

EVOLUTIONARY GENOMICS

Algae's complex origins

The nuclear genomes of two of nature's most complex cells have been sequenced. The data will help to determine the evolutionary path from symbioses between species to a multi-compartmental unicellular organism.

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The orchestration of cellular function from genetic information is a marvel, even in a typical cell that has a single genome. But a few unusual members of a group of microorganisms called protists pack four genomes, each of unique origin, into a single cell. In a paper published on *Nature's* website today, Curtis *et al.*¹ report the nuclear-genome sequences for two of these organisms: the unicellular algae *Guillardia theta* and *Bigelowiella natans*. These species are each the derivatives of symbiotic relationships between multiple cells, and their endowment of genetic and cellular relicts from these

symbioses makes them the most complex cells known. The genome sequences are the final pieces in a long-standing puzzle, and they will enrich our understanding of how this arrangement evolved and how the cells manage their intricate biochemistry.

The unusual biology of these algae arose from endosymbiosis, which is when a cell lives in irreversible symbiosis within another cell. Endosymbiosis is an important concept — the photosynthetic organelles (plastids) of all photosynthetic eukaryotes ultimately trace back to a cyanobacterium that took up residence within a eukaryotic host more than 1.2 billion years ago². (Eukaryotes are organisms with nucleated cells, such as animals,

plants and fungi, whereas cyanobacteria are prokaryotes, whose cells do not have a nucleus or other membrane-bound organelles.) This fateful encounter is called the primary endosymbiosis (Fig. 1a), because it led to all plastids known today.

Plastids are usually surrounded by two membranes, which are derived from the inner and outer cell membranes of that ancestral cyanobacterium³. But the plastids of some algae have additional membranes — a further two in the case of the algal groups studied by Curtis *et al.* — and the origin of these extra membranes is the story of secondary endosymbiosis⁴. Secondary endosymbiosis occurs when a eukaryotic host acquires a eukaryotic alga as its endosymbiont; the extra membranes around the plastid are the telltale relicts of what was once a whole eukaryotic cell that became an organelle of another (Fig. 1b).

The idea of secondary endosymbiosis was proved correct by the discovery of nucleomorphs — the remnant nuclei of these eukaryotic endosymbionts. Nucleomorph-carrying organisms retain not only the membranes of this relict nucleus, but also many of its genes: nucleomorph genomes consist of three linear 'bonsai'

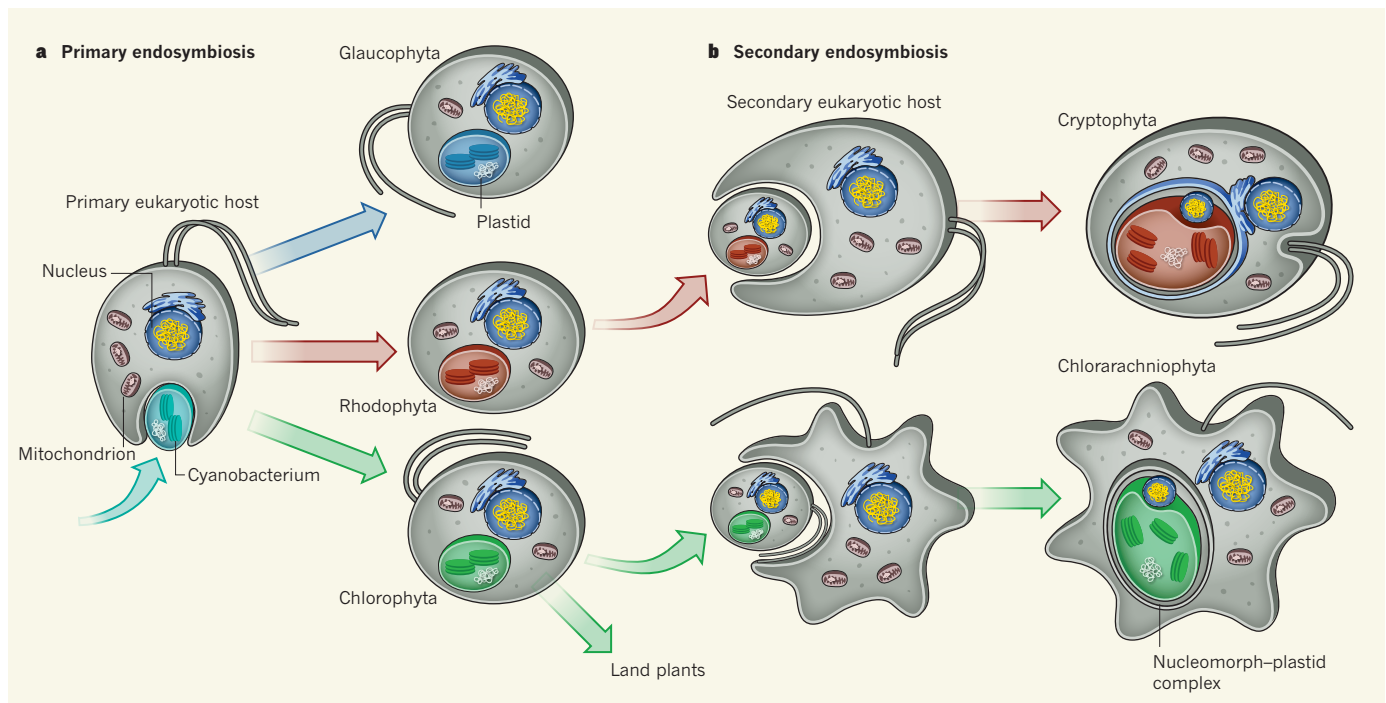


Figure 1 | The rise of photosynthetic eukaryotes. a, During an evolutionary event referred to as primary endosymbiosis, a eukaryotic cell irreversibly assimilated a prokaryotic cell called a cyanobacterium. This generated three lineages of alga (glaucophyta, rhodophyta and chlorophyta) that contain a photosynthetic organelle (plastid), which is the remnant of the cyanobacterium, and mitochondria, which are organelles derived from the integration of other prokaryotes early in eukaryotic evolution. Members of one of these lineages, the chlorophytes, gave rise

to all extant complex land plants. **b,** In two independent evolutionary events, other non-photosynthetic eukaryotic hosts then engulfed cells from both the rhodophyta (red algae) and chlorophyta (green algae) lineages. This secondary wave of endosymbiosis led to two further algal groups, the cryptophytes and the chlorarachniophytes. These unicellular organisms contain not only plastids and mitochondria, but also an organelle called the nucleomorph, which is the relict of the nucleus of the engulfed algae.

chromosomes, each less than 200 kilobases in size (for *Guillardia* species) and are the smallest eukaryotic genomes known⁵. Many extant protist groups display organellar evidence of secondary endosymbiotic events, such as the dinoflagellates (the cause of red tides) and the apicomplexa (parasitic organisms that include the *Plasmodium* species that cause malaria).

The algae that Curtis *et al.* studied are from two groups that independently evolved to bear nucleomorphs — the cryptophytes (*G. theta*) and the chlorarachniophytes (*B. natans*). These cells express, and make use of, four distinct sets of protein-synthesis machinery known as ribosomes. Two types of ribosome are prokaryotic, belonging to the plastids and the mitochondria (which are eukaryotic organelles also originally derived from endosymbiotic prokaryotes). The other two types are eukaryotic: the cytoplasmic ribosome and that of the periplastidial compartment, which is the remnant cytoplasm of the engulfed alga. At some evolutionary stage, these organisms had five genomes: two nuclear and two mitochondrial (from the two eukaryotic symbionts) and one plastidal. Over time, however, the mitochondrial genome of the endosymbiont was lost, leaving four genomes in the extant cells. Sounds complex? It is! Which is why Curtis and colleagues' genome sequences provide rich insight into cell and genome evolution, and the opportunity to study sophisticated intracellular communication and molecular logistics.

The different types of membrane-bound compartment in nucleomorph-bearing cells all have to be serviced by proteins coded for by nuclear genes, and some of these proteins have to cross five membranes before they reach their destination⁶. The number of these compartment-destined proteins predicted from Curtis and colleagues' nuclear genome is much greater than expected. This may be the final curtain for an old idea that some genes fail to be transferred from organelles to the nucleus because the hydrophobic nature of their protein products would make it difficult for them

to be transported across multiple membranes⁷. Further experiments will be needed to clarify the biochemical compartmentalization in these cells and how the complex trafficking of proteins and metabolites is organized.

Curtis *et al.* also found that RNA transcripts produced by *Bigeloviella* algae undergo more alternative splicing than any other unicellular eukaryote that has been studied. (Alternative splicing is when segments of a transcript can be joined in different conformations to produce different forms of a given protein or lead to different subcellular localization.) Why this organism splices so wildly and how this influences its biology are questions for the years to come.

Both algal genomes also harbour what evolutionary biologists call conflicting phylogenetic signals: there are a surprisingly large number of genes of apparent green-algal origin in *Guillardia*, which actually harbours the remnants of a red alga, and too many red-algal genes in the green-alga-harbours *Bigeloviella* (Fig. 1b). This Pandora's box was first opened when such 'green signals' were identified in related heterokontophyte algae, and additional layers of endosymbiotic events were proposed to explain this apparent conflict⁸. However, more recent analyses favour phylogenetic errors and sampling bias as the explanation^{9,10}. As more red algal genomes are sequenced and a more balanced set of reference genomes becomes available, the conundrum of what early evolutionary track these complex cells took will probably be solved.

Perhaps the most exciting insight revealed by the two genomes is why these cells have not lost the nucleomorph — a question that has puzzled researchers since the discovery of these organelles. Eukaryotic nuclear genomes are typically replete with copies of mitochondrial or plastid DNA¹¹. Humans, for example, have about 500 kilobases of mitochondrial DNA strewn across their chromosomes¹². These insertions arise when one of these organelles lyses and releases its DNA, fragments of which then enter the nucleus,

with some being successfully integrated into the nuclear DNA.

But what if there were only one mitochondrion per cell? Its lysis would mean no mitochondrion-bearing progeny and the death of that cell lineage (because mitochondria are essential for cellular energy provision). Notably, only one nucleomorph-plastid complex exists per cell in *Guillardia* and *Bigeloviella*. So, although Curtis *et al.* find mitochondrial DNA in both nuclear genomes, indicating that recent gene transfer from the mitochondrial organelles has occurred, they find no recently transferred copies of nucleomorph or plastid DNA. This implies that the presence of a single nucleomorph-plastid complex prevents any further gene transfer in that cell, such that the nucleomorph is evolutionarily frozen. This constellation tells us that it takes two to tango on the route to becoming an enduring secondary organelle. ■

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