

# The Early Evolution of Eukaryotes: A Geological Perspective

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Molecular phylogenies of eukaryotic organisms imply patterns of biological and environmental history that can be tested against the geological record. As predicted by sequence comparisons, Precambrian rocks show evidence of episodic increases in biological diversity and atmospheric oxygen concentrations. Nonetheless, complete integration of the two records remains elusive and may require that the earliest macroscopic organisms be recognized as extinct experiments in eukaryotic multicellularity.

The Eucarya, or eukaryotes, constitute one of the three principal domains of life (1). They encompass the conspicuous organisms of our daily experience—plants, animals, and fungi—as well as a phylogenetically diverse array of microorganisms and sea weeds, often grouped paraphyletically as protists, or protoctists (2). Defined by a membrane-bounded nucleus, eukaryotes are distinguished evolutionarily by endocytosis, the ability of a dynamic cytoskeletal and membrane system to engulf external materials and bring them into the cell and, in several lines, by a remarkable capacity for multicellular growth and differentiation. These two attributes account for much of the evolutionary success of the group.

The roots of the Eucarya lie phylogenetically among the protists and temporally within the long Precambrian era. In recent years, molecular data have permitted a new understanding of the former, while paleontological and geochemical discoveries have shed increasing light on the latter. Paleontology and molecular phylogeny describe the same biological history, and although they illuminate different aspects, the two records should be complementary. Inferences made on the basis of one approach should find tests in the other. In this article I examine the goodness of fit between rapidly advancing molecular phylogenies and the emerging view of early eukaryote evolution provided by geology.

## Molecular Phylogeny of the Eucarya

In 1965, Zuckerkandl and Pauling (3) argued that sequence comparisons of informational macromolecules permit the evaluation of evolutionary relatedness, thereby fomenting a phylogenetic revolution. Current phylogenies of the Eucarya depend principally on small (4–6) or large (7)

subunit ribosomal RNA (rRNA), although 5S rRNA and a number of protein sequences also inform phylogenetic interpretation. The principal features of eukaryote evolution inferred from molecular analyses can be summarized as follows (Fig. 1):

1) The Eucarya is an extremely ancient group, as old as the prokaryotic Bacteria (eubacteria) and Archaea (archaeobacteria), or nearly so (4, 8). Branching relationships among the three domains have been debated since the archaeobacteria were first recognized more than a decade ago. Commonly, universal trees have been unrooted, with the suggestion that the three kingdoms differentiated more or less simultaneously from a common ancestor (8). Other hypotheses have also been championed (9). Sequence analyses of protein-encoding genes that duplicated before the divergence of the domains now suggest that life's tree should be rooted between the eubacteria and archaeobacteria, with eukaryotes bearing a specific phylogenetic relationship to the latter group (10, 11). Regardless of branching topology, the retention in eukaryotic cells of molecular features interpreted as primitive (12) implies that the group became distinct early in the history of life.

2) The lowermost branches of the eukaryotic tree contain aerotolerant anaerobes, most of which live parasitically within animal hosts (4, 13). These organisms have a well-defined nucleus and flagellar apparatus, but no mitochondria or chloroplasts. They have relatively simple cytoskeletons and exhibit a number of ultrastructural and biochemical characters more similar to those of prokaryotes than to other eukaryotes.

3) Protists occupying the middle branches of the eukaryotic tree commonly contain mitochondria, but no chloroplasts. The exception is the euglenids, about one-third of whose species are photosynthetic. Euglenid chloroplasts may be derived from symbiotic green algae (14), implying a relatively late acquisition of photosynthesis

within this group. Although predominantly aerobic, organisms in this section of the tree commonly thrive under relatively oxygen-poor conditions (15).

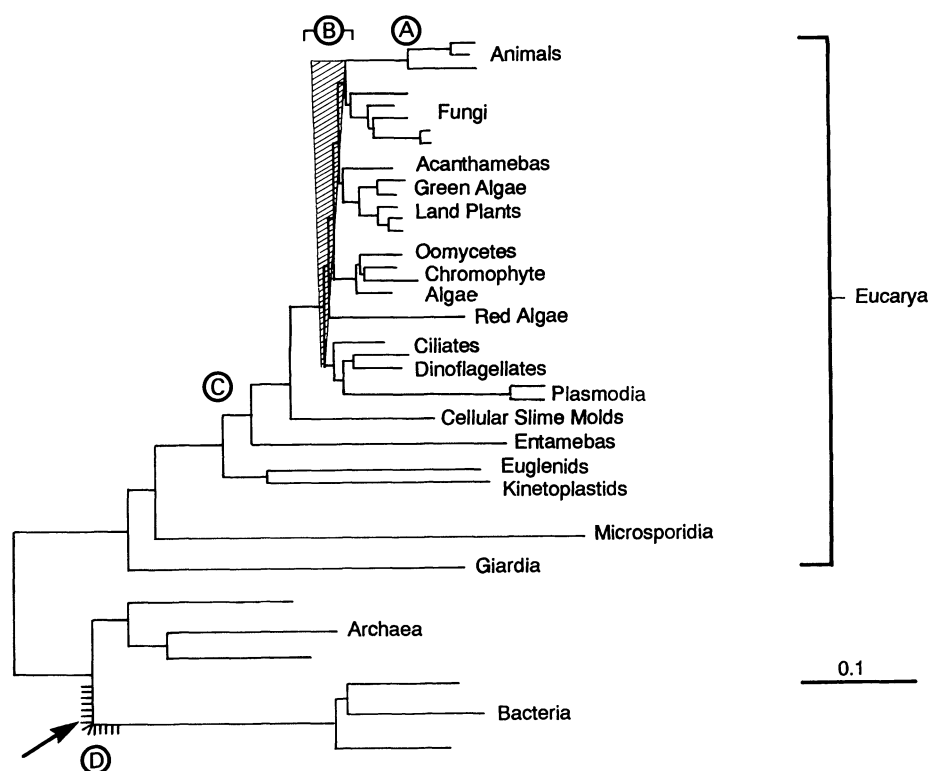
4) Most eukaryotic diversity is nested within the densely branched crown of the phylogenetic tree (4–7). Major clades that branch near a common point include the animals, fungi, green algae and land plants, chromophyte algae and related heterotrophs, red algae, and a complex comprising the ciliates, dinoflagellates, and plasmodia. A rapid burst of evolution is inferred. Because of rapid diversification, branching order within the crown remains uncertain (4–7). With the exception of the euglenids, photosynthetic eukaryotes occur in these upper branches.

5) Mitochondria and chloroplasts have molecular sequences that ally them to the eubacterial proteobacteria and cyanobacteria, respectively (16, 17). The sequence data complement ultrastructural and biochemical evidence supporting the endosymbiotic theory for the origins of these organelles (18). Molecular data also support the multiple origins of chloroplasts, with some plastids originating from eukaryotic symbionts (19). Chloroplasts may have arisen as many as six or more times, with separate symbioses giving rise to the rhodophytes, chlorophytes, chromophytes, cryptophytes, photosynthetic euglenids, and photosynthetic dinoflagellates [which may contain several discrete chloroplast lineages within the group (15)].

6) Coelomate animal phyla diverged in a later episode of rapid diversification; however, the coelomate phyla were preceded by acoelomate triploblastic animals and diploblasts (20–22). Christen *et al.* (22) suggest that the evolutionary split between diploblastic (placozoa, sponges, cnidarians, and ctenophores) and triploblastic (bilaterally symmetric) animals occurred as part of the greater rapid radiation of higher eukaryotes.

The patterns of form and physiology that characterize this (still emerging) phylogenetic tree provide predictions that may be tested independently in the geological record. As Woese (8, p. 252) wrote, "What this preliminary phylogeny begins to suggest is that major epochs in eukaryote evolution corresponded to major periods of Earth history." The implication is that the paleontological record should show several stepwise bursts of

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**Fig. 1.** The molecular phylogeny of eukaryotic organisms based on sequence comparisons of small subunit rRNA and constructed by distance matrix methods (4–6). The scale bar corresponds to a distance of ten changes per 100 nucleotide positions. The arrow and hatched lines indicate the position of the tree root, as inferred from analysis of protein sequences (9). Letters correspond to points in the phylogeny that may be tied to the geological record: A, the radiation of coelomate animal phyla, about 560 to 540 Ma; B, the major radiation of phenotypic diversity in eukaryotes, suggested on the basis of fossils to be 1100 to 1000 Ma, but possibly substantially earlier (the figure emphasizes the uncertainty of branching order among the principal clades of higher eukaryotes); C, the acquisition of mitochondria, inferred from the geochemical record to be 2400 to 2800 Ma, but possibly earlier; and D, the diversification of the major domains, 3500 Ma or earlier.

diversity increase and that these steps should correspond to independently identified periods of environmental change.

### The Fossil Record of Early Eukaryote Evolution

Fossils are the traditional documents of record concerning evolutionary history. In plant and animal evolution, the record has proven indispensable, regardless of progress in molecular studies because (i) it provides examples of character combinations that differ from those found today, (ii) it provides a direct record of time in evolution, and (iii) it can be combined with sedimentological and geochemical data to provide an indication of the environmental circumstances attendant on major evolutionary events. All of these attributes apply equally well to Precambrian evolution.

*Interpreting the early fossil record.* Before proceeding to a discussion of Precambrian biological history, a word of caution is necessary. Fossil preservation is facilitated by mineralized hard parts and structures composed of organic materials that resist

degradation. Many organisms have neither and so have little chance of fossilization. Those that do may only live in certain environments or become fossilized under specific sedimentary conditions. Thus, fossil presence and absence cannot be treated symmetrically.

The presence of *Eoentophysalis belcherensis* in cherts of the Belcher Supergroup, Canada, provides unambiguous evidence that cyanobacteria existed 2000 million years (Ma) ago; however, the absence of brown algae from the same deposit may reflect any of several circumstances. Possibly, brown algae had not yet evolved. Alternatively, they may have existed but had a low probability of preservation. Or they may have existed but not inhabited the semiarid tidal flat environments represented by Belcher rocks. They may even have lived in the environment and become fossilized, but having lost all traces of pigment and ultrastructure be unrecognizable as brown algae.

Clearly, the paleobiological interpretation of absence requires extensive sampling of the geological record. This means that

for any time interval, one needs numerous samples representing a range of sedimentologically defined environments, exhibiting good fossil preservation, and prepared in ways that maximize likelihood of recovery. By this criterion, only the Neoproterozoic Era (1000 to 540 Ma) can be said to be at all well sampled (23). The record of the preceding 1000 Ma is patchy, with instances of good preservation but thin paleoenvironmental coverage and almost no *Lagerstätten* (exceptionally well-preserved fossil assemblages) comparable to those found in Neoproterozoic successions. Sedimentary rocks older than 2000 Ma are relatively rare, commonly metamorphosed, and so poorly sampled that evolutionary interpretation of the fossil record per se becomes a risky undertaking.

*The earliest records of eukaryotic life.* The oldest fossils whose interpretation as protistan seems probable are large (40 to 200  $\mu\text{m}$ ) spheromorph acritarchs (24) from the Chuanlinggou Formation, China, dated with some uncertainty at 1800 to 1900 Ma (25). In general morphology, size frequency distribution, and sedimentary distribution, this population is similar to assuredly eukaryotic fossils in younger rocks; nonetheless, its systematic interpretation is a statistical likelihood, not a taxonomic certainty. Cyanobacterial sheaths can reach the size of the Chuanlinggou fossils, and a few blue-green envelopes exhibit regular tears that resemble the median split excystment structures of protists. Higher in the same succession, Hofmann and Chen (26) have reported millimeter-scale organic disks and 70- to 250- $\mu\text{m}$  wide filaments preserved as compressions on shale surfaces. These fossils may also be eukaryotic, but like the smaller spheromorphs, classification at even the phylum level is impossible.

Independent evidence for late Paleoproterozoic (2500 to 1600 Ma) eukaryotes comes from steranes in bitumens of the about 1690-Ma-old Barney Creek Formation, northern Australia (26). Steranes are derived from sterols, membrane components characteristic of eukaryotic cells (28). Together, paleontological and biogeochemical evidence suggest that eukaryotic cells were significant features of ecosystems at least as early as 1700 to 1900 Ma ago.

The temptation to view this as the evolutionary first appearance of eukaryotic cells is best resisted. As noted above, the earlier paleobiological record is simply too meager to support any such conclusion. There are no rigorous biomarker studies of rocks older than the Barney Creek Formation, and only a handful of well-studied microfossil assemblages (29). While deposits such as the 2000-Ma-old Gunflint Iron Formation, Ontario, contain no unequivocally eukaryotic remains, they do contain

problematic taxa such as *Eosphaera tylei* (30) and *Leptoteichos golubicii* (31) that could be either eukaryotic or eubacterial.

**Mesoproterozoic (1600 to 1000 Ma) eukaryotes.** Acritarchs of probable eukaryotic origin are abundant in Mesoproterozoic shales, but their diversity remains limited to simple spheroidal morphologies that provide few clues to classification (32–34) (Fig. 2A). Only near to the end of the era do the first modestly sculptured forms appear (23, 34). Despite the limited diversity of probable eukaryotic unicells, the fossil record contains evidence of diversification in the form of macroscopic organisms preserved as organic compressions and impressions in sandstones and shales. *Grypania* is a morphologically regular organic tube, about 1 to 2 mm wide and up to 80 mm long, found in 1400-Ma-old shales from China, India, and North America (35) (Fig. 3B). It is uncontroversially eukaryotic, but in the absence of cell preservation, its taxonomic affinities are (once again) uncertain. More complex but equally problematic organiza-

tion is implied by strings of millimeter-scale beads preserved as impressions in Mesoproterozoic sediments from Australia and North America. Grey and Williams (36) have suggested that these impressions may reflect articulated organisms reminiscent of the modern seaweed *Hormosira*.

The oldest multicellular protist (or, indeed, eukaryote of any kind) that can be assigned with confidence to an extant phylum is a bangiophyte red alga preserved in silicified carbonates on Somerset Island, Canada, indirectly dated as 1260 to 950 Ma (37) (Fig. 3C). This population provides one of the most significant tiepoints for the molecular and fossil records of early eukaryotic evolution. Collectively, these Meso- to earliest Neoproterozoic fossils refute the notion that the latest Proterozoic radiation of animals was the first evolutionary expression of complex multicellularity in eukaryotes.

**Neoproterozoic (1000 to 540 Ma) eukaryotes.** The taxonomic diversity of Neoproterozoic eukaryotes far exceeds that of any

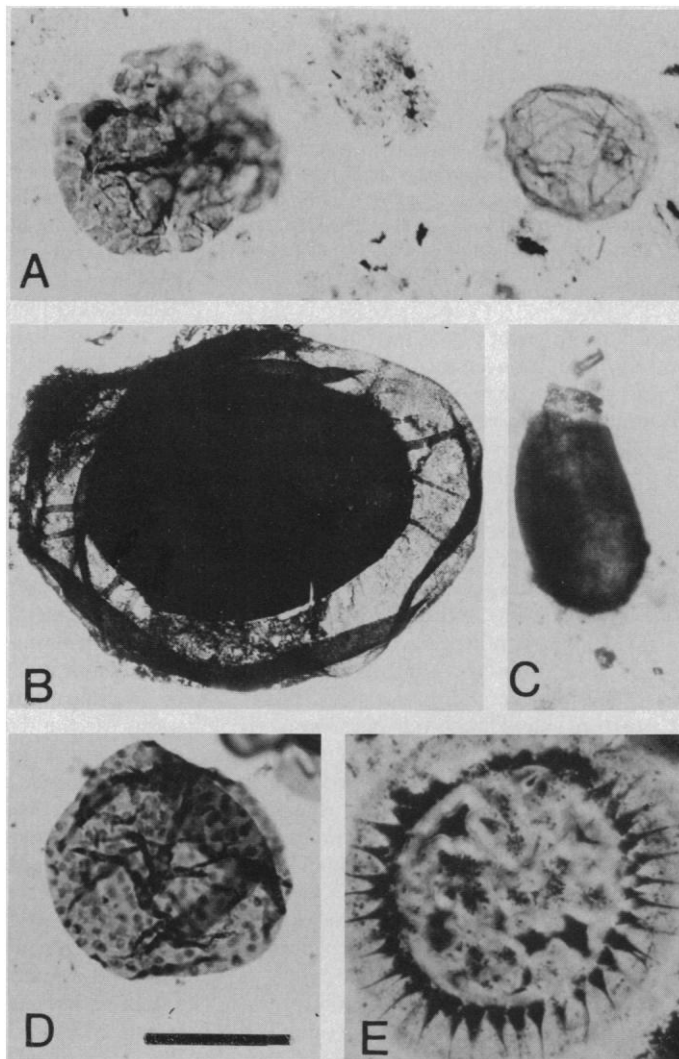
earlier era (23). Multicellular algae are well known from a handful of Neoproterozoic *Lagerstätten*. The spectacular fossil assemblage recovered from mudstones of the 900- to 1000-Ma Lakhanda Group, eastern Siberia, contains half a dozen cellularly preserved metaphytes (38) (Fig. 3, D and E), all of uncertain systematic relationships. It also includes *Eosaccharomyces ramosus*, a colonial fossil whose web-like network of oriented cells resembles the aggregation swarms of certain slime molds (Fig. 3A). While neither this analogy nor its name provides any necessary clue to the systematics of *Eosaccharomyces*, the preserved orientation of cells along bedding planes clearly indicates a sophisticated level of intercellular chemical communication and behavioral response.

Somewhat younger shales of the (~750 Ma) Svanbergfjellet Formation, Spitsbergen, preserve an even larger sampling of multicellular algae (39, 40), including forms that bear significant morphological similarity to green algae such as *Cladophora* and *Coelastrum*. As recent molecular phylogenies of the green algae show *Cladophora* and its relatives to be a late branching group (41), these fossils, if correctly interpreted, indicate that the major features of green algal diversity were already established 750 Ma ago. Macroscopic compressions of problematic, multicellular eukaryotes are additionally known from a dozen or more formations ranging in age from 850 Ma to the Precambrian-Cambrian boundary (42). Such fossils may reflect a Neoproterozoic algal radiation, but the absence of comparable *Lagerstätten* in older rocks undermines confidence in this interpretation.

Neoproterozoic radiation is better documented by single-celled protists, and again, the Lakhanda and slightly younger Miroyedicha biotas of Siberia provide a baseline for interpretation (38). In addition to the simple acritarchs that characterize older formations, these shales contain a variety of verrucate, corrugated, or otherwise modestly sculptured vesicles (Fig. 2D), as well as *Trachyhystrichosphaera*, a genus of spiny acritarchs more than 300  $\mu\text{m}$  in diameter (Fig. 3B). The Svanbergfjellet (39, 40) and overlying Draken (43) formations also contain *Trachyhystrichosphaera* (up to 2700  $\mu\text{m}$  in diameter), but contain as well an increased diversity of morphologically complex acritarchs ranging in size from 10 to 20  $\mu\text{m}$  to millimeter scale. Vase-shaped vesicles (100 to 250  $\mu\text{m}$  long) with distinct oral apertures are also common in rocks younger than 850 Ma (Fig. 2C).

Among the most distinctive of Neoproterozoic microfossils are morphologically diverse disk-like scales found in 610- to 750-Ma-old cherts from the upper Tindir

**Fig. 2.** Proterozoic protists. (A) Simple acritarchs from the Mesoproterozoic Roper Group, Australia; (B) *Trachyhystrichosphaera aimika* from the 900- to 1000-Ma Lakhanda Group, Siberia; (C) A vase-shaped protist from the 700- to 800-Ma old Akademikerbreen Group, Spitsbergen; (D) a modestly ornamented acritarch from the 850- to 950-Ma Miroyedicha Formation, Siberia; and (E) a large spine-bearing acritarch from the 590-Ma Doushantuo Formation, China. Scale bar in (D) represents 50  $\mu\text{m}$  for (A) and (C), 120  $\mu\text{m}$  for (B) and (E), and 25  $\mu\text{m}$  for (D).



Group, Yukon Territory (44, 45). But for their larger size (up to 30  $\mu\text{m}$ ), they resemble the siliceous scales of chrysophyte algae. The Tindir fossils, along with calcareous multicellular algae from Namibia (46) and, possibly, California (47), indicate that eukaryotic biomineralization significantly predates the Precambrian-Cambrian boundary.

The acme of Neoproterozoic acritarch diversity is found in rocks deposited after the great Varanger Ice Age (590 to 610 Ma) but prior to the principal radiation of Ediacaran-grade metazoans (48–50). In these assemblages, the large, morphologically complex protists that characterize the Neoproterozoic record are represented by more than two dozen taxa (Fig. 2E), in addition to smaller spiny forms and the simple sphaeromorphs that are ubiquitous in Proterozoic deposits. Increasing evidence supports the hypothesis that most morphologically complex forms do not persist until the end of the Proterozoic (49–51). Well-sampled records from the East European Platform (52) and elsewhere indicate that acritarch diversity decreased sharply at about the time of the major Ediacaran animal radiation. Globally, latest Proterozoic biotas are characterized by simple, thin-walled sphaeromorphs and rare, small acanthomorph forms; renewed diversification of prasinophytes and acritarchs parallels the Cambrian radiation of coelomate animals (53).

The systematic affinities of most Neoproterozoic eukaryotes remain uncertain, although one can make educated guesses. For example, the degradation-resistant cysts of *Trachyhystrichosphaera* show evidence of vegetative growth within an expanding cyst wall; a volumetric size increase of more than two orders of magnitude can be documented (43). Among living algae, this combination of features characterizes the phycmata of prasinophyte algae (54). Prasinophyte affinities have repeatedly been suggested for Neoproterozoic acritarchs; undoubted prasinophytes occur in basal Cambrian rocks (53). Biomarker studies independently indicate that both green and red algae were abundant in Neoproterozoic ecosystems (27).

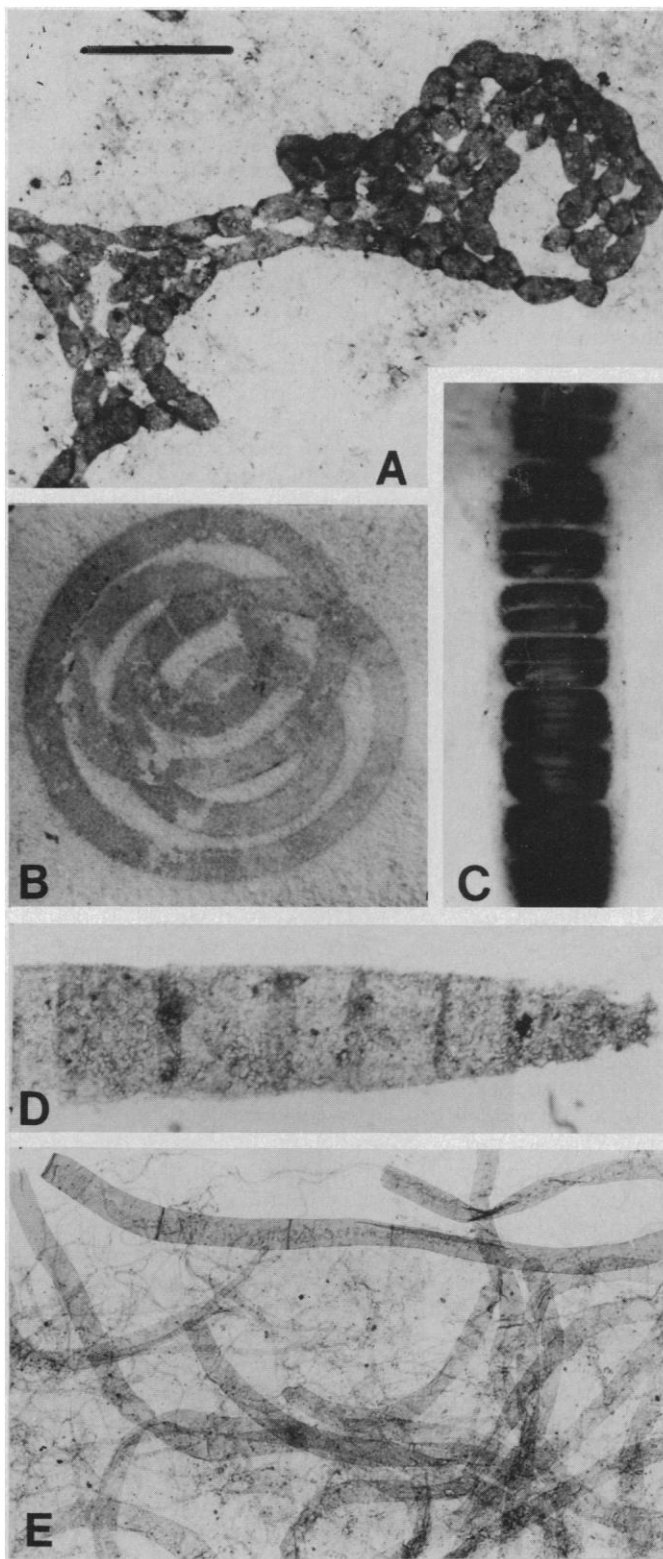
Despite inadequate sampling of Mesoproterozoic strata, it is clear that the Neoproterozoic radiation of single-celled eukaryotes is a real phenomenon and not an artifact of sampling. On a practical note, the evolutionary dynamics of Neoproterozoic protists permit the biostratigraphic subdivision and correlation of Neoproterozoic sedimentary successions (23, 36, 55).

### Environmental Evolution

It is generally agreed that surface environments changed appreciably through early

Earth history, but given a limited number of empirical constraints, varying interpretations of the timing and magnitude of these changes have been entertained. This is particularly true with regard to the physiologically important history of atmospheric oxygen. Several recent findings, however, constrain the broad outlines of Precambrian

oxygen evolution. Models of the primitive Earth indicate that prior to the evolution of cyanobacterial photosynthesis, the partial pressure of oxygen ( $p\text{O}_2$ ) in the atmosphere must have been low, perhaps as low as  $10^{-10}$  atm (56). Following the evolution of oxygenic photosynthesis,  $p\text{O}_2$  increased to concentrations capable of supporting aro-



**Fig. 3.** Early colonial and multicellular eukaryotes. (A) *Eosaccharomyces ramosus*, detail of a large colony from the 900- to 1000-Ma Lakhanda Group, Siberia; (B) *Grypania spiralis* from the 1400-Ma-old Gaoyuzhuang Formation, China; (C) detail of a bangiophyte red alga from the 1250- to 950-Ma Hunting Formation, Canada; (D and E) filamentous algae from the Lakhanda Group. In (E), the small string-like filaments interwoven with the alga are cyanobacterial sheaths. Scale bar in (A) represents 25  $\mu\text{m}$  for (A), 5 mm for (B), 50  $\mu\text{m}$  for (C), 400  $\mu\text{m}$  for (D), and 250  $\mu\text{m}$  for (E).

bic respiration; this occurred at least 2800 to 2400 Ma ago (57) and possibly earlier (58). However, high rates of oxygen consumption kept  $pO_2$  at 1 to 2% of the present-day atmospheric level (PAL) until about 1900 Ma, when paleoweathering surfaces indicate a rise to 15% PAL or higher (59). The Mesoproterozoic history of atmospheric oxygen is poorly constrained, but increasing geochemical data indicate that the period just prior to the emergence of macroscopic animals was a time of major biogeochemical change, including, quite possibly, major increases in  $pO_2$  (60, 61). Thus, a picture has begun to emerge of long intervals of relative environmental stasis separated by brief periods of biologically significant increase in  $pO_2$ .

### The Fit of the Data

As predicted by molecular phylogeny, the Precambrian geological record shows evidence for both episodic environmental change and sharp increases in fossil diversity. However, inferred physical and biological changes do not always coincide. Can we reconcile the two geological records with each other and with the phylogenetic relationships of living organisms?

*The earliest eukaryotes.* The oldest fossils of eukaryotes are between 1700 and 1900 Ma old, in apparent agreement with some molecular clock estimates of eukaryote-prokaryote divergence based on protein sequences (62). In contrast, molecular phylogenies based on other proteins and rRNA (4, 8, 10, 11) imply that the Eucarya emerged as a distinct clade early in the history of life, and paleobiological evidence clearly indicates that complex ecosystems fueled by photosynthesis existed at least 3400 to 3500 Ma ago (63). Although this incongruity might be interpreted as evidence against the eukaryotic antiquity implied by molecular-derived phylogenetic trees, an alternative explanation lies in the extreme poverty of the Archean (>2500 Ma) and earliest Proterozoic fossil records, as well as the low preservational potential of most lower eukaryotes. (Protists that branch below the crown of the eukaryotic tree have left few Phanerozoic fossils, let alone Precambrian occurrences.) The anaerobic metabolism of basal eukaryotes is also consistent with an origin early in Earth history.

*The acquisition of mitochondria and plastids.* Phylogenetic trees imply the sequential acquisition of mitochondria and plastids, but it is not clear what biological features would ensure this order of incorporation. As Cavalier-Smith (64) has noted, any cell capable of engulfing a proto-mitochondrial proteobacterium could also swallow a proto-chloroplastic cyanobacterium. The step-

wise environmental history outlined in the previous section may provide at least a partial solution.

Aerobic metabolism would have been impossible in the oxygen-poor environments hypothesized for the early Earth. However, once atmospheric  $pO_2$  increased to about 1 to 2% PAL, aerobic respiration became possible, and the seeds of proto-mitochondrial symbiosis were sown. What prevented the simultaneous acquisition of plastids?

During the long interval when  $pO_2$  stood at 1 to 2% PAL, fixed nitrogen would have been scarce in the surface ocean. Ammonia would have had a very short half-life, and for  $CO_2$  concentrations consistent with biological and geochemical data, abiological nitrate production would have been much lower than today (57). Heterotrophs, including eukaryotes, could have obtained nitrogen easily enough, but autotrophs would likely have had to fix their own nitrogen. Most bacterial autotrophs can indeed fix nitrogen, but for incompletely understood reasons nitrogen-fixation does not occur in chloroplasts within eukaryotic cytoplasm (65). The implication is that the evolutionarily stable acquisition of protochloroplast symbionts may not have been favored until atmospheric oxygen reached levels where nitrate production is much higher, about 10% PAL or more. As noted above, geochemical evidence from the weathering of sideritic iron formations indicates that this occurred about 1900 Ma ago (59), approximately coincident with the cessation of iron formation deposition. Although the poor early fossil record precludes confident interpretation, it may not be a coincidence that the earliest fossilizable and recognizable eukaryotes appear in rocks 1700 to 1900 Ma old.

*The big bang of eukaryotic evolution.* The timing of the evolutionary burst that forms the crown of the eukaryotic tree is one of the most difficult issues in Precambrian paleobiology. The logic presented in the previous paragraph might suggest that the burst began some 1900 Ma ago with the acquisition of chloroplasts. After all, algae occur in more than half of the major branches of this crown. Considering that most marine ciliates and many diploblastic animals contain algal symbionts, the importance of photosynthesis in eukaryote diversification seems clear. Nonetheless, the most obvious diversification in the paleontological record began only about 1200 to 1000 Ma ago. At least three explanations are possible.

First, if we accept the logic of the previous section, it is possible that the Neoproterozoic radiation, insofar as it is real, represents diversification within a few easily fossilized groups rather than among all

branches in the crown. If the simple but abundant acritarchs and rarer macroscopic fossils in Mesoproterozoic rocks are algae, as conventionally interpreted, then the later increase in diversity is most easily interpreted in this way.

Alternatively, it is possible that the logic of the preceding section is flawed or that other factors inhibited diversification after the chloroplast problem had disappeared. Not only is the crown of the eukaryotic tree studded with photoautotrophs, it is also where complex multicellularity resides within the domain. Further, evidence from population genetic studies suggests that whatever their capacity for sexual reproduction, many lower eukaryotes are effectively asexual (66). Thus, chloroplast acquisition is not the only possible trigger for explosive eukaryotic diversification. One possibility is that a relatively late establishment of sexual population structures—long after the capacities for plastid acquisition and multicellular development were established—led to an explosive radiation of extant (and, doubtless, some now extinct) groups.

In this case, early Neoproterozoic diversification could be equated to the evolutionary burst inferred from the crown. Mesoproterozoic acritarchs would be interpreted as archaic protists and macroscopic fossils as extinct lineages of multicellular Eucarya. In many ways, this is an attractive, if speculative, possibility. Organic geochemical studies of Proterozoic bitumens are limited in number, but analyses published to date suggest a marked increase in the abundance and diversity of eukaryotic biomarkers beginning 900 to 1200 Ma ago (27). More generally, the fossil record is littered with extinct branches broken from the tree of life. This possibility highlights the principal limitation of molecular phylogeny in historical inquiry—it cannot tell us anything about extinct groups of organisms.

The third, and in many ways least attractive, possibility is that anomalously slow rates of RNA evolution in “crown” eukaryotes provide a misleading sense of elapsed time.

*The emergence of animals.* The clearest radiations in the early fossil record are those of architecturally simple Ediacaran animals about 580 Ma ago and of diverse coelomate invertebrates some 40 Ma later. These events postdate the big bang of eukaryotic evolution by several hundred million years. Therefore, if the clade defined today by animals diverged from other eukaryotes during the big bang, then the Ediacaran event is properly viewed not as the differentiation of a kingdom but more narrowly as the evolution of macroscopic size within the kingdom. Many authors have suggested

that the relatively late appearance of large animals was occasioned by a latest Proterozoic increase in  $pO_2$  (67–69). What is new is that, for the first time, independent geological data provide support for this hypothesis (60, 61). In this case, phylogeny, fossils, and environmental history find a good match.

## Conclusions

Much of what we know about eukaryotic phylogeny and the early fossil record has become available only in the last 6 years. The idea of long-term environmental change throughout the Archean and Proterozoic eons is old, but much of the geological data permitting evaluation of these ideas is not. That there should be points of discrepancy among the emerging records of paleontology, environmental history, and comparative biology is not surprising. Indeed, at the present state of inquiry, the goodness of fit among records is gratifyingly high. Nonetheless, the discrepancies show that a better understanding of all three records will be necessary before their common evolutionary history is revealed. Others can list the desiderata for molecular phylogeny better than I, although the list must surely include further sampling of taxa, particularly eukaryotes that live in environments thought to compare with those of the distant past; phylogenies based on additional molecules; and improved analytical methods for evaluating molecular data. For the geological record, the needs include far better paleontological and organic geochemical sampling of the Paleo- and Mesoproterozoic records, as well as improved techniques for reconstructing the environmental history of the early Earth. With continued research and a little good luck, fossils, environmental history, and comparative biology may soon converge on a single integrated account of early eukaryotic evolution.

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