhat similar to the cyanobacteria of today. This must have been so, Taylor reasoned, because cyanobacteria, algae, and plants all use the same form of photosynthesis. It seemed unlikely that this complex process could have independently evolved in each different line. Of course, this means that animals and fungi must have lost the ability to photosynthesize at some later point in evolutionary history (Figure 3.5).

Similar to some cyanobacteria of today, the ancestral cell had a complex system of internal membranes, embedded with the enzymes and pigments used in photosynthesis and respiration, Taylor claimed. Because increasing the surface area of membranes makes a cell more efficient, natural selection favored the evolution of an increasingly elaborate membrane system. Taylor also assumed that DNA was found in several places in the primitive cell. There was a central **nucleoid** where most of the genetic material was located, but several smaller loops of DNA also were scattered throughout the cell. This was a reasonable assumption, because small accessory molecules of DNA are common among bacteria today.

Chloroplasts and mitochondria were formed by two simultaneous evolutionary processes: compartmentalization and specialization. Sections of the elaborately folded membrane system sometimes broke away to form separate, enclosed compartments. Small pieces of DNA and some ribosomes were often trapped inside these bodies. This explains why organelles today contain some protein-making machinery. At the same time, different membrane-bound compartments became specialized to accomplish specific metabolic tasks. Some became centers of photo-

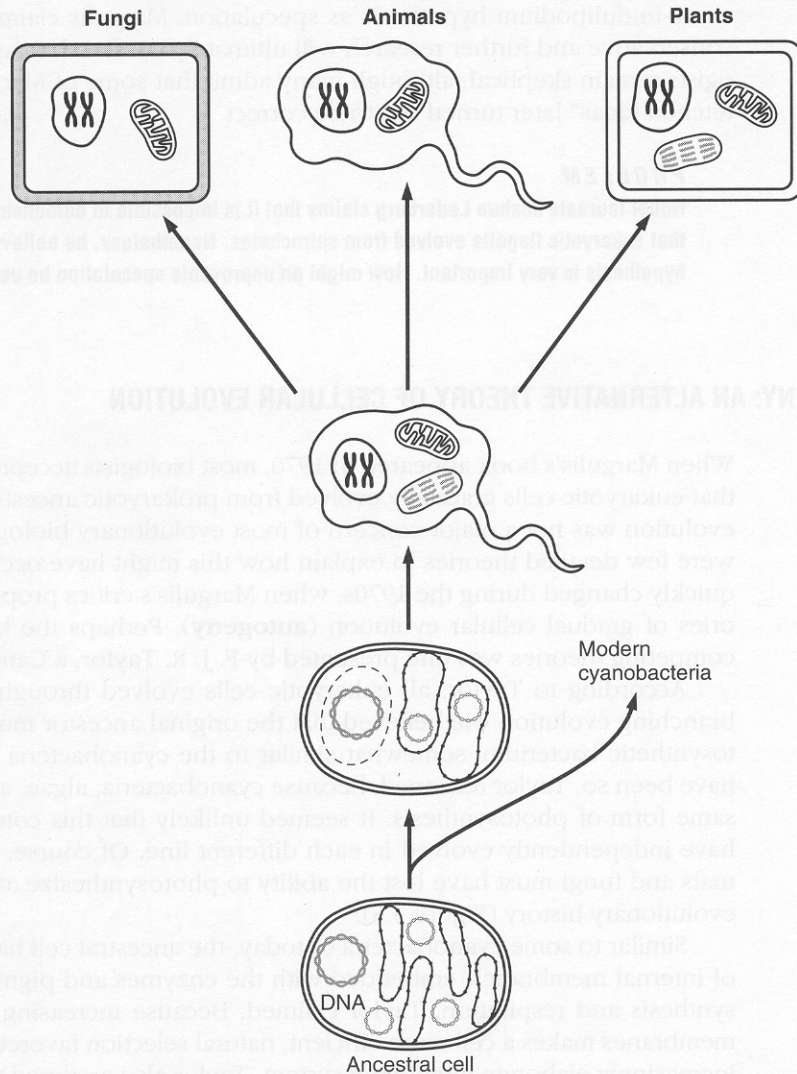


FIGURE 3.5 Autogenous evolution of eukaryotic cells. According to this theory, all cells gradually evolved from a single, primitive photosynthetic cell similar to the cyanobacteria of today. Both mitochondria and chloroplasts evolved from internal membrane systems found in the ancestral cell. Photosynthesis was later lost in the evolutionary lines leading to animals and fungi.

synthesis and eventually evolved into chloroplasts. Others became specialized for respiration and eventually evolved into mitochondria.

During the course of evolution, eukaryotic cells became larger and lost their prokaryotic cell walls. Complex, eukaryotic flagella may also have gradually evolved from simpler bacterial flagella. Presumably, the intermediate forms, which were less efficient for locomotion, disappeared. At least some of the new eukaryotic cells evolved the ability to engulf food particles. These cells became predators, feeding on

bacteria and other tiny organisms. The loss of chloroplasts, which occasionally happened, would not necessarily have been a problem for these evolving predators. Other cells evolved new protective outer walls, quite different from prokaryotic cell walls. This adaptation must have evolved quite late and in two separate branches of the evolutionary tree, because the cell walls of plants and fungi are structurally distinct.

RESOLVING THE CONTROVERSY

During the 1970s, scientists were presented with two broad, competing theories of cellular evolution: the SET and autogeny. At the end of this decade, when Margulis published a second book on endosymbiosis, much of her theory was widely accepted. What had been an unpopular theory a decade before was now part of mainstream biology. Autogeny, although not completely dismissed, seemed less plausible. Why was Margulis so successful at persuading her colleagues to radically change their thinking about the evolution of cells? What problems prevented the widespread acceptance of autogeny?

Margulis claims that, unlike Wallin's unsuccessful theory, both the SET and autogeny make predictions that can be tested. According to her, the SET survived these tests while autogeny did not. This is a plausible explanation, but choosing between two general theories involves more than a single experiment or set of observations. Many of the contrasting predictions made by the SET and autogeny are extremely difficult to test. For example, according to the SET, few intermediates between prokaryotic and eukaryotic cells should be found. If, on the other hand, eukaryotic cells evolved gradually, then many "missing links" should have existed. As it turns out, the dividing line between prokaryotes and eukaryotes is quite sharp, which seems to support the SET. But this is not conclusive evidence against autogeny. Although many fossils of unicellular organisms have been found, including those of some of the earliest prokaryotes, there is general agreement that fossilization is a relatively rare event. Therefore, intermediates between prokaryotes and eukaryotes may yet be discovered.

If simple tests of the SET and autogeny provide ambiguous results, how can scientists choose between the two alternatives? Like most complex theories, the SET and autogeny were evaluated not on the basis of a single test but rather on the basis of multiple lines of evidence. If many seemingly unrelated pieces of evidence can be easily explained by one theory but not by the other, scientists will usually choose the stronger theory—even when it cannot be conclusively proven. For example, if the SET is correct, then numerous similarities should exist between free-living bacteria and eukaryotic organelles. Indeed, in some important ways, organelles should be more similar to free-living bacteria than to the rest of the eukaryotic cell. Few, if any, such similarities should be found if eukaryotic organelles evolved autogenously. The success of the SET was largely due to Margulis's ability to demonstrate so many of the expected similarities. Supporters of autogeny had difficulty explaining why these similarities should be found.

As Wallin had pointed out half a century earlier, mitochondria and chloroplasts reproduce by dividing, much the way bacteria do. What he could not know in the

1920s was that the DNA in mitochondria and chloroplasts is structurally similar to the single circular molecule found in bacteria. Unlike DNA in the nucleus, neither bacterial DNA nor DNA in eukaryotic organelles forms chromosomes. The protein-synthesizing ribosomes found in mitochondria and chloroplasts are smaller than those found elsewhere in eukaryotic cells, but they are about the same size as ribosomes found in free-living bacteria. The nucleotide sequences of RNA molecules in the ribosomes of chloroplasts are also more similar to bacterial sequences than to the sequences of other eukaryotic ribosomes. Some antibiotics that interfere with protein synthesis in bacteria have the same effects on mitochondria and chloroplasts but do not inhibit protein synthesis in the rest of the eukaryotic cell. Finally, as would be expected if these organelles evolved symbiotically, cells experimentally deprived of their mitochondria and chloroplasts are unable to reproduce them. Although most of the genetic instructions for making organelles resides in the nucleus, some necessary genes remain in the mitochondria and chloroplasts. None of these individual bits of evidence is convincing. Taken together, however, they make a strong case for some important parts of the SET.

General theories are usually not completely accepted, at least initially. Margulis's SET is an excellent example of this type of piecemeal acceptance. Nearly all biologists, including many of Margulis's original critics, now believe that mitochondria and chloroplasts evolved from bacteria. But most biologists still reject the claim that eukaryotic flagella (undulipodia) evolved from symbiotic spirochetes. It is generally believed that this hypothesis is too speculative and that there is insufficient evidence to support it. Margulis herself admits that this is the most difficult part of the theory to accept, although she believes that eventually enough evidence will accumulate to convince even skeptical biologists.

Whatever the eventual fate of the spirochete-undulipodium hypothesis, the SET has generated much research on cellular structure and function. Through the process of testing and evaluating competing theories, biologists have learned a great deal about cells and how they evolved. Cell biology and evolutionary biology, two fields that previously seemed to have little in common, now share an important, unifying theory.

□ EPILOGUE

A cornerstone of the SET has been Margulis's belief that cellular evolution was primarily the result of adaptations to an aerobic environment. This claim has recently been challenged by some biologists who study rare eukaryotic cells that do not have mitochondria. More than 1,000 species of protozoans and a few species of fungi lack these respiratory organelles.

Some of the protozoans may be "living fossils" similar to the earliest eukaryotic cells. These simple protists also lack such characteristic eukaryotic organelles as Golgi bodies and the endoplasmic reticulum. According to some biologists, similar protozoans may have existed for millions of years before the evolution of mitochondria. Because these unicellular organisms would have been predators, this explains how the bacterial precursors of mitochondria entered their hosts—they were eaten.

But it undermines Margulis's claim that atmospheric oxygen was the driving force behind the evolution of eukaryotic cells. Perhaps mitochondria evolved later, after other important eukaryotic characteristics were already established. If substantiated, this new claim would be an important modification of Margulis's original theory.

Another interesting group of anaerobic protozoans includes those that have reinvaded such anaerobic environments as the sediments at the bottoms of ponds, lakes, and oceans. These protozoans have lost true mitochondria, but they contain organelles that are structurally similar to these respiratory organelles. These mitochondrionlike structures now function in anaerobic metabolism. Because of their structural similarities, it seems likely that they evolved from mitochondria.

Still another anaerobic protozoan, the ciliate *Strombidium purpureum*, was discovered harboring bacteria capable of both photosynthesis and respiration. In the light, the ciliate avoids even trace amounts of oxygen, but it moves to areas where the light is optimal for bacterial photosynthesis. In the dark, the ciliate migrates to environments where small amounts of oxygen exist. The symbiotic bacteria use the oxygen to produce ATP by respiration. Here is a case of a single symbiotic bacterium playing the roles of both chloroplast and mitochondrion. This may be a biological oddity that does not closely resemble any of the early stages of cellular evolution. On the other hand, studying these present-day examples of endosymbiosis may provide useful models for understanding how eukaryotic organelles evolved over two billion years ago.

QUESTIONS AND ACTIVITIES

1. What does this case show about the following aspects of doing biology?
 - criticism and the revision of theories
 - piecemeal acceptance of new theories
 - resolution of scientific controversies
 - interrelationships of different scientific disciplines
2. Analogies are frequently used by scientists to justify new theories. For example, Margulis claims that hypothetical organisms from the past are somewhat similar to present-day cases of endosymbiosis (e.g., *Paramecium bursaria* and *Myxotricha paradoxa*). How convincing is each of Margulis's analogies? What other forms of evidence might be used to strengthen each analogy?
3. Why does Margulis believe that endosymbiosis was the result of adaptation to an aerobic environment? If this assumption turns out to be incorrect, how might it affect the scientific acceptance of the SET?
4. How might supporters of the SET and autogeny explain each of the following observations? Decide how strongly each observation supports the SET, autogeny, or both.
 - a. Mitochondria and chloroplasts contain DNA molecules similar to those found in bacteria.
 - b. The earliest prokaryotic fossils date from about 3.5 billion years ago, and the earliest eukaryotic fossils date from about 2 billion years ago.

- c. Both chloroplasts and mitochondria contain two sets of membranes, an outer membrane and a greatly folded inner membrane.
- d. Oxygen was not found in the primitive atmosphere, but was formed as a by-product of photosynthesis beginning about 2.5 billion years ago.
- e. Both chloroplasts and mitochondria contain ribosomes smaller than typical eukaryotic ribosomes but about the same size as bacterial ribosomes.
- f. Chloroplasts and mitochondria are about the same size as some bacteria.
- g. Some cyanobacteria are found living inside eukaryotic organisms.
- h. Mitochondria and chloroplasts are incapable of independent existence.
- i. DNA and ribosomes have not been found in eukaryotic flagella.
- j. Some cyanobacteria contain complex systems of internal photosynthetic membranes somewhat similar to those of chloroplasts.
- k. Prior to cell division, chloroplasts and mitochondria reproduce by binary fission, just as bacteria do.
- l. If mitochondria or chloroplasts are removed, the cell cannot regenerate them.
- m. In some cases, spirochetes are attached to the plasma membranes of eukaryotic host cells. These spirochetes look and sometimes act like flagella.
- n. Antibiotics that inhibit protein synthesis on ribosomes in bacteria also have this effect on mitochondrial ribosomes in eukaryotic cells. But the cytoplasmic ribosomes of eukaryotic cells are not inhibited by the antibiotics.

SUGGESTED READING

- Darden, L., and N. Maull. 1977. "Interfield Theories." *Philosophy of Science* 44: 43–64.
- Douglas, A. E. 1994. *Symbiotic Interactions*. Oxford, England: Oxford University Press.
- Fenchel, T., and B. J. Finley. 1994. "The Evolution of Life without Oxygen." *American Scientist* 82: 22–29.
- Maienschein, J. 1991. "Cytology in 1924: Expansion and Collaboration." In K. R. Benson, J. Maienschein, and R. Rainger, eds., *The Expansion of American Biology*. New Brunswick, NJ: Rutgers University Press.
- Mann, C. 1991. "Lynn Margulis: Science's Unruly Earth Mother." *Science* 252: 378–381.
- Margulis, L. 1993. *Symbiosis in Cell Evolution: Microbial Communities in the Archean and Proterozoic Eons*. 2nd ed. San Francisco: Freeman.
- McDermott, J. 1989. "A Biologist Whose Heresy Redraws Earth's Tree of Life." *Smithsonian* 20(5): 72–81.
- Sapp, J. 1994. *Evolution by Association: A History of Symbiosis*. New York: Oxford University Press.
- Taylor, F. J. R. 1979. "Symbioticism Revisited: A Discussion of the Evolutionary Impact of Intracellular Symbioses." *Proceedings of the Royal Society of London Series B* 204: 267–286.