Author/s:
Collier, SL;Farrell, SN;Goodman, CD;McFadden, GI

Title:
Modes and mechanisms for the inheritance of mitochondria and plastids in pathogenic protists

Date:
2025-01-01

Citation:
Collier, S. L., Farrell, S. N., Goodman, C. D. & McFadden, G. I. (2025). Modes and mechanisms for the inheritance of mitochondria and plastids in pathogenic protists. Plos Pathogens, 21 (1), https://doi.org/10.1371/journal.ppat.1012835.

Persistent Link:
https://hdl.handle.net/11343/359776

PLOS PATHOGENS

REVIEW

Modes and mechanisms for the inheritance of mitochondria and plastids in pathogenic protists

Sophie L. Collier[¤*], **Sarah N. Farrell, Christopher D. Goodman**[‡], **Geoffrey I. McFadden**[‡]

School of BioSciences, The University of Melbourne, Parkville, Victoria, Australia

¤ Current address: Department of Biochemistry & Pharmacology, Bio21 Molecular Science and Biotechnology Institute, The University of Melbourne, Australia
‡ These authors are joint senior authors on this work.
* sophie.collier@unimelb.edu.au

OPEN ACCESS

Citation: Collier SL, Farrell SN, Goodman CD, McFadden GI (2025) Modes and mechanisms for the inheritance of mitochondria and plastids in pathogenic protists. PLoS Pathog 21(1): e1012835. https://doi.org/10.1371/journal.ppat.1012835

Editor: Bjorn F. C. Kafsack, Joan and Sanford I Weill Medical College of Cornell University, UNITED STATES OF AMERICA

Published: January 23, 2025

Funding: This work was supported by an Australian Research Council Laureate Fellowship (FL1700008) to GIM and a National Health and Medical Research Council Investigator Award (GNT2016391) to GIM. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Competing interests: The authors have declared that no competing interests exist.

Abstract

Pathogenic protists are responsible for many diseases that significantly impact human and animal health across the globe. Almost all protists possess mitochondria or mitochondrion-related organelles, and many contain plastids. These endosymbiotic organelles are crucial to survival and provide well-validated and widely utilised drug targets in parasitic protists such as *Plasmodium* and *Toxoplasma*. However, mutations within the organellar genomes of mitochondria and plastids can lead to drug resistance. Such mutations ultimately challenge our ability to control and eradicate the diseases caused by these pathogenic protists. Therefore, it is important to understand how organellar genomes, and the resistance mutations encoded within them, are inherited during protist sexual reproduction and how this may impact the spread of drug resistance and future therapeutic approaches to target these organelles. In this review, we detail what is known about mitochondrial and plastid inheritance during sexual reproduction across different pathogenic protists, often turning to their better studied, nonpathogenic relatives for insight.

Protists are important—And so are their endosymbiotic organelles

Protists are abundant, diverse, predominantly unicellular eukaryotes. They are ubiquitous, and many are pathogenic to humans, animals, plants, and even other protists. Almost all protists contain a mitochondrion, or derived version thereof, and many contain a plastid. Mitochondria and plastids arose from endosymbiotic bacteria and retain a relic of the bacterial genome of their predecessors. In parasitic protists, these organelles provide essential metabolic pathways that can make them important drug targets. Consequently, mutations in the organelle genome can confer drug resistance. It is, therefore, important that we understand the inheritance of mitochondria and plastids in these microorganisms. In this review, we explore what is known about mitochondrial and plastid inheritance during sexual reproduction across pathogenic protists, in many instances turning to their better studied nonpathogenic relatives or other more distant, but better-characterised eukaryotes for insight.

Except for *Monocercomonoides*, all protists possess mitochondria, or reduced forms of mitochondria known as mitochondrion-related organelles (MROs) [1]. These structures are a feature of both mutualistic and parasitic protists belonging to the Alveolata clade and protist supergroups including Euglenozoa, Metamonada, Amoebozoa, and Percolozoa (Fig 1 and Table 1). It is widely held that the mitochondrion was acquired through primary endosymbiosis of an α-proteobacterium that was internalised by another cell 1.5 to 2 billion years ago [2,3]. The resulting chimeric cell was ancestral to all modern-day eukaryotes. A hallmark of mitochondrial endosymbiosis is the presence of 2 membranes surrounding every mitochondrion.

Many protists contain a second organelle of endosymbiotic origin known as a plastid (Table 1). Plastids originated through endosymbiotic internalisation of a cyanobacterium by a eukaryotic phagotroph approximately 1.2 billion years ago [4–6]. Over time, this ancestral, photosynthetic eukaryote diverged into glaucophytes (none of which are pathogenic), green algae and land plants (various of which are parasitic, even to humans), and red algae (a few of which are parasitic on other red algae). At some undefined point, an early ancestor of Apicomplexa, and likely also the sister group of dinoflagellates, engulfed a eukaryotic red alga which became an established plastid within the host and eventually evolved into the non-photosynthetic apicoplast within parasites like *Plasmodium* and *Toxoplasma* [7] (Fig 1). The presence of 4 membranes surrounding the apicoplast is a signature of this secondary endosymbiosis [8]. The inner 2 membranes are the cyanobacterial (gram negative) derived membranes of the red algal plastid, the third membrane is hypothesised to be derived from the plasma membrane of the engulfed red alga, and the outer membrane is a remnant of the host phagosome that enclosed the endosymbiont during initial endocytosis [4].

Dinoflagellates are close relatives of Apicomplexa that have adopted numerous lifestyles including parasitism. Some free-living dinoflagellates are also pathogenic to humans via the toxins they produce. Many dinoflagellates possess plastids of red algal origin also acquired through a secondary endosymbiosis event. These plastids differ from apicoplasts in that they are surrounded by 3 membranes. Many are photosynthetic, containing chlorophylls *a* and *c*, and the light-harvesting carotenoid pigment peridinin [59–61]. Peridinin plastids, and plastids in the related chromerid algae, likely share a common endosymbiotic origin with apicoplasts of Apicomplexa [62–65] (Fig 1). Somewhat surprisingly, many dinoflagellates have undergone further endosymbiotic events to replace their original peridinin plastid with new photosynthetic endosymbionts derived from a wide range of microorganisms including green algae, cryptophytes, haptophytes, or diatoms [66–68]. Such replacements can be secondary, tertiary, or even higher order endosymbioses [69]. Other dinoflagellates retain plastids that lack photosynthesis altogether, instead leading heterotrophic or parasitic lifestyles. At least one of these species, *Haematodinium*, has lost the plastid altogether [70,71].

Euglenids such as *Euglena*, procured plastids by secondary endosymbiosis independently of the alveolate dinoflagellates and apicomplexans [72]. The euglenids are part of the Euglenozoa supergroup that includes parasitic kinetoplastids such as *Trypanosoma* and *Leishmania* (Fig 1). Plastids were at one time thought to occur in trypanosomes [73], but this remains unsubstantiated.

Unique characteristics of endosymbiotic organelles

Mitochondria and plastids are semi-autonomous, containing their own genomes that are inherited independently from the nucleus, often in a non-Mendelian pattern. For both organelles, a uniparental (typically maternal) pattern of inheritance predominates (reviewed in [76,77]). While organellar inheritance of mitochondria is considered strictly maternal in the

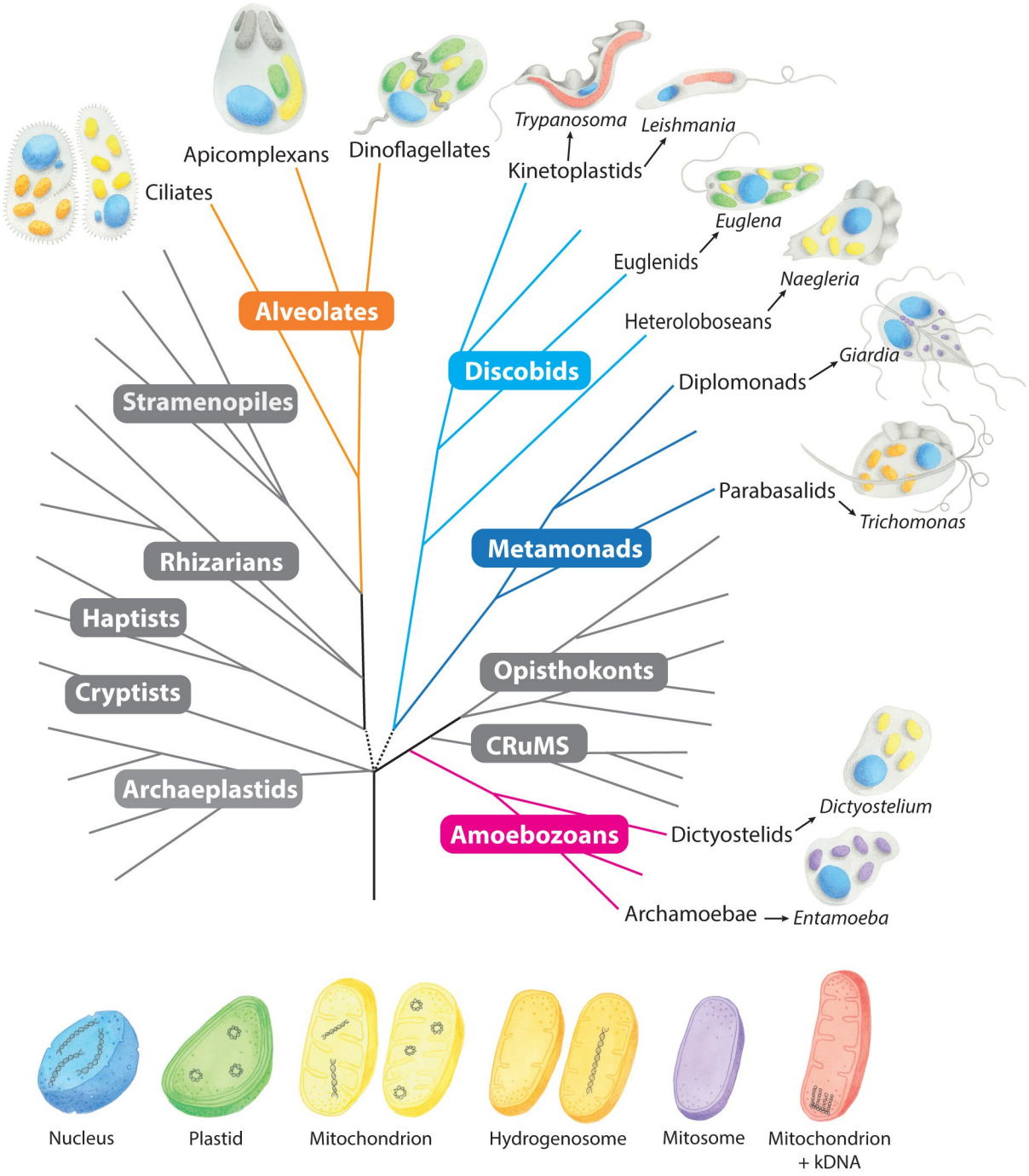


Fig 1. A phylogenetic tree of eukaryotes showing the variety of endosymbiotic organelles in pathogenic and nonpathogenic protists. The main lineages are redrawn according to Keeling and Burki (2019) and Husnik and colleagues (2021) and are depicted according to current consensus phylogeny [74,75]. Protists and organelles are hand-painted by Sarah N. Farrell. Clades and supergroups of interest are highlighted in colour. Each protist contains a nucleus (or a micronucleus and macronucleus in Ciliata) and endosymbiotic organelles. Organelles are colour coded according to the legend at the bottom of the figure. Classical mitochondria (yellow) can contain either linear or circular organellar genomes (as specified for each protist in Table 1 and the body text). Hydrogenosomes (orange) may also contain a linear genome or lack a mitochondrial genome entirely. The mitochondrion of *Trypanosoma* and *Leishmania* contains kinetoplast DNA (kDNA) comprised of DNA minicircles and maxicircles (red).

https://doi.org/10.1371/journal.ppat.1012835.g001

Table 1. Summary of endosymbiotic organelles and organellar genome contents of the protists discussed in this review.

Protist group	Species	Endosymbiotic organelles	Organelle genome size (kb)	Organelle genome features	Source
Dinoflagellates	Various	Mitochondria	6–10 fragments (up to ~326 total in *Symbiodinium minutum*)	Linear, highly fragmented	[Reviewed in 9–11]
		Plastids	1.8–6 kb per minicircle&	Numerous minicircles&	[Reviewed in 12–15]
Apicomplexans	*Plasmodium*	Mitochondrion	6.0	Linear, single fragment	[16,17]
		Apicoplast	29.6–34.2*	Circular, single fragment	[16,18,19]
	Toxoplasma	Mitochondrion	5.9	Linear, fragmented	[20,21]
		Apicoplast	35.0	Circular, single fragment	[19,22]
	Eimeria	Mitochondrion	6.2	Linear, single fragment	[23,24]
		Apicoplast	34.8	Circular, single fragment	[19,25]
	Babesia	Mitochondria	6.2–11.1*	Linear, single fragment	[26–30]
		Apicoplast	28.7–35.1*	Circular, single fragment	[19,26,31–34]
	Theileria	Mitochondria	6.6–8.2*	Linear, single fragment	[30,35–38]
		Apicoplast	31.7–47.8*	Circular, single fragment	[19,37]
	Cryptosporidium	Mitosome	None	N/A	[Reviewed in 39]
	Gregarines	Mitosome	None	N/A	[40]
Ciliates	*Nyctotherus ovalis*	Hydrogenosomes	~41.6 (incomplete sequence)	Hypothesised to be linear (not directly studied)	[41–43]
	Paramecium	Mitochondria	40.0–44.0*	Linear, single fragment	[44,45]
Percolozoans	*Naegleria*	Mitochondria	49.5	Circular, single fragment	[46]
Euglenozoans	*Euglena*	Mitochondria	~1.0–9.0 (incomplete sequence)	Linear, fragmented	[47–49]
		Plastids	143.2	Circular, single fragment	[48,50]
	Leishmania	Mitochondrion	20–40 (maxicircles) ~1–2.5 (minicircles)	Comprised of 1,000 s of mini and 25–50 maxicircles	[51–53]
	Trypanosoma	Mitochondrion	20–40 (maxicircles) ~1–2.5 (minicircles)	Comprised of 1,000 s of mini and 25–50 maxicircles	[52–54]
Metamonads	*Giardia*	Mitosomes	None	N/A	[55]
	Trichomonas	Hydrogenosomes	None	N/A	[56]
Amoebozoans	*Dictyostelium*	Mitochondria	55.6	Circular, single fragment	[57]
	Entamoeba	Mitosomes	None	N/A	[58]

*Species dependent.

&In peridinin plastids specifically, plastid genomes vary in other "replacement plastid" species.

https://doi.org/10.1371/journal.ppat.1012835.t001

animal kingdom, this is not true across other kingdoms of Life, where numerous cases of distinct paternal or biparental inheritance are reported for both mitochondria and plastids (reviewed in [78]). Moreover, the dogma of strict maternal mitochondrial inheritance in animals is being overturned by increasingly common reports of paternal leakage in diverse animal species [79–81]. Nevertheless, a preference for uniparental inheritance of mitochondria and plastids during mating is evident throughout Nature, suggesting that possession of a single organellar genotype is advantageous.

It is postulated that uniparental inheritance evolved to minimise the spread of fast-replicating, deleterious, or selfish organellar genome mutations that would create direct competition between divergent intracellular organellar genotypes to the detriment of the host [82–84]. The establishment of endosymbiotic organelles is routinely accompanied by gene transfer of many organellar genes to the nuclear genome of the host, while others are lost entirely, apparently no

longer being required by the symbiotic partnership. Nuclear-encoded proteins, both those derived from the endosymbiont genes transferred to the nucleus, and those newly directed from host to endosymbiont, are imported into their respective organelles and are essential for their function, so cooperation and co-adaption between the nuclear and organellar genomes is imperative to the survival of the symbiotic amalgam [85]. Furthermore, uniparental inheritance encourages co-adaption between the 2 genomes and helps to discourage or eliminate genetic conflicts that may arise between them, thus avoiding genetic incompatibility [83,86,87].

Mechanisms for uniparental inheritance

Although uniparental inheritance is widespread, and apparently favourable to the survival of most eukaryotic organisms, there is staggering diversity in the mechanisms that facilitate this uniparental inheritance during sexual reproduction. Intriguingly, variation in mechanisms is observed even in closely related species, suggesting that uniparental inheritance has arisen independently numerous times, perhaps even being lost and regained along multiple evolutionary trajectories. To add to this complexity, there are 3 major stages during sexual reproduction in which uniparental inheritance mechanisms are reported to be implemented: 1/ gametogenesis, 2/ fertilisation, and 3/ post-fertilisation (reviewed in [5,76,77,81,88–90]). It is important to highlight that many organisms use a combination of different mechanisms across these unique checkpoints to increase the chances that paternal organelles are eliminated entirely. Given this, we believe that the timing of these mechanisms directly influences the possibility of a breakdown in maternal inheritance, which is known as paternal leakage. For example, pre-fertilisation mechanisms that degrade or decrease organellar DNA (oDNA) or organelles in gametes prior to fertilisation should be the most effective in deterring paternal leakage because exclusion of organelles from gametes of one sex (canonically males) ensures that they are not delivered to the zygote during mating. In this scenario, any persisting paternal organelles must pass through 2 subsequent phases of sexual reproduction (and their accompanying mechanisms) to become established within the offspring. Mechanisms that occur during later stages of reproduction have fewer opportunities to eliminate one parent's organelles and, therefore, should be more prone to paternal leakage. Fig 2 details previously reported mechanisms known to facilitate maternal inheritance at each stage of sexual reproduction across the unique kingdoms of Life (reviewed in [5,76,77,81,88–90]) and our view of how efficient we expect them to be at ensuring uniparental inheritance.

Paternal leakage—Why is it important?

While uniparental inheritance apparently confers some—as yet undefined—fitness benefit, it also carries its own set of evolutionary limitations. The mutation rate of oDNA is frequently higher than in nuclear DNA, likely due to continuous exposure to reactive oxygen species (ROS), limited DNA repair mechanisms, or intrinsic characteristics of organellar ancestry [91–93]. Uniparental inheritance eliminates the opportunity for any kind of purifying selection or recombination between organellar genomes, inevitably leading to an accumulation of deleterious mutations via Muller's ratchet [76,94]. However, a growing body of evidence suggests that occasional biparental transmission (paternal leakage) may provide opportunities for sporadic recombination to counteract these deleterious effects [83,95–97].

Although uniparental inheritance predominates in sexual crosses within the same species, studies of interspecies hybridisation indicate that control of mitochondrial and plastid inheritance is more tenuous in crosses between different species, and even between individuals from divergent populations of the same species (reviewed in [77,95,98]). This may be due to a

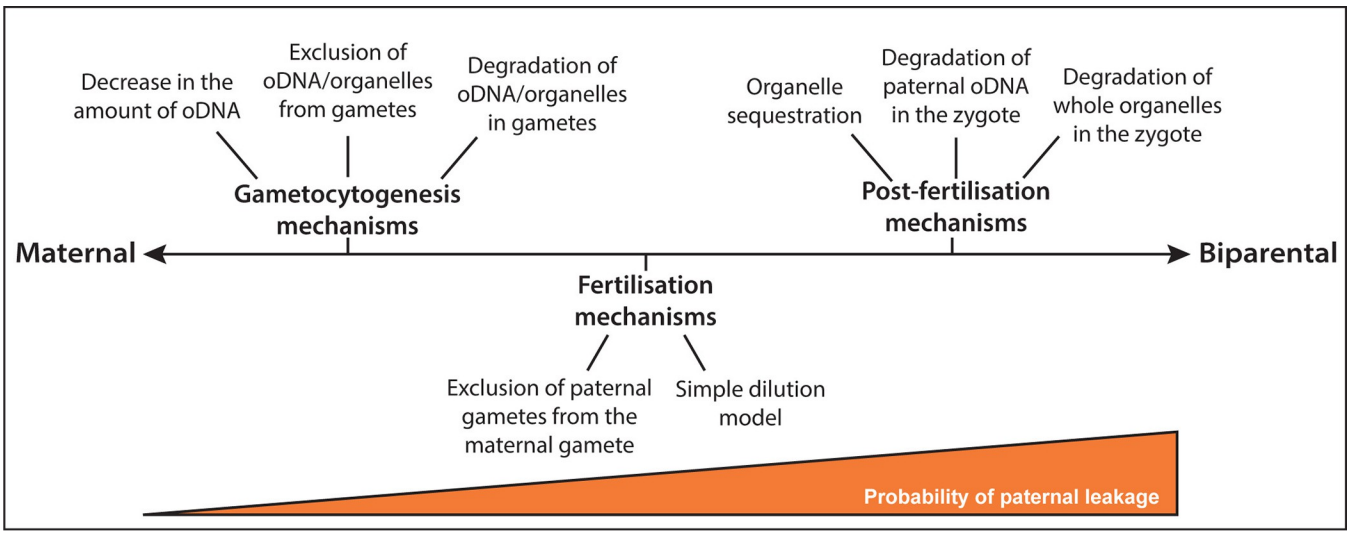


Fig 2. Mechanisms used to achieve uniparental (maternal) inheritance and the probability of paternal leakage or biparental inheritance. This model details the mechanisms commonly used to achieve uniparental (maternal) organellar inheritance of mitochondria and plastids at distinct time points during sexual reproduction, as previously reported (reviewed in [5,76,77,81,88–90]). The orange ramp at the bottom represents our proposal that mechanisms implemented earlier in sexual reproduction should more effectively prevent paternal leakage than those during or after fertilisation. In other words, late mechanisms are potentially more prone to failure resulting in leakage of male derived organelles into sexually produced offspring.

https://doi.org/10.1371/journal.ppat.1012835.g002

breakdown of, or distinct differences in, the mechanisms used to control uniparental inheritance between the 2 individuals and creates the opportunity for recombination of oDNA from a parent that does not usually persist or contribute to the offspring [99,100]. There is direct evidence of paternal leakage of mtDNA after hybridisation in *Drosophila* [101], periodical cicadas [102], mice [103,104], blue mussels [105,106], sea turtles [107], potato cyst nematodes [108], pine trees [109], and maidens' tears (*Silene vulgaris*) [110]. Similarly, indirect evidence of paternal leakage has been detected through the discovery of recombinant mtDNA genotypes in the great tit [111], conifers [112], silk moths [113], and the cryptococcosis causing fungus *Cryptococcus gatti* [114].

Paternal leakage can also rescue hybrids suffering from cytonuclear incompatibility, where the nuclear and organellar genomes are mismatched and unable to function together [115,116]. Plant hybrids experiencing plastid-specific cytonuclear incompatibilities exhibit traits such as chlorotic leaves and high rates of hybrid mortality. However, such phenotypes were rescued in hybrids where paternal leakage was detected [117–119] because one parental plastid genome is more compatible with the hybrid nuclear background than the other [120–122]. Paternal leakage can rescue cytonuclear incompatibilities by introducing genetic variation among organellar haplotypes and increasing the likelihood that a compatible organellar genome is inherited [76,123]. Interestingly, a similar phenomenon occurs in some parasitic protists that demonstrate transient biparental organellar inheritance followed by segregation of organelles in the offspring (see below [124–126]).

Mitochondria and plastids as drug targets in parasitic protists

In most, if not all protists, mitochondria and plastids underpin metabolic processes essential for survival. Thus, in parasites such as *Plasmodium* and *Toxoplasma*, the apicoplast and mitochondrion are useful drug targets due to their bacterial-like ancestry and essentiality to the parasite throughout the life cycle [127,128]. Apicomplexan mitochondria possess unique electron transport chain components, metabolic pathways, and protein translation machinery that

can be selectively inhibited without interfering with the mitochondria of the host. Likewise, the absence of apicoplasts or plastid-like organelles in human cells makes this compartment a prime target for the development of therapeutics to combat apicomplexan parasites, and several commonly used antimalarial drugs perturb mitochondrial or apicoplast functions to kill parasites [129,130]. However, mutations in organellar genomes can render parasites resistant to some of these drugs (reviewed in [131]), challenging our ability to control parasitic diseases. Thus, it is important to understand how organellar genomes, and the resistance polymorphisms contained within them, are inherited during parasite sexual reproduction and what impact uniparental inheritance has on the spread of resistance through a population of parasites.

Pathogenic protists with unusual organelles and mysterious sexual cycles

Understanding mitochondrial and plastid inheritance during sexual reproduction in protist pathogens requires knowledge of the morphology and genomic contents of the endosymbiotic organelles, as well as the protist's sexual cycle. Our understanding of organellar biology is often more advanced than our understanding of sex in many euglenozoans, metamonads, amoebozoans, and percolozoans (summarised in Fig 3). There are some studies that indirectly describe the sexual cycles of these protists, but they provide little basis for making hypotheses about the inheritance of mitochondria and plastids during mating.

An instructive example is the common photosynthetic euglenozoan *Euglena* that contains around 10 chloroplasts [132,133]. The chloroplast genome of *Euglena gracilis* was among the first ever characterised organellar genomes and a genome sequence followed [50,134]. Alongside these chloroplasts, *Euglena* possesses a massive, reticulated mitochondrion that undergoes extensive morphological modulation throughout the life cycle. During cell division and later stages of development, the mitochondrion fragments into 200 to 500 individual mitochondria per cell [135,136]. These organelles contain distinct mitochondrial genomes, which have been sequenced [137,138]. However, despite extensive study of the endosymbiotic organelles within these protists, evidence of sexual reproduction is confined to a single paper by Biecheler (1937), where she describes sex in a single species of *Euglena* [139], and investigations of organellar inheritance are not available.

Naegleria fowleri is a unicellular free-living percolozoan that opportunistically infects the central nervous system of humans, resulting in the fatal disease primary amoebic meningoencephalitis [140]. *N. fowleri* exists in aerobic freshwater environments and contains multiple mitochondria during all life stages [141–146]. The mitochondrial genomes of *N. fowleri*, and the related *N. gruberi*, have been sequenced and suggest both aerobic and anaerobic life-stages within these protists [147,148]. It remains unconfirmed whether *Naegleria* is sexual, but some evidence suggests that these organisms do—or at least did—participate in sexual reproduction. For example, the genome of *N. gruberi* encodes homologs of functional meiosis-specific genes [147]. Likewise, the *N. gruberi* NEG strain is heterozygotic—a strong indication that these parasites are the product of mating [147]. Moreover, isoenzyme studies suggest that the related, but nonpathogenic, *N. lovaniensis* participates in some form of sexual reproduction [149].

Many parasitic protists lack canonical mitochondria but instead contain MROs such as mitosomes and hydrogenosomes [150]. These MROs are typically found in anaerobic protists and have reduced structure and functions compared to canonical mitochondria. In many cases, they lack an organellar genome altogether, with the entire proteome being encoded by the nucleus and imported from the cytoplasm. The metamonad *Giardia intestinalis* is an anaerobic protist containing 40 to 50 mitosomes that lack mtDNA, possess a reduced

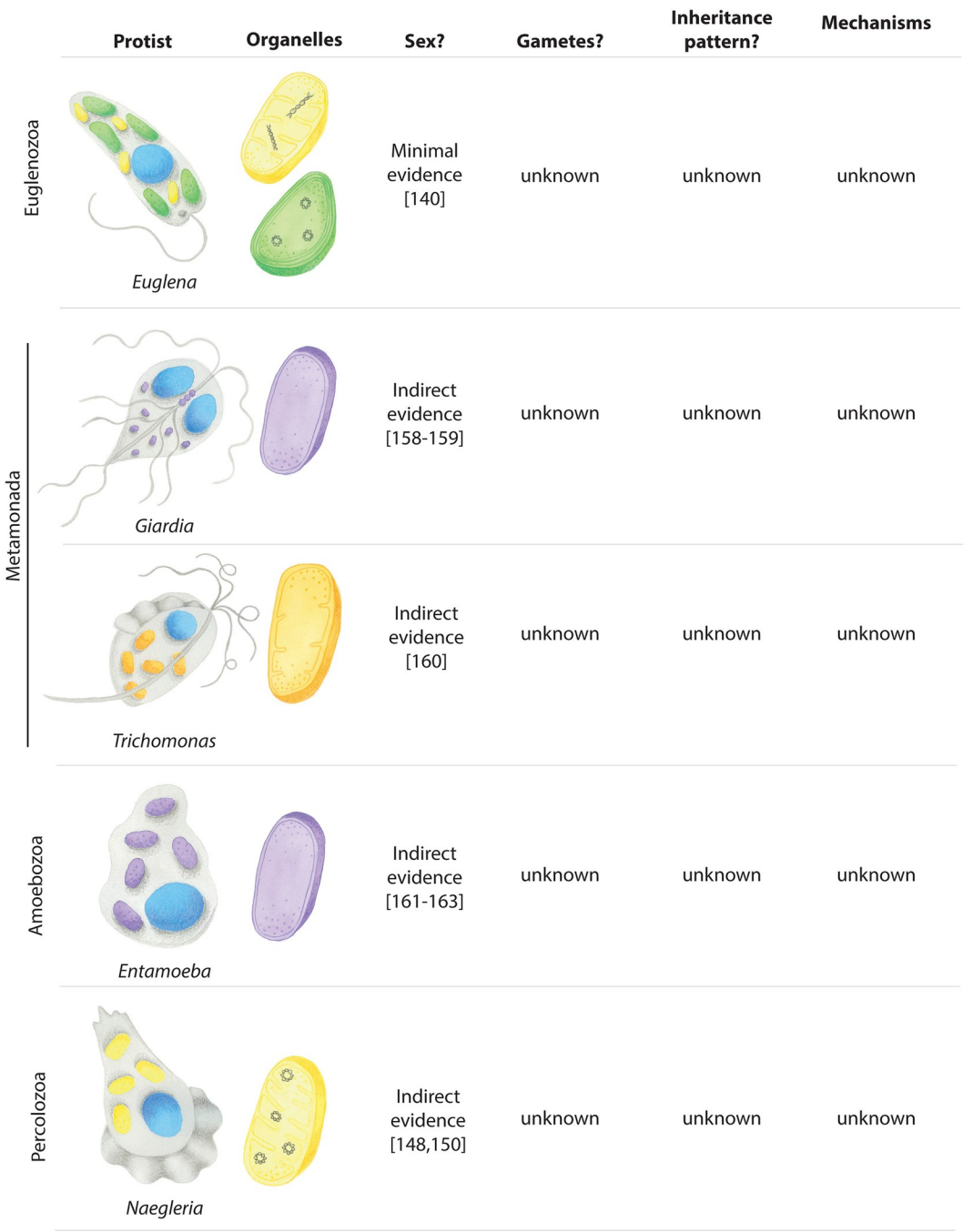


Fig 3. Pathogenic protists with unknown sexual cycles and mitochondrial and plastid inheritance patterns during mating. This figure illustrates the endosymbiotic organelles found in each protist. Protists and organelles are hand-painted by Sarah N. Farrell. Mitochondria (yellow) contain either circular or linear genomes as depicted. Hydrogenosomes (orange) and mitosomes (purple) do not contain an organellar genome. *Euglena* plastids (green) possess a circular genome. References are provided in brackets below the text where relevant.

https://doi.org/10.1371/journal.ppat.1012835.g003

proteome [55] and function specifically in iron-sulphur cluster biosynthesis [151]. *Entamoeba histolytica* possess a mitosome that also lacks an organellar genome, but, unlike *Giardia*, the *E. histolytica* mitosome does not generate iron-sulphur clusters but is instead implicated in

sulfate activation pathways [152]. Both giardiasis and amebiasis can be treated using the pyruvate:ferredoxin oxidoreductase inhibitor nitazoxanide, which disrupts anaerobic energy metabolism in the mitosomes of these protists [153,154]. The metamonad *Trichomonas vaginalis* possesses an MRO called the hydrogenosome, which lacks a genome, cristae, and an electron transport chain (ETC) but produces ATP and molecular hydrogen under anaerobic conditions [56,155]. *Trichomonas* causes the sexually transmitted disease trichomoniasis, which can be treated using hydrogenosome targeting compounds such as 5-nitroimidazole and nitazoxanide [153,156]. Again, nothing is known how *Trichomonas* passes down its hydrogenosomes to progeny.

Sexual reproduction has not been directly observed in any of these parasites, but several lines of genetic evidence suggest a current, or recently lost, sexual stage. Population genetics and fluorescence *in situ* hybridisation studies give evidence of sexual outcrossing and genetic recombination in *Giardia*, *Trichomonas*, *Entamoeba*, and *Naegleria* [157–159]. The presence of meiosis-related genes infers a sexual cycle in *Trichomonas*, *Entamoeba*, and *Naegleria* [147,159–161] as do the observed sexual cycles in the close relative of *Entamoeba*, namely, *Dictyostelium* [162]. Furthermore, the existence of retrotransposons in the *Trichomonas* genome [159] and the presence of isozymes in *Naegleria* [149] also provide evidence for a sexual cycle.

While the above evidence suggests that each of these protists does, or at least has the capacity to participate in sexual reproduction, organellar inheritance cannot be studied until their sexual cycles are described. If found to be sexual, many of these protists provide exciting opportunities to study the modes and mechanisms for mitochondrial and plastid inheritance during mating in the context of atypical plastids and mitochondria. For example, it would be particularly intriguing to track the inheritance of MROs such as the mitosome and hydrogenosome to ascertain whether the same rules of mitochondrial and plastid inheritance apply to organelles without a genome, and if the evolutionary pressure to regulate the proliferation of the organellar genome plays a role in the loss of oDNA.

We now turn our focus to protists in which sexual reproduction is documented, and which may provide clues as to the sorts of inheritance patterns and mechanisms we expect to find in these understudied pathogenic protists.

Pathogenic and nonpathogenic protists with known sexual cycles but unknown patterns of mitochondrial and plastid inheritance

In some groups of protists, including dinoflagellates and ciliates, sexual reproduction is well characterised, and we have valuable insights into the presence, morphology, and behaviour of mitochondria and plastids during pre- and post-zygotic stages of their life cycles. Although mitochondrial and plastid inheritance during mating is yet to be directly studied in these protists, we can begin to form hypotheses of possible modes and mechanisms for mitochondrial and plastid inheritance by drawing parallels with related organisms in which these processes are better understood (summarised in Fig 4).

Dinoflagellates

Dinoflagellates play important and diverse roles in marine and freshwater ecosystems around the globe. Some species are primarily free-living autotrophs or mixotrophs (e.g., *Amphidinium* and *Pyrocystis*), whereas others survive as symbionts, forming mutualistic relationships with marine invertebrates such as corals and anemones (e.g., *Symbiodinium*) [163,164]. Dinoflagellates can also be heterotrophic or parasitic in nature, infecting various organisms including marine crustaceans, molluscs, salps, tunicates, rotifers, annelids, algae, protozoans, and fishes, but not humans [165–170]. Some dinoflagellates are pathogenic to humans, however,

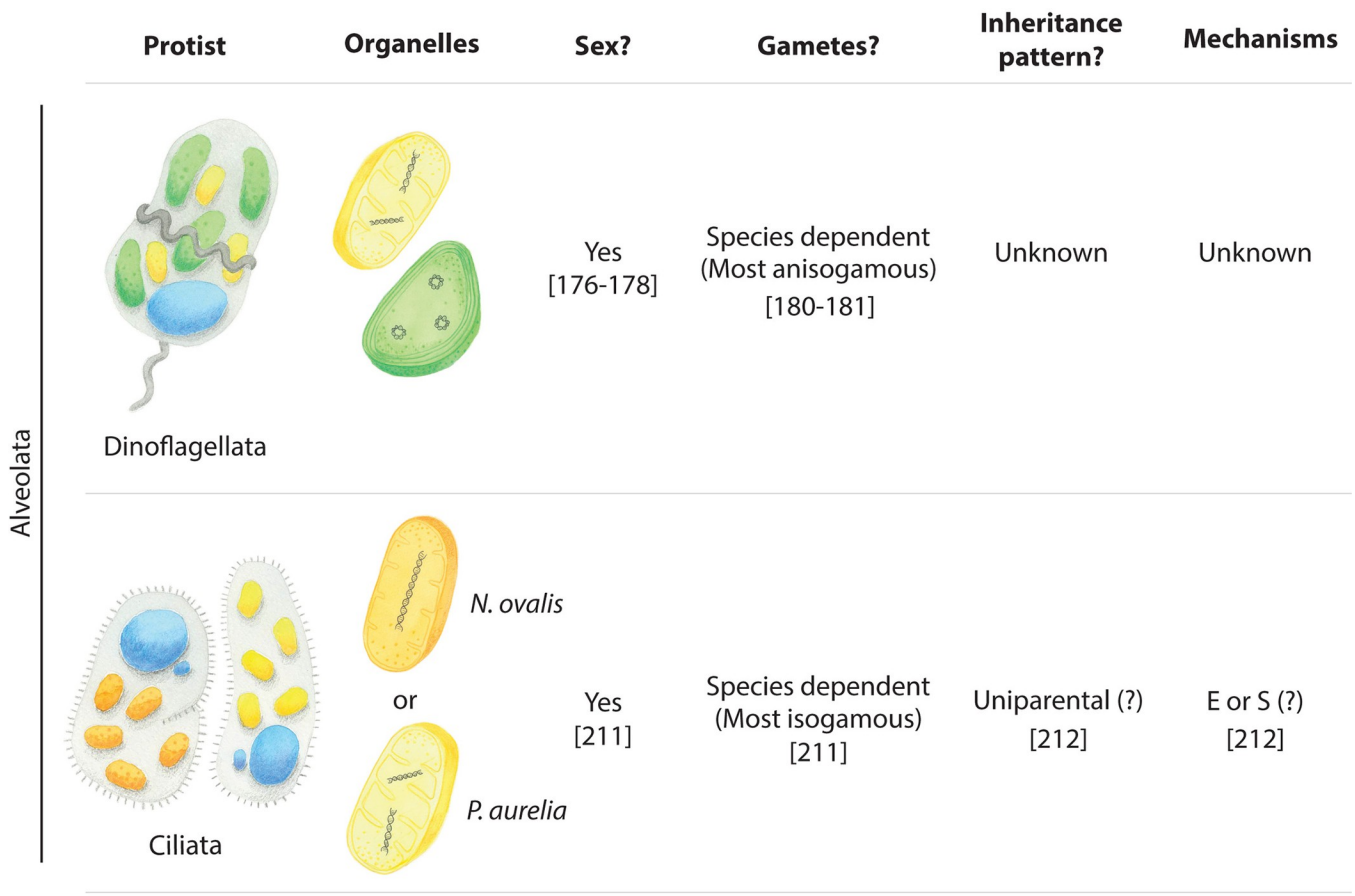


Fig 4. Nonpathogenic protists with known sexual cycles but unstudied mitochondrial and plastid inheritance patterns. Protists and organelles are hand-painted by Sarah N. Farrell. Within the group Ciliata, some ciliates contain hydrogenosomes (orange) with linear genomes (e.g., *N. ovalis*) while others possess classical mitochondria (yellow) with linear genomes (e.g., *P. aurelia*). Dinoflagellates possess both mitochondria and plastids (green), each harbouring their own genome. Potential mechanisms for uniparental inheritance in these groups include exclusion (E) and sequestration (S). References are provided in brackets below the text where relevant.

https://doi.org/10.1371/journal.ppat.1012835.g004

producing toxins that accumulate in seafoods and cause ciguatera poisoning or paralytic shellfish poisoning [171].

Dinoflagellate mitochondria are similar to those in animals. There are multiple mitochondria in each cell that contain tubular cristae and discrete genomes [172]. Dinoflagellates plastids are, however, more varied and possess unique features. Most dinoflagellates possess photosynthetic peridinin-containing plastids that are the product of secondary endosymbiosis of a red alga [173]. Intriguingly, the genome of these plastids is uniquely broken into multiple minicircles that are a mere 2 to 3 kb in size and typically encode a single gene each [174]. Some dinoflagellates have discarded their original peridinin-containing plastid for alternative photosynthetic bodies [69,174]. Further, many heterotrophic or mixotrophic dinoflagellates contain plastids that have lost their photosynthetic capabilities [174], and at least 1 dinoflagellate has lost its plastid altogether [71]

Dinoflagellates are capable of sexual reproduction [175–177]. Most reproduction in dinoflagellates is asexual, but certain environmental conditions or stressors induce sexual development [177,178]. Many dinoflagellates produce morphologically similar, or isogamous, gametes, meaning that both mating types are indistinguishable from one another. Others, such as *Noctiluca* and *Pyrophacus steinii*, are anisogamous, meaning they produce morphologically

distinct gametes [179,180]. While mitochondria are yet to be observed in dinoflagellate gametes, various studies report the presence of multiple plastids [181]. Interestingly, dinoflagellate gametes are widely reported to possess a reduced number of plastids compared to asexual stage vegetative cells [182–189]. However, ultrastructural studies have revealed the presence of multiple plastids and mitochondria in zygotes and post-zygotic life stages [181,190,191]. Conversely, the number and size of mitochondria appear to decrease as the zygote develops, and plastids are reported to partially degenerate or decrease in number [190,192]. Despite this, multiple mitochondria and plastids have been observed in planomeiocytes (a temporary, motile form that emerges from the hypnozygote shell under favourable conditions) during later stages of sexual development [192]. This is noteworthy because in many other organisms, decrease in the number of mitochondria, plastids, or organellar nucleoids belonging to a particular parental gamete is used as a mechanism for uniparental inheritance. For example, in the Japanese rice fish, *Oryzias latipes*, the number of paternal mtDNA nucleoids is decreased during spermatogenesis to aid in maternal inheritance [193]. Similarly, in the fern *Pteris vittata*, paternal chloroplasts undergo a dramatic 1/15 decrease in volume during spermatogenesis. Subsequently, paternal chloroplasts divide in the absence of chloroplast DNA (cpDNA) replication, further reducing the overall number of cpDNA nucleoids present [79,194]. It is plausible that similar mechanisms may be implemented in dinoflagellate gametes to underpin a uniparental inheritance pattern for these endosymbiotic organelles. That said, studies into the fate of mitochondria and plastids in sexual dinoflagellates, and their inheritance patterns during mating, are yet to be conducted.

Overall, it is plausible that organelles may be inherited from a single parent in dinoflagellates (particular in anisogamous species), and that the degenerating plastids observed in post-zygotic stages may belong to one mating type and be selectively degraded to ensure uniparental inheritance of these organelles. Such phenomena have been observed for mitochondria in *Caenorhabditis elegans* [81,195], mitochondria and plastids in *Volvox carteri* [196], mitochondria in *Dictyoshaeria cavernosa* [197], mitochondria and plastids in several isogamous green algae (reviewed in [79]) and *Chlamydomonas reinhardtii* [198–200]. Understanding the fate of these organelles and their genomes across different dinoflagellate species will offer insight into both the patterns and mechanisms of organellar inheritance and how different lifestyles, organelle origin, and specific function impact the fidelity and mechanisms of uniparental inheritance.

Ciliates

Ciliates are unicellular protists defined by dimorphic nuclei and the presence of cilia during at least 1 stage of their life cycle. *Balantidium coli* is the only ciliate known to parasitise humans, residing in our gastrointestinal tract. While *B. coli* was first described to contain mitochondrion-like bodies lacking cristae or tubules [201], ultrastructural studies of an isolate from the Philippines described the presence of a mitochondrion with cristae [202]. It remains unclear whether this mitochondrion harbours a mitochondrial genome or possesses its own bacterial-like translational machinery. However, the 30S ribosomal subunit inhibitor tetracycline is used to treat balantidiasis, and therefore may function by impeding protein translation in the mitochondrion of this anaerobic ciliate [203].

Unlike their apicomplexan and dinoflagellate relatives, ciliates lack any plastid [204]. However, many ciliates participate in kleptoplasty, where they retain and utilise plastids of an ingested algal prey. The stolen plastids in ciliates appear unable to divide and must be replaced from newly engulfed prey following cell division, so it seems unlikely that they have a dedicated mechanism of inheritance [205,206].

It is well-established that ciliates can occupy both aerobic and anaerobic environments. Aerobic ciliates tend to possess large numbers of classical mitochondria (e.g., *Paramecium*), whereas anaerobic species contain hydrogenosomes, some of which still contain their own mitochondrial genome (e.g., *Nyctotherus ovalis*) [207–209]. Given that there is almost no exchange of cytoplasm or other organelles between 2 conjugants (or gametes) during the unusual ciliate mating [210], mitochondrial inheritance is perceived to be uniparental [211]. As a result, exclusion or sequestration mechanisms appear to be implemented by default in these organisms to achieve uniparental inheritance. Ciliates are amenable to genetic modifications, and in future, ultrastructure or microscopy studies could be applied to test this conclusion or to uncover other mechanisms used to achieve their uniparental inheritance pattern.

Pathogenic protists with known sexual cycles and traceable patterns of mitochondrial and plastid inheritance

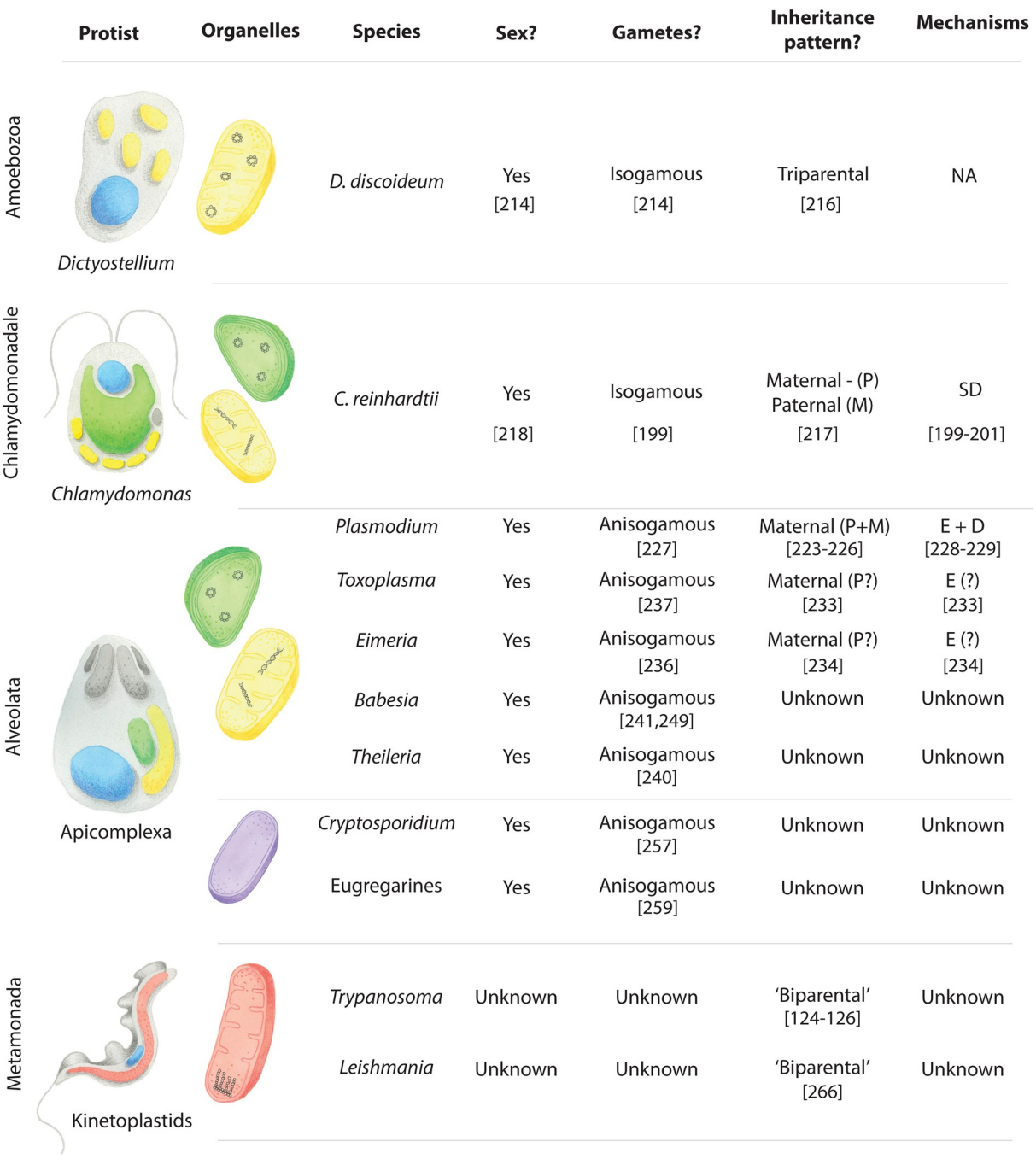
The few protists where mitochondrial and plastid inheritance during mating has been studied offer some valuable insights into the modes of organellar inheritance and accompanying mechanisms we might expect to find in other related alveolates, metamonads, euglenozoans, and amoebozoans (summarised in Fig 5). Furthermore, the experimental approaches used to study these processes can be applied in other protists to broaden our understanding of mitochondrial and plastid inheritance during mating in these understudied groups.

Dictyostelium: A protist related to the pathogenic *Entamoeba* with a peculiar pattern of mitochondrial inheritance

Dictyostelium is a bacterivorous amoebozoan that proliferates as single cells but undergoes significant changes in behaviour and development when starved, forming intimate contacts with surrounding cells of the same strain. During the sexual development that follows, these amoebae differentiate into 3 distinct mating types [212,213]. Gamete fusion in *Dictyostelium* is promiscuous, often leading to the formation of multinucleated syncytia due to a lack of mechanisms to prevent fusion between multiple gametes. Over the next few hours, these syncytia split to form binucleate cells prior to fusion of the nuclei and downstream microcyst development [214]. This results in amalgamation of the cytoplasmic contents of multiple individuals following their fusion. Fusion events between more than 2 gametes frequently leads to triparental mitochondrial inheritance, as the haploid progeny often inherits nuclear DNA from 2 parents alongside the mitochondrial genome of a third [215]. While uniparental inheritance is commonly regarded as the universal and preferred mode of mitochondrial and plastid inheritance during mating in many model organisms, less well-studied organisms such as *Dictyostelium* highlight that alternative modes of inheritance do exist and may be more common than first anticipated.

Chlamydomonas—An example of diverse patterns and mechanisms for uniparental inheritance

The unicellular green algae *Prototheca* (which causes protothecosis in humans and dogs) and the closely related *Helicosporidium* (which parasitises insect guts) possess mitochondria and non-photosynthetic plastids, but nothing is known about inheritance of these endosymbiotic organelles. *Chlamydomonas* is a nonpathogenic, well-characterised relative of these pathogens that may provide clues as to the modes and mechanisms for the inheritance of endosymbiotic organelles during sexual reproduction in these pathogens. *Chlamydomonas reinhardtii* displays interesting patterns of mitochondrial and plastid inheritance in which cpDNA is



Protist	Organelles	Species	Sex?	Gametes?	Inheritance pattern?	Mechanisms
Amoebozoa						
Dictyostellium		*D. discoideum*	Yes [214]	Isogamous [214]	Triparental [216]	NA
Chlamydomonadale						
Chlamydomonas		*C. reinhardtii*	Yes [218]	Isogamous [199]	Maternal - (P) Paternal (M) [217]	SD [199-201]
Alveolata						
Apicomplexa		*Plasmodium*	Yes	Anisogamous [227]	Maternal (P+M) [223-226]	E + D [228-229]
		Toxoplasma	Yes	Anisogamous [237]	Maternal (P?) [233]	E (?) [233]
		Eimeria	Yes	Anisogamous [236]	Maternal (P?) [234]	E (?) [234]
		Babesia	Yes	Anisogamous [241,249]	Unknown	Unknown
		Theileria	Yes	Anisogamous [240]	Unknown	Unknown
		Cryptosporidium	Yes	Anisogamous [257]	Unknown	Unknown
		Eugregarines	Yes	Anisogamous [259]	Unknown	Unknown
Metamonada						
Kinetoplastids		*Trypanosoma*	Unknown	Unknown	'Biparental' [124-126]	Unknown
		Leishmania	Unknown	Unknown	'Biparental' [266]	Unknown

Fig 5. Pathogenic protists with known sexualities and better studied organellar inheritance patterns. This figure illustrates the endosymbiotic organelles and presence or absence of an organellar genome within each listed protist. Protists and organelles are hand-painted by Sarah N. Farrell. Organellar genomes are linear or circular as depicted. While many apicomplexans possess both an apicoplast (green) and a classical mitochondrion (yellow), *Cryptosporidium* and the eugregarines instead contain a single mitosome (purple) and no apicoplast. The mitochondrion found in *Trypanosoma* and *Leishmania* (red) harbours kinetoplast DNA comprised of DNA minicircles and maxicircles. (P) = plastid, (M) = mitochondria. Different mechanisms such as selective degradation of organellar DNA (SD), degradation of whole organelle structures (D) and exclusion (E) are used to ensure uniparental inheritance. References are provided in brackets below the text where relevant.

https://doi.org/10.1371/journal.ppat.1012835.g005

inherited from the maternal (mt^+) mating type and mtDNA is inherited from the paternal (mt^-) mating type [216,217]. Within an hour of zygote formation, mt^- cpDNA is selectively degraded by an mt^+-gamete–specific Ca^{2+}-dependent nuclease that is preferentially imported into the mt^- chloroplasts [200]. Early work by Sager and colleagues demonstrated that mt^+ cpDNA was highly methylated, whereas mt^- cpDNA was not [218]. Moreover, a DNA methyltransferase CrMET1 is expressed in mt^+ gametes and targeted to the plastid [200]. Together, this suggests that methylation of mt^+ cpDNA may protect it from digestion. This is supported by additional studies demonstrating that treatment with methylation inhibitors increases the rate of biparental and paternal inheritance in *C. reinhardtii* [219]. However, the link between methylation and maternal inheritance remains controversial, as other studies indicate that selective methylation or hypomethylation of cpDNA does not necessarily correlate with maternal inheritance [220,221]. By contrast, mtDNA from both parents persists in zygotes until the beginning of meiosis in *C. reinhardtii*. After meiosis is induced, the maternal mt^+ mtDNA is selectively degraded, leaving only paternal mt^- mtDNA nucleoids [198,199]. The mechanism for selective degradation of mt^+ mtDNA in the zygote is currently unknown. The difference in timing between the degradation of cpDNA and mtDNA suggests that uniparental inheritance of the respective organelles is controlled by distinct mechanisms in *C. reinhardtii* [198].

The poster child for studying mitochondrial and plastid inheritance in parasitic protists—*Plasmodium*

Mitochondrial and plastid inheritance has been most thoroughly characterised in apicomplexans because their sexual life cycles can be recapitulated experimentally and is often fundamental for disease transmission. All *Plasmodium* species harbour a single mitochondrion and a single non-photosynthetic plastid called the apicoplast. Both organelles contain their own reduced genomes and are believed to follow a uniparental pattern of inheritance [16]. Early genetic cross and DNA blot studies suggested that mitochondria and apicoplasts are maternally inherited in *Plasmodium* [222–224]. Subsequently, maternal inheritance of the mitochondrion has been confirmed in sexual crosses tracking mitochondrion-encoded resistance to the antimalarial drug atovaquone [225]. In *P. falciparum*, maternal inheritance of both organelles is likely implemented by exclusion of apicoplasts and mitochondria from male microgametes (sperm). Imaging revealed that both organelles remain lodged within the residual body of male gametocytes during exflagellation (the process in which microgametes/sperm are produced [226]), suggesting that sperm exclusion mechanisms ensure maternal inheritance in *P. falciparum* [227]. Intriguingly, Stanway and colleagues reported the absence of the apicoplast in male *P. berghei* gametocytes entirely [228]. In this instance, the presence of an additional degradation mechanism suggests that maternal inheritance of the apicoplast may be a more tightly regulated and strict process in *P. berghei* compared to other *Plasmodium* species.

In many organisms that employ selective-degradation or exclusion mechanisms to eliminate organelles, the organellar genome is degraded prior to the elimination of the organelle structure. For example, in the green alga *Bryopsis maxima*, preferential destruction of both paternal plastid DNA and mtDNA occurs during late gametogenesis. The remaining plastid structures are eliminated by lysosome-like organelles 24 to 48 h after mating [229,230]. Similarly, the mitochondrial endonuclease G CSP-6 mediates degradation of paternal mtDNA and accelerates breakdown of empty mitochondria in *C. elegans* [231]. While such organellar genome reductions are yet to be reported in *Plasmodium*, this may serve as an additional mechanism to ensure maternal inheritance of the apicoplast and mitochondrion in these protists, as depicted in Fig 2.

The absence of apicoplasts and apicoplast-specific markers in male gametes also suggests a maternal inheritance pattern for the apicoplast in *Toxoplasma gondii* and *Eimeria tenella* [232,233]. Interestingly, electron microscopy of *Toxoplasma*—and also *Eimeria*—indicates that mitochondria are present in both male gametocytes and gametes [234]. However, studies of these mitochondrial genomes, and how they are inherited, are yet to be done. In future, it would be intriguing to determine whether the mitochondria packaged into the sperm of these anisogamous [235,236] protists are carried into the zygote, or whether they are excluded from the female gamete during fertilisation as observed in the Chinese hamster *Cricetulus griseus* [237]. Excitingly, recent breakthroughs in inducing sexual development in *T. gondii* in mice—thus eliminating the need to harvest sexual forms of the parasite from infected felines—offer a promising and tractable system to study mitochondrion and apicoplast inheritance in this excellent apicomplexan model [238].

Babesia and *Theileria* are apicomplexan parasites responsible for livestock and companion animal diseases throughout much of the world [239,240]. Ultrastructural studies of asexual stage parasites indicate that various *Babesia* species possess 1 or 2 mitochondria and a single apicoplast [241,242]. However, the morphology and behaviour of these organelles, particularly during sexual stages of development, is limited to preliminary electron microscopy data for *Babesia* gametocytes and no immunofluorescence, or live cell microscopy studies are available [243]. In *Theileria*, ultrastructural studies have identified the presence of mitochondria in various life stages, and genome sequencing of *T. parva* and *T. annulate* confirms the presence of the apicoplast in these apicomplexan parasites [35,244]. Surprisingly, the *Theileria* apicoplast is yet to be visualised through electron microscopy or other imaging techniques and is only known through sequencing of the apicoplast genome [245,246]. Overall, very little is known about the inheritance or mechanisms for inheritance of the apicoplast and mitochondria in these organisms, although recent developments in methods to recover *Babesia* gametes in vitro [247] suggests that investigating mitochondrial and apicoplast inheritance in this apicomplexan may soon be possible.

Cryptosporidium is the "black sheep" of the apicomplexan family as it has lost the apicoplast and contains a genome-less MRO termed the mitosome [248,249]. Intriguingly, while select gastroinstestine-inhabiting cryptosporidians such as *C. muris* and *C. andersonii* possess a complete set of tricarboxylic acid (TCA) cycle associated enzymes and a truncated electron transport chain, these are lost entirely from other intestinal-type species such as *C. parvum* and *C. hominis* [248,250–252]. Ultrastructural analysis of *C. muris* revealed a double-membraned mitosome with highly developed cristae like *T. gondii* [253]. By contrast, the mitosomes of *C. parvum* and *C. hominis* are reduced in size and possess atypical cristae [248,254]. Together, this highlights that diversity in mitosome structure and function between different cryptosporidians is most likely based on their unique metabolic requirements and the host environments they occupy. Mitosome inheritance during sexual reproduction is yet to be studied in *Cryptosporidium* species. Sex studies in *Cryptosporidium* have recently become tractable [255], and it will be fascinating to dissect whether the mitosome is uniparentally inherited like the mitochondrion in *Plasmodium*, or if such inheritance patterns become unnecessary in the absence of organellar genomes.

Eugregarines are another group of anisogamous apicomplexans that lack an apicoplast, but these enigmatic parasites are understudied compared to other apicomplexan groups [256,257]. Distinctive mitochondria with tubular cristae have been identified multiple times in gregarines [256,258–262]. Despite this, transcriptomics revealed that mitochondrial metabolism is reduced to differing degrees in many gregarines, with most lacking respiratory complexes III and IV, and some lacking the ETC and TCA cycle entirely [39,40]. As described for *Cryptosporidium*, gregarines present another exciting opportunity to investigate differences in

mitochondrial inheritance patterns and/or mechanisms between apicomplexans that do or do not contain a mitochondrial genome and lack an apicoplast.

The overarching theme among apicomplexans is that while most possess an apicoplast and mitochondrion throughout their complex life cycles, patterns of inheritance and mechanisms used to facilitate their inheritance are yet to be properly explored in all but a few tractable models, and some intriguing instances of organelle and/or organelle genome loss pose fascinating scenarios of possible inheritance modes.

Euglenozoans with peculiar mitochondria and mitochondrial inheritance patterns

The parasitic protists *Trypanosoma* and *Leishmania* contain a single mitochondrion that harbours an unusual network of circular DNA termed the kinetoplast. Kinetoplast DNA (kDNA) is positioned adjacent to the basal body of the single flagellum and comprises an interconnected network of approximately 25 to 50 maxicircles (23 kb in size) and several thousand minicircles (1 kb in size) that together form a disc-like structure [52,53]. Maxicircles encode 20 genes, many of which are cryptogenes whose primary mRNA transcripts require editing to become translatable. Minicircles encode guide RNAs that mediate editing of these maxicircle genes [263]. Interestingly, in *Trypanosoma brucei*, lab-derived hybrid clones contained heterogeneous kDNA networks consisting of minicircles from both parents but maxicircles from a single parent, indicating some form of partial uniparental inheritance [124,125]. It is postulated that the whole kDNA network is initially biparentally inherited but homogeneity of maxicircles is quickly achieved through random DNA segregation during subsequent rounds of mitotic division [124–126]. Conversely, minicircles are not randomly segregated between daughter networks, maintaining the heterogeneity of the originally inherited network [124–126]. Similar patterns of biparental minicircle and maxicircle are also observed in *Leishmania* [264]. In future, it would be intriguing to observe kinetoplast inheritance in these protists during their natural sexual reproduction.

Final remarks and future directions

Overall, staggeringly little is known about mitochondrial and plastid inheritance during sexual reproduction, or the mechanisms used to achieve this inheritance, in the pathogenic protists discussed in this review. This is largely attributed to our current inability to culture many of these organisms *in vitro* or to easily induce their sexual development in the lab. However, many genetic and molecular biology tools created to study the biogenesis, dynamics, and segregation of mitochondria and plastids in apicomplexan parasites could be applied in other protists to study mitochondrial and plastid inheritance during mating. Fluorescent protein markers have been used extensively in *Plasmodium* and other apicomplexans such as *Toxoplasma gondii* and *Eimeria tenella* to track organelles across the life cycle, and setting up laboratory sexual crosses should now be possible to explore inheritance [247,265–271]. In instances where *in vitro* induction of sexual development or genetic modification is not possible, well-validated live cell mitochondrial stains with different colours such as MitoTracker could be utilised to localise and track the inheritance of mitochondria in gametes, zygotes, and post-zygotic stages.

Ultimately, an understanding of the sex lives of these parasites is fundamental to the study of mitochondrial and plastid inheritance during mating, particularly in the context of reduced organelles such as mitosomes and hydrogenosomes. *Toxoplasma* and *Cryptosporidium* seem ripe for study of organelle inheritance, with the latter presenting an intriguing test case of whether a likely ancestral mechanism of maternal inheritance persists or has been rendered

defunct with the loss of the apicoplast and no genome in the mitosome. Protistologists have a lot to learn about the inheritance of these important organelles, so many of which are useful drug targets.

Author Contributions

Conceptualization: Sophie L. Collier, Christopher D. Goodman, Geoffrey I. McFadden.

Formal analysis: Sophie L. Collier.

Funding acquisition: Geoffrey I. McFadden.

Investigation: Sophie L. Collier.

Visualization: Sarah N. Farrell.

Writing – original draft: Sophie L. Collier, Sarah N. Farrell.

Writing – review & editing: Sophie L. Collier, Sarah N. Farrell, Christopher D. Goodman, Geoffrey I. McFadden.

References

1. Karnkowska A, Vacek V, Zubáčová Z, Treitli SC, Petrželková R, Eme L, et al. A Eukaryote without a Mitochondrial Organelle. Curr Biol. 2016; 26:1274–1284. https://doi.org/10.1016/j.cub.2016.03.053 PMID: 27185558
2. Sagan L. On the origin of mitosing cells. J Theor Biol. 1967; 14:225–274. https://doi.org/10.1016/0022-5193(67)90079-3 PMID: 11541392
3. Martin W, Müller M. The hydrogen hypothesis for the first eukaryote. Nature. 1998; 392:37–41. https://doi.org/10.1038/32096 PMID: 9510246
4. McFadden GI. Primary and secondary endosymbiosis and the origin of plastids. J Phycol. 2001; 37:951–959. https://doi.org/10.1046/j.1529-8817.2001.01126.x
5. Birky CW Jr. The Inheritance of Genes in Mitochondria and Chloroplasts: Laws, Mechanisms, and Models. Annu Rev Genet. 2001; 35:125–148. https://doi.org/10.1146/annurev.genet.35.102401.090231 PMID: 11700280
6. Keeling PJ. The endosymbiotic origin, diversification and fate of plastids. Philos Trans R Soc Lond B Biol Sci. 2010; 365:729–748. https://doi.org/10.1098/rstb.2009.0103 PMID: 20124341
7. Kalanon M, McFadden GI. Malaria, *Plasmodium falciparum* and its apicoplast. Biochem Soc Trans. 2010; 38:775–782. https://doi.org/10.1042/BST0380775 PMID: 20491664
8. Köhler S, Delwiche CF, Denny PW, Tilney LG, Webster P, Wilson RJM, et al. A Plastid of Probable Green Algal Origin in Apicomplexan Parasites. Science. 1997; 275:1485–1489. https://doi.org/10.1126/science.275.5305.1485 PMID: 9045615
9. Gagat P, Mackiewicz D, Mackiewicz P. Peculiarities within peculiarities–dinoflagellates and their mitochondrial genomes. Mitochondrial DNA Part B. 2017; 2:191–195. https://doi.org/10.1080/23802359.2017.1307699 PMID: 33473765
10. Waller RF, Jackson CJ. Dinoflagellate mitochondrial genomes: Stretching the rules of molecular biology. Bioessays. 2009; 31:237–245. https://doi.org/10.1002/bies.200800164 PMID: 19204978
11. Berná L, Rego N, Francia ME. The Elusive Mitochondrial Genomes of Apicomplexa: Where Are We Now? Front Microbiol. 2021; 12. https://doi.org/10.3389/fmicb.2021.751775 PMID: 34721355
12. Tang L, Tam NFY, Lam W, Lee TCH, Xu SJL, Lee CL, et al. Interpreting the complexities of the plastid genome in dinoflagellates: a mini-review of recent advances. Plant Mol Biol. 2024; 114. https://doi.org/10.1007/s11103-024-01511-3 PMID: 39432142
13. Howe CJ, Nisbet RER, Barbrook AC. The remarkable chloroplast genome of dinoflagellates. J Exp Bot. 2008; 59:1035–1045. https://doi.org/10.1093/jxb/erm292 PMID: 18319241
14. Wisecaver JH, Hackett JD. Dinoflagellate genome evolution. Annu Rev Microbiol. 2011; 65:369–387. https://doi.org/10.1146/annurev-micro-090110-102841 PMID: 21682644
15. Matsuo E, Morita K, Nakayama T, Yazaki E, Sarai C, Takahashi K, et al. Comparative Plastid Genomics of Green-Colored Dinoflagellates Unveils Parallel Genome Compaction and RNA Editing. Front Plant Sci. 2022; 13. https://doi.org/10.3389/fpls.2022.918543 PMID: 35898209

16. Wilson RJM, Williamson DH. Extrachromosomal DNA in the Apicomplexa. Microbiol Mol Biol Rev. 1997; 61:1–16. https://doi.org/10.1128/mmbr.61.1.1-16.1997 PMID: 9106361

17. Vaidya AB, Akella R, Suplick K. Sequences similar to genes for two mitochondrial proteins and portions of ribosomal RNA in tandemly arrayed 6-kilobase-pair DNA of a malarial parasite. Mol Biochem Parasitol. 1989; 35:97–108. https://doi.org/10.1016/0166-6851(89)90112-6 PMID: 2549417

18. Wilson I, Denny PW, Preiser PR, Rangachari K, Roberts K, Roy A, et al. Complete Gene Map of the Plastid-like DNA of the Malaria Parasite *Plasmodium falciparum*. J Mol Biol. 1996; 261:155–172. https://doi.org/10.1006/jmbi.1996.0449 PMID: 8757284

19. Arisue N, Hashimoto T. Phylogeny and evolution of apicoplasts and apicomplexan parasites. Parasitol Int. 2015; 64:254–259. https://doi.org/10.1016/j.parint.2014.10.005 PMID: 25451217

20. Namasivayam S, Baptista RP, Xiao W, Hall EM, Doggett JS, Troell K, et al. A novel fragmented mitochondrial genome in the protist pathogen *Toxoplasma gondii* and related tissue coccidia. Genome Res. 2021; 31:852–865. https://doi.org/10.1101/GR.266403.120 PMID: 33906963

21. Namasivayam S, Sun C, Bah AB, Oberstaller J, Pierre-Louis E, Etheridge RD, et al. Massive invasion of organellar DNA drives nuclear genome evolution in *Toxoplasma*. Proc Natl Acad Sci U S A. 2023; 120. https://doi.org/10.1073/pnas.2308569120 PMID: 37917792

22. Williamson DH, Denny PW, Moore PW, Sato S, Mccready S, Wilson RJMI. The *in vivo* conformation of the plastid DNA of *Toxoplasma gondii*: Implications for replication. J Mol Biol. 2001; 306:159–168. https://doi.org/10.1006/jmbi.2000.4385 PMID: 11237591

23. Tian SQ, Cui P, Fang SF, Liu GH, Wang CR, Zhu XQ. The complete mitochondrial genome sequence of *Eimeria magna* (Apicomplexa: Coccidia). Mitochondrial DNA. 2015; 26:714–715. https://doi.org/10.3109/19401736.2013.843088 PMID: 24328820

24. Lin RQ, Qiu LL, Liu GH, Wu XY, Weng YB, Xie WQ, et al. Characterization of the complete mitochondrial genomes of five *Eimeria species* from domestic chickens. Gene. 2011; 480:28–33. https://doi.org/10.1016/j.gene.2011.03.004 PMID: 21402132

25. Cai X, Fuller AL, McDougald LR, Zhu G. Apicoplast genome of the coccidian *Eimeria tenella*. Gene. 2003; 321:39–46. https://doi.org/10.1016/j.gene.2003.08.008 PMID: 14636990

26. Brayton KA, Lau AOT, Herndon DR, Hannick L, Kappmeyer LS, Berens SJ, et al. Genome sequence of *Babesia bovis* and comparative analysis of apicomplexan hemoprotozoa. PLoS Pathog. 2007; 3:1401–1413. https://doi.org/10.1371/journal.ppat.0030148 PMID: 17953480

27. Schelp C, Böse R, Friedhoff KT. Demonstration of extrachromosmal DNA from *Babesia equi* merozoites. Parasitol Res. 1992; 78:707–708. https://doi.org/10.1007/BF00931526 PMID: 1480611

28. Hotzel I, Kabakoff R, Ozaki LS. Small extrachromosomal nucleic acid segments in protozoan parasites. Vet Parasitol. 1995; 57:57–60. https://doi.org/10.1016/0304-4017(94)03110-i PMID: 7597793

29. Hikosaka K, Kita K, Tanabe K. Diversity of mitochondrial genome structure in the phylum Apicomplexa. Mol Biochem Parasitol. 2013; 188:26–33. https://doi.org/10.1016/j.molbiopara.2013.02.006 PMID: 23466751

30. Hikosaka K, Watanabe YI, Tsuji N, Kita K, Kishine H, Arisue N, et al. Divergence of the Mitochondrial Genome Structure in the Apicomplexan Parasites, *Babesia and Theileria*. Mol Biol Evol. 2010; 27:1107–1116. https://doi.org/10.1093/molbev/msp320 PMID: 20034997

31. Wang X, Wang J, Liu J, Liu A, He X, Xu J, et al. Comparative analysis of apicoplast genomes of *Babesia* infective to small ruminants in China. Parasit Vectors. 2019; 12. https://doi.org/10.1186/s13071-019-3581-x PMID: 31234937

32. Wang T, Guan G, Korhonen PK, Koehler AV, Hall RS, Young ND, et al. The apicoplast genomes of two taxonomic units of *Babesia* from sheep. Vet Parasitol. 2017; 233:123–128. https://doi.org/10.1016/j.vetpar.2016.11.004 PMID: 27916258

33. Garg A, Stein A, Zhao W, Dwivedi A, Frutos R, Cornillot E, et al. Sequence and annotation of the apicoplast genome of the human pathogen *Babesia microti*. PLoS ONE. 2014; 9. https://doi.org/10.1371/journal.pone.0107939 PMID: 25280009

34. Huang Y, He L, Hu J, He P, He J, Yu L, et al. Characterization and annotation of *Babesia orientalis* apicoplast genome. Parasit Vectors. 2015; 8. https://doi.org/10.1186/s13071-015-1158-x PMID: 26474853

35. Gardner MJ, Bishop R, Shah T, De Villiers EP, Carlton JM, Hall N, et al. Genome Sequence of *Theileria parva*, a Bovine Pathogen That Transforms Lymphocytes. Science (1979). 2005; 309:134–137. https://doi.org/10.1126/science.1110439 PMID: 15994558

36. Yam J, Bogema DR, Micallef ML, Djordjevic SP, Jenkins C. Complete Genomes of *Theileria orientalis* Chitose and Buffeli Genotypes Reveal within Species Translocations and Differences in ABC Transporter Content. Pathogens. 2022; 11. https://doi.org/10.3390/pathogens11070801 PMID: 35890045

37. Kappmeyer LS, Thiagarajan M, Herndon DR, Ramsay JD, Caler E, Djikeng A, et al. Comparative genomic analysis and phylogenetic position of *Theileria equi*. BMC Genomics. 2012; 13:1–13. https://doi.org/10.1186/1471-2164-13-603 PMID: 23137308

38. Hayashida K, Hara Y, Abe T, Chisato Yamasaki CY, Toyoda A, Kosuge T, et al. Comparative genome analysis of three eukaryotic parasites with differing abilities to transform leukocytes reveals key mediators of *theileria*-induced leukocyte transformation. MBio. 2012; 3. https://doi.org/10.1128/mBio.00204-12 PMID: 22951932

39. Mathur V, Wakeman KC, Keeling PJ. Parallel functional reduction in the mitochondria of apicomplexan parasites. Curr Biol. 2021; 31:2920–2928.e4. https://doi.org/10.1016/j.cub.2021.04.028 PMID: 33974849

40. Salomaki ED, Terpis KX, Rueckert S, Kotyk M, Varadínová ZK, Čepička I, et al. Gregarine single-cell transcriptomics reveals differential mitochondrial remodeling and adaptation in apicomplexans. BMC Biol. 2021; 19. https://doi.org/10.1186/s12915-021-01007-2 PMID: 33863338

41. Boxma B, de Graaf RM, van der Staay GWM, van Alen TA, Ricard G, Gabaldón T, et al. An anaerobic mitochondrion that produces hydrogen. Nature. 2005; 434:74–79. https://doi.org/10.1038/nature03343 PMID: 15744302

42. de Graaf RM, Ricard G, van Alen TA, Duarte I, Dutilh BE, Burgtorf C, et al. The Organellar Genome and Metabolic Potential of the Hydrogen-Producing Mitochondrion of *Nyctotherus ovalis*. Mol Biol Evol. 2011; 28:2379–2391. https://doi.org/10.1093/molbev/msr059 PMID: 21378103

43. van Hoek AHAM, Akhmanova AS, Huynen MA, Hackstein JHP. A Mitochondrial Ancestry of the Hydrogenosomes of *Nyctotherus ovalis*. Mol Biol Evol. 2000; 17:202–206. https://doi.org/10.1093/oxfordjournals.molbev.a026234 PMID: 10666720

44. Tsukii Y. Evolution of mitochondrial DNA in *Paramecium caudatum*. Jpn J Genet. 1994; 69:307–319. https://doi.org/10.1266/jjg.69.307

45. Pritchard AE, Seilhamer JJ, Mahalingam R, Sable CL, Venuti SE, Cummings DJ. Nucleotide sequence of the mitochondrial genome of *Paramecium*. Nucleic Acids Res. 1990; 18. https://doi.org/10.1093/nar/18.1.173 PMID: 2308823

46. Aurongzeb M, Rashid Y, Naqvi SHA, Malik HMT, Azim MK, Hassan SS, et al. Insights into genome evolution, pan-genome, and phylogenetic implication through mitochondrial genome sequence of *Naegleria fowleri* species. Sci Rep. 2022; 12. https://doi.org/10.1038/s41598-022-17006-4 PMID: 35909191

47. Spencer DF, Gray MW. Ribosomal RNA genes in *Euglena gracilis* mitochondrial DNA: fragmented genes in a seemingly fragmented genome. Mol Genet Genomics. 2011; 285:19–31. https://doi.org/10.1007/s00438-010-0585-9 PMID: 20978909

48. Ebenezer TGE, Carrington M, Lebert M, Kelly S, Field MC. *Euglena gracilis* genome and transcriptome: Organelles, nuclear genome assembly strategies and initial features. Advances in Experimental Medicine and Biology. Springer New York LLC; 2017. p. 125–140. https://doi.org/10.1007/978-3-319-54910-1_7 PMID: 28429320

49. Dobáková E, Flegontov P, Skalický T, Lukeš J. Unexpectedly streamlined mitochondrial genome of the euglenozoan *Euglena gracilis*. Genome Biol Evol. 2015; 7:3358–3367. https://doi.org/10.1093/gbe/evv229 PMID: 26590215

50. Hallick RB, Ling H, Drager RG, Favreau MR, Monfort A, Orsat B, et al. Complete sequence of *Euglena gracilis* chloroplast DNA. Nucleic Acids Res. 1993; 21:3537–3544. https://doi.org/10.1093/nar/21.15.3537 PMID: 8346031

51. Camacho E, Rastrojo A, Sanchiz Á, González-De La Fuente S, Aguado B, Requena JM. *Leishmania* mitochondrial genomes: Maxicircle structure and heterogeneity of minicircles. Genes (Basel). 2019; 10. https://doi.org/10.3390/genes10100758 PMID: 31561572

52. Shapiro TA. Kinetoplast DNA maxicircles: Networks within networks. Proc Natl Acad Sci U S A. 1993; 90:7809–7813. https://doi.org/10.1073/pnas.90.16.7809 PMID: 8395055

53. Lukeš J, Guilbride DL, Votýpka J, Zíková A, Benne R, Englund PT. Kinetoplast DNA network: Evolution of an improbable structure. Eukaryot Cell. 2002; 1:495–502. https://doi.org/10.1128/EC.1.4.495-502.2002 PMID: 12455998

54. Callejas-Hernández F, Herreros-Cabello A, del Moral-Salmoral J, Fresno M, Gironès N. The Complete Mitochondrial DNA of *Trypanosoma cruzi*: Maxicircles and Minicircles. Front Cell Infect Microbiol. 2021; 11. https://doi.org/10.3389/fcimb.2021.672448 PMID: 34268138

55. Jedelský PL, Doležal P, Rada P, Pyrih J, Šmíd O, Hrdý I, et al. The minimal proteome in the reduced mitochondrion of the parasitic protist *Giardia intestinalis*. PLoS ONE. 2011; 6:e17285. https://doi.org/10.1371/journal.pone.0017285 PMID: 21390322

56. Lindmark DG, Shio H. Hydrogenosomes in *Trichomonas vaginalis*. J Parasitol. 1975; 61:552–554. https://doi.org/10.2307/3283598

57. Ogawa S, Yoshino R, Angata K, Iwamoto M, Pi M, Kuroe K, et al. The mitochondrial DNA of *Dictyostelium discoideum*: complete sequence, gene content and genome organization. Mol Gen Genet. 2000; 263:514–519. https://doi.org/10.1007/pl00008685 PMID: 10821186

58. Kazama M, Ogiwara S, Makiuchi T, Yoshida K, Nakada-Tsukui K, Nozaki T, et al. Behavior of DNA-lacking mitochondria in *Entamoeba histolytica* revealed by organelle transplant. Sci Rep. 2017; 7. https://doi.org/10.1038/srep44273 PMID: 28287148

59. Jeffrey SW, Sielicki M, Haxo FT. Chloroplast Pigment Patterns in Dinoflagellates. J Phycol. 1975; 11:374–384. https://doi.org/10.1111/j.1529-8817.1975.tb02799.x

60. Zapata M, Fraga S, Rodríguez F, Garrido JL. Pigment-based chloroplast types in dinoflagellates. Mar Ecol Prog Ser. 2012; 465:33–52. https://doi.org/10.3354/meps09879

61. MacPherson AN, Hiller RG. Light-Harvesting Systems in Chlorophyll c-Containing Algae. In: Green BR, Parson WW, editors. Light-Harvesting Antennas in Photosynthesis Advances in Photosynthesis and Respiration. Dordrecht: Springer; 2003.

62. Dorrell RG, Howe CJ. Integration of plastids with their hosts: Lessons learned from dinoflagellates. Proc Natl Acad Sci U S A. 2015; 112:10247–10254. https://doi.org/10.1073/pnas.1421380112 PMID: 25995366

63. Janouškovec J, Tikhonenkov DV, Burki F, Howe AT, Kolísko M, Mylnikov AP, et al. Factors mediating plastid dependency and the origins of parasitism in apicomplexans and their close relatives. Proc Natl Acad Sci U S A. 2015; 112:10200–10207. https://doi.org/10.1073/pnas.1423790112 PMID: 25717057

64. Janouškovec J, Horák A, Oborník M, Lukeš J, Keeling PJ. A common red algal origin of the apicomplexan, dinoflagellate, and heterokont plastids. Proc Natl Acad Sci U S A. 2010; 107:10949–10954. https://doi.org/10.1073/pnas.1003335107 PMID: 20534454

65. Dorrell RG, Howe CJ. Functional remodeling of RNA processing in replacement chloroplasts by pathways retained from their predecessors. Proc Natl Acad Sci U S A. 2012; 109:18879–18884. https://doi.org/10.1073/pnas.1212270109 PMID: 23112181

66. Cavalier-Smith T. Principles of protein and lipid targeting in secondary symbiogenesis: Euglenoid, dinoflagellate, and sporozoan plastid origins and the eukaryote family tree. J Eukaryot Microbiol. 1999; 46:347–366. https://doi.org/10.1111/j.1550-7408.1999.tb04614.x PMID: 18092388

67. Delwiche CF. Tracing the Thread of Plastid Diversity through the Tapestry of Life. Am Nat. 1999; 154: S164–S177. https://doi.org/10.1086/303291 PMID: 10527925

68. Bhattacharya D, Yoon HS, Hackett JD. Photosynthetic eukaryotes unite: Endosymbiosis connects the dots. Bioessays. 2004; 26:50–60. https://doi.org/10.1002/bies.10376 PMID: 14696040

69. Waller RF, Kořený L. Plastid complexity in dinoflagellates: a picture of gains, losses, replacements and revisions. Adv Bot Res. 2017; 84:105–143. https://doi.org/10.17863/CAM.16938

70. Jeong HJ, du Yoo Y, Kim JS, Seong KA, Kang NS, Kim TH. Growth, feeding and ecological roles of the mixotrophic and heterotrophic dinoflagellates in marine planktonic food webs. Ocean Sci J. 2010; 45:65–91. https://doi.org/10.1007/s12601-010-0007-2

71. Gornik SG, Febrimarsa, Cassin AM, MacRae JI, Ramaprasad A, Rchiad Z, et al. Endosymbiosis undone by stepwise elimination of the plastid in a parasitic dinoflagellate. Proc Natl Acad Sci U S A. 2015; 112:5767–5772. https://doi.org/10.1073/pnas.1423400112 PMID: 25902514

72. Sibbald SJ, Archibald JM. Genomic insights into plastid evolution. Genome Biol Evol. 2020; 12:978–990. https://doi.org/10.1093/gbe/evaa096 PMID: 32402068

73. Hannaert V, Saavedra E, Duffieux F, Szikora J-P, Rigden DJ, Michels PAM, et al. Plant-like traits associated with metabolism of *Trypanosoma* parasites. Proc Natl Acad Sci U S A. 2003; 100:1067–1071. https://doi.org/10.1073/pnas.0335769100 PMID: 12552132

74. Husnik F, Tashyreva D, Boscaro V, George EE, Lukeš J, Keeling PJ. Bacterial and archaeal symbioses with protists. Curr Biol. 2021; 31:R862–R877. https://doi.org/10.1016/j.cub.2021.05.049 PMID: 34256922

75. Keeling PJ, Burki F. Progress towards the Tree of Eukaryotes. Curr Biol. 2019; 29:PR808–R817. https://doi.org/10.1016/j.cub.2019.07.031 PMID: 31430481

76. Greiner S, Sobanski J, Bock R. Why are most organelle genomes transmitted maternally? Bioessays. 2015; 37:80–94. https://doi.org/10.1002/bies.201400110 PMID: 25302405

77. Xu J. The inheritance of organelle genes and genomes: Patterns and mechanisms. Genome. 2005; 48:951–957. https://doi.org/10.1139/g05-082 PMID: 16391664

78. Ladoukakis ED, Zouros E. Evolution and inheritance of animal mitochondrial DNA: Rules and exceptions. J Biol Res (Thessalon). 2017; 24:1–7. https://doi.org/10.1186/s40709-017-0060-4 PMID: 28164041

79. Kuroiwa T. Review of cytological studies on cellular and molecular mechanisms of uniparental (maternal or paternal) inheritance of plastid and mitochondrial genomes induced by active digestion of organelle nuclei (nucleoids). J Plant Res. 2010; 123:207–230. https://doi.org/10.1007/s10265-009-0306-9 PMID: 20145972

80. Sakamoto W, Miyagishima S, Jarvis P. Chloroplast Biogenesis: Control of Plastid Development, Protein Import, Division and Inheritance. Arabidopsis Book. 2008; 6:1–30. https://doi.org/10.1199/tab.0110 PMID: 22303235

81. Sato M, Sato K. Maternal inheritance of mitochondrial DNA by diverse mechanisms to eliminate paternal mitochondrial DNA. Biochim Biophys Acta Mol Cell Res. 2013; 1833:1979–1984. https://doi.org/10.1016/j.bbamcr.2013.03.010 PMID: 23524114

82. Hastings IM. Population genetic aspects of deleterious cytoplasmic genomes and their effect on the evolution of sexual reproduction. Genet Res. 1992; 59:215–225. https://doi.org/10.1017/s0016672300030500 PMID: 1511870

83. Hoekstra RF. Evolutionary origin and consequences of uniparental mitochondrial inheritance. Hum Reprod. 2000; 15:102–111. https://doi.org/10.1093/humrep/15.suppl_2.102 PMID: 11041518

84. Radzvilavicius A. Beyond the "selfish mitochondrion" theory of uniparental inheritance: A unified theory based on mutational variance redistribution. Bioessays. 2021; 43. https://doi.org/10.1002/bies.202100009 PMID: 33729620

85. Zimorski V, Ku C, Martin WF, Gould SB. Endosymbiotic theory for organelle origins. Curr Opin Microbiol. 2014; 22:38–48. https://doi.org/10.1016/j.mib.2014.09.008 PMID: 25306530

86. Hadjivasiliou Z, Pomiankowski A, Seymour RM, Lane N. Selection for mitonuclear co-adaptation could favour the evolution of two sexes. Proc Biol Sci. 2012; 279:1865–1872. https://doi.org/10.1098/rspb.2011.1871 PMID: 22158961

87. Bock R, Timmis JN. Reconstructing evolution: Gene transfer from plastids to the nucleus. Bioessays. 2008; 30:556–566. https://doi.org/10.1002/bies.20761 PMID: 18478535

88. Birky CW. Uniparental inheritance of mitochondrial and chloroplast genes: Mechanisms and evolution. Proc Natl Acad Sci U S A. 1995; 92:11331–11338. https://doi.org/10.1073/pnas.92.25.11331 PMID: 8524780

89. Mishra P, Chan DC. Mitochondrial dynamics and inheritance during cell division, development and disease. Nat Rev Mol Cell Biol. Nature Publishing Group; 2014. p. 634–646. https://doi.org/10.1038/nrm3877 PMID: 25237825

90. Breton S, Stewart DT. Atypical mitochondrial inheritance patterns in eukaryotes. Genome Canadian Science Publishing; 2015. p. 423–431. https://doi.org/10.1139/gen-2015-0090 PMID: 26501689

91. Lynch M, Koskella B, Schaack S. Mutation Pressure and the Evolution of Organelle Genomic Architecture. Science (1979). 2006; 311:1727–1730. https://doi.org/10.1126/science.1118884 PMID: 16556832

92. Neiman M, Taylor DR. The causes of mutation accumulation in mitochondrial genomes. Proc Biol Sci. 2009; 276:1201–1209. https://doi.org/10.1098/rspb.2008.1758 PMID: 19203921

93. Bogenhagen DF. Repair of mtDNA in Vertebrates. Am J Hum Genet. 1999; 64:1276–1281. https://doi.org/10.1086/302392 PMID: 10205257

94. Muller HJ. The relation of recombination to mutational advance. Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis. 1964; 1:2–9. https://doi.org/10.1016/0027-5107(64)90047-8 PMID: 14195748

95. Barr CM, Neiman M, Taylor DR. Inheritance and recombination of mitochondrial genomes in plants, fungi and animals. New Phytol. 2005; 168:39–50. https://doi.org/10.1111/j.1469-8137.2005.01492.x PMID: 16159319

96. Birky CW. Uniparental inheritance of organelle genes. Curr Biol. 2008; 18:R692–R695. https://doi.org/10.1016/j.cub.2008.06.049 PMID: 18727899

97. Parakatselaki ME, Ladoukakis ED. mtDNA heteroplasmy: Origin, detection, significance, and evolutionary consequences. Life. 2021; 11:633. https://doi.org/10.3390/life11070633 PMID: 34209862

98. Rokas A, Abbot P. Harnessing genomics for evolutionary insights. Trends Ecol Evol. 2009; 24:192–200. https://doi.org/10.1016/j.tree.2008.11.004 PMID: 19201503

99. Ramsey AJ, Mandel JR. When one genome is not enough: Organellar heteroplasmy in plants. Annu Plant Rev Online. 2019; 2:619–658. https://doi.org/10.1002/9781119312994.apr0616

100. Mccauley DE. Paternal leakage, heteroplasmy, and the evolution of plant mitochondrial genomes. New Phytol. 2013. p. 966–977. https://doi.org/10.1111/nph.12431 PMID: 23952142

101. Kondo R, Satta Y, Matsuura ET, Takahatat N, Chigusa SI. Incomplete Maternal Transmission of Mitochondrial DNA in *Drosophila*. Genetics. 1990; 126:657–663. https://doi.org/10.1093/genetics/126.3.657 PMID: 2249764

102. Fontaine KM, Cooley JR, Simon C. Evidence for paternal leakage in hybrid periodical cicadas (Hemiptera: *Magicicada* spp.). PLoS ONE. 2007; 2:e892. https://doi.org/10.1371/journal.pone.0000892 PMID: 17849021

103. Gyllensten U, Whartont D, Josefsson A, Wilsont AC. Paternal inheritance of mitochondrial DNA in mice. Nature. 1991; 352:255–257. https://doi.org/10.1038/352255a0 PMID: 1857422

104. Shitara H, Hayashi J-I, Takahama S, Kaneda H, Yonekawa H. Maternal Inheritance of Mouse mtDNA in Interspecific Hybrids: Segregation of the Leaked Paternal mtDNA Followed by the Prevention of Subsequent Paternal Leakage. Genetics. 1998; 148:851–857. https://doi.org/10.1093/genetics/148.2.851 PMID: 9504930

105. Zouros E, Ball AO, Saavedra C, Freeman KR. An unusual type of mitochondrial DNA inheritance in the blue mussel *Mytilus*. Genetics. 1994; 91:7463–7467. https://doi.org/10.1073/pnas.91.16.7463 PMID: 8052604

106. Ramon PD, Secor CL, Hilbish TJ. The Effects of Natural Hybridization on the Regulation of Doubly Uniparental mtDNA Inheritance in Blue Mussels (*Mytilus* spp.). Genetics. 1996; 144:241–248. https://doi.org/10.1093/genetics/144.1.241 PMID: 8878689

107. Vilaça ST, Maroso F, Lara P, de Thoisy B, Chevallier D, Arantes LS, et al. Evidence of backcross inviability and mitochondrial DNA paternal leakage in sea turtle hybrids. Mol Ecol. 2023; 32:628–643. https://doi.org/10.1111/mec.16773 PMID: 36336814

108. Hoolahan AH, Blok VC, Gibson T, Dowton M. Paternal leakage of mitochondrial DNA in experimental crosses of populations of the potato cyst nematode *Globodera pallida*. Genetica. 2011; 139:1509–1519. https://doi.org/10.1007/s10709-012-9650-0 PMID: 22555855

109. Wagner DB, Carlson MR, Yanchuk AD. Paternal leakage of mitochondrial DNA in *Pinus*. Theor Appl Genet. 1991; 82:510–514. https://doi.org/10.1007/BF00588607 PMID: 24213270

110. Pearl SA, Welch ME, McCauley DE. Mitochondrial heteroplasmy and paternal leakage in natural populations of *Silene vulgaris*, a gynodioecious plant. Mol Biol Evol. 2009; 26:537–545. https://doi.org/10.1093/molbev/msn273 PMID: 19033259

111. Kvist L, Martens J, Nazarenko AA, Orell M. Paternal leakage of mitochondrial DNA in the great tit (*Parus major*). Mol Biol Evol. 2003; 20:243–247. https://doi.org/10.1093/molbev/msg025 PMID: 12598691

112. Jaramillo-Correa JP, Bousquet J. Mitochondrial genome recombination in the zone of contact between two hybridizing conifers. Genetics. 2005; 171:1951–1962. https://doi.org/10.1534/genetics.105.042770 PMID: 16118197

113. Arunkumar KP, Metta M, Nagaraju J. Molecular phylogeny of silkmoths reveals the origin of domesticated silkmoth, *Bombyx mori* from Chinese *Bombyx mandarina* and paternal inheritance of *Antheraea proylei* mitochondrial DNA. Mol Phylogenet Evol. 2006; 40:419–427. https://doi.org/10.1016/j.ympev.2006.02.023 PMID: 16644243

114. Xu J, Yan Z, Guo H. Divergence, hybridization, and recombination in the mitochondrial genome of the human pathogenic yeast *Cryptococcus gattii*. Mol Ecol. 2009; 18:2628–2642. https://doi.org/10.1111/j.1365-294X.2009.04227.x PMID: 19457185

115. Azhagiri AK, Maliga P. Exceptional paternal inheritance of plastids in *Arabidopsis* suggests that low-frequency leakage of plastids via pollen may be universal in plants. Plant J. 2007; 52:817–823. https://doi.org/10.1111/j.1365-313X.2007.03278.x PMID: 17931353

116. Postel Z, Van Rossum F, Godé C, Schmitt E, Touzet P. Paternal leakage of plastids rescues inter-lineage hybrids in *Silene nutans*. Ann Bot. 2023. https://doi.org/10.1093/aob/mcad196 PMID: 38141228

117. Ruf S, Karcher D, Bock R. Determining the transgene containment level provided by chloroplast transformation. Proc Natl Acad Sci U S A. 2007; 104:6998–7002. https://doi.org/10.1073/pnas.0700008104 PMID: 17420459

118. Metzlaff M, Pohlheim F, Börner T, Hagemann R. Hybrid Variegation in the Genus *Pelargonium*. Curr Genet. 1982; 5:245–249. https://doi.org/10.1007/BF00391813 PMID: 24186302

119. Greiner S, Rauwolf U, Meurer J, Herrmann RG. The role of plastids in plant speciation. Mol Ecol. 2011; 20:671–691. https://doi.org/10.1111/j.1365-294X.2010.04984.x PMID: 21214654

120. Sakamoto W. Leaf-variegated mutations and their responsible genes in *Arabidopsis thaliana*. Genes Genet Syst. 2003; 78:1–9. https://doi.org/10.1266/ggs.78.1 PMID: 12655133

121. Bogdanova VS. Inheritance of organelle DNA markers in a pea cross associated with nuclear-cytoplasmic incompatibility. Theor Appl Genet. 2007; 114:333–339. https://doi.org/10.1007/s00122-006-0436-6 PMID: 17080258

122. Weihe A, Apitz J, Pohlheim F, Salinas-Hartwig A, Börner T. Biparental inheritance of plastidial and mitochondrial DNA and hybrid variegation in *Pelargonium*. Mol Genet Genomics. 2009; 282:587–593. https://doi.org/10.1007/s00438-009-0488-9 PMID: 19787375

123. Barnard-Kubow KB, McCoy MA, Galloway LF. Biparental chloroplast inheritance leads to rescue from cytonuclear incompatibility. New Phytol. 2017; 213:1466–1476. https://doi.org/10.1111/nph.14222 PMID: 27686577

124. Turner CMR, Hide G, Buchanan N, Tait A. *Trypanosoma brucei*: Inheritance of Kinetoplast DNA Maxicircles in a Genetic Cross and Their Segregation during Vegetative Growth. Exp Parasitol. 1995; 80:234–241. https://doi.org/10.1006/expr.1995.1029 PMID: 7895834

125. Gibson W, Garside L. Kinetoplast DNA minicircles are inherited from both parents in genetic hybrids of *Trypanosoma brucei*. Mol Biochem Parasitol. 1990; 42:45–54. https://doi.org/10.1016/0166-6851(90)90111-x PMID: 2233899

126. Gibson W, Crow M, Kearns J. Kinetoplast DNA minicircles are inherited from both parents in genetic crosses of *Trypanosoma brucei*. Parasitol Res. 1997; 83:483–488. https://doi.org/10.1007/s004360050284 PMID: 9197397

127. Mather M, Henry K, Vaidya A. Mitochondrial Drug Targets in Apicomplexan Parasites. Curr Drug Targets. 2006; 8:49–60. https://doi.org/10.2174/138945007779315632 PMID: 17266530

128. Ralph SA, D'Ombrain MC, McFadden GI. The apicoplast as an antimalarial drug target. Drug Resist Updat. 2001; 4:145–151. https://doi.org/10.1054/drup.2001.0205 PMID: 11768328

129. Goodman CD, Buchanan HD, McFadden GI. Is the Mitochondrion a Good Malaria Drug Target? Trends Parasitol. 2017; 33:185–193. https://doi.org/10.1016/j.pt.2016.10.002 PMID: 27789127

130. Mukherjee A, Sadhukhan GC. Anti-malarial drug design by targeting apicoplasts: New perspectives. Australas J Pharm. 2016; 19:7–15. https://doi.org/10.3831/KPI.2016.19.001 PMID: 27280044

131. Shibeshi MA, Kifle ZD, Atnafie SA. Antimalarial drug resistance and novel targets for antimalarial drug discovery. Infect Drug Resist. 2020; 13:4047–4060. https://doi.org/10.2147/IDR.S279433 PMID: 33204122

132. Gojdics M. The Cell Morphology and Division of *Euglena deses* EHRGB. Trans Am Microsc Soc. 1934; 53:299–310. https://doi.org/10.2307/3222381

133. Ben-Shaul Y, Schiff JA, Epstein HT. Studies of Chloroplast Development in *Euglena* VII: Fine Structure of the Developing Plastid. Plant Physiol. 1964; 39:231. https://doi.org/10.1104/pp.39.2.231 PMID: 16655903

134. Brawerman G, Eisenstadt JM. Deoxyribonucleic Acid from the Chloroplasts of *Euglena Gracilis*. Biochim Biophys Acta. 1964; 91:477–485. https://doi.org/10.1016/0926-6550(64)90077-5 PMID: 14254019

135. Schmidt GW, Lyman H. Inheritance and Synthesis of Chloroplasts and Mitochondria of *Euglena gravcilis*. In: Lewin RA, editor. The Genetics of Algae. Berkeley: University of California Press; 1976. p. 257–299.

136. Hayashi Y, Ueda K. The shape of mitochondria and the number of mitochondrial nucleoids during the cell cycle of *Euglena gracilis*. J Cell Sci. 1989; 93:565–570. https://doi.org/10.1242/jcs.93.3.565

137. Edelman M, Epstein HT, Schiff JA. Isolation and characterization of DNA from the mitochondrial fraction of *Euglena*. J Mol Biol. 1966; 17:463–469. https://doi.org/10.1016/S0022-2836(66)80156-0 PMID: 5963078

138. Fonty G, Bernadi G, Crouse EJ, Stutz E. The Mitochondrial Genome of *Euglena gracilis*. Eur J Biochem. 1975; 54:367–372. https://doi.org/10.1111/j.1432-1033.1975.tb04147.x PMID: 809268

139. Biecheler B. Sur l'existence d'une copulation chez une Euglène verte et sur les conditions globales qui la déterminent. Comptes Rendus des Séances de la Société de Biologie. 1937; 124:12664–1266.

140. Visvesvara GS, Moura H, Schuster FL. Pathogenic and opportunistic free-living amoebae: *Acanthamoeba* spp., *Balamuthia mandrillaris*, *Naegleria fowleri*, and *Sappinia diploidea*. FEMS Immunol Med Microbiol. 2007; 50:1–26. https://doi.org/10.1111/j.1574-695X.2007.00232.x PMID: 17428307

141. Martinez AJ, Nelson EC, Jones MM, Duma RJ, Rosenblum WI. Experimental *Naegleria* Meningoencephalitis in Mice. An Electron Microscopy Study Laboratory Investigation. 1971; 25:465–475.

142. Fulton C. Ameba-flagellates as Research Partners: The Laboratory Biology of *Naegleria* and *Tetramitus*. In: Prescott DM, editor. Methods in Cell Physiology. New York: Academic Press; 1970. p. 341–468.

143. Schuster F. An Electron Microscope Study of the Amoebo-flagellate, *Naegleria gruberi* (Schardinger). I. The Amoeboid and Flagellate Stages. J Protozool. 1963; 10:297–313. https://doi.org/10.1111/j.1550-7408.1963.tb01681.x PMID: 14059620

144. Schuster F. Ultrastructure of Cysts of *Naegleria* spp: A Comparative Study. J Protozool. 1975; 22:352–359. https://doi.org/10.1111/j.1550-7408.1975.tb05185.x

145. Schuster F. An Electron Microscope Study of the Amoebo-flagellate, *Naegleria gruberi* (Schardinger). II The Cyst Stage. J Protozool. 1963; 10:313–320. https://doi.org/10.1111/j.1550-7408.1963.tb01682.x PMID: 14059621

146. Patterson M, Woodworth TW, Marciano-Cabral F, Bradley SG. Ultrastructure of *Naegleria fowleri* Enflagellation. J Bacteriol. 1981; 147:217–226. https://doi.org/10.1128/jb.147.1.217–226.1981

147. Fritz-Laylin LK, Prochnik SE, Ginger ML, Dacks JB, Carpenter ML, Field MC, et al. The Genome of *Naegleria gruberi* Illuminates Early Eukaryotic Versatility. Cell. 2010; 140:631–642. https://doi.org/10.1016/j.cell.2010.01.032 PMID: 20211133

148. Herman EK, Greninger AL, Visvesvara GS, Marciano-Cabral F, Dacks JB, Chiu CY. The mitochondrial genome and a 60-kb nuclear DNA segment from *naegleria fowleri*, the causative agent of primary amoebic meningoencephalitis. J Eukaryot Microbiol. 2013; 60:179–191. https://doi.org/10.1111/jeu.12022 PMID: 23360210

149. Pernin P, Ataya A, Cariout ML. Genetic structure of natural populations of the free-living amoeba, *Naegleria lovaniensis*. Evidence for sexual reproduction Heredity (Edinb). 1992; 68:173–181. https://doi.org/10.1038/hdy.1992.26 PMID: 29231202

150. Santos HJ, Makiuchi T, Nozaki T. Reinventing an Organelle: The Reduced Mitochondrion in Parasitic Protists. Trends Parasitol. 2018; 34:1038–1055. https://doi.org/10.1016/j.pt.2018.08.008 PMID: 30201278

151. Tovar J, León-Avila G, Sánchez LB, Sutak R, Tachezy J, Van Der Giezen M, et al. Mitochondrial remnant organelles of *Giardia* function in iron-sulphur protein maturation. Nature. 2003; 426:172–176. https://doi.org/10.1038/nature01945 PMID: 14614504

152. Santos HJ, Nozaki T. The mitosome of the anaerobic parasitic protist *Entamoeba histolytica*: A peculiar and minimalist mitochondrion-related organelle. J Eukaryot Microbiol. 2022; 69. https://doi.org/10.1111/jeu.12923 PMID: 35588086

153. Gilles HM, Hoffman PS. Treatment of intestinal parasitic infections: a review of nitazoxanide. Trends Parasitol. 2002; 18:95–97. https://doi.org/10.1016/s1471-4922(01)02205-x PMID: 11854075

154. Anderson VR, Curran MP, Alam S, Young JD. Nitazoxanide: A Review of its Use in the Treatment of Gastrointestinal Infections. Drugs. 2007; 67:1947–1967. https://doi.org/10.2165/00003495-200767130-00015 PMID: 17722965

155. Muller M. The hydrogenosome. J Gen Microbiol. 1993; 139:2879–2889. https://doi.org/10.1099/00221287-139-12-2879 PMID: 8126416

156. Fung HB, Doan T-L. New Drug Tinidazole: A Nitroimidazole Antiprotozoal Agent. Clin Ther. 1966; 27:1859–1884. https://doi.org/10.1016/j.clinthera.2005.12.012 PMID: 16507373

157. Cooper MA, Adam RD, Worobey M, Sterling CR. Population Genetics Provides Evidence for Recombination in *Giardia*. Curr Biol. 2007; 17:1984–1988. https://doi.org/10.1016/j.cub.2007.10.020 PMID: 17980591

158. Poxleitner MK, Carpenter ML, Mancuso JJ, Wang CJR, Dawson SC, Cande WZ. Evidence for karyogamy and exchange of genetic material in the binucleate intestinal parasite *Giardia intestinalis*. Science (1979). 2008; 319:1530–1533. https://doi.org/10.1126/science.1153752 PMID: 18339940

159. Bradic M, Carlton JM. Does the common sexually transmitted parasite *Trichomonas vaginalis* have sex? PLoS Pathog. 2018; 14:e1006831. https://doi.org/10.1371/journal.ppat.1006831 PMID: 29518151

160. Gilchrist CA, Ali IKM, Kabir M, Alam F, Scherbakova S, Ferlanti E, et al. A multilocus sequence typing system (MLST) reveals a high level of diversity and a genetic component to *Entamoeba histolytica* virulence. BMC Microbiol. 2012; 12:1–14. https://doi.org/10.1186/1471-2180-12-151 PMID: 22839995

161. Weedall GD, Clark CG, Koldkjaer P, Kay S, Bruchhaus I, Tannich E, et al. Genomic diversity of the human intestinal parasite *Entamoeba histolytica*. Genome Biol. 2012; 13:1–13. https://doi.org/10.1186/gb-2012-13-5-r38 PMID: 22630046

162. Bloomfield G. Sex in Dictyostelia. In: Romeralo M, Baldauf S, Escalante R, editors. Dictyostelids. Berlin, Heidelberg: Springer; 2013. p. 129–148.

163. Walker G, Dorrell RG, Schlacht A, Dacks JB. Eukaryotic systematics: A user's guide for cell biologists and parasitologists. Parasitology. 2011; 138:1638–1663. https://doi.org/10.1017/S0031182010001708 PMID: 21320384

164. Hackett JD, Anderson DM, Erdner DL, Bhattacharya D. Dinoflagellates: A remarkable evolutionary experiment. Am J Bot. 2004; 91:1523–1534. https://doi.org/10.3732/ajb.91.10.1523 PMID: 21652307

165. Stentiford GD, Shields JD. A review of the parasitic dinoflagellates *Hematondinium* species and *Hematodinium*-like infections in marine crustaceans. Dis Aquat Organ. 2005; 66:47–70. https://doi.org/10.3354/dao066047 PMID: 16175968

166. Robledo F, Vasta JA, Record GR. Protozoan Parasites of Bivalve Molluscs: Literature Follows Culture. PLoS ONE. 2014; 9:100872. https://doi.org/10.1371/journal.pone.0100872.t001

167. Lom J. Fish Invading Dinoflagellates: A Synopsis of Existing and Newly Proposed Genera. Folia Parasitol. 1981; 28:3–11. PMID: 7194841

168. Shields JD. The Parasitic Dinoflagellates of Marine Crustaceans. Annu Rev Fish Dis. 1994; 4:241–271. https://doi.org/10.1016/0959-8030(94)90031-0

169. Coats DW. Parasitic Life Styles of Marine Dinoflagellates. J Eukaryot Microbiol. 1999; 46:402–409. https://doi.org/10.1111/j.1550-7408.1999.tb04620.x

170. Horiguchi T. Diversity and Phylogeny of Marine Parasitic Dinoflagellates. In: Ohtsuka S, Suzaki T, Horiguchi T, Suzuki N, editors. Marine Protists Diversity and Dynamics. Tokyo: Springer Japan; 2015. p. 397–420.

171. Dhruve D, Soni A, Jatav SK, Katara S. Naturally occurring marine shellfish and finfish toxins: A review. The Pharma Innovation Journal. 2023; 12:1969–1976. Available from: https://www.researchgate.net/publication/371730611.

172. Gagat P, Bodył A, MacKiewicz P, Stiller JW. Tertiary plastid endosymbioses in dinoflagellates. Endosymbiosis. Springer-Verlag Wien; 2013. p. 233–290. https://doi.org/10.1007/978-3-7091-1303-5_13

173. Falkowski PG, Katz ME, Knoll AH, Quigg A, Raven JA, Schofield O, et al. The Evolution of Modern Eukaryotic Phytoplankton. Science (1979). 2004; 305:354–360. https://doi.org/10.1126/science.1095964 PMID: 15256663

174. Hackett JD, Bhattacharya D. The Genomes of Dinoflagellates. Genomics and Evolution of Microbial Eukaryotes. Oxford: Oxford University Press; 2006. p. 48–63.

175. Figueroa RI, Howe-Kerr LI, Correa AMS. Direct evidence of sex and a hypothesis about meiosis in Symbiodiniaceae. Sci Rep. 2021; 11. https://doi.org/10.1038/s41598-021-98148-9 PMID: 34552138

176. Bravo I, Figueroa RI. Towards an ecological understanding of dinoflagellate cyst functions. Microorganisms. 2014; 2:11–32. https://doi.org/10.3390/microorganisms2010011 PMID: 27694774

177. Phiester LA, Anderson DM. Dinoflagellate Reproduction. In: Taylor FJR, editor. The Biology of Dinoflagellates. Cambridge: Blackwell Scientific Publications; 1987. p. 611–649.

178. Kremp A. Diversity of dinoflagellate life cycles: facets and implications of complex strategies. In: Lewis JM, Marret F, Bradley LR, editors. Biological and Geological Perspectives of Dinoflagellates. London: The Geological Society of London; 2013.

179. Schnepf E, Drebes G. Anisogamy in the dinoflagellate *Noctiluca*? Helgoländer Meeresuntersuchungen. 1993; 47:265–273. Available from: https://hmr.biomedcentral.com/articles/10.1007/BF02367168.

180. Pholpunthin P, Fukuyo Y, Matsuoka K, Nimura Y. Life History of a Marine Dinoflagellate *Pyrophacus steinii* (Schiller) Wall *et* Dale. Botanica Marina. 1999; 42:189197–1999. https://doi.org/10.1515/BOT.1999.022

181. Berdieva M, Kalinina V, Lomert E, Knyazev N, Skarlato S. Life Cycle Stages and Evidence of Sexual Reproduction in the Marine Dinoflagellate *Prorocentrum minimum* (Dinophyceae, Prorocentrales). J Phycol. 2020; 56:941–952. https://doi.org/10.1111/jpy.12989 PMID: 32170721

182. Spector DL, Pfiester LA, Triemer RE. Ultrastructure of the Dinoflagellate *Peridinium cinctum f. ovoplanum*. II. Light and Electron Microscopic Observations on Fertilization. Am J Bot. 1981; 68:34–43. https://doi.org/10.1002/j.1537-2197.1981.tb06353.x

183. Chesnick JM, Cox ER. Fertilization and Zygote Development in the Binucleate Dinoflagellate *Peridinium balticum*. Am J Bot. 1989; 76:1060–1072. https://doi.org/10.1002/j.1537-2197.1989.tb15087.x

184. Sake Y, Nakanishi M, Konda T, Ishida Y, Kadota H, Shrestha K, et al. Life Cycle of *Peridinium* sp. B3 (Dinophyceae) Isolated from Lake Begnas, Nepal. Bulletin of Japanese Society of Microbial Ecology. 1986; 1:19–27. https://doi.org/10.1264/MICROBES1986.1.19

185. Sako Y, Ishida Y, Kadota H, Hata Y. Sexual Reproduction and Cyst Formation in the Freshwater Dinoflagellate *Peridinium cunningtonii*. Bulletin of the Japanese Society of Scientific Fisheries. 1983; 50:743–750. Available from: https://www.jstage.jst.go.jp/article/suisan1932/50/5/50_5_743/_article/-char/en

186. Pfiester LA. Sexual Reproduction of *Peridinium gatunense* (Dinophyceae). J Phycol. 1977; 13:92–95. https://doi.org/10.1111/j.1529-8817.1977.tb02893.x

187. Pfiester LA. Sexual Reproduction of *Peridinium willei* (Dinophyceae). J Phycol. 1976; 12:234–238. https://doi.org/10.1111/j.1529-8817.1976.tb00507.x

188. Pfiester LA. Sexual Reproduction of *Peridinium cinctum f. ovoplanum* (Dinophyceae). J Phycol. 1975; 11:259–265. https://doi.org/10.1111/j.1529-8817.1975.tb02776.x

189. Kita T, Fukuyo Y, Tokuda H, Hirano R. Sexual Reproduction of *Alexandrium hiranoi* (Dinophyceae). Bull Plankton Soc Japan. 1993; 39.

190. Xiaoping G, Dodge JD, Lewis J. An ultrastructural study of pianozygotes and encystment of a marine dinoflagellate, *scrippsiella* sp. Brit Phycol J. 1989; 24:153–165. https://doi.org/10.1080/00071618900650151

191. Koike K, Nishiyama A, Saitoh K, Imai K, Koike K, Kobiyama A, et al. Mechanism of gamete fusion in *Dinophysis fortii* (Dinophyceae, Dinophyta): Light microscopic and ultrastructural observations. J Phycol. 2006; 42:1247–1256. https://doi.org/10.1111/j.1529-8817.2006.00288.x

192. Fritz L, Anderson DM, Triemer RE. Ultrastructural Aspects of Sexual Reproduction in the Red Tide Dinoflagellate *Gonyaulax Tamarensis*. J Phycol. 1989; 25:95–107. https://doi.org/10.1111/j.0022-3646.1989.00095.x

193. Nishimura Y, Yoshinari T, Naruse K, Yamada T, Sumi K, Mitani H, et al. Active digestion of sperm mitochondrial DNA in single living sperm revealed by optical tweezers. Proc Natl Acad Sci U S A. 2006; 103:1382–1387. https://doi.org/10.1073/pnas.0506911103 PMID: 16432229

194. Kuroiwa H, Sugai M, Kuroiwa T. Behavior of Chloroplasts and Chloroplast Nuclei During Spermatogenesis in the Fern, *Pteris vittata L Protoplasma*. 1988; 146:89–100. Available from: https://link.springer.com/article/10.1007/BF01405917.

195. Sato M, Sato K. Degradation of Paternal Mitochondria by Fertilization-Triggered Autophagy in *C. elegans*. Source: Science, New Series. 2011; 334:1141–1144. https://doi.org/10.1126/science.l208619

196. Kuroiwa H, Nozaki H, Kuroiwa T. Preferential Digestion of Chloroplast Nuclei in Sperms before and during Fertilization in *Volvox carteri*. Cytologia (Tokyo). 1993; 58:281–291. https://doi.org/10.1508/CYTOLOGIA.58.281

197. Kuroiwa T, Enomoto S, Shihira-Ishikawa I. Preferential destruction of chloroplast nucleoids in zygotes in green algae *Dictyosphaeria cavernosa* and *Acetabularia calyculus*. Experientia. 1985; 41:1178–1179. Available from: https://link.springer.com/article/10.1007/BF01951716.

198. Aoyama H, Hagiwara Y, Misumi O, Kuroiwa T, Nakamura S. Complete elimination of maternal mitochondrial DNA during meiosis resulting in the paternal inheritance of the mitochondrial genome in *Chlamydomonas* species. Protoplasma. 2006; 228:231–242. https://doi.org/10.1007/s00709-006-0155-5 PMID: 16838082

199. Nakamura S, Aoyama H, Van Woesik R. Strict paternal transmission of mitochondrial DNA of *Chlamydomonas* species is explained by selection against maternal nucleoids. Protoplasma. 2003; 221:205–210. https://doi.org/10.1007/s00709-002-0053-4 PMID: 12802627

200. Nishimura Y, Misumi O, Kato K, Inada N, Higashiyama T, Momoyama Y, et al. An mt+ gamete-specific nuclease that targets mt- chloroplasts during sexual reproduction in *C. reinhardtii*. Genes Dev. 2002; 16:1116–1128. https://doi.org/10.1101/gad.979902 PMID: 12000794

201. Zaman V. *Balantidium coli*. In: Kreier JP, editor. Parasitic protozoa. New York: Academic Press; 1978. p. 633–653.

202. Nilles-Bije ML, Rivera WL. Ultrastructural and molecular characterization of *Balantidium coli* isolated in the Philippines. Parasitol Res. 2010; 106:387–394. https://doi.org/10.1007/s00436-009-1673-9 PMID: 19902250

203. Schuster FL, Ramirez-Avila L. Current world status of *Balantidium coli*. Clin Microbiol Rev. 2008; 21:626–638. https://doi.org/10.1128/CMR.00021-08 PMID: 18854484

204. Archibald JM. The Puzzle of Plastid Evolution. Curr Biol. 2009; 19. https://doi.org/10.1016/j.cub.2008.11.067 PMID: 19174147

205. Schoener DM, McManus GB. Plastid retention, use, and replacement in a kleptoplastidic ciliate. Aquat Microb Ecol. 2012; 67:177–187. https://doi.org/10.3354/ame01601

206. Johnson MD. Acquired phototrophy in ciliates: A review of cellular interactions and structural adaptations. J Eukaryot Microbiol. 2011; 58:185–195. https://doi.org/10.1111/j.1550-7408.2011.00545.x PMID: 21518077

207. Embley MT, Finlay BJ, Dyal PL, Hirt RP, Wilkinson M, Williams AG. Multiple Origins of Anaerobic Ciliates with Hydrogenosomes within the Radiation of Aerobic Ciliates. Proc Biol Sci. 1995; 262:87–93. https://doi.org/10.1098/rspb.1995.0180 PMID: 7479994

208. Martin W. The missing link between hydrogenosomes and mitochondria. Trends Microbiol. 2005; 13:457–459. https://doi.org/10.1016/j.tim.2005.08.005 PMID: 16109488

209. Corliss JO. The Ciliated Protozoa: characterization, classification, and guide to the literature. 2nd ed. Oxford: Pergamon Press; 1979.

210. Phadke SS, Zufall RA. Rapid diversification of mating systems in ciliates. Biol J Linn Soc. 2009; 98:187–197. https://doi.org/10.1111/j.1095-8312.2009.01250.x

211. Meyer E, Garnier O. Non-Mendelian Inheritance and Homology-Dependent Effects in Ciliates. Adv Genet. 2002; 46:305–337. https://doi.org/10.1016/s0065-2660(02)46011-7 PMID: 11931229

212. Hedgethorne K, Eustermann S, Yang J-C, Ogden TEH, Neuhaus D, Bloomfield G. Homeodomain-like DNA binding proteins control the haploid-to-diploid transition in *Dictyostelium*. Sci Adv. 2017; 3: e1602937. https://doi.org/10.1126/sciadv.1602937 PMID: 28879231

213. Bloomfield G, Skelton J, Ivens A, Tanaka Y, Kay RR. Sex determination in the social Amoeba *dictyostelium discoideum*. Science (1979). 2010; 330:1533–1536. https://doi.org/10.1126/science.1197423 PMID: 21148389

214. Ishida K, Hata T, Urushihara H. Gamete fusion and cytokinesis preceding zygote establishment in the sexual process of *Dictyostelium discoideum*. Dev Growth Differ. 2005; 47:25–35. https://doi.org/10.1111/j.1440-169x.2004.00776.x PMID: 15740584

215. Bloomfield G, Paschke P, Okamoto M, Stevens TJ, Urushihara H. Triparental inheritance in *dictyostelium*. Proc Natl Acad Sci U S A. 2019; 116:2187–2192. https://doi.org/10.1073/pnas.1814425116 PMID: 30670662

216. Boynton JE, Harris EH, Burkhart BD, Lamerson PM, Gillhamt NW. Transmission of mitochondrial and chloroplast genomes in crosses of *Chlamydomonas*. Proc Natl Acad Sci U S A. 1987; 84:2391–2395. https://doi.org/10.1073/pnas.84.8.2391 PMID: 3031682

217. Goodenough U, Lin H, Lee JH. Sex determination in *Chlamydomonas*. Semin Cell Dev Biol. 2007; 18:350–361. https://doi.org/10.1016/j.semcdb.2007.02.006 PMID: 17643326

218. Sager R, Lane D. Molecular Basis of Maternal Inheritance. Proc Natl Acad Sci U S A. 1972; 69:2410–2413. https://doi.org/10.1073/pnas.69.9.2410 PMID: 4506760

219. Umen JG, Goodenough UW. Chloroplast DNA methylation and inheritance in *Chlamydomonas*. Genes Dev. 2001; 15:2585–2597. https://doi.org/10.1101/gad.906701 PMID: 11581163

220. Bolen PL, Grant DM, Swinton D, Boynton JE, Gillham NW. Extensive Methylation of Chloroplast DNA by a Nuclear Gene Mutation Does Not Affect Chloroplast Gene Transmission in *Chlamydomonas*. Cell. 1982; 28:335–343. https://doi.org/10.1016/0092-8674(82)90351-8 PMID: 7060134

221. Feng T-Y, Chiang K-S. The persistence of maternal inheritance in *Chlamydomonas* despite hypomethylation of chloroplast DNA induced by inhibitors. Genetics. 1984; 81:3438–3442. https://doi.org/10.1073/pnas.81.11.3438 PMID: 6203123

222. Creasey AM, Mendis K, Carlton J, Williamson D, Wilson Iain, Carter R. Maternal inheritance of extrachromosomal DNA in malaria parasites. Mol Biochem Parasitol. 1994; 65:95–98. https://doi.org/10.1016/0166-6851(94)90118-x PMID: 7935632

223. Creasey AM, Ranford-Cartwright LC, Moore DJ, Williamson DH, Wilson RJM, Walliker D, et al. Uniparental inheritance of the mitochondrial gene *cytochrome b* in *Plasmodium falciparum*. Curr Genet. 1993; 23:360–364. https://doi.org/10.1007/BF00310900 PMID: 8467535

224. Vaidya AB, Morrisey J, Plowe C V, Kaslow DC, Wellems2 TE. Unidirectional Dominance of Cytoplasmic Inheritance in Two Genetic Crosses of *Plasmodium falciparum*. Mol Cell Biol. 1993; 13:7349–7357. https://doi.org/10.1128/mcb.13.12.7349–7357.1993 PMID: 8246955

225. Goodman CD, Siregar JE, Mollard V, Vega-Rodríguez J, Syafruddin D, Matsuoka H, et al. Parasites resistant to the antimalarial atovaquone fail to transmit by mosquitoes. Science. (1979). 2016; 352:349–353. https://doi.org/10.1126/science.aad9279 PMID: 27081071

226. Sinden RE, Canning EIU, Bray RS, Smalley ME. Gametocyte and gamete development in *Plasmodium falciparum*. Proc R Soc Lond B Biol Sci. 1978; 201:375–399. https://doi.org/10.1098/rspb.1978.0051 PMID: 27809

227. Okamoto N, Spurck TP, Goodman CD, McFadden GI. Apicoplast and mitochondrion in gametocytogenesis of *Plasmodium falciparum*. Eukaryot Cell. 2009; 8:128–132. https://doi.org/10.1128/EC.00267-08 PMID: 18996983

228. Stanway RR, Witt T, Zobiak B, Aepfelbacher M, Heussler VT. GFP-targeting allows visualization of the apicoplast throughout the life cycle of live malaria parasites. Biol Cell. 2009; 101:415–435. https://doi.org/10.1042/BC20080202 PMID: 19143588

229. Kuroiwa T, Kawano S, Watanabe M, Hori T. Preferential digestion of chloroplast DNA in male gametangia during the late stage of gametogenesis in the anisogamous alga *Bryopsis maxima*. Protoplasma. 1991; 163:102–113. https://doi.org/10.1007/BF01323334

230. Kuroiwa T, Hori T. Preferential Digestion of Male Chloroplast Nuclei and Mitochondrial Nuclei During Gametogenesis of *Bryopsis maxima* Okamura. Protoplasma. 1986; 133:85–87. https://doi.org/10.1007/BF01293191

231. Zhou Q, Li H, Li H, Nakagawa A, Lin JLJ, Lee ES, et al. Mitochondrial endonuclease G mediates breakdown of paternal mitochondria upon fertilization. Science (1979). 2016; 353:394–399. https://doi.org/10.1126/science.aaf4777 PMID: 27338704

232. Ferguson DJP, Henriquez FL, Kirisits MJ, Muench SP, Prigge ST, Rice DW, et al. Maternal inheritance and stage-specific variation of the apicoplast in *Toxoplasma gondii* during development in the intermediate and definitive host. Eukaryot Cell. 2005; 4:814–826. https://doi.org/10.1128/EC.4.4.814–826.2005

233. Ferguson DJP, Campbell SA, Henriquez FL, Phan L, Mui E, Richards TA, et al. Enzymes of type II fatty acid synthesis and apicoplast differentiation and division in *Eimeria tenella*. Int J Parasitol. 2007; 37:33–51. https://doi.org/10.1016/j.ijpara.2006.10.003 PMID: 17112527

234. Scholtyseck E, Mehlhorn H, Hammond DM. Electron Microscope Studies of Microgametogenesis in Coccidia and Related Groups. Z Parasitenkd. 1972; 38:95–131. https://doi.org/10.1007/BF00329023 PMID: 4622927

235. McDougald LR, Jeffers TK. Comparative *In Vitro* Development of Precocious and Normal Strains of *Eimeria tenella* (Coccidia). J Protozool. 1976; 23:530–534. https://doi.org/10.1111/j.1550-7408.1976.tb03834.x PMID: 1003341

236. Tomasina R, Francia ME. The Structural and Molecular Underpinnings of Gametogenesis in *Toxoplasma gondii*. Front Cell Infect Microbiol. 2020; 10:1–8. https://doi.org/10.3389/fcimb.2020.608291 PMID: 33365279

237. Yanagimachi R, Kamiguchi Y, Sugawara S, Mikamo K. Gametes and Fertilization in the Chinese Hamster. Gamete Res. 1983; 8:97–117. https://doi.org/10.1002/mrd.1120080202

238. Di Genova BM, Wilson SK, Dubey JP, Knoll LJ. Intestinal delta-6-desaturase activity determines host range for *Toxoplasma* sexual reproduction. PLoS Biol. 2019; 17. https://doi.org/10.1371/journal.pbio.3000364 PMID: 31430281

239. Kiara H, Steinaa L, Nene V, Svitek N. *Theileria* in Ruminants. In: Florin-Christensen M, Schnittger L, editors. Parasitic Protozoa of Farm Animals and Pets. Switzerland: Springer; 2018. p. 187–215.

240. Ganzinelli S, Rodriguez A, Schnittger L, Florin-Christensen M. *Babesia* in Domestic Ruminants. In: Florin-Christensen M, Schnittger L, editors. Parasitic Protozoa of Farm Animals and Pets. Switzerland: Springer; 2018. p. 215–241.

241. Rudzinska MA, Trager W, Lewengrub SJ, Gubert E. An Electron Microscopic Study of *Babesia microti* Invading Erythrocytes. Cell Tissue Res. 1976; 169:323–334. https://doi.org/doi.org/10.1007/BF00219605

242. del Carmen Terrón M, González-Camacho F, González LM, Luque D, Montero E. Ultrastructure of the *Babesia divergens* free merozoite. Ticks Tick Borne Dis. 2016; 7:1274–1279. https://doi.org/10.1016/j.ttbdis.2016.07.001 PMID: 27430965

243. Weber G, Friedhoff K. Preliminary Observations on the Ultrastructure of Supposed Sexual Stages of *Babesia bigemina* (Piroplasmea). Z Parasitenkd. 1977; 53:83–92. https://doi.org/10.1007/BF00383118 PMID: 919690

244. Pain A, Renauld H, Berriman M, Murphy L, Yeats CA, Weir W, et al. Genome of the host-cell transforming parasite *Theileria annulata* compared with *T. parva*. Science (1979). 2005; 309:131–133. https://doi.org/10.1126/science.1110418 PMID: 15994557

245. Fawcett DW, Conrad PA, Grootenhuist JG, Morzaria SP. Ultrastructure of the Intra-erythrocytic stage of *Theileria* Species from Cattle and Waterbuck. Tissue Cell. 1987; 19:643–655. https://doi.org/10.1016/0040-8166(87)90071-1 PMID: 3122362

246. Jura WGZO, Brown CGD, Kelly B. Fine Structure and Invasive Behaviour of the Early Developmental Stages of *Theileria annulata in vitro*. Vet Parasitol. 1983; 12:31–44. https://doi.org/10.1016/0304-4017(83)90085-7 PMID: 6407181

247. Hussein HE, Johnson WC, Taus NS, Ueti MW. Expression of sex-specific molecular markers by *Babesia bovis* gametes. Parasit Vectors. 2024; 17. https://doi.org/10.1186/s13071-024-06185-w PMID: 38374075

248. Putignani L, Tait A, Smith HV, Horner D, Tovar J, Tetley L, et al. Characterization of a mitochondrion-like organelle in *Cryptosporidium parvum*. Parasitology. 2004; 129:1–18. https://doi.org/10.1017/S003118200400527X PMID: 15267107

249. Zhu G, Marchewka MJ, Keithly JS. *Cryptosporidium parvum* appears to lack a plastid genome. Microbiology (Reading). 2000; 146:315–321. https://doi.org/10.1099/00221287-146-2-315 PMID: 10708370

250. Mogi T, Kita K. Diversity in mitochondrial metabolic pathways in parasitic protists *Plasmodium* and *Cryptosporidium*. Parasitol Int. 2010; 59:305–312. https://doi.org/10.1016/j.parint.2010.04.005 PMID: 20433942

251. Liu S, Roellig DM, Guo Y, Li N, Frace MA, Tang K, et al. Evolution of mitosome metabolism and invasion-related proteins in *Cryptosporidium*. BMC Genomics. 2016; 17. https://doi.org/10.1186/s12864-016-3343-5 PMID: 27931183

252. Abrahamsen MS, Templeton TJ, Enomoto S, Abrahante JE, Zhu G, Lancto CA, et al. Complete Genome Sequence of the Apicomplexan, *Cryptosporidium parvum*. Science (1979). 2004; 304:441–445. https://doi.org/10.1126/science.1095455 PMID: 15031438

253. Uni S, Iseki M, Maekawa T, Moriya K, Takada S. Ultrastructure of *Cryptosporidium muris* (strain RN 66) parasitizing the murine stomach. Parasitol Res. 1987; 74:123–132. https://doi.org/10.1007/BF00536023 PMID: 2964037

254. Rosales MJ, Arnedo T, Mascaró C. Ultrastructural Details of *Cryptosporidium parvum* Development in Calf Intestine. Mem Inst Oswaldo Cruz. 1998; 93:847–850. https://doi.org/10.1590/S0074-02761998000600027 PMID: 9921314

255. Tandel J, English ED, Sateriale A, Gullicksrud JA, Beiting DP, Sullivan MC, et al. Life cycle progression and sexual development of the apicomplexan parasite *Cryptosporidium parvum*. Nat Microbiol. 2019; 4:2226–2236. https://doi.org/10.1038/s41564-019-0539-x PMID: 31477896

256. Toso MA, Omoto CK. *Gregarina niphandrodes* may lack both a plastid genome and organelle. J Eukaryot Microbiol. 2007; 54:66–72. https://doi.org/10.1111/j.1550-7408.2006.00229.x PMID: 17300522

257. Jameson P. The Chromosome Cycle of Gregarines, with Special Reference to *Diplocystis schneideri* Kunstler. J Cell Sci. 1920; 2:207–262. https://doi.org/10.1242/jcs.s2-64.254.207

258. Schrevel J, Caigneaux E, Gros D, Philippe M. The Three Cortical Membranes of the Gregarines: I. Ultrastructural Organisation of *Gregarina blaberae*. J Cell Sci. 1983; 61:151–174. https://doi.org/10.1242/jcs.61.1.151 PMID: 6411745

259. Reger JF. The Fine Structure of the Gregarine *Pyxinoides balani* Parasitic in the Barnacle *Balanus tintinnabulum*. J Protozool. 1967; 14:488–497. https://doi.org/10.1111/j.1550-7408.1967.tb02034.x PMID: 4963675

260. Walsh RD, Callaway CS. The Fine Structure of the Gregarine *Lankesteria culicis* Parasitic in the Yellow Fever Mosquito *Aedes aegypti*. J Protozool. 1969; 16:536–545. https://doi.org/10.1111/j.1550-7408.1969.tb02313.x PMID: 4981008

261. Landers SC. The fine structure of the gamont of *Pterospora floridiensis* (Apicomplexa: Eugregarinida). J Eukaryot Microbiol. 2002; 49:220–226. https://doi.org/10.1111/j.1550-7408.2002.tb00526.x PMID: 12120987

262. Lucarotti CJ. Cytology of *Leidyana canadensis* (Apicomplexa: Eugregarinida) in *Lambdina fiscellaria fiscellaria* larvae (Lepidoptera: Geometridae). J Invertebr Pathol. 2000; 75:117–125. https://doi.org/10.1006/jipa.1999.4911 PMID: 10772324

263. Hajduk S, Ochsenreiter T. RNA editing in kinetoplastids. RNA Biol. 2010; 7:229–236. https://doi.org/10.4161/rna.7.2.11393 PMID: 20220308

264. Ferreira TR, Sacks DL. Experimental Hybridization in *Leishmania*: Tools for the Study of Genetic Exchange. Pathogens. 2022; 11:580. https://doi.org/10.3390/pathogens11050580 PMID: 35631101

265. Gubbels MJ, Striepen B. Studying the cell biology of apicomplexan parasites using fluorescent proteins. Microsc Microanal. 2004; 10:568–579. https://doi.org/10.1017/S1431927604040899 PMID: 15525431

266. McGovern OL, Rivera-Cuevas Y, Kannan G, Narwold AJ, Carruthers VB. Intersection of endocytic and exocytic systems in *Toxoplasma gondii*. Traffic. 2018; 19:336–353. https://doi.org/10.1111/tra.12556 PMID: 29437275

267. Marugan-Hernandez V, Long E, Blake D, Crouch C, Tomley F. *Eimeria tenella* protein trafficking: Differential regulation of secretion versus surface tethering during the life cycle. Sci Rep. 2017; 7. https://doi.org/10.1038/s41598-017-04049-1 PMID: 28676667

268. Ebrahimzadeh Z, Mukherjee A, Richard D. A map of the subcellular distribution of phosphoinositides in the erythrocytic cycle of the malaria parasite *Plasmodium falciparum*. Int J Parasitol. 2018; 48:13–25. https://doi.org/10.1016/j.ijpara.2017.08.015 PMID: 29154995

269. Tomlins AM, Ben-Rached F, Williams RA, Proto WR, Coppens I, Ruch U, et al. *Plasmodium falciparum* ATG8 implicated in both autophagy and apicoplast formation. Autophagy. 2013; 9:1540–1552. https://doi.org/10.4161/auto.25832 PMID: 24025672

270. Rotmann A, Sanchez C, Guiguemde A, Rohrbach P, Dave A, Bakouh N, et al. *Pf*CHA is a mitochondrial divalent cation/H+ antiporter in *Plasmodium falciparum*. Mol Microbiol. 2010; 76:1591–1606. https://doi.org/10.1111/j.1365-2958.2010.07187.x PMID: 20487273

271. Pastor-Fernández I, Pegg E, Macdonald SE, Tomley FM, Blake DP, Marugán-Hernández V. Laboratory Growth and Genetic Manipulation of *Eimeria tenella*. Curr Protoc Microbiol. 2019; 53. https://doi.org/10.1002/cpmc.81 PMID: 30811108