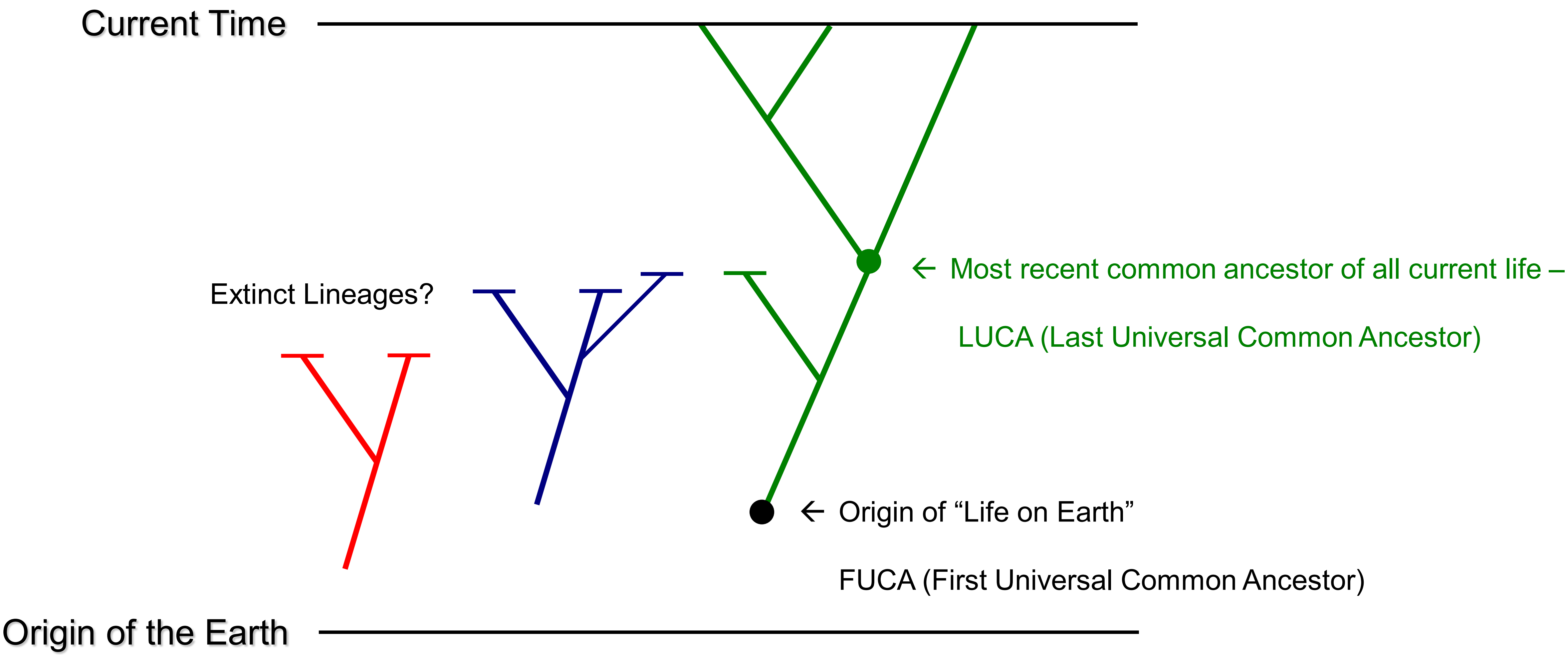
The Unity of Life



Current Time

Extinct Lineages?

← Most recent common ancestor of all current life –

LUCA (Last Universal Common Ancestor)

← Origin of “Life on Earth”

FUCA (First Universal Common Ancestor)

Origin of the Earth

The Baseline Timescale of Key Events

- Life was established on Earth ~4 billion years ago (BYA), with eukaryotes appearing ~3 BYA.

- The common ancestor that eventually gave rise to today’s eukaryotes formed the base of the eukaryotic tree ~2 BYA.

- Each of these time points has a level of uncertainty of a few hundred million years.

Potentially biogenic carbon preserved in a 4.1 billion-year-old zircon

Elizabeth A. Bell[a,1], Patrick Boehnke[a], T. Mark Harrison[a,1], and Wendy L. Mao[b]

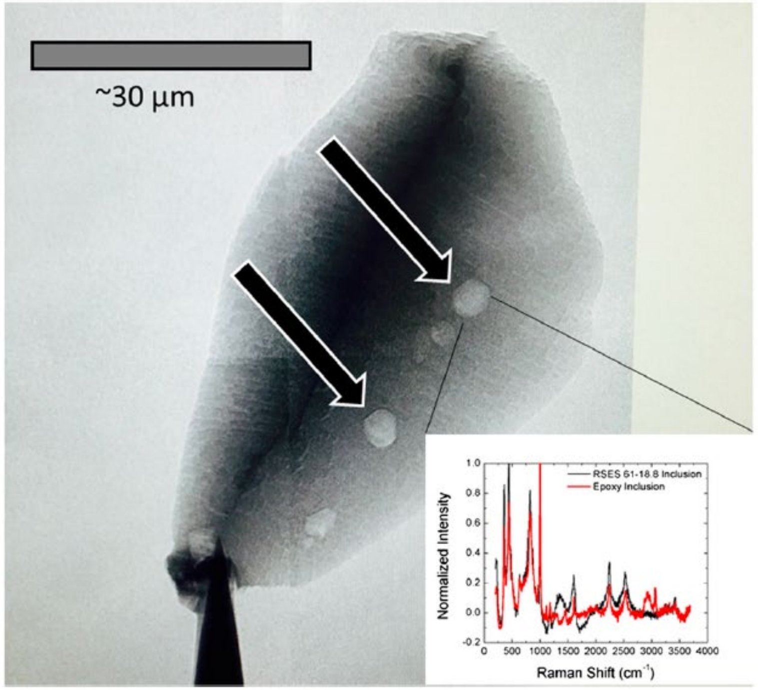


Fig. 1. Transmission X-ray image of RSES 61-18.8 with graphite indicated. (*Inset*) Raman spectra for the top inclusion and for an epoxy "inclusion" from another investigated zircon. The broadened "D-band" at ~1,400 cm^{-1} indicates disordered graphite (39); C–H stretch bands at ~2,800–3,100 cm^{-1} (39) are observed in epoxy but not graphite.

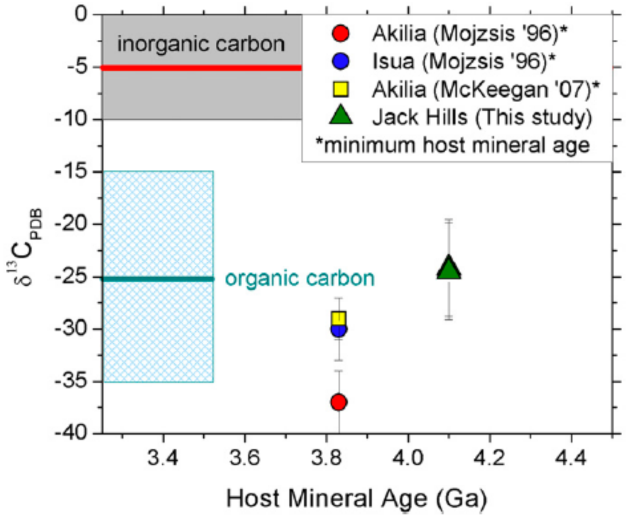


Fig. 2. $\delta^{13}C$ for Eoarchean–Hadean carbon samples measured via SIMS vs. host mineral age compared with inorganic and organic carbon (organic carbon values from ref. 13; inorganic from ref. 14).

Organic-walled microfossils in 3.2-billion-year-old shallow-marine siliciclastic deposits

Emmanuelle J. Javaux[1], Craig P. Marshall[2] & Andrey Bekker[3] Nature (2010)

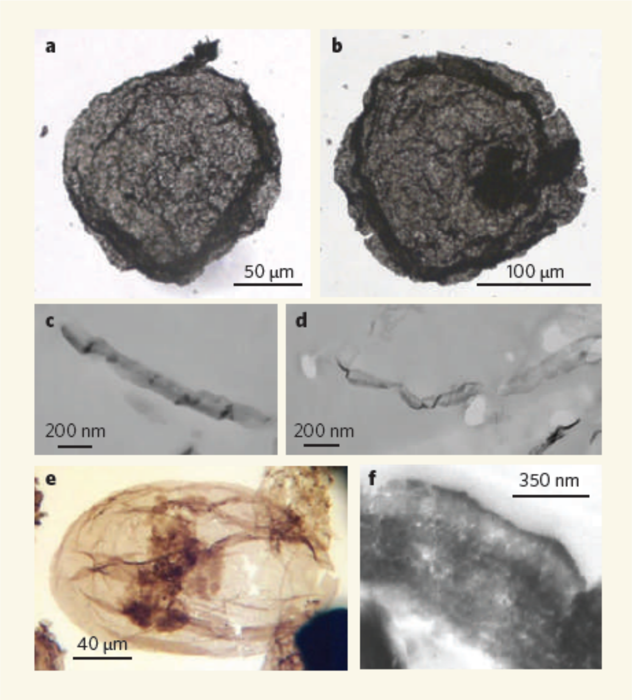
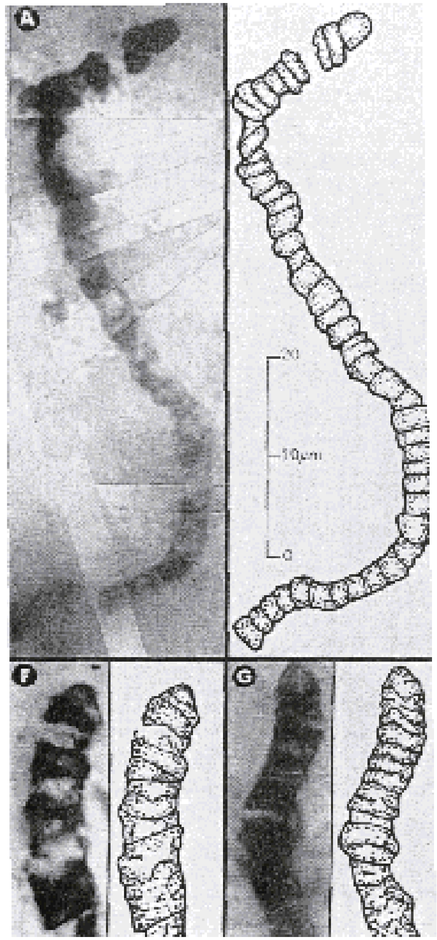
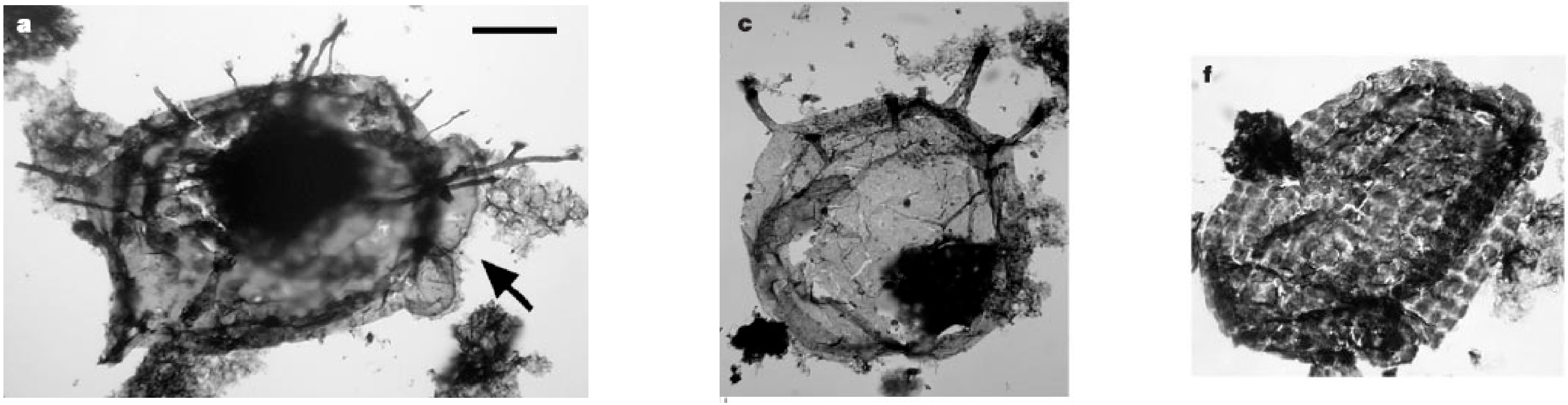


Figure 1 | Archaean and other acritarchs. **a**, **b**, Examples of Archaean acritarchs extracted from the ~3.2-billion-year-old Moodies Group, South Africa, by Javaux and her colleagues[2]. **c**, **d**, Transmission electron microscope images of the wall ultrastructure of these Archaean examples showing a simple monolayered organization. These specimens can be contrasted with **e**, a Mesoproterozoic acritarch (*Leiosphaeridia tenuissima*) from the ~1.4-billion-year-old Roper Group, northern Australia. **f**, The wall ultrastructure of the acritarch in **e**, showing a complex, multilayered organization with four distinct structural regions of differing thickness, indicating a eukaryotic origin. By contrast, the Archaean acritarchs' simple wall ultrastructure doesn't prove a eukaryotic origin, but nor does it preclude it. (Images from E. Javaux.)



3.5 billion-year old filamentous "Cyanobacteria"
Rocks in western Australia

Definitive Evidence for Eukaryotes from 1.5 BY-old Fossils?



Protist (?) microfossils, western Australia

From: Javaux et al., 2001, Nature.

Large colonial organisms with coordinated growth in oxygenated environments 2.1 Gyr ago

Abderrazak El Albani[1], Stefan Bengtson[2], Donald E. Canfield[3], Andrey Bekker[4], Roberto Macchiarelli[5,6],

Nature (2010)

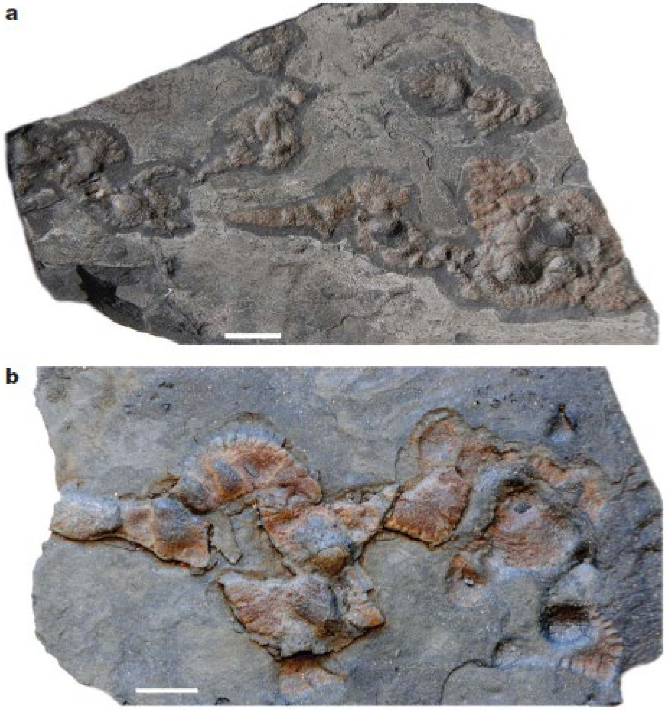


Figure 2 | Examples of black shale bedding surfaces. **a**, **b**, Bearing macrofossils in colony form from the FB2 level. Scale bars, 1.0 cm.

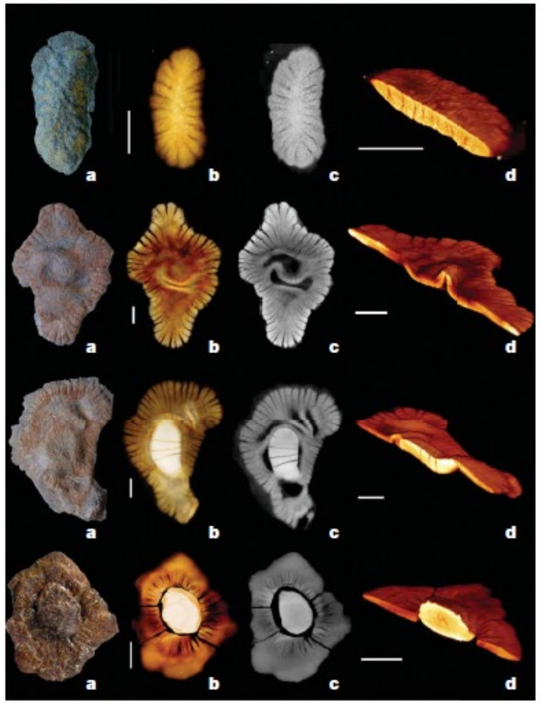
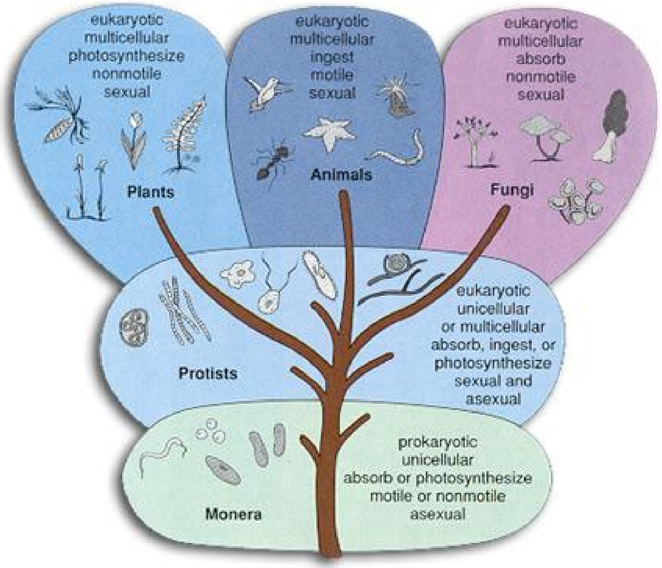


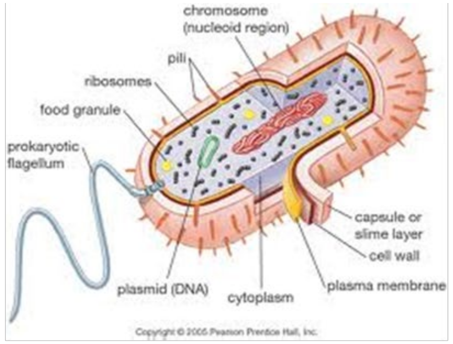
Figure 4 | Micro-CT-based reconstructions and virtual sections of four specimens from the FB2 macrofossil record of Gabon. Samples show a disparity of forms based on: external size and shape characteristics; peripheral radial microfabric (missing in view **d**); patterns of topographic thickness distribution; general inner structural organization, including occurrence of folds (seen in views **b** and **c**) and of a nodular pyrite concretion in the central part of the fossil (absent in views **a** and **b**). **a**, Original specimen. **b**, Volume rendering in semi-transparency. **c**, Transverse (axial) two-dimensional section. **d**, Longitudinal section running close to the estimated central part of the specimen. Scale bars, 5 mm. Specimens from top to bottom: G-FB2-f-mst1.1, G-FB2-f-mst2.1, G-FB2-f-mst3.1, G-FB2-f-mst4.1.

Ancient rocks from Gabon suggest an early origin of multicellularity.

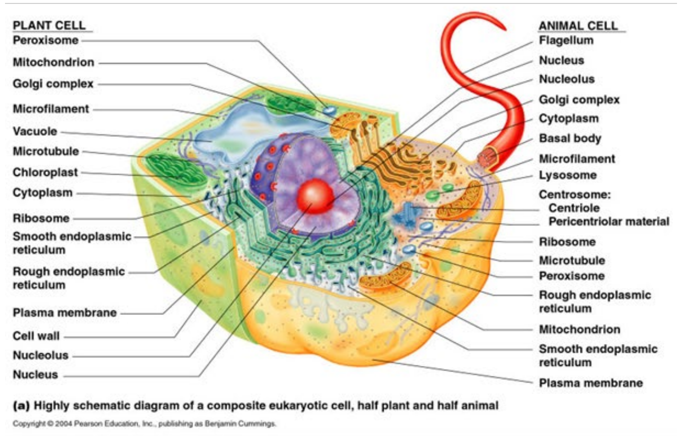
The Old Views



The Five Kingdoms of Life (1950s)



Prokaryote

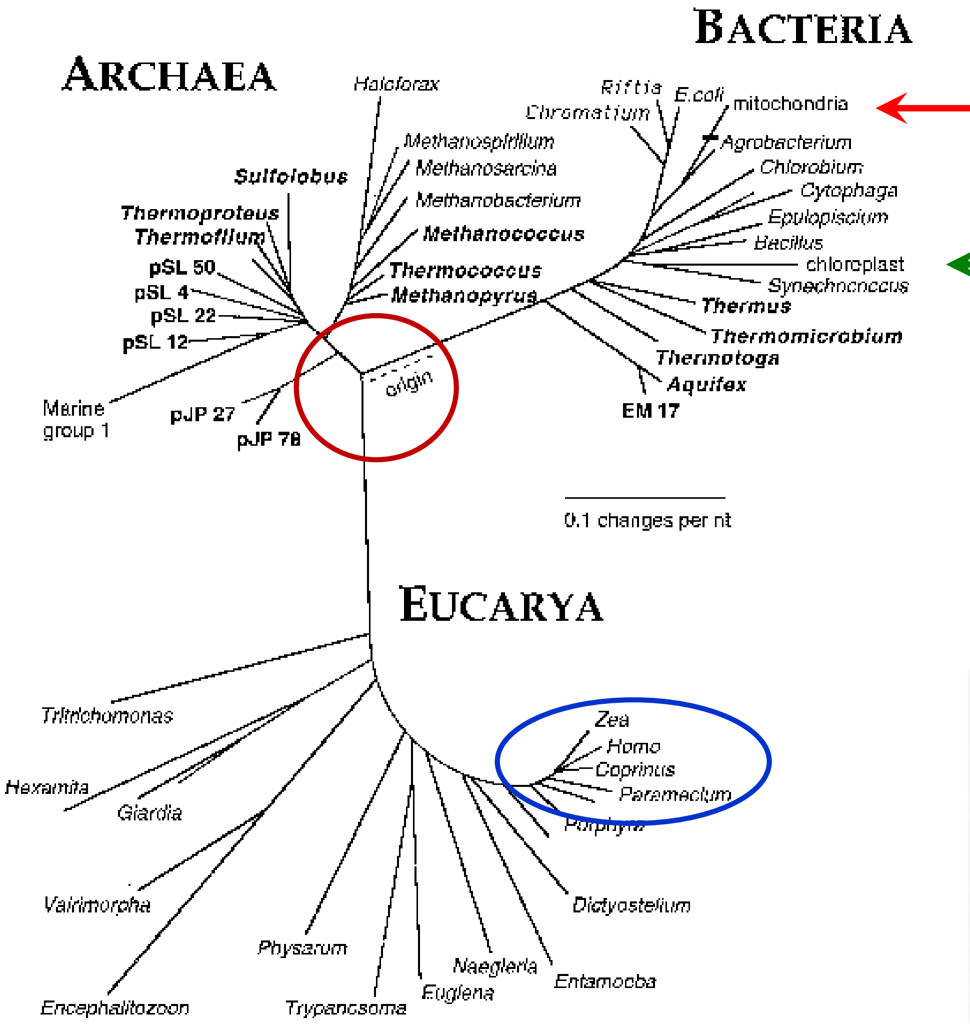


(a) Highly schematic diagram of a composite eukaryotic cell, half plant and half animal

Eukaryote

1977 – Using sequences of ribosomal RNA, Woese and Fox discover that prokaryotes consist of two deeply divergent clades, as different from each other as from eukaryotes.

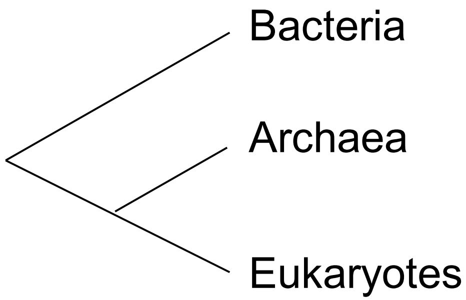
- Although life has classically been divided into eukaryotes and prokaryotes, molecular analyses indicate that these are not meaningful phylogenetic labels.
- The three-domains hypothesis has given way to a two-domains view, with the two prokaryotic groups, bacteria and archaea, appearing on opposite sides of the root of the Tree of Life.
- Eukaryotes are the most recent newcomer, emerging from a member of the archaea.

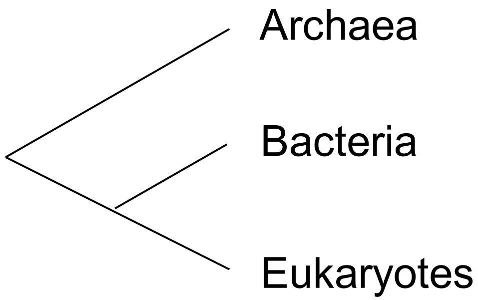


Carl Woese
University of Illinois

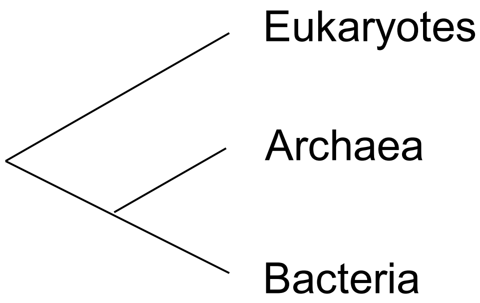
How Are The Three Major Domains Of Life Related?

- Are eukaryotes more closely related to the archaea or the bacteria?

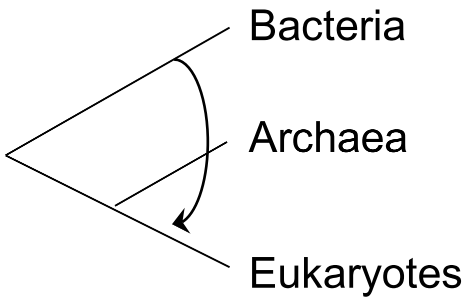




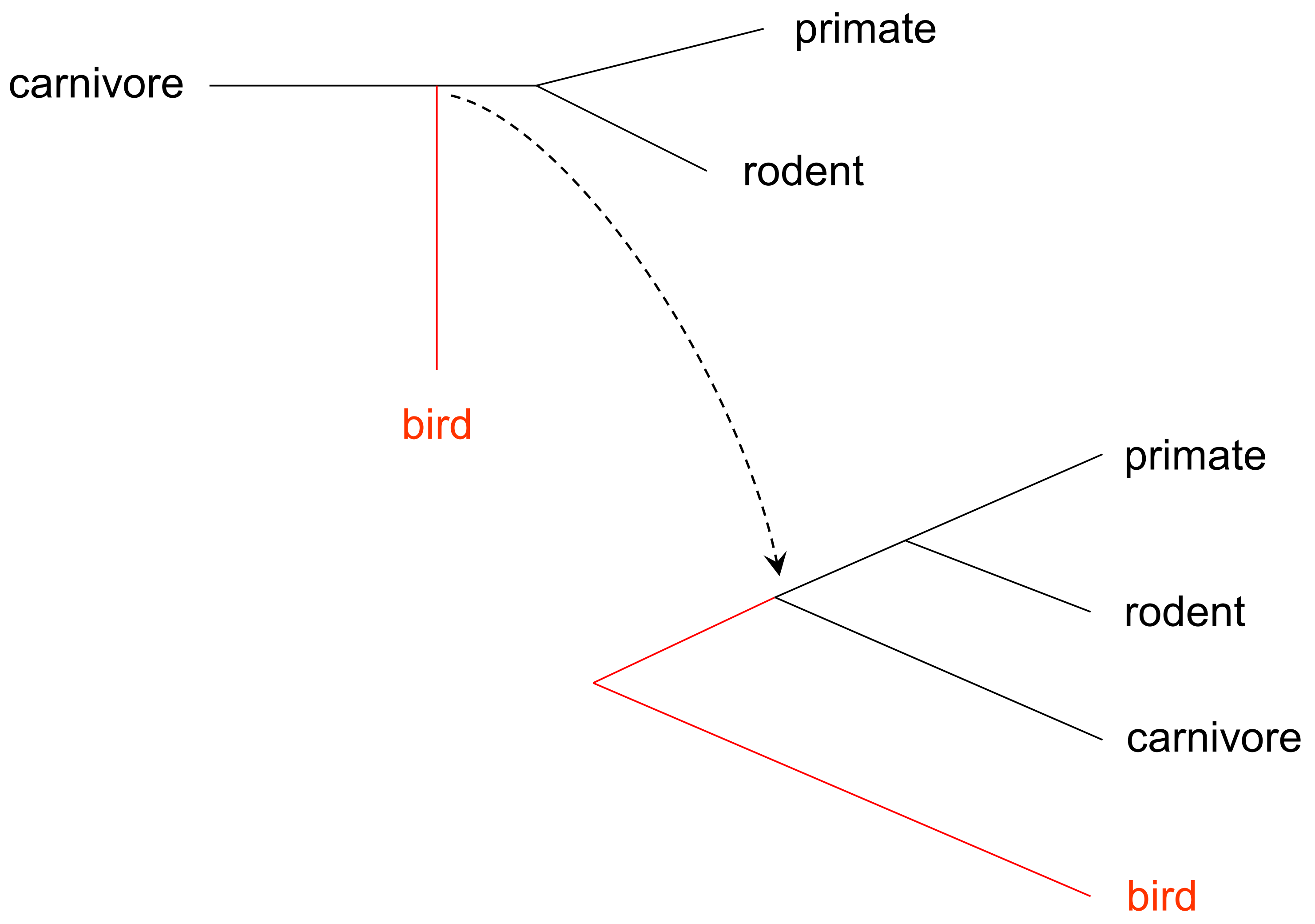
- Are prokaryotes derived, simplified eukaryotes or a monophyletic sister group?



- Are eukaryotes fully distinct from both prokaryotic clades, or has there been gene transfer from one or both prokaryotic groups?



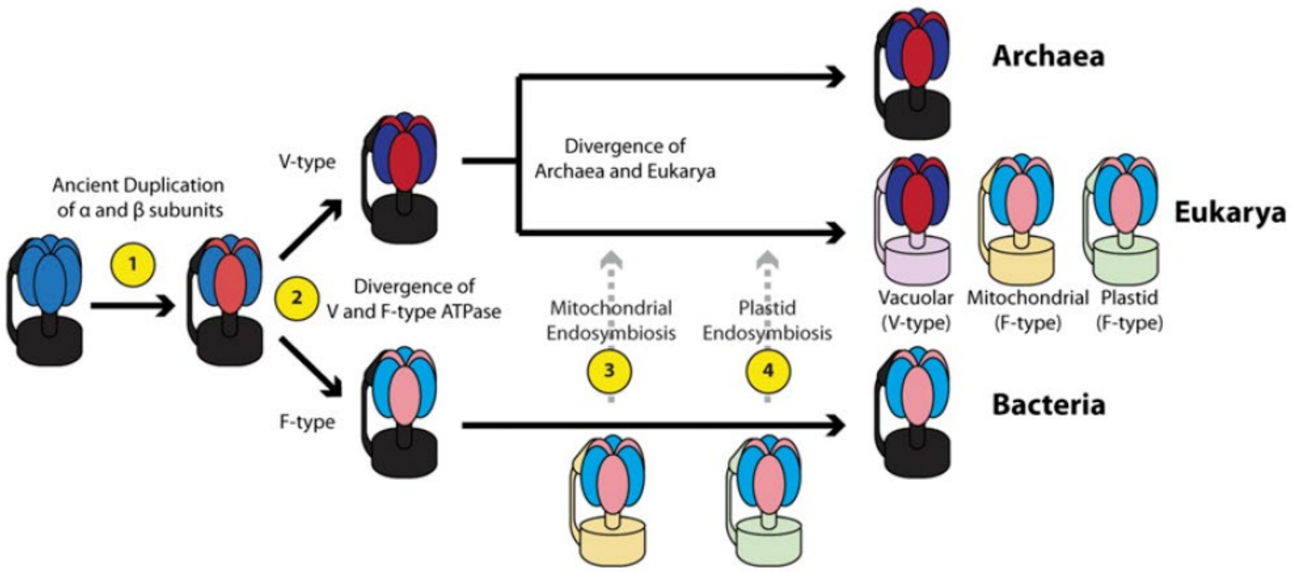
Rooting a Phylogenetic Tree with an Outgroup



Rooting the Tree of Life with Duplicate Genes

- The reciprocal rooting of a phylogenetic tree using a pair of ancient duplicate genes.
- If the duplication event occurred prior to the divergence of species, identical topologies are expected for each gene.
- The connecting branch reflects the divergence of the two copies within the ancestral species.

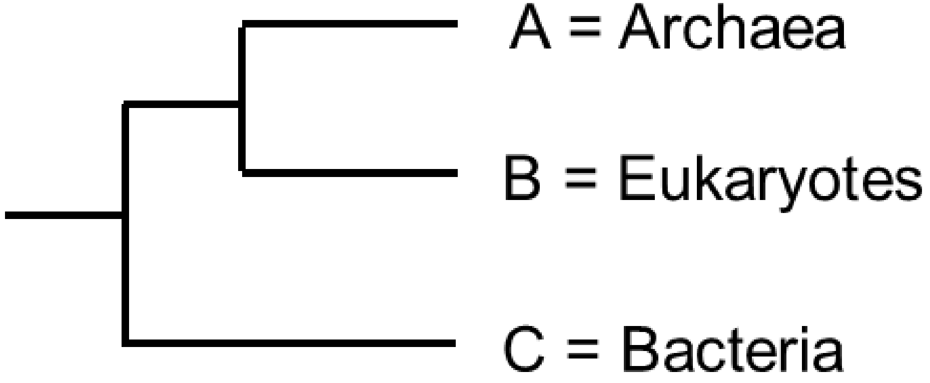
Reciprocal Rooting Often Supports the Grouping of Archaea and Eukaryotes



Duplication of proteins in the catalytic subunit of ATP synthase predates LUCA

(Shih and Matzke, 2013, PNAS)

- Other support from elongation factor duplicates, tRNA synthetases, GTP triphosphatases, etc.



A = Archaea

B = Eukaryotes

C = Bacteria

An archaeal origin of eukaryotes supports only two primary domains of life

Tom A. Williams[1], Peter G. Foster[2], Cymon J. Cox[3] & T. Martin Embley[1]

- Eukaryotes are derived Archaea.

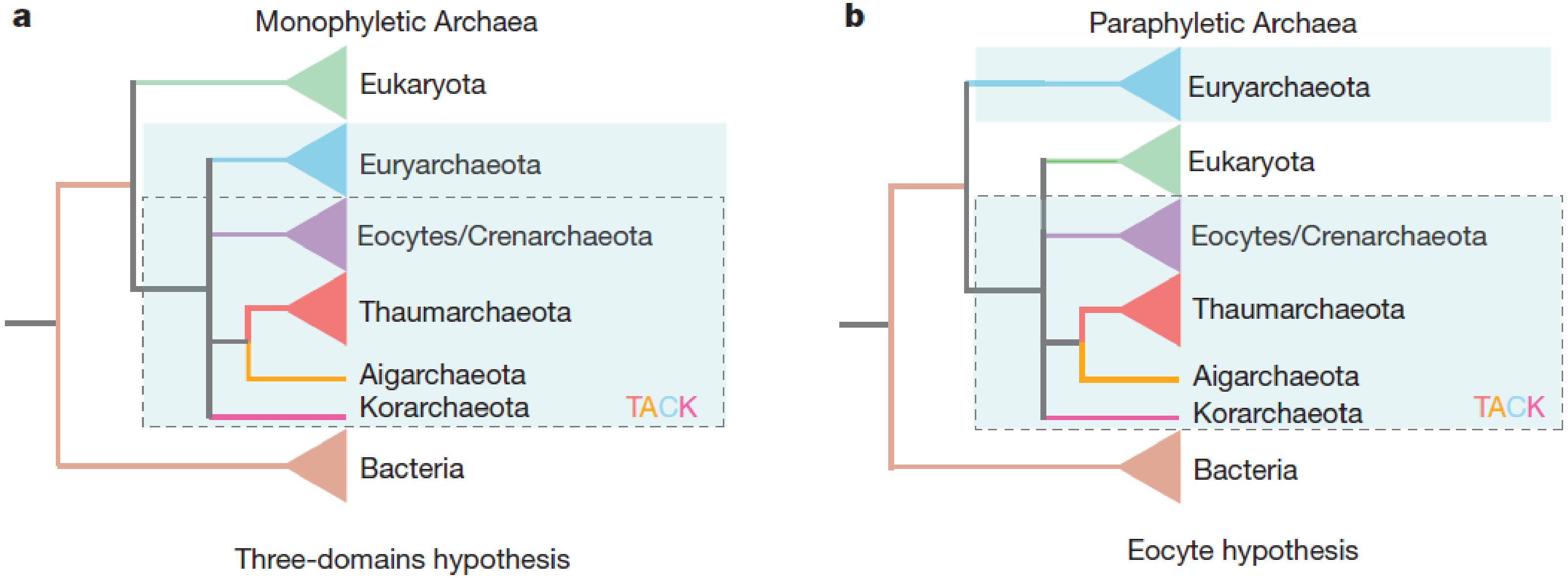
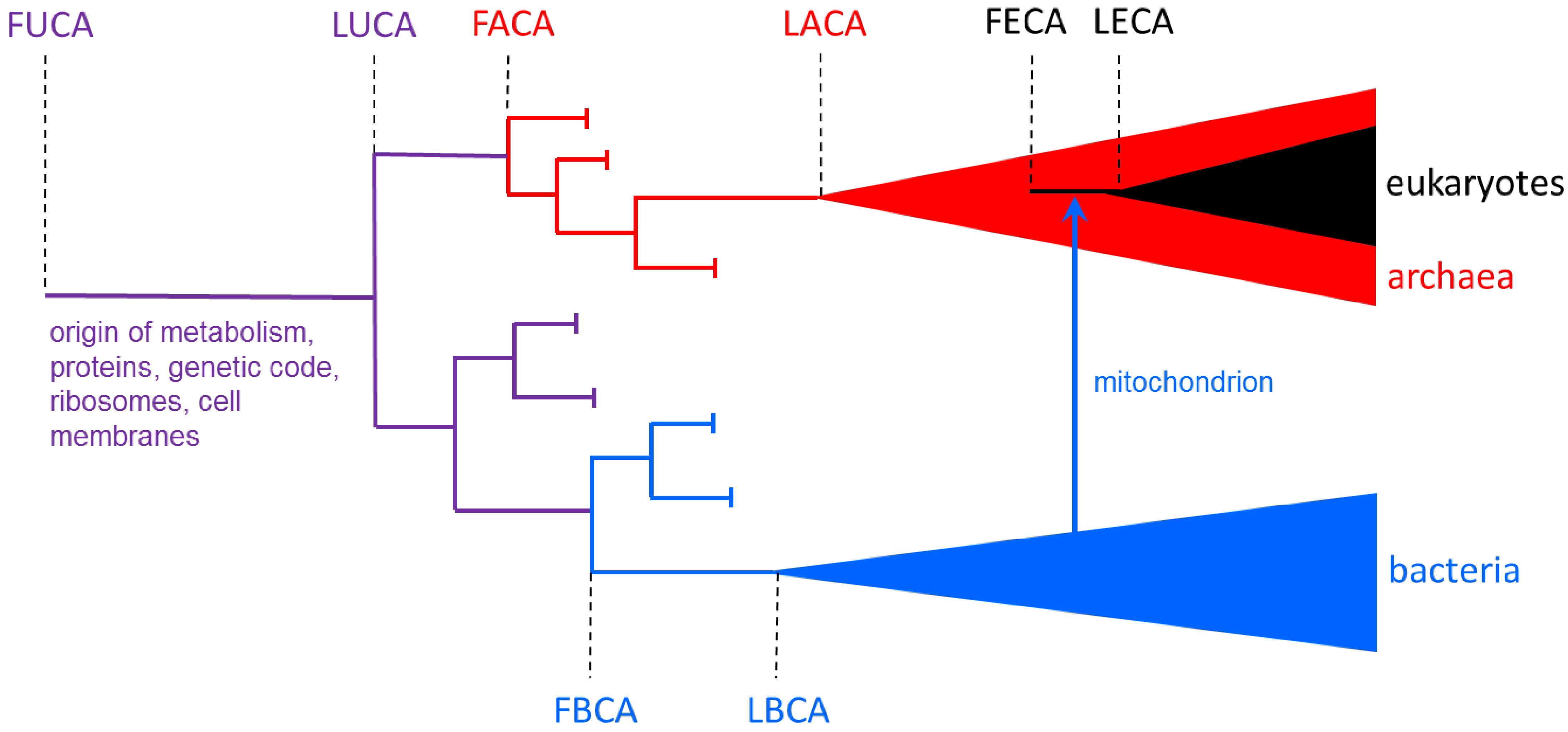


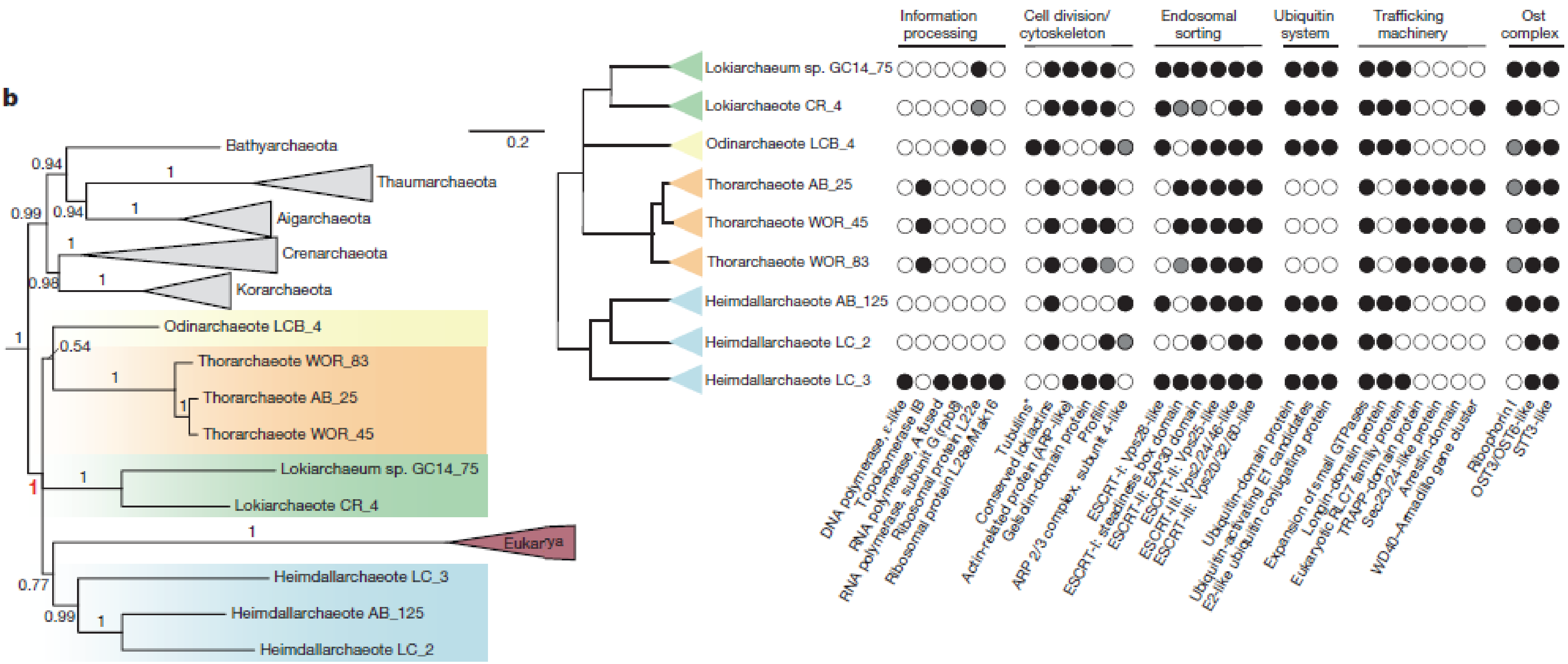
Figure 1 | Competing hypotheses for the origin of the eukaryotic host cell. **a**, The rooted three-domains tree[14] depicts cellular life divided into three major monophyletic groups or domains: the Bacteria, Archaea and Eukaryota—the latter representing the host lineage, sometimes also called the nuclear or nucleo-cytoplasmic lineage[5], that acquired the mitochondrial endosymbiont. In this tree the Archaea and Eukaryota are most closely related to each other because they share a common ancestor that is not shared with Bacteria. **b**, The rooted eocyte tree recovers the host-cell lineage nested within the Archaea as a sister group to the eocytes (which Woese *et al.*[14] called the Crenarchaeota); this implies that, on the basis of the small set of core genes, there are only two primary domains of life—the Bacteria and the Archaea. In its modern formulation shown here the eocyte hypothesis implies that the closest relative of the eukaryotic nuclear lineage is one, or all, of the TACK Archaea, which include newly discovered relatives of the eocytes/Crenarchaeota. Both trees have been traditionally rooted on the bacterial stem, consistent with some published analyses[5–8].

Williams et al. (2013, Nature)

Notational Summary



Signature Eukaryotic Genes in Our Archaeal Relatives (the Asgard Archaea)



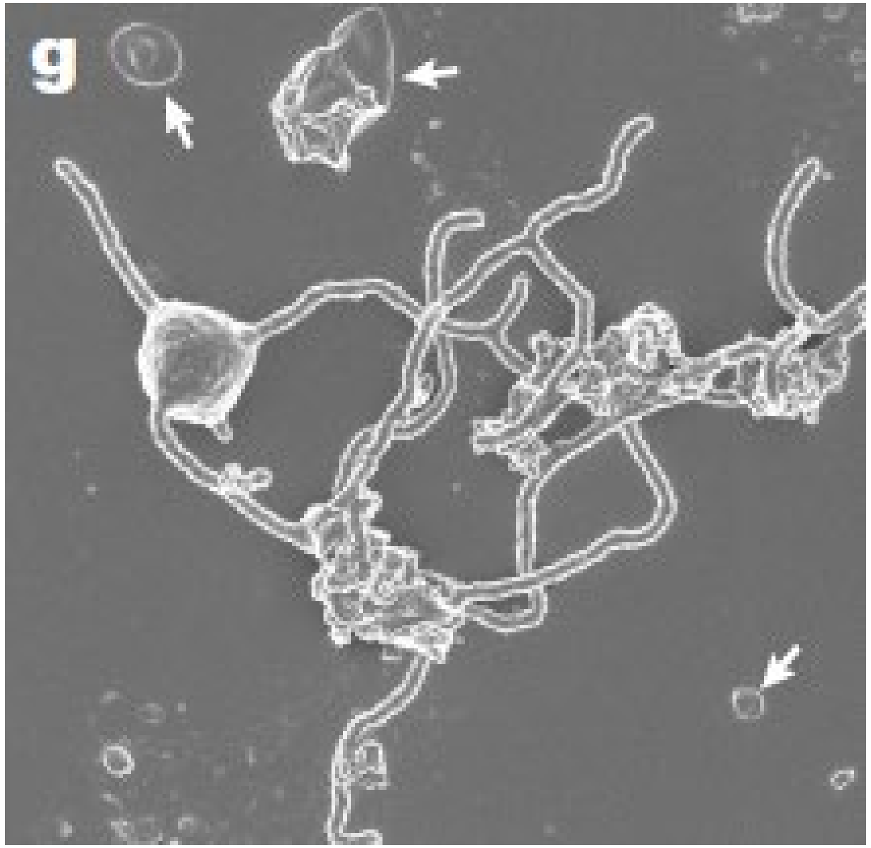
	Information processing						Cell division/ cytoskeleton						Endosomal sorting						Ubiquitin system			Trafficking machinery							Ost complex		
	DNA polymerase, ε-like	Topoisomerase IB	RNA polymerase, A fused	RNA polymerase, subunit G (rpb8)	Ribosomal protein L22e	Ribosomal protein L28e/Mak16	Tubulins*	Conserved lokiactins	Actin-related protein (ARP-like)	Gelsolin-domain protein	Profilin	ARP 2/3 complex, subunit 4-like	ESCRT-I: Vps28-like	ESCRT-I: steadiness box domain	ESCRT-II: EAP30 domain	ESCRT-II: Vps25-like	ESCRT-III: Vps2/24/46-like	ESCRT-III: Vps20/32/60-like	Ubiquitin-domain protein	Ubiquitin-activating E1 candidates	E2-like ubiquitin conjugating protein	Expansion of small GTPases	Longin-domain protein	Eukaryotic RLC7 family protein	TRAPP-domain protein	Sec23/24-like protein	Arrestin-domain protein	WD40–Armadillo gene cluster	Ribophorin I	OST3/OST6-like	STT3-like
Lokiarchaeum sp. GC14_75	○	○	○	○	●	○	○	●	●	●	●	○	●	●	●	●	●	●	●	●	●	●	●	●	○	○	○	○	●	●	●
Lokiarchaeote CR_4	○	○	○	○	◐	○	○	●	●	●	●	○	●	◐	◐	○	●	●	●	●	●	●	●	●	○	○	○	●	●	●	○
Odinarchaeote LCB_4	○	○	○	●	●	○	●	●	○	○	●	◐	●	○	●	●	●	●	●	●	●	●	●	●	○	○	○	○	◐	●	●
Thorarchaeote AB_25	○	●	○	○	○	○	○	●	○	●	●	○	○	●	●	●	●	●	○	○	○	●	○	●	●	●	●	●	◐	●	●
Thorarchaeote WOR_45	○	●	○	○	○	○	○	●	○	●	●	○	○	●	●	●	●	●	○	○	○	●	○	●	●	●	●	●	◐	●	●
Thorarchaeote WOR_83	○	●	○	○	○	○	○	●	○	●	◐	○	○	◐	●	●	●	●	○	○	○	●	○	●	●	●	●	●	◐	●	●
Heimdallarchaeote AB_125	○	○	○	○	○	○	○	●	○	○	○	●	●	○	●	●	●	●	●	●	●	●	●	●	○	○	○	○	●	●	●
Heimdallarchaeote LC_2	○	○	○	○	○	○	○	●	○	○	●	◐	○	○	●	○	●	●	●	●	●	●	●	○	○	○	○	○	○	●	●
Heimdallarchaeote LC_3	●	○	●	●	●	●	○	○	●	●	●	○	●	●	●	●	●	●	●	●	●	●	●	●	○	○	○	○	○	●	●

Zaremba-Niedzwiedzka et al., Nature, 2017

Isolation of an archaeon at the prokaryote–eukaryote interface

Imachi et al. (2020,Nature)

- Isolated from deep marine sediment.
- Anaerobic, methane-fed bioreactor for 5.5 years.
- Grows very slowly, and lives by degrading amino acids in syntrophy with other microbes.
- Has no internal eukaryote-like structure.



‘*Candidatus* Prometheoarchaeum syntrophicum’

One Hypothesis for How the Mitochondrion Might Have Arisen

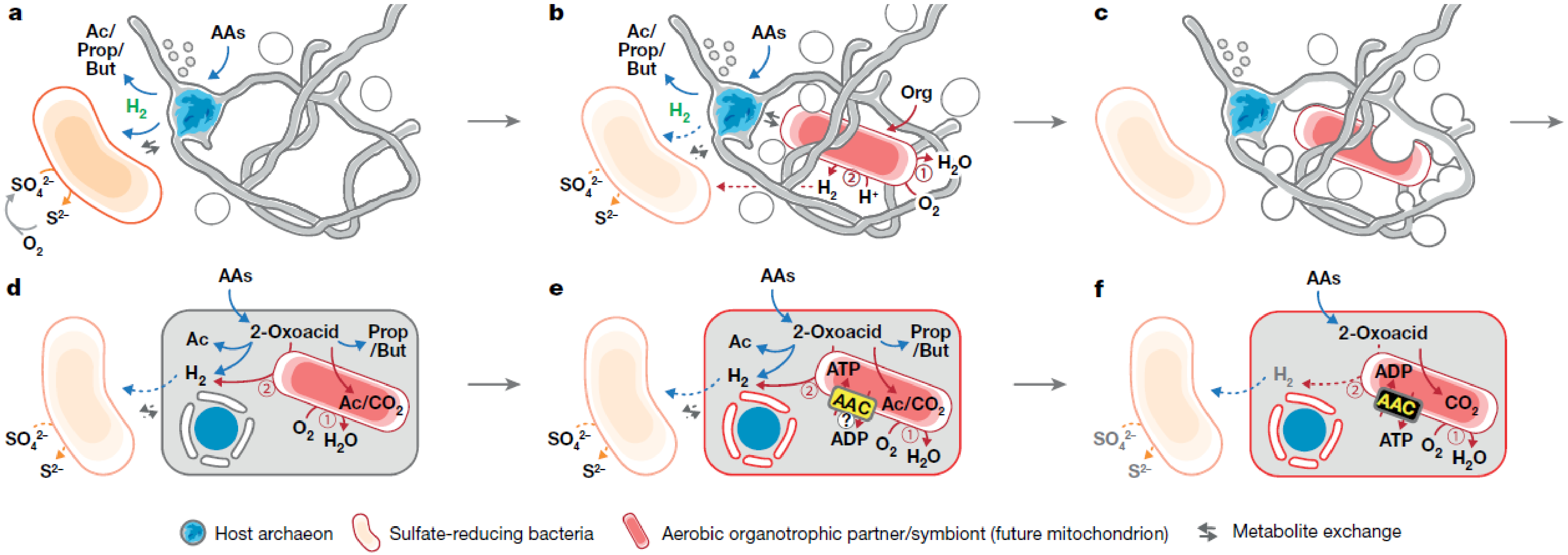
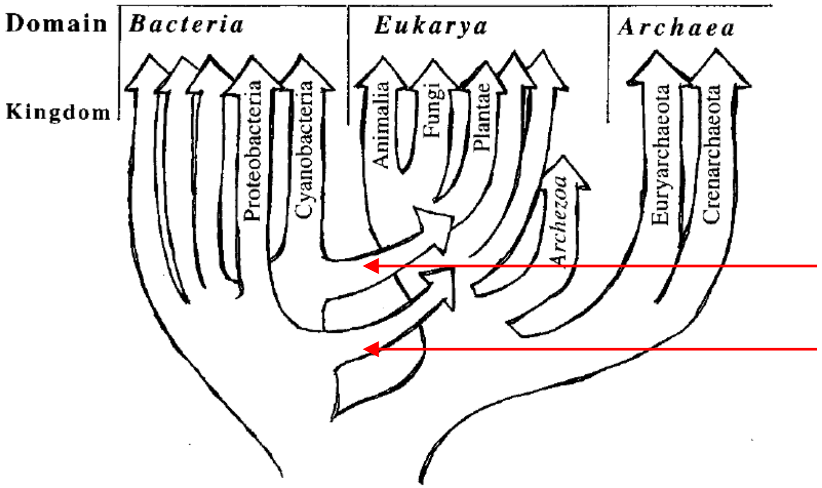


Fig. 5 | Proposed hypothetical model for eukaryogenesis. a, The syntrophic/fermentative host archaeon is suggested to degrade amino acids to short-chain fatty acids and H_2, possibly by interacting with H_2-scavenging (and indirectly O_2-scavenging) SRB (orange; see Supplementary Note 6). **b**, The host may have further interacted with a facultatively aerobic organotrophic partner that could scavenge toxic O_2 (the future mitochondrion; red). Continued interaction with SRB could have been beneficial but not necessarily essential; dotted arrows indicate the interaction; see Supplementary Note 7. **c**, Host external structures could have interacted (for example, mechanical or biological fusion[50]) with the aerobic partner to enhance physical interaction and further engulf the partner for simultaneous development of endosymbiosis and a primitive nucleoid-bounding membrane. **d**, After engulfment, the host and symbiont could have continued the interaction shown in **b** as a primitive type of endosymbiosis. **e**, Development of ADP/ATP carrier (AAC) by the endosymbiont (initial direction of ATP transport remains unclear; see Supplementary Note 9). **f**, Endogenization of partner symbiosis by the host through delegation of catabolism and ATP generation to the endosymbiont and establishment of a symbiont-to-host ATP channel.

Is Life a Web Rather Than a Tree?



chloroplast

mitochondrion

Fig. 2. The current consensus or standard model. Only a few of the "kingdoms" of the "domain" Bacteria are shown. Branching orders of several kingdoms within Bacteria and Eukarya remain in dispute. Mitochondrial and chloroplast endosymbioses are indicated by lower and upper diagonal arrows, respectively. Archezoa, as a subkingdom composed of primitively amitochondriate protists, may be extinct. For SSU rRNA trees with much more detail, see (*5*).

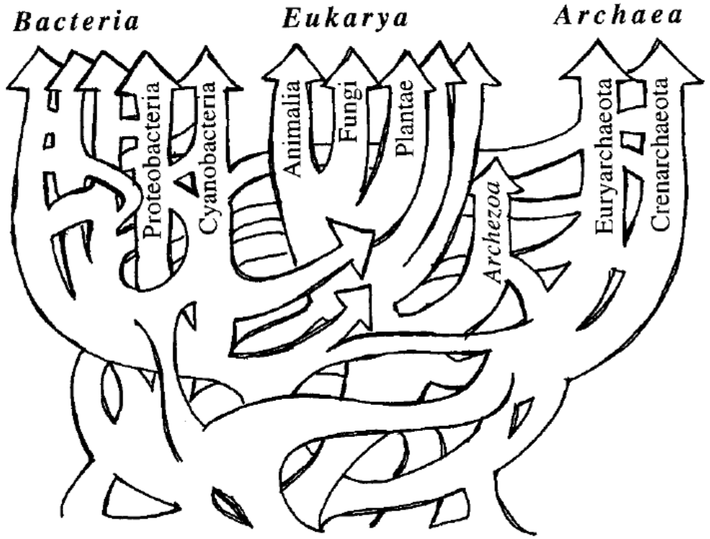


Fig. 3. A reticulated tree, or net, which might more appropriately represent life's history. Martin (*16*), in a review covering many of the same topics as this one, has presented some striking colored representations of such patterns.

From: W. F. Doolittle, 1999, Science.

The Chimeric Nuclear Genome of Eukaryotes: different trees for different genes.

- Eukaryotic genes involved in information processing (e.g., transcription and translation) tend to be more similar to orthologous genes of archaea.

- Eukaryotic genes with house-keeping functions (e.g., metabolism) tend to be more similar to orthologous genes of bacteria.

References: Brown and Doolittle 1997; Rivera et al. 1998; Leipe et al. 1999; Brown et al. 2001; Horiike et al. 2001.

Use of Different Phospholipids in the Major Lineages

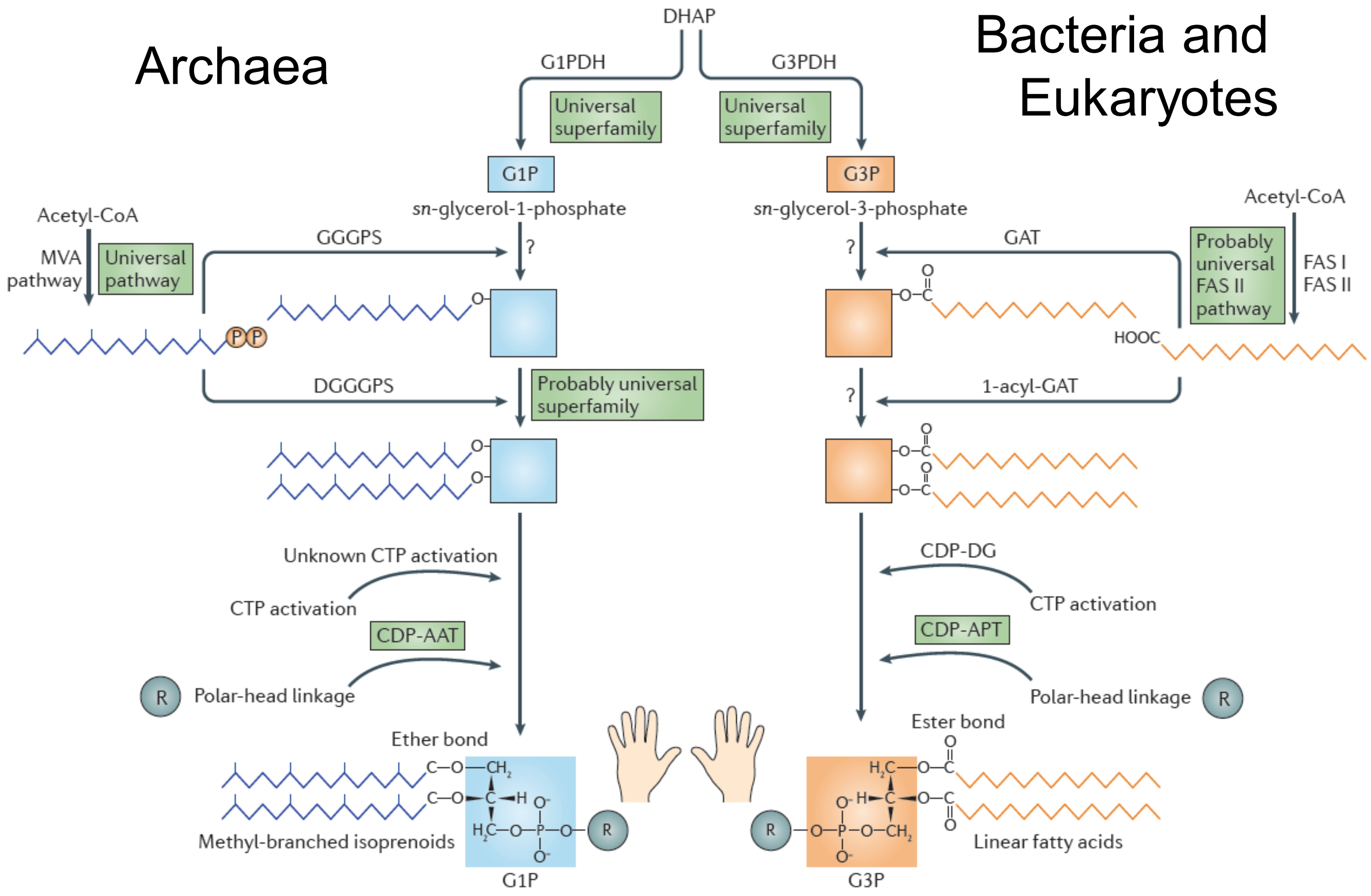
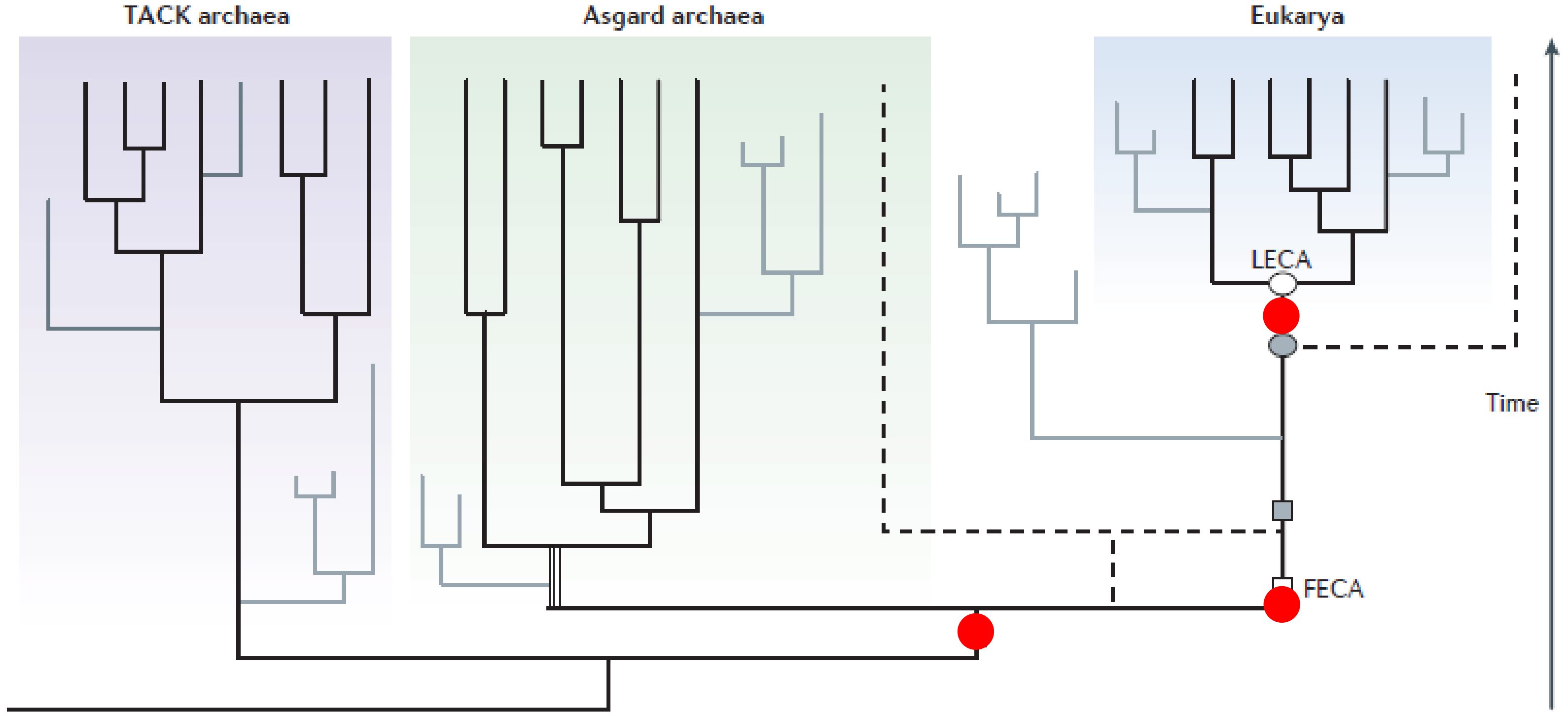
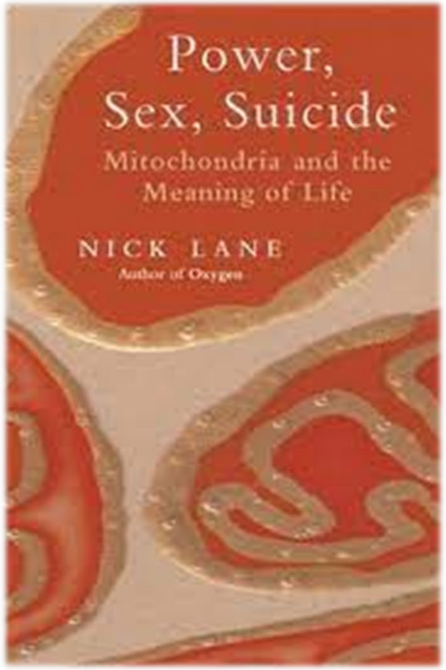


Figure 1 | **Phospholipid biosynthesis pathways in archaea, bacteria and eukaryotes.** Phospholipid components and the enzymes that synthesize them are different in modern archaea versus modern bacteria and eukaryotes. For some steps in the pathways, there is phylogenomic evidence either supporting the hypothesis that homologous enzymes carried out a particular step in the cenancestor (universal proteins or pathways) or indicating that the presence of the relevant enzymes in the cenancestor cannot be excluded (probably universal proteins or pathways). Cytidine diphosphate-alcohol archaetidyltransferase (CDP-AAT) and CDP-alcohol phosphatidyltransferase (CDP-APT) are homologous in the two pathways. Polar head radicals can be serine, ethanolamine or glycerol, among others. A question mark indicates that information is unknown. CDP-DG, CDP-diacylglycerol synthase; CTP, cytidine triphosphate; DGGGPS, digeranylgeranylglyceryl phosphate synthase; DHAP, dihydroxyacetone phosphate; G1P, *sn*-glycerol-1-phosphate; G1PDH, G1P dehydrogenase; G3P, *sn*-glycerol-3-phosphate; G3PDH, G3P dehydrogenase; GAT, G1P acyltransferase; GGGPS, geranylgeranylglyceryl phosphate synthase; FAS, fatty acid synthesis; MVA, mevalonate.

Lombard et al. 2012,
Nature Rev. Microbiol.

Did the Mitochondrion Play an Essential, Causal Role in the Emergence of Eukaryotes?





- Did the mitochondrion establish before or after other eukaryotic-specific innovations?
- Did the mitochondrion endow LECA with extraordinary bioenergetic powers?
- Was the primordial mitochondrion a mutualist or an energy parasite?

Eme et al. 2017

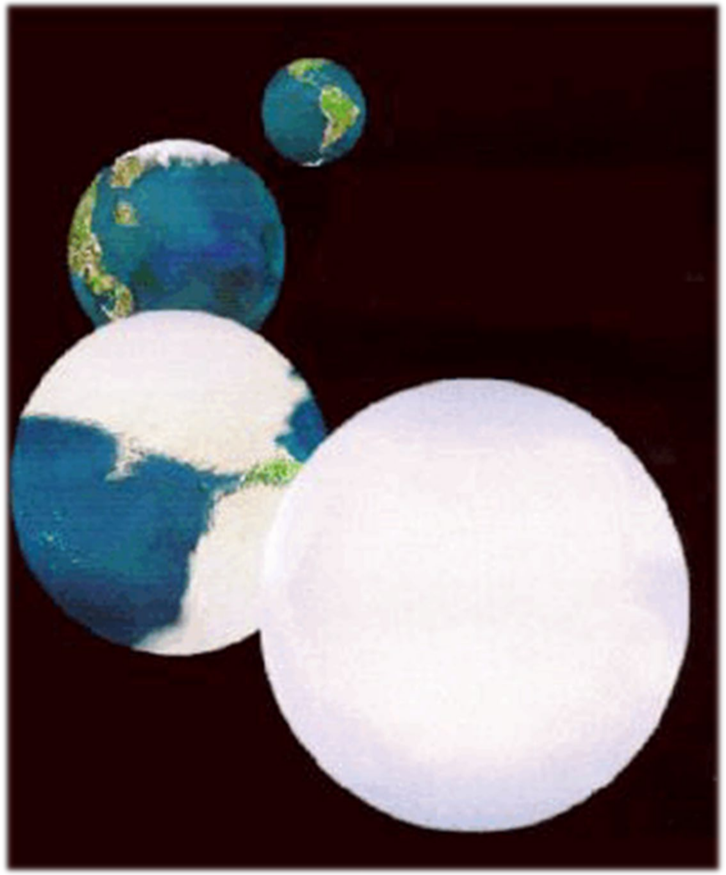
The Emergence of Eukaryotes Marked a Dramatic Epoch of Morphological Diversification in Earth's History

- The stem eukaryote was a complex cell, with dozens of eukaryote-specific changes having become established prior to LECA.
  - LECA (last eukaryotic common ancestor) had a mitochondrion, a cytoskeleton, a nuclear membrane, and vesicle transport machinery.
  - Had linear chromosomes, a mechanism for preventing erosion of chromosome ends during replication (telomeres), multiple origins of replication, a means for organized chromosomal inheritance (mitosis), and perhaps for gamete production and cell fusion (meiosis).
  - Contained intragenic spacers (introns) in a large number of protein-coding genes, and therefore harbored a spliceosome.
  - Harbored mobile genetic elements, both RNA-based retrotransposons and DNA-based transposons.
  - Massive increase in cell size?
- The order in which these features arose remains unknown, and many of them are difficult to explain with adaptive arguments.

The Snowball Earth Hypothesis

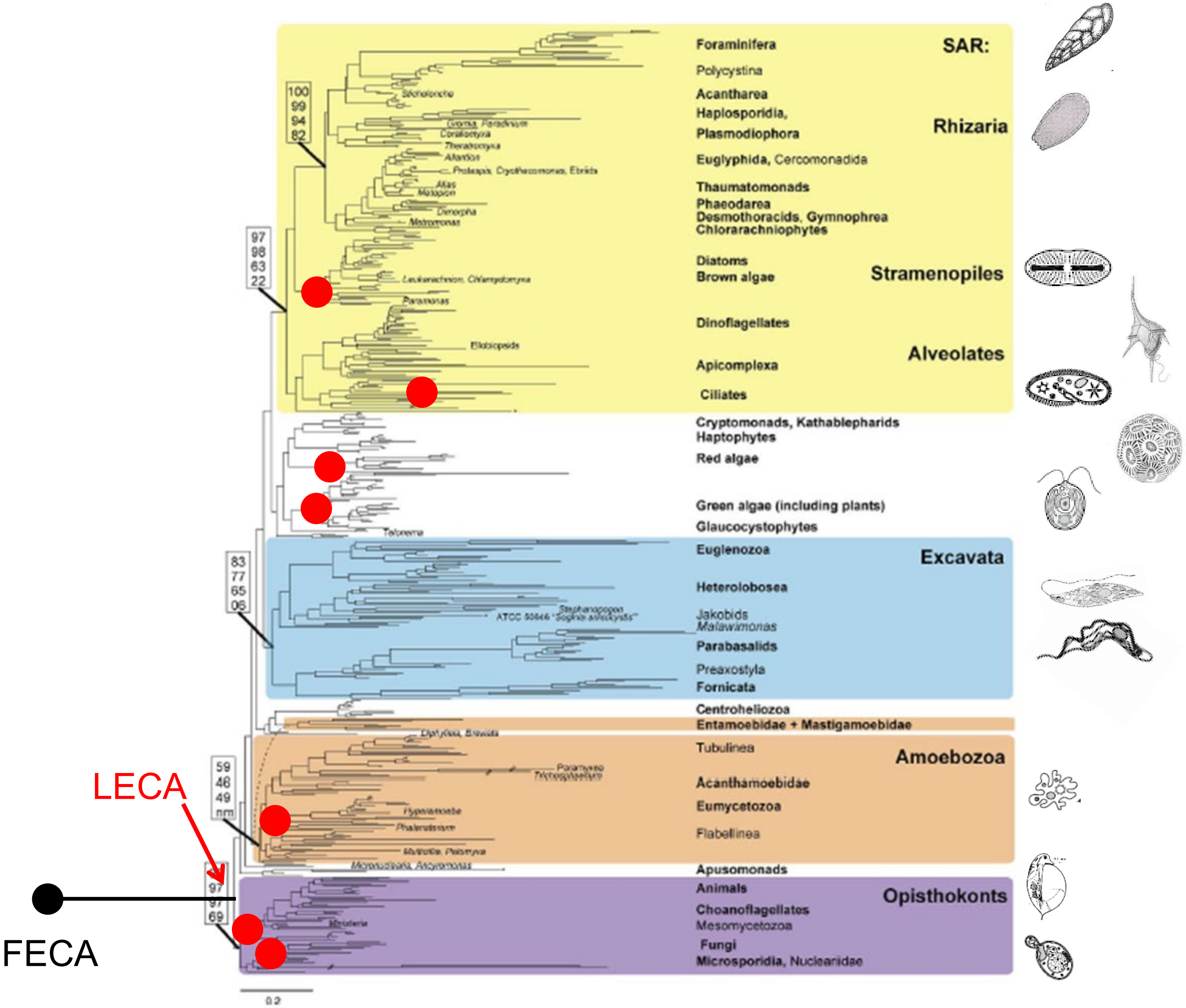
Two major episodes of global glaciation: ~2.4 and ~0.7 BYA.

- Mean surface temperatures approached -50ºC, and the oceans were frozen over with up to a kilometer of ice for ~35 MY.
- Periods of prolonged population bottlenecks – few habitats with significant sunlight would have been coincident with liquid water and oxygen.



References: Hoffman et al. (1998); Kirschvink et al. (2000); Hoffman and Schrag (2002).

Deep and Explosive Divergence Among the Major Eukaryotic Lineages

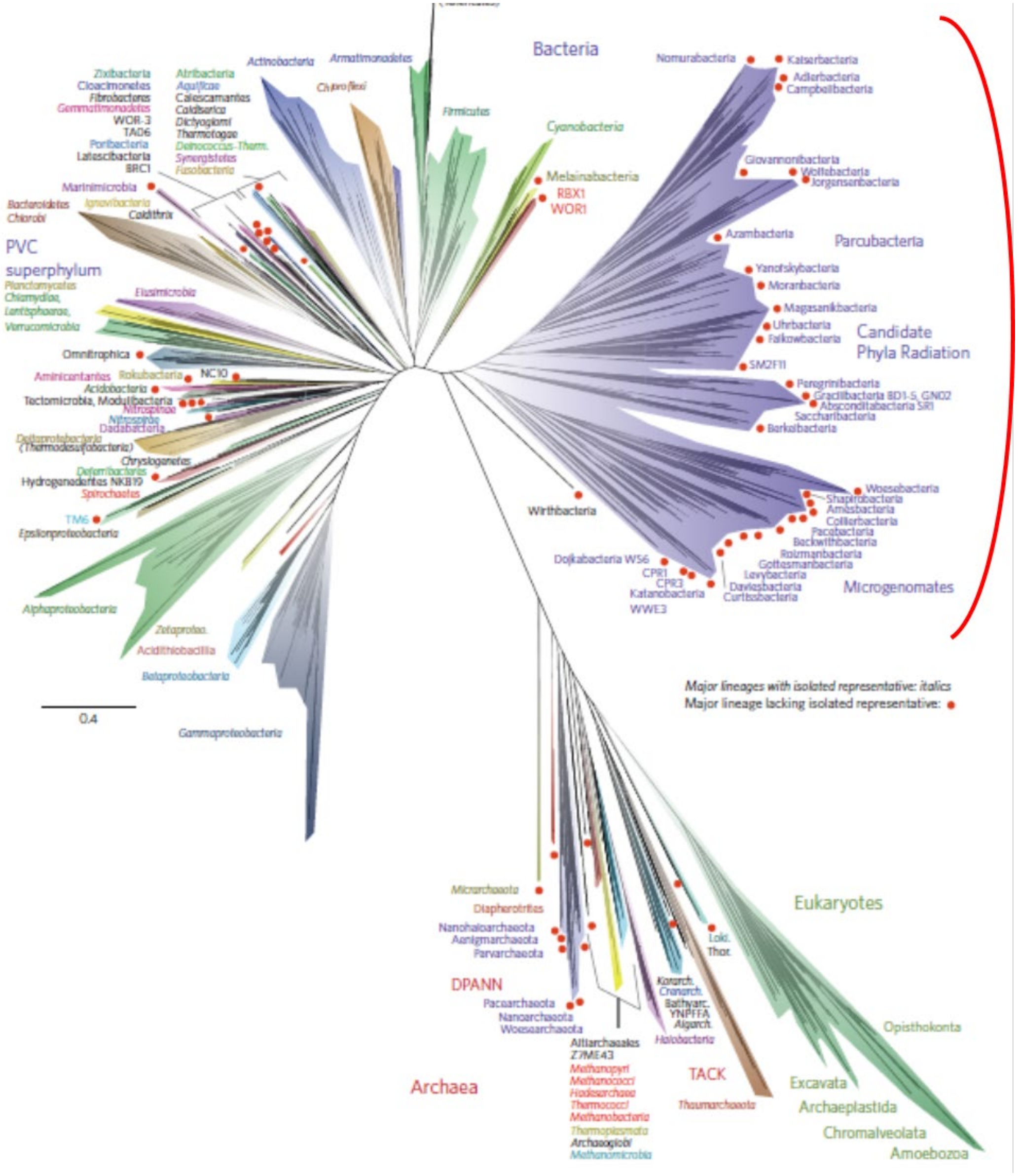


- Approximate phylogenetic tree for the major eukaryotic groups, using concatenated sequences of 29 genes.
- Branch lengths are proportional to amino-acid substitutions on the lineage and denote variation in rates of molecular evolution.

● Numerous Independent origins of multicellularity

From: Parfrey et al. (2010).

Many New Lineages Remain to be Discovered



From environmental sampling, including uncultured and unobserved organisms.

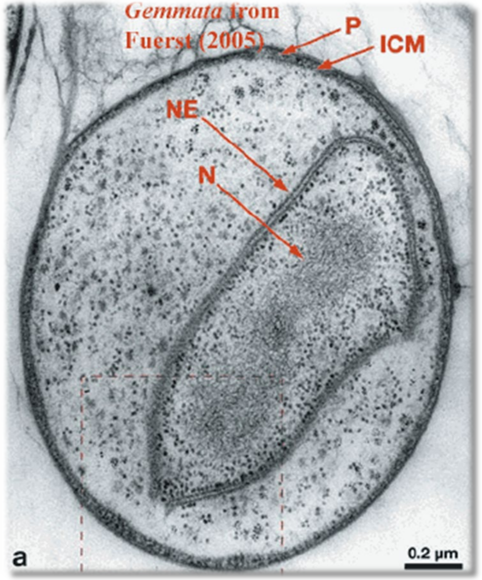
Hug et al., 2016, Nature Microbiol.

The Full Suite of Eukaryotic Innovations Need Not Have Arisen *De Novo*

Features that set eukaryotic genomes apart from those of prokaryotes, and their exceptions:

Presence of a nuclear membrane.	Internal membranes in the planctomycetes.
Organelles derived from endosymbionts.	Also present in the betaproteobacteria.
Cytoskeleton and vesicle transport machinery.	Tubulin-related proteins exist in archaea.
Introns in protein-coding genes, and a complex for excising them.	Rare self-splicing introns in prokaryotes, but almost never in coding DNA.
Multiple linear chromosomes capped with telomeres.	Single linear chromosomes in a few eubacteria.
Addition of poly(A) tails to all mRNAs.	Rare and nonessential polyadenylation of transcripts.
Expansion in gene number.	The largest prokaryotic genomes contain more genes than the smallest eukaryotic genomes.
Expansion of cell size and number.	A few prokaryotes have very large cell sizes (e.g., *Thiomargarita*), and several produce multiple cell types (e.g., cyanobacteria).

Members of the Bacterial Group Planctomycetes, and Broader Verrucomicrobia, Have Internal Membranes



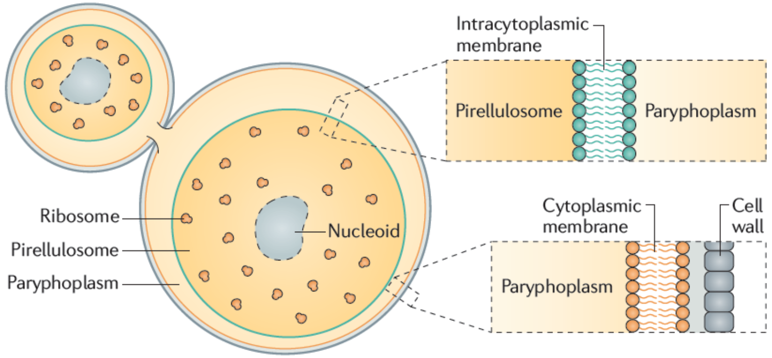
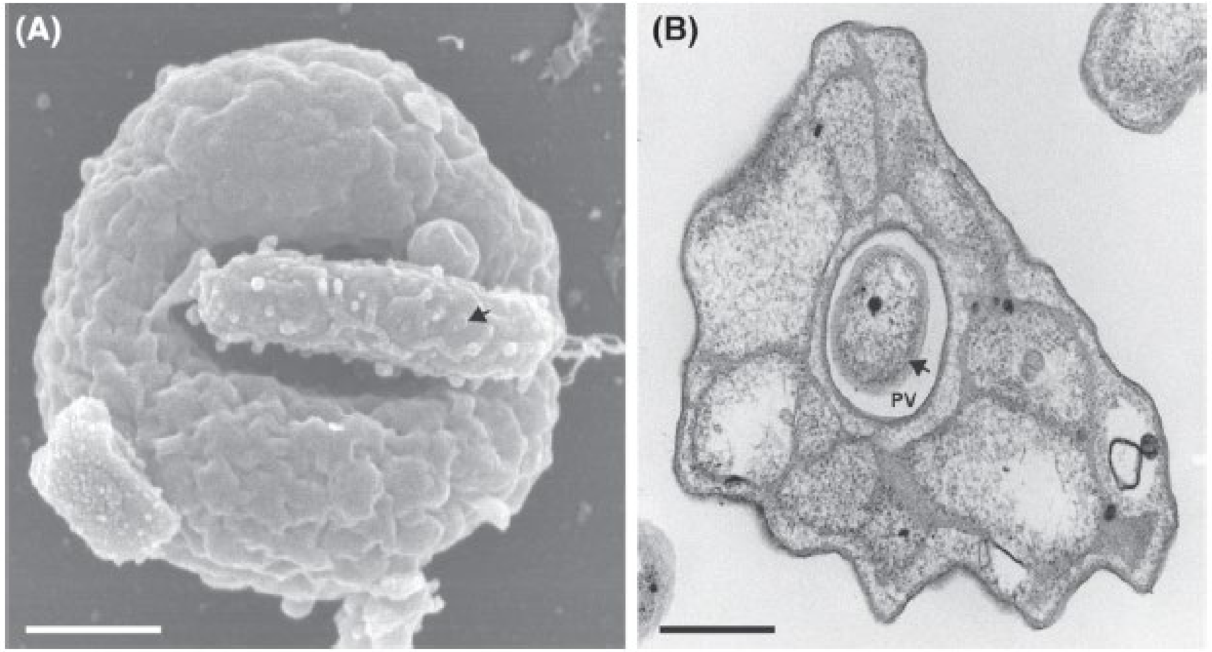


Figure 1 | **An idealized planctomycete in the process of cell division.** The unusual compartmentalized cell structure of the planctomycetes is shown, detailing the protein cell wall, the cytoplasmic membrane lipid bilayer and the intracytoplasmic membrane lipid bilayer in relation to the ribosome-free paryphoplasm and the ribosome-containing pirellulosome that contains condensed nucleoid DNA. The exact mechanism of bud formation and membrane transfer into the bud has not been determined in the simplest type of planctomycete shown here (but see REF. 72 for data relating to the more complex *Gemmata obscuriglobus*).

- Reproduce by a budding process.
- Capable of uptake from the external environment by endocytosis.
- Oxidize ammonia within a compartment called the anammoxosome.
- Some phylogenetic trees place the group at the base of bacteria in the tree of life.
- Other related groups (verrucomicrobia and chlamydiae) also have endomembranes.

A Marine Member of the Planctomycetes Capable of Phagocytosis



Trends in Microbiology

Figure 1. Phagocytosis-like Ingestion of Bacteria by '*Candidatus* Uab amorphum'. (A) Scanning electron micrograph showing bacterial cell (highlighted by arrowhead) in the process of being ingested by the organism. (B) Transmission electron micrograph showing interior of '*Ca.* Uab amorphum' with ingested cell (arrowhead) inside a phagosome-like vacuole (PV). Scale bars = 500 nm. Images kindly provided by Takashi Shiratori.

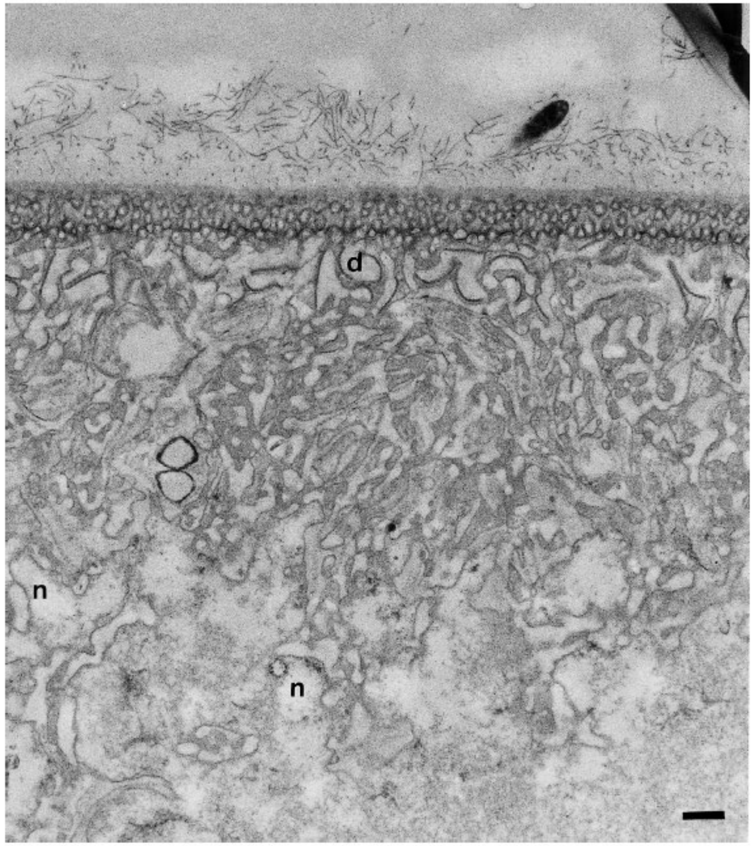
- A giant predatory bacterium, up to 10 microns in diameter.
- Engulfs and digests bacteria and pico-eukaryotes.
- Clearly indicates that establishment of a mitochondrion was not a precondition for the origin of internal membranes or of phagocytosis.

Shiratori et al. (2019, Nat. Comm.)

Epulopiscium – a giant symbiotic bacterium inhabiting triggerfish guts

Length on the order of 1 mm

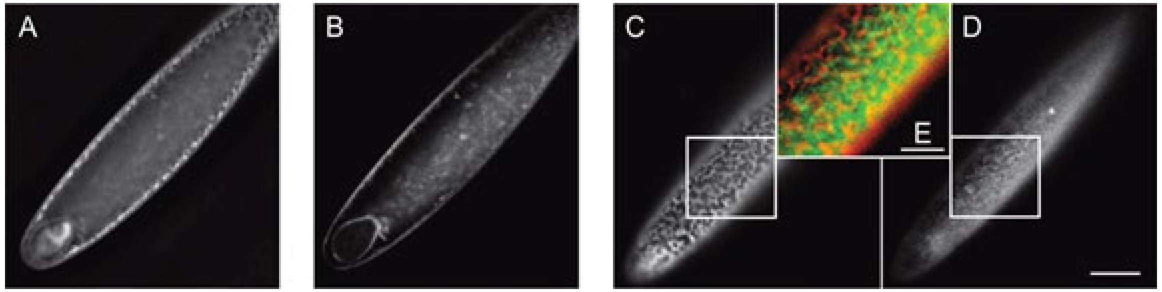
Fig. 6 Electron micrograph of a section of the cortex of another A morphotype. The cloud-like, faintly speckled shapes near the bottom are profiles of nucleoids (*n*). Above these is a labyrinth of interconnected spaces. Further up there is yet another labyrinth where the open spaces are lined with much denser membranes (*d*) than are those of the main labyrinth below. The outer border of the cortex appears to be formed by roughly circular profiles of irregularly stacked vesicles (bar 1.0 µm)



Stacked “vesicles”

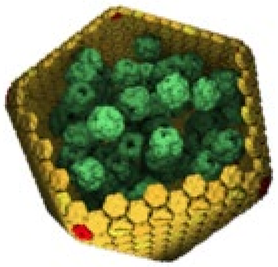
(Robinow and Angert 1998, Arch. Microbiol.)

Each cell contains ~10,000 copies of the genome.

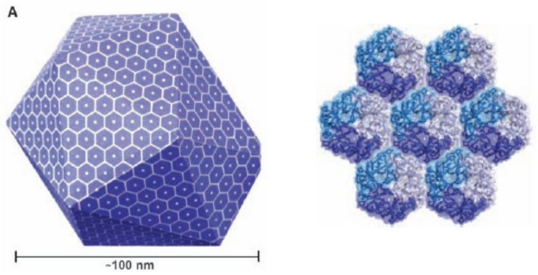


Other Intracellular “Organelles” in Bacteria:

The carboxysome sequesters the photosynthetic machinery in cyanobacteria.

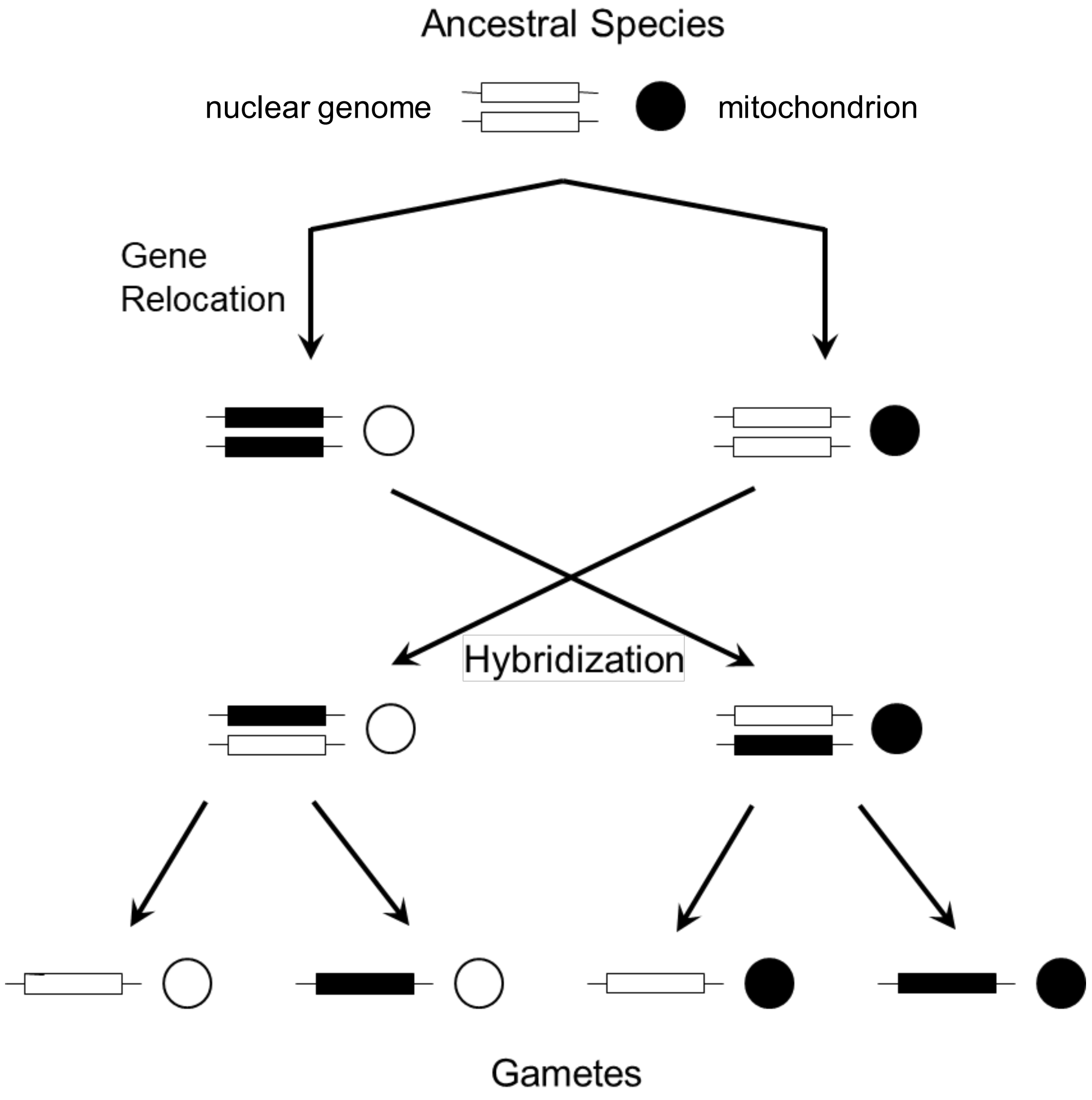


A microcompartment for ethanolamine metabolism in *E. coli* consists of hexameric subunits.



Magnetosomes are magnetite crystals enclosed by phospholipid membranes in magnetotactic bacteria.

The Potential for Passive, Rapid Phylogenetic Diversification Following the Establishment of Mitochondria



- Once established, LECA gave rise to an explosive radiation of the major eukaryotic groups on a relatively short time scale.
- This rapid episode of lineage isolation may have had little to do with ecological factors, instead being an inevitable consequence of two pre-LECA genomic upheavals:
  - the origin of the mitochondrion;
  - rampant nuclear gene duplication.
- Combined with the evolution sex, these changes would have led to the passive accumulation of gene relocations and reproductive isolation in ways that would have been inoperable in prior lineages of asexual prokaryotes.