Red algal parasites: Models for a life history evolution that leaves photosynthesis behind again and again

*Nicolas A. Blouin** *and Christopher E. Lane*

Many of the most virulent and problematic eukaryotic pathogens have evolved from photosynthetic ancestors, such as apicomplexans, which are responsible for a wide range of diseases including malaria and toxoplasmosis. The primary barrier to understanding the early stages of evolution of these parasites has been the difficulty in finding parasites with closely related free-living lineages with which to make comparisons. Parasites found throughout the florideophyte red algal lineage, however, provide a unique and powerful model to investigate the genetic origins of a parasitic lifestyle. This is because they share a recent common ancestor with an extant free-living red algal species and parasitism has independently arisen over 100 times within this group. Here, we synthesize the relevant hypotheses with respect to how these parasites have proliferated. We also place red algal research in the context of recent developments in understanding the genome evolution of other eukaryotic photosynthesizers turned parasites.

Keywords:
genome reduction; life history; parasite evolution; red algae

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Department of Biological Sciences, University of Rhode Island, Kingston, RI, USA

***Corresponding author:**
Nicolas A. Blouin
E-mail: nblouin@mail.uri.edu

Introduction

Parasitology is one of the oldest fields of medical research and continues to be an essential area of study on organisms that kill millions annually, either directly or through agricultural loss. In the early genomics era, parasites were some of the initial eukaryotes to have their genomes sequenced. The combination of medical interest and small genome size (due to genome compaction [1]) has resulted in a relatively large number of sequenced genomes from these taxa. The range of relationships that exist between parasites and comparative free-living taxa, however, complicates understanding the evolution of eukaryotic parasitism. In some cases (such as apicomplexans, which cause malaria, cryptosporidiosis and toxoplasmosis, among other diseases) entire lineages appear to have a common parasitic ancestor [2].

Parasitism as a life history strategy has, by several orders of magnitude, more independent origins than the combined origins of autotrophy (e.g. [3–5]) and multicellularity [5, 6]. Many of the most virulent eukaryotic pathogens and parasites, however, have either directly evolved from a photosynthetic ancestor or are hypothesized to have plastid-containing ancestry. The apicomplexans are an example of a formerly photosynthetic lineage, turned parasitic. Additionally, both the human pathogen *Blastocystis* and the oomycetes that infect animals and devastate numerous plant species are members of the stramenopile lineage. This entire group (which includes diatoms and brown algae) is hypothesized to be derived from a photosynthetic ancestor, but this idea remains controversial [3, 7]. It is not clear why or how photosynthetic organisms so readily become parasites. However, at its core, photosynthesis is an enslavement of another organism. Multiple types of plastids exist, but they have originated either based on a eukaryote taking up a cyanobacteria and turning it into a plastid (primary endosymbiosis), or a eukaryotic organism being taken up by another eukaryote (secondary and tertiary endosymbiosis) to the same end [8]. These events start out as endosymbioses, but as the ''host'' exerts greater control, thousands of endo-

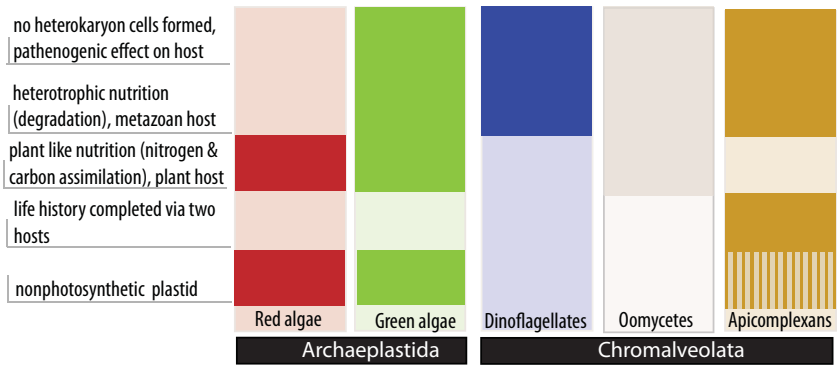


Figure 1. Parasitism has evolved many times from a photosynthetic ancestry, both in lineages containing plastids derived from primary endosymbiosis (Archaeplastida) and those with plastids derived from secondary endosymbiosis (Chromalveolata). Whereas many of these parasites share some commonalities, red algal parasites are by far the most recently diverging (darker shading indicates presence of trait).

symbiont genes are transferred to the host nucleus. There they are encoded with a 5′ targeting signal that routes their proteins back to the organelle [7]. The organellar targeting signal is similar to the signal that sends proteins to the secretory pathway [9–11], i.e. the pathway that parasites use to secrete proteins to control host cells.

From all that we know about obligate parasites or pathogens across the spectrum of eukaryotes, the vast majority of well-studied parasites are members of diverse clades with only distantly related free-living species (Fig. 1) [12–14]. Therefore, describing the "end result" of genomic consequences for parasites has been possible [1, 15, 16], but little is currently known about the process of getting there. Genome sequencing technology has developed considerably in recent years and the costs have fallen precipitously. This has opened the door to investigatons of novel systems with less direct applicability to human health. As a result, it is now possible to examine the early evolutionary stages of certain life histories and the variety of evolutionary pathways taken to arrive at a strategy, such as parasitism. In particular, the tools for closely examining gene loss, gene family expansions, and organellar reduction that accompany a switch to a parasitic lifestyle, are becoming available.

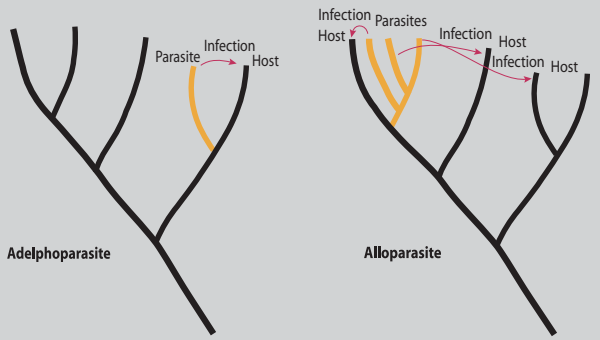
Parasites from within the red algae lineage provide a unique and powerful model for investigating the origins of a parasitic lifestyle for two important reasons. First, most red algal parasites share a recent common ancestor with an extant free-living red algal species. These close relatives are almost always their host, earning them the title adelphoparasites (adelphose is the Greek term for "kin"). Because of this sister-species relationship between parasite and host, a single pair of organisms can provide direct comparative data on the cellular and genomic changes occurring early in the evolution of a parasite. Additionally, this can yield information on host/parasite co-evolutionary dynamics. Second, a range of red algal parasites exists that extends from the highly specific adelphoparasites just described, to promiscuous alloparasites – those parasites that are capable of infecting distantly related hosts (Box 1). Therefore, the evolutionary spectrum and independent origins of these parasites provides a natural system with which to examine the earliest stages of parasite evolution. Throughout this paper, the term "red algal parasite" will refer only to those parasites that have evolved from within canonical free-living red algal taxa, and not all parasites of red algae.

Red algae easily become parasites

The red algae are an ancient photosynthetic lineage that arose through primary endosymbiosis, with a fossil record dating back over 1.2 billion years [17, 18]. To date, very little is known

Box 1

Relationship of adelpho- and alloparasites and their hosts within the red algae



Traditionally, parasites were classified as adelphoparasites if their eruptant development mimicked a life-history stage of their host. In many cases these relationships remain robust, based on more recent molecular work. However, re-examinations of many of these taxa show that morphology is a poor indicator of the relationship between parasite and host. Species considered adelphoparasites based on this criterion often attack not only sister taxa, but also genera from the same family or tribe [31, 47, 97]. Conversely, to be truly considered alloparasites, it seems that a host species must have substantial evolutionary distance from its parasite, often from a different family or tribe [28, 47]. The majority of red algal parasites exist in the continuum between these poles of single host adelphoparasites and promiscuous alloparasites (see above image). This brings into question whether or not the designation of particular taxa as adelpho- or alloparasites is still useful in the context of molecular investigations, with several authors calling for the terms to be avoided altogether [26, 31, 42, 47].

Glossary

Adelphoparasite: A red algal parasite that grows on a closely related host organism(s) and receives nutrition from the host via secondary pit connections. *Sensu stricto* this includes only those taxa that are sister species in phylogenetic analyses and are likely to share conserved developmental mechanisms for all reproductive stages. In practice, molecular evidence of the required resolution is often lacking, therefore often adelphoparasites are labeled as such based on morphological diagnoses.

Alloparasite: A red algal parasite that grows on a host that does not share a recent common ancestor. *Sensu stricto*, those organisms that parasitize hosts from different families, tribes, or orders. Alloparasitism also occurs within the family level; however, good phylogenetic resolution is required for this assessment.

Promiscuous alloparasite: A red algal parasite that grows on several hosts in nature, with at least one of the hosts not closely related to the parasite (cf. alloparasite).

Primary pit connection: In the red algae new cells that are the result of mitotic division retain a connection to their parent cell that is subsequently filled in with a proteinaceous plug. No intercellular exchange has ever been detected through primary pit connections as occurs through the plasmodesmata found in higher plants.

Secondary pit connection: Cell fusions between adjacent cells in the red algae, not resulting from mitosis. Developing red algal carposporophytes form cellular connections with female gametophyte cells through which they can transfer carposporophyte nuclei and receive nutrition from the female gametophyte. A similar process is observed in red algal parasite development (see Figs. 2A and 5).

about red algal genomes, despite their ecological, economic, and evolutionary importance. Carrageenans and agars extracted from red algae have many applications in food products, industry, and pharmaceuticals as stabilizers for emulsions and for their gelling and thickening properties [19]. The bangiophyte red alga, *Porphyra* – a staple food crop in Asia (''nori'') – represents a multi-billion dollar per year industry. However, availability of genome-scale data from red algae lags far behind other lineages with equivalent ecological and economical importance.

Red algae are distinguished from other algal groups by their complete lack of flagella in any of their life-history stages and their maintenance of accessory photosynthetic pigments. These are derived from their cyanobacteria endosymbiont that later evolved into the plastid. Whereas the rhodophyte lineage is both ancient and diverse, multicellular forms only exist in the bangiophytes (e.g. *Porphyra*) and florideophytes (e.g. *Chondrus*). Parasitism is only known from the Florideophyceae, where there exists a peculiar triphasic life history (Fig. 2A) [20]. In addition to haploid and diploid life-history stages found in many algae and lower plants, florideophytes have a third life-history stage, termed a ''carposporophyte''. This stage consists of diploid spores arising from a zygote retained post fertilization that is surrounded by a protective and nutritive layer of haploid cells originating from the female gametophyte (Fig. 2C). This life-history stage has been thought to be a way to amplify fertilization events that may be rare given that the red algae lack flagellated spores ([21], but see [22]). Additionally, the carposporophyte is often referred to as a ''parasitic'' life-history stage in the literature because nutrients flow from the haploid ''host'' female gametophyte into the developing diploid carposporophyte (Fig. 2) [21].

Parasites within the florideophyte red algae were initially classified based on morphological characteristics in the early 20th century [23]. Consistent with an idea popular at the time for parasitism in animals, they were assumed to have evolved from their hosts [24–26]. Starting in the mid 20th century, many parasites were re-classified based on reproductive development. This shift confirmed that both allo- and adelphoparasitism exists within the florideophytes [27–29]. More recent applications of molecular tools have helped to identify previously cryptic relationships. There are currently 116 species of red algal parasites described that represent 8% of the described genera (66 genera) in the florideophyte algae. This should be considered an estimate pending further molecular investigation [30]. Even with new taxa being described or combined, it is obvious that parasitism is widespread within the florideophytes, including parasitic taxa in 8 of the currently recognized 24 orders (Fig. 3). Whether or not taxonomic distribution is correlated to the development of a parasitic lifestyle has never been tested. However, from their taxonomic distribution, it is evident that parasitism has independently evolved within the red algae at least 14 times and possibly more than 100 times [31]. From an evolutionary standpoint, the number of times parasitism has arisen among red algae compels us to question why the red algae are so prone to assuming this lifestyle, and why so many attack their closest relatives (Fig. 4)?

Red algal parasites have multiple possible origins

While sharing some commonalities with other parasites, red algal parasites have many features that make them unique, resulting from their life history. Two features of florideophyte red algae, secondary pit connections and carposporophytes, may be instrumental in the multiple origins of a parasitic lifestyle. Additionally, they explain why expanding to hosts outside the Florideophyceae is unlikely. Primary pit connections result from cell division, which is incomplete in red algae. A small pore remains after cytokinesis that is subsequently filled with a glycoprotein plug. Secondary pit connections are formed via cell fusions between adjacent cells, not resulting from cell division. This mechanism removes one important barrier for red algal parasites, namely getting into the host cell.

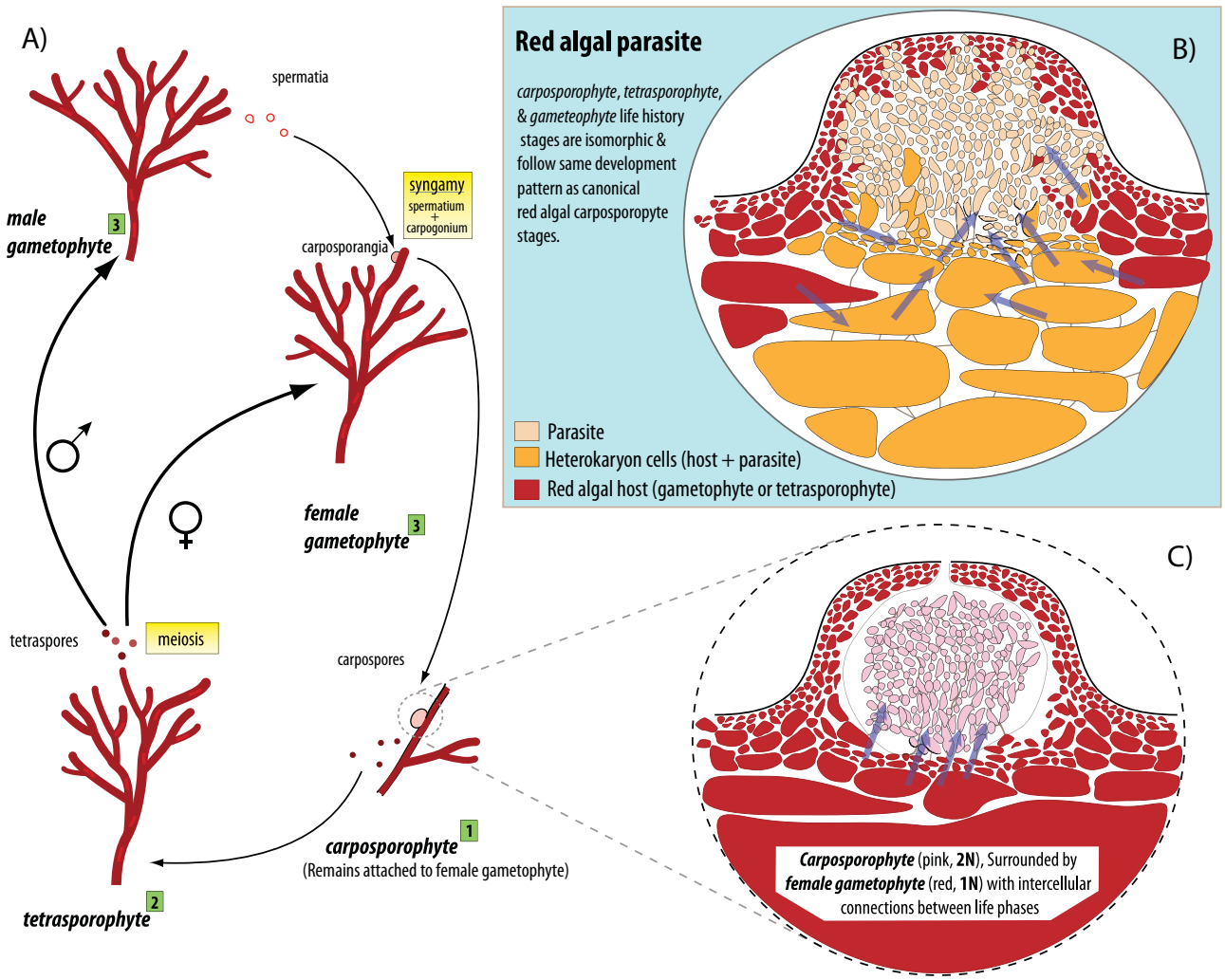


Figure 2. A typical florideophyte life history showing: **A1:** the carposporophyte, **A2:** tetrasporophyte, and **A3:** gametophyte life-history stages. Magnifications show the similarity between **B:** the development of red algal parasites and **C:** the carposporophyte life-history stage of free-living red algae. Arrows indicate source and sink for carbohydrate trafficking that occurs through cellular connections (i.e. ''secondary pit connections''), **B**: between parasite and host or **C**: between life-history stages.

Many parasites have evolved unique mechanisms to enter their hosts. Apicomplexans get their name from the apical complex, which is a specialized structure at the apical end of the cell, used for host entry [32]. It is a conserved feature across the diverse apicomplexan lineage and probably the strongest morphological feature uniting the group [33]. Another large lineage including many pathogens that infect a broad range of taxa is the oomycetes. Plant pathogenic members of this group use structures called appressoria to penetrate the cuticle and gain access to the host [34]. Red algal parasites, however, circumvent the need to invent host entry mechanisms, because they already possess the ability to form secondary pit connections with host cells to facilitate their attack. In support of the importance of this feature, florideophyte orders that lack cell fusions (e.g. Nemaliales) also lack parasitic members. Furthermore, the Palmariales produce simple cell fusions – which may be an intermediary step toward development of true secondary pit connections – and this order contains only one confirmed parasitic species (*Rhodophysema kjellmanii* [35]).

In all red algal parasites studied thus far, host infection and subsequent proliferation of the parasite closely resembles the developmental pattern of the florideophyte carposporophyte life-history stage [36, 37]. This has led to the supposition that parasites likely arose from a short circuit of the triphasic life history and subsequent loss of photosynthesis. The carposporophyte stage is recapitulated in the absence of the gametophyte and tetrasporophyte life-history stages (Fig. 2) [38]. However, these parasites retain a complete red algal triphasic life history even though their visible morphology is reduced to resemble only the carposporophyte stage, in comparison to free-living relatives (e.g. [39–41]). Therefore, the situation is more complicated than a self-replicating carposporophyte-like stage gone haywire.

An alternative hypothesis invokes a progression toward parasitism from endophytism and epiphytism, which are common strategies among macroalgae. Endophyte rametes or

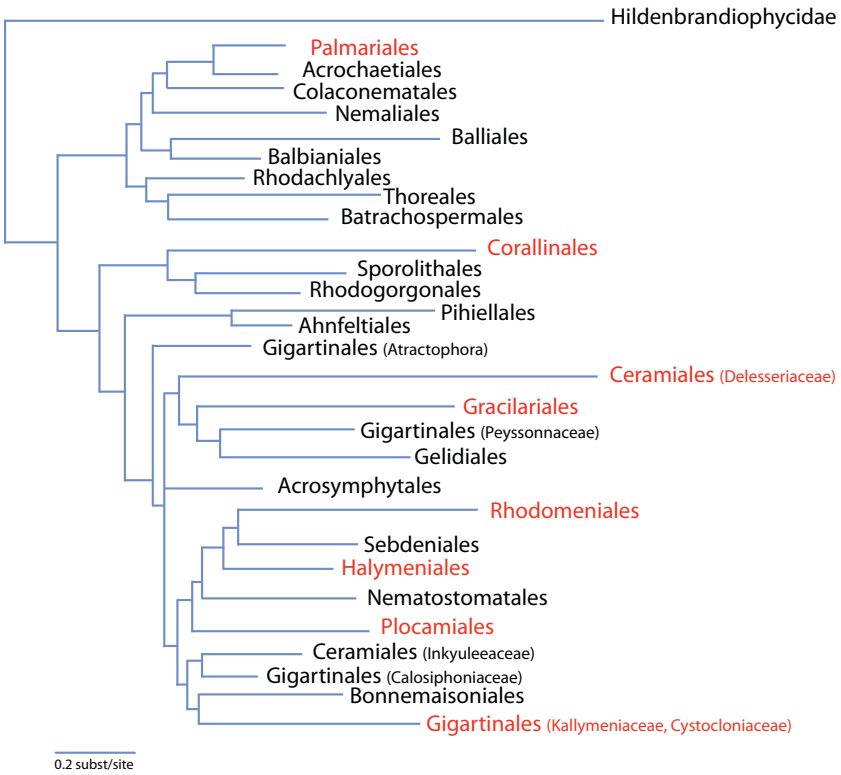


Figure 3. Ordinal florideophyte red algal tree with current taxonomic classification based on [30]. Red text indicates orders that contain parasitic taxa. Both the Gigartinales and Ceramiales appear to be paraphyletic and thus appear multiple times in the tree.

epiphyte rhizoids from proto-red algal parasites have been proposed to make secondary pit connections that are tolerated by closely related species. This situation is much like how typical parasites establish connections with their host [23, 28, 42]. This trend of increasing dependence is recapitulated in other protist parasite lineages as well. The free-living oomycetes are saprobic decomposers. The closest relative of apicomplexans, *Chromera velia*, is a coral symbiont [43]. Likewise, the closest relative of the green algal parasites *Prototheca* and *Helicosporidium*, *Chlorella variabilis*, is an intracellular photobiont [44]. Perhaps tellingly, the distribution of red algal parasites appears particularly enriched in those lineages that contain large numbers of epiphytic taxa (e.g. Ceramiales). By taking advantage of morphological features that are part of the canonical florideophyte life cycle, parasitism may be a relatively simple transition in red algae. Unlike parasites that invade organisms from other phyla and must evade host detection, the barrier to infection is likely quite low for a parasite derived from a recent common ancestor of the host. The genetic similarity between parasite and host may be the key in host evasion. This genomic compatibility could lead to rapid gene loss by the evolving parasite, based on genetic redundancy and the ability to use host proteins. This, however, remains to be tested formally.

Currently, adelphoparasites are believed to be more recently diverged than alloparasites, and there is some molecular data to support this idea [26, 31, 45–48]. In this context, red algal parasite lineages must initially evolve as an adelphoparasite with a single host. Subsequently, they gain the ability to form relationships with other closely related hosts, and eventually infect more distantly related taxa (Box 1). However, independent origins for allo- and adelphoparasites cannot be ruled out at this time, especially given the apparent number of times parasitism has evolved in red algae and the evidence that somatic growth and proliferation of the parasite may differ between alloparasites and adelphoparasites (Fig. 5).

Nascent parasites have the potential for genome compaction

Where detailed investigations have been performed, the nuclear genomes of eukaryotic parasites are substantially reduced in their coding capacity, compared to canonical genomes [1, 49]. The reasons for this are mainly related to genome streamlining and the abandonment of metabolic pathways whose products are supplied by the host. Whereas little is known about the genomes of red algal parasites, it is clear that parasite nuclei are physically smaller than the nuclei of their hosts, based on DAPI staining of heterokaryon cells [50]. Both the host and parasite maintain haploid and diploid stages, eliminating the possibility that ploidy is solely responsible for the observed differences.

There may, however, be critical nuclear changes that occur once a parasite begins to broaden its host range. Microscopy studies have demonstrated that adelphoparasite nuclei exhibit large nucleolar regions. This observation suggests that these genomes are functioning normally using their own translational machinery. Adelphoparasite nuclei are also evenly distributed throughout the cell [50]. Conversely, in studied alloparasites, the transferred parasite nuclei are markedly

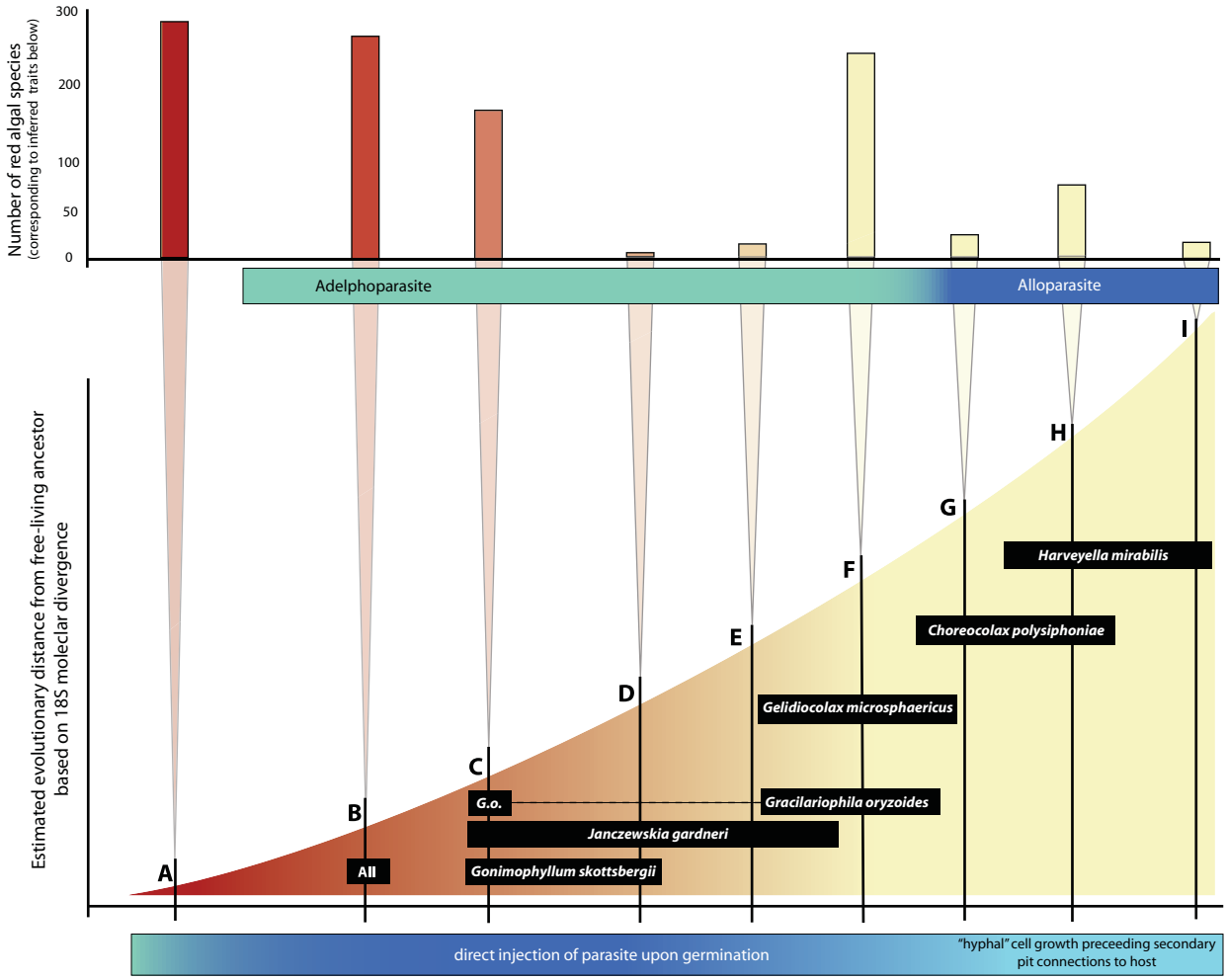


Figure 4. Relationship of key red algal parasite evolutionary characters to number of taxa containing each character. Example taxa are included in black boxes. **A:** Non-parasitic epiphyte or endophyte; **B:** carbon trafficking from host to parasite, loss of parasite plastid; **C:** single closely-related host, parasite and host nuclei equally dispersed throughout heterokaryon cells; **D:** maintenance of functioning plastids derived from host; **E:** transient photosynthesis; **F:** complete loss of photosynthetic ability; **G:** distantly related host; **H:** parasite and host nuclei segregating into zones in heterokaryon cells; **I:** infection of multiple distantly related hosts. 18S rDNA divergence based on [47]. Number of red algal species estimated from taxonomic records in ref. [98].

compacted in comparison to the host nuclei. They exhibit no apparent nucleolus, and form a strong effective gradient between the two nuclei types within the heterokaryon cell [51]. This may indicate that the host cell environment is less permissive for the distantly-related parasite, partially compartmentalizing the infection that results in low transcriptional activity in the alloparasite nucleus.

It is well documented that genome compaction occurs through the loss of intergenic sequences and gene loss during the establishment of an endosymbiont [1, 52–55]. Similar genome compaction seems to occur through gene loss in many parasites, most notably the fungal pathogen *Encephalitozoon cuniculi*, but also in euglenozoans and apicomplexans [56–61]. Despite a general trend toward reduction, there are expansions in those gene families that allow some parasites to avoid host defenses and scavenge nutrients [59, 62, 63]. A broad genomic/transcriptomic sampling of red algal parasites will identify if there are trends in genome compaction and gene family expansions. This will either confirm or refute the idea that alloparasites represent older evolutionary events within the red algae than adelphoparasites.

Parasites direct nutrition delivery from hosts and have tightly constrained organellar relationships

In the red algae, the developing carposporophyte relies on nutritive cells of the parent to accumulate and deliver floridean starch. These nutritive cells stain darkly with hematoxilin blue and other protein stains, and are thought to indicate

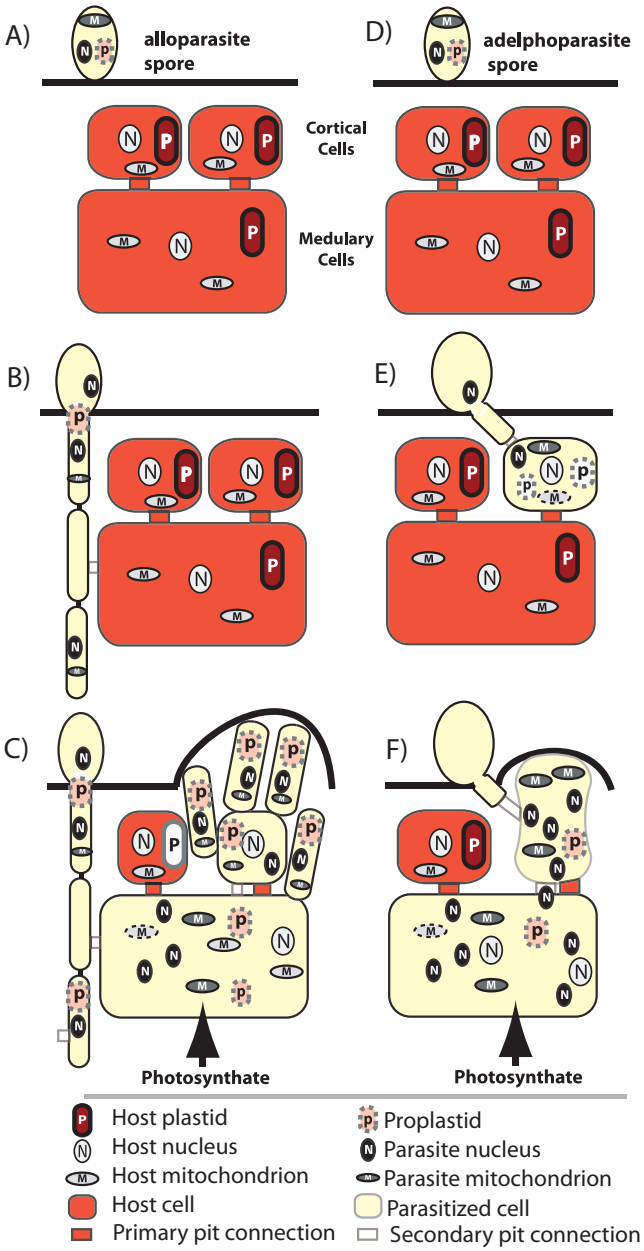


Figure 5. A-C: Diagram showing germination and development for an alloparasite (*C. polysiphoniae*), and **D-F:** an adelphoparasite (*G. oryzoides*) showing how proliferation is mediated through secondary pit connections.

that a range of nutrition is fed to the carposporophyte [21, 27, 64]. A similar trend occurs during growth of the parasite, but nutrient transfer is more diffuse because of a larger number of secondary pit connections (Figs. 2B and C). In the case of parasites, transfer appears to occur either through host cells that are intermixed within eruptant parasite tissue, or through a gradation of non-photosynthetic parasite cells overlaying host-parasite heterokaryons that overlays uninfected host tissue [50]. The host cells adjacent to parasite cells exhibit a lower concentration of starch and proteins than found in carposporophyte nutritive cells. This is considered an indication that there is a constant gradient for nutrient transfer upon infection [41]. Rather than all of the parasite cells storing starch in large quantities, as the carpospores do, starch accumulates in large amounts in the reproductive cells of the parasite [65]. The movement of at least five sugar species and phosphate esters from heterokaryon cells into the alloparasite *Harveyella mirabilis* indicates that parasites must be actively directing nutrients to their cells and that this results from the presence of parasitic nuclei in source cells [66]. It is still not known which metabolites, beyond photosynthate, are trafficked to the parasite from the host. If metabolite trafficking is extensive, alloparasites would likely streamline their own genomes and depend on the host genome and machinery for the necessary products for survival [67, 68].

Parasite organelles

Another unusual feature of red algal parasites is that they maintain host-derived, photosynthetically inactive plastids (“proplastids” [69]), resulting in the requirement for exogenous starch to compensate for the lack of endogenous photosynthate. There are many other parasites that maintain a non-photosynthetic plastid (e.g. the “apicoplast” of apicomplexans, the green algal parasites in the genera *Helicosporidium* and *Prototheca*, and the parasitic liverwort *Aneura mirabilis* [12, 70, 71]). Aside from photosynthesis, plastids have been shown to be required for amino acid metabolism, fatty acid biosynthesis [72–75], and pyrimidine biosynthesis ([76, 77] but see [78]). The ability of red algal parasites to retain host-derived proplastids, and in some cases reactivate them in culture, suggests that nuclear-encoded plastid-targeted genes are still functionally active in some adelphoparasites [69, 79–81]. Most parasites, however, cannot rescue photosynthesis in their proplastids, indicating that evolutionary divergence from the host plays a significant role in the parasite’s interaction with the proplastid.

Parasite mitochondria are maintained separately from the host copy, and upon infection, parasites transfer their mitochondria into the host where they rapidly divide [26, 69]. Given that parasites co-opt host plastids, the maintenance of their own mitochondria indicates that parasites cannot utilize the host mitochondria. In heterokaryon cells, parasite mitochondria rapidly divide prior to parasite nuclei duplication, strongly suggesting that these are the source for the ATP required to replicate parasite nuclei.

Evidence for the nuclear control of mitochondrial compatibility has been reported in closely related species of *Paramecium* [74], interspecific hybrids in *Solanum* [82, 83], and even in intraspecific somatic cell nuclear transfer in bovines [84]. These studies demonstrated that even small differences in ATP content in mitochondrial haplotypes reduced transfer efficiency, providing evidence that nuclear-mitochondria relationships are tightly constrained. A recent mitochondrial comparative genomic study between the red algal parasite, *Gracilariophila oryzoides*, and its host, *Gracilariopsis andersonii*, showed that the two genomes differed in their gene content and organization [48]. The differences in mitochondrial genome content/organization between *G. oryzoides* and *G. andersonii* may result in differing poly-

cistronic RNA products that are part of species-specific nuclear-mitochondrion interactions. This alone could explain why the parasite maintains its own mitochondria.

Organellar genomes

In terms of gene content, *atp*8 and *sdh*C are not present in the *G. oryzoides* mitochondrial genome, but they are present in all other sequenced red algal mitochondrial genomes, including unicellular representatives of the earliest diverging red algal groups [85–87]. Interestingly, *atp*8 is also missing from the mitochondrial genome of *Plocamiocolax pulvinata*, another red algal parasite [48]. *Atp*8 codes for a membrane-bound component of ATP synthase that is thought to form part of the coupling structure between the F_0 and F_1 subunits [88]. The core structure of the F_0 is still poorly resolved in the red algae, particularly the proteins that form the connectors between the F_0 barrel and the F_1 catalytic subunit. The *ymf*39 gene present in both parasite mitochondrial genomes may be able to replace the function normally performed the *atp*8 protein product [89], but this requires further investigation. Additionally, there is one other difference in gene content between *G. andersonii* and *G. oryzoides* mitochondria that deserves attention: *G. oryzoides* contains histidine tRNA and its host appears to have lost its copy [48]. Histidyl-tRNA expression is required for healthy growth of the blood pathogen *Trypanosoma brucei*. It would not appear that the loss of histidine tRNA affects growth of the *G. andersonii*. However, further mitochondrial genome studies may provide evidence as to whether or not histidine tRNA retention is common in red algal parasites (it is not present in the *P. pulvinata* mitochondrion genome) in comparison to their host species.

In parasites derived from photosynthetic lineages (apicomplexans, red algae, green algae, and stramenopiles including *Blastocystis* and oomycetes) for which genomic information is available, only those derived from green algal lineages have kept their full complement of mitochondrial electron transport genes. In red algae and apicomplexan parasites, as well as *Blastocystis*, components of the F_0F_1 ATP synthetase complex are missing, and they appear incapable of completing the citric acid (TCA) cycle. Indeed, the lack of *sdh*C and *atp*8 genes in the *G. oryzoides* mitochondrial genome, the fact that *atp*8 is conserved in the mitochondrial genomes of all red algae studied thus far, and our inability to detect *sdh*C or *atp*8 in 454 (genomic) and Illumina (RNA) datasets [48,unpublished data], are consistent with the idea that electron transport is the first mitochondrial casualty in degenerative evolution of this organelle [48, 90].

Organellar proteomes

Examination of alloparasite mitochondrial proteomes will provide an interesting comparison to see if the deletions found in adelphoparasite mitochondria are more extensive with respect to electron transport. These data will likely provide context for the organellar reduction observed in more anciently evolved parasites. Both oomycetes and apicomplexan pathogens have reduced electron transport and TCA cycle genetic compliments in comparison to red algal parasites. The reductions are to the point that alternative strategies for maintaining an ATP pool must be considered [91, 92]. This has been demonstrated between many intracellular parasites and their hosts through non-mitochondrial ADP/ATP transport [93, 94]. Some red algae are known to contain non-mitochondrial ADP/ATP translocases in addition to organellar-bound translocases [95]. We are in the process of examining this gene family in free-living and parasitic red algae to see if the loss of electron transport components covaries with gene enrichment or expression in these genes, or their intracellular location. This will help us understand whether or not the mitochondrion is losing genes in a similar trajectory as that reconstructed for other parasites, and provide the basis for understanding organellar retrograde signaling to the nucleus. This will be particularly interesting given that signals such as redox states and tetrapyrrole pools are likely quite different in parasitic organelles, compared to those of their free-living counterparts [96].

Conclusions

The millions of years of evolution separating well-characterized eukaryotic parasites and free-living relatives complicate the interpretation of genomic innovations. The combination of ancient divergence and accelerated rates of protein evolution in most parasites conspires against their direct comparison to closely related free-living taxa. This is especially true for well-studied parasites with photosynthetic roots, like apicomplexans, because closely related free-living taxa are unknown. Red algal parasites provide a way around these difficulties and specifically address questions regarding the establishment of the parasitic lifestyle. Red algal data will be particularly useful for understanding the vital biochemistry of organellar function and protein trafficking during an infection. Additionally, the red algal infection cycle will help us answer questions regarding the tolerance of cells to genomic chimericism, e.g. how do two nuclei from different species co-exist in the same cell after the parasite nucleus is injected into the host? Even if the host and parasite are closely related, genomic changes in both organisms following their divergence should cause cellular incompatibilities. All red algal parasites have found a way to alter the host cell or trick the host cellular machinery in order to propagate themselves, whether they are closely or distantly related to the host. An understanding of the breakdown of self-recognition factors within the host may provide intriguing hypotheses for other parasite systems or for fields as distant as research on cancer and autoimmune diseases. The transition from photosynthetic to parasitic is a well-trodden evolutionary path throughout the tree of life. Therefore, establishing red algal parasites as a model system for comparative genomic studies for early parasitism will produce broad-reaching data for years to come.

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