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Organelle genomes of *Sargassum confusum* (Fucales, Phaeophyceae): mtDNA vs cpDNA

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Abstract

The drifting biomass of golden tide in the Yellow Sea of China mainly consisted of *Sargassum horneri* with a small fraction composed of *Sargassum confusum* thalli. In this study, the circular-mapping organelle genomes (mtDNA and cpDNA) of *S. confusum* were sequenced and coupled with comparative genomic and phylogenomic analyses within the *Sargassum* genus. This revealed 34,721-bp mitochondrial and 124,375-bp chloroplast genomes of *S. confusum* harboring 65 and 173 genes, respectively, figures which are highly comparable to those reported in other *Sargassum* species. The mtDNA of *S. confusum* displayed lower values in A+T and intergenic spacer contents than cpDNA. Mitochondrial phylogenomics revealed a close relationship between *Sargassum muticum* and *S. confusum*. The *Sargassum* mtDNAs had an approximately three-fold greater mutation rate than cpDNAs indicating a higher evolution rate in mtDNAs than cpDNAs for *Sargassum* species. Therefore, mtDNA is a more effective molecular marker and could aid in tracking the source of the golden tides.

Keywords Organelle genomes · Golden tide · Phaeophyceae · Evolution · *Sargassum* · Phylogenomics

Introduction

Sargassum C.Agardh is a genus in the order Fucales, comprising 360 brown algal species (Mattio and Payri 2011; Guiry and Guiry 2018). Most species are distributed within intertidal and subtidal regions of temperate and tropical oceans, forming important ecological structures known as marine forests that provide food and shelter to a diverse range of invertebrates, fishes, sea turtles, and mammals (Laffoley et al. 2011; Witherington et al. 2012; Duncan 2013; Milledge and Harvey 2017). Some *Sargassum* species (e.g., *Sargassum horneri*, *Sargassum natans*, and *Sargassum fluitans*) can maintain high growth rates while in a floating state within the pelagic zone, forming harmful macroalgal blooms known as "golden tides" (e.g., Komatsu et al. 2008; Smetacek and Zingone 2013; Komatsu et al. 2014). The reoccurrence of large-scale drifting *Sargassum* biomass, or golden tides, in the Yellow Sea of China in recent years has caused considerable damage to the local environment and economy (China Xinhua News 2017). The *Sargassum* biomass mainly consisted of drifting *S. horneri* thalli, with a small fraction composed of *Sargassum confusum* thalli (Liu et al. 2018).

Organelle genomes carry important genetic information crucial for comparative genomics and understanding the evolutionary history of brown algae (e.g. Le Corguillé et al. 2009; Yotsukura et al. 2010; Liu et al. 2017a, b). Known brown algal mitochondrial and chloroplast genomes (mtDNAs and cpDNAs) typically consist of single circular molecules. These include an ever-increasing number of

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mitochondrial (e.g., *cox3*, *cox1*, *nad6*, *atp9*, *nad1*, *nad4*, *rns*, and *rnl*) and plastid (e.g., *rbc*L, *rbc*LSspacer, *psb*A, and *psa*A) DNA markers that have been investigated and used in phylogenetic analyses in brown algae (e.g., Phillips et al. 2005; Lane et al. 2007; Engel et al. 2008; Boo et al. 2011; Silberfeld et al. 2014). Previous work has attempted to unveil some organelle genome sequences in the order Fucales. The size of sequenced mtDNAs in Fucales ranges from 34.6 to 36.4 kb and the cpDNAs from 124.1 to 125.0 kb (Supplementary material: Table S1). These mtDNAs contain a total of 65–67 genes including 35 protein-coding genes (PCGs), 25–26 tRNA genes, three rRNA genes, and two to three conserved open reading frames (ORFs) (Oudot-Le Secq et al. 2006; Liu et al. 2015; Liu and Pang 2016b, c; Liu et al. 2016a, b; Bi and Zhou 2016; Hughey and Gabrielson 2017), whereas the cpDNAs carry a total of 173 genes including 137 PCGs, 28 tRNA genes, six rRNA genes, and two conserved ORFs (Le Corguillé et al. 2009; Liu and Pang 2016a; Yang et al. 2016; Bi et al. 2017; Graf et al. 2017).

Thus far, the complete mitochondrial genome has been sequenced for 12 species of *Sargassum*, and 3 species have had their plastid genomes sequenced (Supplementary material: Table S1). *Sargassum horneri* is the first *Sargassum* species to have its mtDNA and cpDNA sequenced (Liu et al. 2015; Liu and Pang 2016a). Comparison of the fully sequenced mitochondrial and plastid genomes showed that genome size, gene content, and architecture are highly conserved within the *Sargassum* genus (Liu et al. 2017a, b). A major important advantage of mtDNA and cpDNA over the diploid nuclear genome in brown algae is that organelle genomes are effectively haploid, with no heterozygosity. This suggests that this genetic information could potentially be used as effective molecular markers to track the source of the drifting *Sargassum* biomass to manage golden tides (Amaral-Zettler et al. 2017; Liu et al. 2017b). Furthermore, the richness of the organelle genomic data can greatly promote our understanding of the evolutionary history and phylogenetics of brown algae (Liu et al. 2017a, b).

In this work, we report the complete mitochondrial and chloroplast genomes of the brown alga *S. confusum* and performed comparative genomic and phylogenomic analysis in the ecologically important genus *Sargassum*.

Materials and methods

Sampling and DNA extraction

The drifting adult plants of *Sargassum confusum* C.Agardh were collected in the Yellow Sea (32° 40′–34° 00′ N, 120° 30′–121° 45′ E) on a cruise on the R/V Sutongyu-01026 from 4 March to 12 March 2017 surveying the abundance of drifting *Sargassum horneri* biomass. *Sargassum confusum* was a minor constituent of the golden tide biomass in the Yellow Sea in spring 2017 (Liu et al. 2018). Algal thalli were transported to the laboratory in coolers (5–8 °C) after collection. Frozen tissue from the original algal samples was used for DNA extraction. Algal tissue was ground to fine powder in liquid nitrogen. Total DNA was extracted using a Plant Genomic DNA Kit (Tiangen Biotech, China) according to the manufacturer's instructions. The quality and concentration of isolated DNA were evaluated using the electrophoresis on a 1.0% agarose gel.

Genome sequencing and assembly

The whole mitochondrial and chloroplast genome sequences of *S. confusum* were determined using long PCR and primer walking sequencing techniques (Cheng et al. 1994). Six primer sets were used to amplify the complete *S. confusum* mitochondrial genome into six large fragments and 15 primer sets to amplify the chloroplast genome into 15 fragments according to Liu et al. (2015) and Liu and Pang (2016a), respectively. Long PCRs were performed in 50 μL reaction mixtures containing 10 μL of 5× PrimeSTAR GXL buffer (5 mM Mg^{2+} plus, Takara, Japan), 4 μL of dNTP mixture (2.5 mM each), 1 μL of each primer (10 μM), 1 μL of PrimeSTAR GXL DNA polymerase (1.25 units μL^{-1}, Takara, Japan), 1 μL of DNA template (approximate 50 ng), and 32 μL of sterile distilled H_2O. PCR amplification was carried out on a TC1000-G Thermal Cycler (Scilogex, USA) with an initial denaturation at 94 °C for 3 min, followed by 30–35 cycles of denaturation at 94 °C for 20 s, annealing at 50–52 °C for 50 s, extension at 68 °C for 1 min kb^{-1}, and a final extension at 68 °C for 10 min. PCR products were purified using a QIAquick Gel Extraction Kit (Qiagen, Germany). Sequencing reactions were performed using ABI 3730 XL automated sequencers (Applied Biosystems, USA). The DNA sequences were manually edited and assembled using BioEdit v7.1.9 (Hall 1999) and Geneious 7.1 software (Biomatters, http://www.geneious.com). DNA sequences of the complete mitochondrial and chloroplast genomes were determined by comparison with published mtDNA and cpDNA sequences of *Sargassum* species. This resulted in the 34,721-bp scaffold for mtDNA and the 124,375-bp scaffold for cpDNA.

Genome annotation and analysis

The locations of the protein-coding genes (PCGs) were determined with DOGMA (Wyman et al. 2004), Open Reading Frame Finder (http://www.ncbi.nlm.nih.gov/gorf/orfig.cgi), and Blastx (GenBank). Transfer RNA (tRNAs) genes were identified by reconstructing their cloverleaf structures using the tRNAscan-SE 1.21 software with default parameters (Lowe and Chan 2016). Ribosomal RNA (rRNAs) genes were

identified by BLAST searches (Altschul et al. 1997) of the nr database at the National Center for Biotechnology Information (NCBI). The mtDNA and cpDNA of *S. confusum* have been deposited in GenBank with the accession numbers of MG459430 and MG459429, respectively.

Base composition and codon usage of mtDNAs and cpDNAs were examined by MEGA 7.0 software (Kumar et al. 2016). Sequence datasets composed of newly sequenced genomes as well as those of previously reported data available in Genbank were subjected to concatenated alignments using a ClustalW with MEGA 7.0. So far, only four *Sargassum* species including *S. confusum*, *S. thunbergii*, *S. horneri*, and *S. vachellianum* have complete mtDNAs and cpDNAs sequenced (Supplementary material: Table S1). Single-gene sequences (nt and aa) in mtDNAs and cpDNAs as well as genome sequences from these four species were separately aligned and manually adjusted using MEGA 7.0. The identity percentages of mitochondrial and chloroplast gene or genome sequences were evaluated using the BioEdit v7.1.9 software (Hall 1999). Analysis of the number of base substitutions per site from averaging overall sequence pairs was conducted using the maximum composite likelihood model by a bootstrap procedure (1000 replicates) using MEGA 7.0.

Phylogenomic analysis

The phylogenies inferred from whole mitochondrial and plastid genomes in Fucales were constructed with 1000 bootstrap replicates with the maximum likelihood (ML) method. The ML tree was obtained based on the Kimura two-parameter model (Kimura 1980). The datasets were further analyzed by Bayesian inference (BI) using MrBayes v.3.2 (Huelsenbeck and Ronquist 2001; Ronquist and Huelsenbeck 2003). Four Markov chains were run for 1,000,000 generations to allow for adequate time for convergence. Every 1000th generation was saved and the first 100 generations were discarded as burn-in. The remaining trees were used to estimate the 50% majority rule consensus tree and the Bayesian posterior probability values. *Fucus* species were selected as out-group taxa. Positions containing gaps and missing data were eliminated in DNA sequences.

Results and discussion

Genome features of mtDNA and cpDNA

The circular-mapping mitochondrial genome of *S. confusum* is 34,721 bp in size and carries 35 PCGs, 25 tRNA genes, three rRNA genes, and two conserved ORFs (Supplementary material: Fig. S1). The tRNA gene situated between *nad4* and *nad5* was annotated as *trnI*(*uau*), which was different from that in *Sargassun aquifolium* [trnK(uuu)] (Liu et al. 2017b). The 124,375-bp cpDNA of *S. confusum* possesses conserved canonical quadripartite structures with a 73,552-bp large single-copy (LSC) region separated from the 39,941-bp small single-copy (SSC) region by two 5441-bp inverted repeats (IRs), each of which encodes three rRNA genes and two tRNA genes. The *S. confusum* cpDNA contains 137 PCGs, 28 tRNA genes, six rRNA genes, and two conserved ORFs (Supplementary material: Fig. S2). Only one intron with the size of 220 bp is detected in the *trnL*(*uaa*) gene in cpDNA. The genome size, gene content, and genome architecture in mtDNA and cpDNA of *S. confusum* bear close resemblance to that of previously reported *Sargassum* species (Supplementary material: Table S1).

The A+T contents of mtDNA and cpDNA in *S. confusum* are 63.43 and 69.65%, respectively. The cpDNAs tend to be much richer in A+T than mtDNAs. This is not unique to *Sargassum* species, with other brown macroalgae exhibiting similar A+T contents (Supplementary material: Table S1). The A+T ranges from 68.81% (in *Dictyopteris divaricata*) to 71.06% (in *Fucus vesiculosus*) in cpDNAs (Liu et al. 2017a), and from 62.01% (in *Pylaiella littoralis*) to 68.01% (in *Colpomenia peregrina*) in mtDNAs (Liu and Pang 2015). The intergenic spacer content in mtDNA (4.59%) is significantly lower than that in cpDNA (13.80%) (Table 1), indicating a higher level of compactness of mtDNA than that of cpDNA.

Nearly all the PCGs and ORFs in *S. confusum* mtDNA and cpDNA have an ATG start codon, with the only exception being the plastid *psbF* which starts with a GTG codon, a conserved phenomenon within all known *Sargassum* cpDNAs (Liu and Pang 2016a; Yang et al. 2016; Bi et al. 2017). The TAA stop codon is the most commonly used in both mtDNA and cpDNA (77.70 vs 67.57%), but the codon usage biases are different in TAG and TGA stop codons. The

Table 1 Comparison of general features of mtDNA and cpDNA in *Sargassum confusum*

	Accession number	Size (bp)	A+T content (%)	Spacer content (%)	Gene content	Start codon (%)		Stop codon (%)		
					rRNAs/tRNAs/PCGs/ORFs	ATG	GTG	TAA	TAG	TGA
mtDNA	MG459430	34,721	63.43	4.59	3/25/35/2	100.00	0.00	67.57	16.22	16.22
cpDNA	MG459429	124,375	69.65	13.80	6/28/137/2	99.28	0.72	77.70	18.71	3.60

mtDNA shows the same usage frequency for TAG (16.22%) and TGA (16.22%), while the cpDNA has a tendency towards TAG (18.71%) compared with TGA (3.60%) (Table 1).

The cpDNAs are more conserved than mtDNAs

The mtDNA and cpDNA sequence pairs in four *Sargassum* species including *S. confusum*, *S. thunbergii*, *S. horneri*, and *S. vachellianum* were compared. These are the only *Sargassum* species to have complete genomic data from both compartments. The identity values in mtDNAs (88.2–92.6%) were much lower than those in cpDNAs (94.7–97.6%) (Table 2), indicating the mtDNAs in the four *Sargassum* species are more variable in nucleotide sequences than the cpDNAs. The average evolutionary divergence over mtDNA sequence pairs within four *Sargassum* species was 10.57 ± 0.13%, which is higher than the value over cpDNA pairs (3.31 ± 0.02%). More base pair substitutions have accumulated in the mtDNAs than those in the cpDNAs. A total of 5991 substitution sites occupied 17.14% of *Sargassum* mtDNAs, while 7574 substitution sites were detected in cpDNAs and accounted for 6.04%.

Functional groups of tRNAs and rRNAs displayed higher average identity values than PCGs (including conserved ORFs), which was the shared feature in *Sargassum* mtDNAs and cpDNAs (Table 2). For each functional gene group, average identity values of cpDNA genes were much higher than those in mtDNA genes, indicating that *Sargassum* chloroplast genes were more conserved than mitochondrial genes in each group. A total of 15 chloroplast tRNA gene sequences were identical for four *Sargassum* species, whereas only one mitochondrial tRNA gene, *trnP*(*ugg*), was completely identical. In four *Sargassum* cpDNAs, *rrn5* was very conserved with an identity value of 100%, followed by *rns* (99.48%), and *rnl* (99.02%). The *rrn5* was the most variable rRNA gene in mtDNAs with an average identity value of 89.58%, compared to *rnl* (95.17%) and *rns* (95.53%).

Two chloroplast PCGs involved in photosystem II, *psbF* and *psbJ*, are the most conserved, with average values of 99.20 and 99.15%, respectively. The two most conserved PCGs in mtDNAs are *atp9* (96.88%) and *atp8* (95.20%), which are involved in ATP synthase. The average identity values in nearly all chloroplast PCGs are > 92.40%, with the exception of *rpl32* (85.33%) (Fig. 1). The *rpl32* in *S. horneri* cpDNA is 51 bps shorter than those present in the other three *Sargassum* species, due to the premature termination codon TAA. Only four (*atp8*, *atp9*, *nad4L*, and *nad3*) of mitochondrial PCGs have identity values of > 92.40%. Two mitochondrial PCGs, *rpl31* and *orf131*, have the lowest identity values of 83.83 and 83.25%, respectively (Fig. 1).

Although the gene content and genome architecture of mtDNAs and cpDNAs were conserved in *Sargassum* (Liu et al. 2017a, b), we did not know which (mtDNAs or cpDNAs) are more conserved in this lineage before. Mitogenomes were originally thought to be more conserved than chloroplast genomes. In contrast to similar studies on terrestrial plants and dinoflagellates, which showed a more conserved mtDNA than cpDNA (Wolfe et al. 1987; Smith and Keeling 2015), a few species from various algal lineages (e.g., glaucophytes, red algae, green algae, and haptophytes) have shown to have higher mutation rates in the mitochondrion than in the chloroplast (Smith 2015). The brown algae are classified within the group of stramenopiles and contain plastids from red algae as the result of a secondary endosymbiotic event (Keeling 2010; Yang et al. 2012). Most lineages with red algal-derived plastids have an mtDNA/ptDNA mutation rate ratio of > 1, but the ratio changed drastically in different lineages (Smith 2015). The mitochondrial genomes have an approximately three-fold greater mutation rate than chloroplast genomes in *Sargassum* species, even in brown algae (data not given). This feature could be effectively used by developing novel molecular markers for evolutionary analyses and phylogenetics, even making function in tracking the source of the golden tides.

Table 2 The comparison of identity percentages (%) in mitochondrial and chloroplast sequence pairs of genome sequences, tRNA genes, rRNA genes, and PCGs (including the conserved ORFs, nt, and aa) within four *Sargassum* species including *S. confusum* (Sco), *S. thunbergii* (Sth), *S. horneri* (Sho), and *S. vachellianum* (Sva)

		Identity (%)					
		Sco/Sth	Sco/Sho	Sco/Sva	Sth/Sho	Sth/Sva	Sho/Sva
Genome	mtDNA	92.6	88.2	89.6	88.6	90.4	88.3
	cpDNA	97.6	95.3	95.9	95.5	96.1	94.7
tRNA	mtDNA	97.6 (2.3)	95.8 (3.0)	96.8 (2.1)	96.0 (2.7)	96.7 (2.2)	96.3 (2.4)
	cpDNA	99.8 (0.5)	99.6 (0.7)	99.4 (0.9)	99.6 (0.7)	99.5 (0.8)	99.6 (0.9)
rRNA	mtDNA	93.7 (4.2)	91.8 (5.7)	91.7 (6.3)	93.6 (2.2)	94.4 (1.6)	95.4 (0.2)
	cpDNA	99.6 (0.4)	99.4 (0.5)	99.5 (0.5)	99.5 (0.4)	99.5 (0.4)	99.5 (0.4)
PCG (nt)	mtDNA	92.4 (2.6)	87.9 (3.7)	89.3 (3.4)	88.7 (3.4)	90.3 (3.0)	88.7 (3.6)
	cpDNA	98.2 (0.9)	96.3 (2.5)	96.8 (1.3)	96.5 (2.3)	97.0 (1.3)	96.0 (2.5)
PCG (aa)	mtDNA	93.8 (4.1)	89.7 (7.3)	90.8 (6.9)	90.0 (6.8)	91.4 (5.8)	89.8 (7.2)
	cpDNA	98.7 (1.6)	97.3 (3.5)	97.8 (2.1)	97.4 (3.5)	97.9 (2.1)	97.1 (3.6)

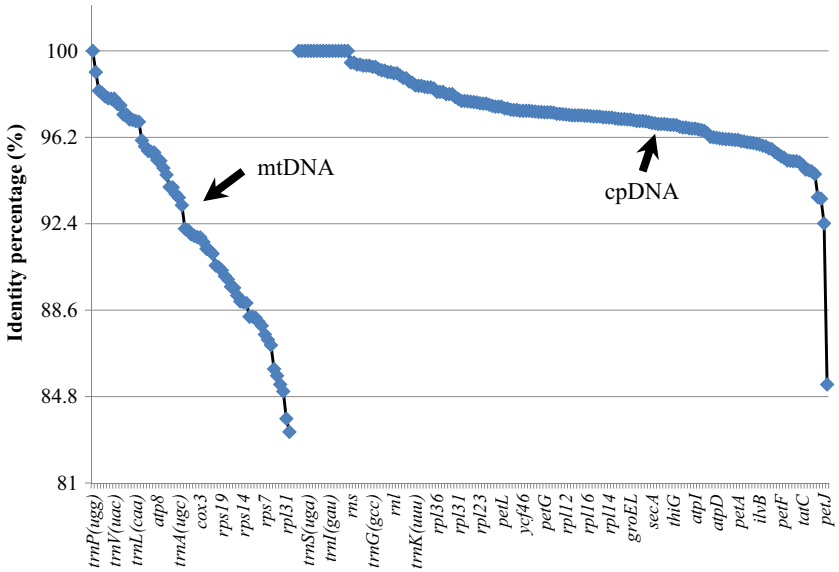

Fig. 1 The comparison of average identity percentages (%) in single mitochondrial (left) and chloroplast (right) gene sequences (nt) in four *Sargassum* species. The mtDNA and cpDNA datasets included 65 and 173 genes, respectively

Phylogenomic analysis

Based on mtDNA and cpDNA datasets, our phylogenomic analysis showed that *S. confusum* combined with other reported *Sargassum* species formed the *Sargassum* clade with high support values (ML/BI = 100%) (Fig. 2). It is worth noting that mitochondrial phylogenomics positioned *S. confusum* in a subclade with *S. muticum*, supporting the close relationship between *S. muticum* and *S. confusum*. The genetic distance of mtDNAs between these two species was only 2.66% (Supplementary material: Table S2). Previously, we found that the *cox3-atp6* spacer regions displayed apparent variation in size among *Sargassum* species (Liu et al. 2017b). However, this spacer region in *S. confusum* was 105 bp in size and identical to that in *S. muticum*. Two species shared 13 tRNA genes with the same nt sequences and four PCGs (*atp8*, *atp9*, *nad4L*, and *orf39*) with identical aa sequences (Table 3). A total

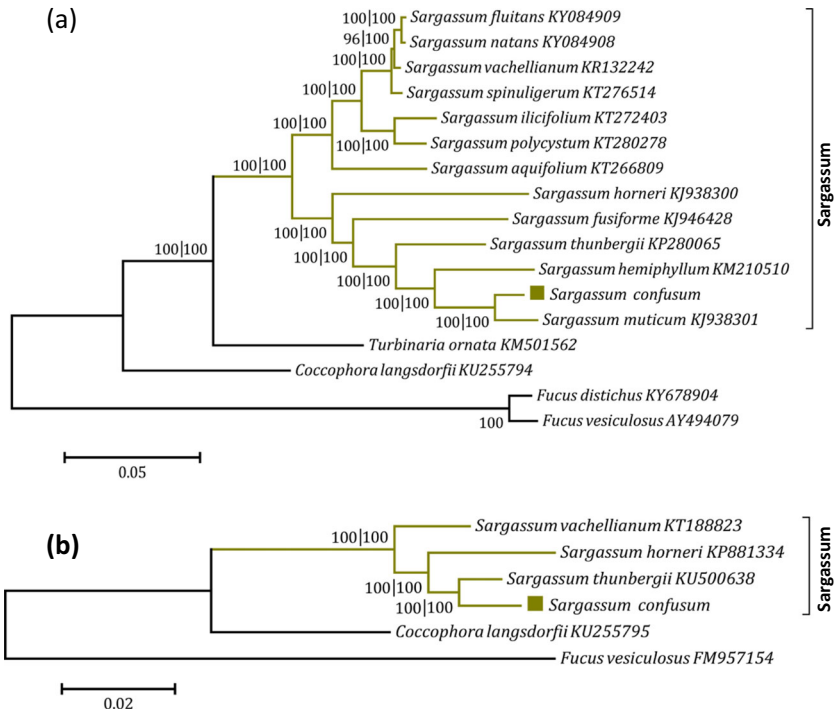

Fig. 2 Phylogenomic trees constructed from analyses of the known mitochondrial (**a**) and chloroplast (**b**) genomes in *Sargassum*. The trees were rooted with *Fucus* species. The numbers at internal nodes (ML/BI) indicated maximum likelihood (ML) bootstrap values, and Bayesian inference (BI) posterior probability values, respectively. The legend below represents the scale for nucleotide substitutions

Table 3 Identity percentages (%) of specific mtDNA sequences from *S. confusum* and *S. muticum*

	Identity (%)			Identity (%)			Identity (%)
	nt	aa		nt	aa		nt
Mitogenome	97.2	–	*rpl16*	97.1	98.5	*trnE(uuc)*	98.6
atp6	97.2	98.7	*rpl31*	95.7	92.8	*trnF(gaa)*	98.6
atp8	98.7	100.0	*rps2*	96.0	97.9	*trnG(gcc)*	98.6
atp9	99.1	100.0	*rps3*	97.2	97.8	*trnH(gug)*	100.0
cob	96.4	98.3	*rps4*	97.7	99.6	*trnI1(gau)*	100.0
cox1	97.7	99.4	*rps7*	95.1	95.1	*trnI2(uau)*	100.0
cox2	96.6	96.0	*rps8*	96.2	98.3	*trnK(uuu)*	100.0
cox3	97.3	99.6	*rps10*	98.2	99.1	*trnL1(caa)*	100.0
nad1	97.6	99.3	*rps11*	96.4	94.6	*trnL2(uaa)*	100.0
nad2	97.6	98.3	*rps12*	97.4	98.4	*trnL3(uag)*	97.5
nad3	97.5	98.3	*rps13*	96.9	98.3	*trnM1(cau)*	100.0
nad4	96.7	98.5	*rps14*	96.2	97.9	*trnM2(cau)*	100.0
nad4L	98.6	100.0	*rps19*	97.9	98.7	*trnM3(cau)*	100.0
nad5	96.8	98.3	*tatC*	95.6	93.8	*trnN(guu)*	95.8
nad6	96.9	96.5	*orf39*	98.3	100.0	*trnP(ugg)*	100.0
nad7	96.9	99.2	*orf131*	97.2	97.7	*trnQ(uug)*	97.2
nad9	97.8	99.4	*rnl* (23S)	98.5	–	*trnR(ucu)*	98.6
nad11	97.1	98.0	*rns* (16S)	98.2	–	*trnS1(gcu)*	98.8
rpl2	97.7	97.1	*rrn5* (5S)	98.4	–	*trnS2(uga)*	98.8
rpl5	97.0	97.1	*trnA(ugc)*	97.2	–	*trnV(uac)*	100.0
rpl6	96.9	97.5	*trnC(gca)*	98.6	–	*trnW(cca)*	100.0
rpl14	97.9	99.2	*trnD(guc)*	97.2	–	*trnY(gua)*	100.0

of 60 genes with identity values of > 96% were detected in mitochondrial genomes of *S. confusum* and *S. muticum*.

Our genomic data stressed the close relationship between *S. confusum* and *S. muticum*, which correlates to morphology and reproductive strategy observations. *Sargassum confusum* and *S. muticum* are two common seaweeds found along the Yellow Sea coasts and share some similar morphological characteristics in terms of stem structure, spherical vesicles, and shape of leaves (Tseng 2009). These two species are monoecious, with the same reproductive strategy (monoecism), setting them apart from other *Sargassum* species which are dioecious (Liu et al. 2013).

Conclusion

The genome size, gene content, and genome architecture of mtDNA and cpDNA were highly conserved for *Sargassum* species. However, the two organelle genomes displayed an array of distinctly different evolutionary features in terms of A+T content, spacer content, and stop codon usage. A key finding in this study was that the mtDNAs for *Sargassum* species are more variable in nucleotide sequences than cpDNAs. This result provides concrete evidence for the use of mtDNA and cpDNA sequences for evolutionary analyses and phylogenetics. Furthermore, based on the higher evolution rate in mtDNAs when compared to cpDNAs for *Sargassum* species, mtDNA is a more effective molecular marker and could aid in tracking the source of the golden tides.

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References

Altschul SF, Madden TL, Schaffer AA, Zhang J, Zhang Z, Miller W, Lipman DJ (1997) Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. Nucleic Acids Res 25: 3389–3402

Amaral-Zettler LA, Dragone NB, Schell J, Slikas B, Murphy LG, Morrall CE, Zettler ER (2017) Comparative mitochondrial and chloroplast genomics of a genetically distinct form of *Sargassum* contributing to recent "Golden Tides" in the Western Atlantic. Ecol Evol 7:516–525

Bi YH, Zhou ZG (2016) Complete mitochondrial genome of the brown alga *Sargassum vachellianum* (Sargassaceae, Phaeophyceae). Mitochondrial DNA A 27:2796–2797

Bi YH, Li JL, Zhou ZG (2017) Complete sequence of chloroplast genome from *Sargassum vachellianum* (Sargassaceae, Phaeophyceae): genome structure and comparative analysis. Aquac Fish 2:157–164

Boo GH, Lindstrom SC, Klochkova NG, Yotsukura N, Yang EC, Kim HG, Waaland JR, Cho GY, Miller KA, Boo SM (2011) Taxonomy and biogeography of the genus *Agarum* (Laminariales, Phaeophyceae) based on nuclear, mitochondrial, and plastid gene sequences. Taxon 60:831–840

Cheng S, Chang SY, Gravitt P, Respess R (1994) Long PCR. Nature 369: 684–685

China Xinhua News (2017) http://www.js.xinhuanet.com/2017-01/11/c_1120284277.htm

Duncan RS (2013) Southern wonder: Alabama's surprising biodiversity. The University of Alabama Press, Tuscaloosa, p 459

Engel CR, Billard E, Voisin M, Viard F (2008) Conservation and polymorphism of mitochondrial intergenic sequences in brown algae (Phaeophyceae). Eur J Phycol 43:195–205

Graf L, Kim YJ, Cho GY, Miller KA, Yoon HS (2017) Plastid and mitochondrial genomes of *Coccophora langsdorfii* (Fucales, Phaeophyceae) and the utility of molecular markers. PLoS One 12(11):e0187104

Guiry MD, Guiry GM (2018) AlgaeBase. World-wide electronic publication. National University of Ireland, Galway http://www.algaebase.org; searched on 7 February 2018

Hall TA (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. Nucl Acid Symp Ser 41:95–98

Huelsenbeck JP, Ronquist F (2001) MrBAYES: Bayesian inference of phylogeny. Bioinformatics 17:754–755

Hughey JR, Gabrielson PW (2017) The complete mitogenome of the rockweed *Fucus distichus* (Fucaceae, Phaeophyceae). Mitochondrial DNA P B 2:203–204

Keeling PJ (2010) The endosymbiotic origin, diversification and fate of plastids. Philos Trans R Soc B 365:729–748

Kimura M (1980) A simple method for estimating evolutionary rate of base substitutions through comparative studies of nucleotide sequences. J Mol Evol 16:111–120

Komatsu T, Matsunaga D, Mikami A, Sagawa T, Boisnier E, Tatsukawa K, Aoki M, Ajisaka T, Uwai S, Tanaka K, Ishida K, Tanoue H, Sugimoto T (2008) Abundance of drifting seaweeds in eastern East China Sea. J Appl Phycol 20:801–809

Komatsu T, Fukuda M, Mikami A, Mizuno S, Kantachumpoo A, Tanoue H, Kawamiya M (2014) Possible change in distribution of seaweed, *Sargassum horneri*, in Northeast Asia under A2 scenario of global warming and consequent effect on some fish. Mar Pollut Bull 85: 317–324

Kumar S, Stecher G, Tamura K (2016) MEGA7: molecular evolutionary genetics analysis version 7.0 for bigger datasets. Mol Biol Evol 33: 1870–1874

Laffoley D'A, Roe HSJ, Angel MV, Ardron J, Bates NR, Boyd LLL, Brooke S, Buck KN, Carlson CA, Causey B, Conte MH, Christiansen S, Cleary J, Donnelly J, Earle SA, Edwards R, Gjerde KM, Giovannoni SJ, Gulick S, Gollock M, Hallett J, Halpin P, Hanel R, Hemphill A, Johnson RJ, Knap AH, Lomas MW, McKenna SA, Miller MJ, Miller PI, Ming FW, Moffitt R, Nelson NB, Parson L, Peters AJ, Pitt J, Rouja P, Roberts J, Roberts J, Seigel DA, Siuda ANS, Steinberg DK, Stevenson A, Sumaila VR, Swartz W, Thorrold S, Trott TM, Vats V (2011) The protection and management of the Sargasso Sea: the golden floating rainforest of the Atlantic Ocean: summary science and supporting evidence case. Sargasso Sea Alliance, St. George's

Lane CE, Lindstrom SC, Saunders GW (2007) A molecular assessment of Northeast Pacific *Alaria* species (Laminariales, Phaeophyceae) with reference to the utility of DNA barcoding. Mol Phylogenet Evol 44:634–648

Le Corguillé G, Pearson G, Valente M, Viegas C, Gschloessl B, Corre E, Bailly X, Peters AF, Jubin C, Vaccherie B, Cock JM, Leblanc C (2009) Plastid genomes of two brown algae, *Ectocarpus siliculosus* and *Fucus vesiculosus*: further insights on the evolution of red-algal derived plastids. BMC Evol Biol 9:253

Liu F, Pang SJ (2015) Mitochondrial genome of *Turbinaria ornata* (Sargassaceae, Phaeophyceae): comparative mitogenomics of brown algae. Curr Genet 61:621–631

Liu F, Pang SJ (2016a) Chloroplast genome of *Sargassum horneri* (Sargassaceae, Phaeophyceae): comparative chloroplast genomics of brown algae. J Appl Phycol 28:1419–1426

Liu F, Pang SJ (2016b) Complete mitochondrial genome of the invasive brown alga *Sargassum muticum* (Sargassaceae, Phaeophyceae). Mitochondrial DNA A 27:1129–1130

Liu F, Pang SJ (2016c) Mitochondrial genome of *Sargassum thunbergii*: conservation and variability of mitogenomes within the subgenus *Bactrophycus*. Mitochondrial DNA A 27:3186–3188

Liu F, Pang SJ, Gao SQ, Shan TF (2013) Intraspecificgenetic analysis, gamete release performance and growth of *Sargassum muticum* (Fucales, Phaeophyta) from China. Chin J Oceanol Limnol 31: 1268–1275

Liu F, Pang SJ, Li X, Li J (2015) Complete mitochondrial genome of the brown alga *Sargassum horneri* (Sargassaceae, Phaeophyceae): genome organization and phylogenetic analyses. J Appl Phycol 27: 469–478

Liu F, Pang SJ, Chen WZ (2016a) Complete mitochondrial genome of the brown alga *Sargassum hemiphyllum* (Sargassaceae, Phaeophyceae): comparative analyses. Mitochondrial DNA A 27:1468–1470

Liu F, Pang SJ, Luo MB (2016b) Complete mitochondrial genome of the brown alga *Sargassum fusiforme* (Sargassaceae, Phaeophyceae): genome architecture and taxonomic consideration. Mitochondrial DNA A 27:1158–1160

Liu F, Jin Z, Wang Y, Bi YP, Melton JT (2017a) Plastid genome of *Dictyopteris divaricata* (Dictyotales, Phaeophyceae): understanding the evolution of plastid genomes in brown algae. Mar Biotechnol 19: 627–637

Liu F, Li XD, Che ZW (2017b) Mitochondrial genome sequences uncover evolutionary relationships of two *Sargassum* subgenera, *Bactrophycus* and *Sargassum*. J Appl Phycol 29:3261–3270

Liu F, Liu XF, Wang Y, Jin Z, Moejes FW, Sun S (2018) Insights on the *Sargassum horneri* golden tides in the Yellow Sea inferred from morphological and molecular data. Limnol Oceanogr. https://doi.org/10.1002/lno.10806

Lowe TM, Chan PP (2016) tRNAscan-SE On-line Search and contextual analysis of transfer RNA genes. Nucleic Acids Res 44:54–57

Mattio L, Payri CE (2011) 190 years of *Sargassum* taxonomy, facing the advent of DNA phylogenies. Bot Rev 77:31–70

Milledge JJ, Harvey PJ (2017) Golden tides: problem or golden opportunity? The valorization of *Sargassum* from beach inundations. J Mar Sci Eng 4:2–19

Oudot-Le Secq MP, Loiseaux-De Goër S, Stam WT, Olsen JL (2006) Complete mitochondrial genome of the three brown algae (Heterokonta: Phaeophyceae) *Dictyota dichotoma*, *Fucus vesiculosus* and *Desmarestia viridis*. Curr Genet 49:47–58

Phillips NE, Smith CM, Morden CW (2005) Testing systematic concepts of *Sargassum* (Fucales, Phaeophyceae) using portions of the *rbc*L-S operon. Phycol Res 53:1–10

Ronquist F, Huelsenbeck JP (2003) MRBAYES 3: Bayesian phylogenetic inference under mixed models. Bioinformatics 19:1572–1574

Silberfeld T, Rousseau F, Reviers B d (2014) An updated classification of brown algae (Ochrophyta, Phaeophyceae). Cryptogam Algol 35: 117–156

Smetacek V, Zingone A (2013) Green and golden seaweed tides on the rise. Nature 504:84–88

Smith DR (2015) Mutation rates in plastid genomes: they are lower than you might think. Genome Biol Evol 7:1227–1234

Smith DR, Keeling PJ (2015) Mitochondrial and plastid genome architecture: reoccurring themes, but significant differences at the extremes. Proc Natl Acad Sci U S A 112:10177–10184

Tseng CK (2009) Seaweeds in Yellow Sea and Bohai Sea of China. Science Press, Beijing, pp 363–372 (in Chinese)

Witherington B, Hirama S, Hardy R (2012) Young sea turtles of the pelagic *Sargassum* dominated drift community: habitat use, population density, and threats. Mar Ecol Prog Ser 463:1–22

Wolfe KH, Li WH, Sharp PM (1987) Rates of nucleotide substitution vary greatly among plant mitochondrial, chloroplast, and nuclear DNAs. Proc Natl Acad Sci U S A 84:9054–9058

Wyman SK, Jansen RK, Boore JL (2004) Automatic annotation of organellar genomes with DOGMA. Bioinformatics 20:3252–3255

Yang EC, Boo GH, Kim HJ, Cho SM, Boo SM, Andersen RA, Yoon HS (2012) Supermatrix data highlight the phylogenetic relationships of photosynthetic stramenopiles. Protist 163:217–231

Yang JH, Graf L, Cho CH, Jeon BH, Kim JH, Yoon HS (2016) Complete plastid genome of an ecologically important brown alga *Sargassum thunbergii* (Fucales, Phaeophyceae). Mar Genomics 28:17–20

Yotsukura N, Shimizu T, Katayama T, Druehl LD (2010) Mitochondrial DNA sequence variation of four *Saccharina* species (Laminariales, Phaeophyceae) growing in Japan. J Appl Phycol 22:243–251