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DOES IT PAY TO COOPERATE?

The origin of eukaryotic cells is a key milestone in life's history on Earth.

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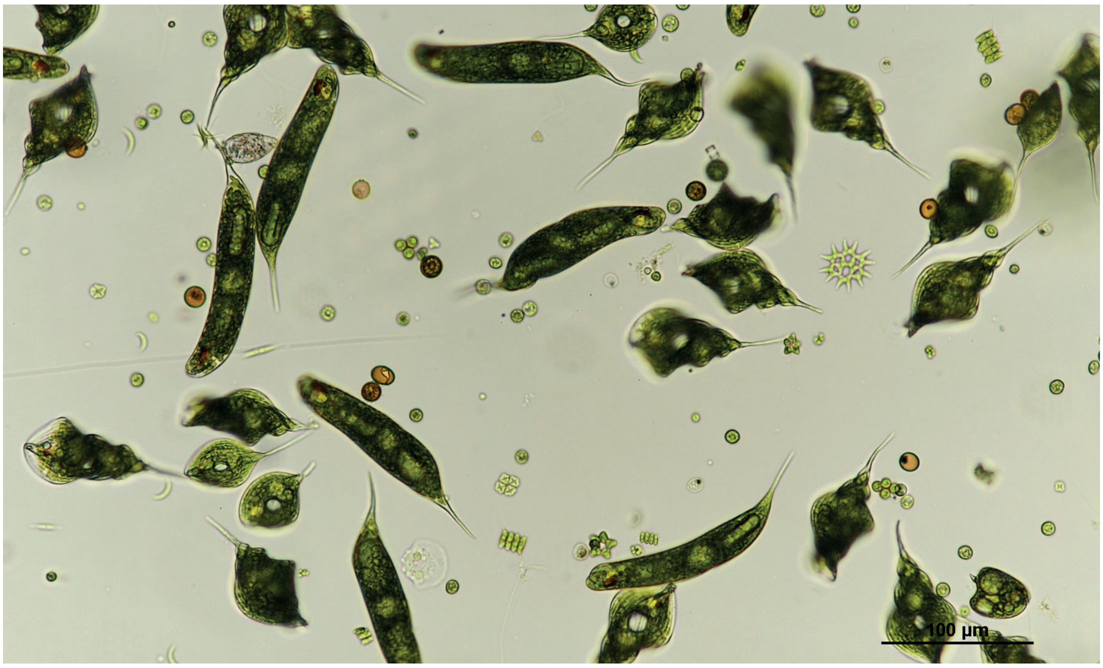
All living organisms are made up of cells. Some, like humans, animals, and plants, are made up of many cells, but the majority of life forms are single-celled organisms. Despite their small size, the cells of both single- and multi-celled eukaryotic organisms – including our own – are remarkably complex. Inside, we find specialized structures known as organelles, each with a distinct role. One of the most important is the nucleus, which houses our DNA: the blueprint of life (the term *eukaryote* comes from the Greek *karyon*, meaning "nucleus"). In addition to the nucleus, eukaryotic cells also contain other vital organelles. Mitochondria, often called the cell's powerhouses, generate energy, while plant and algae cells have chloroplasts, which capture sunlight and convert it into energy. Interestingly, both mitochondria and chloroplasts contain their own distinct DNA, separate from the cell's nuclear genome, a remnant of their ancient endosymbiotic origins. In contrast, most single-celled organisms, such as bacteria and archaea, have much simpler cells. As they lack a nucleus and organelles, we call these organisms *prokaryotes*.

Endosymbiosis

The question of how eukaryotic cells (also known as *Eukaryota*) came to be is a complex one. These events took place so long ago that direct evidence has not sur-



Algae from a freshwater reservoir (including euglenids and green algae) with chloroplasts formed through primary and secondary endosymbiosis

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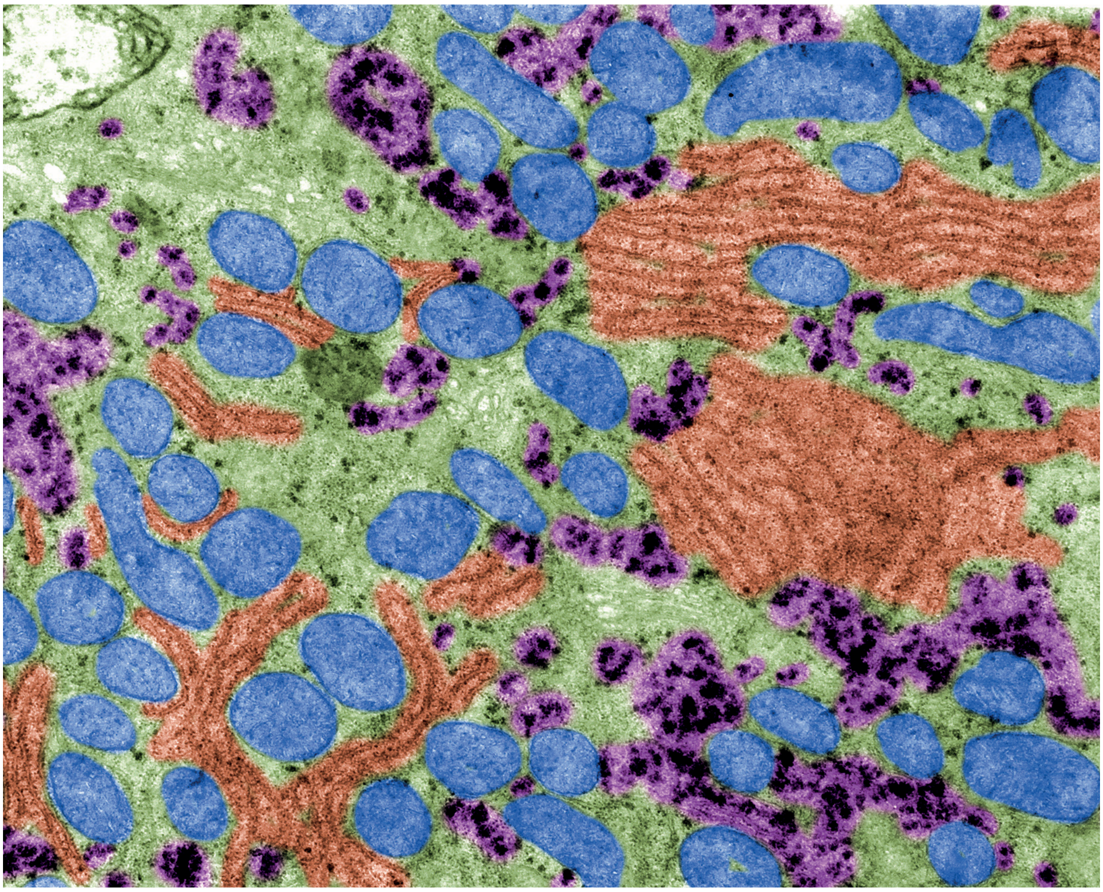

Microscopic image of a liver cell (hepatocyte), with mitochondria highlighted in blue

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vived. However, by studying the diversity of modern Eukaryota and analyzing their genomes, we've been able to piece together significant insights into their evolutionary history and the key role of a cooperative phenomenon called *endosymbiosis*.

The development of complex eukaryotic cells was one of the most pivotal moments in the evolution of life on Earth. The process by which such cells came to exist is called *eukaryogenesis* – but how exactly did it proceed? It is now well understood that eukaryotes evolved after bacteria and archaea, with current evidence suggesting they originated from a specific lineage of archaea. However, bacteria also played a major role in eukaryogenesis and the subsequent evolution of eukaryotic organisms. One of the most extraordinary processes in the evolution of these cells was bacterial endosymbiosis. This phenomenon involves close cooperation between two cells – one of which is engulfed by the other. Rather than being digested, the engulfed cell starts working symbiotically with its host. Over time, this relationship evolved into specialized organelles – mitochondria and chloroplasts.

Mitochondria and chloroplasts retain many features of their bacterial ancestors. For example, they are surrounded by membranes that originated from bacteria, and they still have their own genetic material. However, compared to their free-living bacterial ancestors, they contain far fewer genes. As the symbiotic relationship deepened over time, many endosymbiont genes were lost, as they were no longer essential in their new environment. The endosymbiont began to depend on its host's metabolic processes, making the retention of a complete genome unnecessary. In fact, some of the endosymbiont's genes were transferred to the host's genome over the course of evolution. The host cell produces proteins from these genes and targets them to the organelles to perform their functions. This transferal of genes, known as *endosymbiotic gene transfer*, has further integrated the two partners and allowed the host to exert more control over the endosymbiont. One hypothesis suggests that endosymbionts retain only those genes that enable them to respond quickly to environmental changes and regulate their metabolism, particularly the key processes in organelles, such as electron transfer in the organelle membrane. Reactions of this kind are crucial for producing cellular energy in mitochondria and for photosynthesis in chloroplasts.

Since it became widely accepted that mitochondria and chloroplasts originated through endosymbiosis, scientists have been working to uncover the finer details of this process. In this evolutionary detective work, genetic information has been crucial. Although the genomes of endosymbionts are greatly reduced, they still contain from a few dozen to a few hundred genes. Analyzing these genes has provided compelling evidence that mitochondria originated from alphaproteobacteria and chloroplasts from cyanobacteria. Among today's diverse bacteria, we can even identify living relatives of these ancient organelles. For mitochondria, there are several candidates, while for chloroplasts, the closest known relative is a recently described freshwater cyanobacterium called *Gleomargarita lithophora*.

Evolution never stands still

In addition to experiencing gene loss, chloroplasts and mitochondria have continued to evolve in other fascinating ways. Some of the most striking examples involve the complete loss of the typical functions of these organelles. Mitochondria are best known for producing cellular fuel (ATP) through respiration, a process that relies heavily on oxygen. But not all eukaryotic organisms live in oxygen-rich environments. Many anaerobic eukaryotes have undergone drastic changes in their mitochondria. In extreme cases, these mitochondria are reduced to such an extent that they went undetected for a long time. They lack a mitochondrial genome and the characteristic cristae (folds inside the mitochondria), and they no longer produce ATP. Instead, their primary role, which is also crucial in oxygen-using mitochondria, is to assemble simple molecules known as Fe-S centers (iron-sulfur clusters). These clusters are essential for many vital proteins and play a key role in key cellular reactions by binding and releasing electrons.

The ability to build Fe-S centers is inherited from the bacterial ancestors of mitochondria, and eukaryotic cells rely on mitochondrial machinery to do this. Interestingly, in rare cases, anaerobic eukaryotes have found alternative ways to assemble these centers. They have acquired genetic material from other organisms that also have the capacity to form Fe-S centers, through a process known as *horizontal gene transfer*. In these cases, the Fe-S centers are built in the cytosol rather than in reduced mitochondria. When this happens, mitochondria can become redundant and eventually disappear altogether. To date, this total loss of mitochondria has only been observed in a single group of anaerobic eukaryotes, specifically the protist *Monocercomonoides exilis*, which inhabits the intestines of certain vertebrates.

Similar reductions have occurred with chloroplasts. In nearly every lineage of organisms that contains these organelles, the photosynthetic function has been lost over the course of evolution. Green chloroplasts have transformed into colorless organelles. This radical change is seen in some plants but is most common in single-celled parasites, like the malaria parasite *Plasmodium*. Despite being heavily reduced, the chloroplasts in *Plasmodium* still carry out important metabolic processes. Understanding these changes is not just a scientific curiosity – it could also help in developing antimalarial therapies by targeting these colorless plastids and their proteins, which are absent in host cells that lack chloroplasts.


Paramecium bursaria with symbiotic algae (green). Single-celled algae (desmids) with chloroplasts formed through primary endosymbiosis with cyanobacteria

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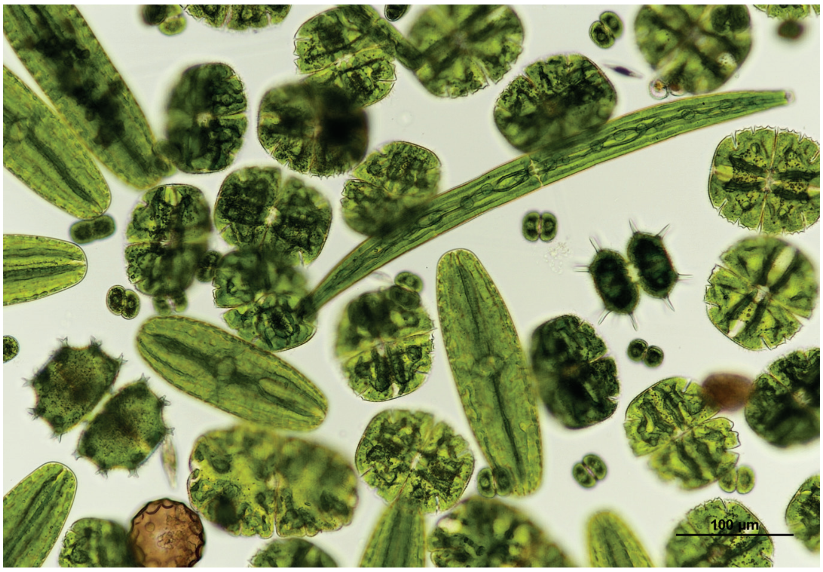

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Unicellular algae (desmids) with chloroplasts formed through primary endosymbiosis with cyanobacteria

Lightning never strikes twice?

Unlike mitochondria, chloroplasts are found in only some evolutionary branches of Eukaryota. In addition to plants, they occur in a wide range of different, often unrelated groups of eukaryotic algae. The endosymbiosis between cyanobacteria and a eukaryotic cell – called *primary endosymbiosis* – gave rise to chloroplasts in the ancestors of green algae, higher plants, red algae, and the lesser-known glaucophytes. Later, single-celled green and red algae themselves became endosymbionts in other eukaryotes through a similar process known as *secondary endosymbiosis*. This led to the acquisition of chloroplasts by many unrelated groups of single-celled microorganisms.

In fact, a recent discovery has added a new twist to the story. Scientists found a small group of shelled amoebas – single-celled organisms that, unlike the amoebas you might remember from school, are encased in a protective shell. Some of these amoebas, specifically those in the genus *Paulinella*, also have cyanobacteria-derived chloroplasts. However, these amoebas must have acquired chloroplasts independently from plants. This endosymbiosis occurred only about 60–200 million years ago, which is relatively recent compared to the much older chloroplast and mitochondrial endosymbiosis events, which happened over 1.5 billion years ago. This discovery has revolutionized our understanding of endosymbiosis. Comparative studies have revealed numerous similarities between these two endosymbiosis events, including gene loss, gene transfer from the endosymbiont to the host genome, and the host's extensive control over the endosymbiont via proteins encoded in its nuclear genome. There is now no reason not to consider the amoeba's endosymbiont a fully functioning organelle.

The world as we experience it is dominated by large eukaryotic organisms: animals, plants, and fungi. These multicellular life-forms share ancestors with an immense variety of eukaryotic microorganisms. Today, we are uncovering more of this hidden diversity thanks to modern sequencing and imaging technologies, though some major groups of eukaryotic microorganisms have been known since the earliest days of light microscopy. These organisms are collectively called protists (previously also known as protozoa, though this term is reserved for heterotrophic protists, while many others are photosynthetic and often referred to as algae). Research on protists has shown that they frequently interact with bacteria, which can become their endosymbionts. Studying these relationships helps us understand how symbiosis between a host and its symbiont begins, shedding light on the ancient endosymbiosis events that gave rise to today's organelles.

Just a few months ago, in 2024, scientists described a new organelle of endosymbiotic origin, which they named the *nitroplast*. As the name suggests, this organelle is involved in nitrogen fixation. So far, it has only been found in a small group of marine algae, but it's possible that nitroplasts exist in other organisms. Moreover, perhaps still other, as yet undiscovered organelles are out there, just waiting to be found by inquisitive researchers. ■

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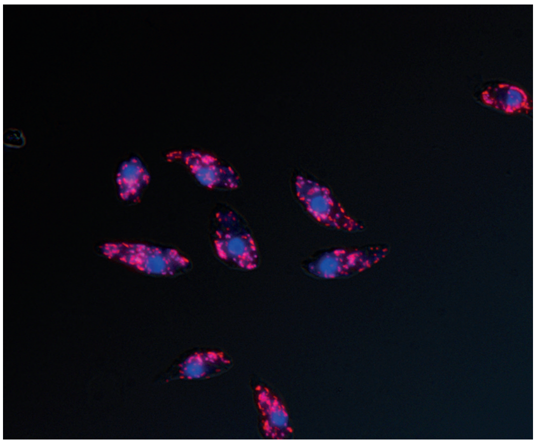

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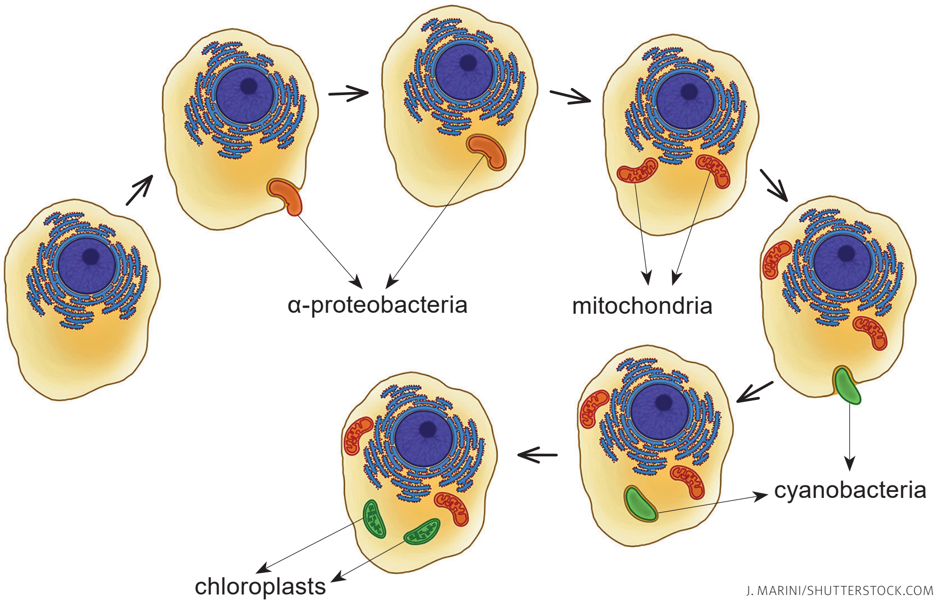
Protist *Diplonema japonicum* with bacterial endosymbionts marked in purple using a fluorescent probe. The cell nucleus is stained blue with DAPI fluorescent dye



Mitochondria originated when a proto-eukaryotic cell engulfed oxygen-using (aerobic) bacteria. Later, chloroplasts formed in a similar process through endosymbiosis with cyanobacteria

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Further reading:

Archibald J.M., Endosymbiosis and Eukaryotic Cell Evolution, *Current Biology*, 2015, https://www.sciencedirect.com/science/article/pii/S0960982215008891

Burki F., Mitochondrial Evolution: Going, Going, Gone, *Current Biology*, 2016, https://www.sciencedirect.com/science/article/pii/S0960982216303542

Husnik F. et al., Bacterial and archaeal symbioses with protists, *Current Biology*, 2021, https://www.sciencedirect.com/science/article/pii/S0960982221007478

Karnkowska A., Maczíszewski K., Should I stay or should I go? Retention and loss of components in vestigial endosymbiotic organelles, *Curr Opin Genet & Dev*, 2019, https://www.sciencedirect.com/science/article/pii/S0959437X19300425

Massana R., The nitroplast: A nitrogen-fixing organelle, *Science*, 2024, https://www.science.org/doi/full/10.1126/science.ado8571