

Reconciling an archaeal origin of eukaryotes with engulfment: a biologically plausible update of the Eocyte hypothesis

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Abstract

An archaeal origin of eukaryotes is often equated with the engulfment of the bacterial ancestor of mitochondria by an archaeon. Such an event is problematic in that it is not supported by archaeal cell biology. We show that placing phylogenetic results within a stem-and-crown framework eliminates such incompatibilities, and that an archaeal origin for eukaryotes (as suggested from recent phylogenies) can be uncontroversially reconciled with phagocytosis as the mechanism for engulfment of the mitochondrial ancestor. This is significant because it eliminates a perceived problem with eukaryote origins: that an archaeal origin of eukaryotes (as under the Eocyte hypothesis) cannot be reconciled with existing cell biological mechanisms through which bacteria may take up residence inside eukaryote cells.

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1. Introduction

A range of models for the origin of the eukaryote cell have been proposed on phylogenetic, genomic and cell biological grounds (reviewed in Embley and Martin, 2006; Martin et al., 2001; Poole and Penny, 2007b; Gribaldo et al., 2010). There is a general agreement that the ancestor of mitochondria¹ was an α -proteobacterium, contributing many (though probably not all—(Esser et al., 2004, 2007; Lester et al., 2006)) genes of bacterial origin to the eukaryote nuclear genetic complement. It is likewise beyond doubt that the mitochondrion was one of many features present in the Last Eukaryotic Common Ancestor (LECA) from which modern eukaryote diversity has

derived. This also appears to be the case for key parts of the machinery for phagocytosis—cell engulfment (Yutin et al., 2009). As summarised in Table 1, the emerging consensus from a range of studies is that the LECA was essentially a fully-fledged eukaryote cell.

Broad agreement on the timing and specific evolutionary origin of mitochondria stands in stark contrast to disagreement concerning the nature of the other partner in this endosymbiosis: the host. Two main views have been expounded in the literature. One is that the host was an archaeon, the other that the host was a protoeukaryote capable of cell engulfment (discussed in Embley and Martin, 2006; Martin et al., 2001; Poole and Penny, 2007b). These seemingly opposing views have been the source of extensive recent debate on two levels: one phylogenetic, one cell biological (Davidov and Jurkevitch, 2007, 2009; Poole and Penny, 2007a,b,c, Hartman and Fedorov, 2002; Kurland et al., 2006; Martin and Koonin, 2006; Lopez-Garcia and Moreira, 2006; Gribaldo et al., 2010).

Phylogenetically, eukaryotes and archaea might each be monophyletic, as suggested by the bacterial rooting of the tree of life (Gogarten et al., 1989; Iwabe et al., 1989; Woese et al., 1990), meaning that eukaryotes and archaea are sister groups

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¹ Mitochondria and related organelles—mitosomes and hydrogenosomes—all derive from a common ancestor (reviewed in van der Giezen and Tovar, 2005). For brevity, we will refer to mitochondria throughout, but references we make to the mitochondrial ancestor will assume that we are talking about the ancestor of all three, irrespective of important differences in their metabolic repertoire.

Table 1
Features of contemporary eukaryote cells proposed to be present in the Last Eukaryotic Common Ancestor (LECA).

Feature	References
Mitochondrion	Embley and Martin, 2006; van der Giezen and Tovar, 2005
Phagocytosis	Cavalier-Smith, 2002b; Jékely, 2003, 2007a; Yutin et al., 2009
Nucleus and nuclear pore complex	Baptiste et al., 2005; Devos et al., 2004, 2006; Mans et al., 2004; Neumann et al., 2010
Endomembrane system	Dacks et al., 2003; Dacks and Field, 2007; Field and Dacks, 2009; Jékely, 2003, 2007a; Neumann et al., 2010
Mitosis and meiosis	Cavalier-Smith, 2002a; Ramesh et al., 2005; Egel and Penny, 2008
Introns and spliceosomal apparatus	Collins and Penny, 2005; Jeffares et al., 2006; Roy and Gilbert, 2005, 2006; Roy and Irimia, 2009
Linear chromosomes and telomerase	Nakamura and Cech, 1998
RNA processing	Collins et al., 2009; Gardner et al., 2010
Peroxisome	Gabalton et al., 2006; Gabalton, 2010
Cytokinesis	Eme et al., 2009

that diverged from some common ancestor (Poole and Penny, 2007b; Pace, 2006; Cavalier-Smith, 2002b; Woese et al., 1990—Fig. 1A). Alternatively eukaryotes may have evolved directly from within archaea (Fig. 1B) (Cox et al., 2008; Embley and Martin, 2006; Martin and Müller, 1998; Rivera and Lake, 1992, 2004). Numerous authors have reported evidence for one or the other general topology, with little sign of an emerging consensus (Gribaldo et al., 2010).

Phylogenetics is central to our understanding of the origin of the eukaryote cell because trees can distinguish between the two tree topologies given in Fig. 1 (Panels A and B). However, the two trees in Fig. 1 have been taken to imply very different (and incompatible) series of cell biological events for the endosymbiotic origin of the mitochondrion and the origin of eukaryotes. The tree in Panel A is equated with the hypothesis that the modern eukaryote cell evolved via a protoeukaryotic host (PEH) cell engulfing an ancient α -proteobacterium (Panel C) (Cavalier-Smith, 2002b, 2009; Poole and Penny, 2007a,b). In contrast, the tree in Panel B has been interpreted to mean that the host (the cell that did the engulfing) must have been an archaeon (Panel D) (Martin and Koonin, 2006; Martin and Müller, 1998). The model in Panel C has the advantage that it relies on cell biological processes known to be in action in the present (i.e. phagocytosis or subversion of phagocytic machinery as a mechanism for host infiltration), whereas the latter (Panel D) currently lacks cell biological evidence because no archaea are known to be capable of phagocytosis, and no archaea have been documented to harbour any bacterial endosymbionts (Poole and Penny, 2007a,b).

The primary point of this paper is to show that the tree topologies (Panels A and B) and the cell biological processes for endosymbiosis (Panels C and D) are not logically connected, despite a historical association between the models in Panels A and C, and between Panels B and D. To illustrate this, we will make the assumption that recent phylogenetic analyses reporting support for the Eocyte tree topology ((Lake, 1988; Rivera and Lake, 1992) — schematically represented in Panel B) (Cox et al., 2008; Foster et al., 2009) correctly recover the evolutionary relationship between archaea and eukaryotes. These analyses place eukaryotes as sister to the crenarchaeota, and, if subsequently corroborated, the implication is that eukaryotes have evolved directly from archaea. We show that, under an archaeal origin of eukaryotes, no

special unknown cell biological mechanisms of the type illustrated in Panel D (Embley and Martin, 2006; Martin and Müller, 1998; Davidov and Jurkevitch, 2009) are required to understand the origins of the eukaryote cell.

2. Ancestors, missing links, stems and crowns

In assuming that the Eocyte tree is correct, the biological problem that we face is as follows. All sequences used to investigate the deep phylogeny of eukaryotes and archaea necessarily come from extant organisms. The evidence for a complex eukaryote at the base of the eukaryote tree (LECA) (Koonin, 2010; Poole, 2010) (Table 1) is resultant from the observation that no intermediate forms are preserved among extant eukaryote lineages (Poole and Penny, 2007b).

In debate over human origins, Sarich (1973) famously remarked, ‘the biochemist knows his molecules have ancestors, while the palaeontologist can only hope that his fossils left descendants’. The problem for those seeking to reconstruct eukaryote evolution is the exact opposite. Comparative molecular and cell biology has painted a surprisingly sharp picture of LECA as a modern eukaryote cell (Table 1), but evolution has left no trace of the intermediate stages. Even tantalising fossils such as the 3.2 billion year old Acritarchs recently reported by Javaux et al. (2010) are difficult to interpret within this framework. While the suspicion that these are stem group eukaryotes has been voiced (Buick, 2010), the issue of whether they are or not is nevertheless unlikely to shed light on the questions raised by the reconstruction of LECA because detailed cell ultrastructure is not discernible in these fossils. It is moreover not possible to relate this find to the timing of the origin of archaea, as there is no firm evidence for the timing of their origin in the fossil record (Brocks et al., 2003).

That eukaryotes possess a multitude of large multiprotein complexes and internal structures that lack counterparts in both archaea and bacteria means eukaryogenesis cannot be understood by reference to cellular features of extant bacteria and archaea, because obvious precursor structures from which those traits could be derived are absent. Are we then restricted to speculation regarding the steps in eukaryogenesis?

The answer is a resounding no. The apparent controversy is perhaps a casualty of only being able to examine the diversity of extant eukaryotes. All extant eukaryote lineages are by

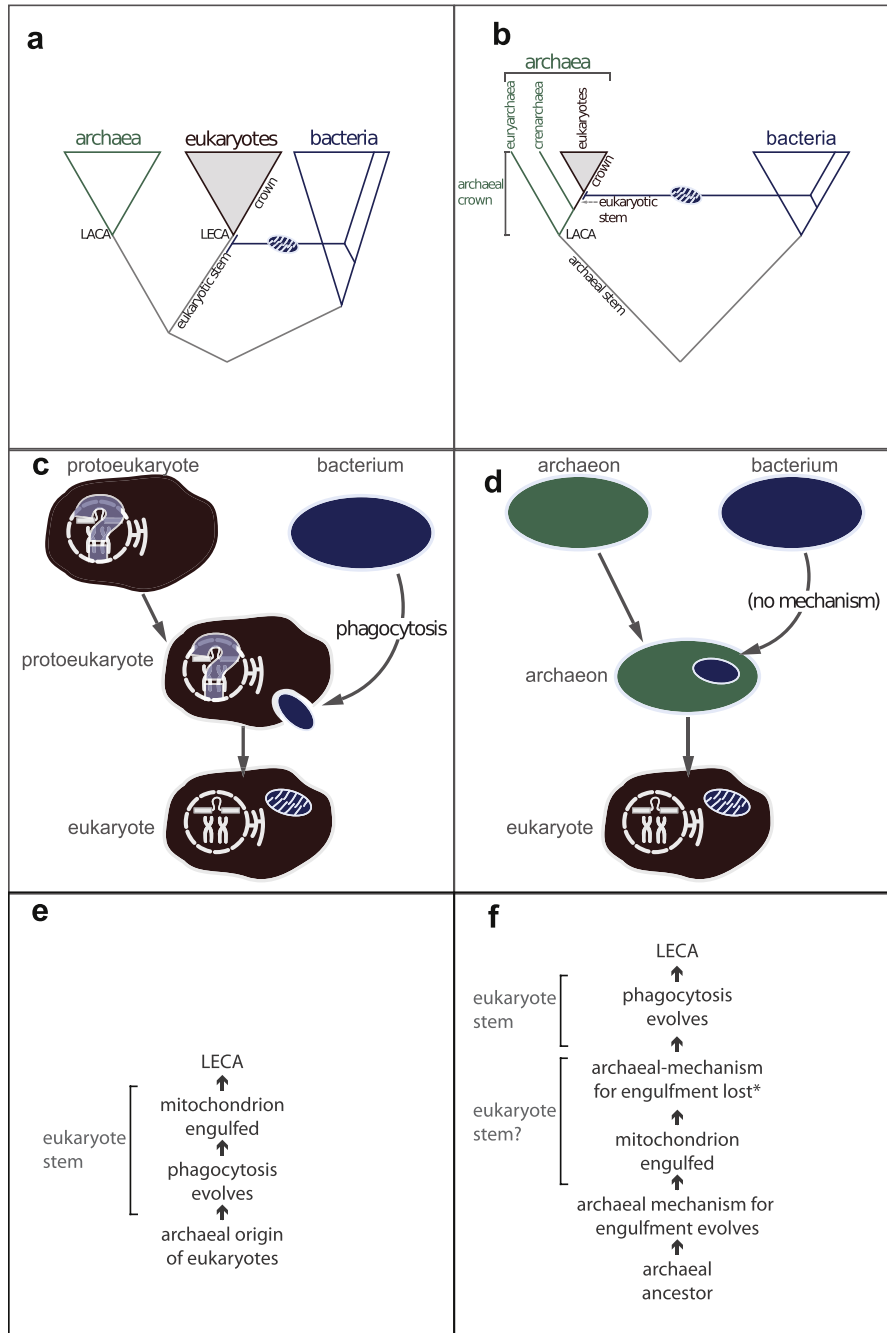


Fig. 1. Panels A and B. Possible phylogenetic relationships between eukaryotes and archaea. A. Eukaryotes and Archaea are each monophyletic. B. Eukaryotes as a derived group emerging within the Archaea. The specific topology shown is the Eocyte tree. Note that, under the Eocyte hypothesis, Eukaryotes are, phylogenetically, a group within archaea, so the eukaryote total group (stem plus crown) is nested within the diversity of the archaeal crown. In both trees, the bacterial contribution to the eukaryote cell via (endosymbiotic) gene transfer is shown for simplicity as a single line. Suggestions on the timing of the acquisition of mitochondria relative to the evolution of other eukaryote-specific traits ranges from the initial event to the final event in formation of the eukaryote cell. Uncertainty as to timing relative to other evolutionary events along the eukaryotic stem is therefore depicted by the line running parallel to the eukaryote stem in both panels. Bacteria are depicted as an outgroup in both trees, and the position of the Last Eukaryotic Common Ancestor (LECA), with the features summarised in Table 1 is shown. LECA: Last Eukaryotic Common Ancestor; LACA: Last Archaeal Common Ancestor. Panels C and D. The origin of the mitochondrion via engulfment by C. a protoeukaryote host and D. a hypothetical engulfment-competent archaeon. The question mark emphasises that the timing of the engulfment of the mitochondrion by a protoeukaryote host is uncertain relative to other eukaryote-specific features (though see Poole, 2010, for further discussion). No cell biological mechanism is currently known to support the model presented in Panel D. Panels E and F. A comparison of the series of events invoked by a protoeukaryotic (Panel E) versus an archaeal host (Panel F) for the origin of the mitochondrion shows that the former is more parsimonious than the latter. The scenario in Panel F is difficult to place precisely within the context of stem and crown. This difficulty arises because of the uncertainty associated with the timing of the emergence of the hypothetical mechanism of archaeal-based engulfment. Under the Eocyte tree in Panel B, if the host was an archaeon, engulfment would have to have evolved in the stem leading to the common ancestor of crenarchaea and eukaryotes. This scenario requires that the mechanism of engulfment is lost twice independently: once in the crenarchaea and once in the lineage leading to eukaryotes, following the engulfment of the mitochondrion (marked with an asterisk). The other alternative is that this 'archaeal' mechanism only evolved in the archaeal lineage leading to eukaryotes, which is technically the eukaryote stem. In the absence of fossil evidence for a distinct, non-phagocytic, mechanism involving a demonstrably archaeal host, this alternative simplifies to the scenario in Panel E, rendering an archaeal-specific mechanism for engulfment completely unnecessary.

definition constituents of the crown, whereas the evolution of those features fully formed and present in LECA must pre-date the diversification of modern eukaryotes (Poole and Penny, 2007a). Under an archaeal origin of eukaryotes (Fig. 1, Panel B) the evolution of all features in Table 1 can be readily attributed to the stem, because the ancestral state is archaeal cell architecture. As there are no intermediate forms among extant lineages, we have no way of directly observing the exact evolutionary steps leading to the origin of the modern eukaryote cell. Under any evolutionary model, these myriad complex features did not arise in a single step. Thus, irrespective of which is the correct tree in Fig. 1, there must be a eukaryote stem. The advantage of the Eocyte tree (Panel B) is that we can determine the direction of evolution: if the tree is correct, then archaeal traits are ancestral and eukaryote characters derived.

Given the logical necessity of a stem group under both trees (Panels A and B), it is uncontroversial to choose between the two scenarios given in Panels C and D. The reasoning is summarised in Panels E and F, and is as follows.

No contemporary eukaryote lineages are known that never possessed mitochondria (van der Giezen and Tovar, 2005), but, contrary to the suggestions of some, this does not weaken the model given in Panel C. This is because mitochondria and phagocytosis can both be traced to LECA (Yutin et al., 2009; Jékely, 2007b, 2008; Cavalier-Smith, 2009c), indicating that both features evolved prior to the diversification of extant eukaryotes. An uncontroversial interpretation is thus that phagocytosis evolved before the engulfment event that gave rise to mitochondria (Panel E). In contrast, the model depicted in Panel D requires that the archaeal ancestor of eukaryotes was capable of cell engulfment by a mechanism different from typical eukaryotic phagocytosis (otherwise there is no reason to consider this ancestor an archaeon, see legend to Fig. 1), allowing the acquisition of the mitochondrion. Such a mechanism would subsequently have to go extinct and be replaced by modern typical eukaryotic phagocytosis along the stem leading to the LECA (scenario in Panel F).

A model proposed by a number of authors (e.g. Davidov and Jurkevitch, 2009; Martin and Koonin, 2006; Martin and Müller, 1998) is that only the direct archaeal ancestors of eukaryotes evolved this mechanism, that is, an early ancestor in the eukaryote stem lineage pre-dating LECA. However, a direct archaeal host requires a convoluted series of events (Fig. 1, Panel F). In brief, a mechanism for engulfment would have to emerge among archaea², lead to engulfment of the mitochondrial ancestor, and then disappear (possibly twice—see legend to Fig. 1) with an unrelated machinery for phagocytosis emerging later in the eukaryote stem. We see no valid scientific reason for favouring such a convoluted series of events when a far simpler, and mechanistically plausible, model is possible, independent of tree topology (Table 2; Fig. 1, Panel E) (Jékely, 2007b).

Table 2

Engulfment of the mitochondrial ancestor by phagocytosis is compatible with both 3-domains and Eocyte topologies, but ‘archaeal engulfment’ is not.

	Sister groups (3-domains)	Eocyte (archaeal origin)
Phagocytosis in LECA	✓	✓
‘Archaeal engulfment’ ^a	×	×

^a Archaeal engulfment here refers to both a hypothetical mechanism by which an archaeon may have engulfed the mitochondrial ancestor and a hypothetical mechanism by which the mitochondrial ancestor may have invaded the host cytoplasm.

Ockham’s razor is frequently invoked with respect in discussions of early evolution as a means of arguing in favour of specific scenarios (Dagan and Martin, 2007; Martin and Müller, 1998). Under this criterion, the scenario in Panel F is clearly unnecessarily complex. However, evaluating the deep evolutionary past by reference to Ockham’s razor is not an ideal approach, especially in the current case where the data do not fit the models equally well. A more crucial consideration in evaluating the scenarios in Panels C and D is that under actualism (interpreting past events by reference to mechanisms in action in the present) the archaeal mode of engulfment involved in Panel D is currently unsupported. This is because it must appear in a specific lineage, and then be replaced by a similar mechanism in a later descendant for that lineage. Given that a rudimentary mechanism had certainly developed early in the evolutionary history of the eukaryote lineage (Yutin et al., 2009), there seems no compelling reason to favour engulfment by a direct archaeal ancestor, especially as such an unsupported cell biological event is not requisite for acceptance of the Eocyte hypothesis.

3. Concluding remarks

Phagocytosis by a protoeukaryote host is the only viable mechanism currently available to explain the origin of the mitochondrion and hence modern crown group eukaryotes. However, phagocytosis as a mechanism for cell engulfment is not associated with a particular phylogenetic topology, and can as easily be reconciled with an archaeal phylogenetic origin for eukaryotes, such as under the Eocyte tree, as it can with the three domains topology. An appreciation of the necessity of a eukaryote stem does not invoke special hypothetical intermediates under either phylogenetic tree. In the case of an archaeal origin of eukaryotes, recognition of the necessary existence of the stem enables us to appreciate that the process of eukaryogenesis from an archaeal ancestor to the LECA requires an enormous number of intermediate forms, none of which are represented among the modern diversity of eukaryotes.

Our intention in this paper is not to advocate a particular phylogenetic relationship between archaea and eukaryotes—we are neutral on whether archaea and eukaryotes are sister groups or whether eukaryotes are directly derived from archaea. Rather, our aim is to show that models favouring the latter do not require special mechanisms. Applying stem-and-crown thinking to this problem demonstrates that phylogenetic origins and the nature of the mitochondrial host are separate issues.

² The issue of whether archaeal lipids preclude engulfment is an issue that, in the absence of evidence either for or against archaeal engulfment or the invasion of archaeal cells by bacteria, remains purely academic.

Consequently, while it may not be possible to work out the exact order of events in eukaryogenesis, there is absolutely no sense in invoking a direct archaeal host for mitochondria. To do so results in the introduction of additional and unnecessary contingent events, where none are required.

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