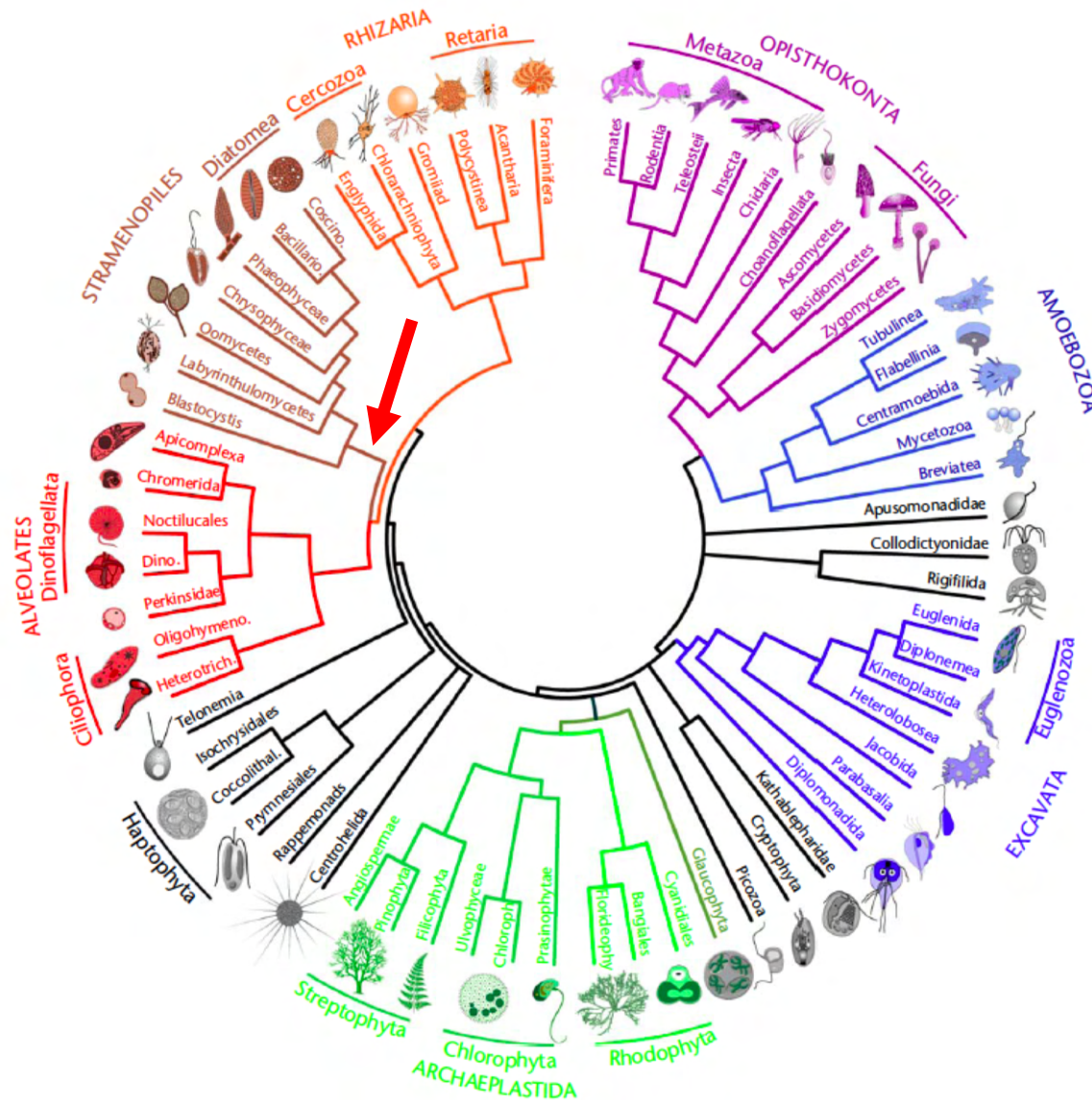


Stramenopiles / Heterokontophyta
clade Ochrophytes

Eukaryotic tree of life



Stramenopiles = Heterokonta

*historically recognized photosynthetic classes

Supergroups	Major taxa	Representative genera	Catalogued diversity
Stramenopiles	→ Bacillariophyta	<i>Thalassiosira, Nitschia</i>	20 000 *
	Bicosoecida	<i>Bicosoeca, Cafeteria</i>	72
	→ Chrysophyceae-Synurophyceae	<i>Chromulina, Ochromonas Synura</i>	1200 *
	→ Dictyochophyceae	<i>Dictyocha</i>	15
	→ Eustigmatales	<i>Vischeria</i>	15
	Hyphochytriales	<i>Hyphochytridium</i>	25
	Labyrinthulomycetes	<i>Labyrinthula, Amphitrema</i>	40
	Oomycetes	<i>Saprolegnia, Phytophthora</i>	676
	Opalinata	<i>Opalina</i>	400
	→ Pelagophyceae	<i>Aureococcus</i>	12
	→ Phaeophyceae	<i>Fucus, Laminaria</i>	1750 *
	→ Phaeothamniophyceae	<i>Phaeothamnion</i>	25
	→ Pinguiphyceae	<i>Pinguiochrysis</i>	5
	→ Raphidophyceae	<i>Goniostomum</i>	20
	→ Xanthophyceae	<i>Botrydium, Vaucheria</i>	6000.5 *

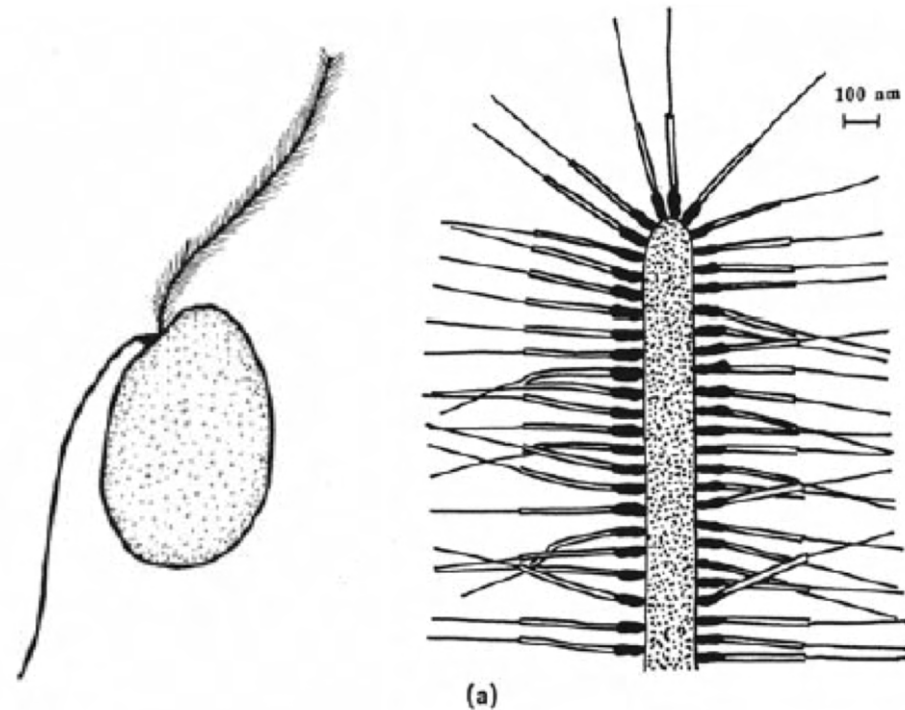
+ other less diversified classes

Bicosoecida: flagellates having tiny flagellar hairs, some genera encased in lorica; **Hyphochytriales**: flagellates having hypha-like structures; **Labyrinthulomycetes**: gliding flagellates producing a network of filaments, amoeboid; **Oomycetes** = **Peronosporomycetes**: fungus-like plant pathogens, parasites of both freshwater and marine algae; **Opalinata**: animal endobionts, flagellates mostly found in frog, with a flat shape like a slice, endocommensals.

Stramenopiles = Heterokonta

- distinctive feature are flagella [if swimming cells are present in their life-cycle]: uneven length and functionally different = heterokont flagella
- tripartite tubular hairs on the long flagellum = stramenopiles; anterior position
- no mastigonemes on the short flagellum (smooth/naked flagellum); posterior position
- but in Pelagoph. only a single, long flagellum with bipartite hairs; in some Pinguiph. a single flagella with no hairs; in Bacillarioph. a single pleuronematic (i.e. with tripartite hairs) long flagellum

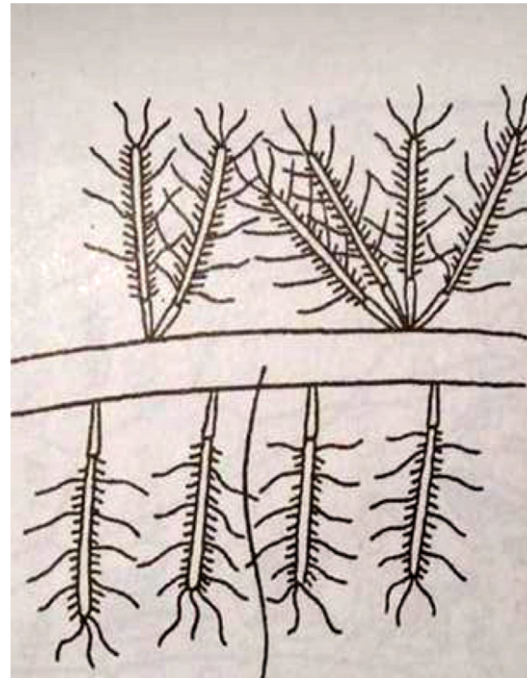
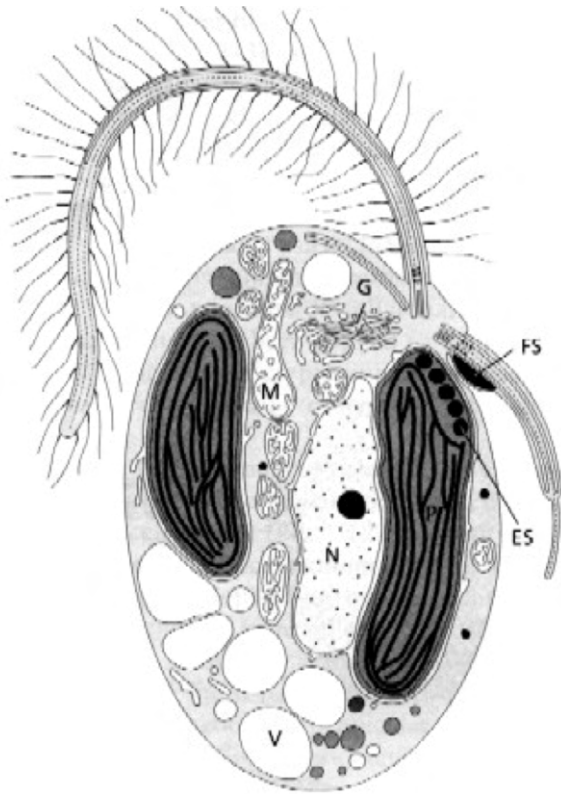
typical stramenopile flagella



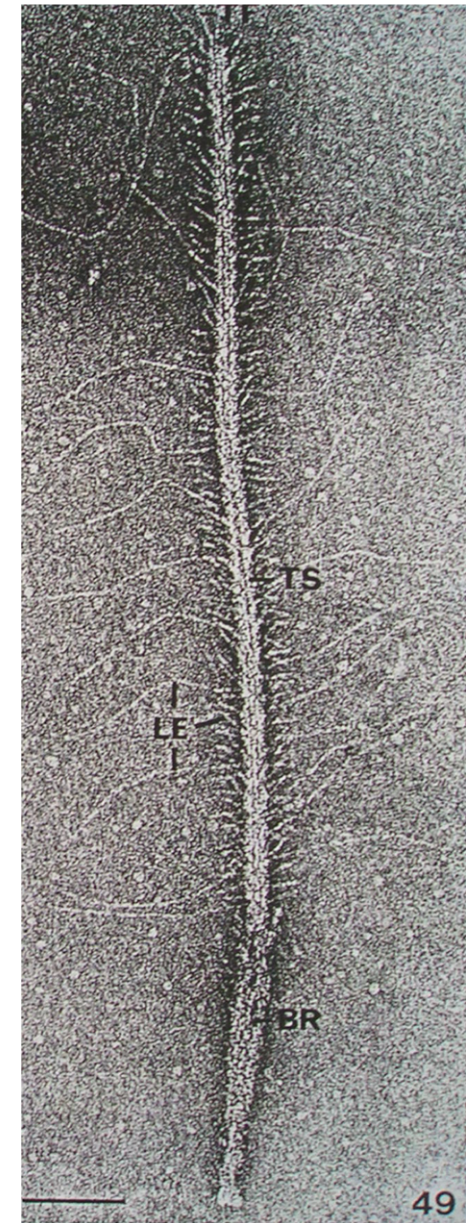
two rows of tripartite tubular hairs (basal part, microtubular part, a terminal hair or hairs), from glycoproteins, assembled in ER

Stramenopiles = Heterokonta

typical long stramenopile flagellum



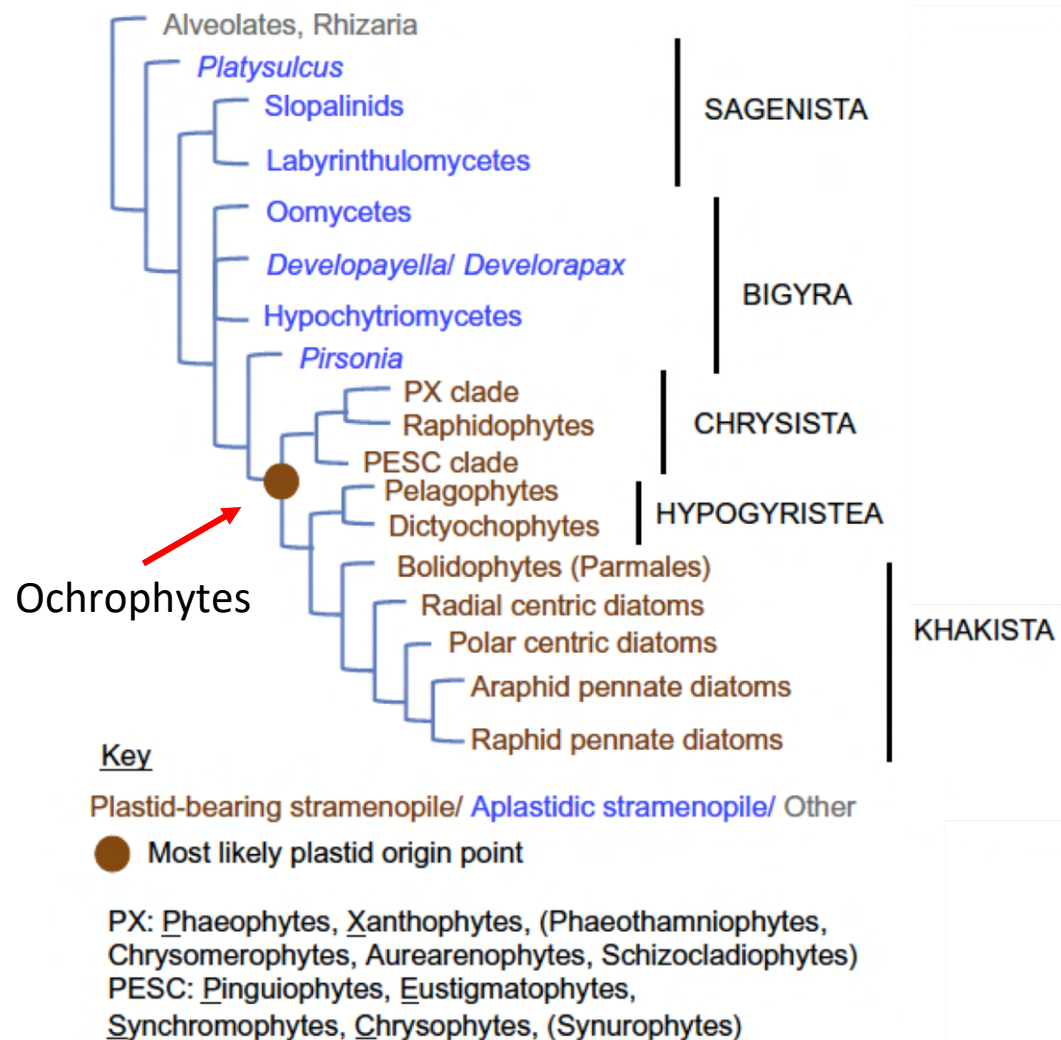
two rows of tripartite tubular hairs
BR: basal part, TS: tubular part, LE: terminal hairs



49

Ochrophytes = Chromophytes

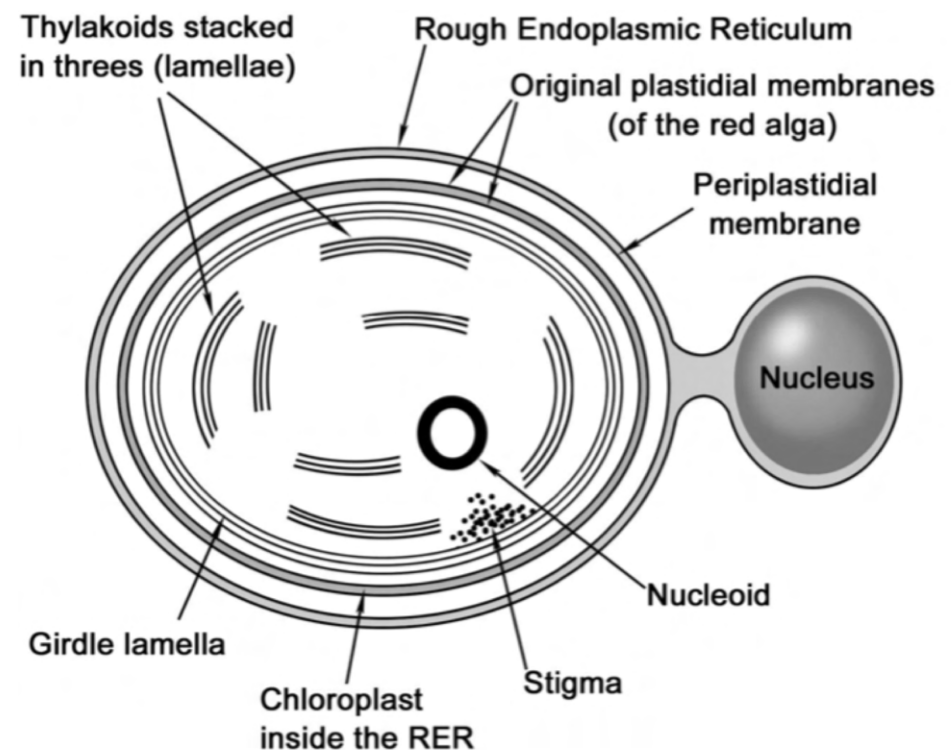
- non-photosynthetic groups basally divergent
- a recent common ancestor of the extant photosynthetic lineages [secondary lost of photosynthesis in some lineages]



Ochrophytes - plastids

- plastids: 4 membranes
[unconventional outer membrane arrangement in Synchromoph. and Aurearenoph.; only two membranes in Chrysoparadoxoph.]; outer plastid ER membrane connected with outer nuclear envelope membrane
- chlorophylls *a* + *c1*, *c2*, and/or *c3*
[*c* absent in Eustigmatoph., not detected in Aurearenoph.], various carotenoids including fucoxanthin [absent in Eustigm, Xanthoph.]
- unique thylakoid organization: girdle lamella [but not found in Eustigmatoph., Synchromoph., Aurearenoph.; some Pinguiph. and Raphidoph. species]

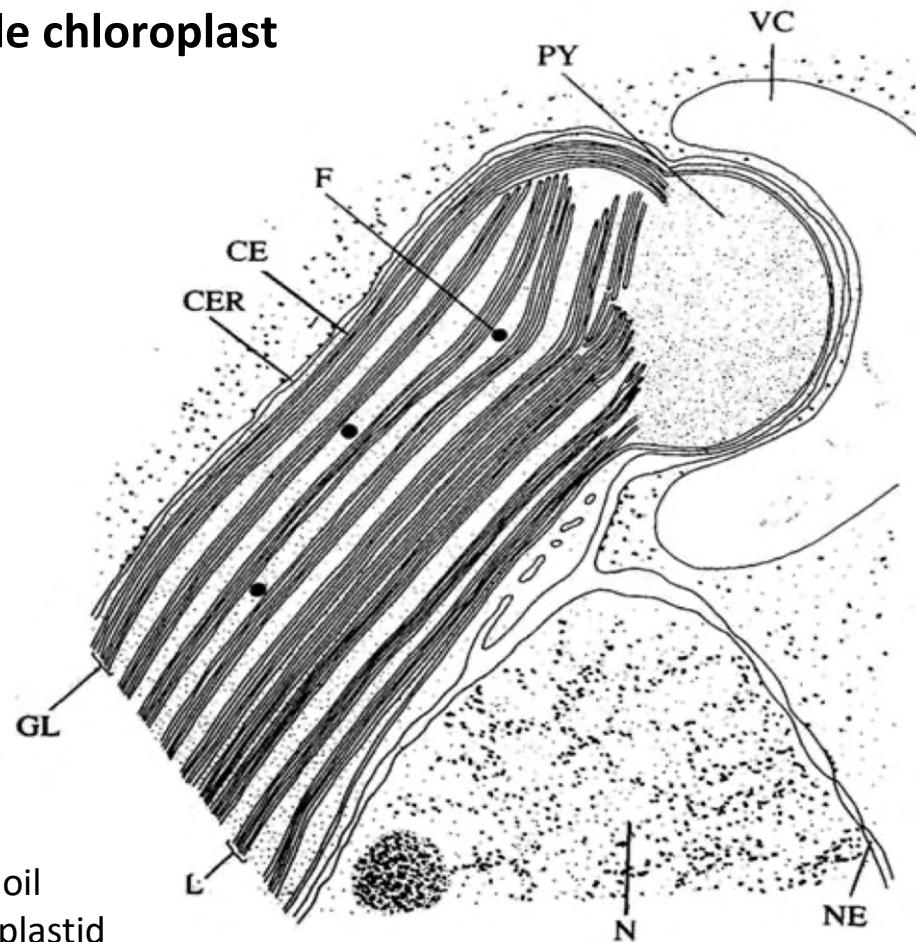
typical stramenopile chloroplast



Ochrophytes - plastids

typical stramenopile chloroplast

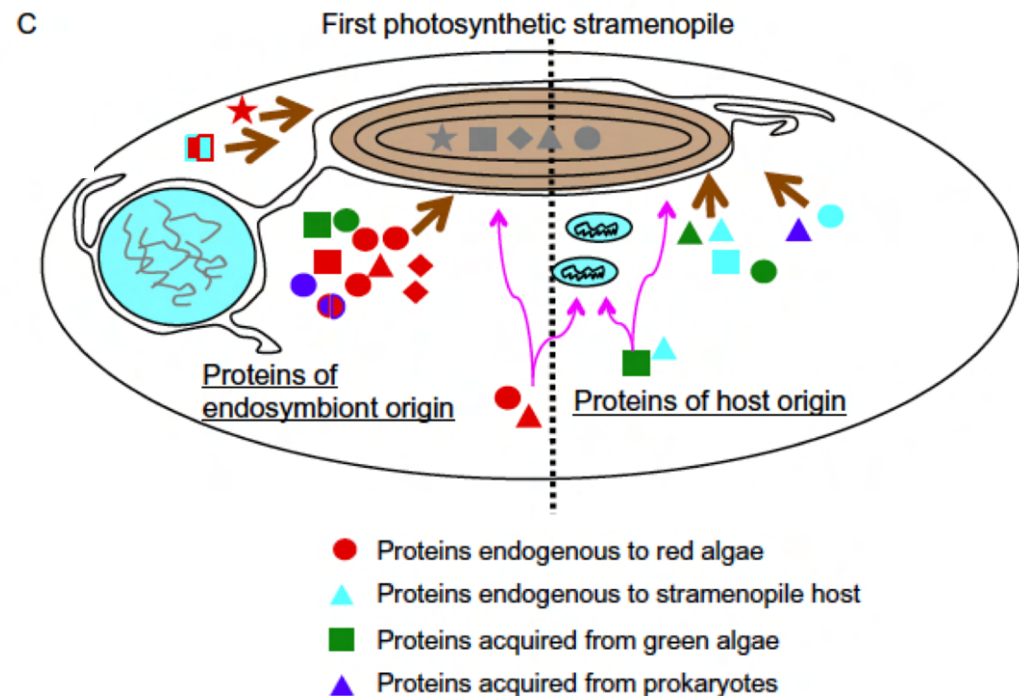
- chrysolaminaran and oil globules as a storage product (absence of starch which is characteristic for Archaeplastida)



VC: chrysolaminaran vacuole, PY: pyrenoid, F: oil globules, CE: 2 layered plastid envelope, CER: plastid ER, GL: girdle lamella, L: lamella (thylakoids stacked in triplets), N: nucleus, NE: nuclear envelope

Ochrophytes - plastids

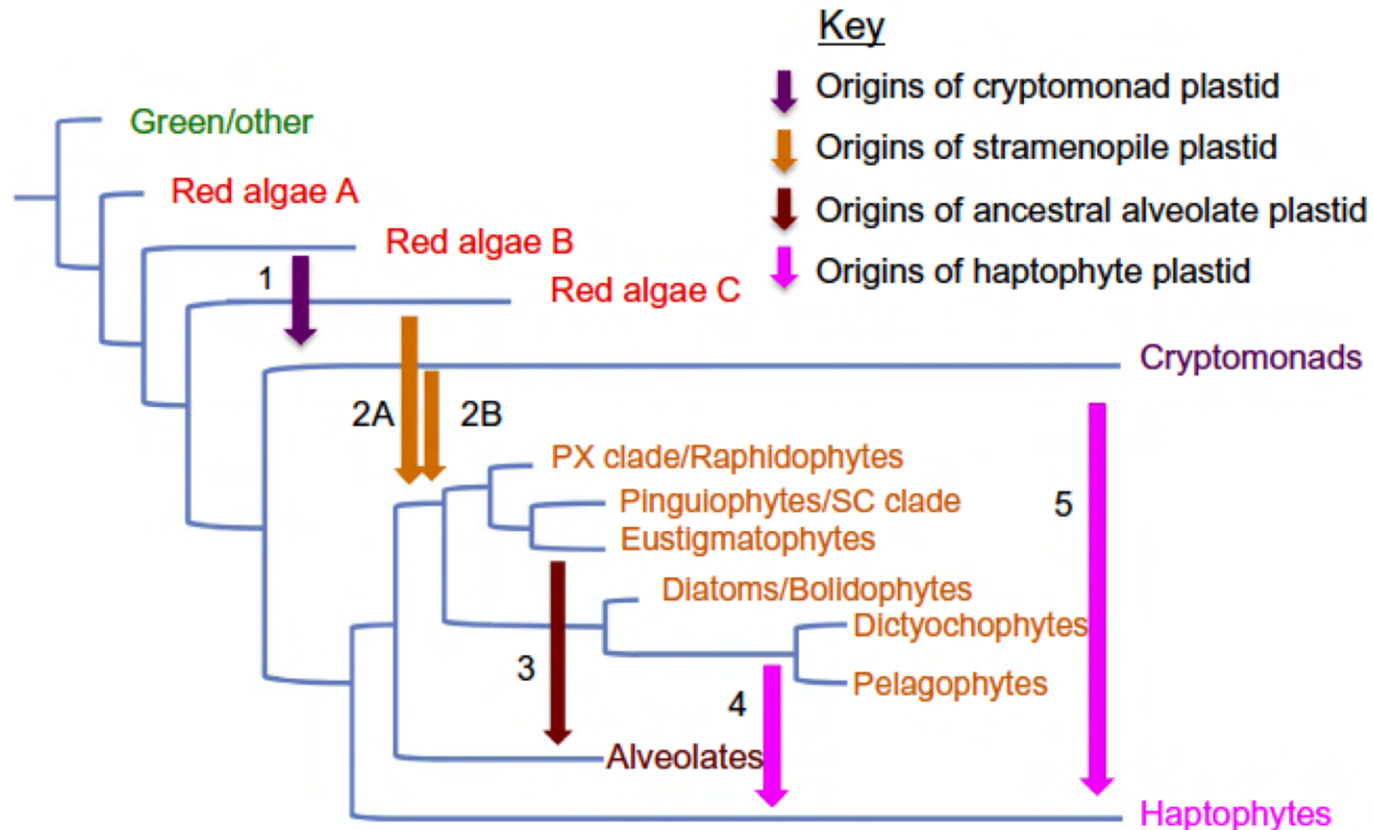
- chimeric origins of plastids:
genes of red algal origin + from
prokaryotes + from green algae
(footprint of a cryptic plastid?)
- physiological capacities known
in green algae but not known in
red algae: xanthophyll cycle
(light-harvesting function and
photoprotective role)
- fucoxanthin/chlorophyll
pigment-binding proteins: both
red and green origin



Mosaic origin of plastids, green alga and/or prokaryotic origin: i) acquired through lateral transfer events or through endosymbiosis; ii) transfer from green algae into red algae or other endosymbiont, which might be a possible vector for introducing green genes into the nucleus.

Ochrophytes - plastids

Possible endosymbiotic connections between stramenopiles and other plastids of red origin.



plastids: secondary or more complex endosymbiosis of a red alga

Ochrophytes - pigments

	Number plastids/ cell	Pyrenoid	Chlorophyll	Fucoxanthin	Xanthophylls	Girdle lamella	Plastid-nucleus membrane connection	Eyespot	ptDNA
Phaeophytes	Multiple	Some	a, c1, c2	Fuc	V/Z	+	+	Some	R
Aurearenophytes	2	+	a, c?	Fuc	D/D	–	+	+	nd
Phaeothamniophytes	1 to 3	–	a, c1, c2	Fuc	D/D, H	+	+	+	R
Schizocladiophytes	1	–	a, c?	Fuc	nd	+	+	+	R
Chrysomerophytes	1	+	a, c1, c2	Fuc	V/Z	+	+	+	R
Xanthophytes	1	Some	a, c1, c2	–	D/D, H, Va	+	+	Some	R
Raphidophytes	Multiple	+	a, c1, c2	Fuc	V/Z or D/D	Some	–	–	S
Pinguicophytes	1 or 2	+	a, c1, c2	Fuc	V/Z	Some	+	–	R/S
Eustigmatophytes	1 or 2	Some	a	–	V/Z, Va	–	+	+	R
Synchromophytes	Multiple	Some	a, c2	Fuc	V/Z	–	+	–	nd
Synurophytes	1 or 2	Some	a, c1	Fuc	V/Z	+	–	–	R
Chrysophytes	1 or 2	+	a, c1, c2	Fuc, hexFuc	V/Z	+	+	+	R
Pelagophytes	Multiple	Some	a, c1, c2	Fuc, butFuc	D/D	+	+	Some	S
Dictyochophytes	Multiple	+	a, c1, c2	Fuc	D/D	+	+	Some	S
Bolidophytes	1	Some	a, c1, c2, c3	Fuc, hexFuc	D/D	+	+	–	R
Centric diatoms	Multiple	+	a, c1, c2	Fuc, hexFuc	D/D	+	Some	–	R
Pennate diatoms	1 or 2	+	a, c1, c2	Fuc, hexFuc	D/D	+	Some	–	R

Fucoxanthins Fuc: fucoxanthin; hexFuc: 19'-hexanoyloxyfucoxanthin; butFuc: 19'-butanoyloxyfucoxanthin
Xanthophylls V/Z: violaxanthin/zeaxanthin; D/D: diatoxanthin/diadinoxanthin; H: heteroxanthin; Va: vaucherioxanthin
Plastid DNA R: ring genophore; S: scattered genophore

Ultrastructural and pigment characteristics.

Ochrophytes - pigments

	Number plastids/ cell	Pyrenoid	Chlorophyll	Fucoxanthin	Xanthophylls
Phaeophytes	Multiple	Some	a, c1, c2	Fuc	V/Z
Aurearenophytes	2	+	a, c?	Fuc	D/D
Phaeothamniophytes	1 to 3	–	a, c1, c2	Fuc	D/D, H
Schizocladophytes	1	–	a, c?	Fuc	nd
Chrysomerophytes	1	+	a, c1, c2	Fuc	V/Z
Xanthophytes	1	Some	a, c1, c2	–	D/D, H, Va
Raphidophytes	Multiple	+	a, c1, c2	Fuc	V/Z or D/D
Pinguiphites	1 or 2	+	a, c1, c2	Fuc	V/Z
Eustigmatophytes	1 or 2	Some	a	–	V/Z, Va
Synchromophytes	Multiple	Some	a, c2	Fuc	V/Z
Synurophytes	1 or 2	Some	a, c1	Fuc	V/Z
Chrysophytes	1 or 2	+	a, c1, c2	Fuc, hexFuc	V/Z
Pelagophytes	Multiple	Some	a, c1, c2	Fuc, butFuc	D/D
Dictyochophytes	Multiple	+	a, c1, c2	Fuc	D/D
Bolidophytes	1	Some	a, c1, c2, c3	Fuc, hexFuc	D/D
Centric diatoms	Multiple	+	a, c1, c2	Fuc, hexFuc	D/D
Pennate diatoms	1 or 2	+	a, c1, c2	Fuc, hexFuc	D/D

Fucoxanthins
Xanthophylls
Plastid DNA

Fuc: fucoxanthin; hexFuc: 19'-hexanoyloxyfucoxanthin; butFuc
V/Z: violaxanthin/zeaxanthin; D/D: diatoxanthin/diadinoxanthin;
R: ring genophore; S: scattered genophore

Xanthophytes, yellow-green

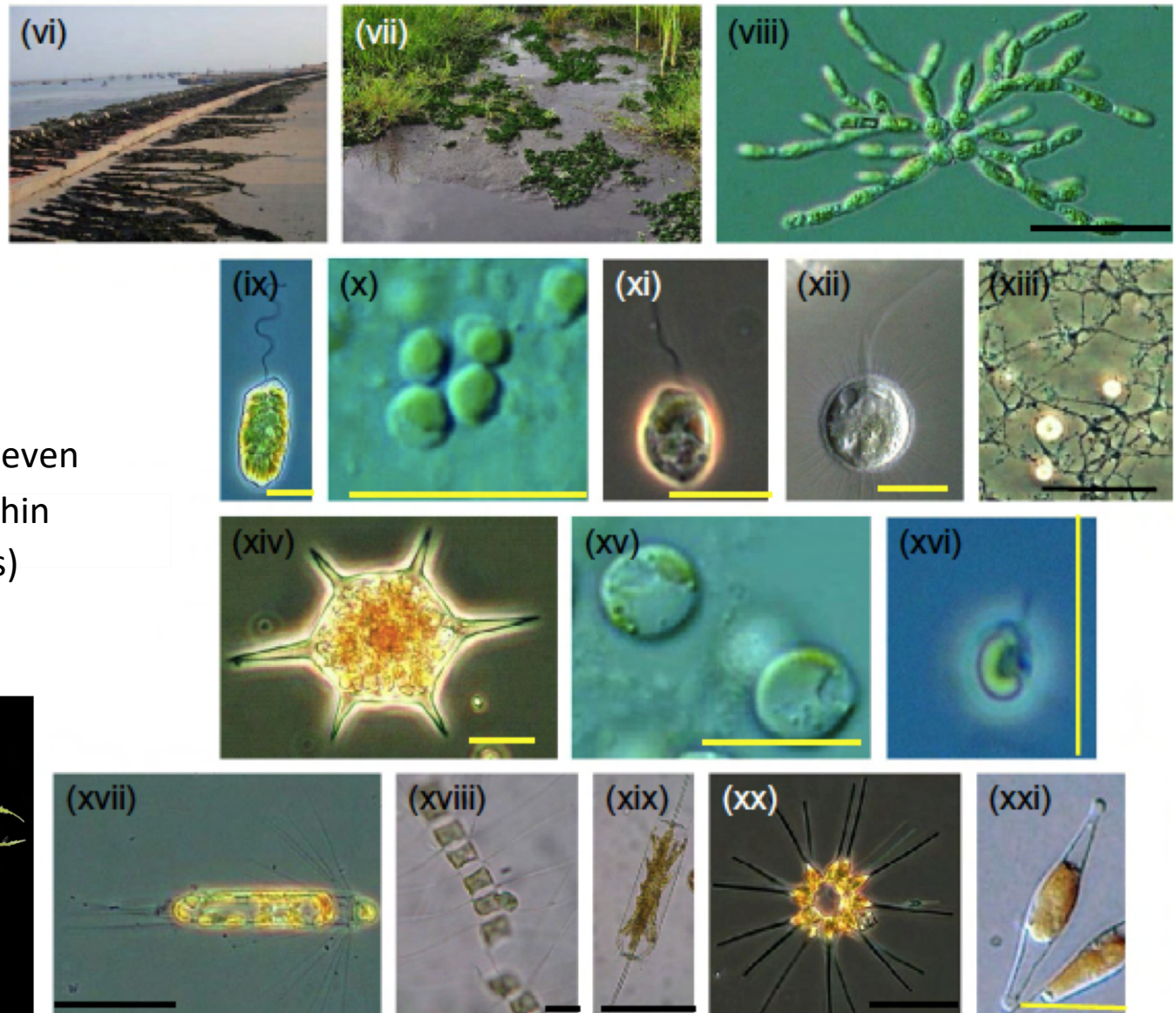


Synurophytes, golden-brown

Ultrastructural and pigment characteristics.

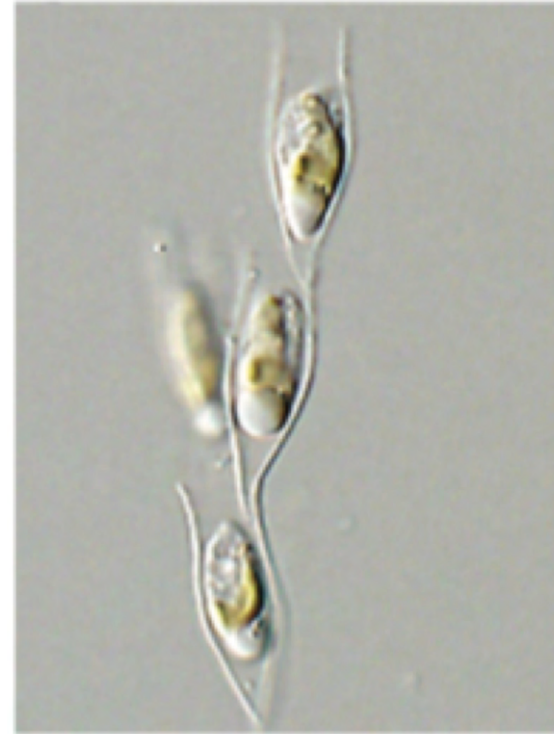
Ochrophytes - diversity

- huge morphological, physiological and phylogenetic diversity (even rivalling that observed within the green algae and plants)



Ochrophytes - trophic

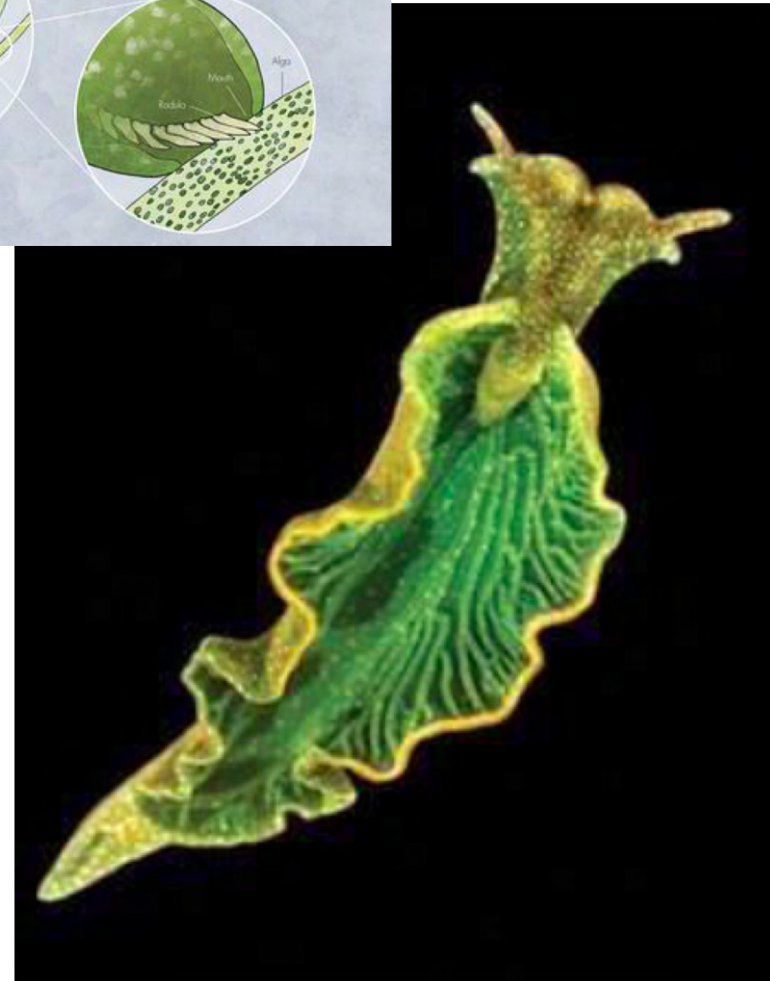
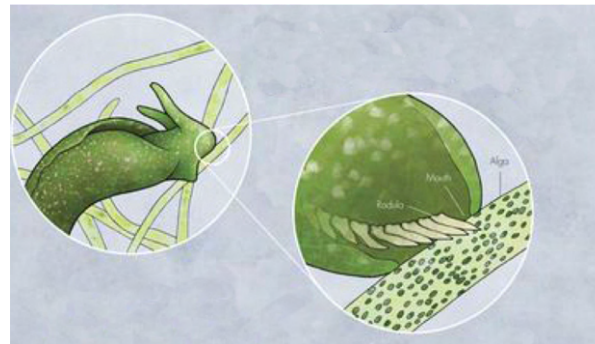
- photoautotrophy
- photo-mixotrophy: supplementing the products of photosynthesis with phagotrophy (Chrysoph., Raphidoph.) or osmotrophy (Pelagoph., Bacillarioph.)
- secondary loss of photosynthesis (e.g., Chrysoph., Dictyochoph., Bacillarioph.; some of them have leucoplasts)



Dinobryon, Chrysophyta

Ochrophytes - symbioses

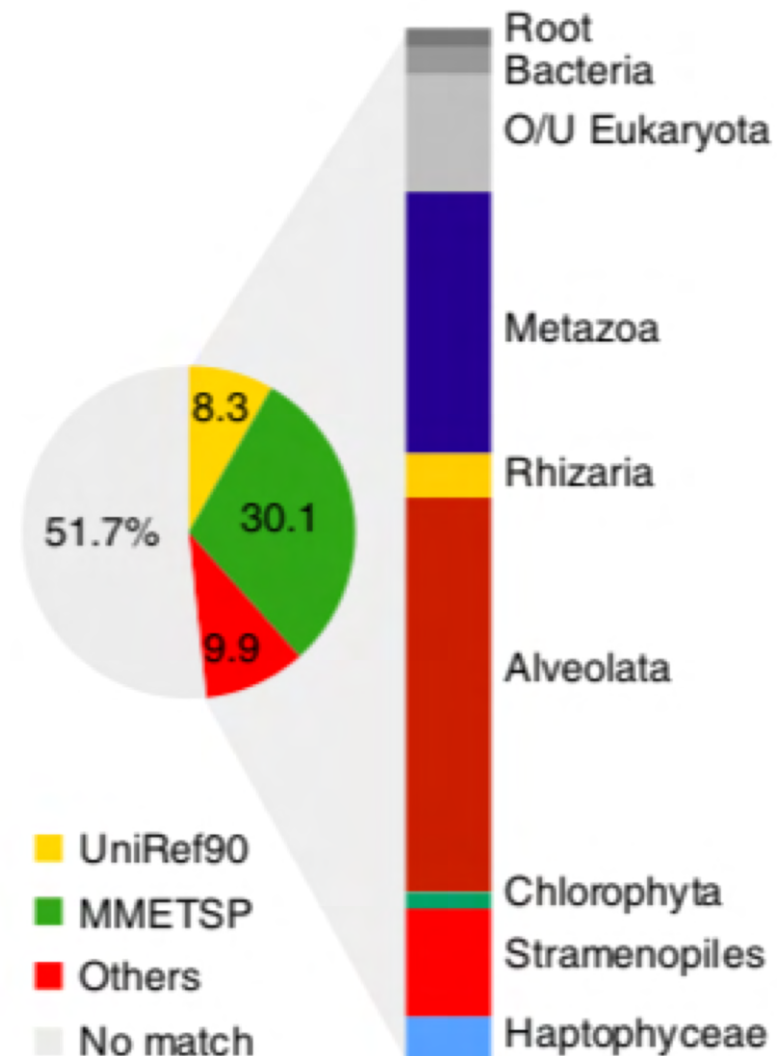
- plastids as transient symbionts by other eukaryotes = kleptoplasts: marine sea slug (*Elisia chlorotica* – from *Vaucheria*), dinoflagellates (from Dictyochoph.); foraminifera (from Bacillarioph.), "marine lichen" (*Verrucaria*: fungi and phaeophyte alga)
- dinotoms: dinoflagellates with permanent plastids of diatom origin



Elisia chlorotica

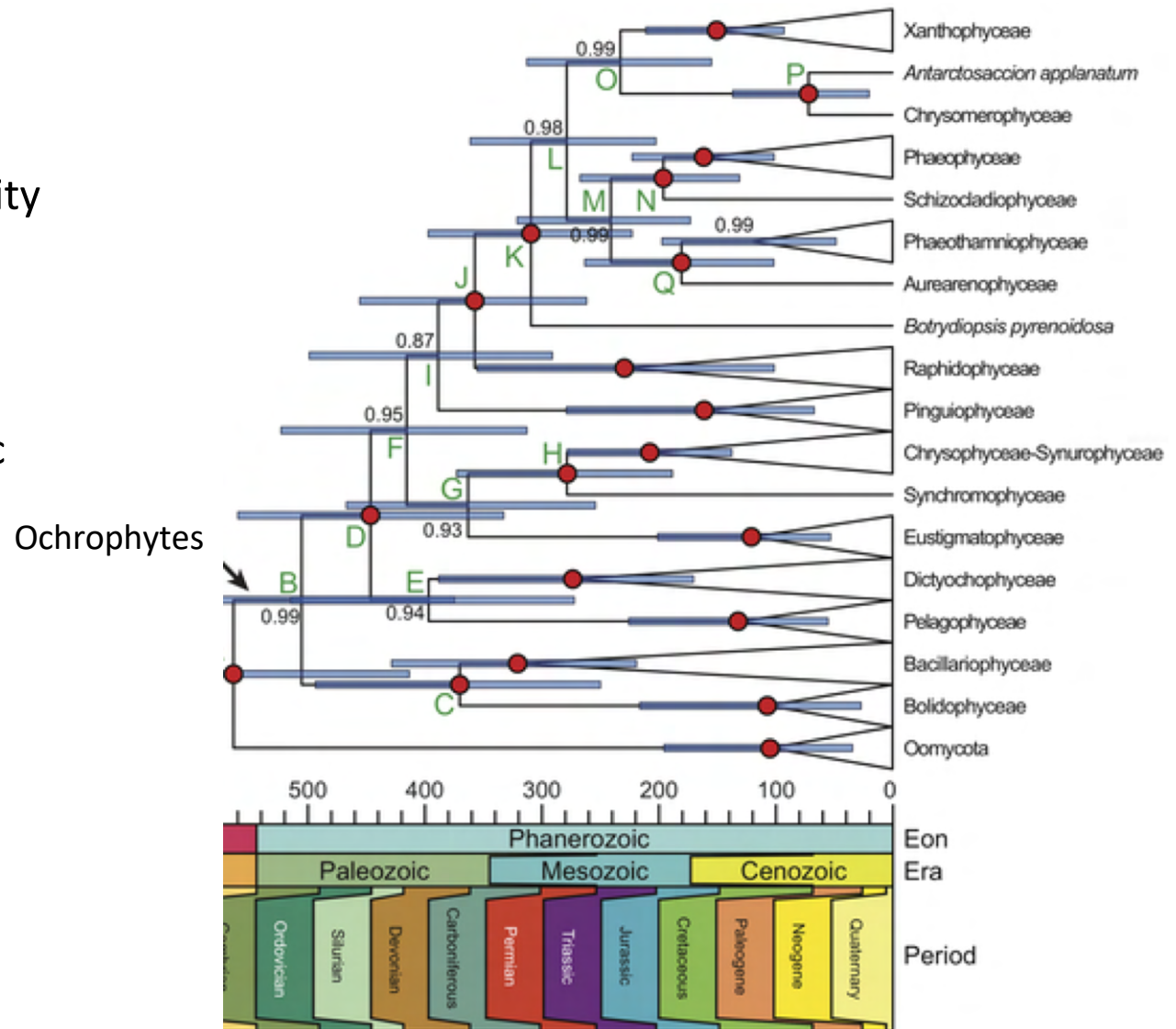
Ochrophyta – habitats

- high diversity and frequency of ochrophytes in marine habitats [but Xanthoph. and Eustigmatoph. predominantly freshwater or terrestrial]
- ecological function (food webs, biogeochemical cycles) of contemporary marine ecosystems are critically dependent on eukaryotic phytoplankton (despite they are numerically inferior to cyanobacteria)

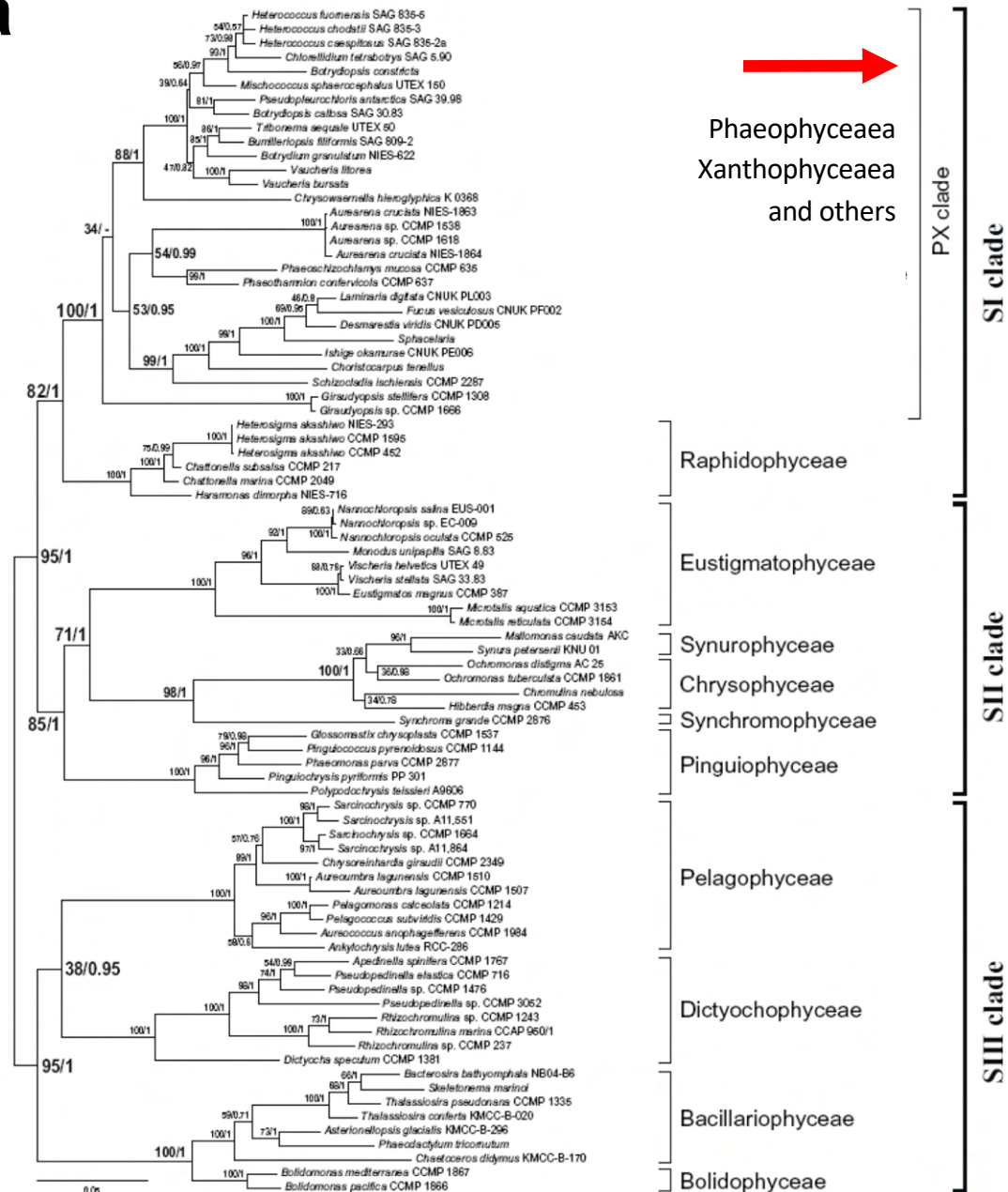


Ochrophytes - history

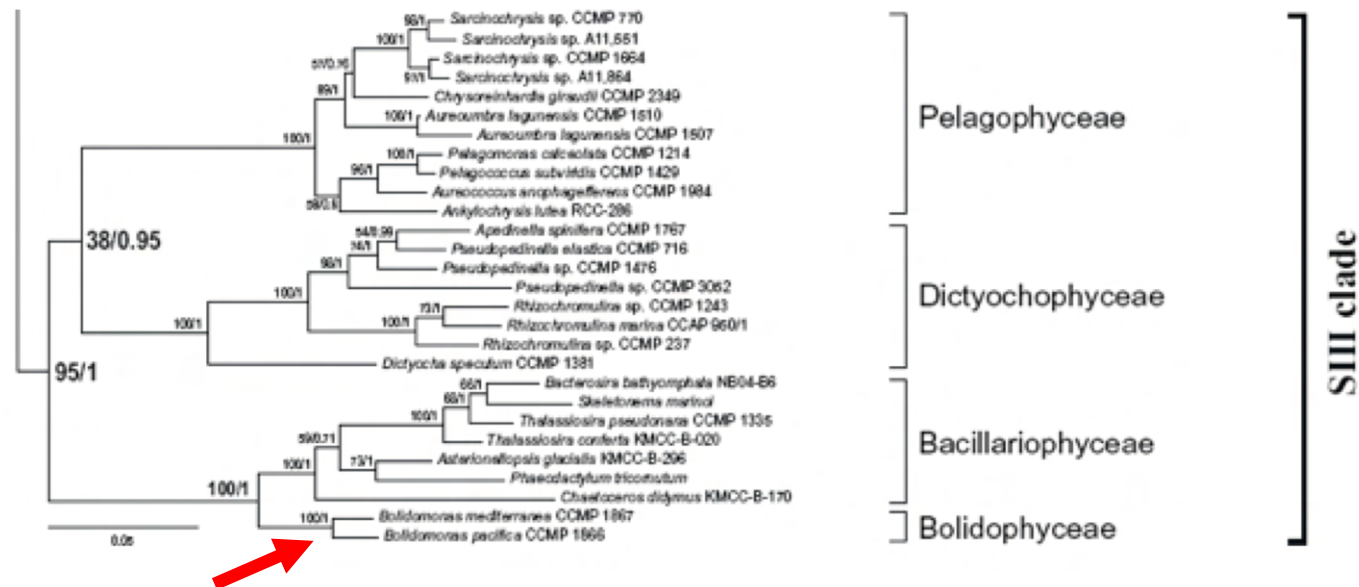
- the radiation of the majority of the known heterokont algal classes occurred throughout the Paleozoic and in the Triassic/Jurassic of the Mesozoic



Ochrophyta

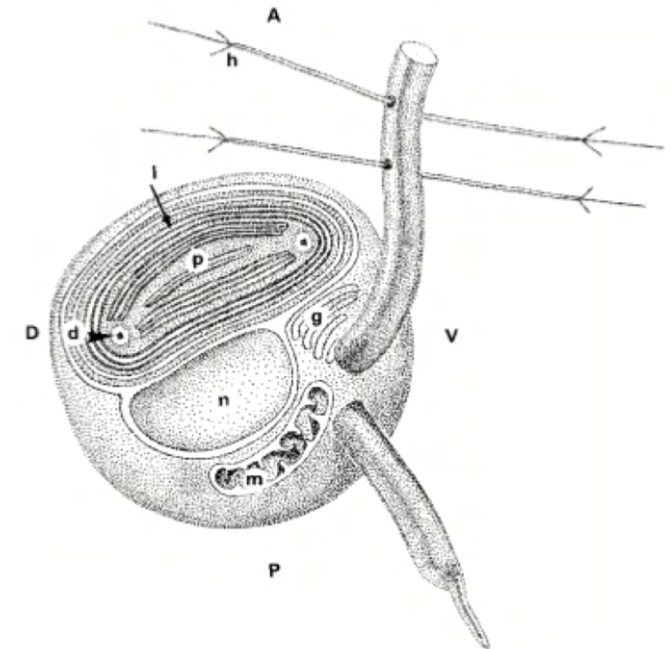


Bolidophyceae



Bolidophyceae

- marine picophytoplankton (~1.2 μm)
- heterokont naked flagellates
- typical stramenopile chloroplast; no eyespot; chlorophyll *a*, *c* + xanthophylls (including fucoxanthin)
- established in 1999, analyses of the SSU rDNA gene
- etymology: the rapid swimming behavior of the cells reminiscent of a racing car
- a sister group of diatoms



Bolidomonas mediterranea

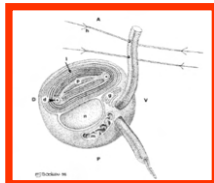
(h) tubular flagellar hair, flagellum appears bipartite because their **basal section is reduced** to a basal disk. plastid (p), nucleus (n), Golgi body (g), mitochondrion (m). The plastid has a ring DNA genophore (d), girdle lamella (l). A = anterior, D = dorsal, P = posterior, V = ventral part

Bolidophyceae ← Parmales

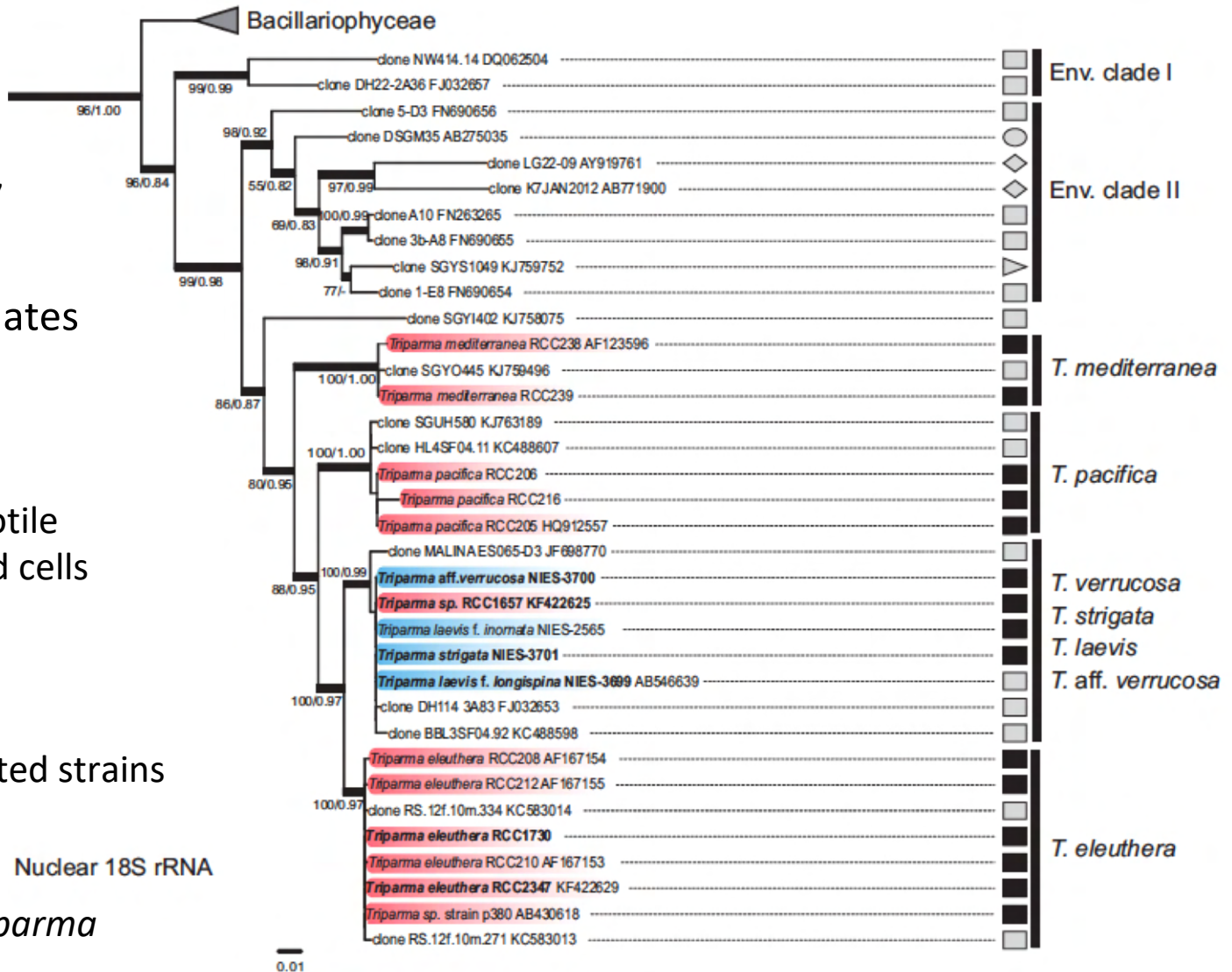
- Parmales (since 1987 within Chrysophyta) multipartite silica plates



non-motile
silicified cells



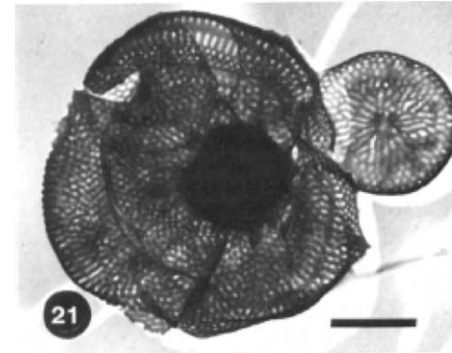
motile
flagellated strains



Bolidomonas = *Triparma*

Bolidophyceae - Parmales

- marine nanophytoplankton (~2-5 μm)
- coccoid non-motile forms
- abundant in polar and subpolar regions
- siliceous walls made up of plates (round, triradiate or oblong shape; often bearing ridges and spines and radiating lines of pores)
- etymology: small round shields



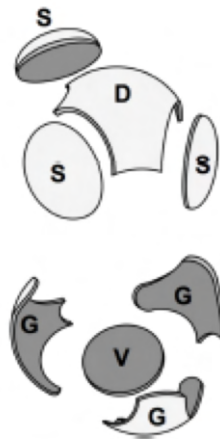
Triparma, TEM



Tetraparmella



Tetraparma: 4 round and 4 triradiate plates



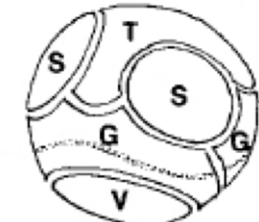
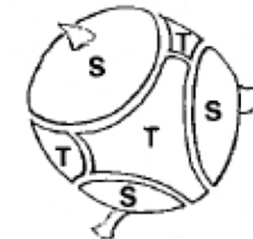
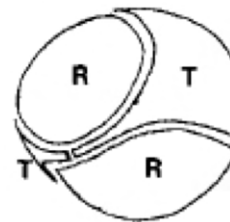
Pentalamina



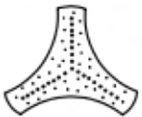
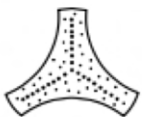
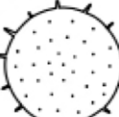
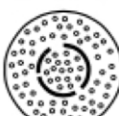
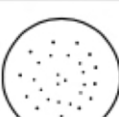
Tetraparma

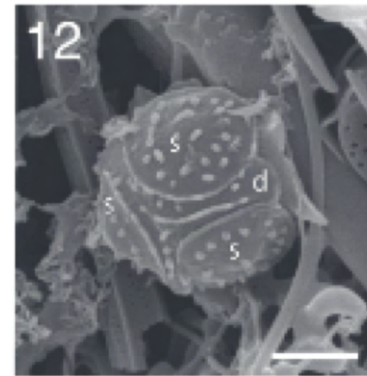


Triparma

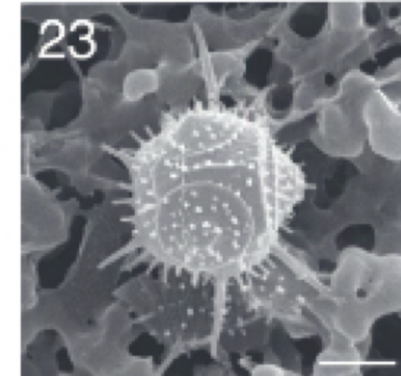


Bolidophyceae - Parmales

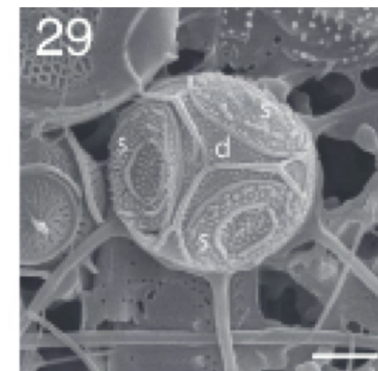
<i>Triparma</i>	dorsal plate	shield plate	girdle plate	ventral plate
<i>strigata</i> f. <i>strigata</i>				
<i>strigata</i> f. <i>subarctica</i>				
<i>strigata</i> f. <i>brevispina</i>				
<i>verrucosa</i> f. <i>verrucosa</i>				
<i>verrucosa</i> f. <i>boothiae</i>				
<i>papillosa</i>				
<i>scotiaensis</i>				



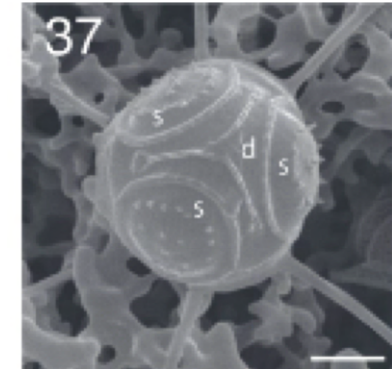
T. strigata f. *brevispina*



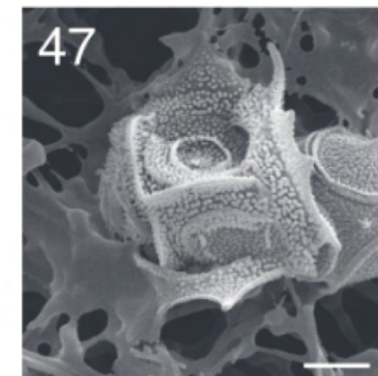
T. strigata f. *subarctica*



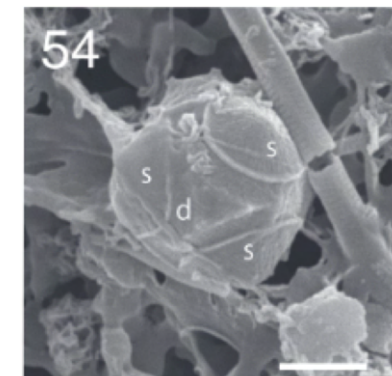
T. verrucosa f. *verrucosa*



T. verrucosa f. *boothiae*



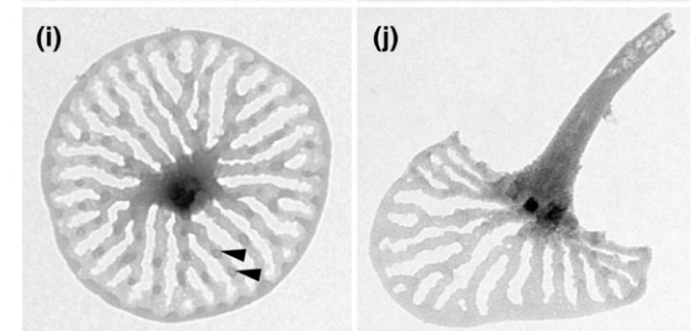
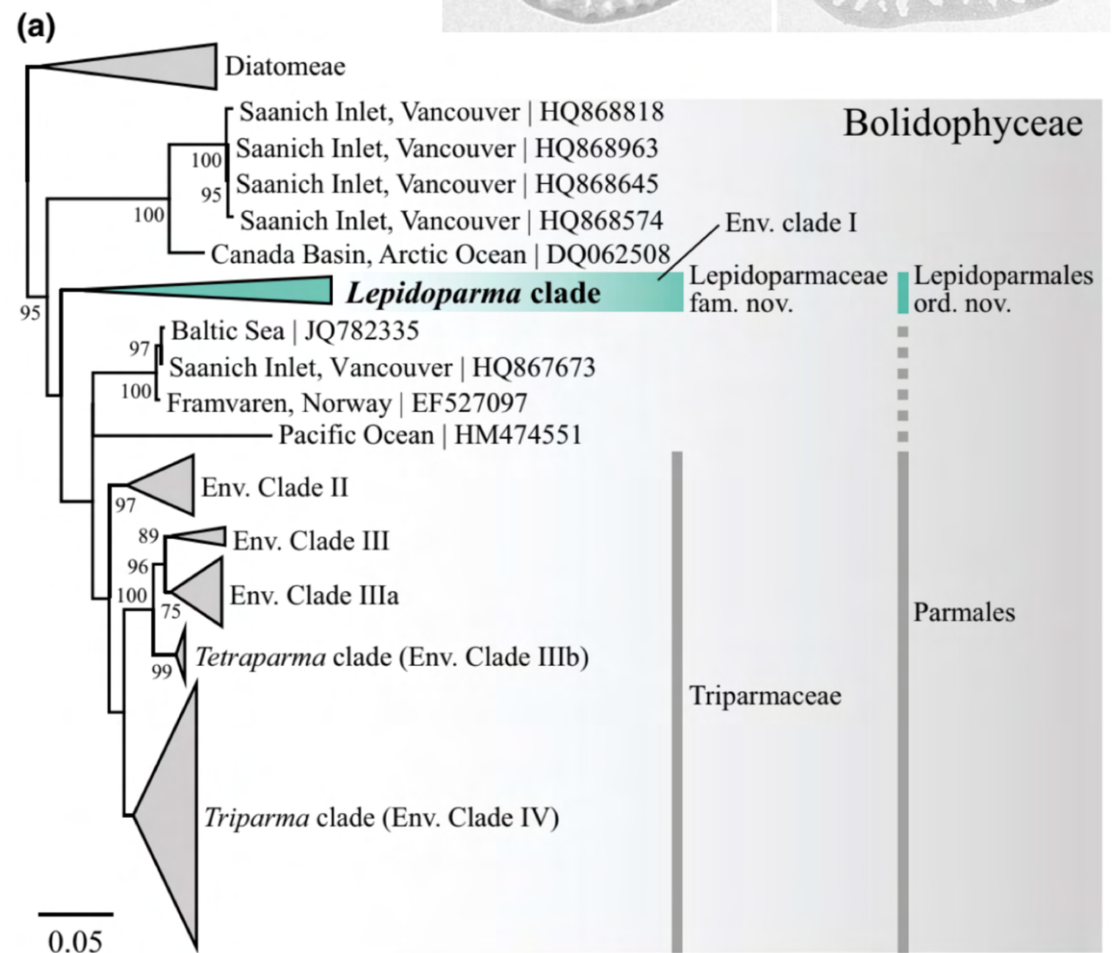
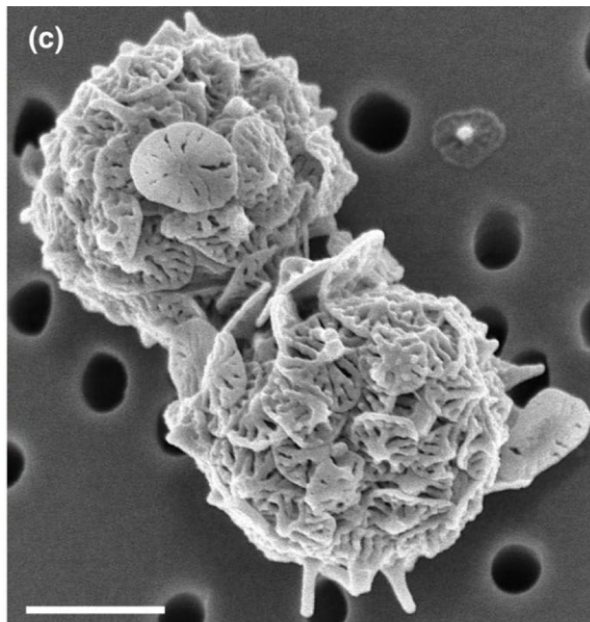
Triparma papillosa



Triparma scotiaensis

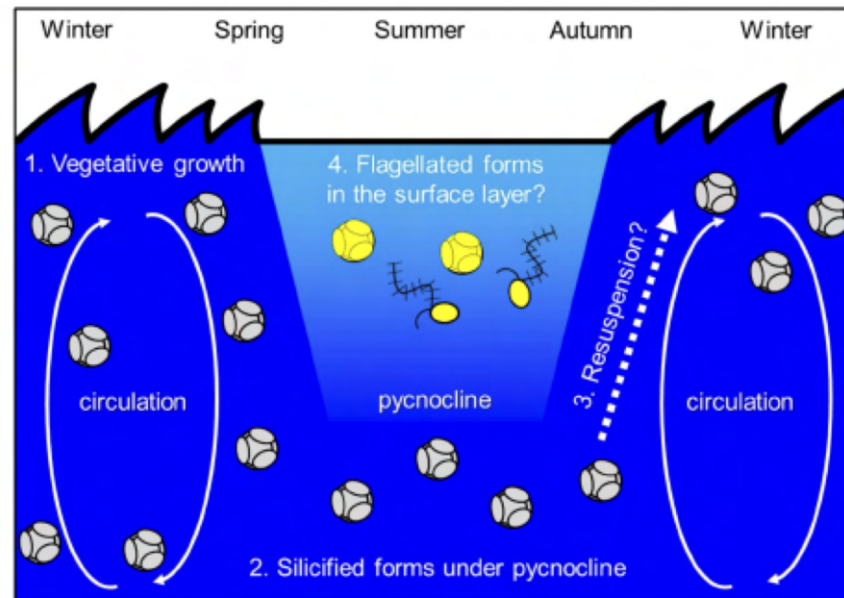
Bolidophyceae - **Lepidoparmales**

- reminiscent of the pre-diatom?
- the cell surface covered with loosely overlapping siliceous scales (radial patterns similar to centric diatoms)



Bolidophyceae - Parmales

- differences in ecological requirements between silicified and flagellated species
- the phylogenetically close relationship between silicified and naked strains (and recent occasional observation of both forms in cultures) -> different life cycle stages of the same species?



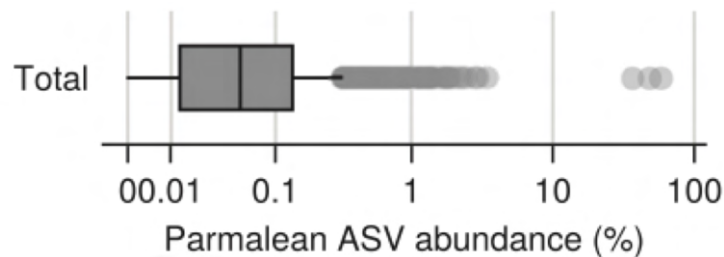
Properties

Bolidophyceae

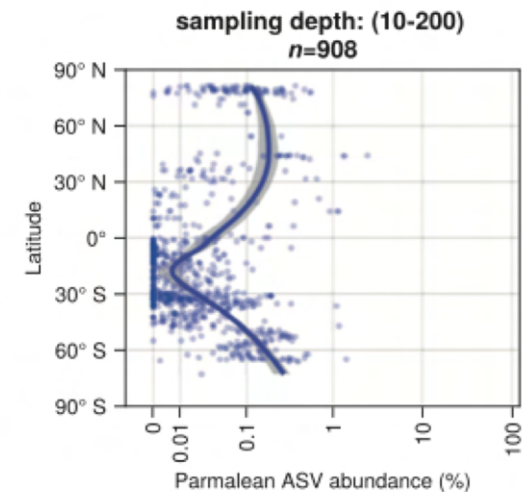
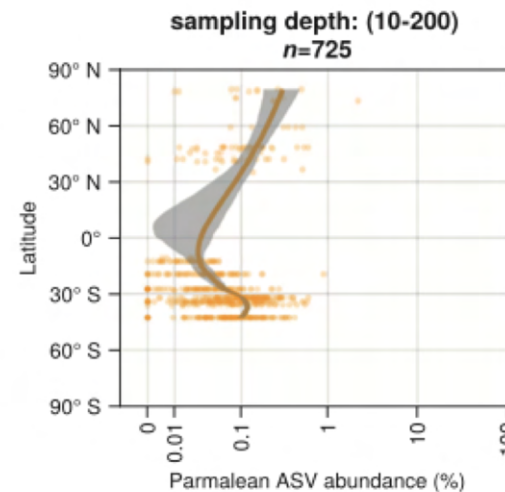
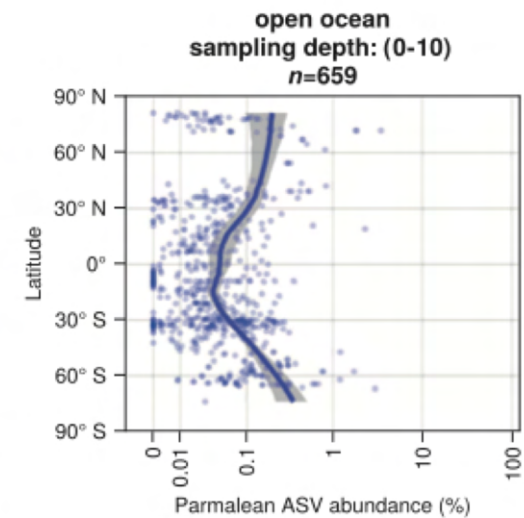
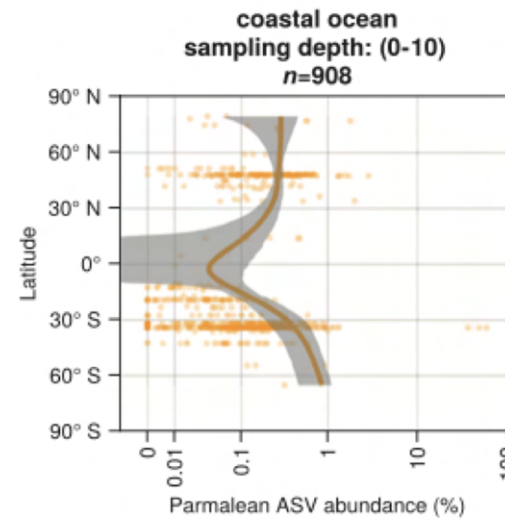
	Silicified species	Flagellated species
Size (µm)	2–5	1–1.7
Level of organization	Unicellular	Unicellular
Silicified cell wall	Yes	No
Flagellate form	Yes	Yes
Number of species	12	3
Oceanic distribution	Ubiquitous, but minor	
Main habitat	Cold eutrophic water (Polar and subpolar region)	Warm oligotrophic water (Tropical or subtropical)

Bolidophyceae - Parmales

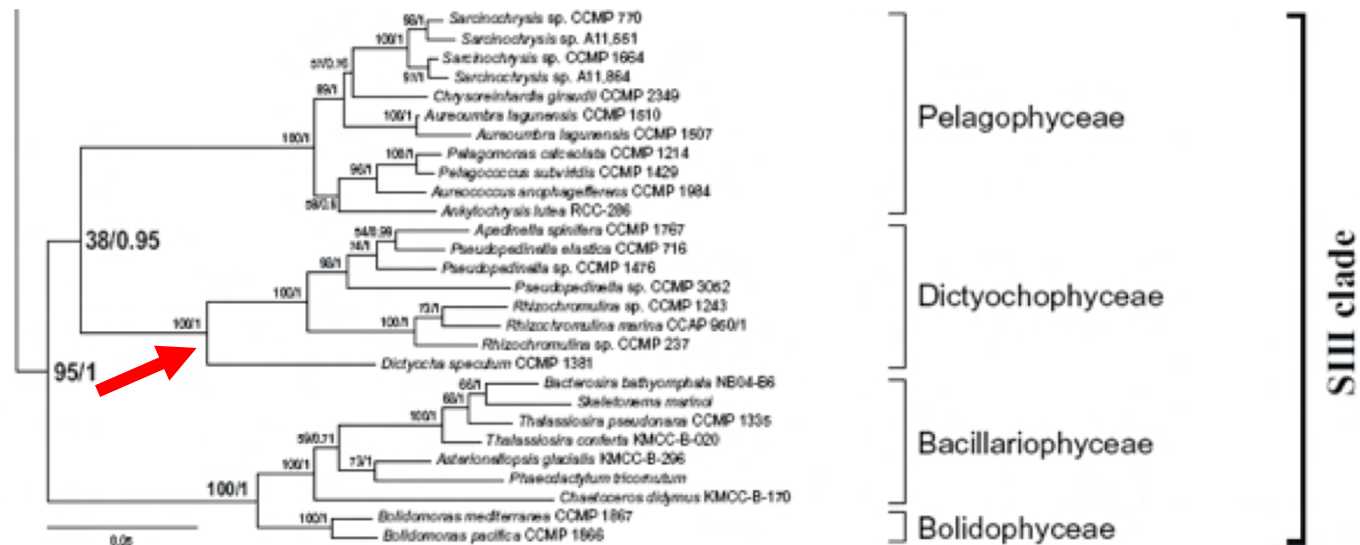
- the average contribution of Bolidoph. to total metabarcodes was less than 0.2%



- the estimated true richness was approximately 316 ASVs, confirming their low abundance and diversity
- the abundance is generally high in the poles and decreases towards the tropics



Dictyochophyceae



Dictyochophyceae

- freshwater and marine environments
- as planktonic autotrophs (some are mixotrophs, and non-photosynthetic bacterivores)
- single cells or colonial; basically amoeboid cells (tentacles/rhizopodia/pseudopodia)
- usually with one flagellum with mastigonemata
- cells naked, with organic scales or with siliceous skeleton
- typical stramenopile chloroplast; chlorophylls *a*, *c1*, *c2*, xanthophylls (including fucoxanthin)
- previously classified in the Chrysophyceae (1950-1986)



Dictyocha speculum

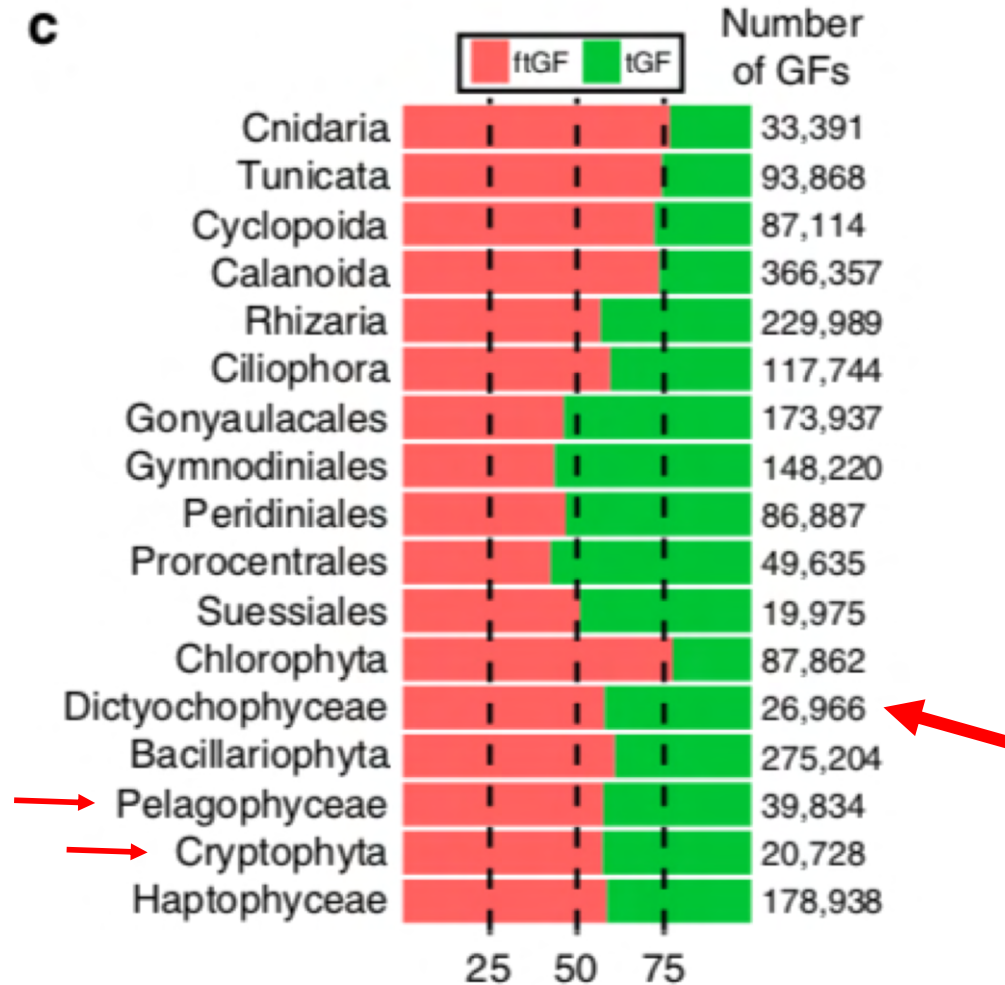


Apedinella

Dictyochophyceae

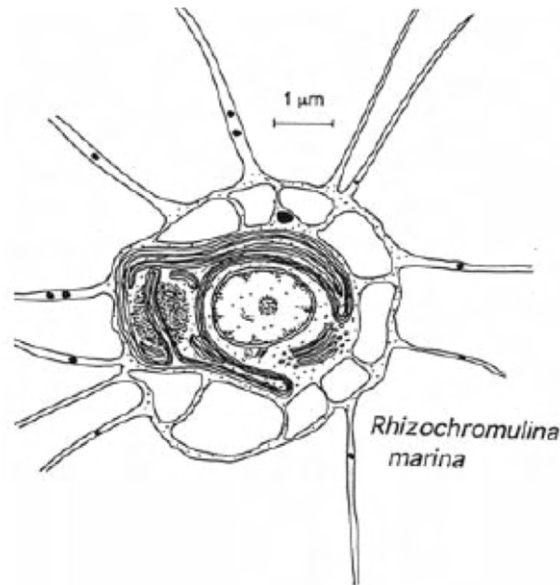
- during the TARA Oceans expedition (to generate a global ocean reference catalog of genes from planktonic eukaryotes) show that Dictyochophyceae is quite abundant and diverse in global oceans, possibly being important planktonic primary producers

percentage of unigenes; ftGF: functional and taxonomical assigned groups; tGF: taxonomically assigned.

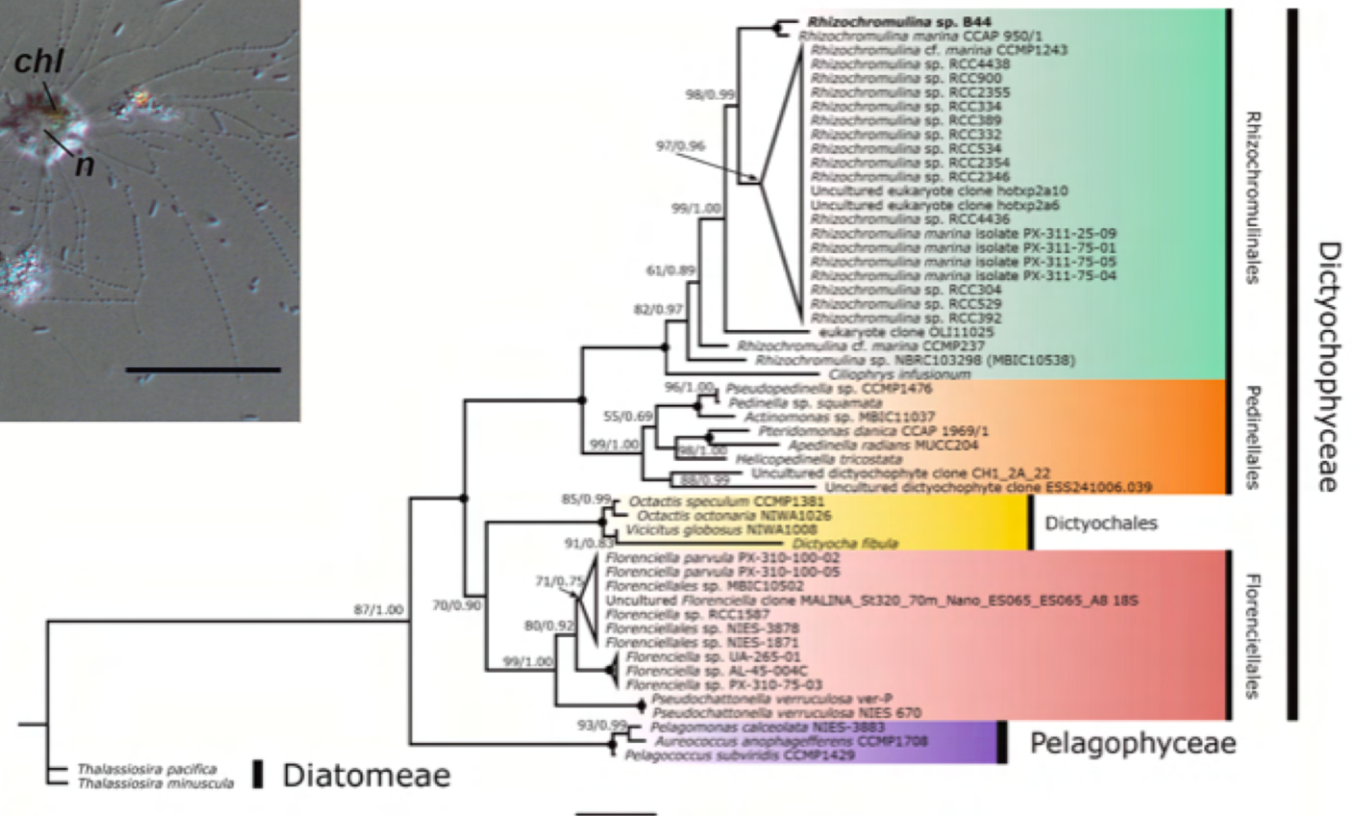
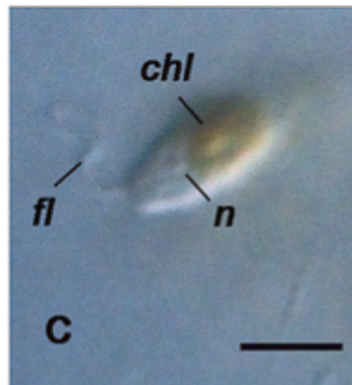
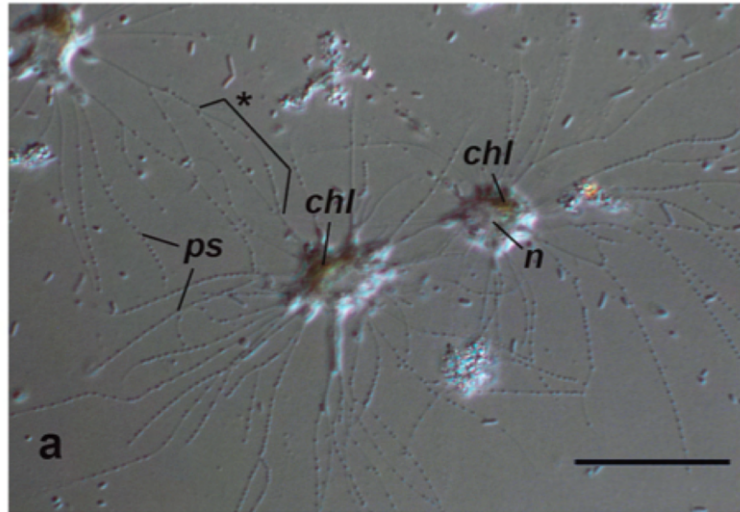


Dictyochophyceae – Rhizochromulinales

- marine (puddles and tidal zones) and freshwater
- amoeboid vegetative cells, fine beaded-filipodia (pseudopods)
- swimming cells with a single flagellum (a second basal body in protoplasm)
- a single golden-brown chloroplast (*Rhizochromulina marina*) or heterotrophic taxa (*Ciliophrys* spp.)



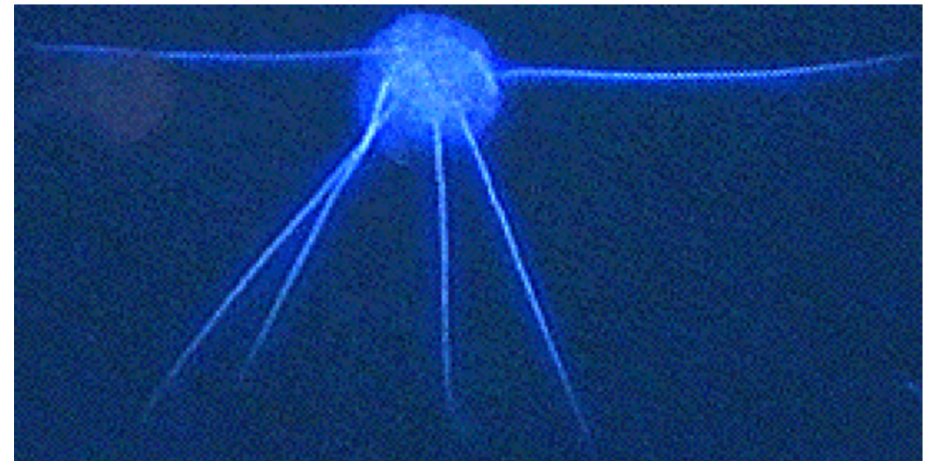
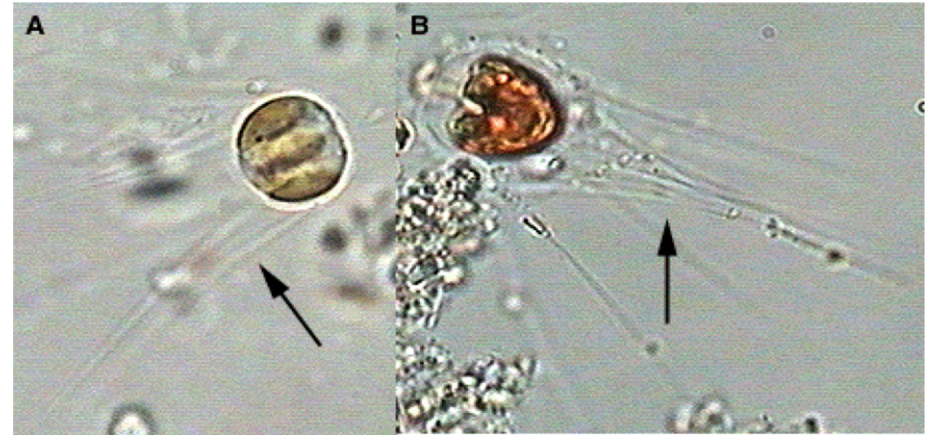
Dictyochophyceae – Rhizochromulinales



Rhizochromulina marina (chl – chloroplasts, fl – flagellum, n – nucleus, ps – pseudopodia)

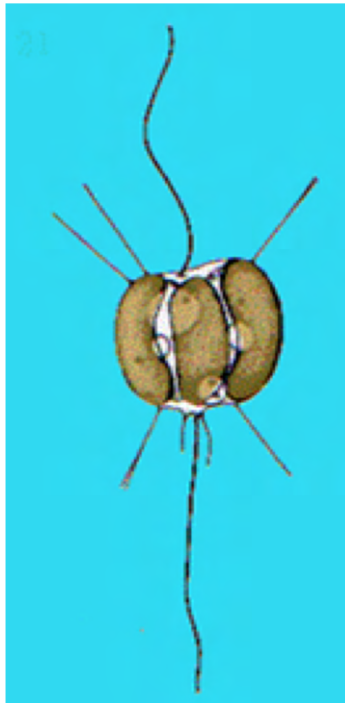
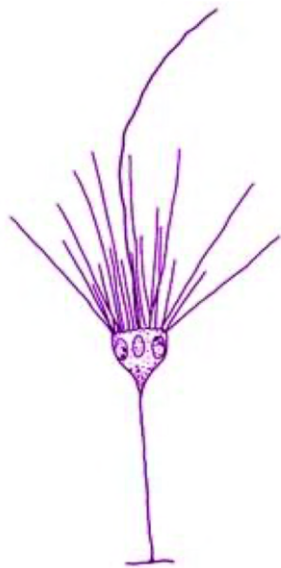
Dictyochophyceae – Pedinellales

- marine and freshwater
- mixotrophic or heterotrophic (phagotrophy of bacteria, *Actinomonas*, *Pteridomonas* - detection of a leucoplast)
- unicells with a long anterior flagellum (a second flagellum reduced to a basal body)
- usually three to six chloroplasts (if chloroplasts are present)
- organically scaled (*Pedinella*, *Apedinella*) or loricate cells



Apedinella - photoautotrophy

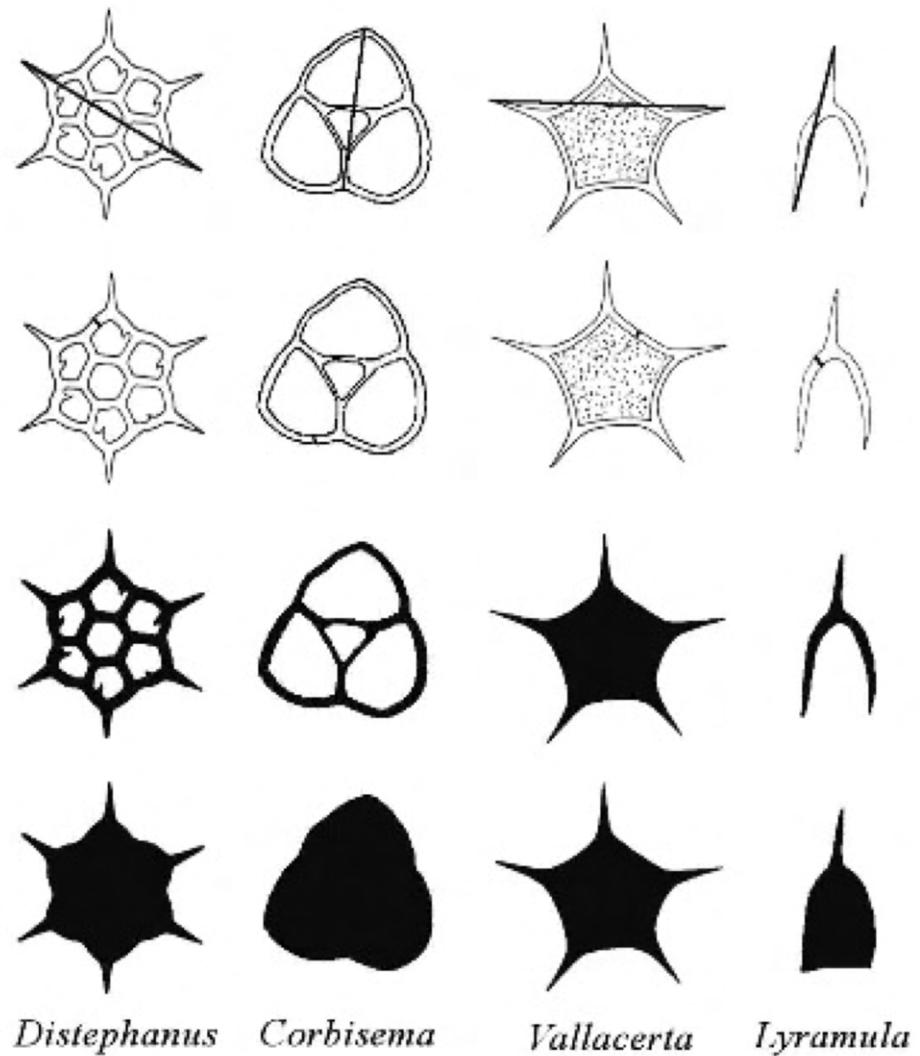
Dictyochophyceae – Pedinellales



Pedinella: mixotrophy, a posterior sticky stalk (cells rotate while swimming, trailing the stalk behind, often adhere a swimming cell to a substrate)

Dictyochophyceae – Dictyochales

- so-called silicoflagellates
- marine habitats, phytoplankton
- external silica skeleton present on at least one life stage [but *Vicicitus globosus*]
- predominantly fossil (first appearing in the Early Cretaceous, currently only four extant siliceous species, genus *Dictyocha*); indication of seawater temperature (predominantly in cold waters)

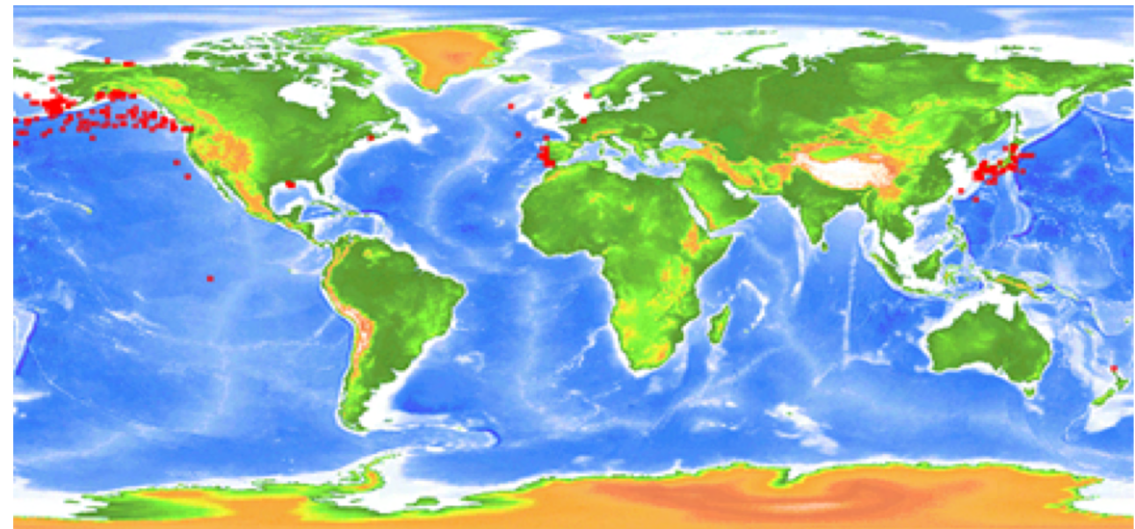
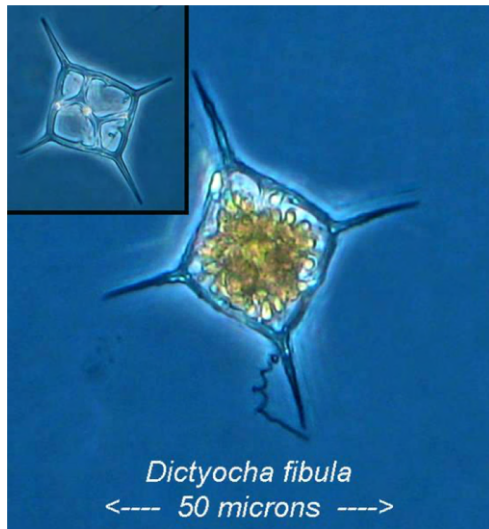
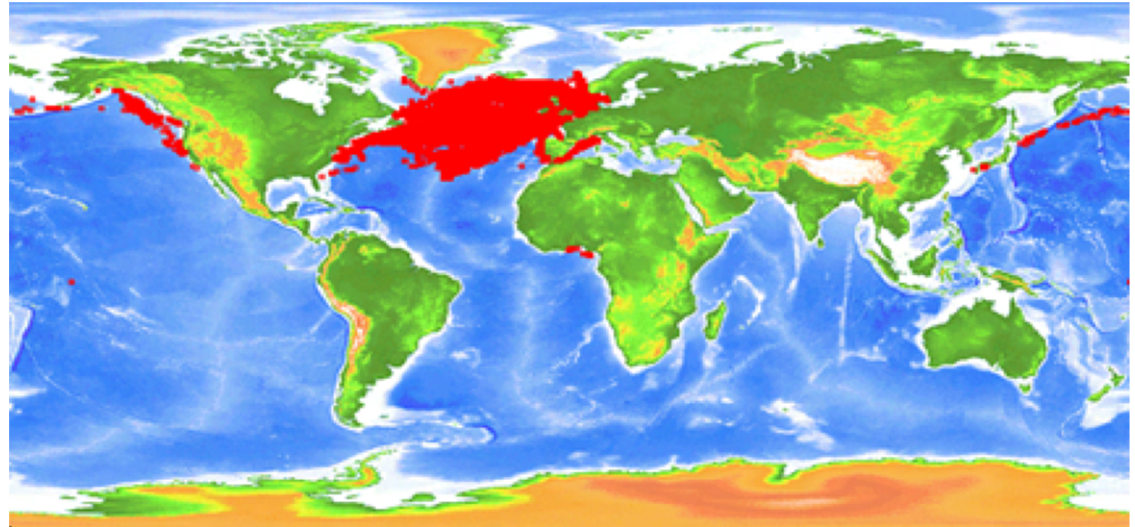


fossil genera, Dictyochales

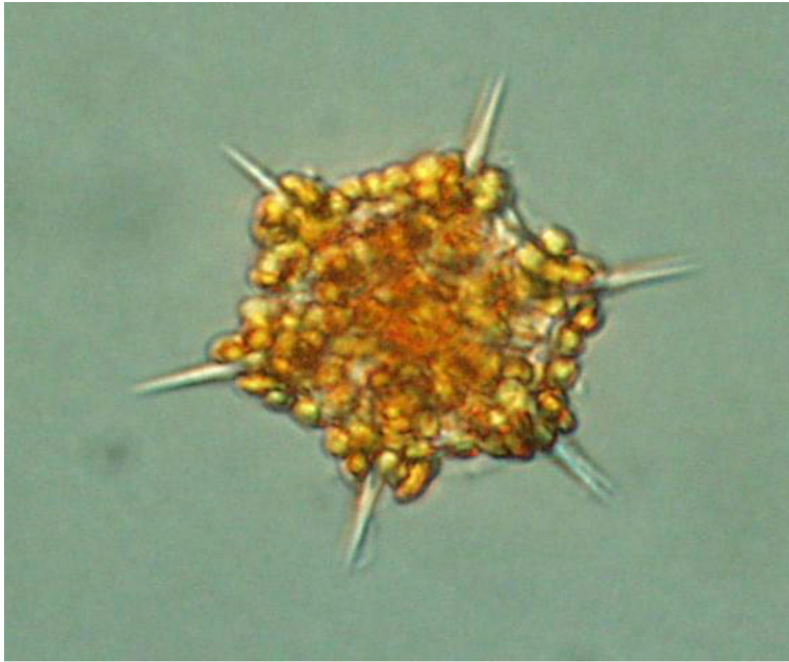
Dictyochophyceae – Dictyochaales



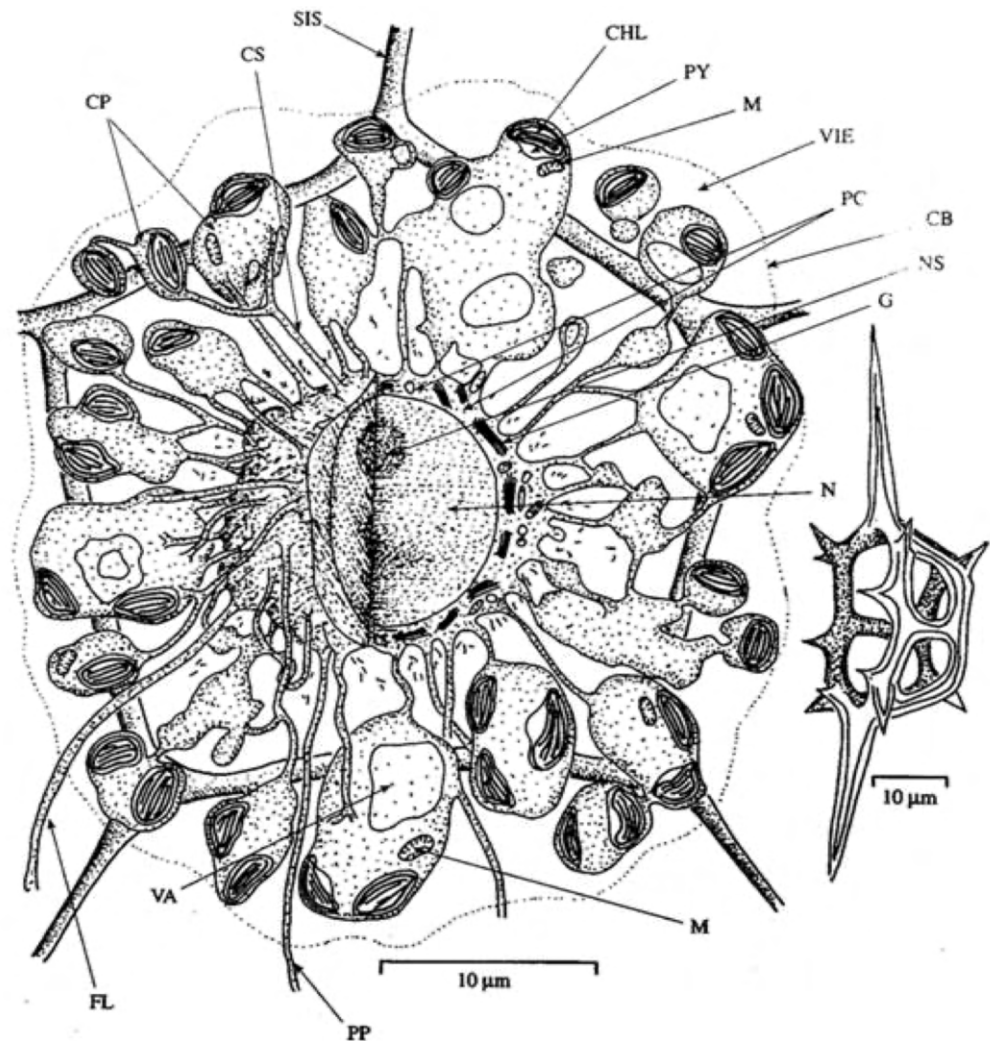
Dictyocha speculum



Dictyochophyceae – Dictyochales

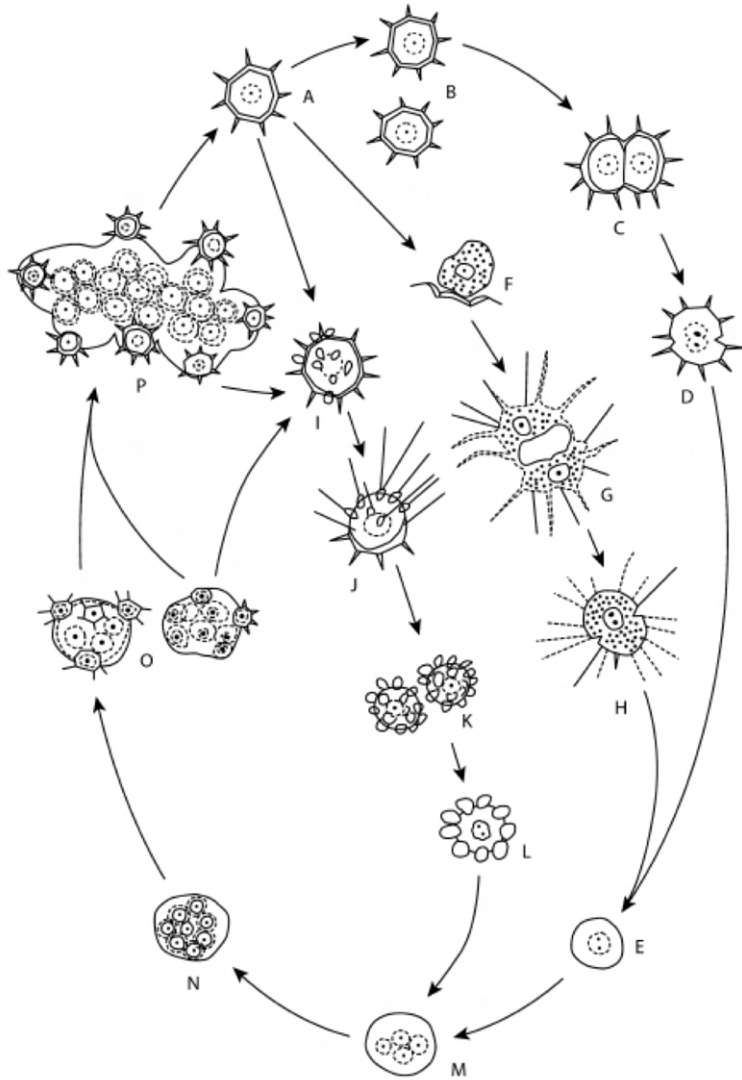


Dictyocha



FL: flagellum; **M:** mitochondria, **N:** nucleus; **CHL:** chloroplast; **PY:** pyrenoid; **SIS:** siliceous skeleton; **PP:** pseudopodium; **CB:** cell boundary; **VIE:** mucilaginous envelope

Dictyochophyceae – Dictyochales



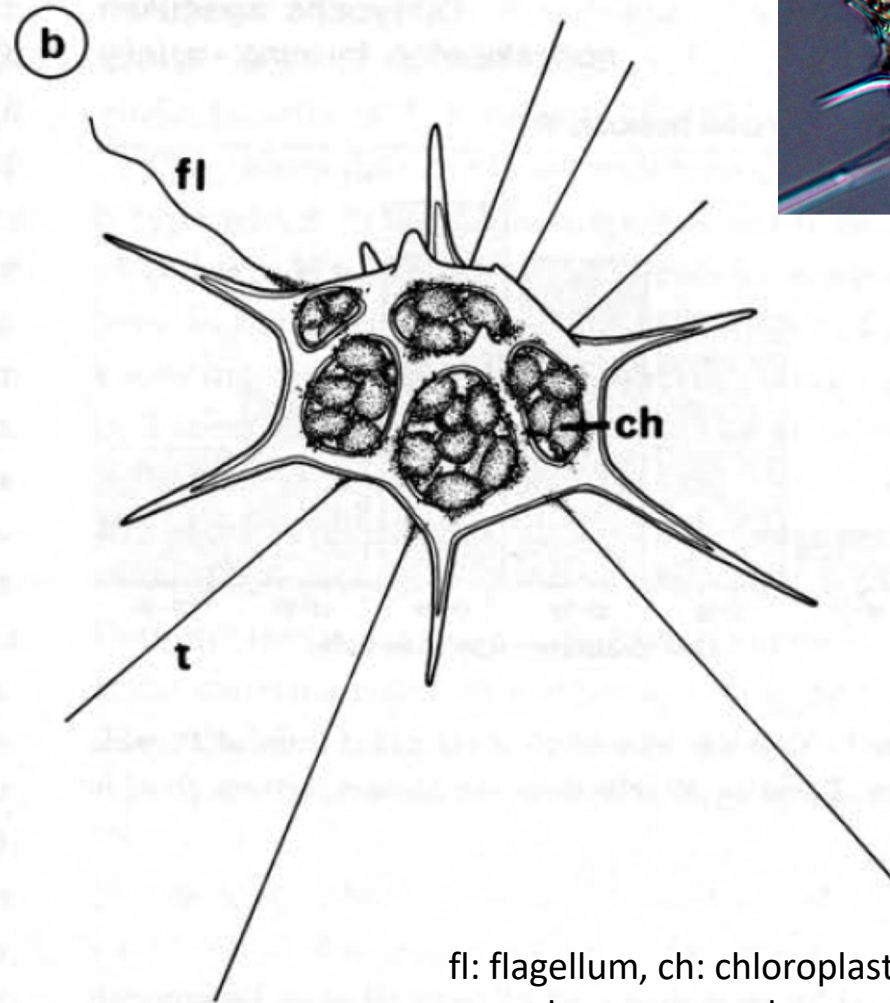
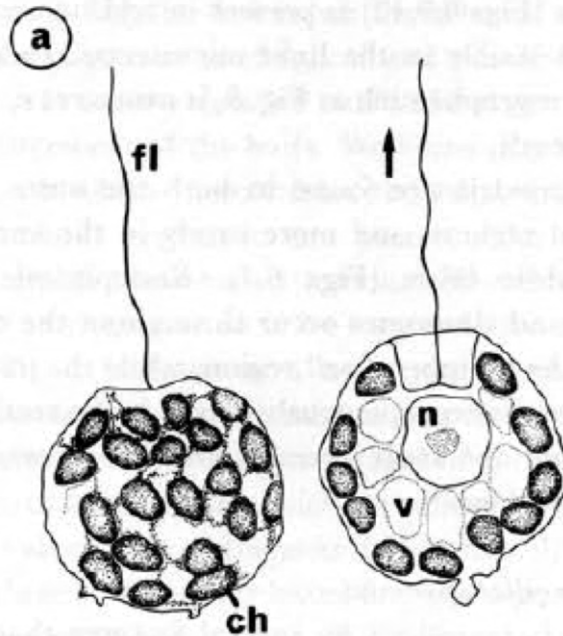
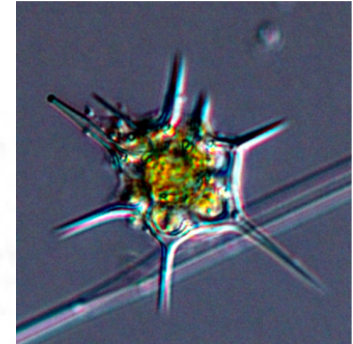
Sexual reproduction of *Dictyocha octonaria*.

A–E + I. Skeletonbearing cells. F. Transformation of a skeletonbearing to amoeboid cell. **G–H. Amoeboid cells J.** Some cells started to show long **mucus filaments** on mucocysts before moving away from the parent skeleton. **K–L.** Fusion of two gamete-like, mucocyst-bearing cells to form a zygote. **M–O. Multinucleate cells P. Flagellated** daughter cells pushed their way through the membrane of the massive **plasmodium-like aggregate**.

With the exception of the zygote, which is in diplophase, all the rest, from uninucleate, gamete-like cells, to daughter cells formed within the multinucleate/parent and plasmodium-like cells, are in haplophase.

Dictyochophyceae – Dictyochaales

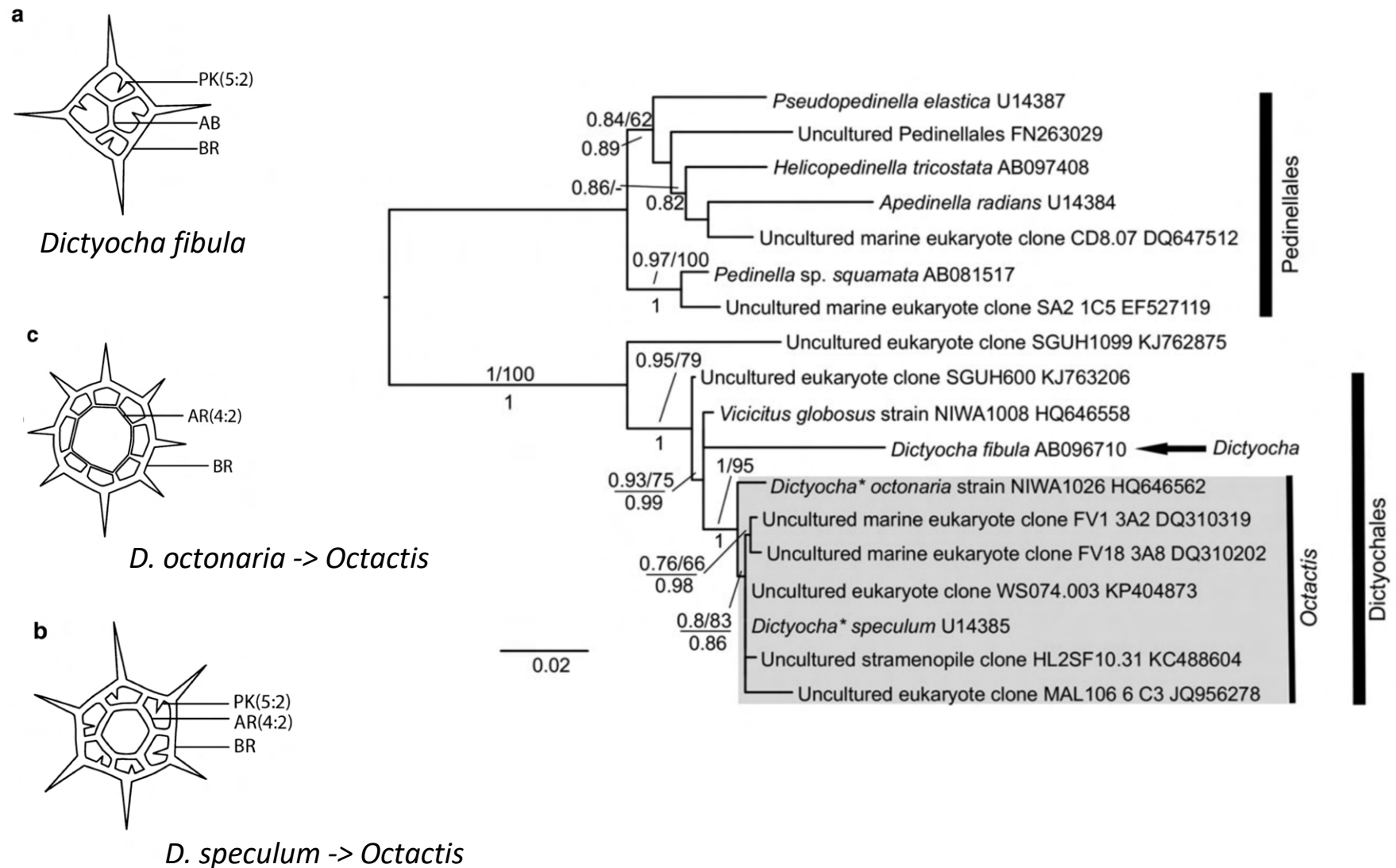
- microtubule-supported pseudopodia



fl: flagellum, ch: chloroplast, v: vacuole,
n: nucleus, t: tentacle=pseudopodium

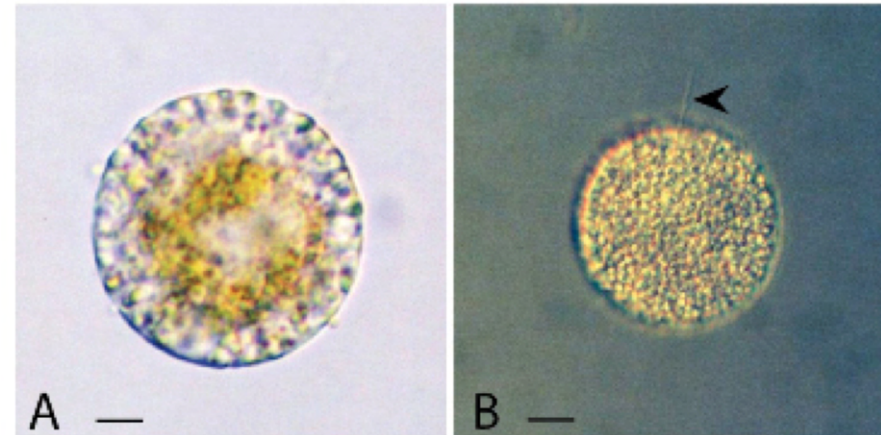
the naked form and skeleton bearing form, *Dictyocha speculum*

Dictyochophyceae – Dictyochaales

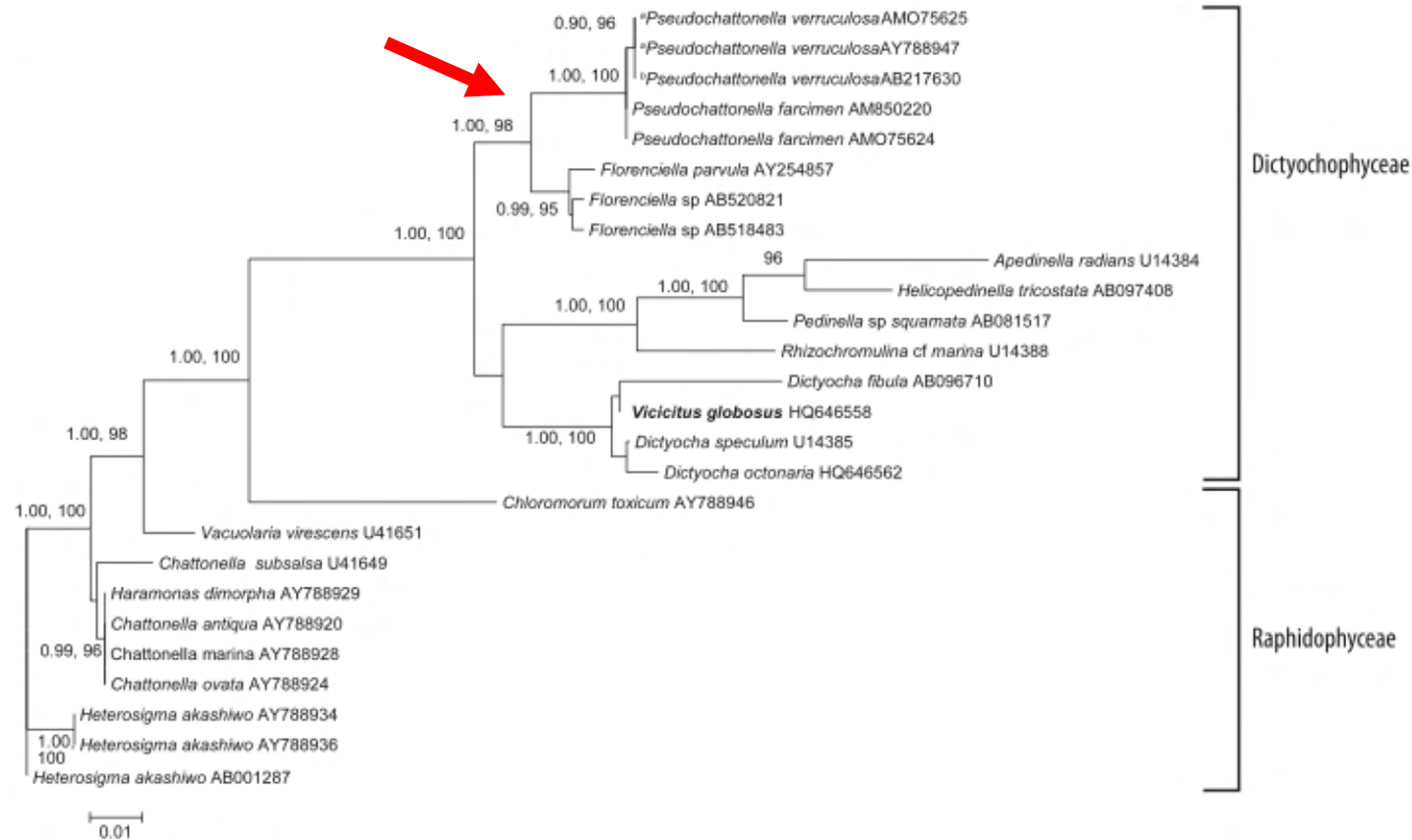


Dictyochophyceae – Dictyochaales

- *Vicicitus globosus* – previously in Raphidophyceae (as *Chatonella globosa*)
- no siliceous skeleton-bearing stage observed in the life history
- mucocysts protruding evenly over the cell membrane (swimming cells can transform swiftly from regular globular shape to amoeboid form in a matter of seconds)



Dictyochophyceae – Florenciellales

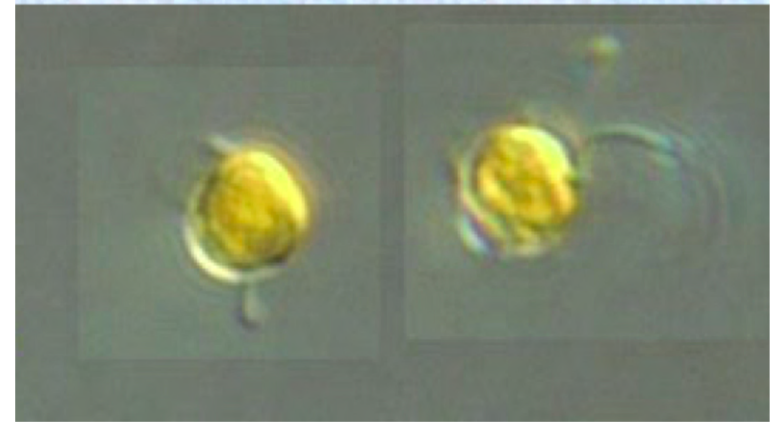


Pseudochattonella ← *Chattonella* aff. *verruculosa*
- confusion with ichthyotoxic Raphidoph.

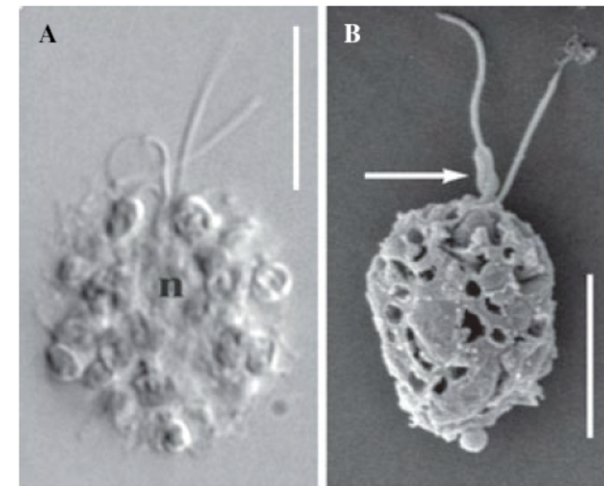
Dictyochophyceae – Florenciellales

- marine environment
- picoflagellate *Florenciella parvula*, described in 2004 (English Channel)
- *Verrucophora* spp. (previously as *Pseudochatonella* ← *Chatonella* aff. *verruculosa*) a bloom forming flagellate in cold waters (Norway, Japan, Chile, New Zealand)

mucocysts evenly distributed on the cell surface – secretion of mucus, fish kills

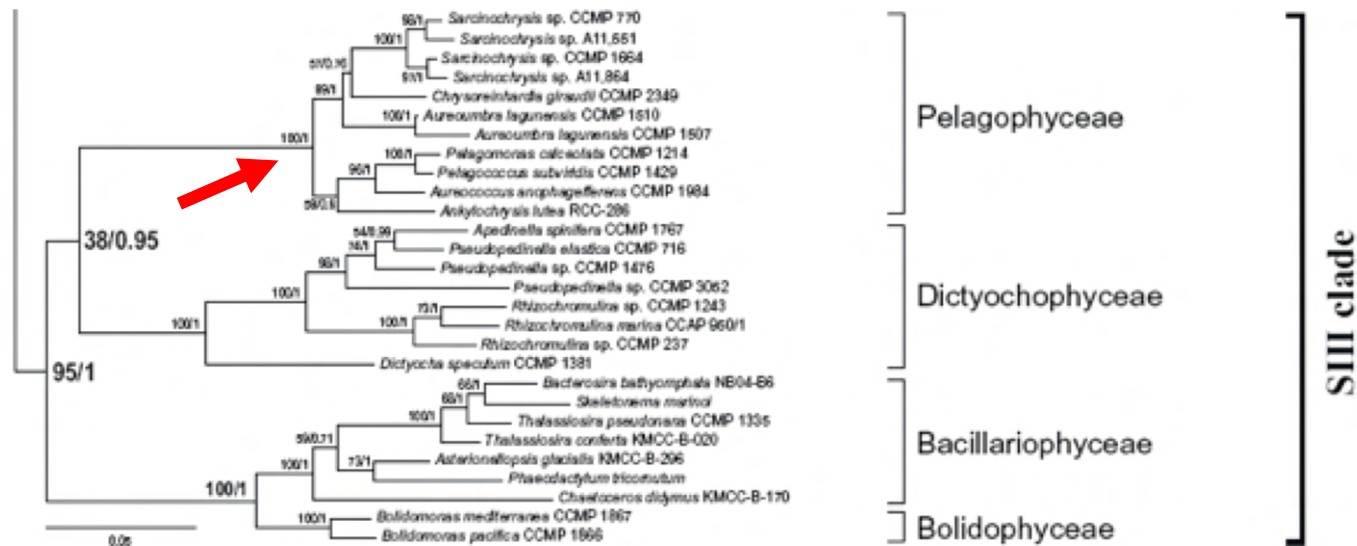


Florenciella parvula



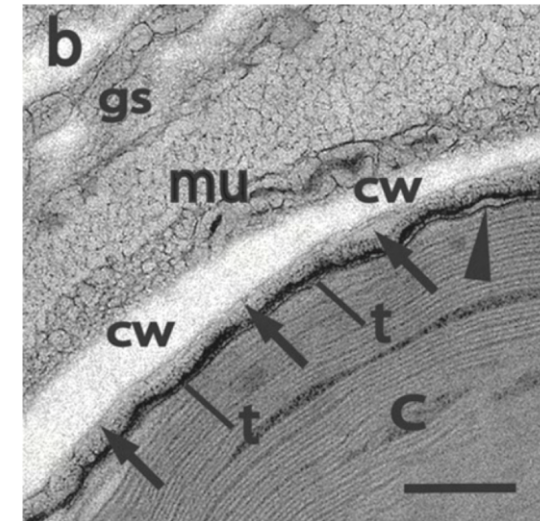
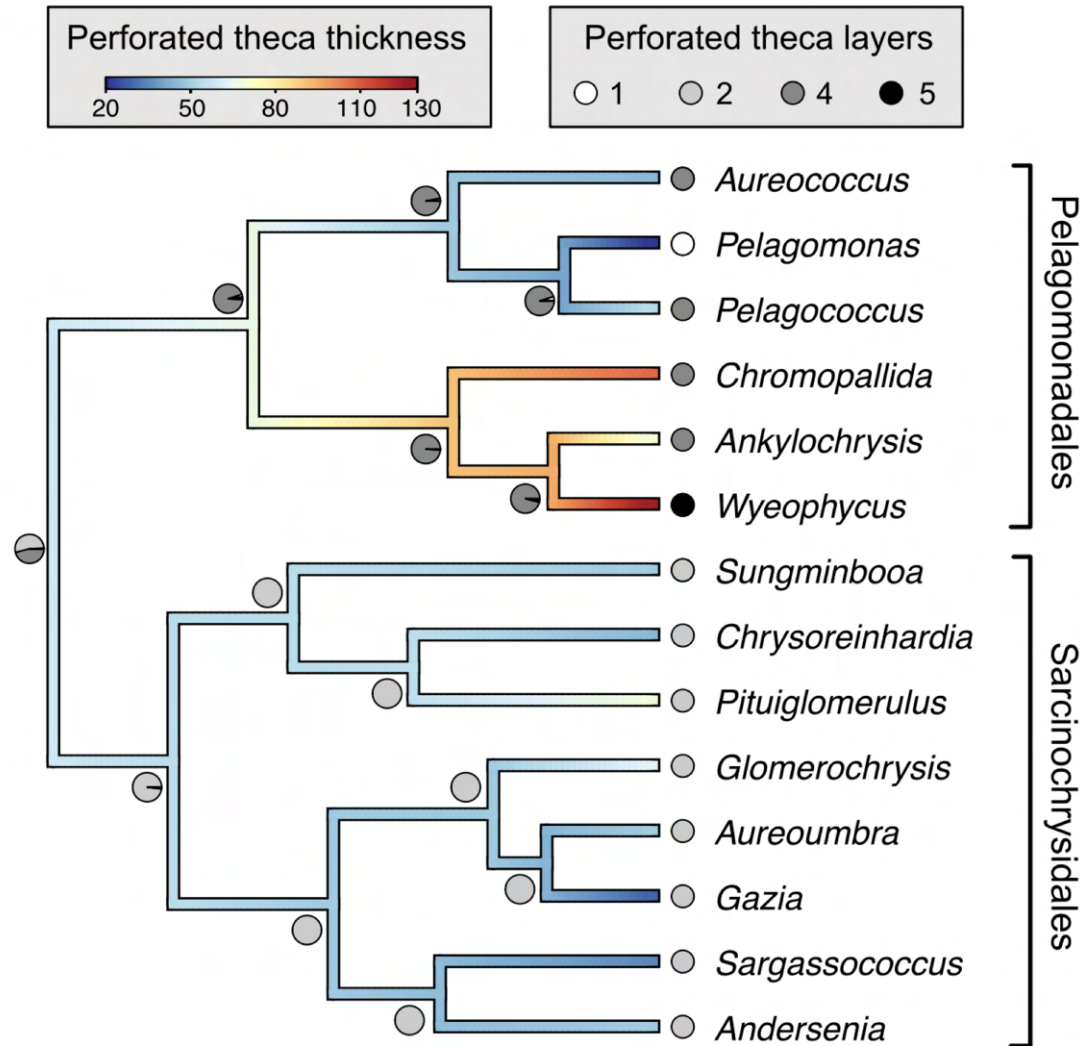
Verrucophora farcimen

Pelagophyceae

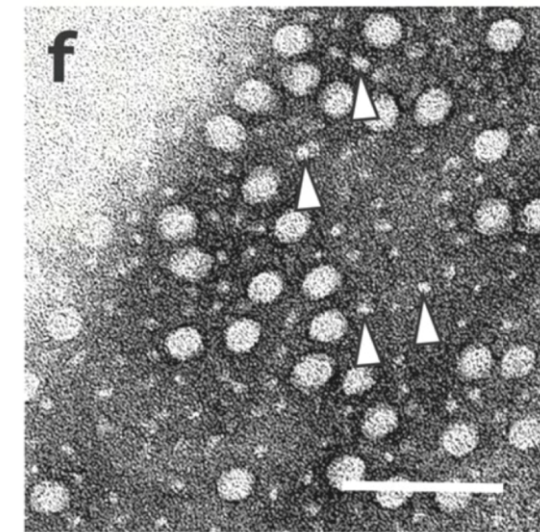


- variable forms: ciliated, coccoid, sarcinoid, capsoid, filamentous
- swimming cells with one or two flagella (bipartite or tripartite tubular hairs)
- typical stramenopile chloroplast; no eyespot; chlorophylls *a*, *c* + xanthophylls (including fucoxanthin)

Pelagophyceae



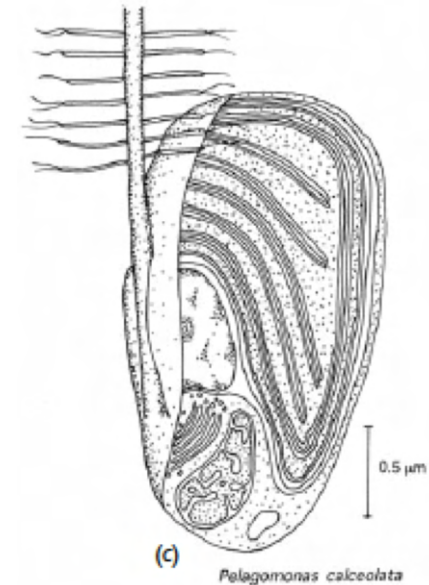
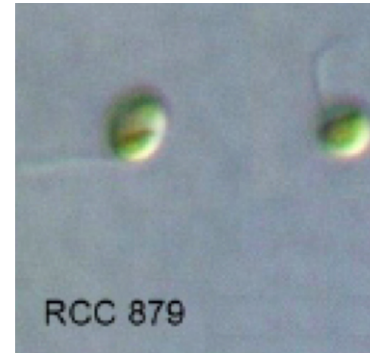
cw: cell wall, **t: theca**, arrowheads: plasma membrane, gs: gel sheat, mu and arrows: mucilaginous layers



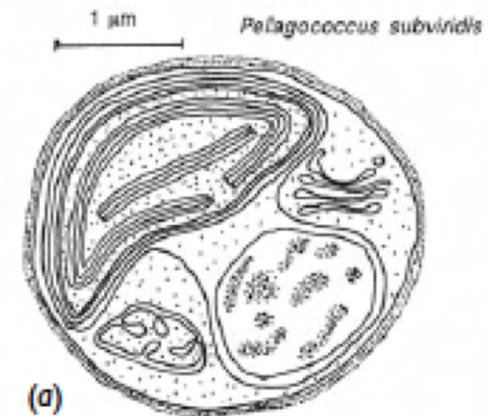
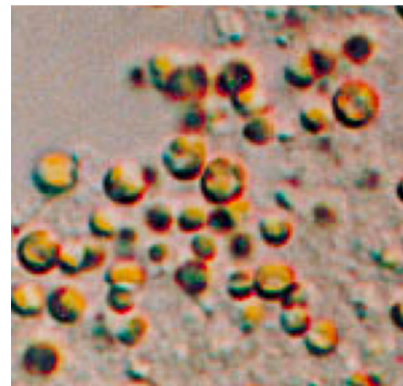
macropores and micropores

Pelagophyceae – Pelagomonadales

- marine picoplankton
- *Pelagomonas*: monadoid (a single flagellum with bipartite hairs, a second basal body absent); a thin organic theca surrounds most of the cell
- *Pelagococcus*: free living or symbiont of foraminifera



Pelagomonas
s



Pelagococcus

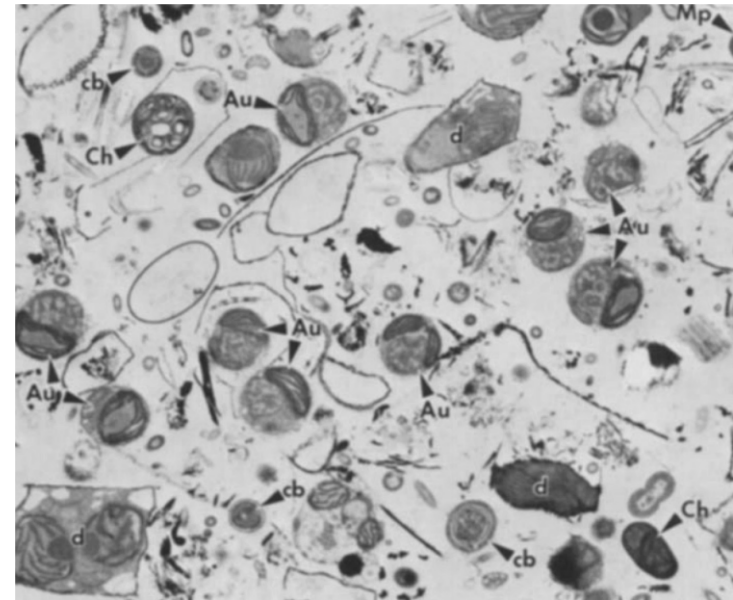
Inhibition of harmful algal blooms caused by *Aureococcus anophagefferens* (Pelagophyceae) using native (*Gracilaria tikvahiae*) and invasive (*Dasysiphonia japonica*) red seaweeds from North America

Colin Benitt¹ · Craig S. Young¹ · Laine H. Sylvers¹ · Christopher J. Gobler¹

Pelagophyceae – Pelagomonadales

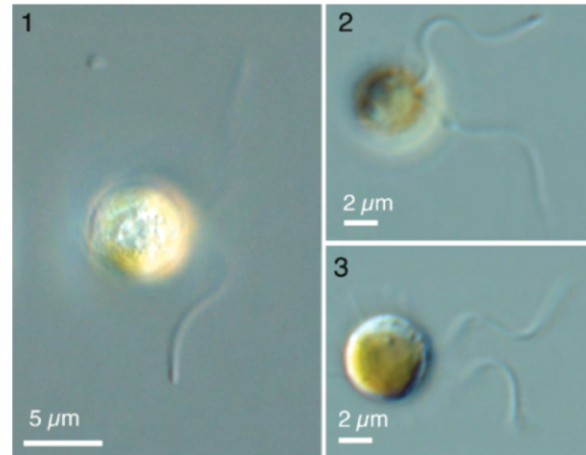
- *Aureococcus anophagefferens*: able to grow at low temperature and survive long periods of darkness
- brown-tides (along the eastern coast of the United States, South Africa), harmful for *Zostera marina* meadows (reduced light penetration) and shellfish (reduced growth rate of filter feeders, unavailable *Zostera* habitat)
- blooms: the importance of mixotrophy (metabolism of dissolved organic carbon) + cells are coated by polysaccharide material (inhibits filtration by grazing animals)
- etymology: golden yellow sphere

Au = *A. anophagefferens*; cb = the cyanobacterium *Synechococcus*; Ch = *Chlorella*-like species; d = diatom

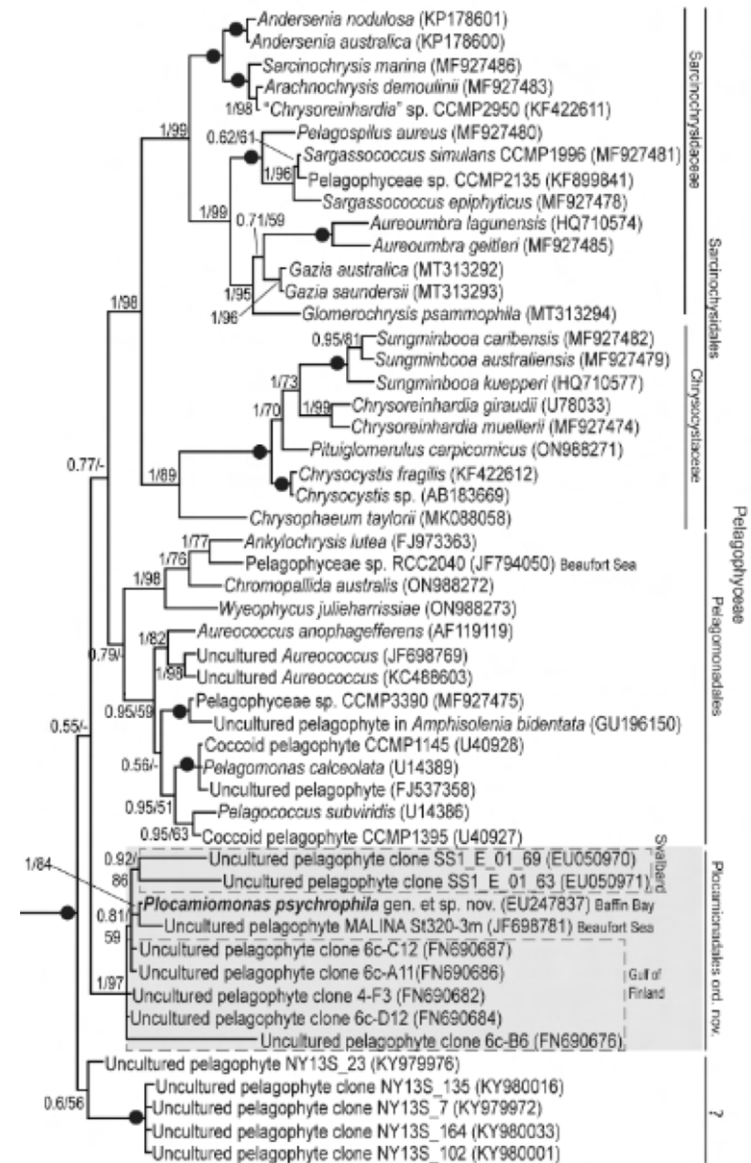


Pelagophyceae – Plocamiomonadales

- an Arctic marine nanoflagellate (7.5 μm , isolated from a small pocket of liquid water on the sea ice)
- euryhaline and stenothermal cold-water species
- did not produce a gel matrix (like Sarcinochrysidales) and have flagellar roots (lacking in Pelagomonadales)

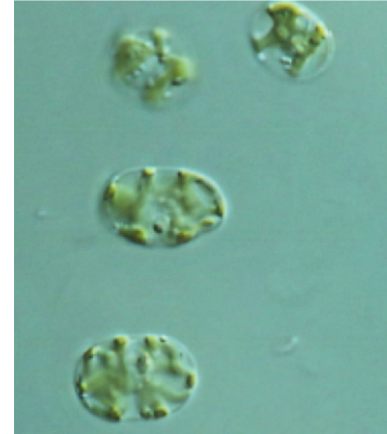


Plocamiomonas psychrophila

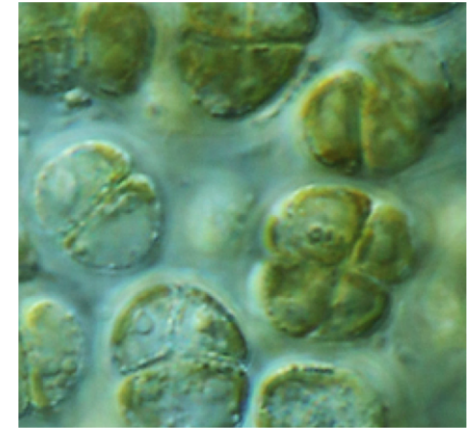


Pelagophyceae – Sarcinochrysidales

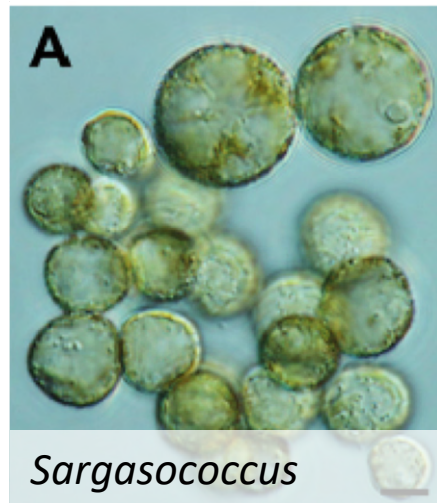
- single cell, filament, sarcinoid colony, clusters of cells
- all the Sarcinochrysidales species are capable of producing gel
- originally an order of Chrysophyceae



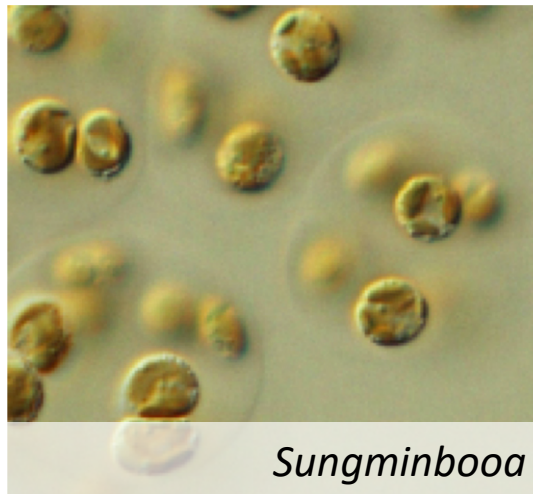
Arachnochrysis



Chrysoreinhardtia



Sargasococcus



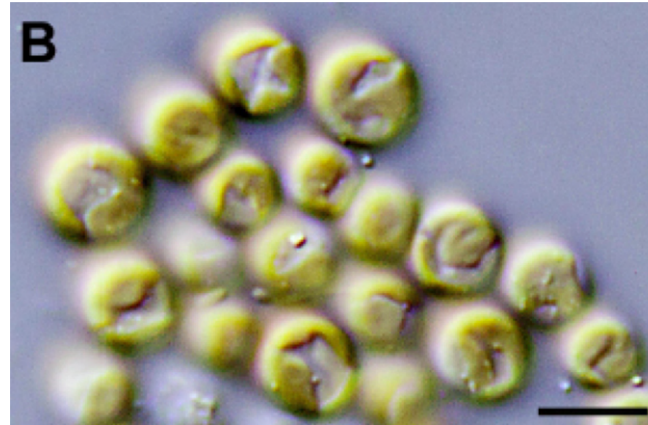
Sungminbooa



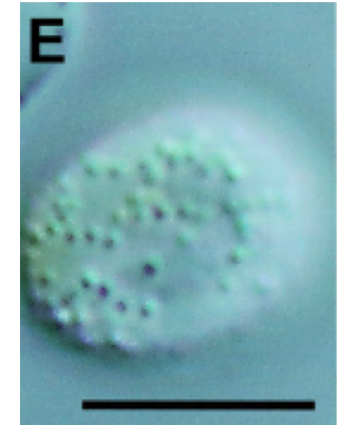
Andersenia

Pelagophyceae – Sarcinochrysidales

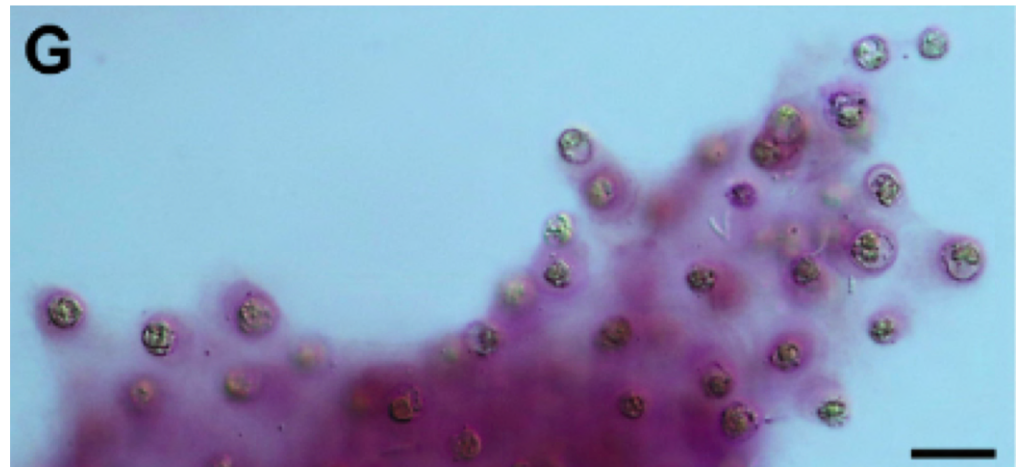
- *Aureoumbra lagunensis*: nonmotile coccoid cells that are covered with a slime layer that reduces predation
- survive and grow in hypersaline conditions
- massive blooms = brown tides
- *Aureoumbra geitlerii*: described in 2018, Canary Islands



Aureoumbra geitlerii



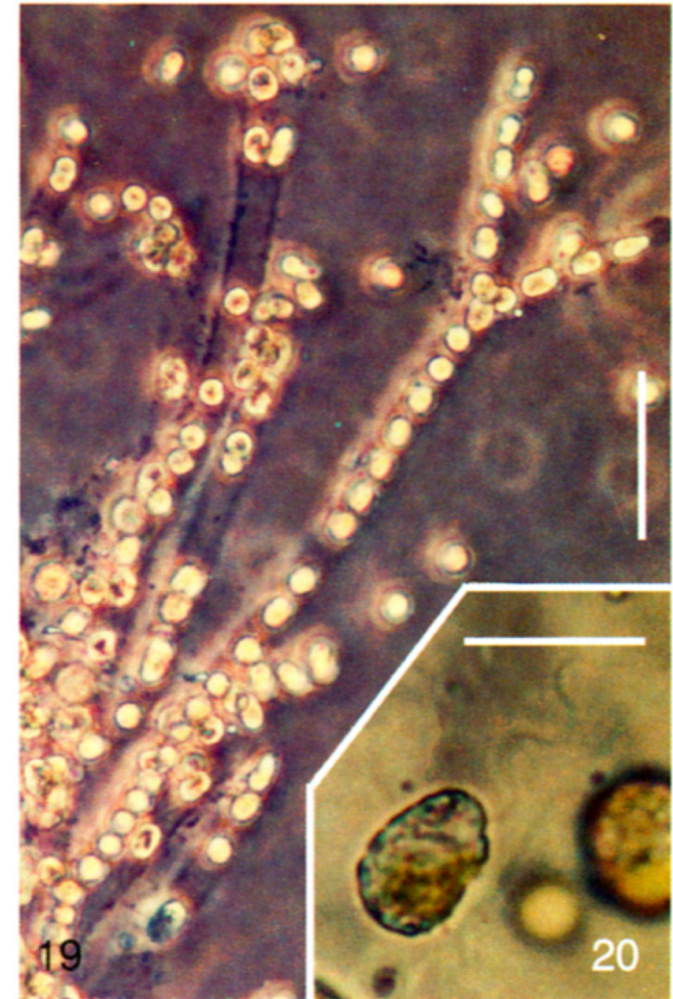
peripheral granules



Aureoumbra geitlerii - gelatinous envelope

Pelagophyceae – Sarcinochrysidales

- *Chrysocystis* (noodle alga)
- colonies forming sacs, cells in a layer of amorphous mucilage
- extensive nuisance blooms on benthic substrates in marine environments



Pelagophyceae – Sarcinochrysidales

- *Chrysophaeum taylorii*:
macroscopic gelatinous colonies
(free-living cells randomly embedded in
the mucilage)
- benthic; invasive spread in the
Mediterranean Sea (from subtropic
and tropic oceans)

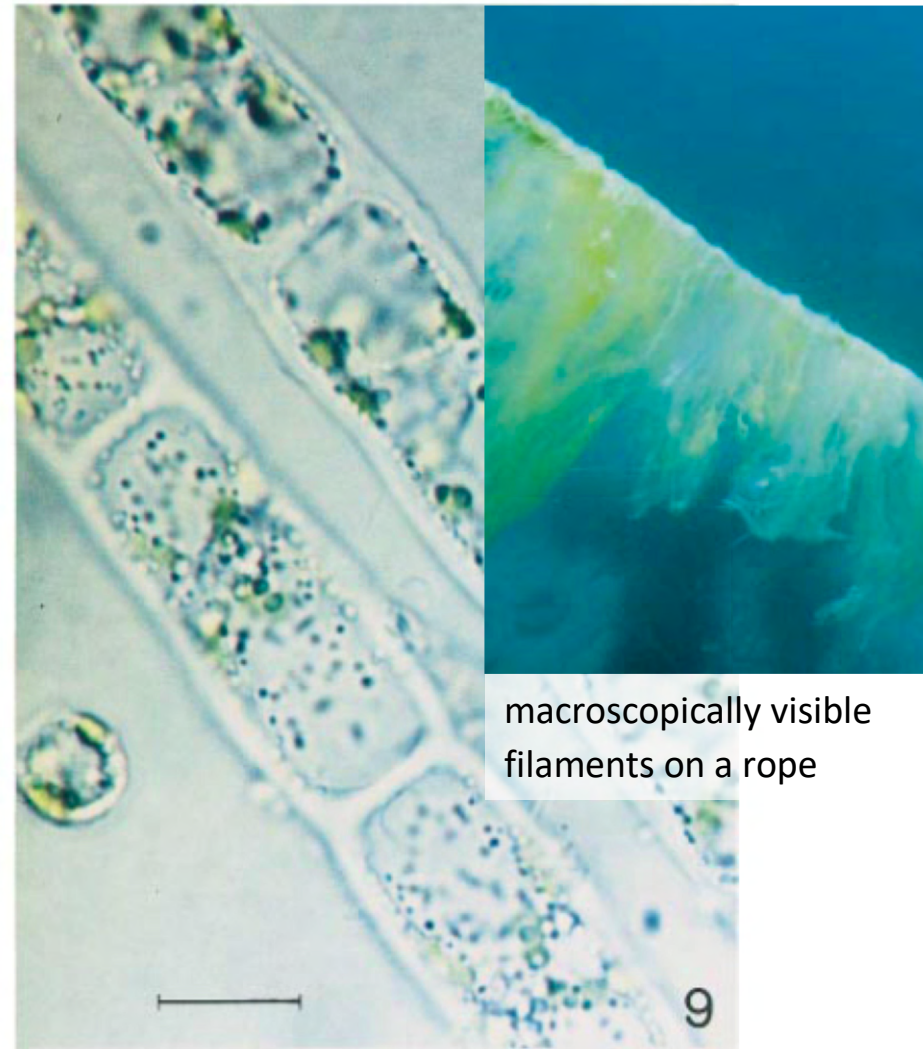


Chrysophaeum taylorii

Pelagophyceae – Sarcinochrysidales



Chrysonephos lewisii

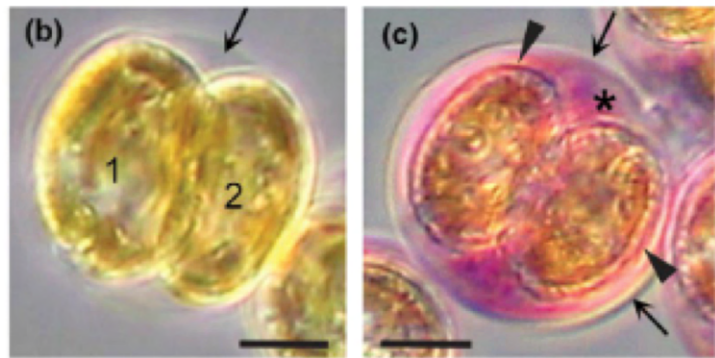


macroscopically visible
filaments on a rope

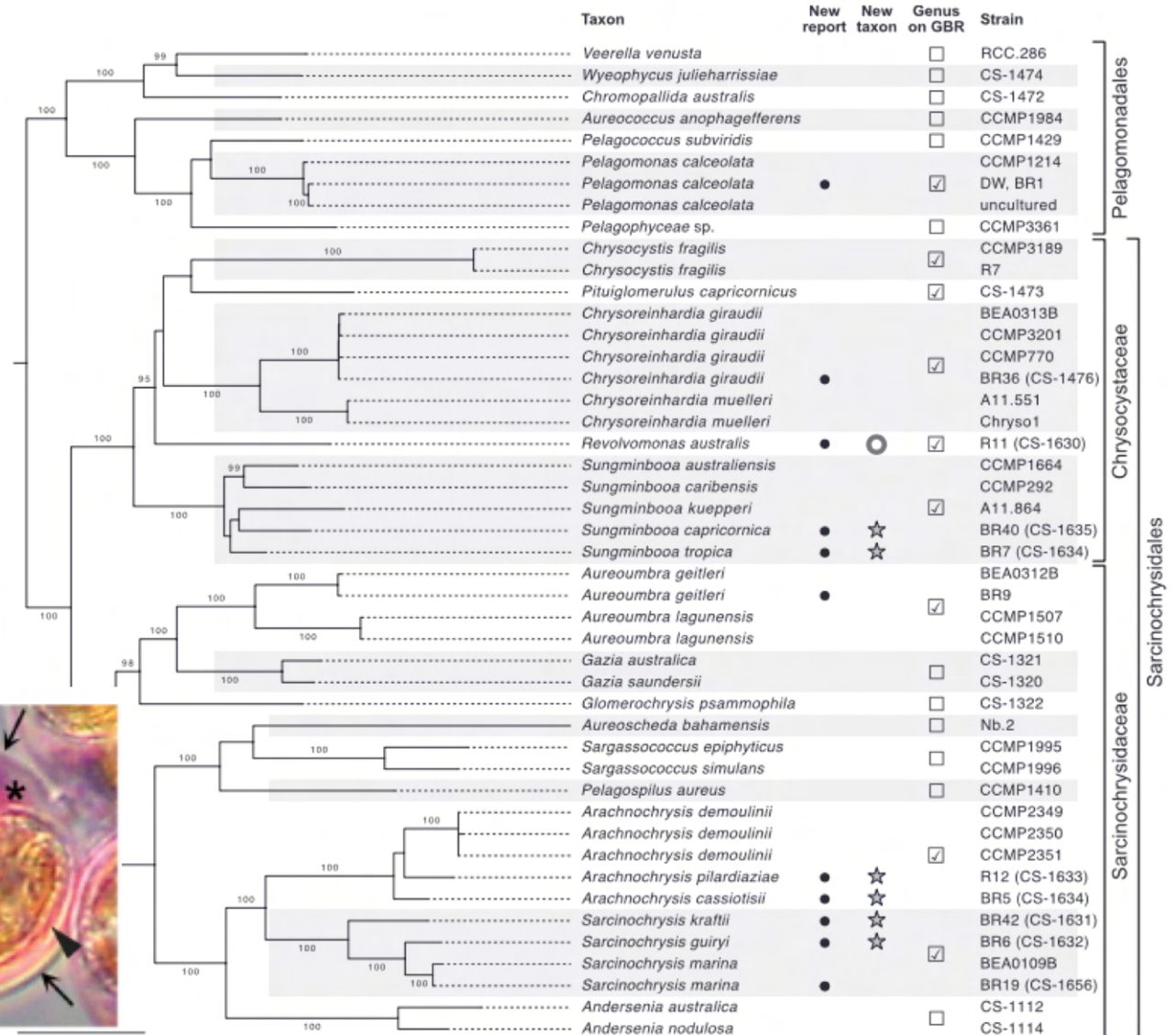
Nematochryopsis marina (*Tribonema marinum*)

Pelagophyceae – Sarcinochrysidales

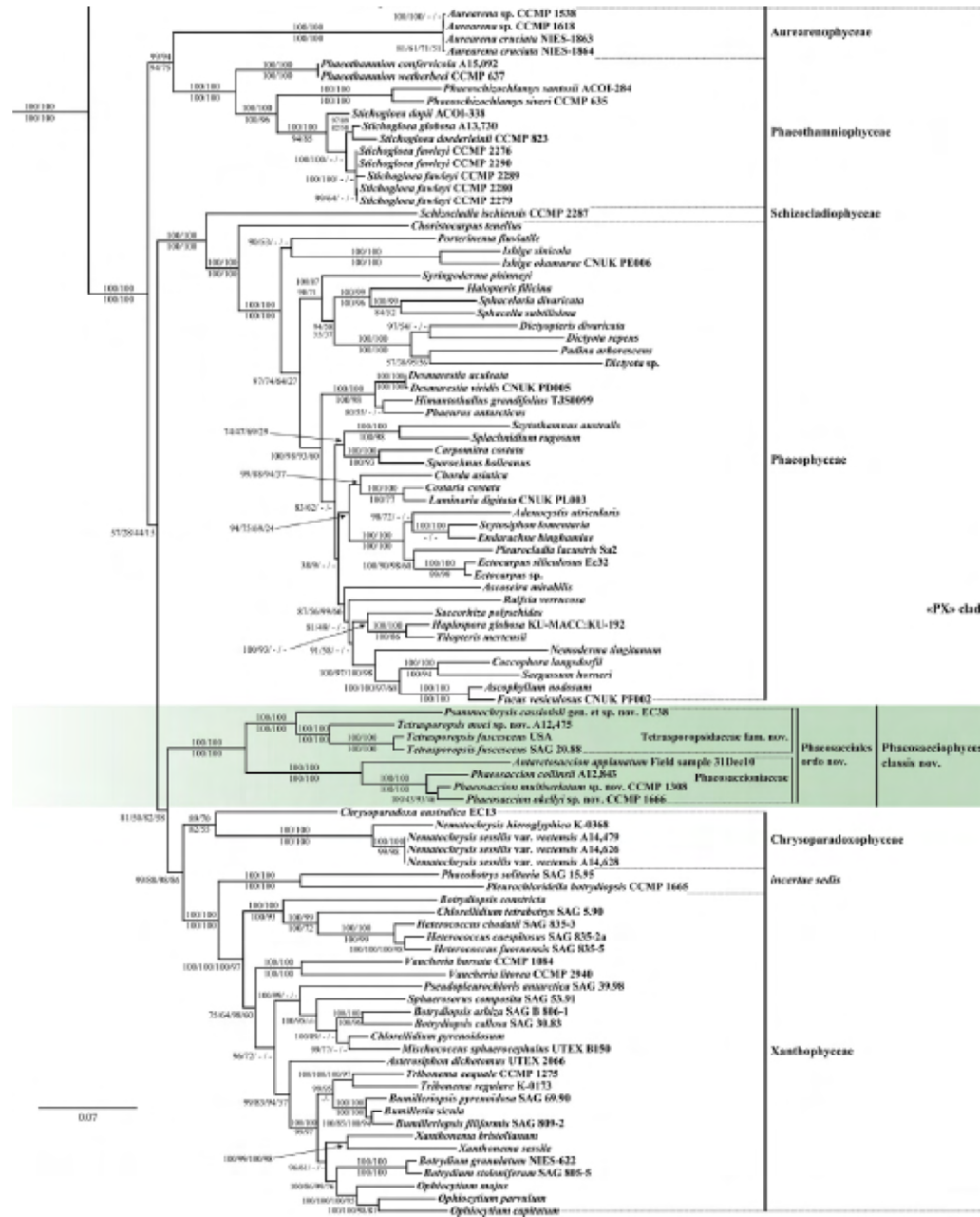
- three short collecting trips to Heron Island (Great Barrier Reef, Australia) - over 40% of the pelagophyte genera (+ a new genus and 7 new species)
- tropical reef sand may be a center for pelagophyte diversity



Revolvomonas australis



PX clade



PX clade

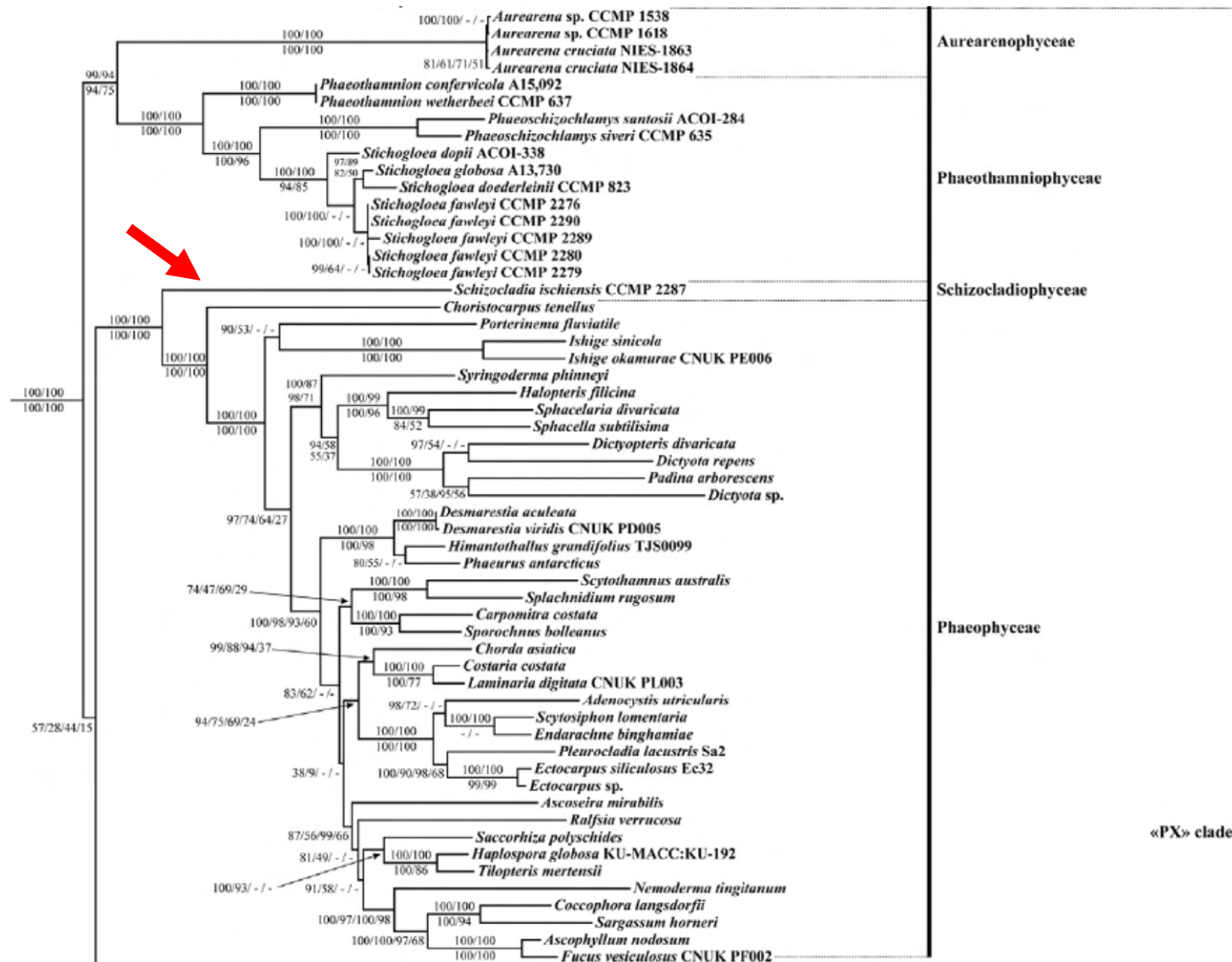
Table 2. Comparison of selected characters among the members of the PX clade

	Organization	Habitat	PO	CF	GL	PN	TF	TH	BR	RH	Chl.c	Fuc	Dia	Vio	Het	Vau
<i>Aurearena</i>	FL–CC	M	+	+	–	S	3	+	–	+	–	+	+	+	–	–
Phaeothamniophyceae s.s.	CC, FI	F	+	–	+	R	1	+	–	–	+	+	+	–	+	–
Xanthophyceae	CC, FI, SI	F (M)	–	–	+	(–) R (S)	2	+	–	+	+	–	+	–	+	+
<i>Pleurochloridella</i>	CC	F	+	?	?	?	?	?	?	?	+	+	+	–	+	–
Chrysomerophyceae	FI	M	–	–	+	R	3	+	+	–	+	+	–	+	–	–
<i>Tetrasporopsis</i>	CC	F	+	–	+	?	?	?	?	?	?	?	?	?	?	?
Schizocladiophyceae	FI	M	–	–	+	R	?	+	?	–?	+	+	?	?	?	?
Phaeophyceae	MC	M (F)	+	–	+	R	1–3	–	+	+	+	+	– (+)	+	–	–

FL = flagellate, CC = coccoid, FI = filamentous, SI = siphonous, MC = multicellular, M = marine, F = freshwater, PO = periplasmic opaque vesicle, CB = chloroplast boundary, GL = girdle lamella, PN = plastid nucleoid (R = ring type, S = scattered type), TF = no. of terminal filaments of mastigoneme, TH = transitional helix, BR = bypassing root, RH = rhizoplast, Fuc = fucoxanthin, Dia = diatoxanthin/diadinoxanthin, Vio = violaxanthin/antheraxanthin/zeaxanthin, Het = heteroxanthin, Vau = vaucherixanthin, asterisks mean very small amount, exceptional example in parentheses.

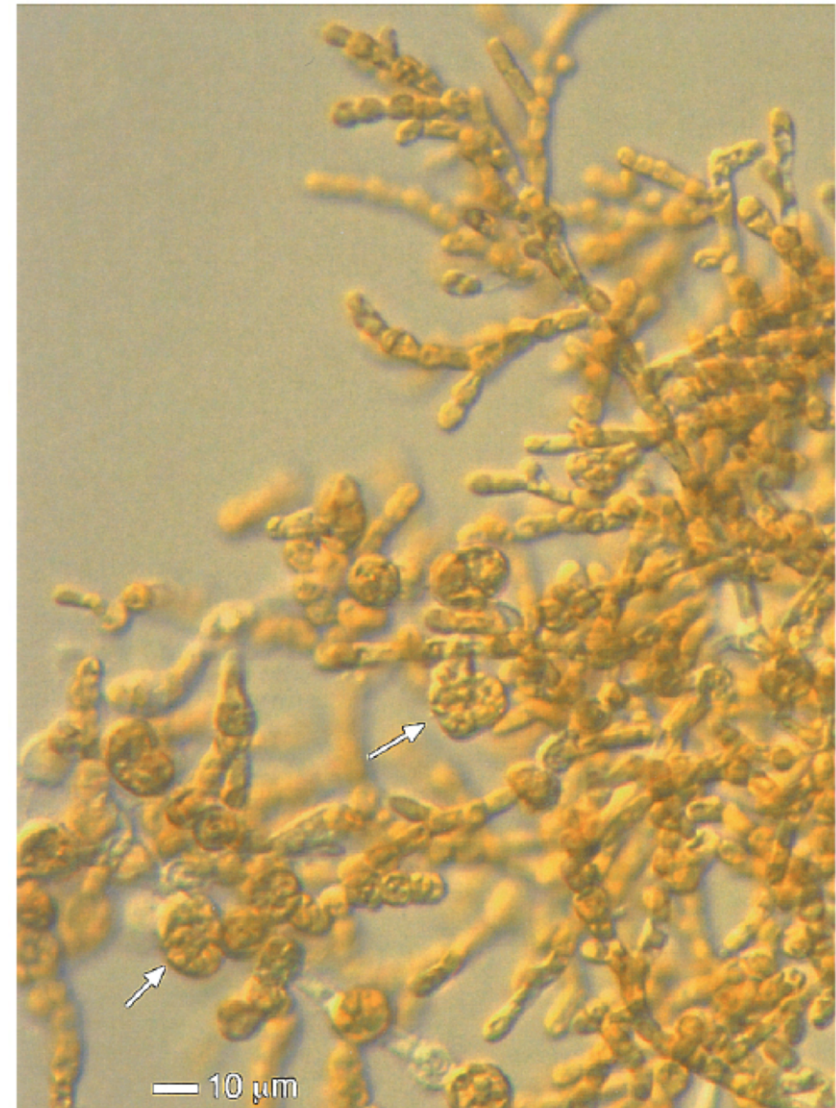
- very diverse life forms and habitats
- emergence of true multicellularity in Phaeophyceae
- absence of flagellate (FL) cells in the vegetative phase
- all members of the PX clade possess a cell wall in the vegetative phase
- presence of electron-opaque vesicles (PO) beneath the plasma membrane

Schizocladiphyceae



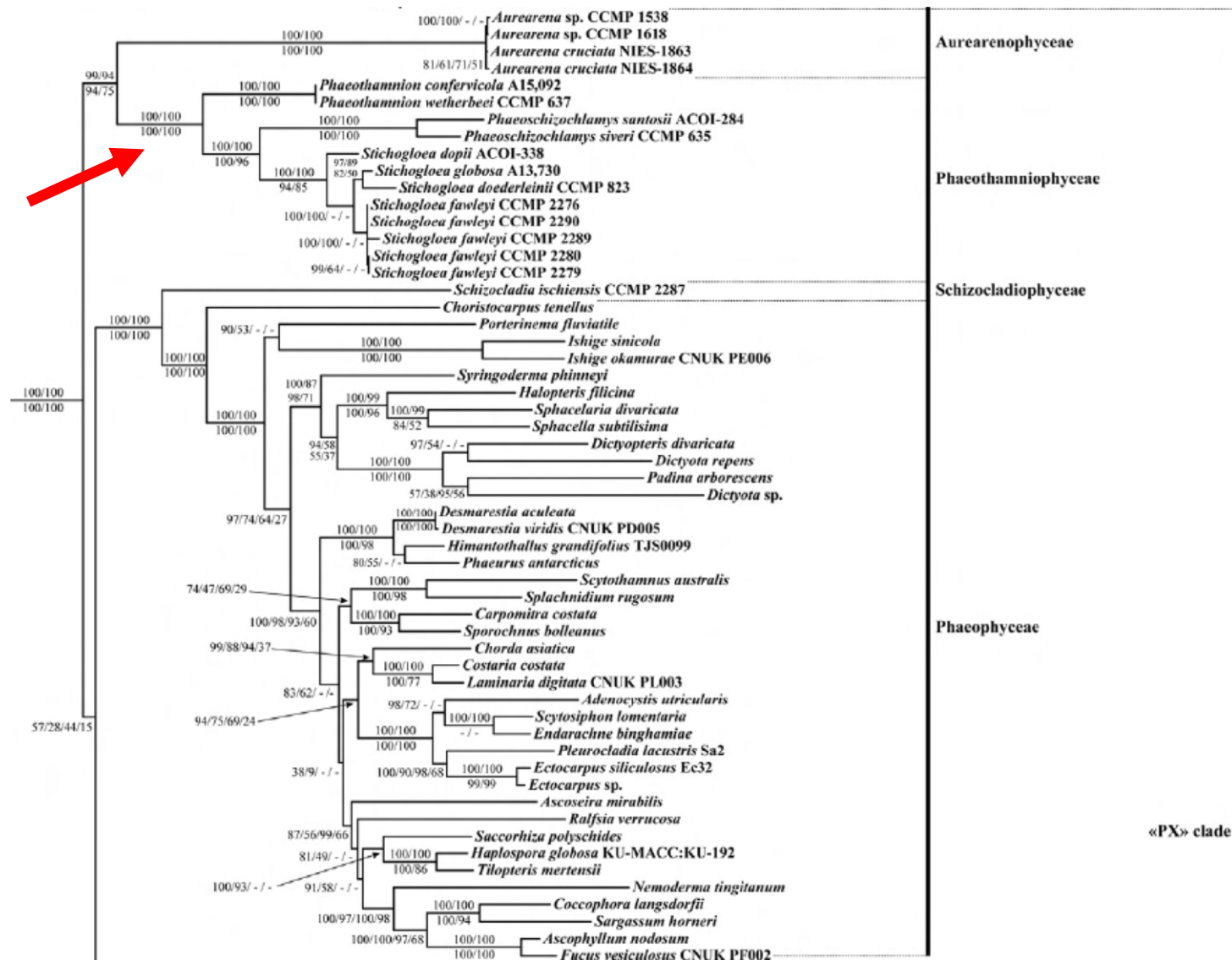
Schizocladiphyceae

- a monotypic class,
Schizocladia ischiensis
- the closest relative of
Phaeophyceae, but probably
not a direct ancestor
- branched filamentous algae
- cell wall containing alginates
(like Phaeohp.) but lacking
cellulose (unlike Xanthoph. and
others) and no plasmodesmata
(unlike Phaeohp.)
- marine, described in 2003
(type locality: Naples, Italy)



Schizocladia ischiensis, vegetative filaments
and swollen reproductive cells (arrows).

Phaeothamniophyceae



Phaeothamniophyceae

- established in 1998 from genera formerly classified in the class Chrysophyceae (but some taxa erroneously classified – polyphyly of Phaeothamniophyceae)
- filamentous, capsoid, palmelloid, ?ciliated, or coccoid
- swimming cells heterokont flagella
- chloroplast with girdle lamella, chlorophyll a, c + xanthophylls (including fucoxanthin)
- alginates present in the cell walls

Phaeothamniophyceae

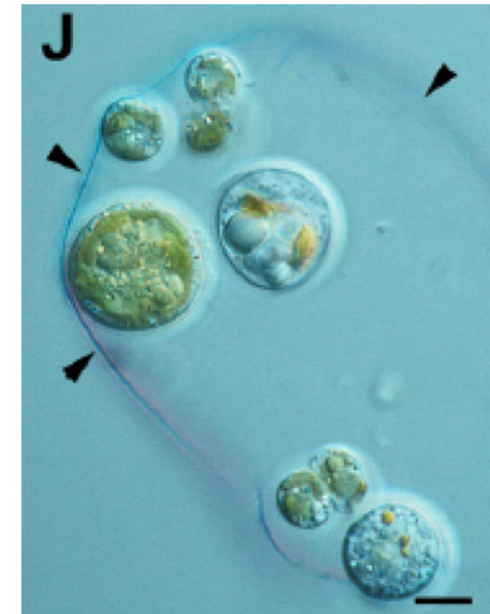
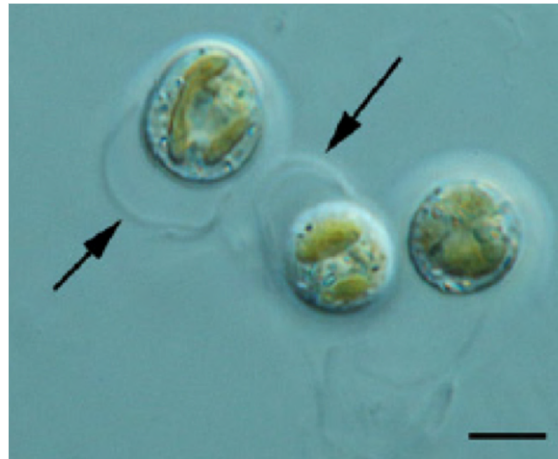
- *Phaeothamnion* spp.
- freshwater bodies, pools and ponds; widely distributed
- filaments with branches, a basal cell attached to substratum (tree like colonies)
- ellipsoidal cells with one or more chloroplasts (no pyrenoid)
- production of heterokont zoospores



Phaeothamniophyceae

Phaeoschizochlamys

- freshwater and brackish waters (pools, lakes)
- colonial within a mucilage
- remnant empty cell walls (cup-like in appearance)
- one or two plastids
- heterokont zoospores

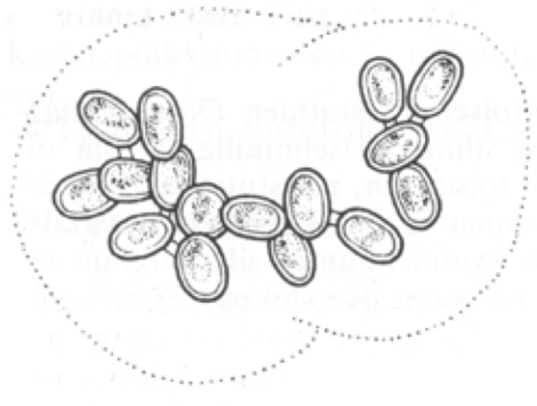


Phaeoschizochlamys santosii

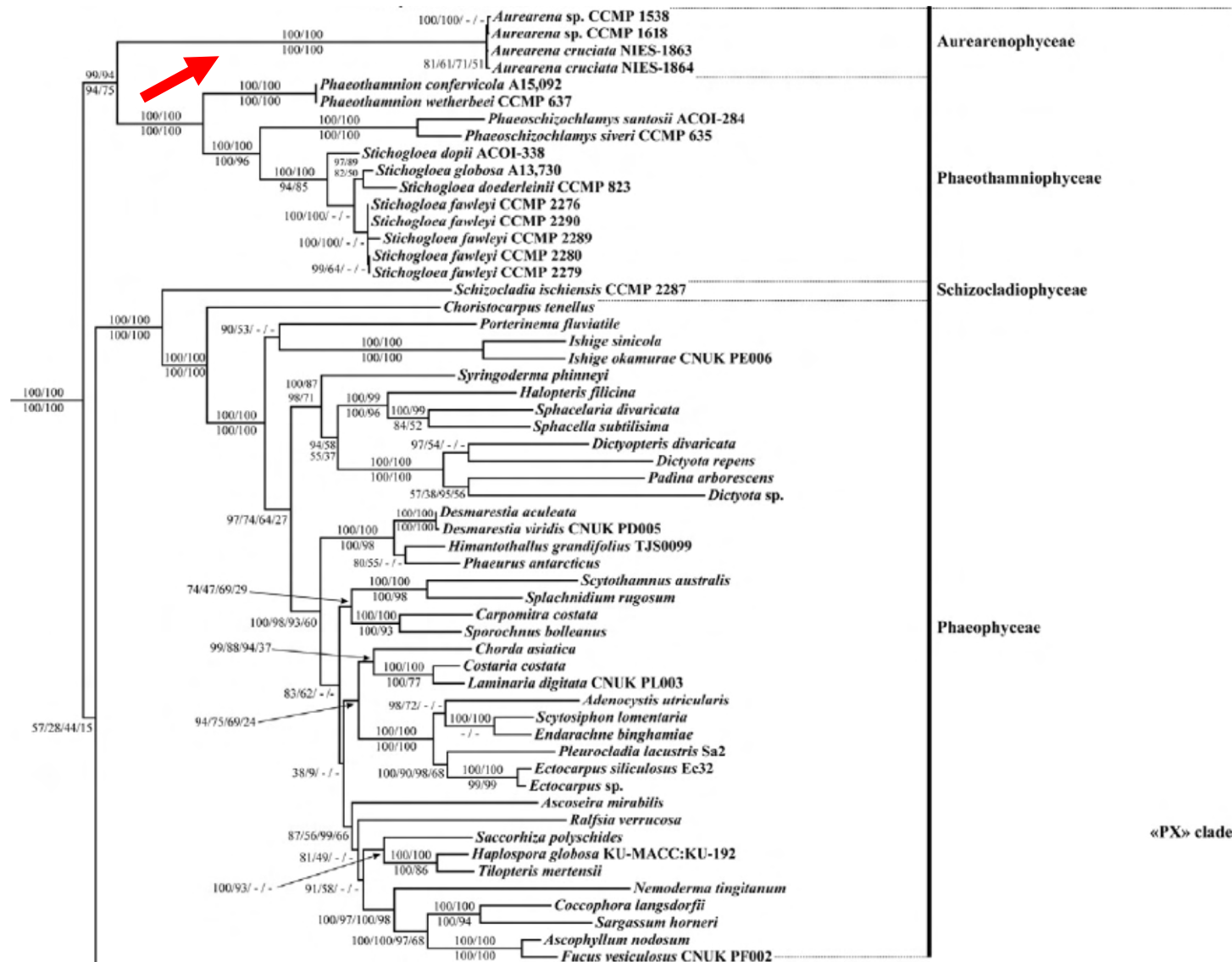
Phaeothamniophyceae

Stichogloea spp.

- freshwater plankton, alpine and northern lakes
- mucilaginous free-floating colonies (4 and 4 cells together)
- 1-2 parietal chloroplasts



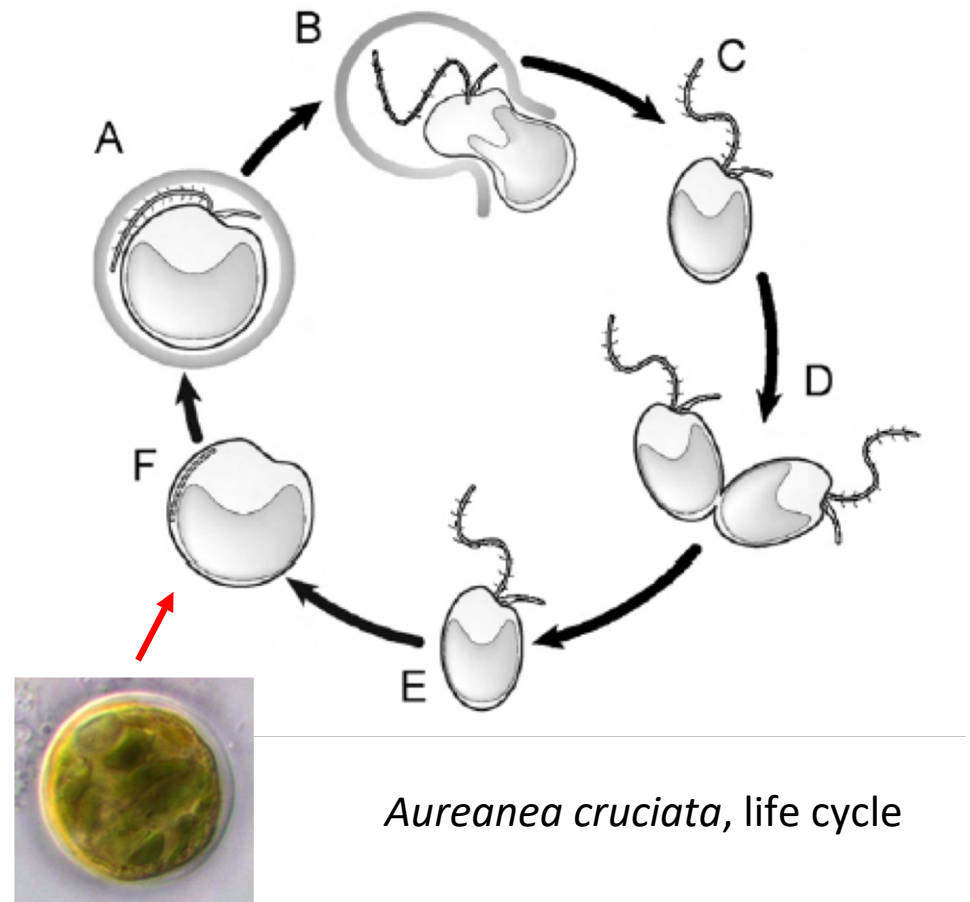
Aurearenophyceae



Aurearenophyceae

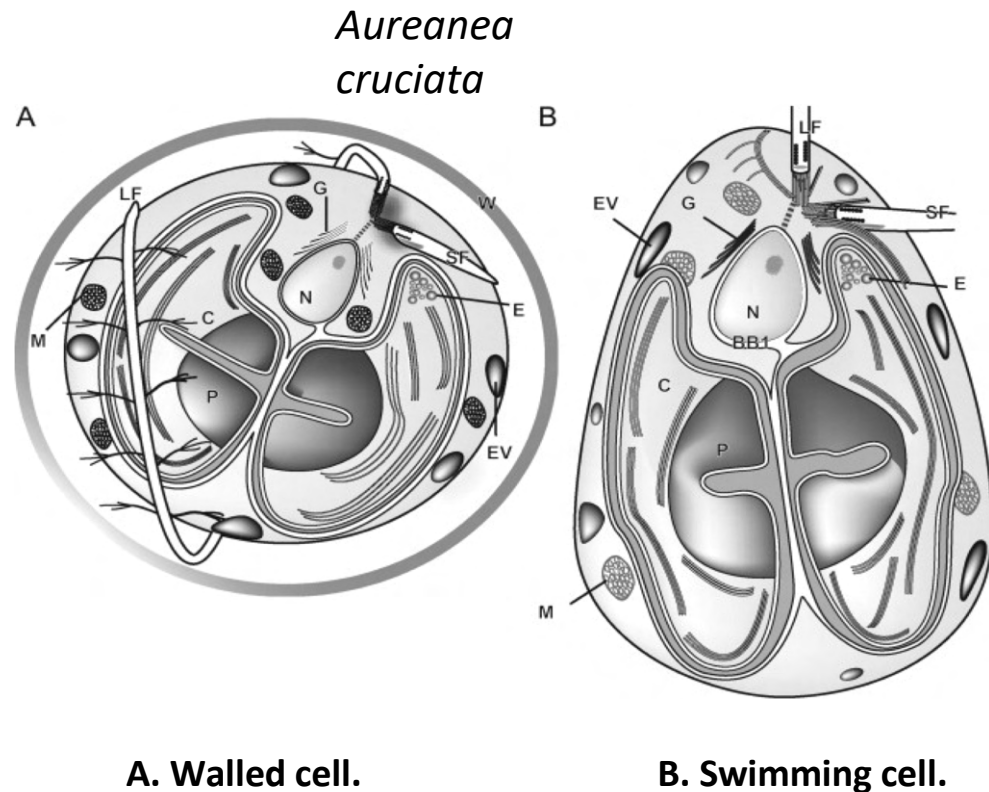
- a monotypic class, *Aureanea cruciata* (described in 2008; type locality: sandy beaches in Japan, etymology: golden sand)
- walled non-motile coccoid cells (flagella underneath the cell wall: not seen elsewhere); swimming cells lack cell walls
- heterokont flagella; golden-yellow chloroplast; with stigma and pyrenoid; lamellae of three thylakoids (no girdle lamella)
- chlorophyll *a* (chl *c* not detected), beta-carotene, various xanthophylls (including fucoxanthin)
- unconventional outer membrane arrangement of plastids

A. A flagellate cell inside a cell wall. B. Naked swimming cell emerging through a pore in the cell wall. D. Cytokinesis proceeding in a swimming cell. E. Swimming cell absorbing flagella into the cell. F. Non-motile cell after absorption of flagella.



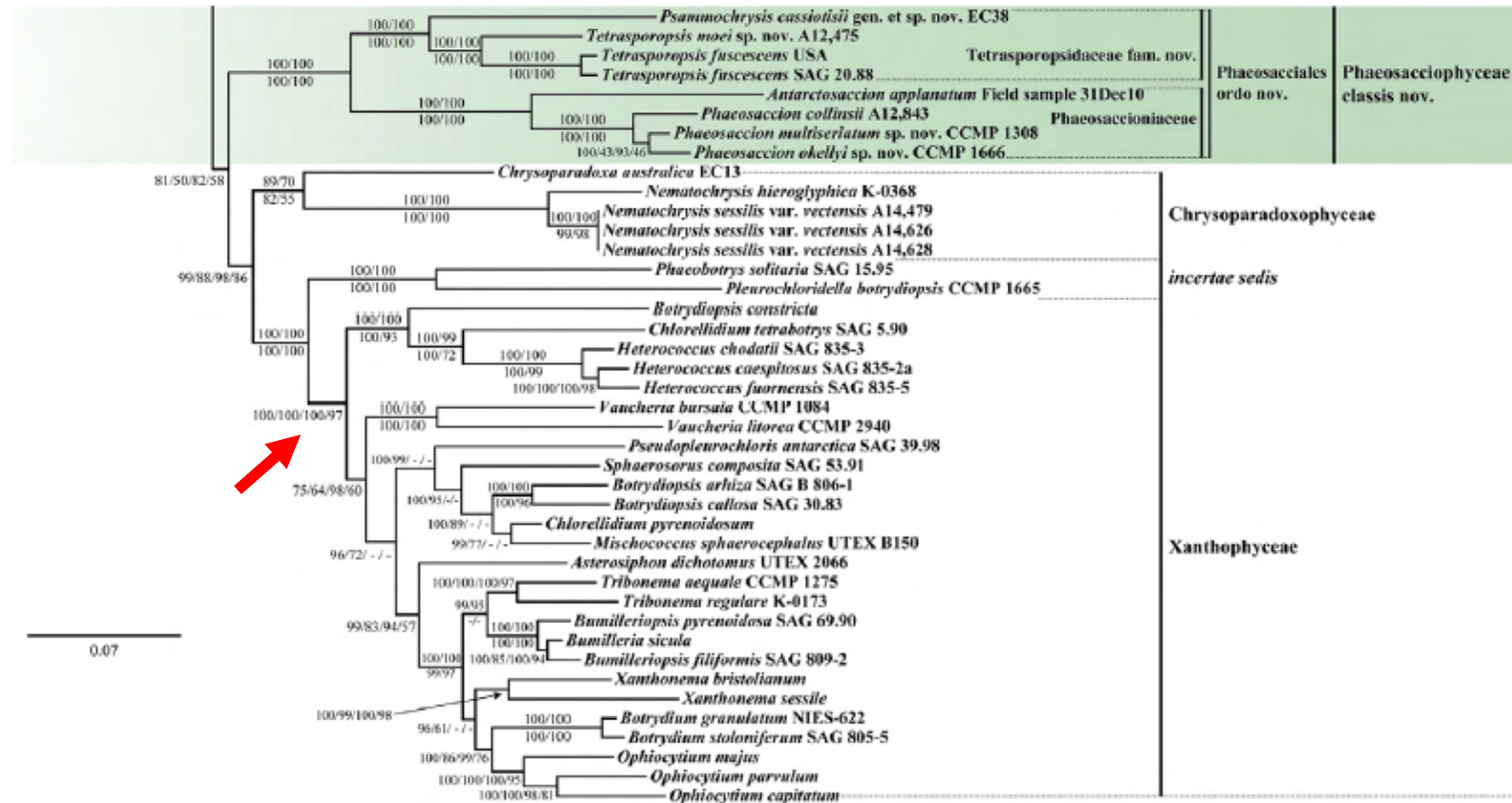
Aurearenophyceae

- a single chloroplast is a consortium of multiple plastids, each surrounded by three inner membranes, is enclosed by a shared outer membrane complex
- this membrane topology has not been reported before



C: chloroplast with cross-shapes furrow (longitudinal = two chloroplast membranes + one ER; transverse = two chloroplast membrane); **E:** eyespot; **G:** Golgi body; **M:** mitochondrion; **N:** nucleus; **P:** central pyrenoid; **W:** cell wall; **EV:** electron opaque vesicle; **BB:** basal body; **LF:** long anterior flagellum.

Xanthophyceae



Xanthophyceae (yellow-green algae)

- predominately coccoid or filamentous, (?amoeboid, ?ciliated or ?capsoid)
- swimming cells with heterokont flagella
- cell walls (cellulose in *Vaucheria*; alginates in the cell walls of *Botrydiopsis*, *Botrydium*, *Tribonema* and *Vaucheria*; cell wall may be impregnated by silica)
- either entire cell wall or two overlapping halves (bipartite cell wall, H-shaped pieces)
- usually several chloroplasts; typical stramenopile ultrastructure; chlorophylls *a*, *c1*, *c2*; xanthophylls



Tribonema, H-shaped pieces of bipartite cell wall

Xanthophyceae (yellow-green algae)

- green to yellow-green color: absence of fucoxanthin (golden/brown color caused by fucoxanthin), chrysolaminaran as a storage product
- chloroplast color: confusions with Chlorophyta (difference: presence of chlorophyll *b* and starch as a storage product) and Eustigmatophyceae (different xanthophyll composition, absence of chlorophyll *c* and fucoxanthin)
- about 600 species, over 90 genera (however, unclear number of true members)



Ophiocythium sp.

Xanthophyceae (yellow-green algae)

- a majority of xanthophyceans reproduce asexually, divide bilaterally (mitotic cell division);
- produce endogenous cysts (impregnated by silica, from two unequal parts) or the akinete (a single celled spore, cell wall from thick parent cell wall)
- zoospores, hemiautospores (without mastigonemata), aplanospores (without flagella); and autospores (small replicates of the parent cell)

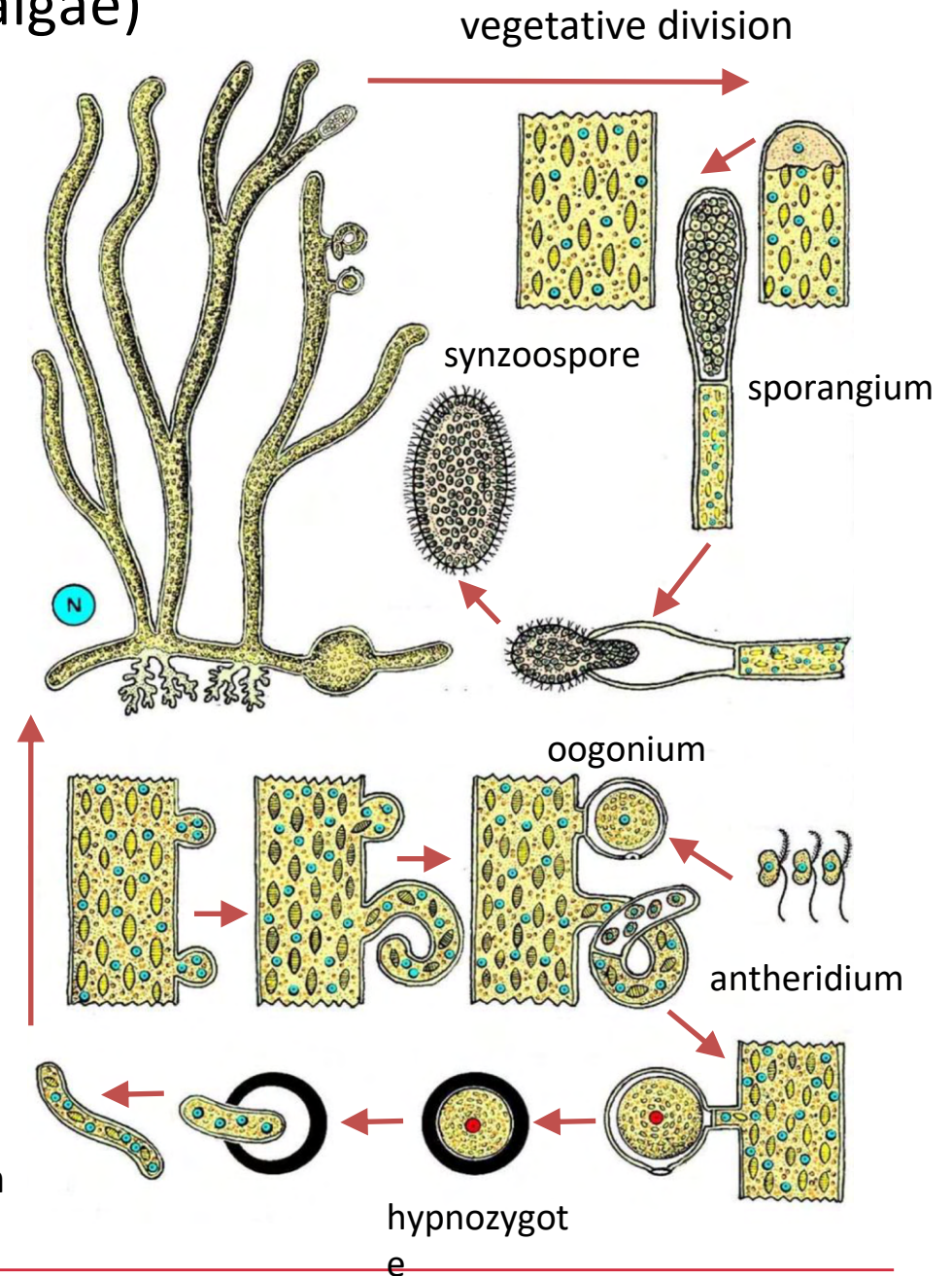


Tribonema, filaments and zoospores

Xanthophyceae (yellow-green algae)

- sexual reproduction observed in *Vaucheria*: oogamy (antheridium: producing a large number of colorless spermatozoids, oogonium: uninucleate, a single oosphere) -> a thick-walled resting zygote (oospore = hypnozygote) -> meiosis, a new siphonous filament
- *Botrydium*: isogametes or anisogametes
- *Tribonema*: isogamous

Vaucheria,
sexual reproduction



Xanthophyceae (yellow-green algae)

- from tropics to polar regions
- predominantly in freshwater as phytoplankton and periphyton + terrestrial habitats [*Vaucheria* in marine/brackish habitats]
- their habitat appear somehow as exceptions (together with Eustigmatoph.) among the Ochrophytes [most groups with red-algal derived plastids dominate in the oceans]
- they resemble ecologically and morphologically Chlorophyta: evolutionary parallelism
- most taxa only rarely (and in small numbers) but some commonly form visible growths (filaments of *Vaucheria* and *Tribonema*) and some are macroscopic (*Botrydium*, *Asterosiphon*)



Botrydium, macroscopic thallus

Xanthophyceae – **systematics**

- traditional classification does not reflect phylogeny: poly- or paraphyletic taxa (order, families, genera)
- coccoid and filamentous taxa with cell wall confirmed as Xanthophyte members
- other species are doubtful and may represent new stramenopile lineages with uncertain phylogenetic position

Xanthophyceae – **systematics**

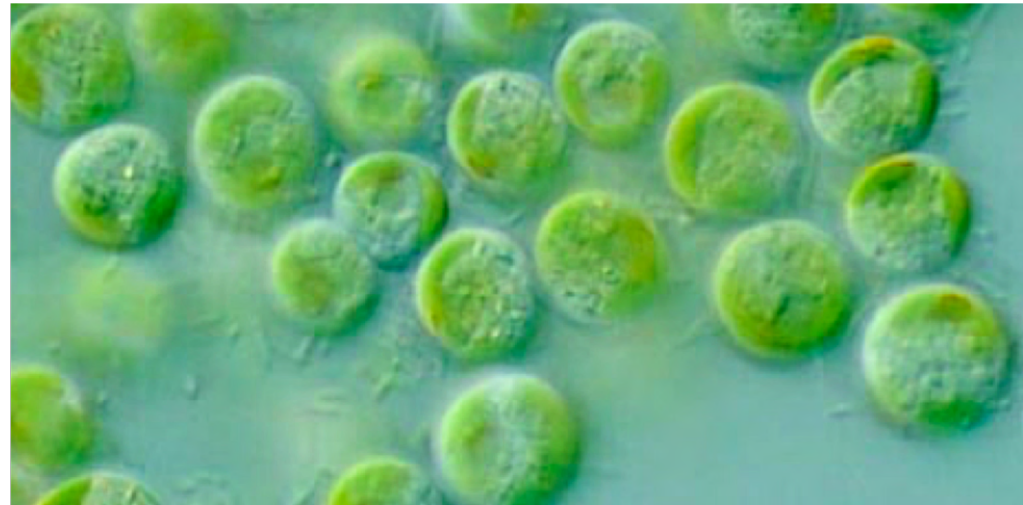
- **doubtful orders of Xanthophytes:** absence of the cell wall
- Heterochloridales = Chloramoebales: unicellular naked flagellates (constant shape or amoeboid); one to several chloroplasts, palmeloid stages, usually freshwater (also found in marine and brackish waters), *Chloromeson* → unavailable molecular data; some representatives transferred to Chrysophyta

Xanthophyceae – **systematics**

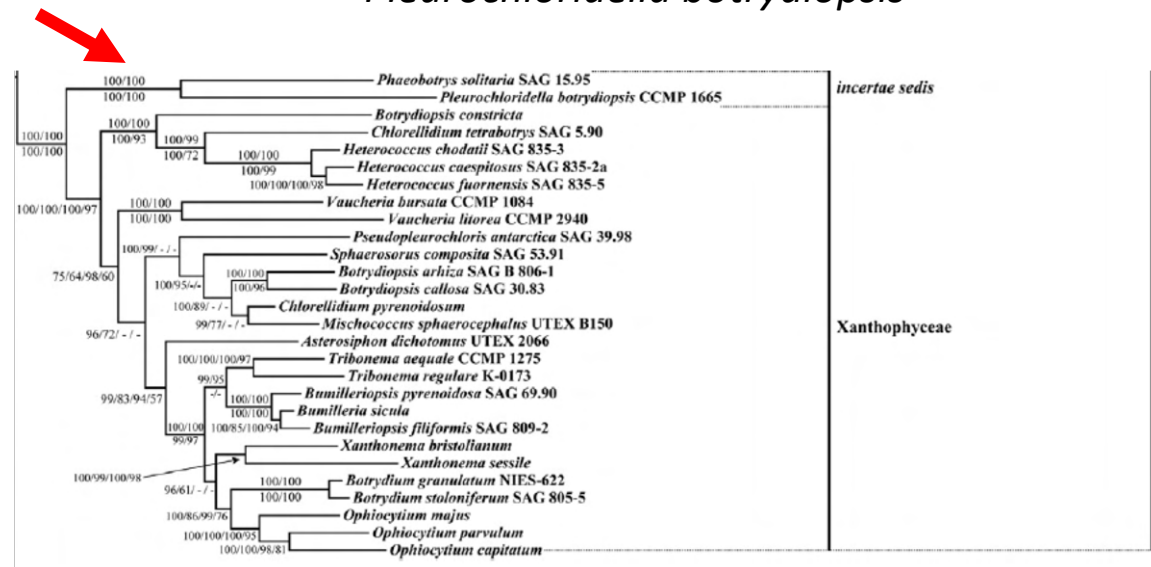
- **doubtful orders of Xanthophytes:** absence of the cell wall
- Heterochloridales = Chloramoebales: unicellular naked flagellates
- Heterogloeales: capsoid (= palmeloid) forms (non-motile cells without wall, surrounded by mucilage or not), a former member *Pleurochloridella*: phylogenetically not included in the class Xanthophyceae

Pleurochloridella

- *Pleurochloridella botrydiopsis*:
marine representative (molecular data not available for a freshwater species *P. vacuolata*)
- described in 1938 as Xanthoph.
-> moved to Phaeothamnioph.
(1998) on the basis of pigment analysis -> phylogenetically related to Xanthophytes (not to Phaeothamnioph.)
- differs from Xanthophytes by possessing fucoxanthin



Pleurochloridella botrydiopsis

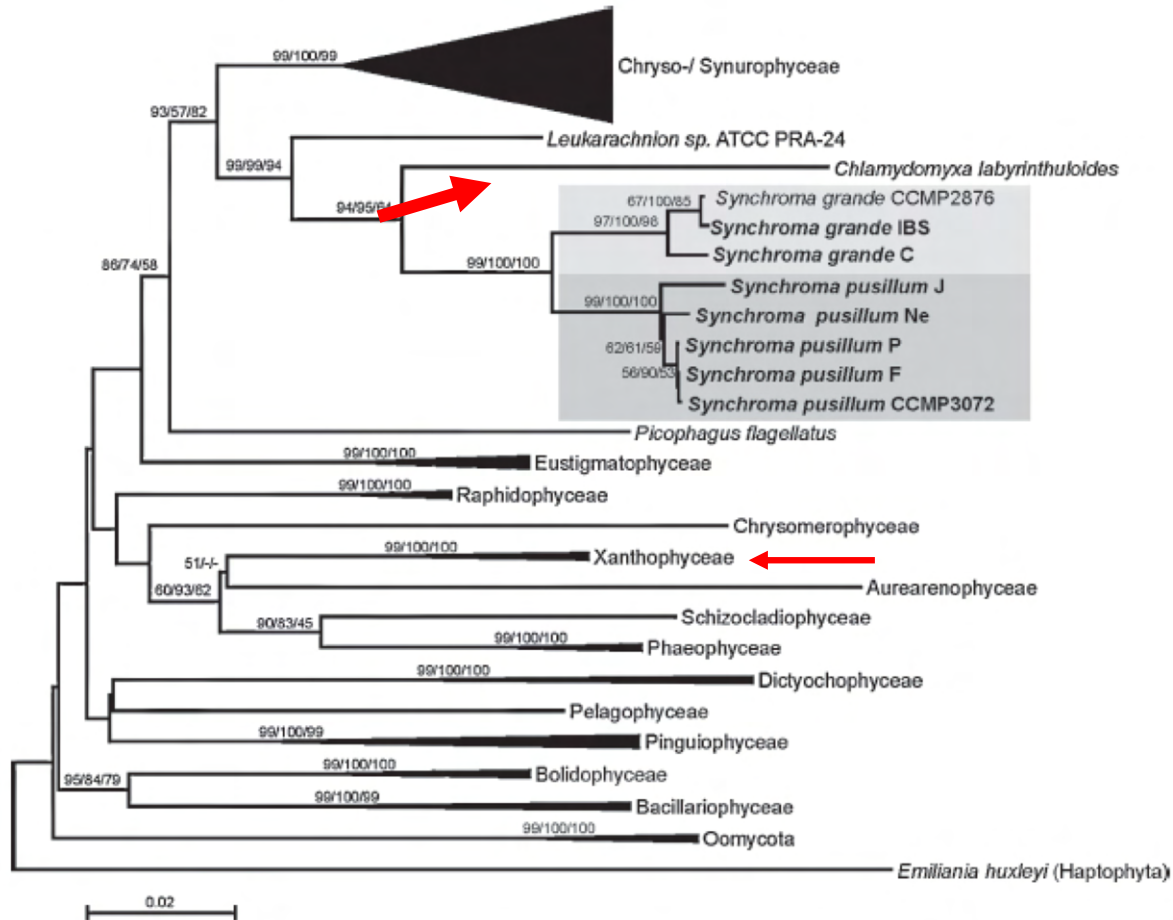


Xanthophyceae – **systematics**

doubtful orders of Xanthophytes: absence of the cell wall

- Heterochloridales = Chloramoebales: unicellular naked flagellates
- Heterogloeales: capsoid (= palmeloid) forms
- Rhizochloridales: rhizopodial forms (naked amoeboid cells with rhizopods); colonial, free living or attached (sometimes loricate); one or more chloroplasts; endogenous cysts; freshwater, marine or brackish; *Rhizochloris* (heterotrophic) and *Myxochloris* (autotrophic): within *Sphagnum* water-holding cells; *Stipitococcus*, *Stipitoporus*: freshwater, live epiphytically on filamentous algae → a former member *Chlamydomyxa labyrinthuloides*: phylogenetically closely related with the class Synchronomophyceae (SII clade)

Chlamydomyxa labyrinthuloides



- chlorophyll a, c + fucoxanthin, violaxanthin



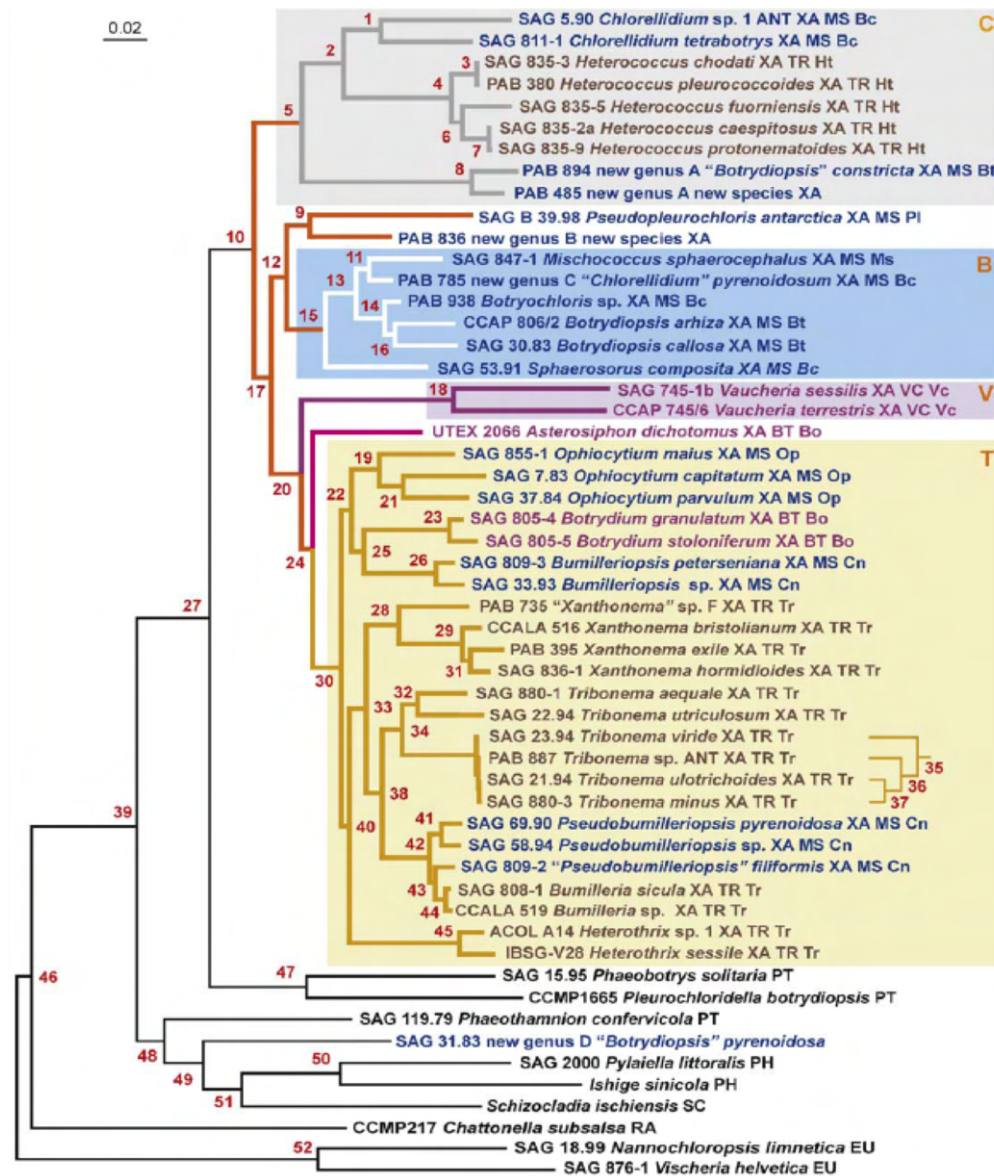
Ch. labyrinthuloides, plasmodium,
on Sphagnum and other water plants

Xanthophyceae – **systematics**

approved representatives of Xanthophytes but polyphyletic orders

- Mischoococcales: coccoid forms
- Tribonematales: filamentous (branched and unbranched forms)
- Botrydiales: siphonous forms
- Vaucheriales: siphonous filaments
- orders and some genera are not monophyletic
- some representatives of Mischoococcales are actually Eustigmatophyceae (*Goniochloris*, *Characiopsis*)
- *Botrydium pyrenoidosa* form a new lineage **outside** of Xanthoph.

Xanthophyceae – systematics



Chlorellidium and others
- coccoid and heterotrichal

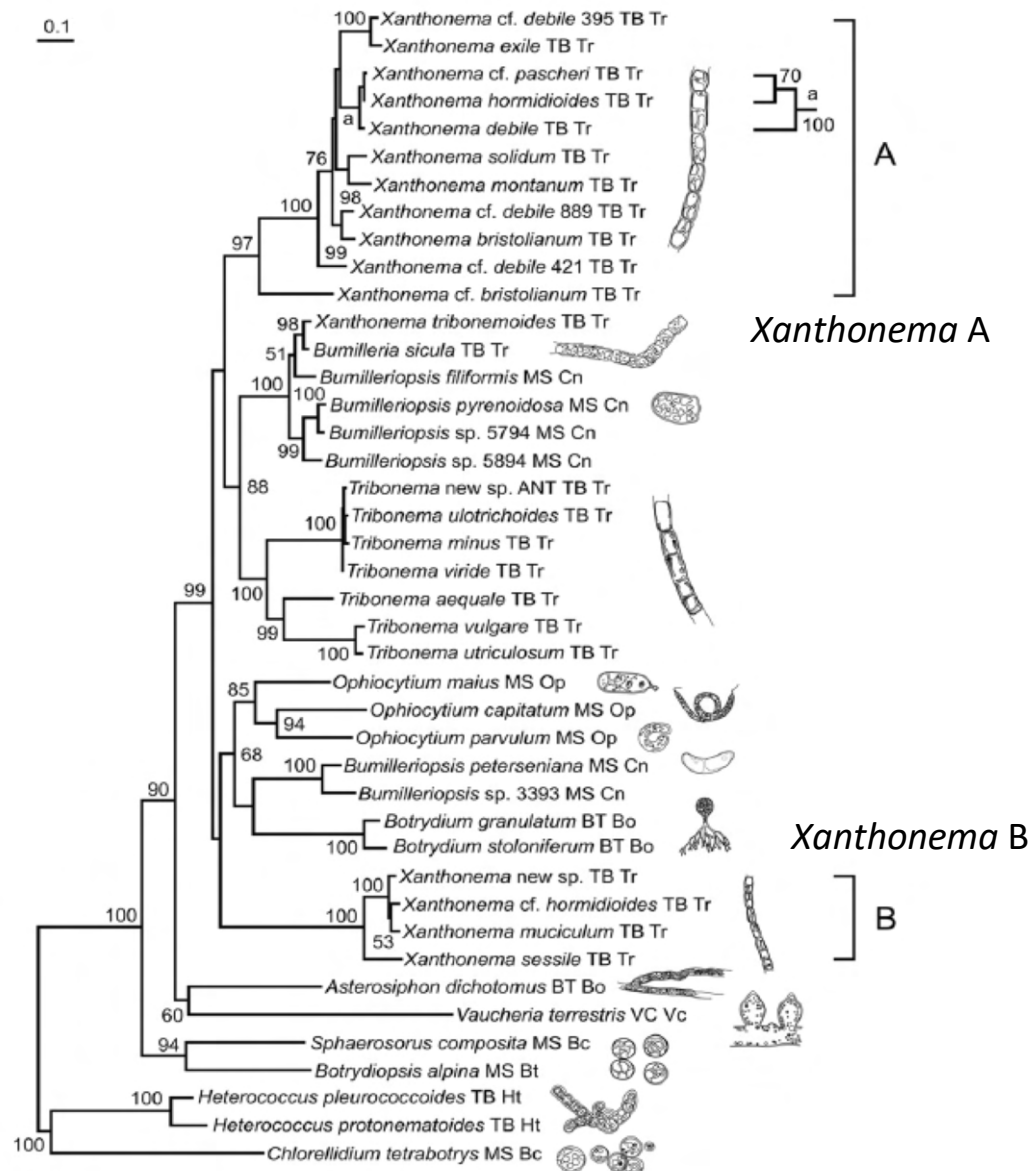
Botrydiopsis and others
- most of the coccoid forms
included in Mischococcales

Vaucheria - siphonous

Tribonema and others
- coccoid, unbranched
filaments, siphonous thalus

4 major clades

Xanthophyceae (yellow-green algae)



Xanthonema



Bumilleria



Tribonema

Xanthophyceae (yellow-green algae)

a split of the filamentous Xanthophyceae in two independent subclades:

- 1) bipartite cell walls of H-shaped pieces; 2) cell walls of one piece

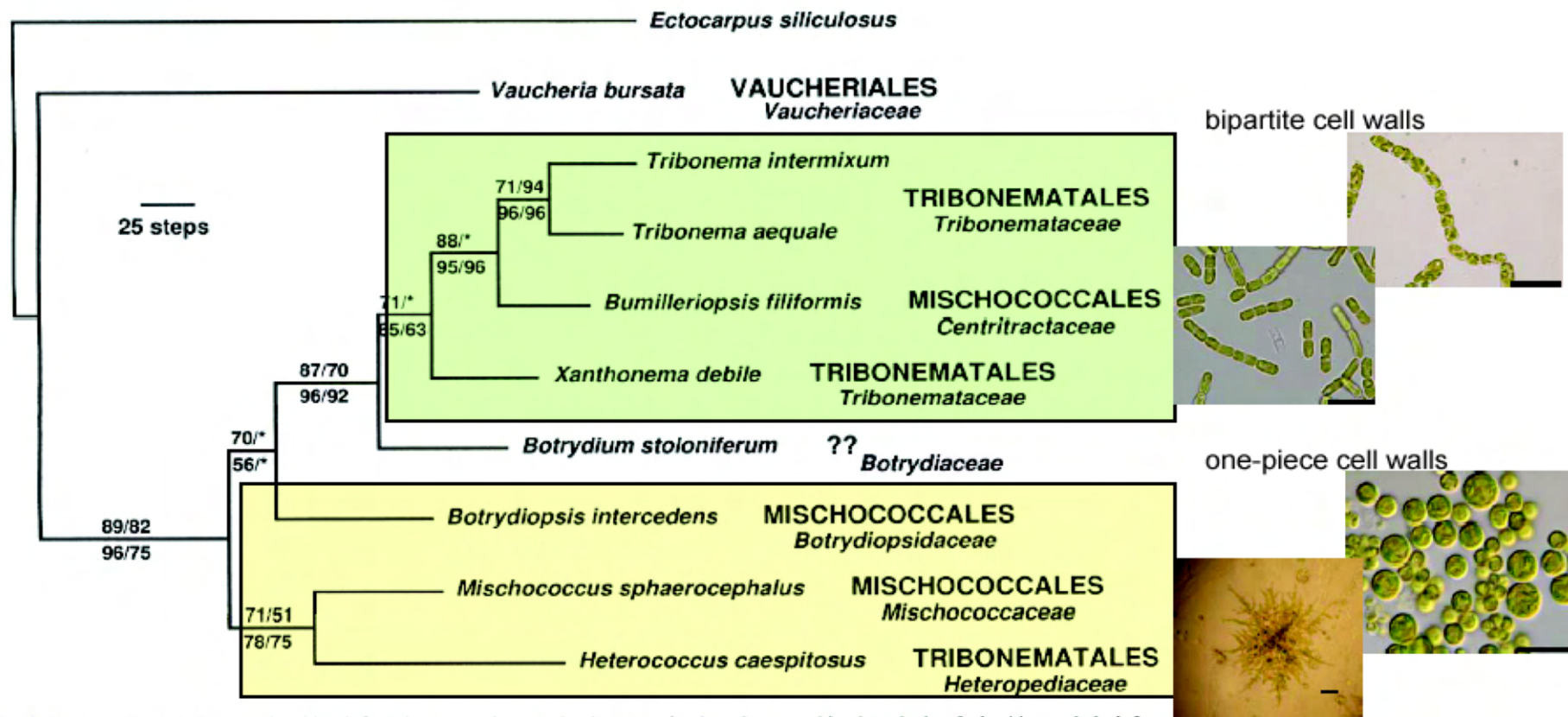
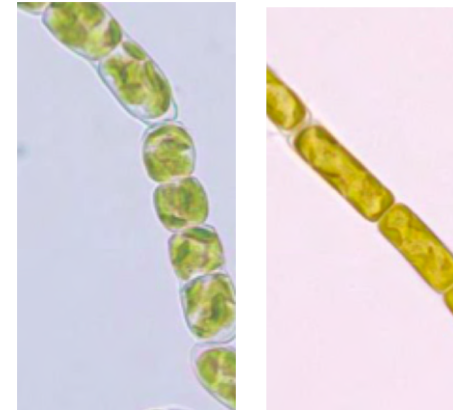
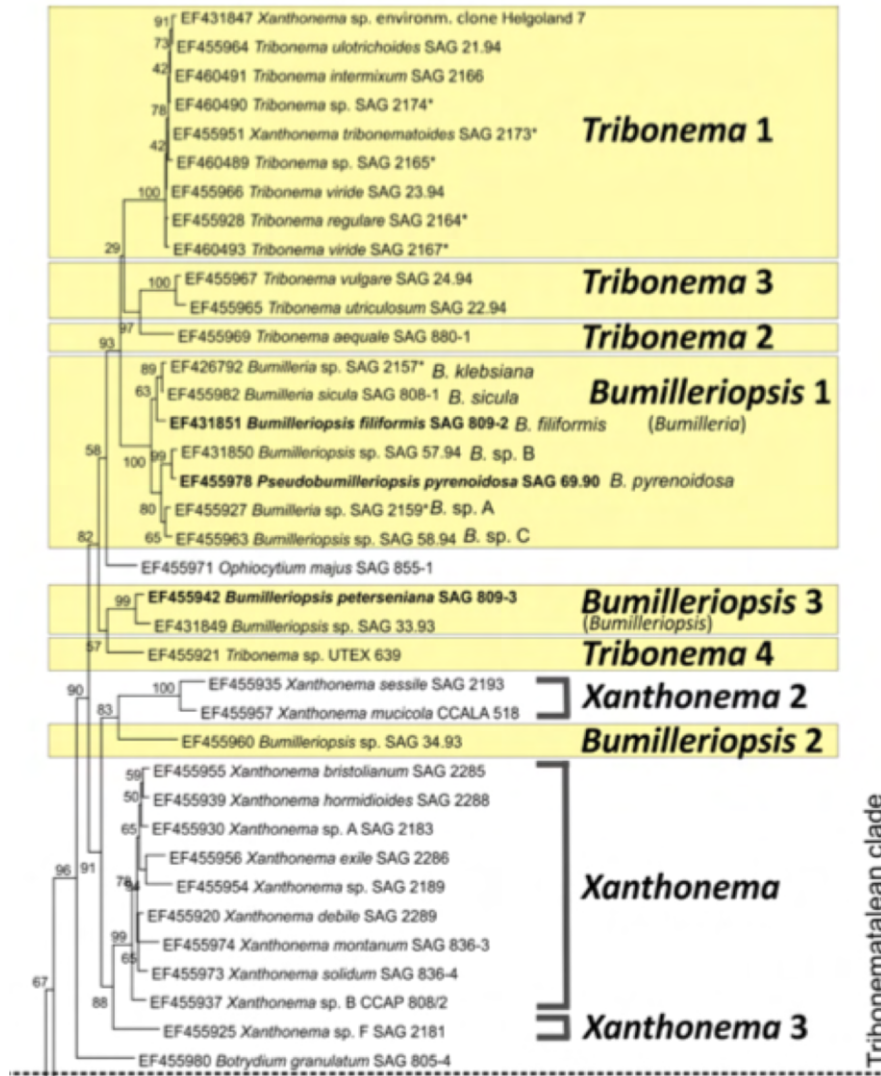
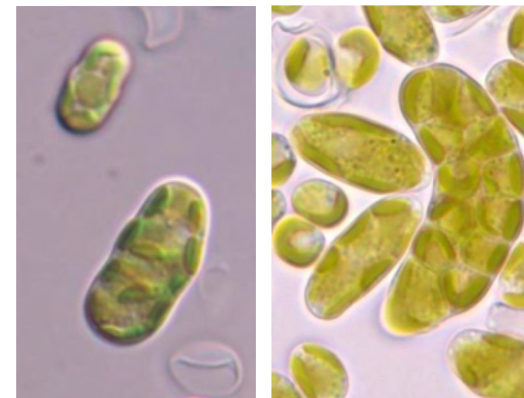


Fig. 3. Phylogram depicting relationships inferred among nine xanthophyte species based on combined analysis of plastid-encoded *rbcL* and nuclear-encoded 18S rRNA gene sequences. Bootstrap values above branches correspond to separate parsimony analyses of the *rbcL* and 18S rRNA data, respectively; values shown below branches are derived from parsimony/ML analyses of the combined data set. *, bootstrap value < 50%; ??, *Botrydium* (Botrydiaceae) has been classified in the Botrydiales (Ettl 1978), Mischococcales (Christensen 1980), and Vaucheriales (Bold & Wynne 1985; Hibberd 1990).

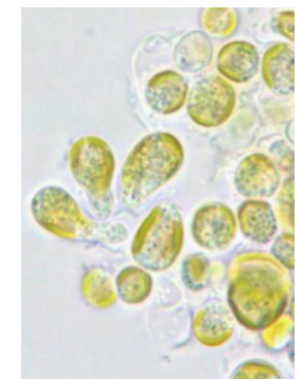
Xanthophyceae – *Bumilleriopsis*



Bumilleriopsis 1 (*Bumilleria*)



Bumilleriopsis 2

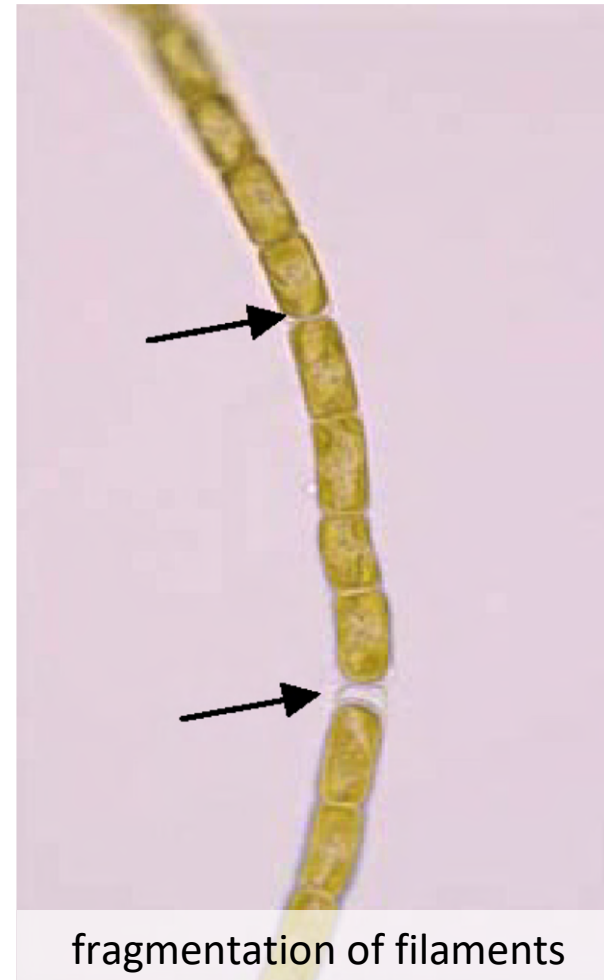


Bumilleriopsis 2 (new genus)

bipartite cells

Xanthophyceae – *Bumilleria*

- unbranched filaments, cylindrical to quadrate cells
- fragmentation of filaments, two H-shaped pieces
- reproduction by zoospores
- freshwater; tychoplanktonic and metaphytic (among other filamentous algae) in creeks, rivers, wetlands, and ponds
- *Tribonema* clade

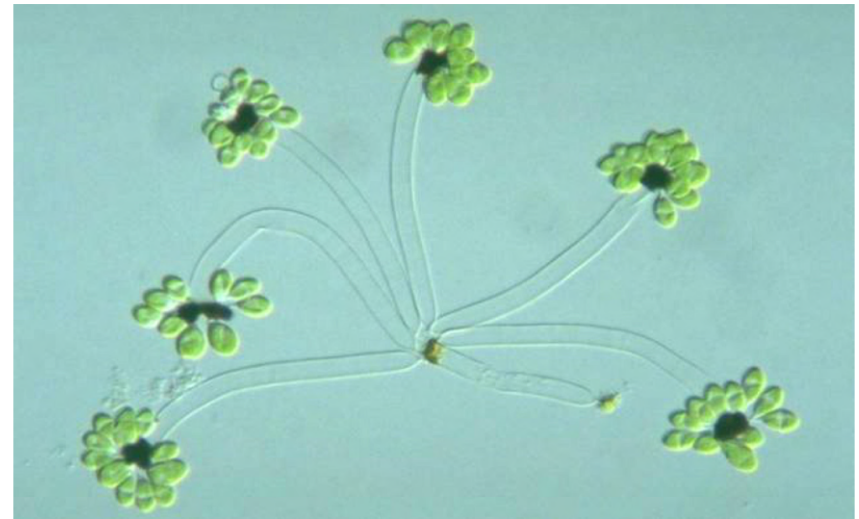


Xanthophyceae - *Ophiocytium*

- freshwater standing waters (euplanktonic, tychoplanktonic, metaphytic, epiphytic)
- solitary or colonial
- elongate-cylindrical cells
- bipartite cell wall; several to many plastids
- autospores, zoospores, aplanospores
- *Tribonema* clade

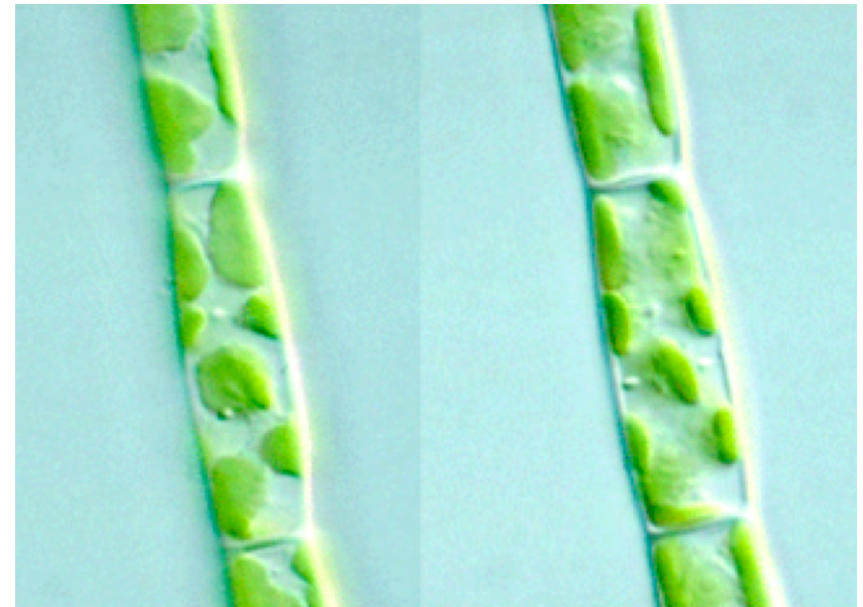


spines on the cell



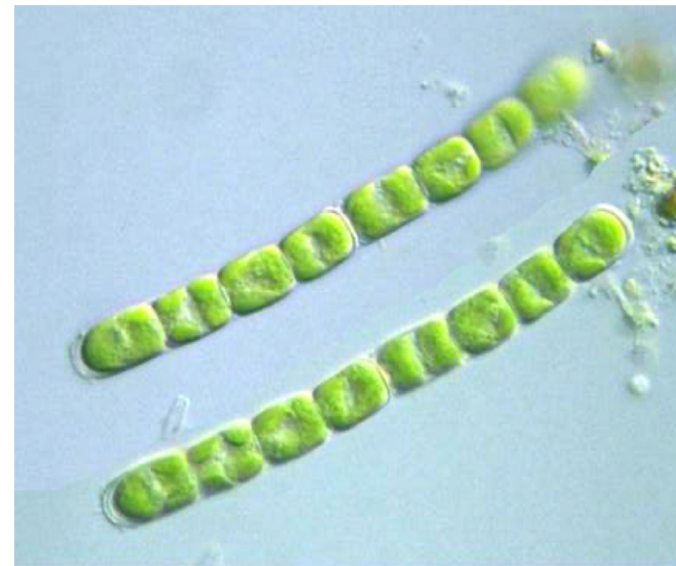
Xanthophyceae - *Tribonema*

- frequently in clear, slightly acidic [some may be found up to pH = 9] freshwaters (standing and running waters, tychoplanktonic and metaphytic)
- unbranched uniseriate filaments; bipartite cell wall (H-shaped pieces), long, cylindrical cells
- one to many chloroplasts, zoospores, aplanospores, resting cysts
- polyphyletic genus
- *Tribonema* clade



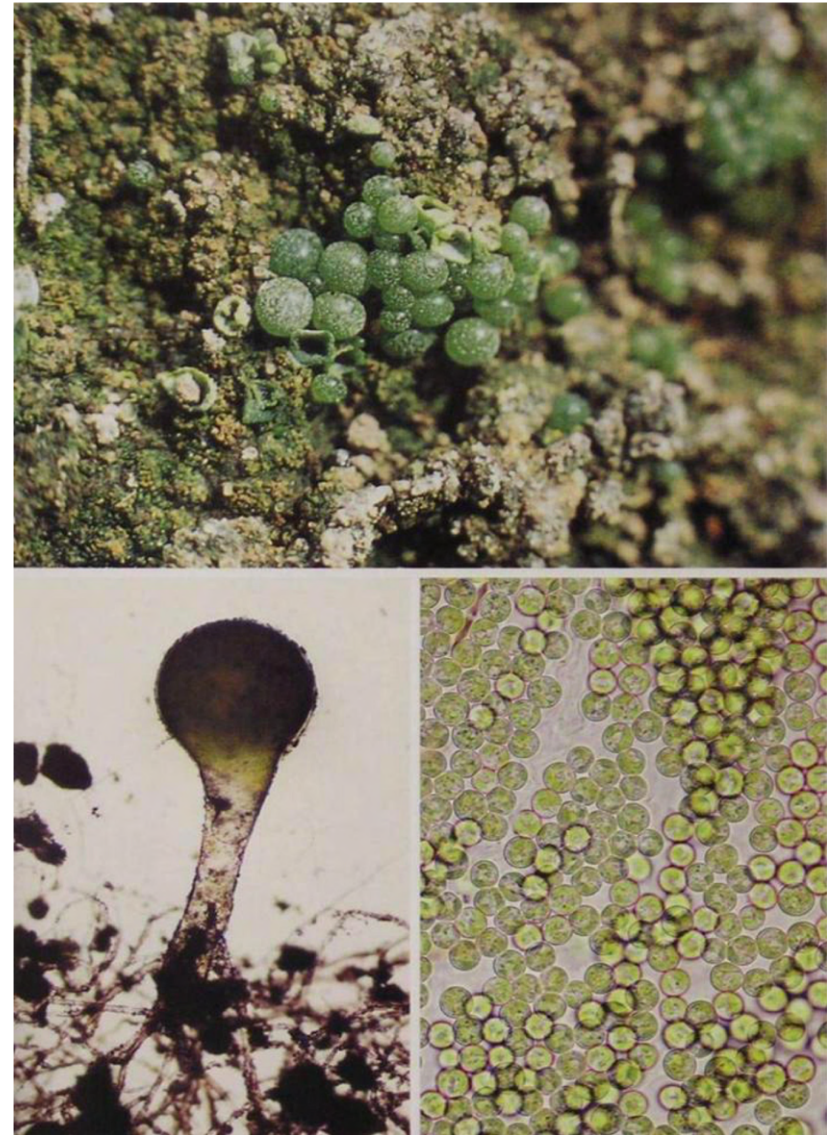
Xanthophyceae - *Xanthonema*

- frequent soil alga, also euplanktonic and metaphytic in freshwater standing waters
- unbranched uniseriate filaments; fragmentation of filaments
- bipartite cell wall (H-shaped pieces), one or two chloroplasts
- polyphyletic genus
- *Tribonema* clade



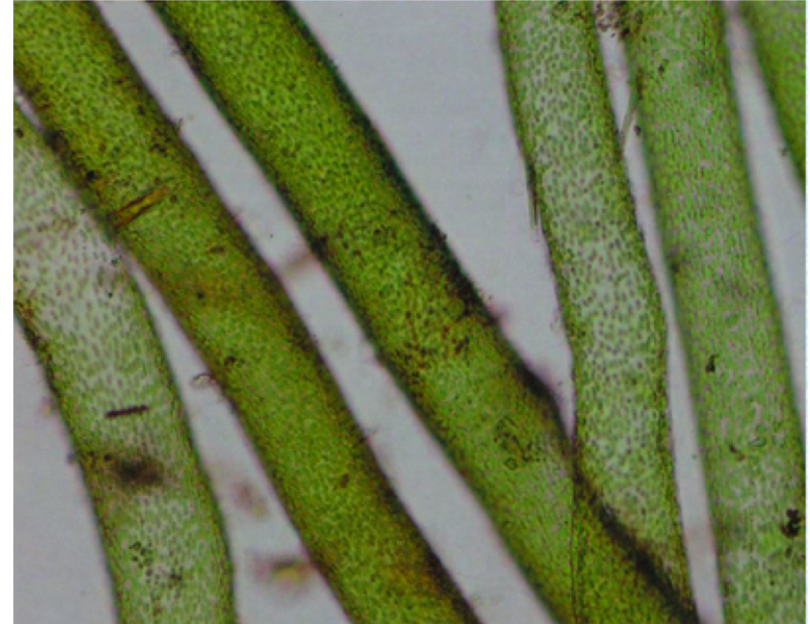
Xanthophyceae - *Botrydium*

- macroscopic sack like vesicles
- aerial habitats (damp soil, drying mud in freshwater environments)
- siphonal thallus: large number of nuclei and chloroplasts, and subterranean colorless branched rhizoids
- zoospores, aplanospores; gametes from undifferentiated vegetative cells
- *Tribonema* clade

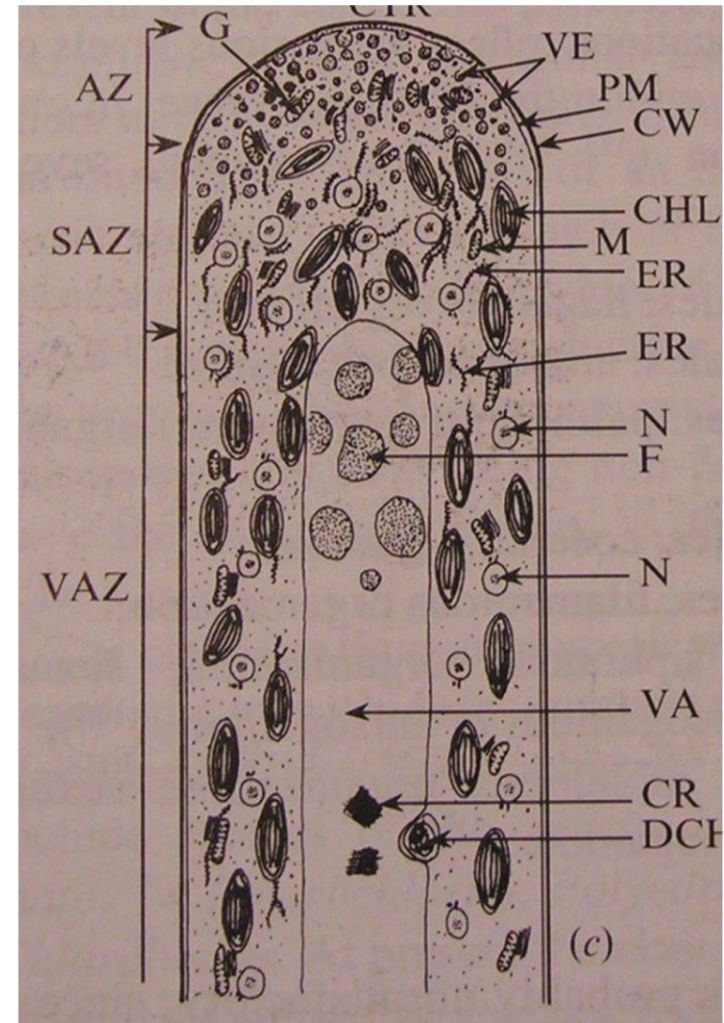
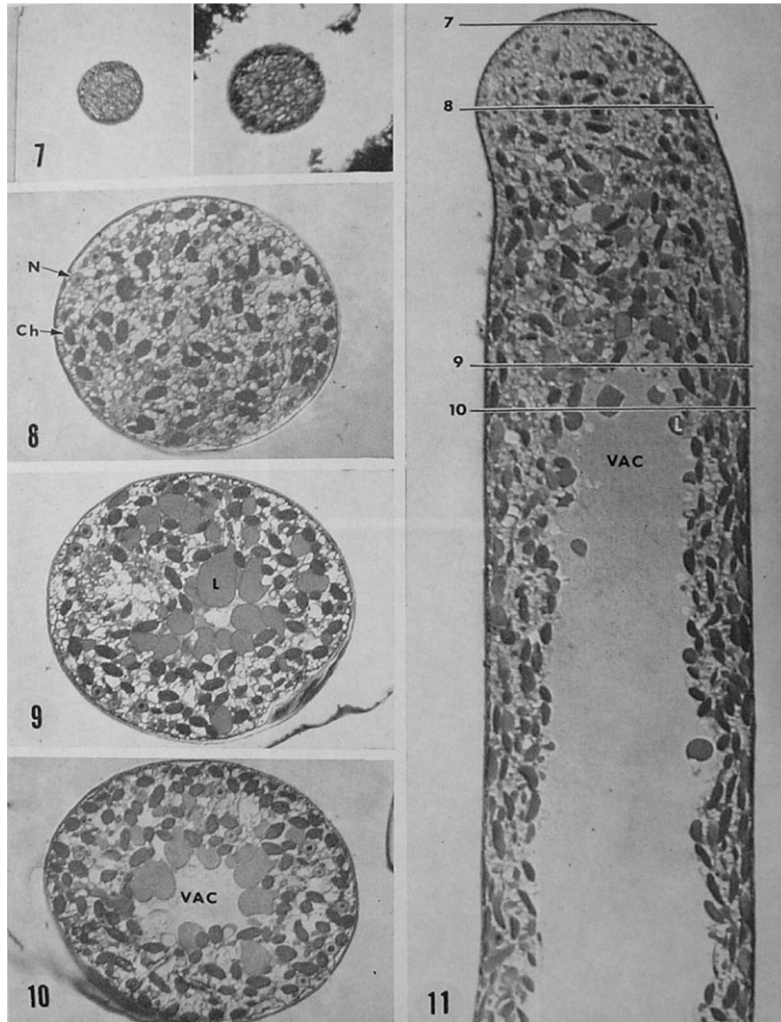


Xanthophyceae - *Vaucheria*

- branched siphonous filaments (unlimited apical growth, cross walls during reproduction)
- large number of chloroplasts and nuclei in peripheral cytoplasmatic layer surrounding a central vacuole
- extensive growths on moist soil (stabilization of soils) or grow submerged (many species amphibious)
- freshwater, marine, and brackish
- *Vaucheria* clade

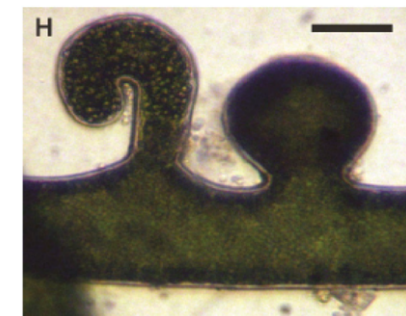
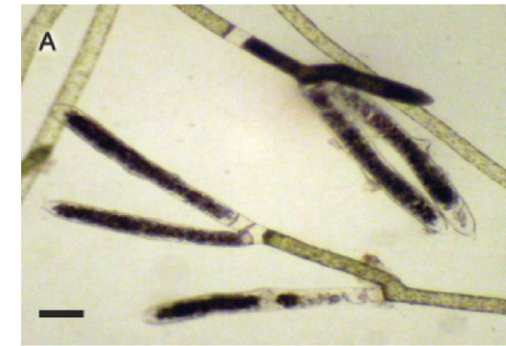
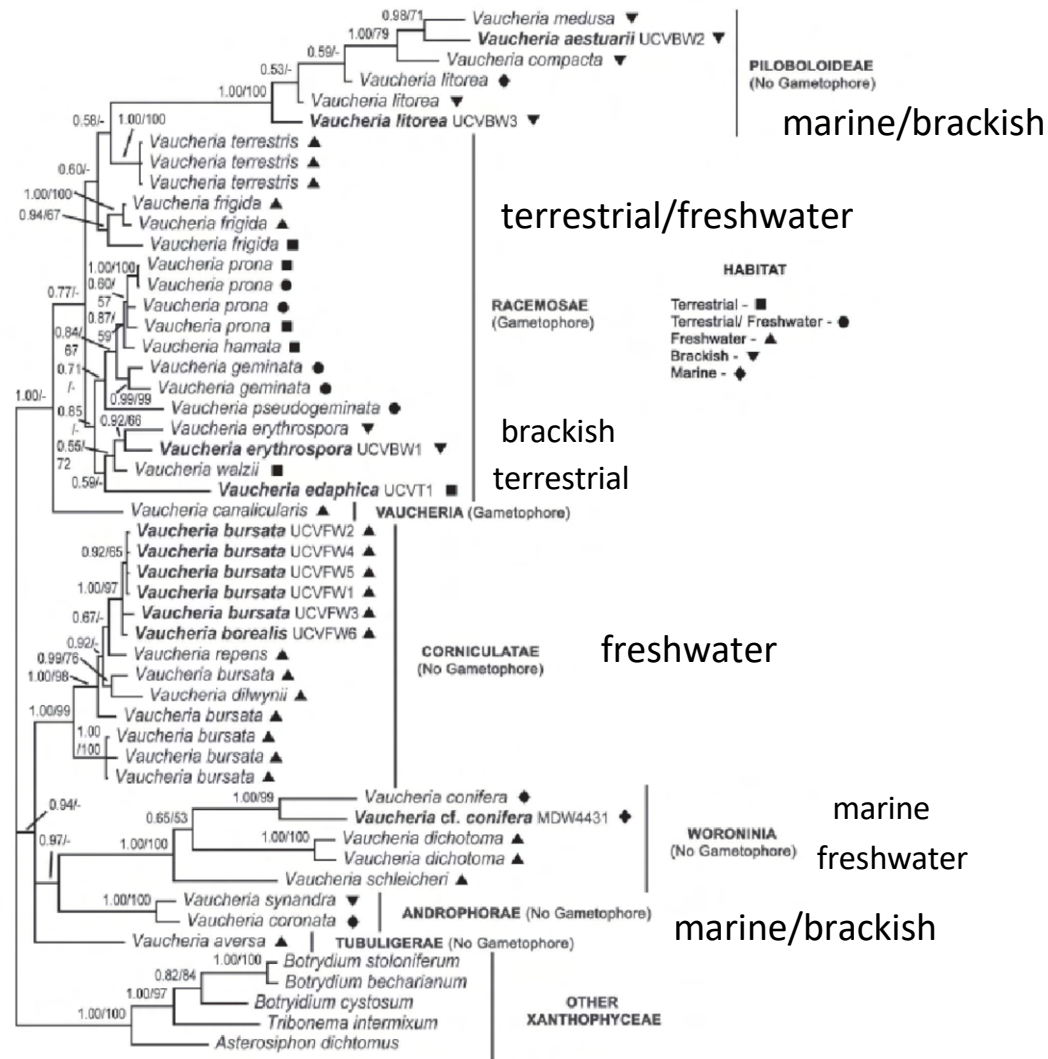


Xanthophyceae - *Vaucheria*



AZ: apical zone, SAZ: subapical zone, VAZ: vacuolised zone, M: mitochondria, VA: vacuole, CHL: chloroplast, N: nucleus, CW: cell wall, PM: plasma membrane, M: mitochondria, ER: endoplasmatic reticulum

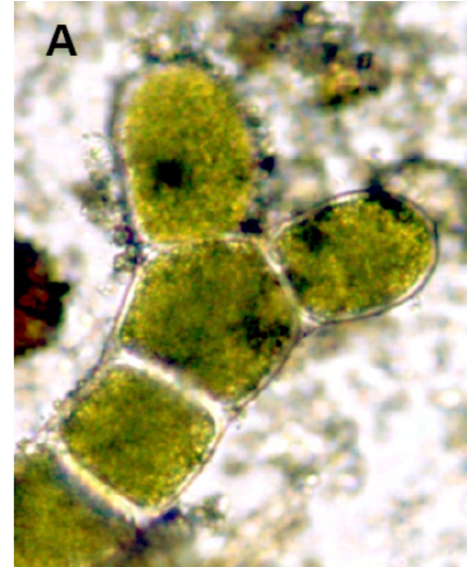
Xanthophyceae - *Vaucheria*



Habitat origin and gametophore formation mapped on phylogeny

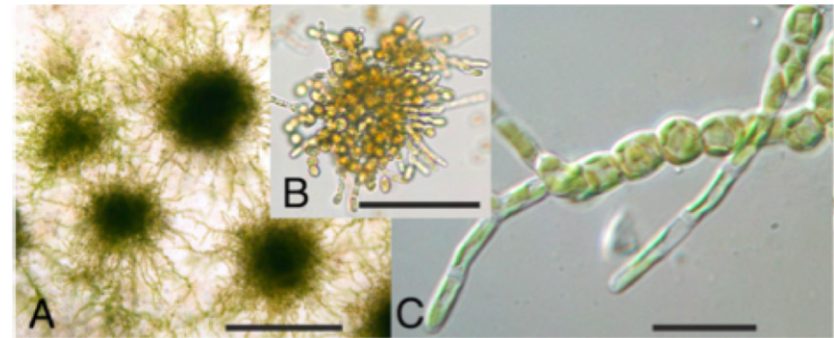
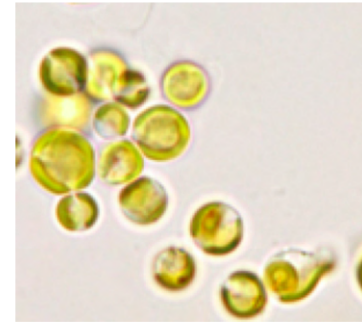
Xanthophyceae - *Asterosiphon*

- filaments as *Vaucheria*
- regular branching forming rosettes (up to 1 cm)
- moist soil
- most likely its phylogenetic position outside the *Vaucheria* clade



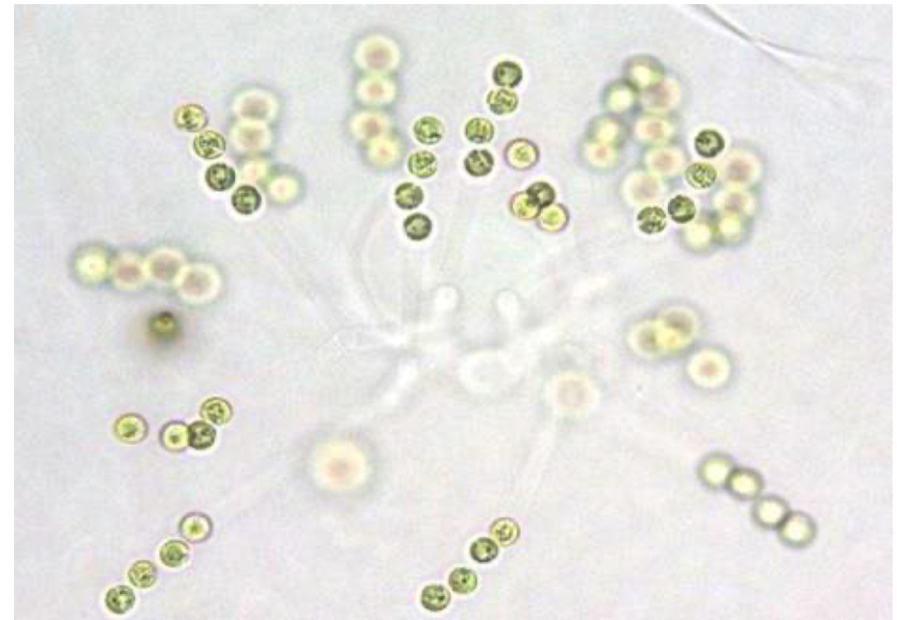
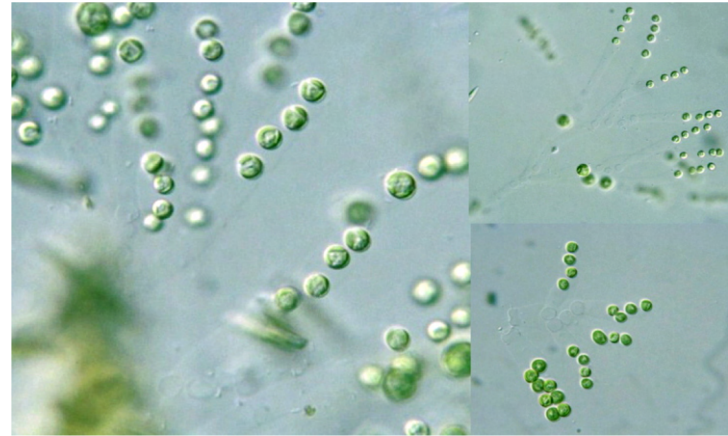
Xanthophyceae - *Heterococcus*

- common and widespread in soils of cold regions [some are freshwater: epiphytic in dystrophic pools]
- unicellular coccoid cells (that are weakly connected) in field samples; branched uni- to multi-seriate filaments (pseudoparenchymatous basal and erect filamentous part) on agar plates
- one-piece cell wall, a single chloroplast; production of zoospores and aplanospores
- *Heterococcus caespitosus*: photobiont of lichen *Verrucaria*
- *Chlorellidium* clade

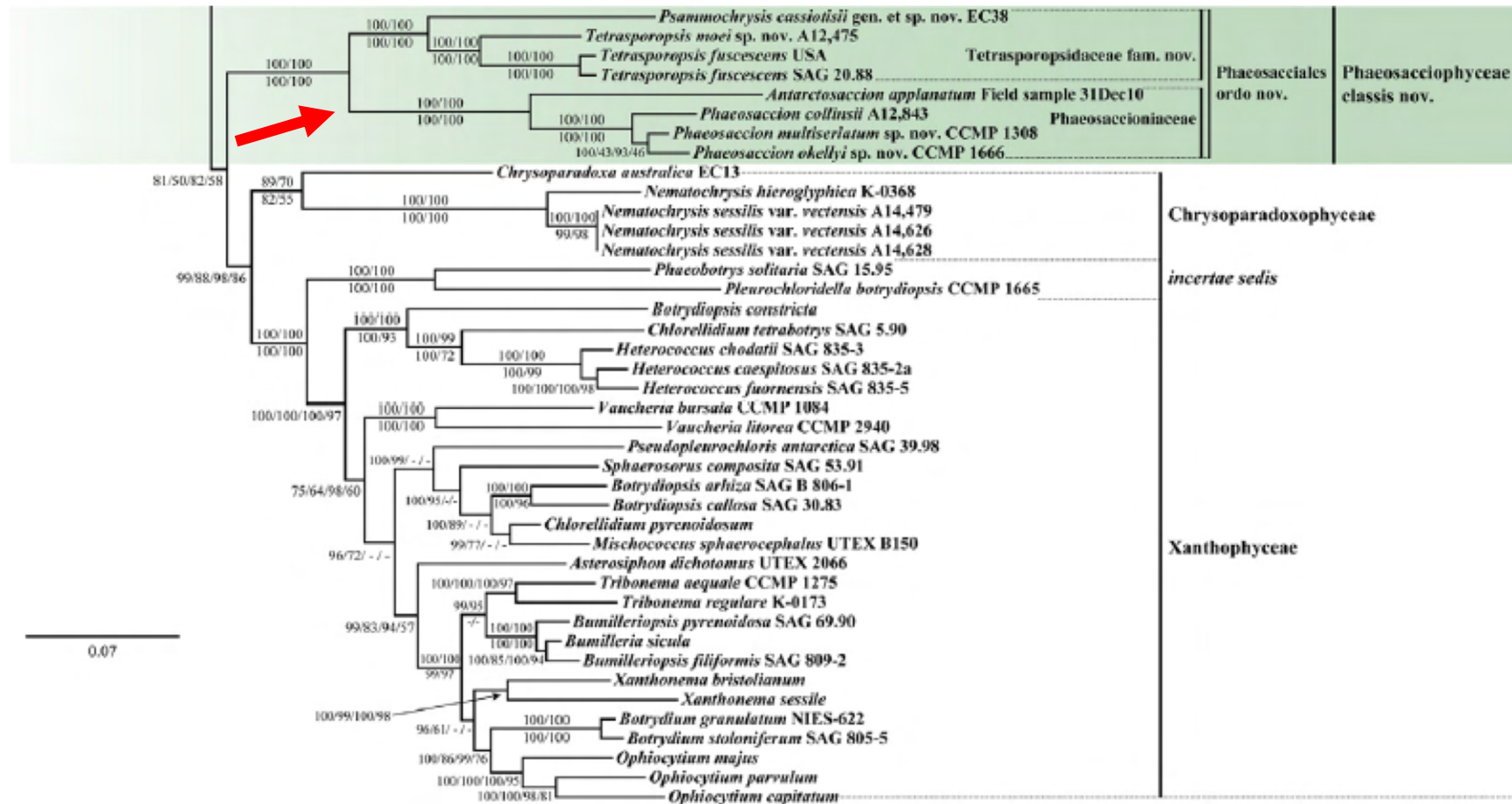


Xanthophyceae - *Mischococcus*

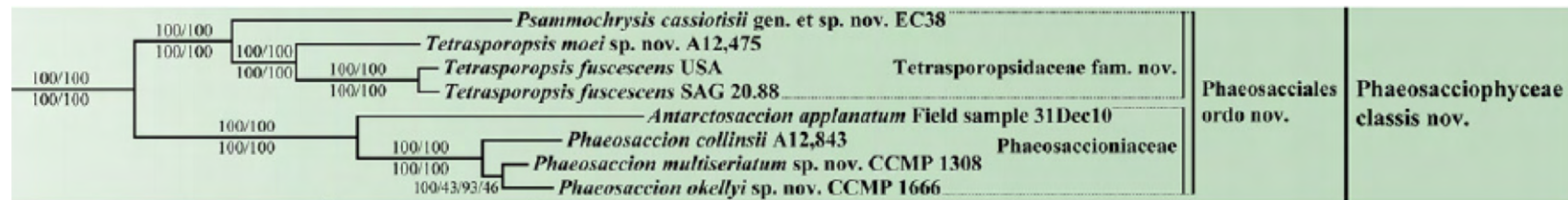
- freshwater (epiphytic or tychoplanktonic)
- colonies on mucilaginous stipes = treelike colonies (2 or 4 cells at the ends of branches)
- one or more plastids
- one-piece cell wall
- autospores, zoospores
- *Botrydiopsis* clade



Phaeosacciophyceae



Phaeosacciophyceae

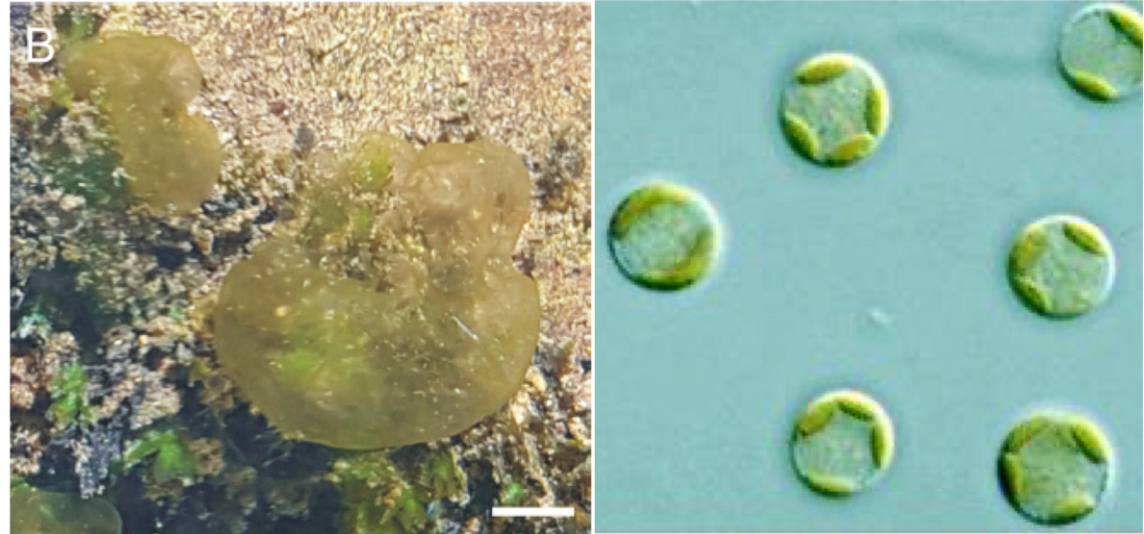


- unicellular, colonial, filamentous (uniseriate or branched multi-seriate) or thallic forms
- cells typically with cell wall
- chloroplasts one to two per cell: three thylakoids per lamella plus a girdle lamella; eyespot
- zoospores heterokont-like
- flagellar apparatus similar to brown algae and xanthophyte algae

Phaeosacciophyceae

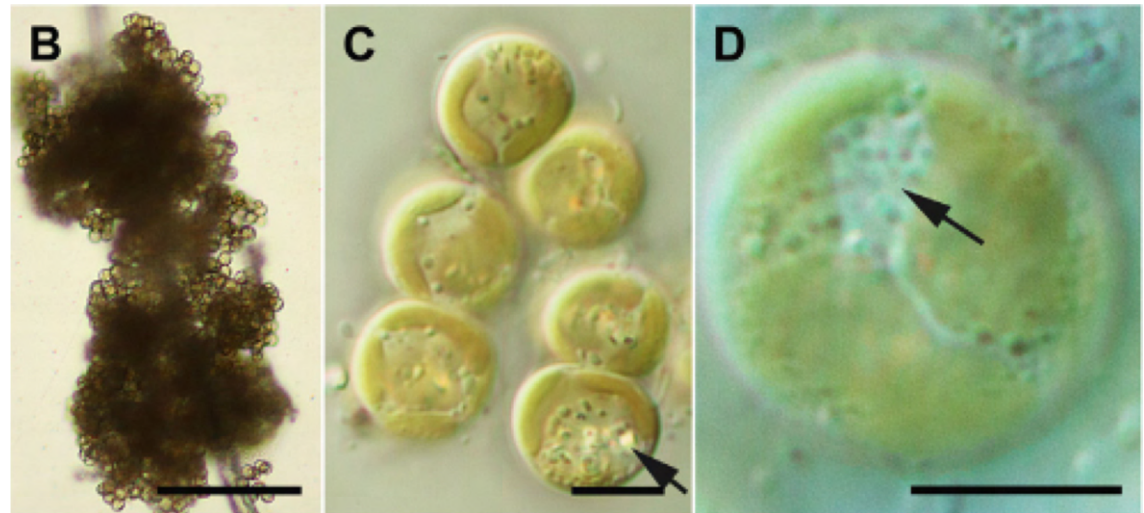
Tetrasporopsis fuscescens

- freshwater environments
- cool and running waters
- colonial in mucilage
- previously within Phaeoph., Chrysoph. and Phaeothamnioph.

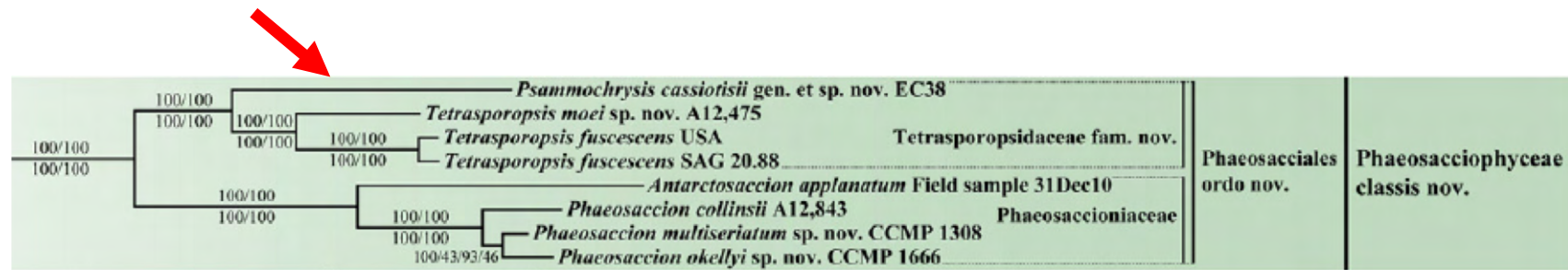


Tetrasporopsis moei

- freshwater (small pool)
- dancing particles located between the chloroplasts
- colonial (irregular cluster of cells)

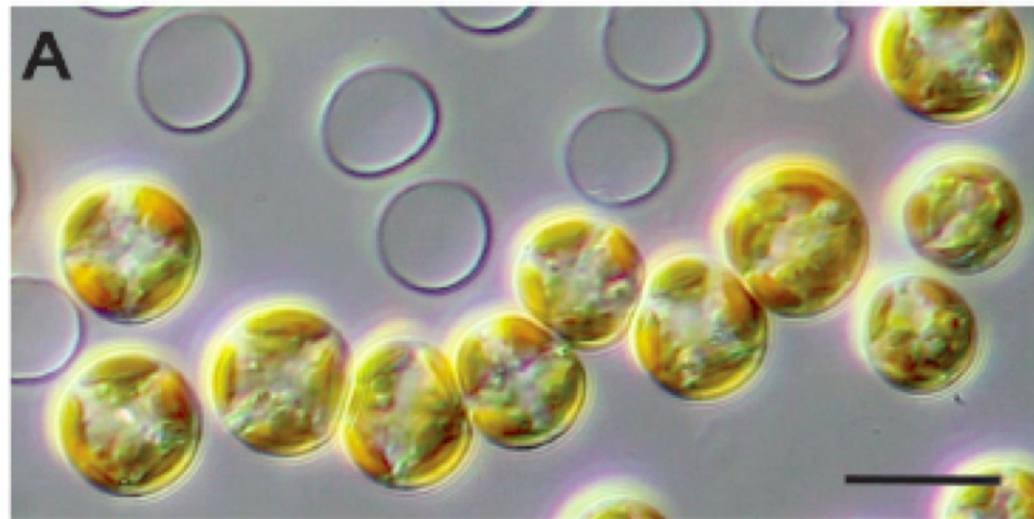


Phaeosacciophyceae



Psammochrysis cassiotissii

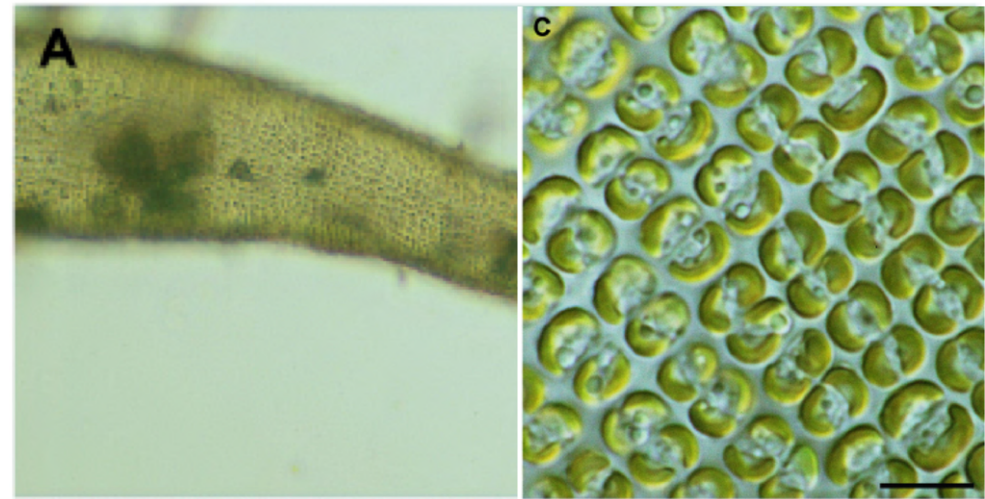
- rounded flattened unicells (as gold coins) adhered to sand grains
- a thick adhesive cell walls
- chloroplasts with 4 lobes
- marine (intertidal pool near Australia)



Phaeosacciophyceae

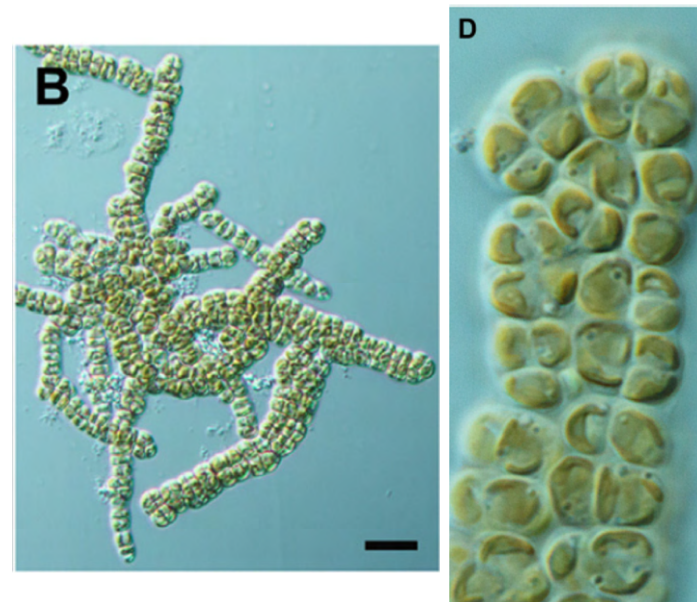
Phaeosaccion collinsii

- macroscopic hollow tubes attached to *Zostera marina*
- block-shaped cells; division in two directions

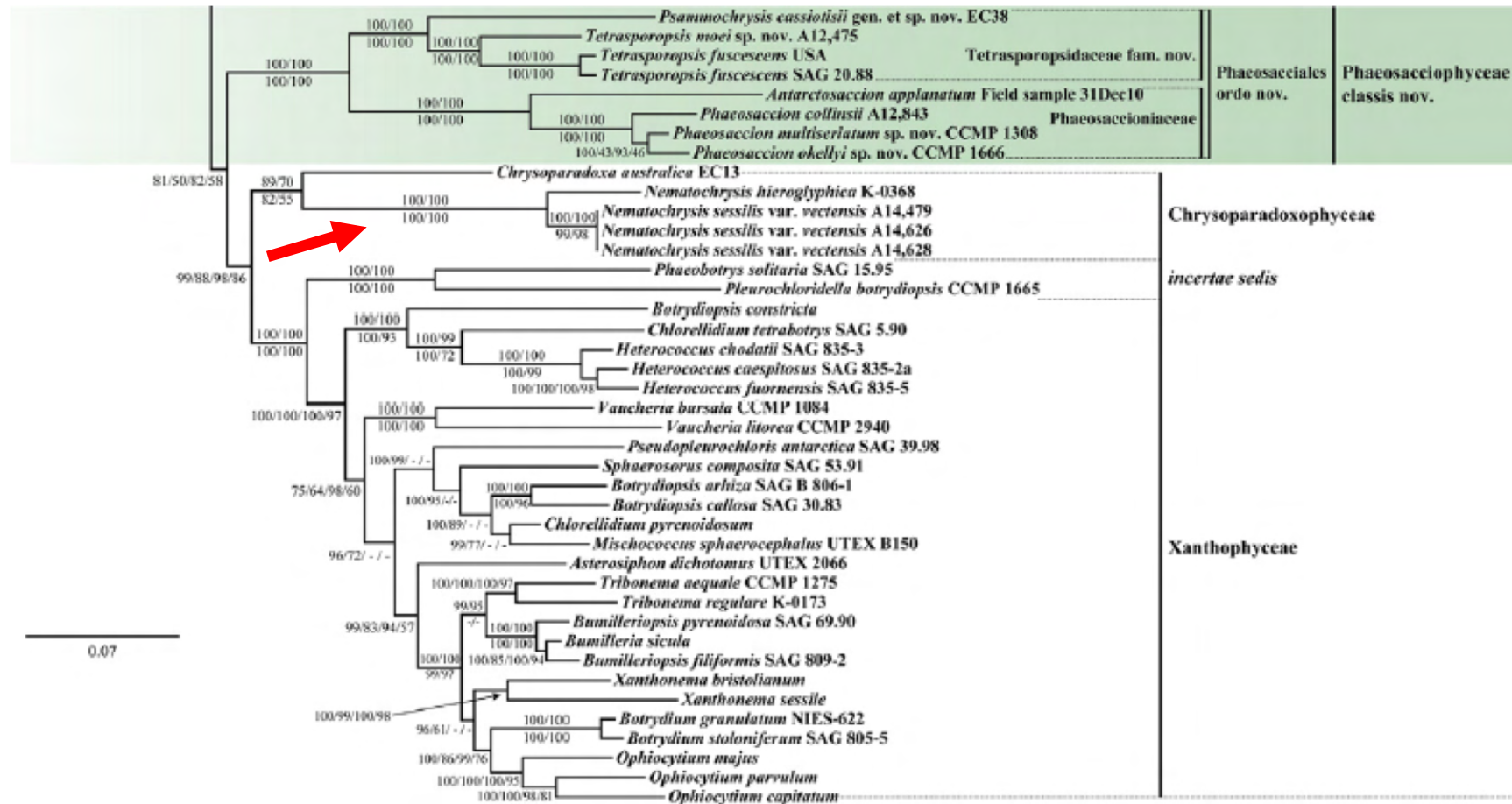


Phaeosaccion multiseriatum

- marine
- uniseriate and multiseriate branched filaments
- block-shaped cells; division not precise (along three axes)



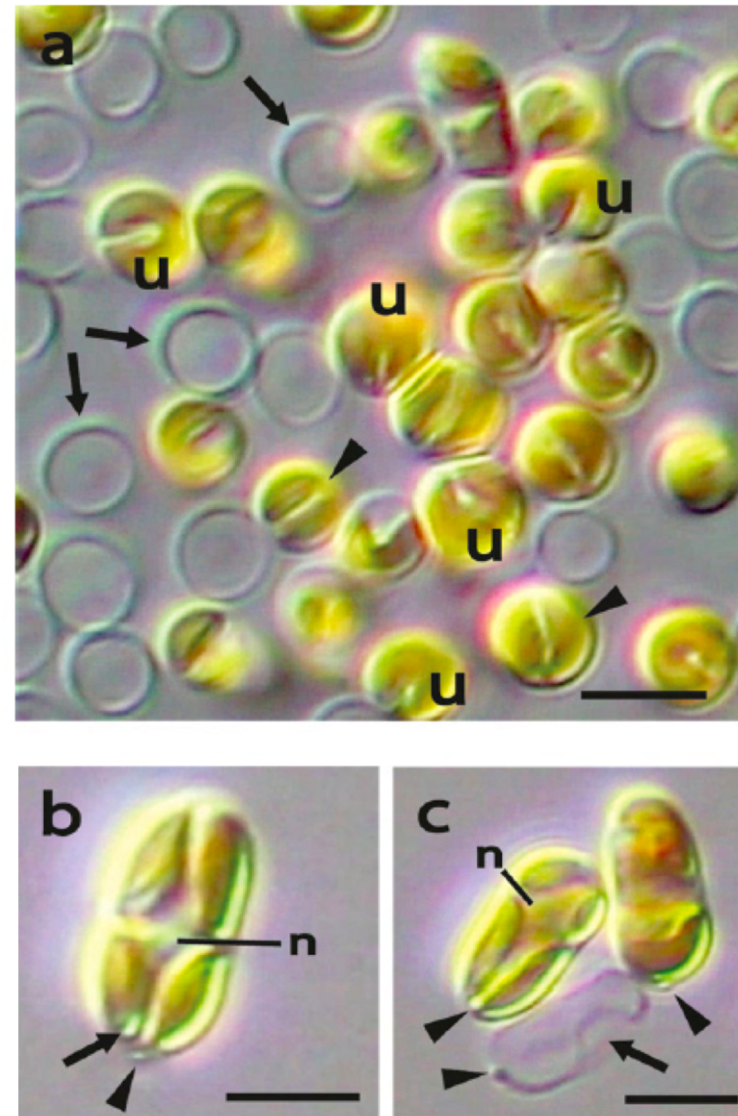
Chrysoparadoxophyceae



Chrysoparadoxophyceae

Chrysoparadoxa australica

- marine, sand dwelling (benthic, attached to the substratum by a complex adhesive plug)
- unicellular, and surrounded by a multilayered cell wall
- etymology: Chryso- = golden; a paradox = chloroplasts are surrounded by only two membranes
- described in 2019, type locality: intertidal pool, Australia

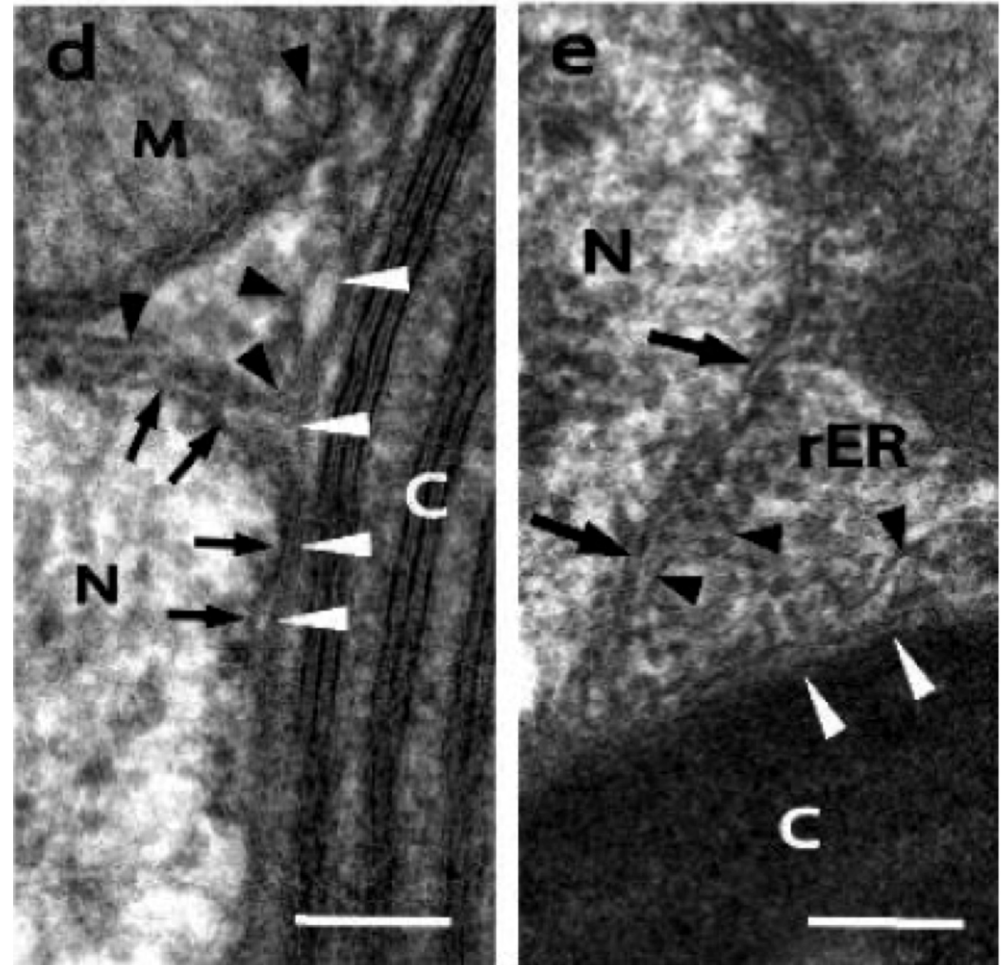


Chrysoparadoxophyceae - *Chrysoparadoxa*

the chloroplast – nucleus interface

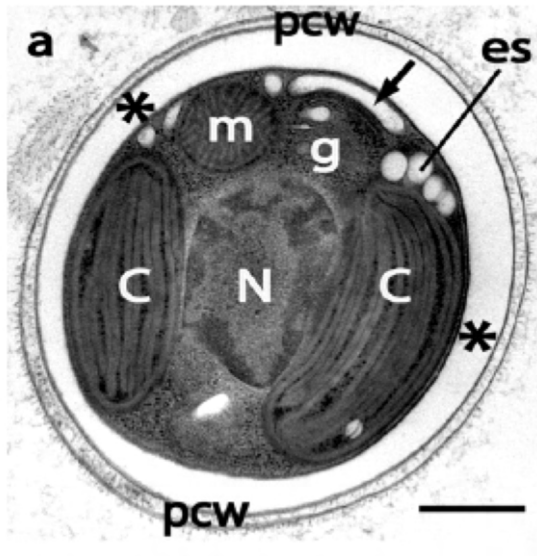
- 2 chloroplasts, surrounded by only two membranes
- chlorophyll *a*, *c1*, *c2*; xanthophylls (including fucoxanthin); presence of pyrenoid, girdle lamella

The outer chloroplast membrane is continuous with the outer membrane of the nuclear envelope (black arrowheads). The inner membrane of the nuclear envelope (arrows) and the inner chloroplast membrane (white arrowheads) separate the two organelles at their interface.

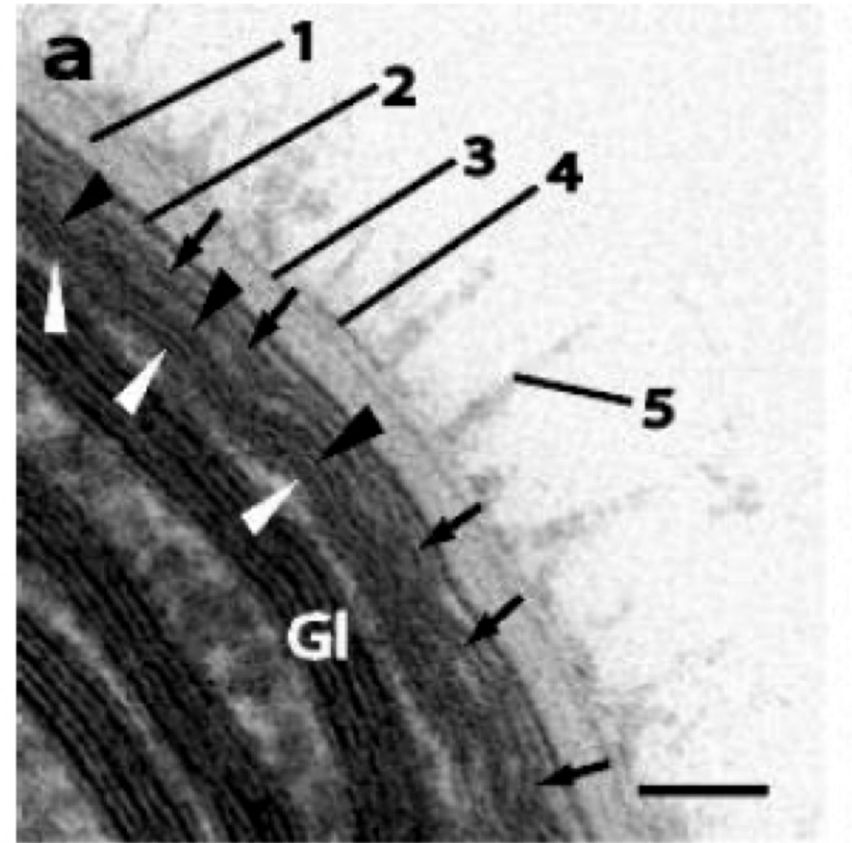


Chrysoparadoxophyceae - *Chrysoparadoxa*

- multilayered cell wall
- primary and secondary cell wall

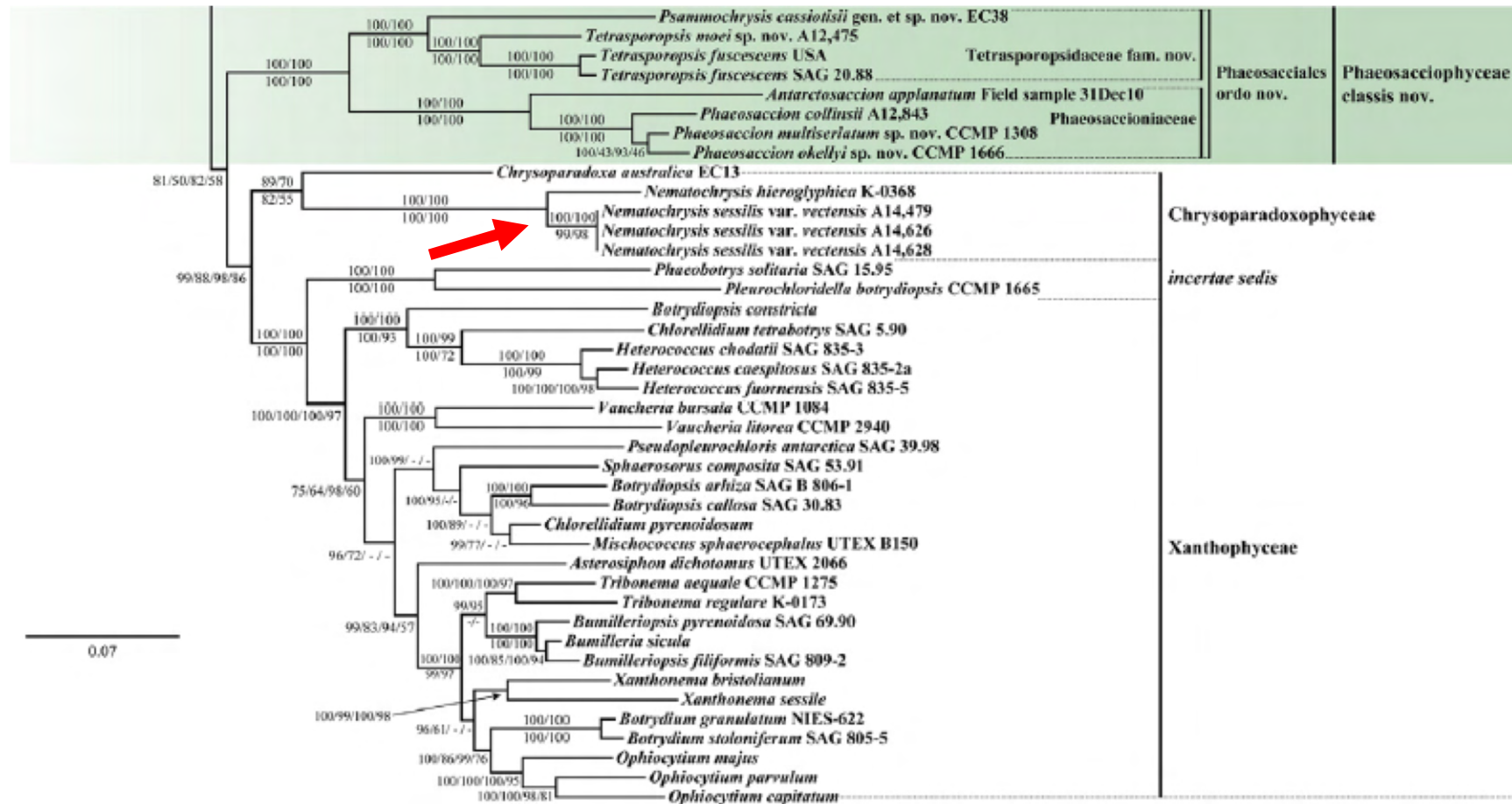


pcw: primary cell wall; * the secondary wall; es: eyespot; m: mitochondria; g: Golgi stack; V: a vesicle; N: nucleus; C: chloroplast



The inner chloroplast membrane (white arrowheads) and outer chloroplast membrane (black arrowheads) lie beneath the PM (arrows). The pcw is multilayered (#1–#5).

¿Chrysoparadoxophyceae?



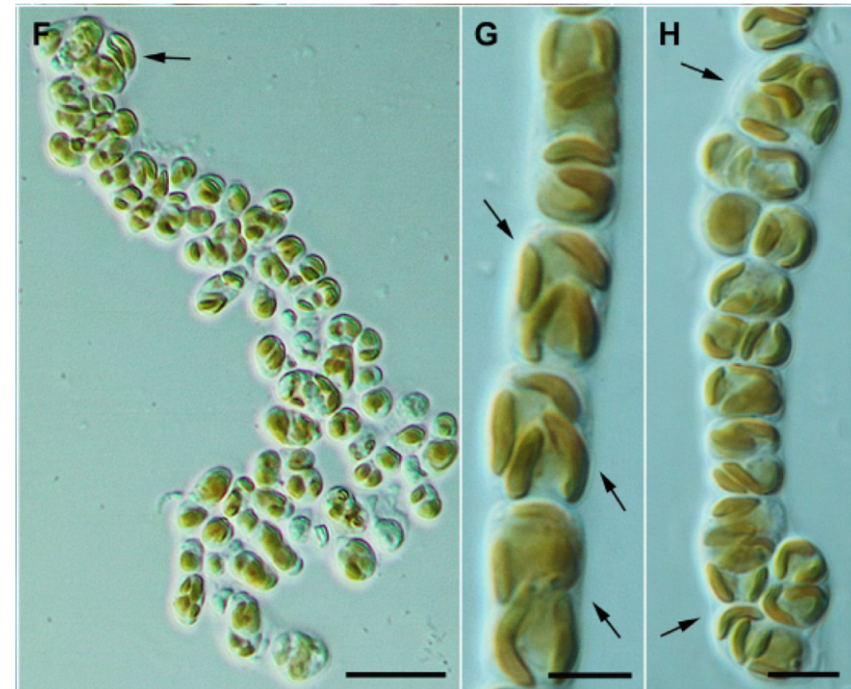
¿Chrysoparadoxophyceae?

Nematochrysis sessilis

- originally described by Pasher (1914)
[found in a Prague tank that was filled with water collected from the Adriatic Sea]
- aquarium tanks and estuaries
[requirement for ammonia except nitrate]

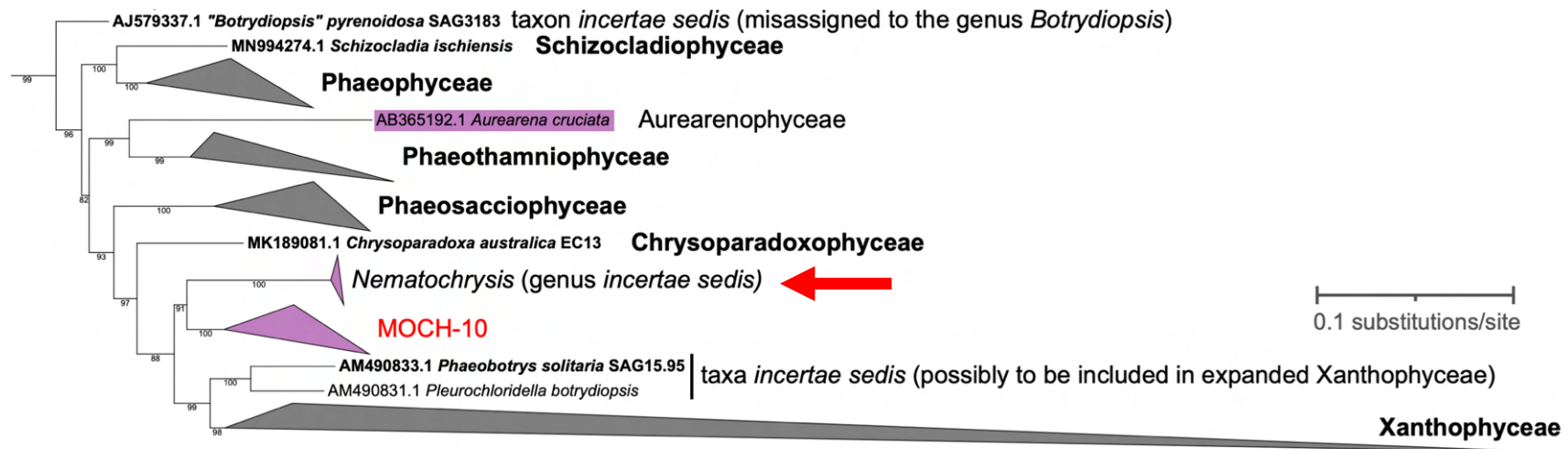
N. (Chrysowaernella) hieroglyphica

- unbranched filaments, uni- to pluri-seriate; surrounded by mucilage
- epiphytic, brackish areas
- previously in Chrysophyta, Sarcinochrysidales - Pelagophyceae, Chrysomerophyceae



N. sessilis var. *vectensis*

¿Chrysoparadoxophyceae?



MOCH - uncultured **M**arine **OCH**rophytes

...future discoveries

