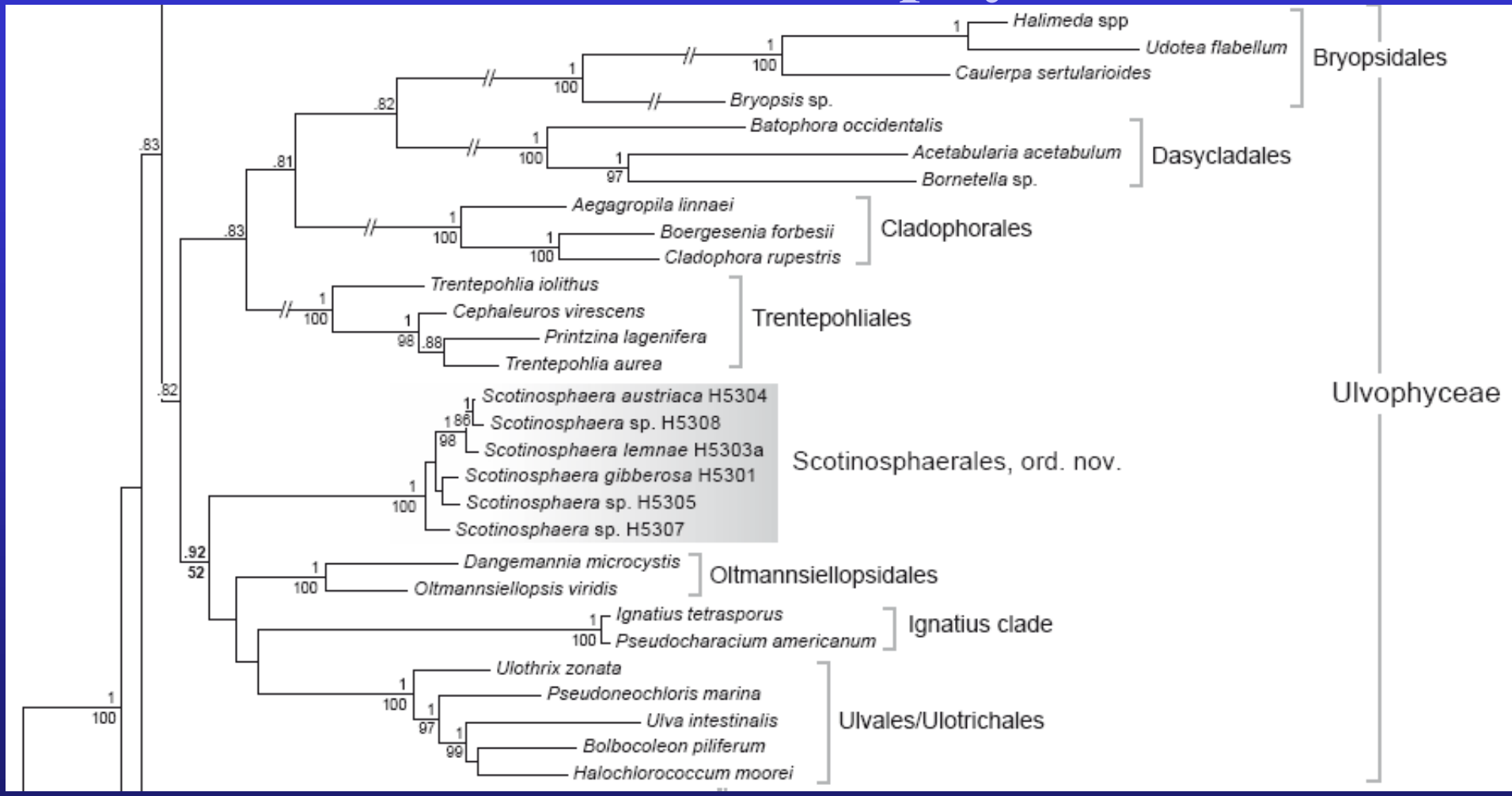
Chlorophyta

Ulvophyceae

Bryopsidales

Dasycladales

Třída: Ulvophyceae



Phylogeny of the Chlorophyta obtained by Bayesian inference of the concatenated SSU-rbcL alignment.

Škaloud et al. 2013

Bryopsidales

Základní charakteristika:

• stélka vždy sifonální (uniaxiální nebo multiaxiální druhy)

• strukturní polysacharidy BS – mannany, xylany, glukany (celulóza není v krystalické formě)

• zoidi 2 nebo 4 bičíky, CCW orientace bičíkových bazí, u některých stefanokontní zoospory

• mitóza uzavřená, perzistující telofázové vřeténko, jádro v telofázi – činkovitý tvar; centrioly mohou, ale nemusí být přítomny

• akcesorické pigmenty sifonein, sifonoxantin

• četné chloroplasty v nástěnné vrstvě cytoplasmy (homoplastidické x heteroplastidické druhy)

• proudění cytoplasmy v podélné ose stélky

• haplontní životní cyklus (haploidní gametofyt, mikroskopická zygota s obrovským diploidním jádrem), anizogamie

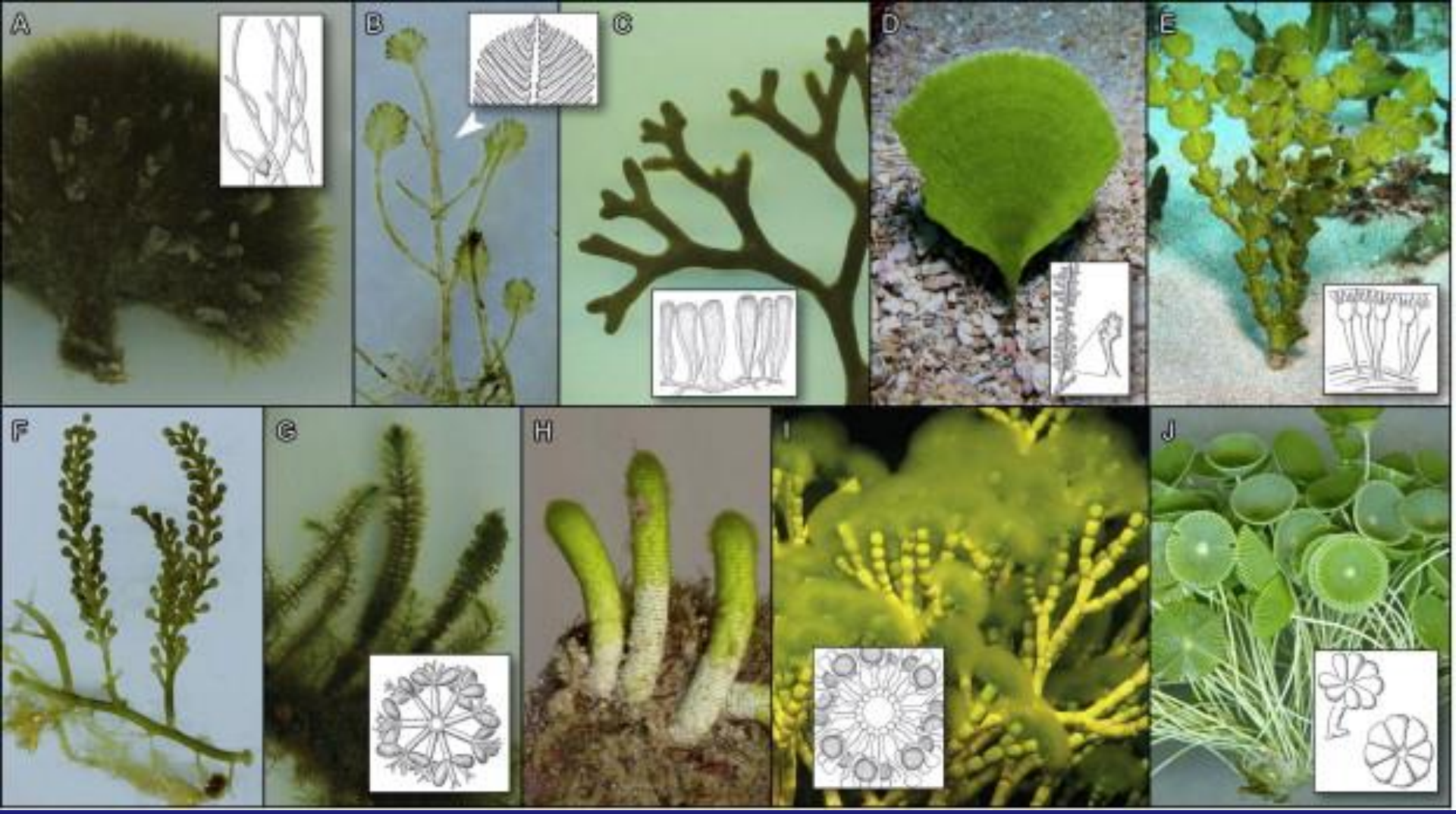
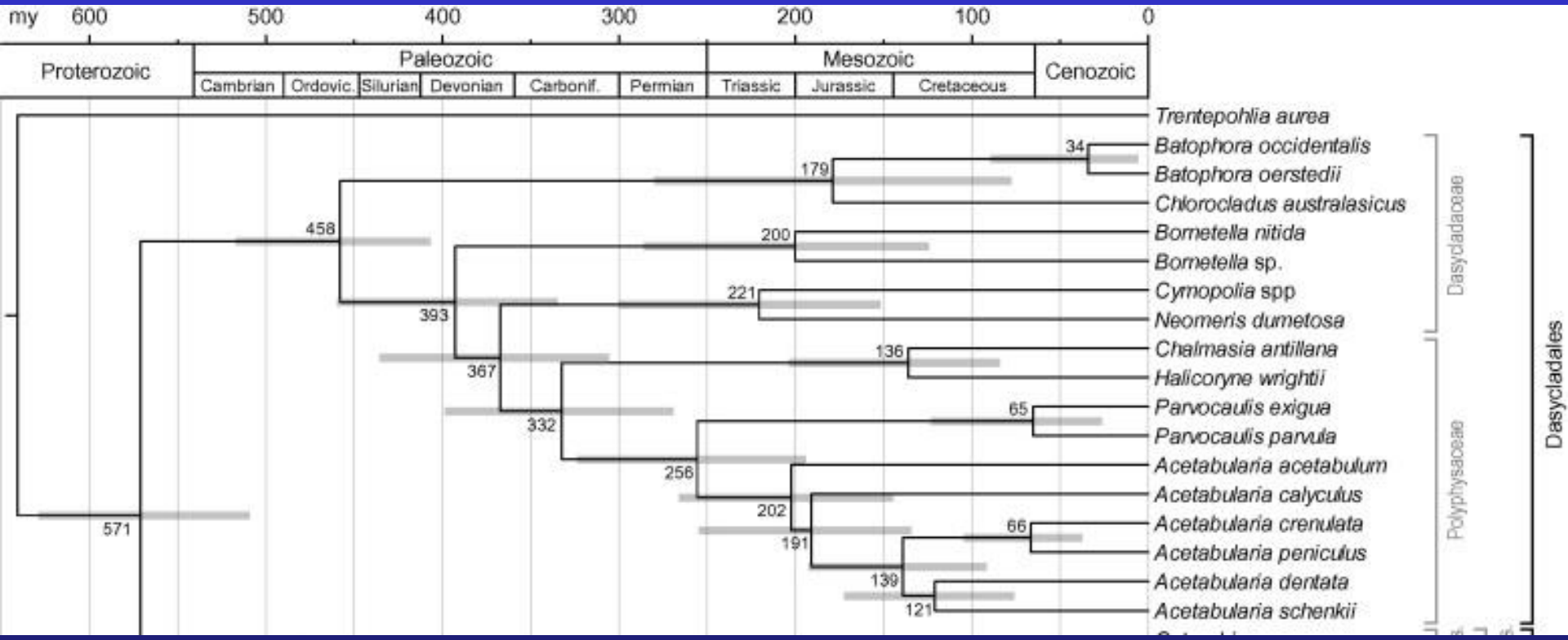
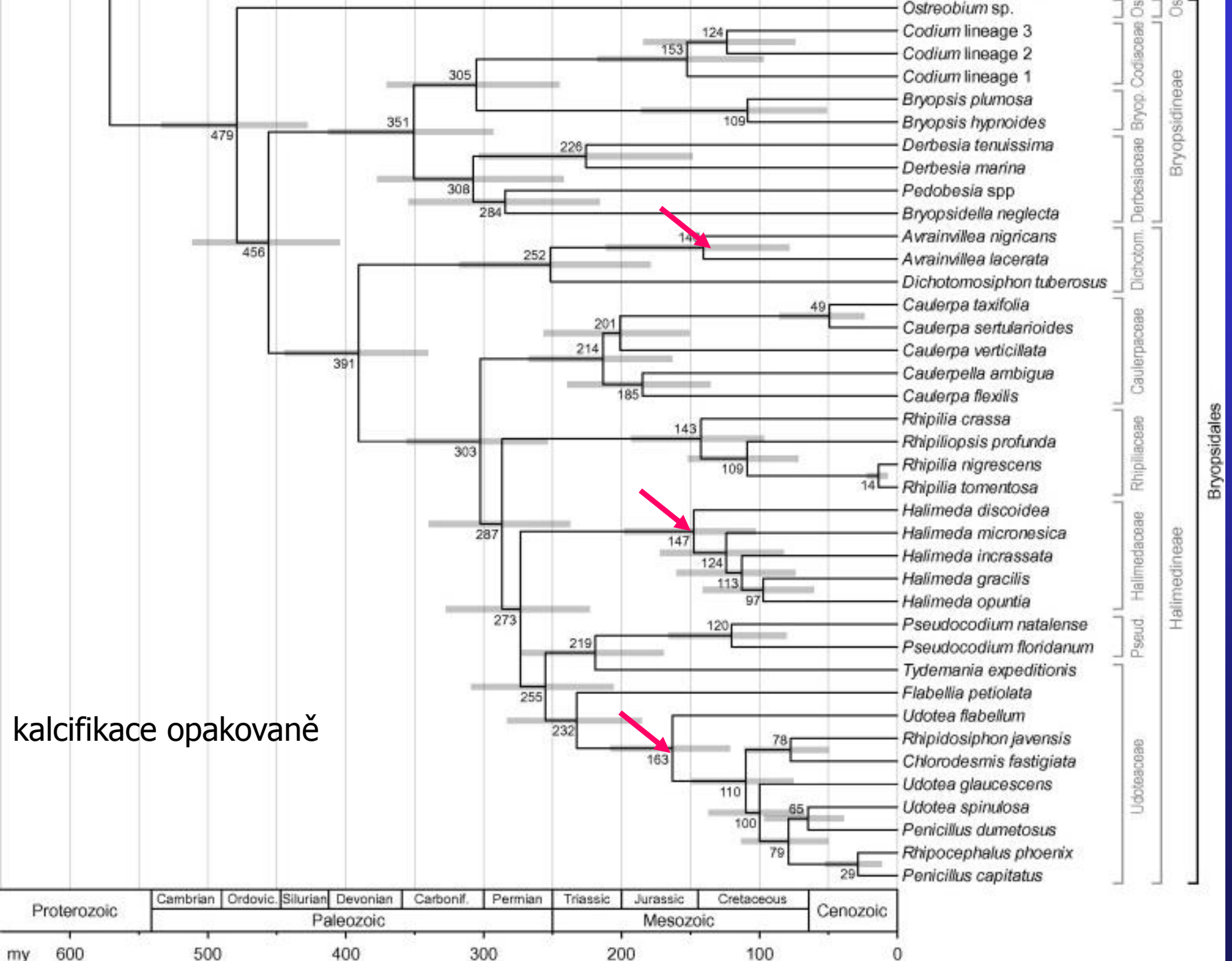


Fig. 1. Morphology and anatomy of the siphonous green algae comprising the orders Bryopsidales (A–F) and Dasycladales (G–J). (A) *Derbesia*, (B) *Bryopsis*, (C) *Codium*, (D) *Udotea*, (E) *Halimeda*, (F) *Caulerpa*, (G) *Batophora*, (H) *Neomeris*, (I) *Cymopolia*, (J) *Acetabularia*.

Verbruggen et al. 2009



Verbruggen et al. 2009

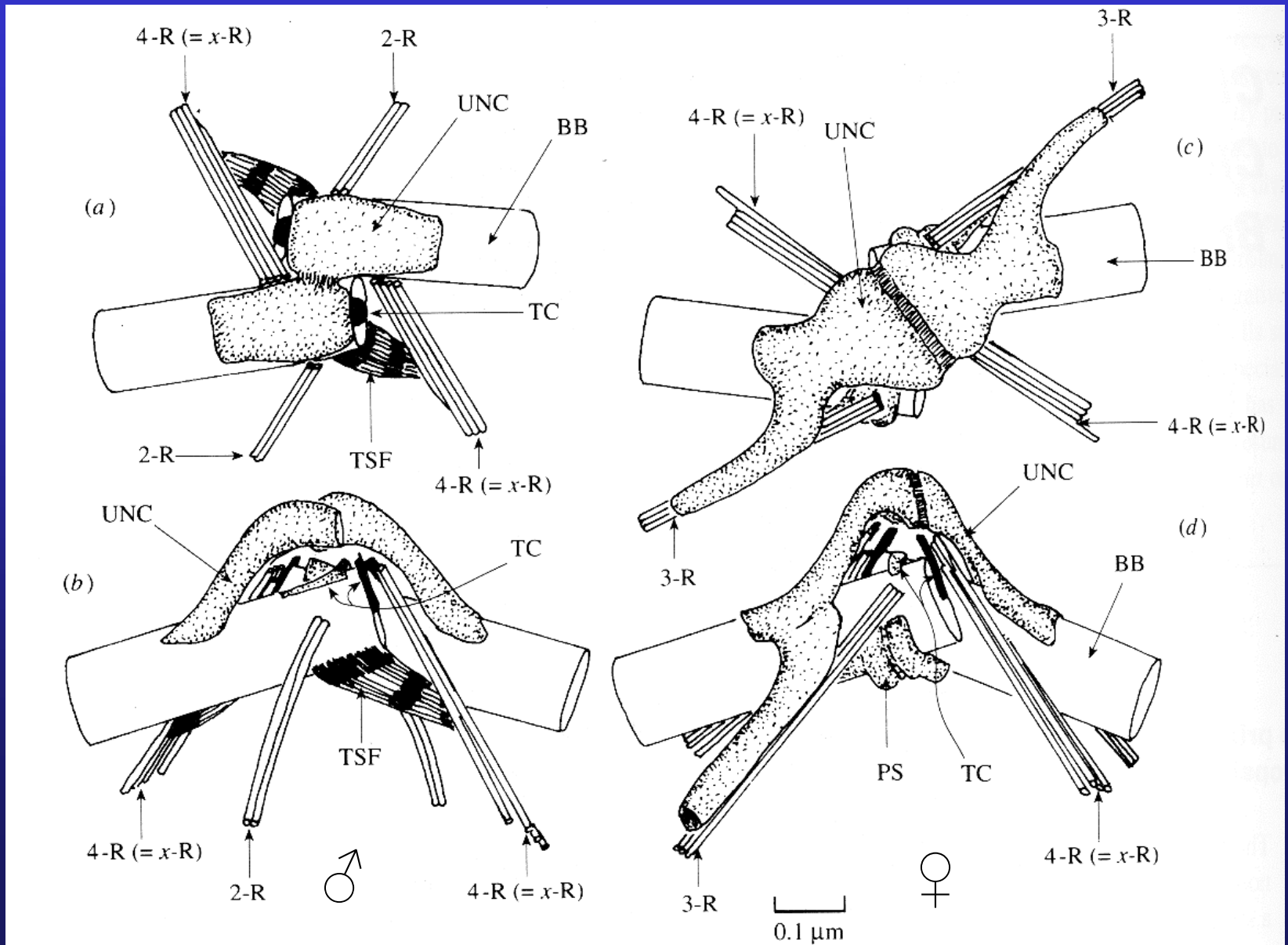


kalcifikace opakovaně

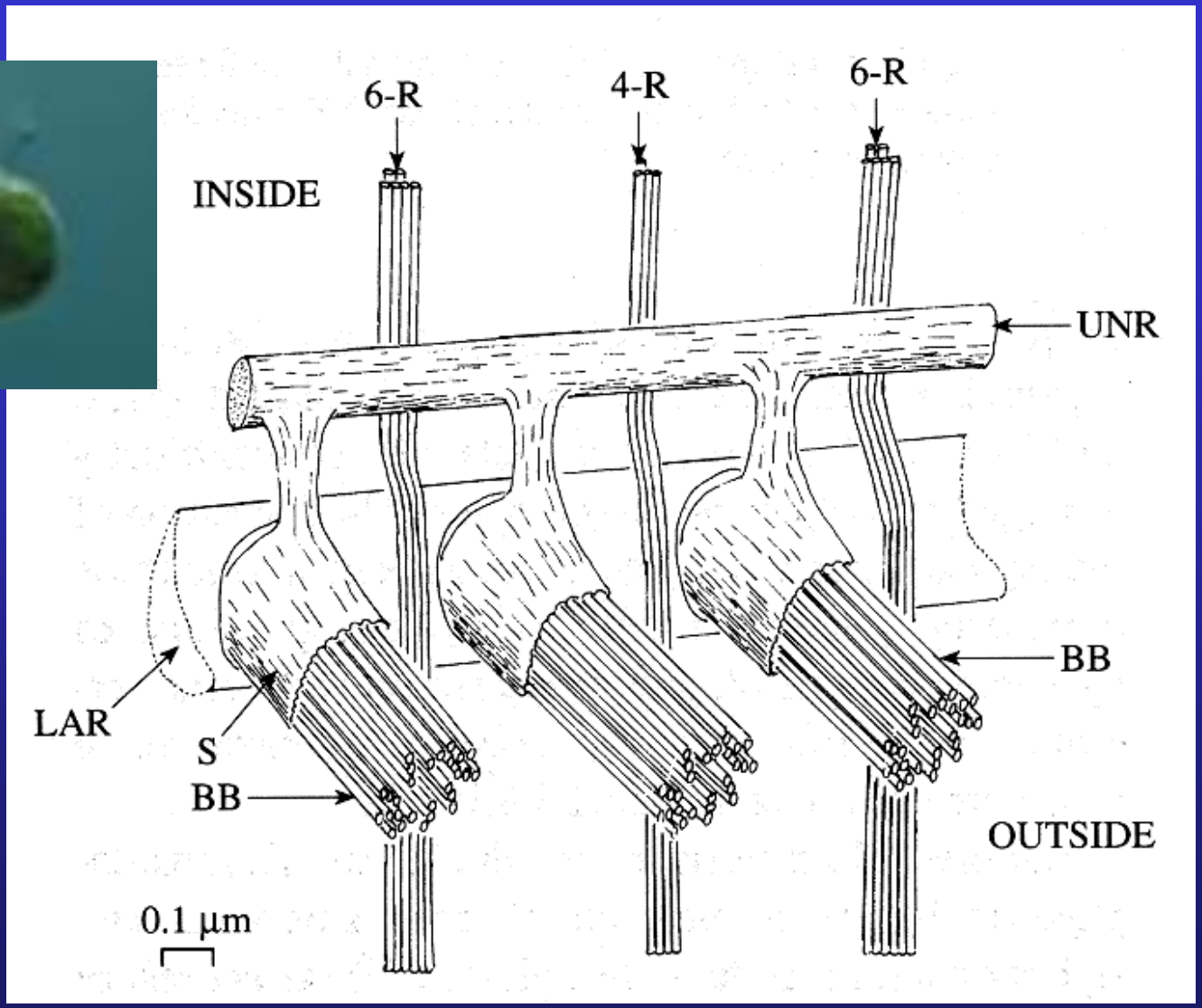
Pseudobryopsis

Flagellar apparatus

Ultrastruktura bičíkového aparátu se u ♀ a ♂gamet *Pseudobryopsis*



Flagellar apparatus of the stephanokont zoospore

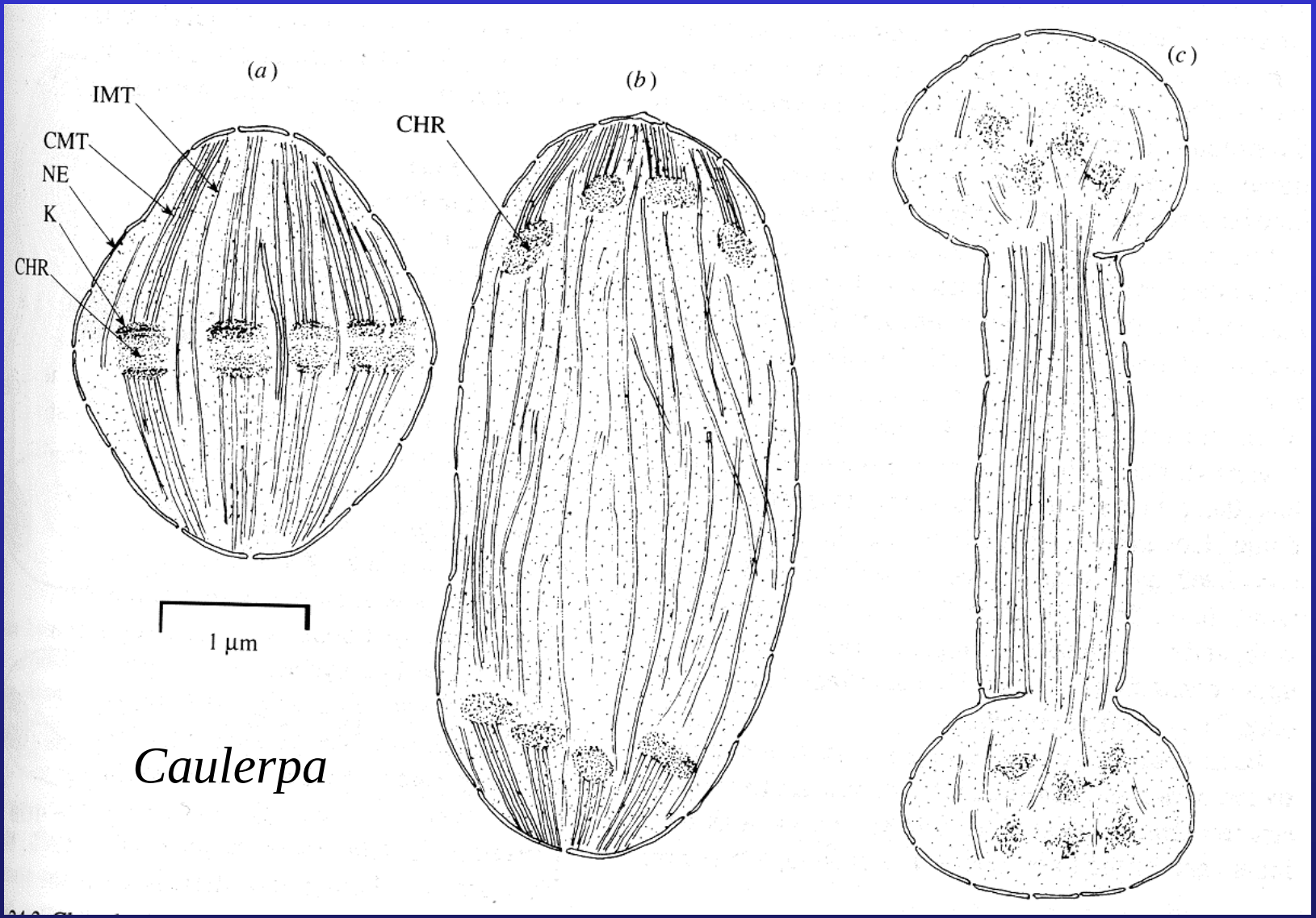


upper non-striated ring

lower amorphous ring

Pseudobryopsis – each stephanokont cell possesses 35 basal bodies

Mitosis



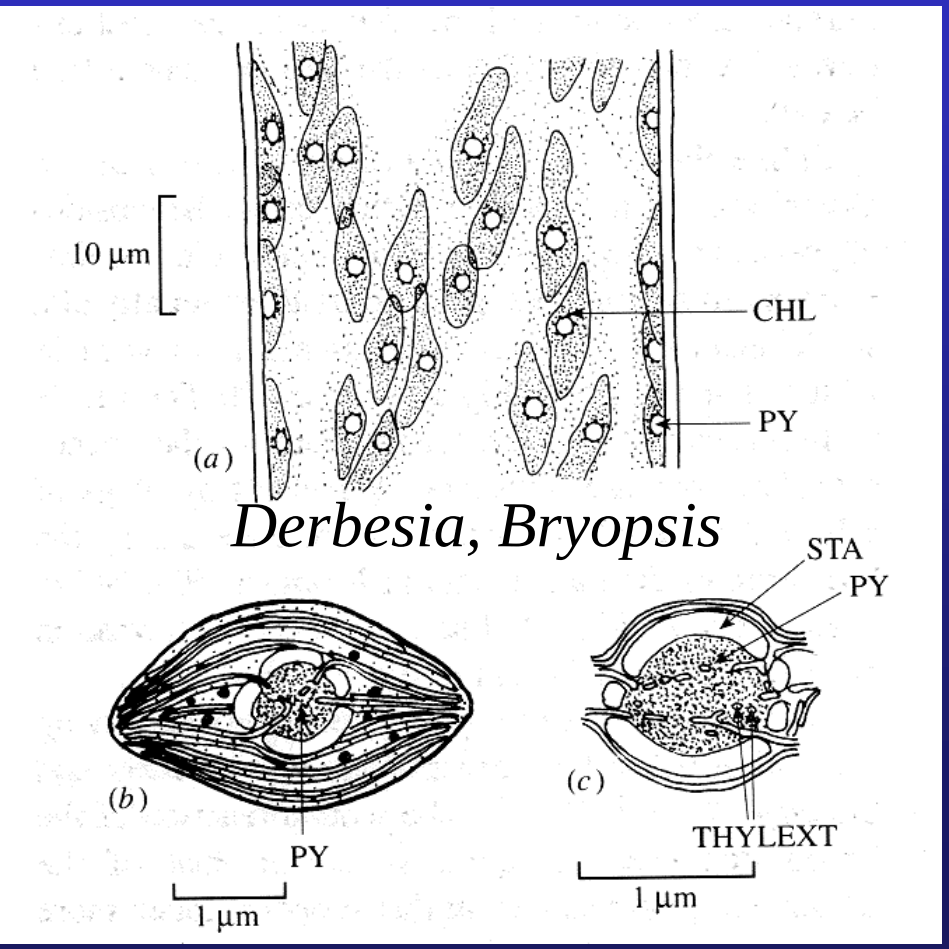
Caulerpa

v telofázi má jádro činkovitý tvar (dumbbell-shaped giant nucleus)

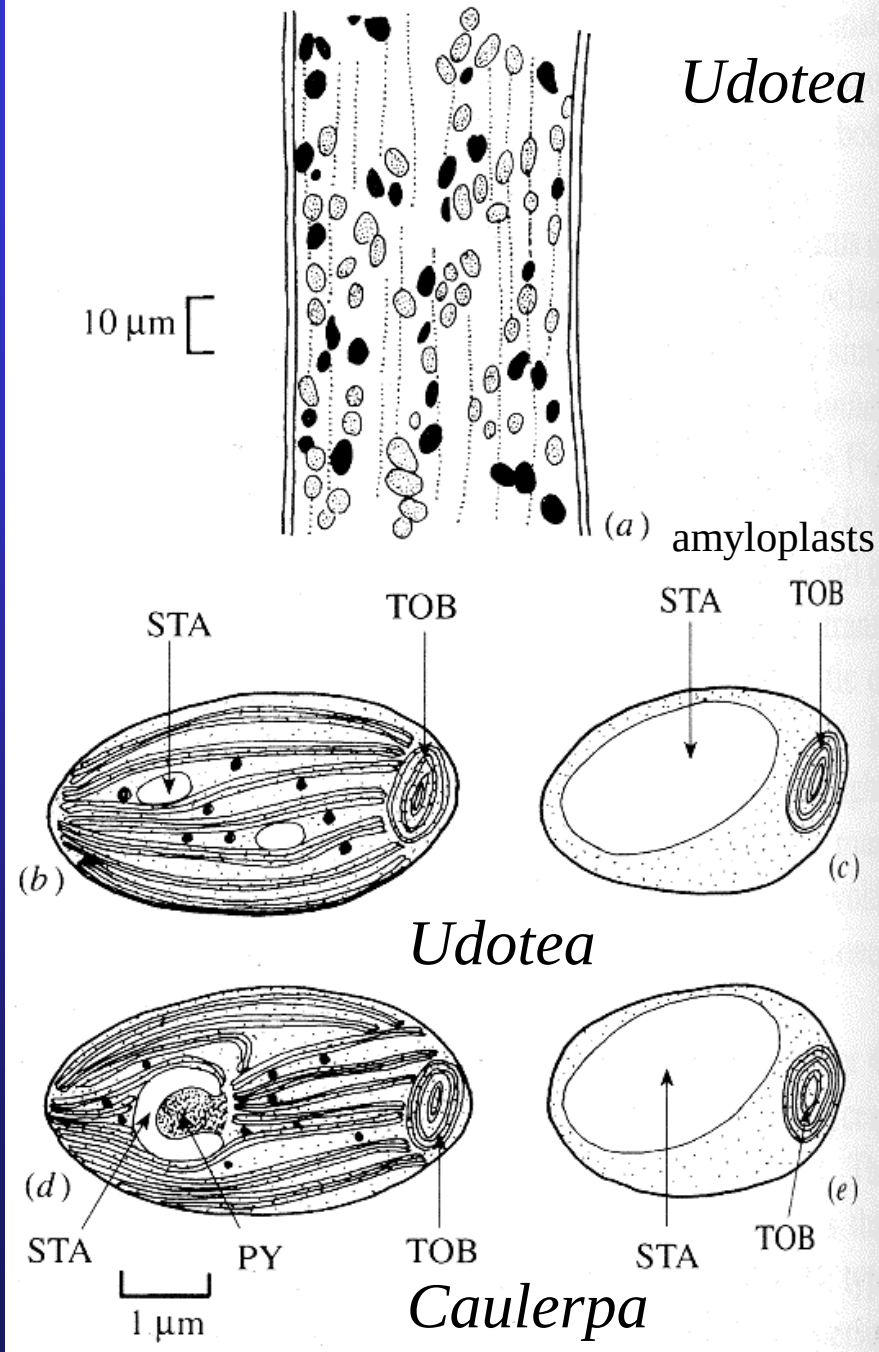
Chloroplasts

heteroplastidic species

homoplastidic species



Derbesia, Bryopsis



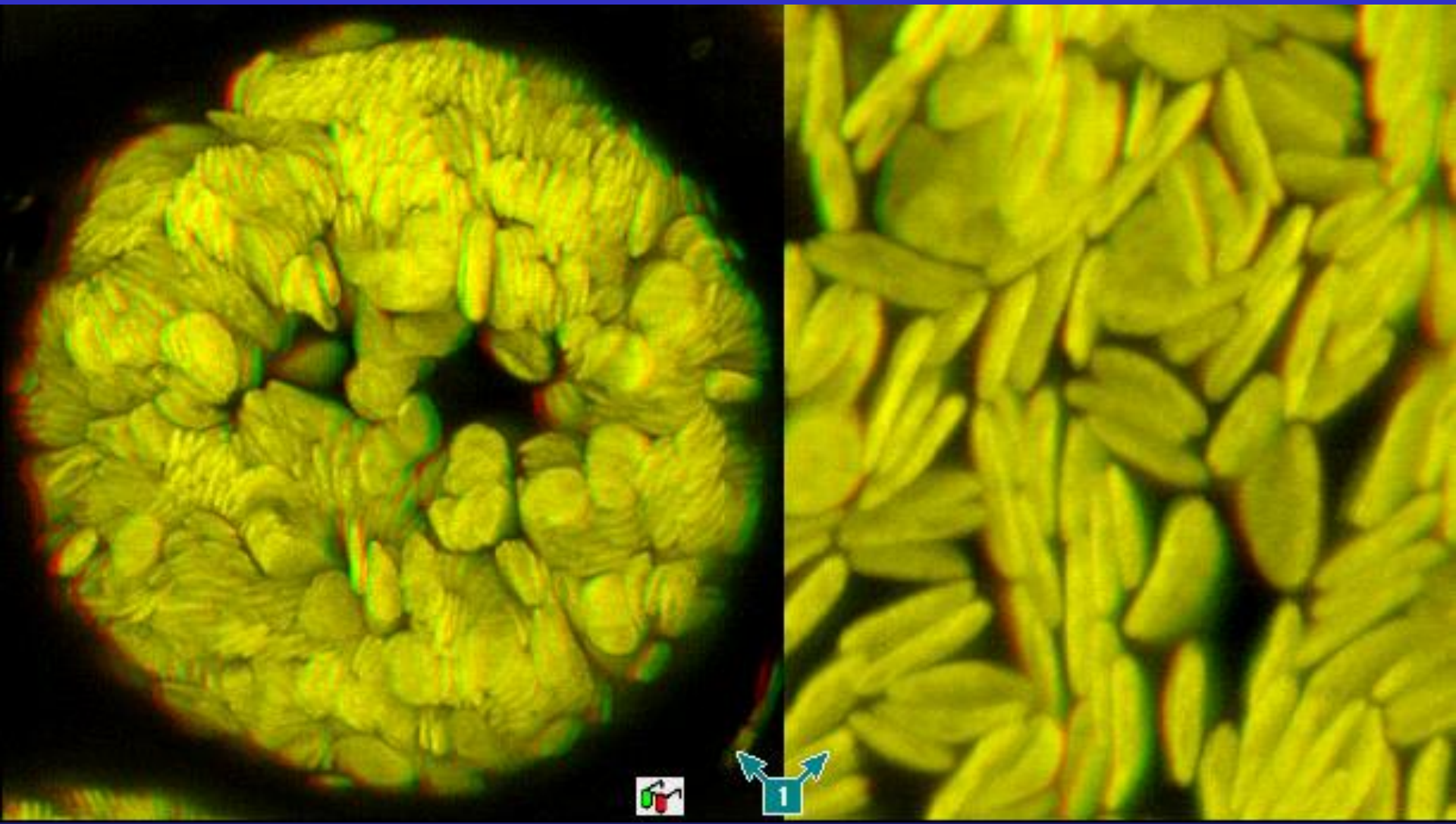
Udotea

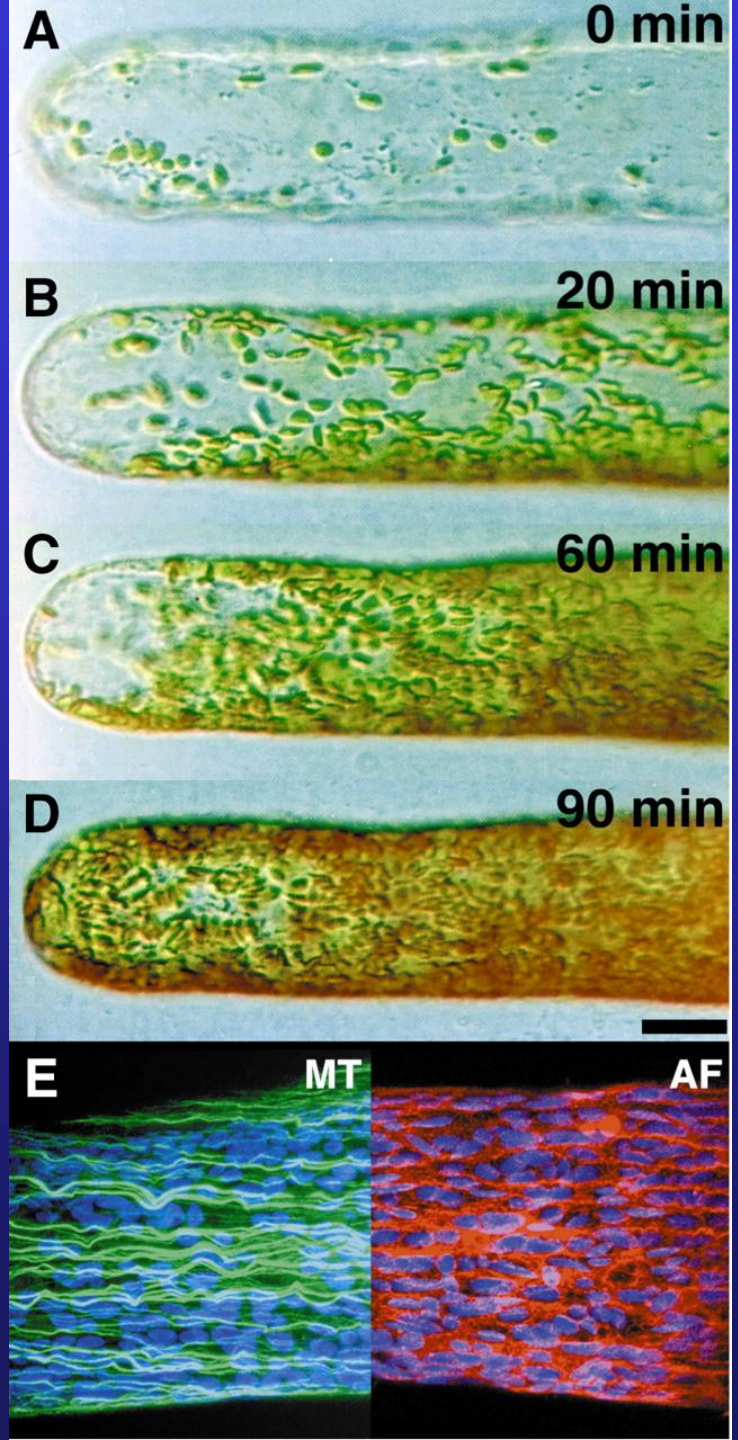
amyloplasts

Udotea

Caulerpa

Chloroplasts of *Codium*
homoplastidic species, chloroplasts without pyrenoid





60 μm/sec

Accumulation of chloroplasts in the cell apex of the coenocytic green alga

Dichotomosiphon

MT – microtubules
AF – actin filaments

Siphonous (coenocytic) thallus

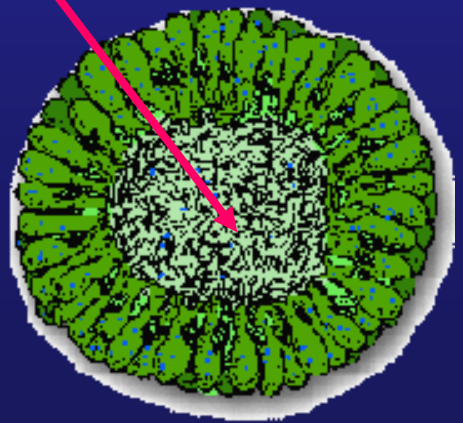
uniaxial

Bryopsis

multiaxial organisation

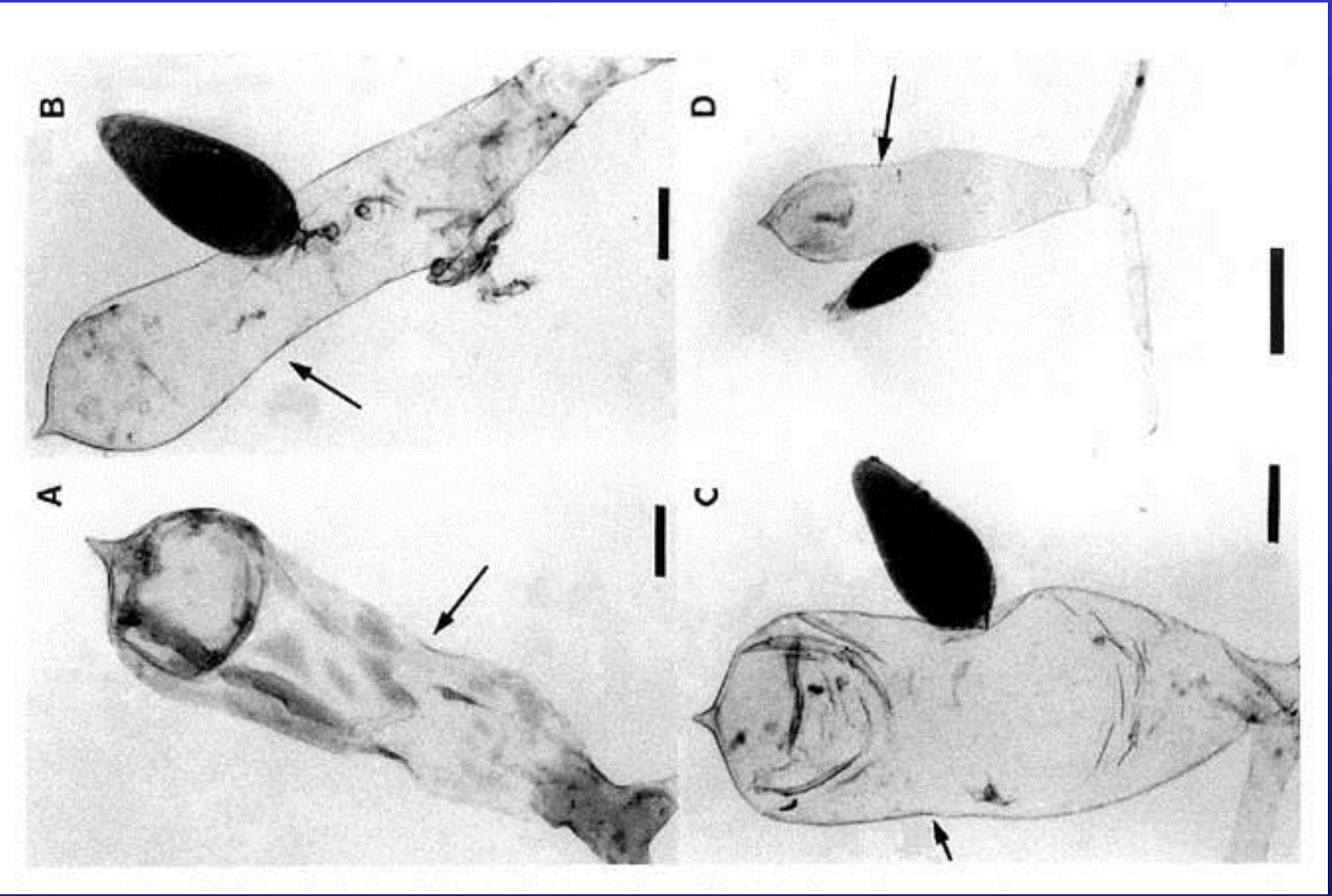
Codium

medulla



utriculus

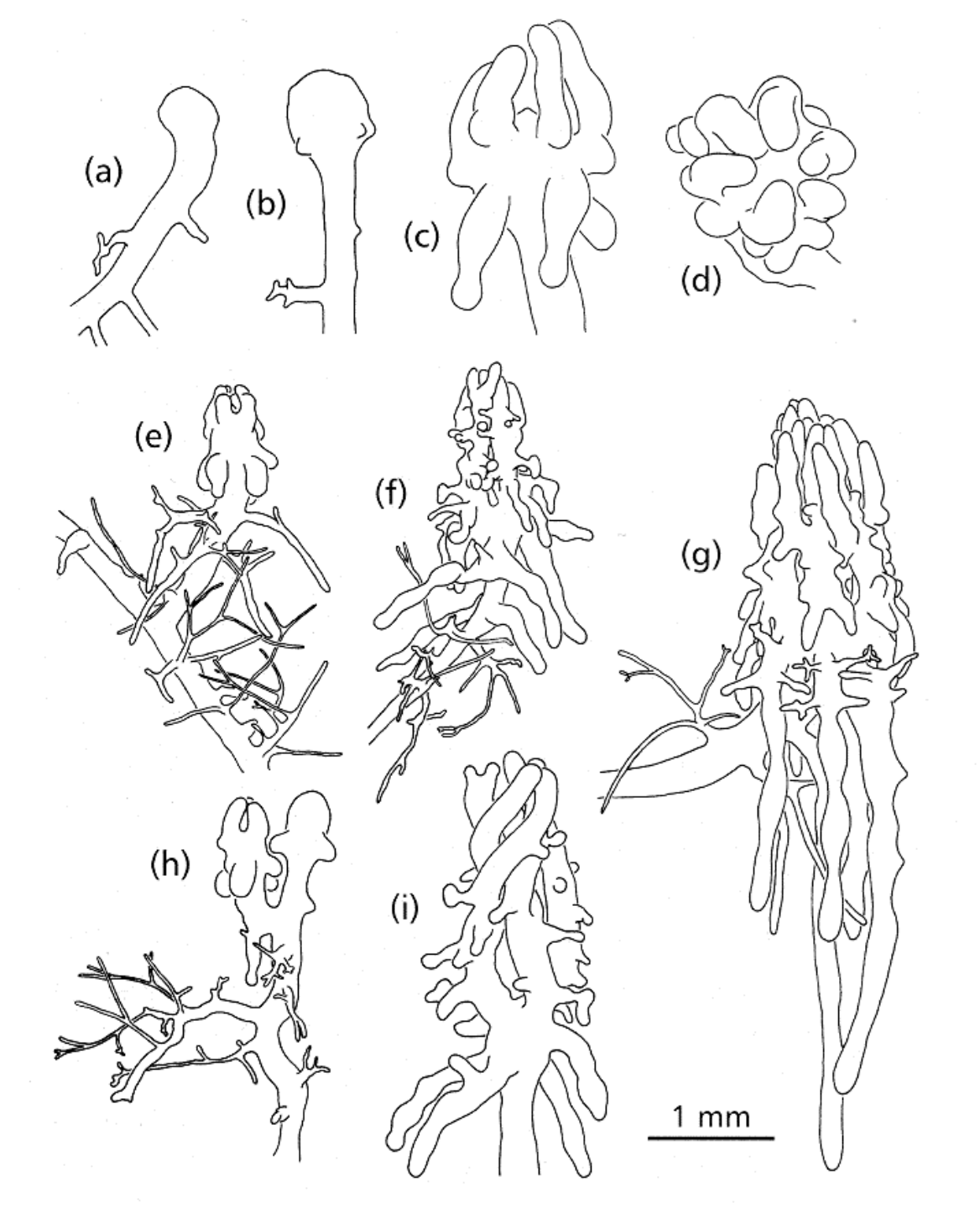
Siphonous (coenocytic) thallus



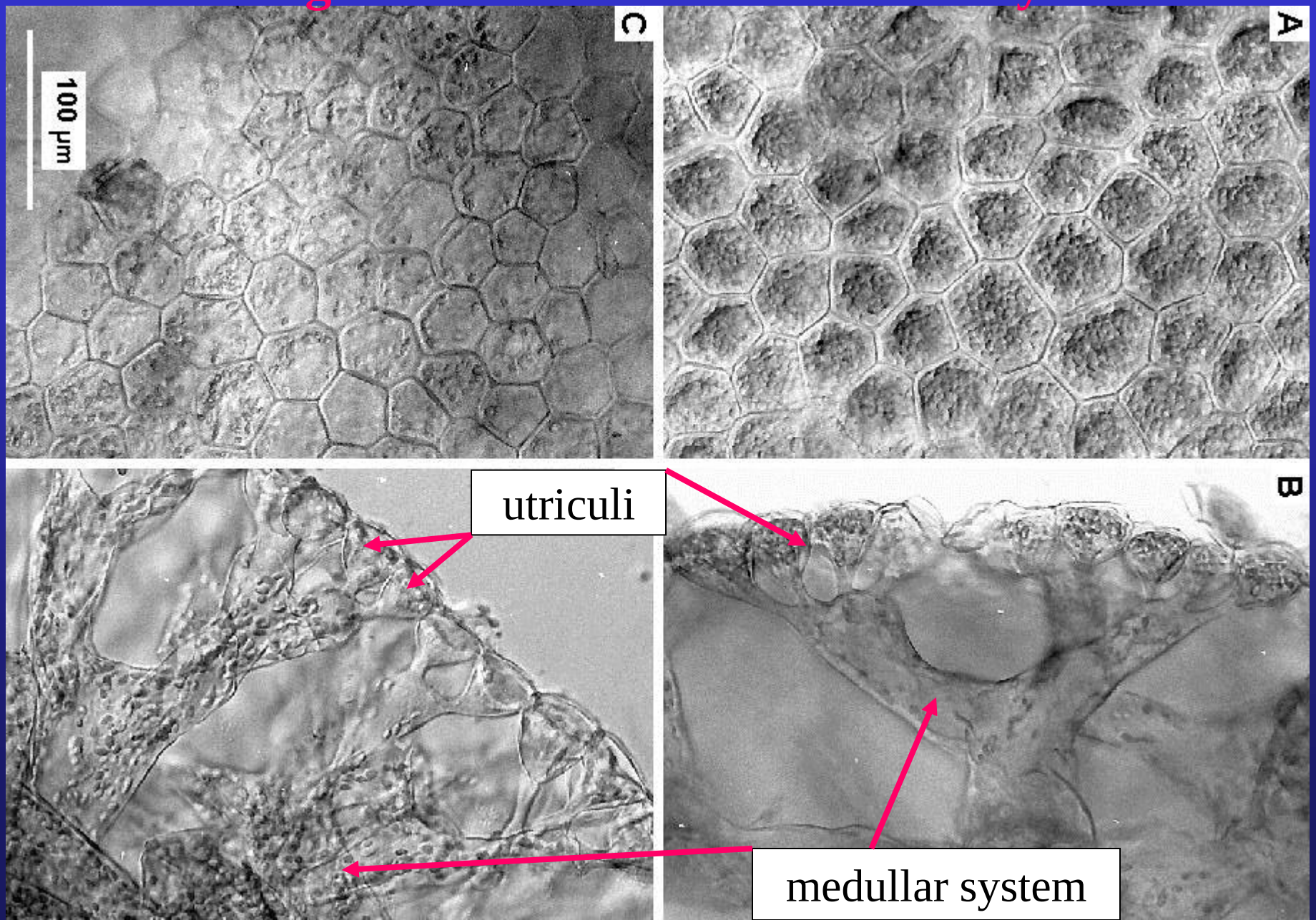
Codium – utriculus bearing gametangium

Siphonous (coenocytic) thallus

multiaxial thallus formation in *Penicillus*



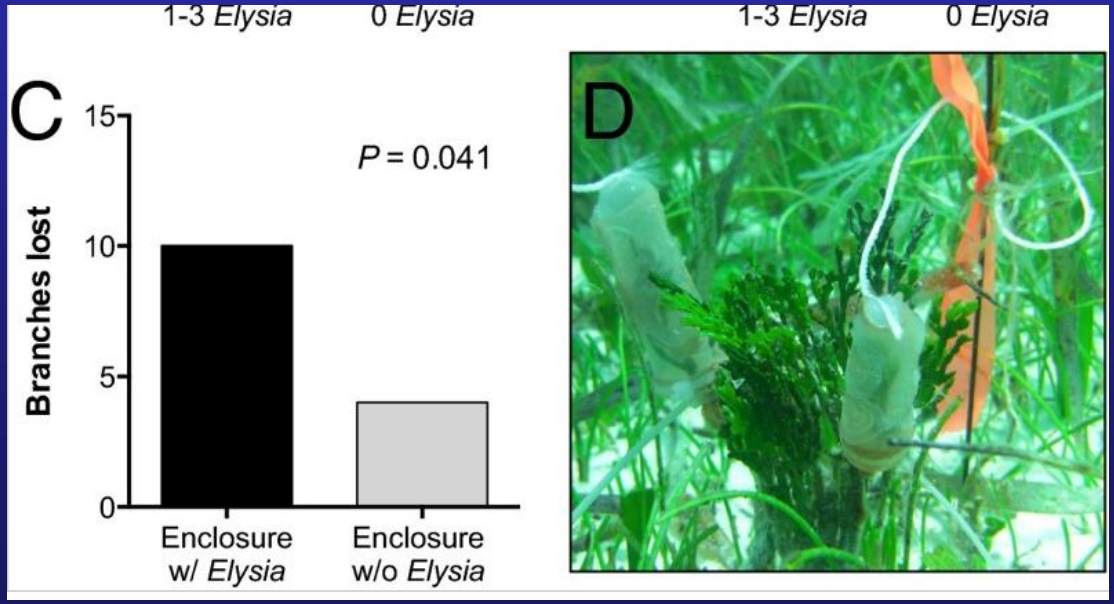
Distribution of chloroplasts in *Halimeda* during the day and night



Elysia pusilla

Elysia pusilla feeds on the calcified green alga *Halimeda* and incorporates unctioning chloroplasts into its body, thus it is known as a solar-powered sea slug. It is found in shallow water in tropical regions of the Indo-Pacific wherever its host species grows.

Halimeda responds by dropping branches occupied by Elysia, apparently to prevent fungal infection associated with Elysia feeding. Elysia is remarkably similar to some terrestrial insects, not only in its hunting strategy, but also its feeding method, defense tactics, and effects on prey behavior and performance



Rasher et al. 2015

Calcification

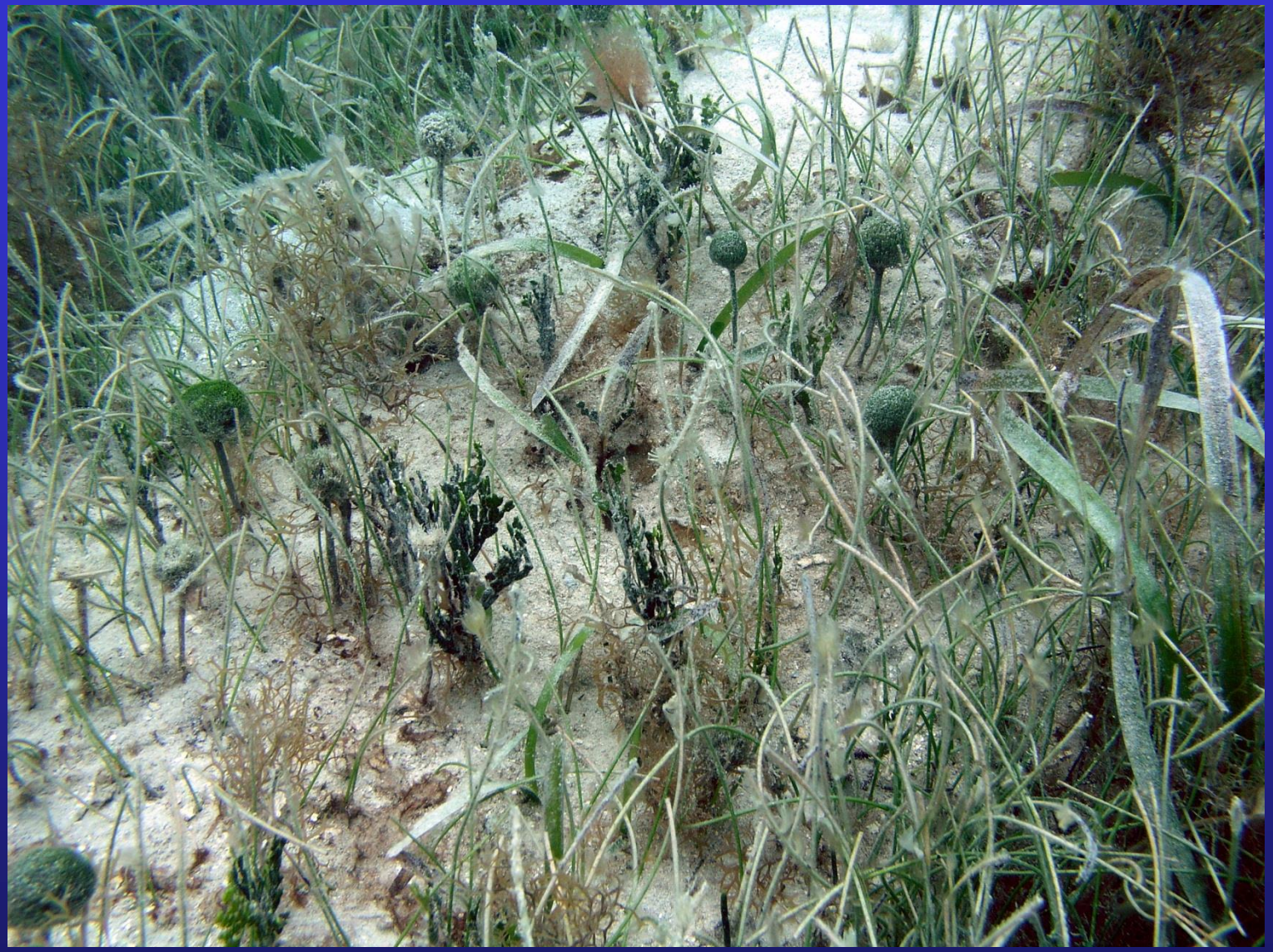
Některé multiaxiální druhy sráží na povrchu sifonů uhličitan vápenatý ve formě krystalků aragonitu

$$Ca^{2+} + 2HCO_3^- \rightarrow CaCO_3 + H_2O + CO_2$$

bicarbonate — calcium carbonate — carbon dioxide

Adhesion to substrate

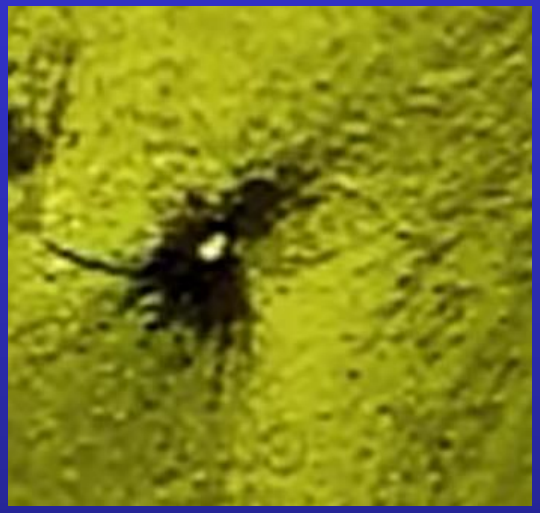
bulbouse base - bulbous mass of rhizoids adhering sand, anchor the adult plant in the sediment



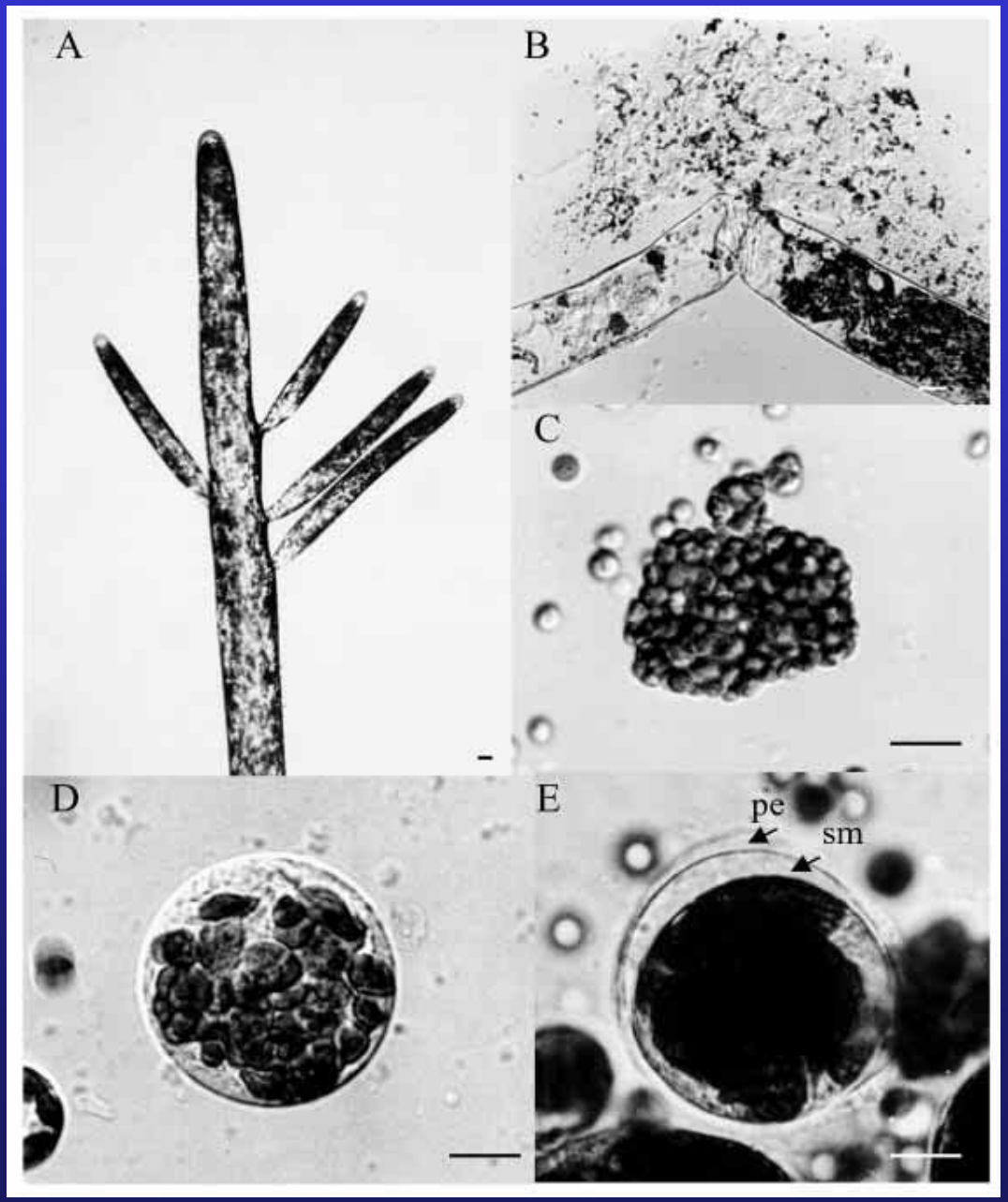
shallows seagrass meadows (*Penicillus*, *Halimeda*)

predation - perforation of CW, effective reparative mechanisms

Derbesia

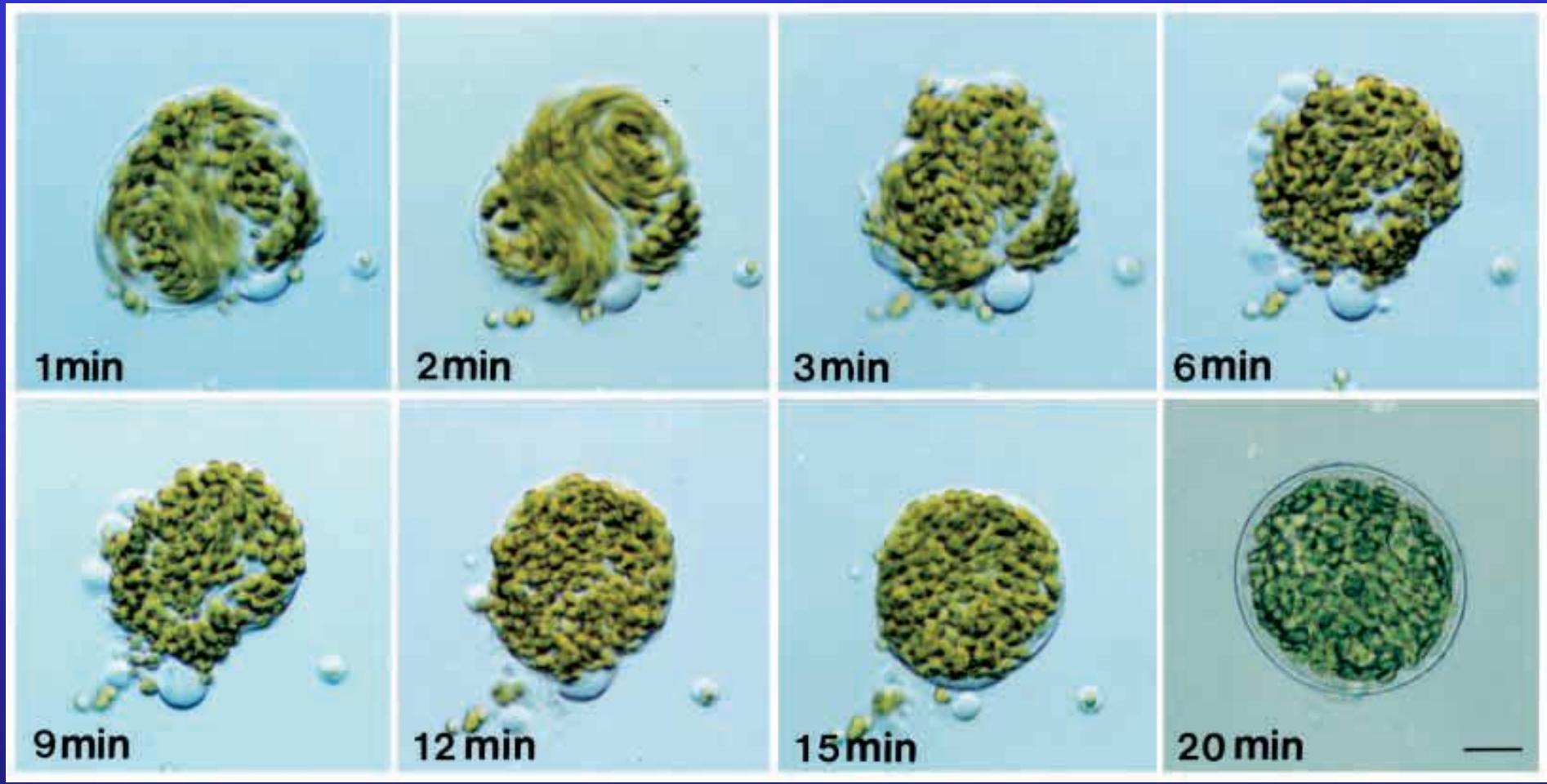


Sub-protoplast regeneration from disintegrated cells of *Bryopsis plumosa*



(A) Vegetative plant with distichous branches. (B) Protoplasm comes out from the wounded cell and spreads in seawater. (C) Aggregation of the extruded cell organelles in seawater. (D) Regenerated sub-protoplast with a primary envelope 20 minutes after wounding. (E) The secondary lipid-based membrane inside the primary envelope 12 hours after wounding

Kim et al. 2001

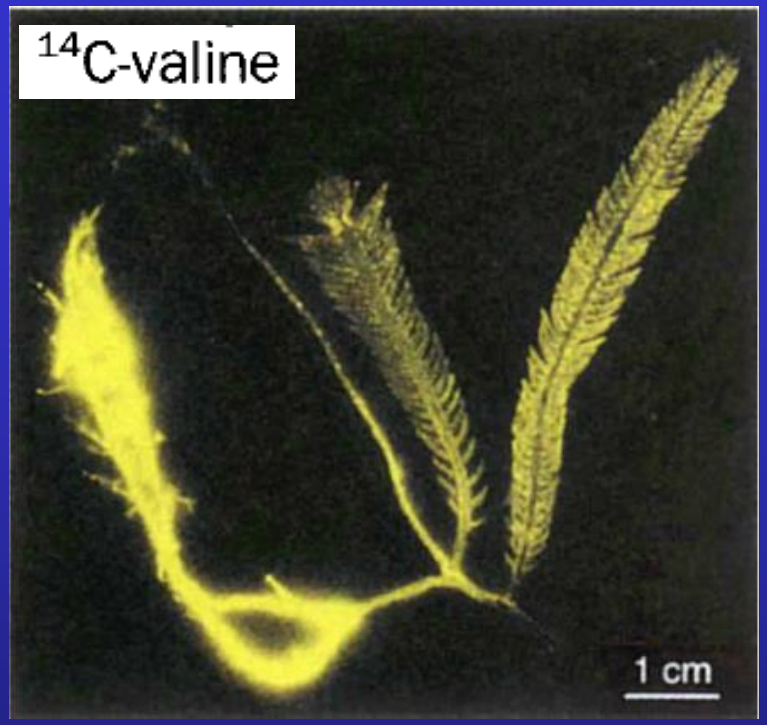


Time-lapse photography of early sub-protoplast regeneration. Bar, 10 mm.

Kim et al. 2001

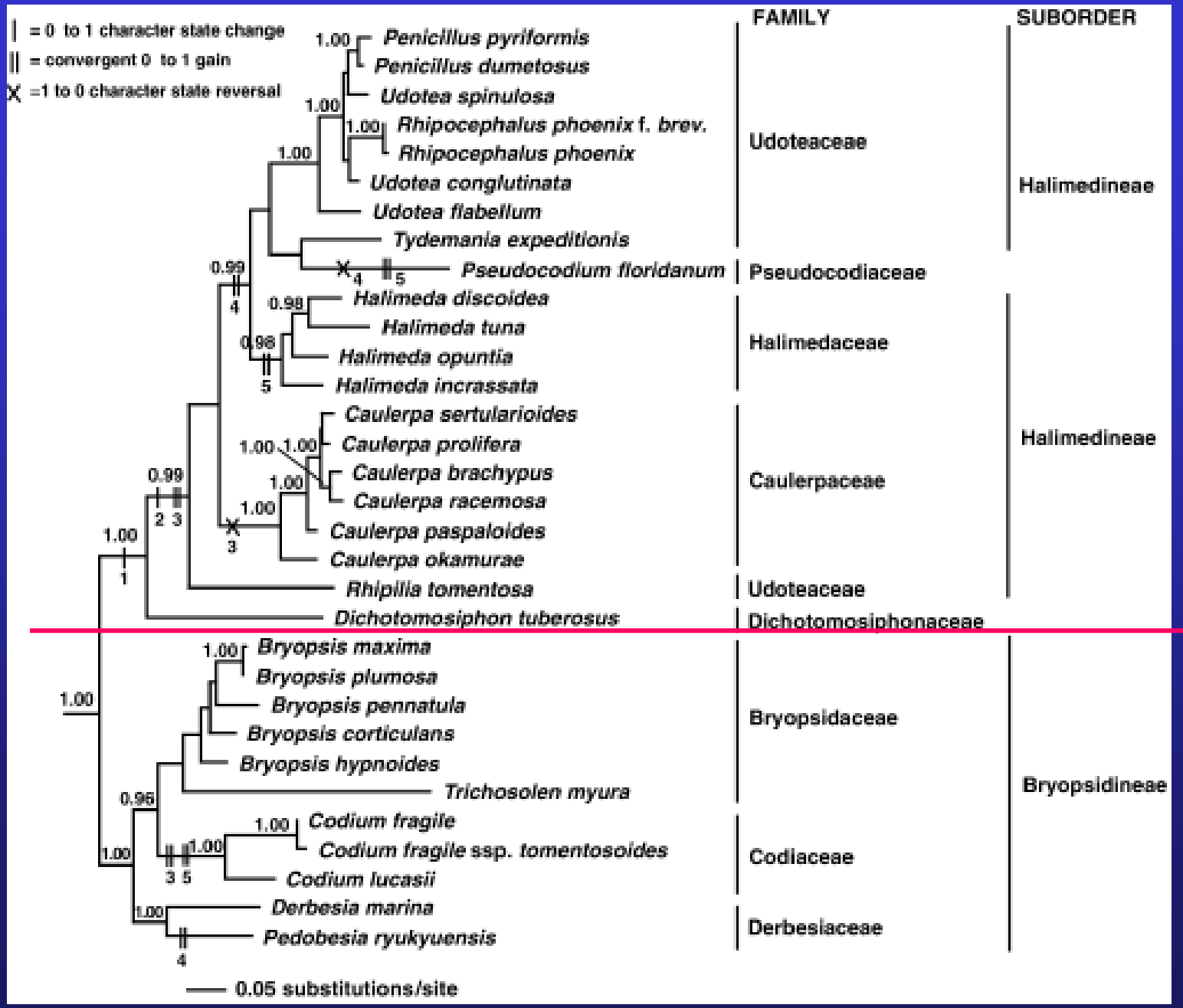
„Roots“ in mixotrophic algae C. taxifolia

Chisholm et al. 1996 Nature



Rhodopseudo monas – *nif* H gene

„root system“ containing endocellular bacteria – up take of inorganic phosphorus, C, organic N from substrata – translocation of nutrient to photoassimilatory organs (fronds)



Optimal tree topology of maximum likelihood analysis

linage - Bryopsidineae

- homoplastidic thalli
- thylakoid organizing body absent from chloroplasts
- production of zoids is non-holocarpic (gametangia and sporangia are formed withis special zones of the thallus; whole thallus in not destroyed)
- many members (though not all) produce stephanokont zoids
- main cell wall polysaccharide is mannan, xylan, cellulose

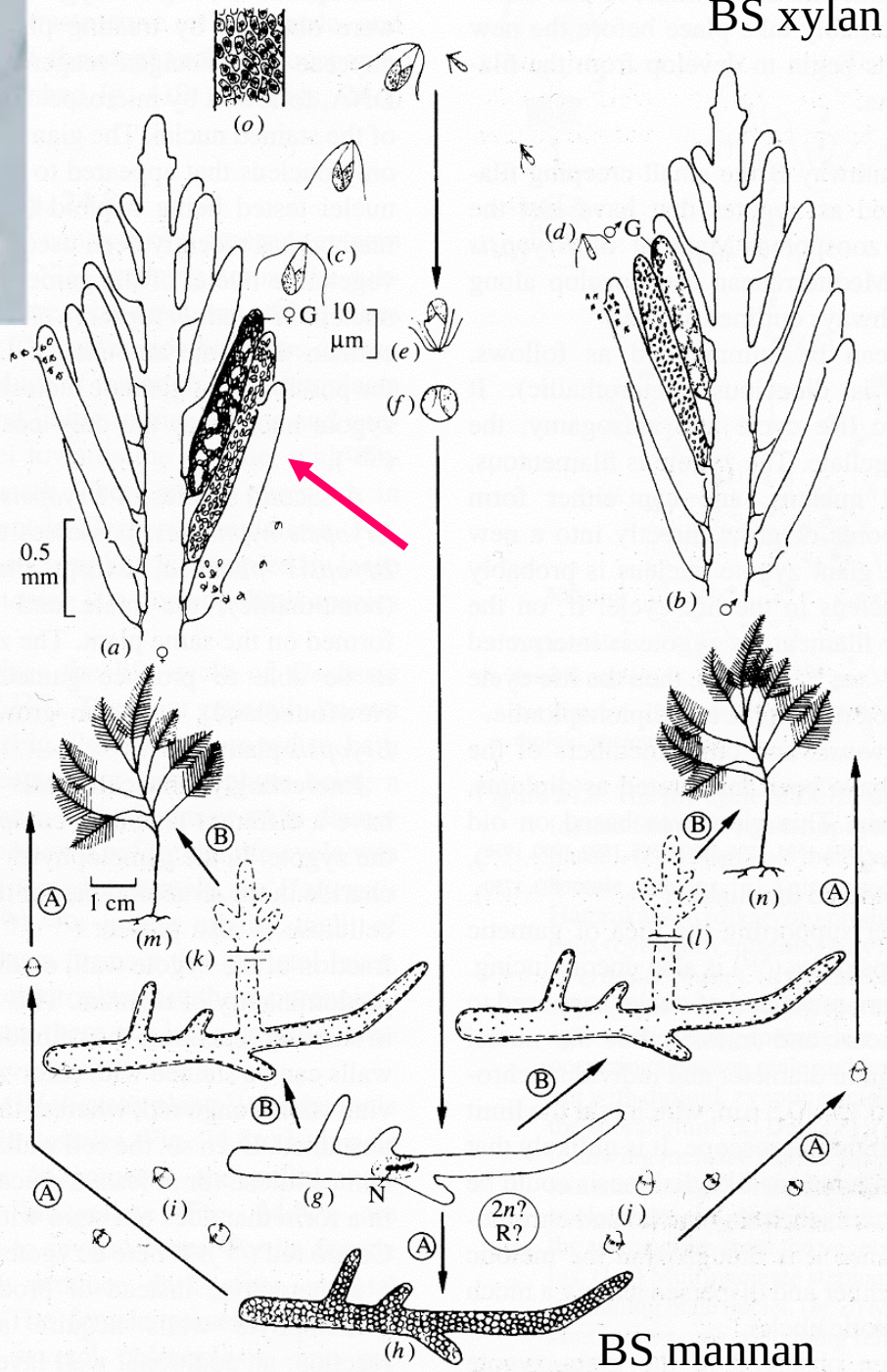
Codium

Bryopsis

Derbesia

Bryopsis plumosa (10-15 cm)

BS xylan



BS mannan

Light and electron micrographs of sporophytes of *Bryopsis plumosa.*

Minamikawa, et al.2005

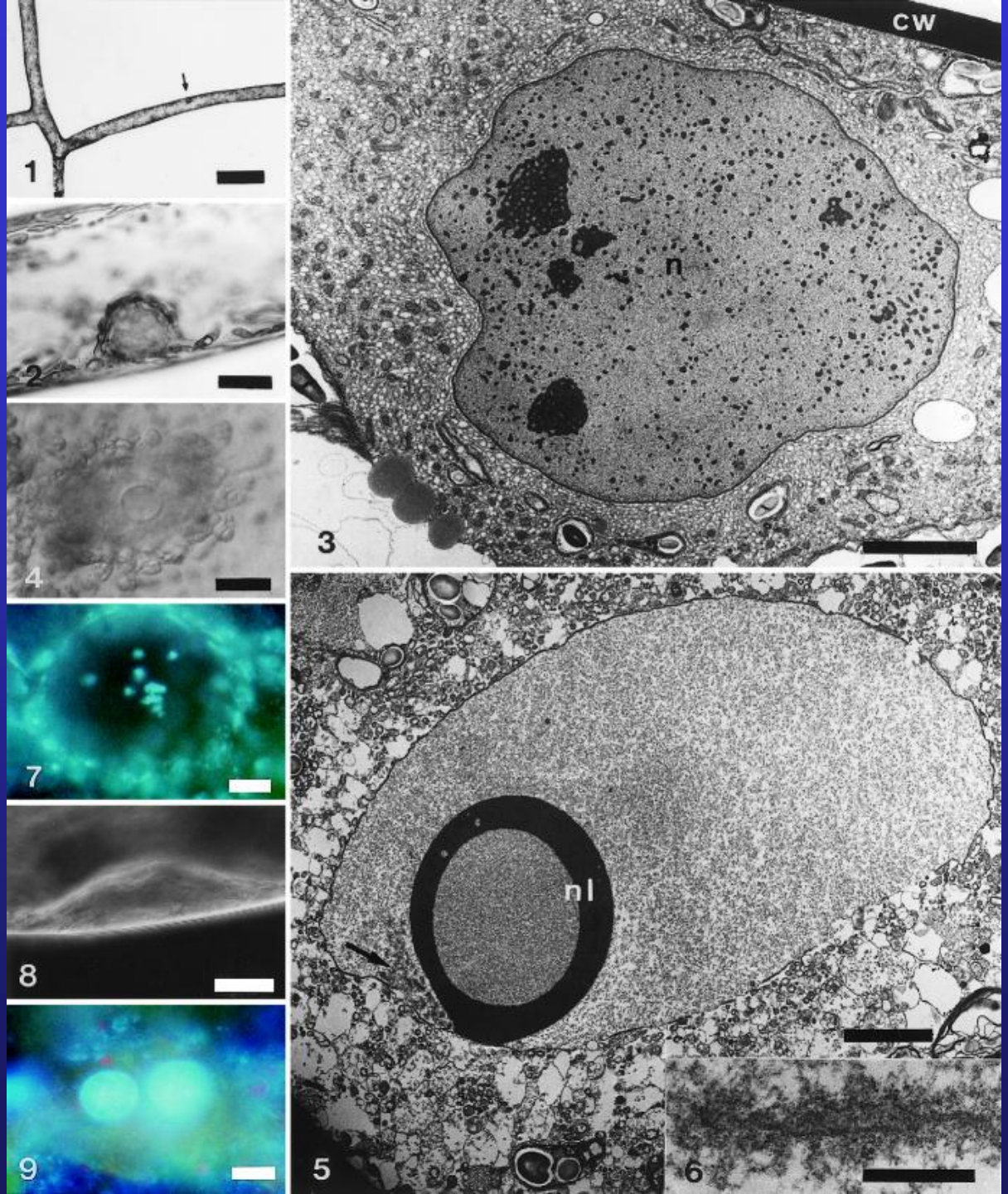
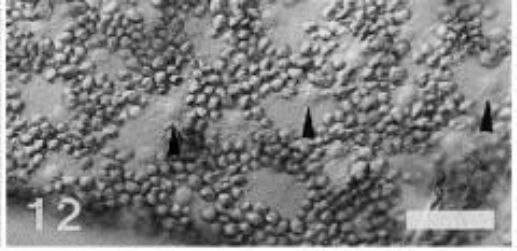
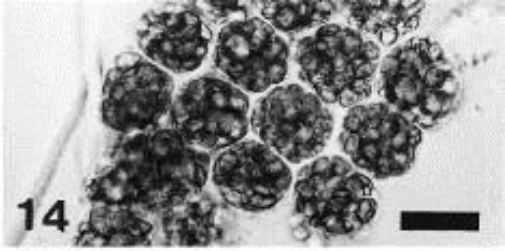


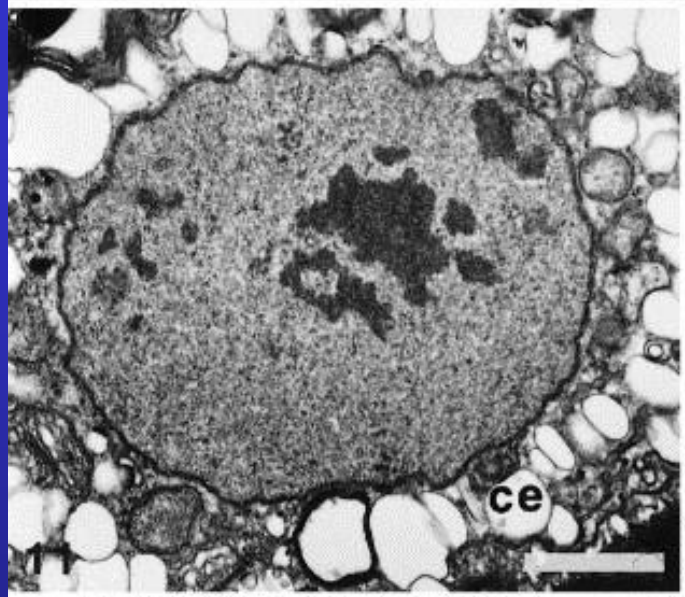
Fig. 1 . *1* a sporophyte, containing a giant nucleus (*arrow*). *2* Giant nucleus, about 25 um in diameter. *3* Electron micrograph of a giant nucleus. *4* Giant nucleus containing a ring-shaped nucleolus. *5* Electron micrograph of *7* Giant nucleus containing chromosomes (Hoechst staining). *8* Dumbbell-shaped giant nucleus. *9* Two small nuclei.

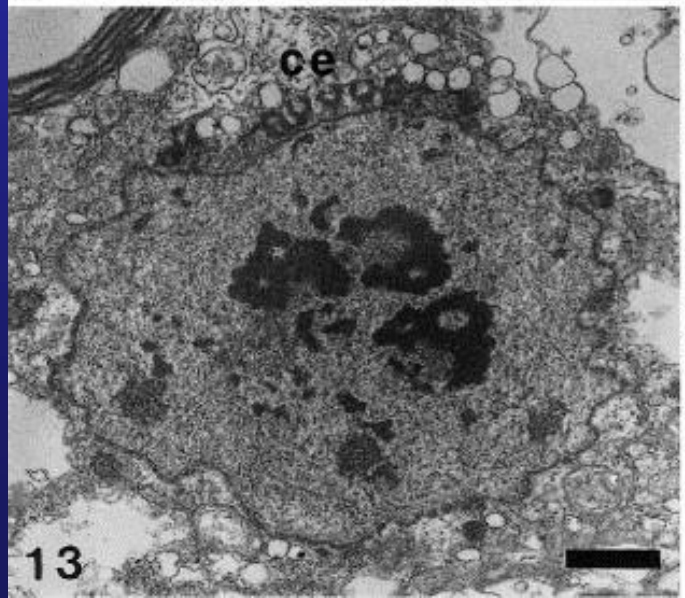
Light and electron micrographs of sporophytes of *Bryopsis plumosa*

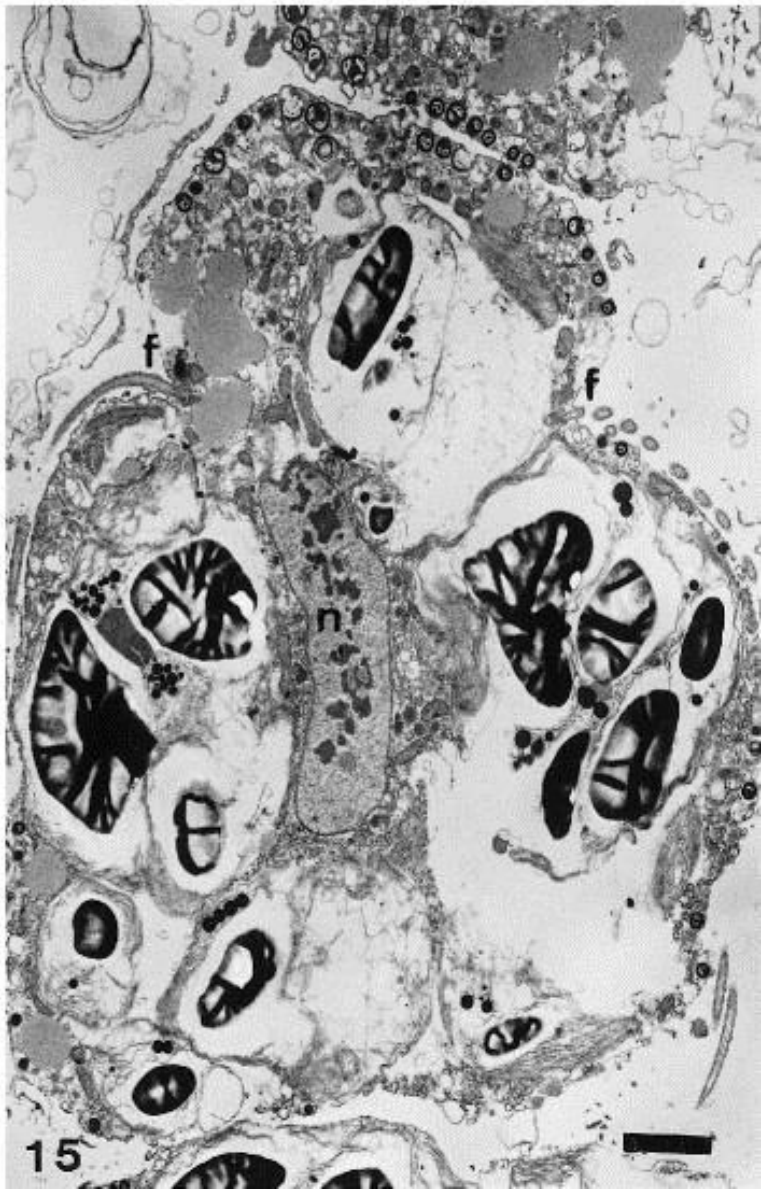
Stephanokontic zoospores just before liberation – 14,15









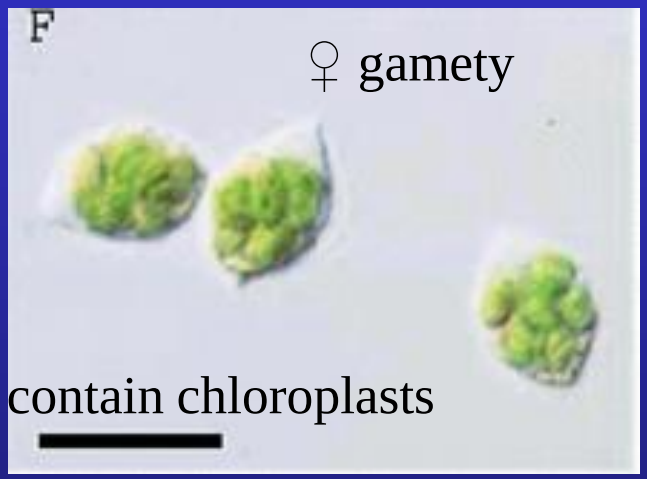


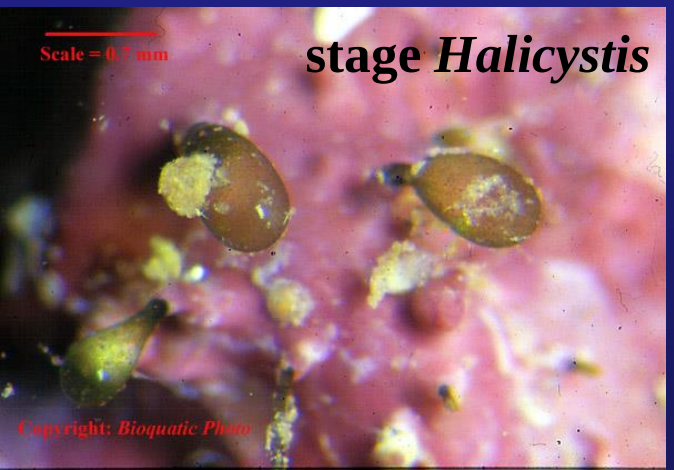
Derbesia

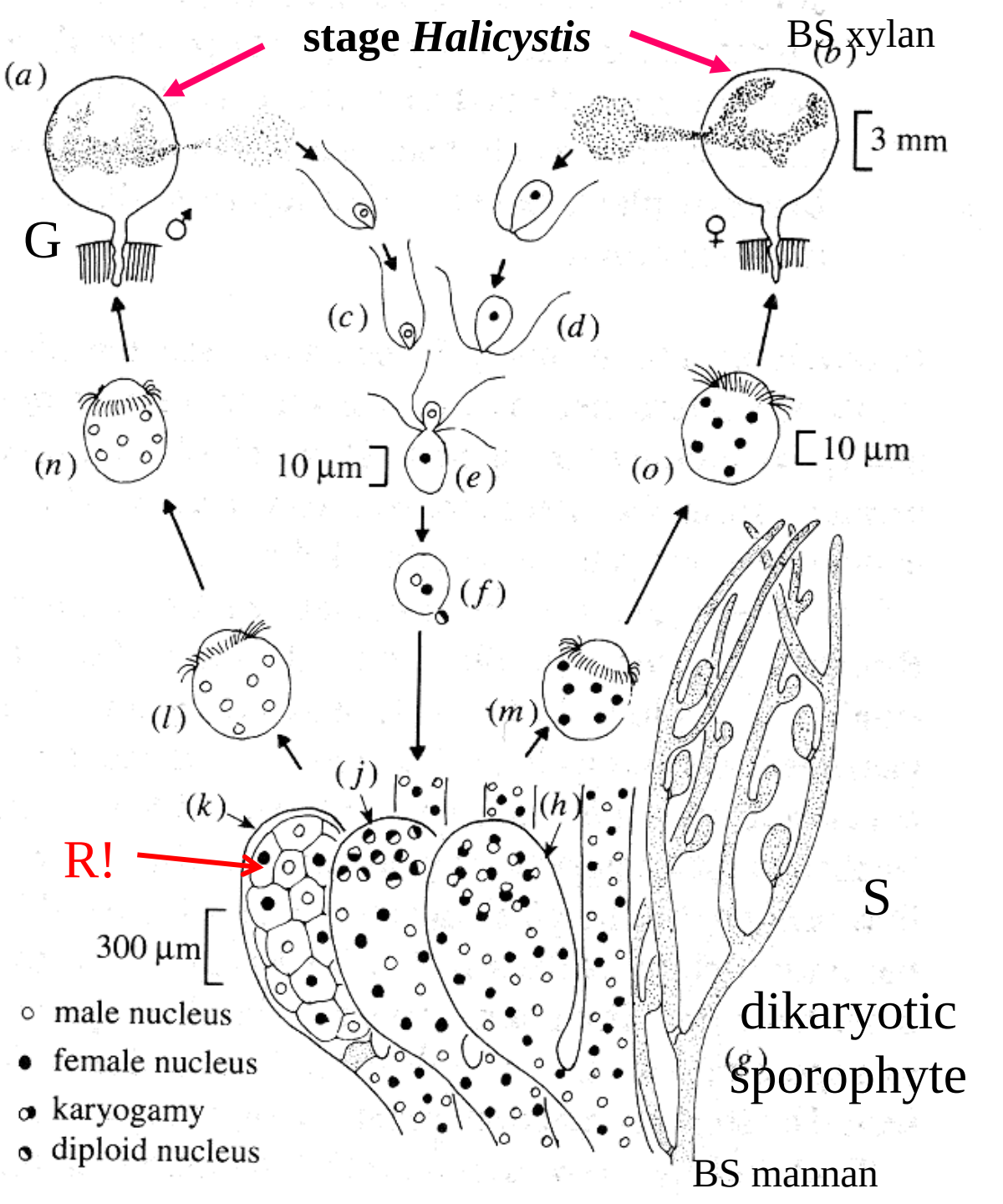
uniaxial thallus

stage *Halicystis*

E
Derbesia


F
♀ gamety
contain chloroplasts


Scale = 0.7 mm
stage Halicystis
Copyright: Bioquatic Photo


stage Halicystis
BS xylan
(a)
(b)
3 mm
G
♂
♀
(c)
(d)
(n)
10 µm
(e)
(o)
10 µm
(f)
(l)
(m)
(j)
(k)
(h)
R!
S
300 µm
male nucleus
female nucleus
karyogamy
diploid nucleus
(g)
dikaryotic
sporophyte
BS mannan

videa

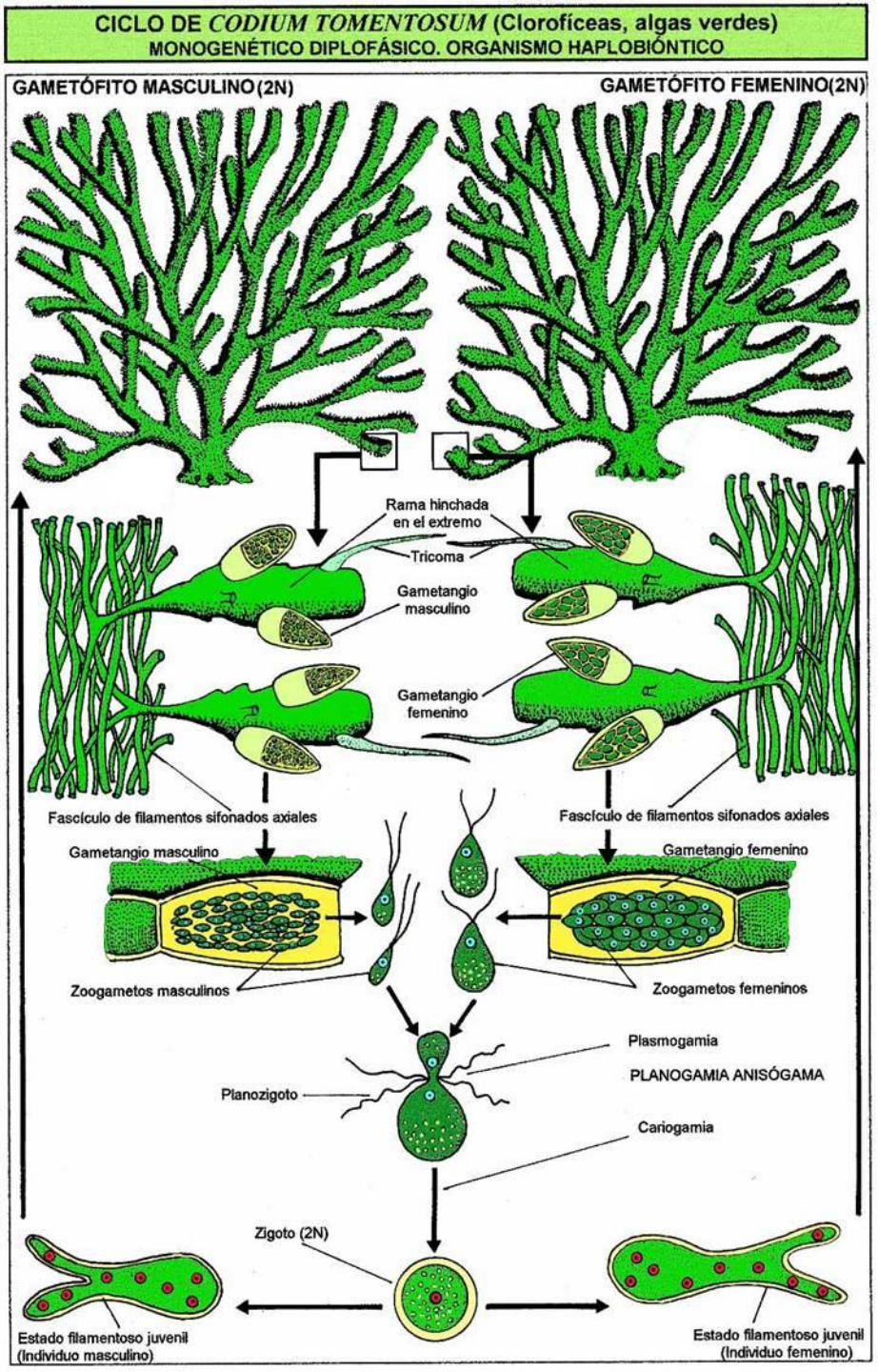
http://www.cytographics.com/

http://www.cytographics.com/clip_articles.html

Codium fragile

multiaxial thallus, medulla, utriculi

Codium

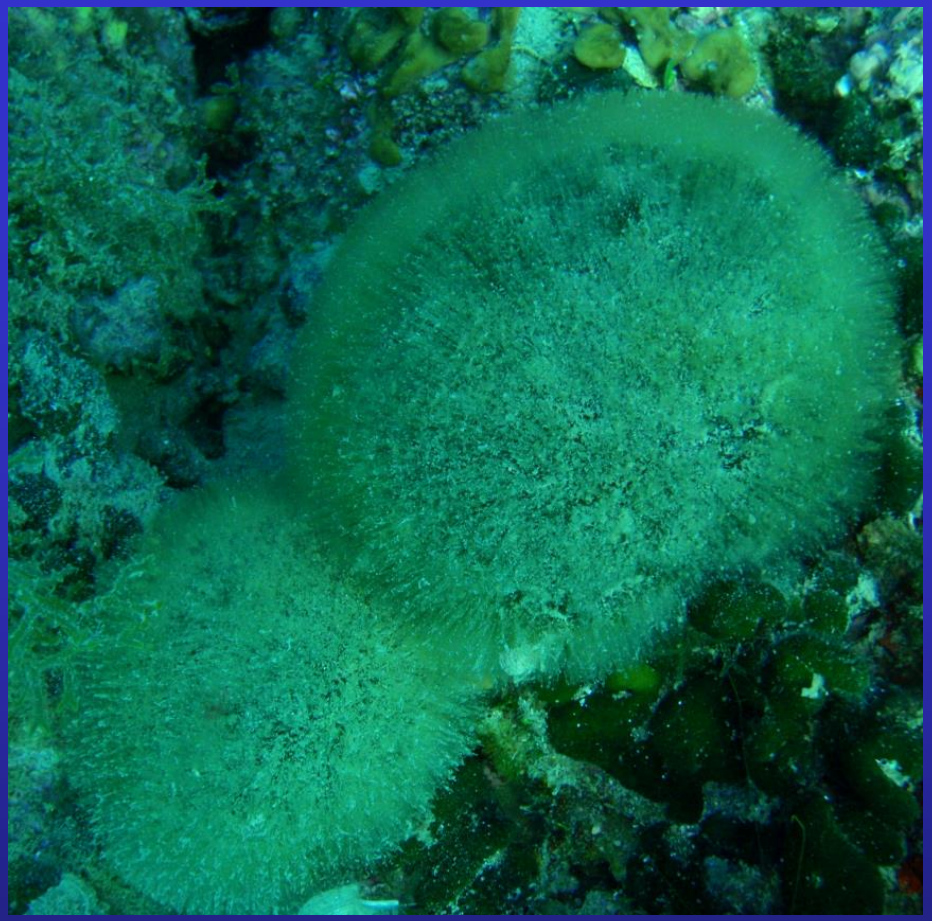


Codium

Codium dimorphum

Codium sp.

Scallop (Argopecten)



Codium – Piran/Punta; Mediterranean Sea

Codium

Codium bursa

C. fragile

linage Halimedinae

- heteroplastidic species (chloroplasts + amyloplasts)
- thylakoid organizing body in both the chloroplasts and amyloplasts
- production of zoids (gametes) holocarpic (whole thallus is entirely transformed to gametes, empty cell wall is left)
- stefanokont zoidy are never produced
- main cell wall polysaccharide xylan (with little or no cellulose)
- mass simultanous release of ♂ and ♀gamets

Simultanous release of ♂ and ♀gamets

Characteristic for *Caulerpa*, *Halimeda*, *Penicillus*, *Rhipocephalus* and *Udotea* (ca 17 species) Clifton, 1997

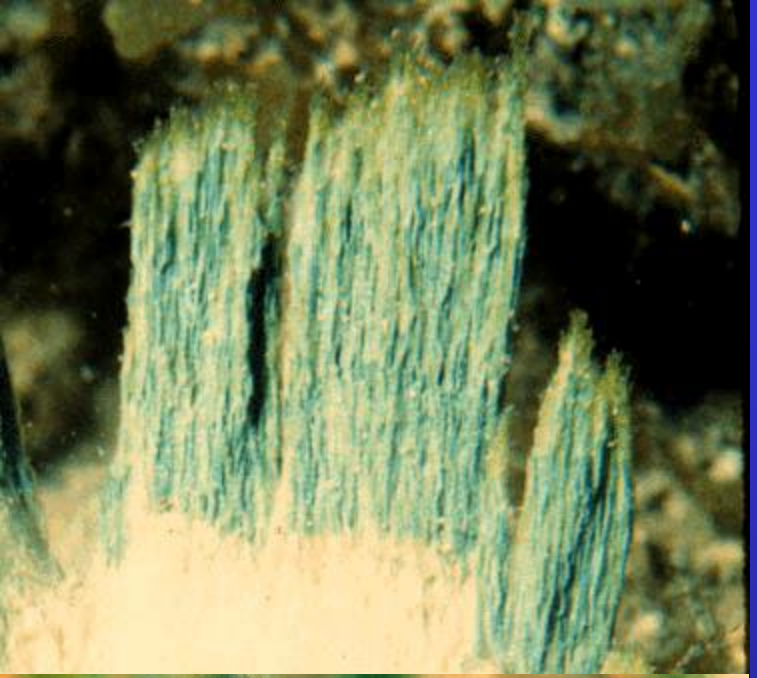
Penicillus. Note the contrast between a non-fertile thallus on the left and a fertile female on the right (dead thallus from earlier reproduction in the middle)

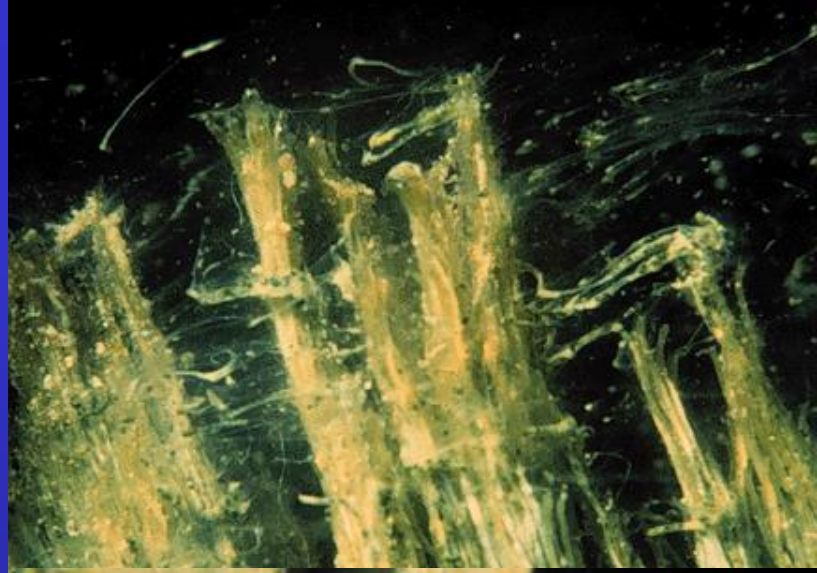
Simultanous release of ♂ and ♀gamets

sterilní *Udotea caribbaea*

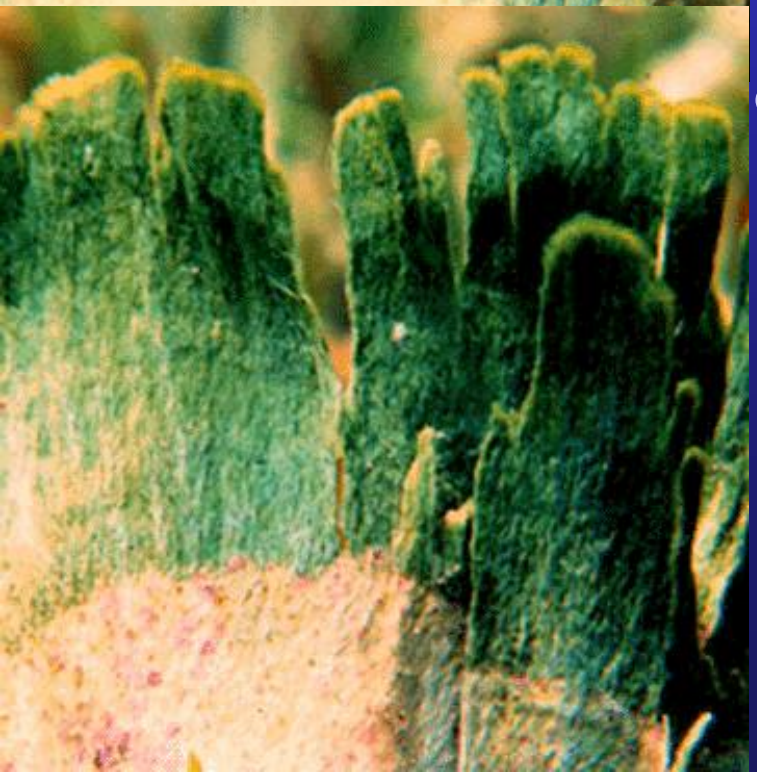
plodná ♀ stélka *Udotea cyathiformes*. Bílé části stélky – migrace protoplasmy do gametangií

Udotea caribaea





♀thallus



♂thallus

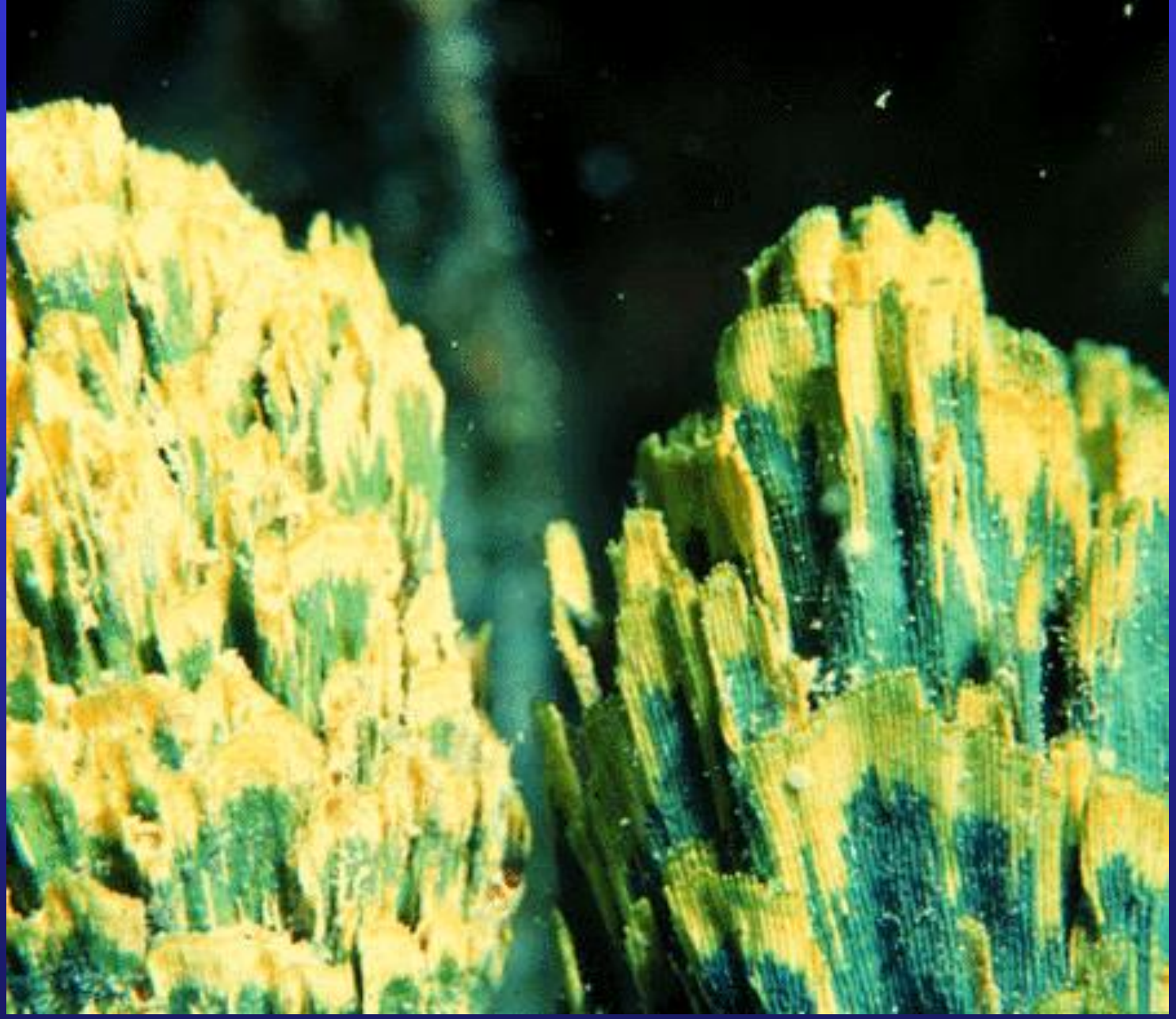
release of gametes →

Simultanous release of ♂ and ♀gamets

♀ **gametangia**

♂ rostlina vypouští gamety

Penicillus



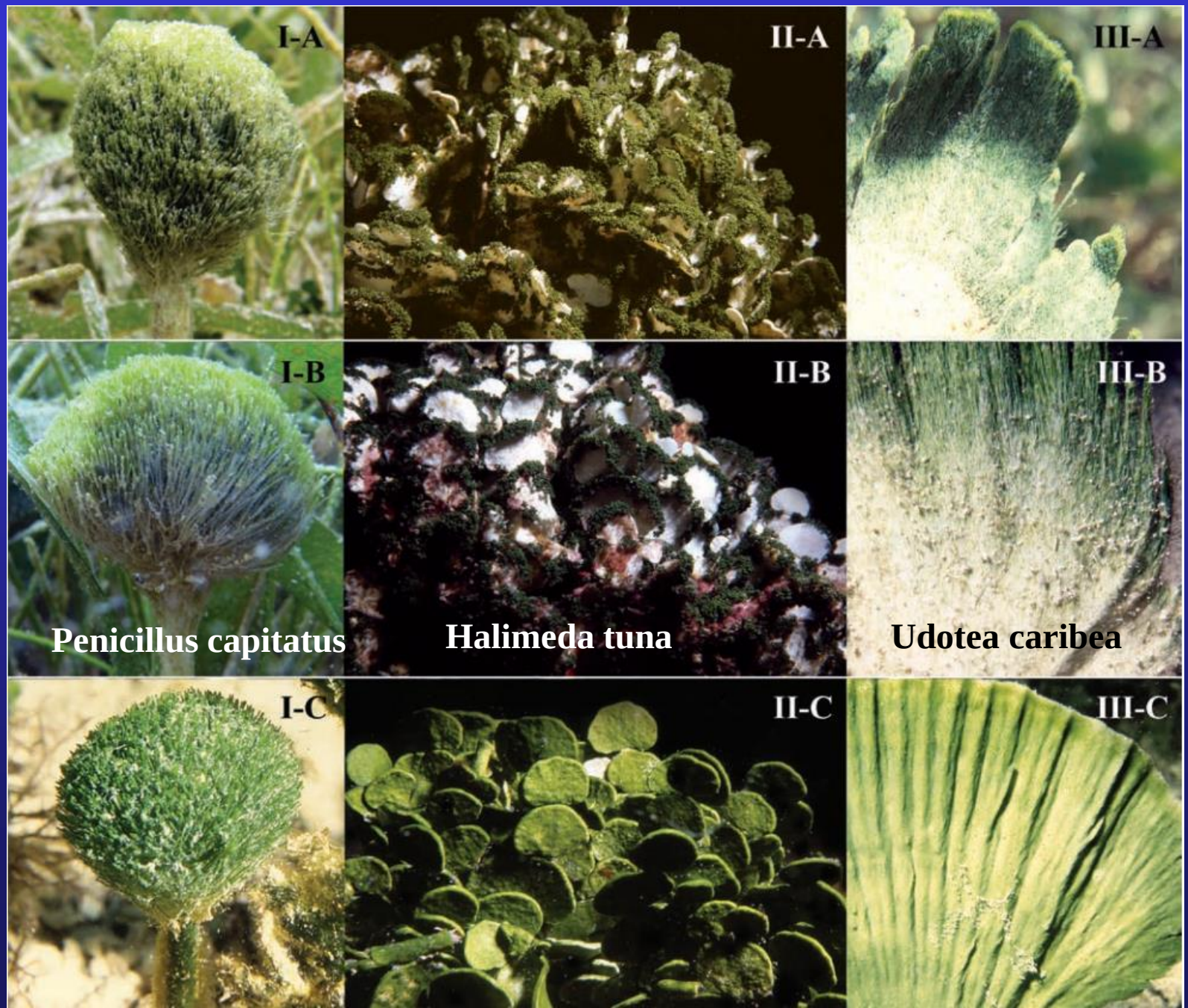
Rhipocephalus phoenix ♂ thallus (left), ♀(right)

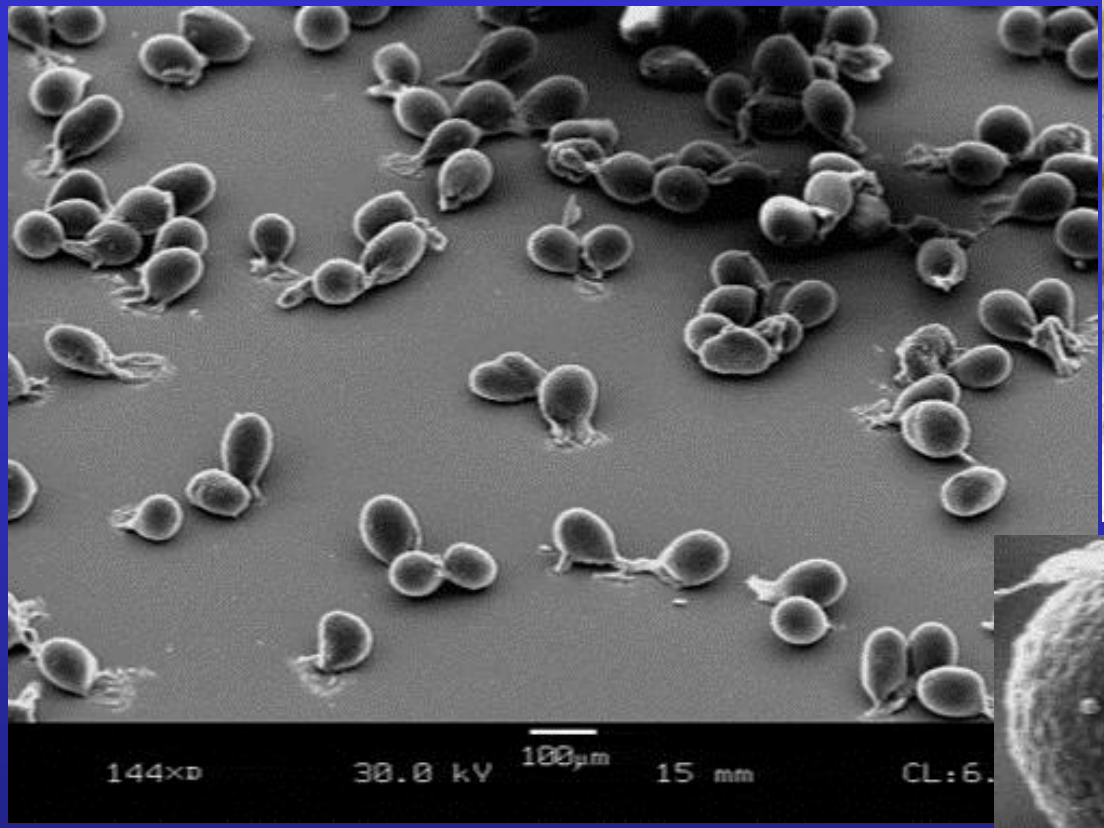
Examples of fertility—evident by the presence of gametangia and color change; Clifton 2013

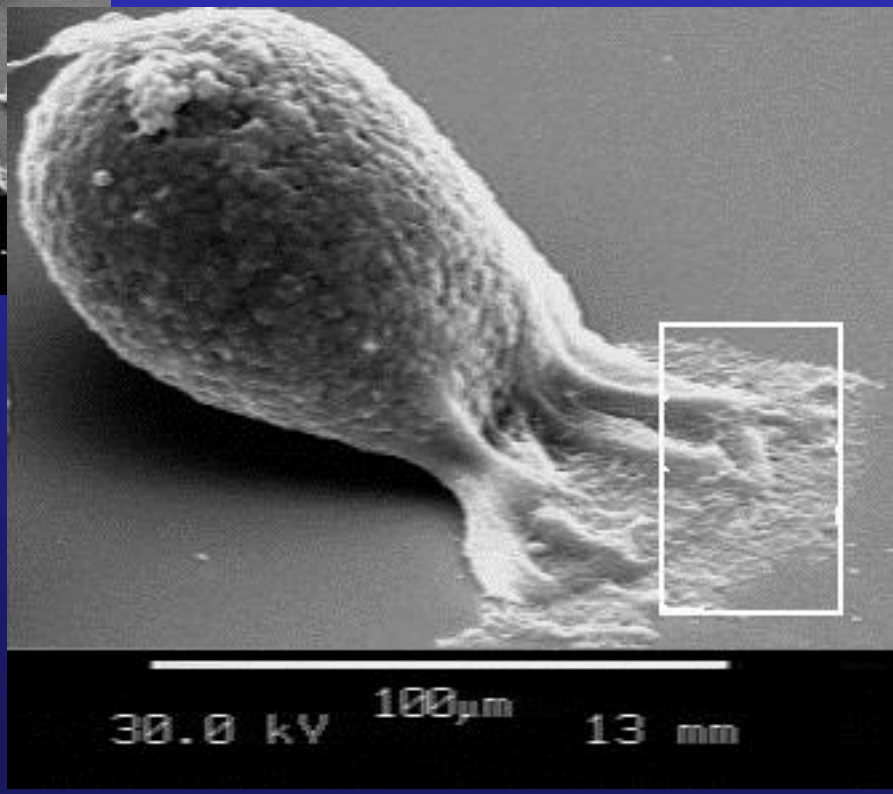
♂

♀

nonfertile thalli





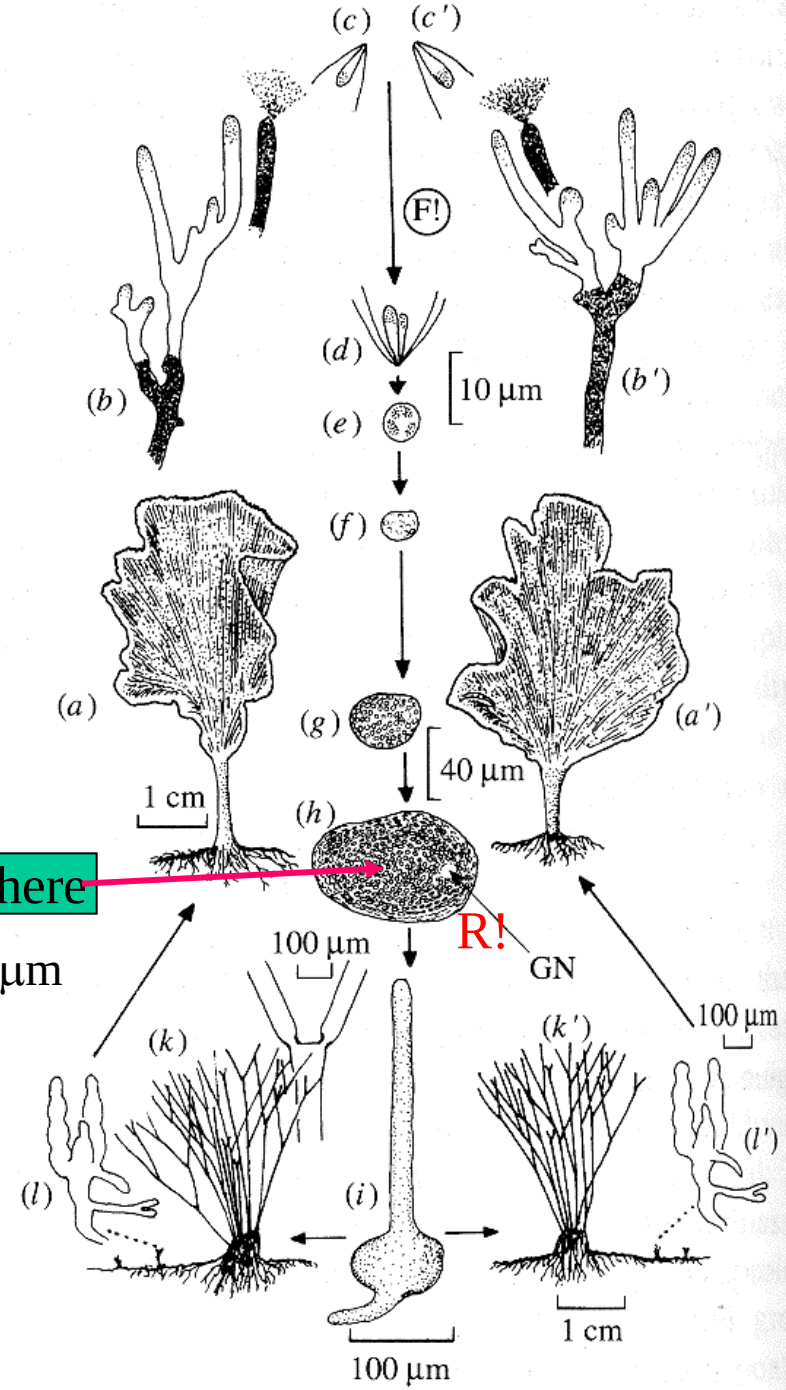


♀ gamety *Rhipocepalus phoenix*

-pinecone alga

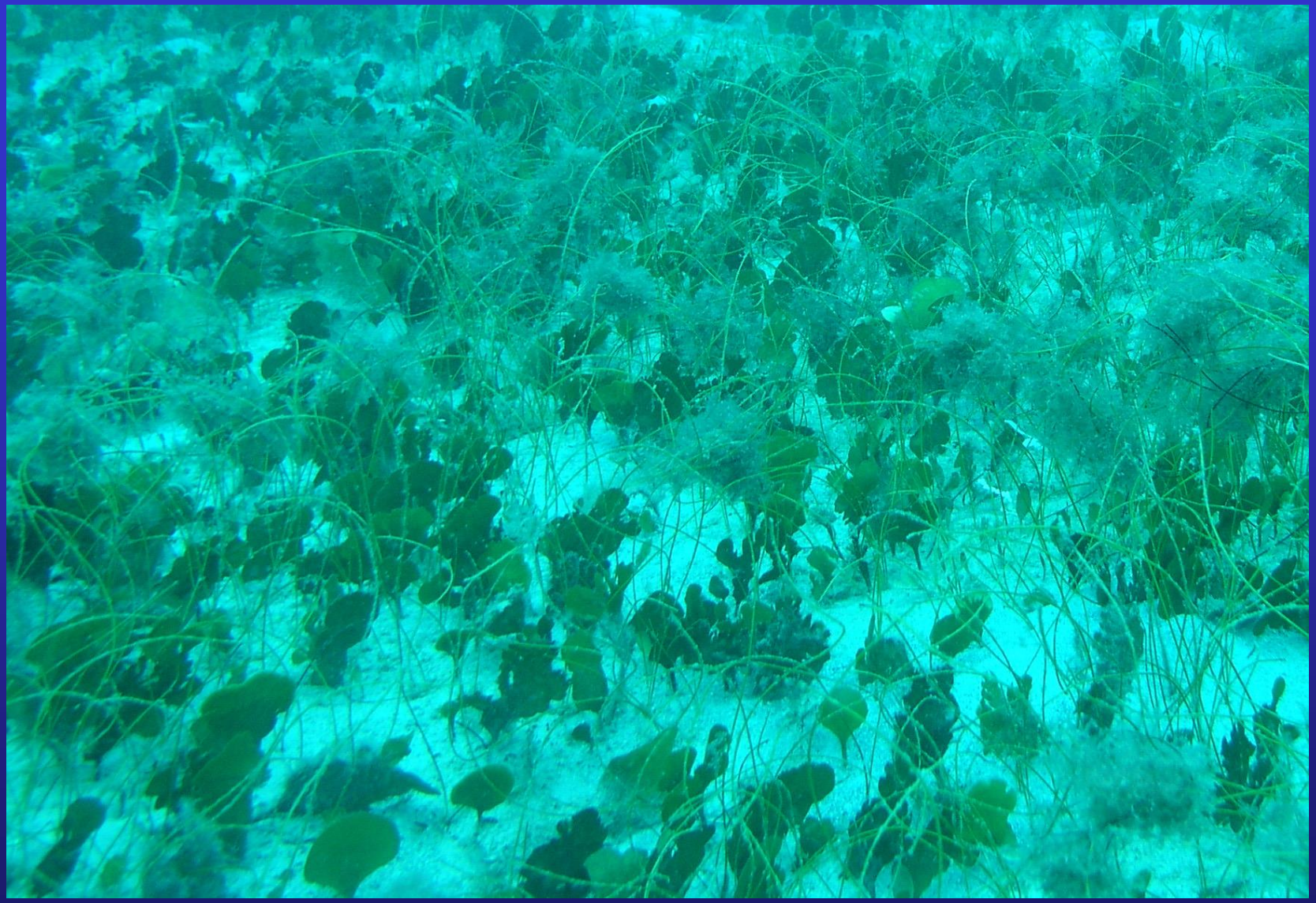
protosphere

50-90 µm



Flabellia petiolata = Udotea petiolata

– Piran/Strunjan - Mediterranean Sea

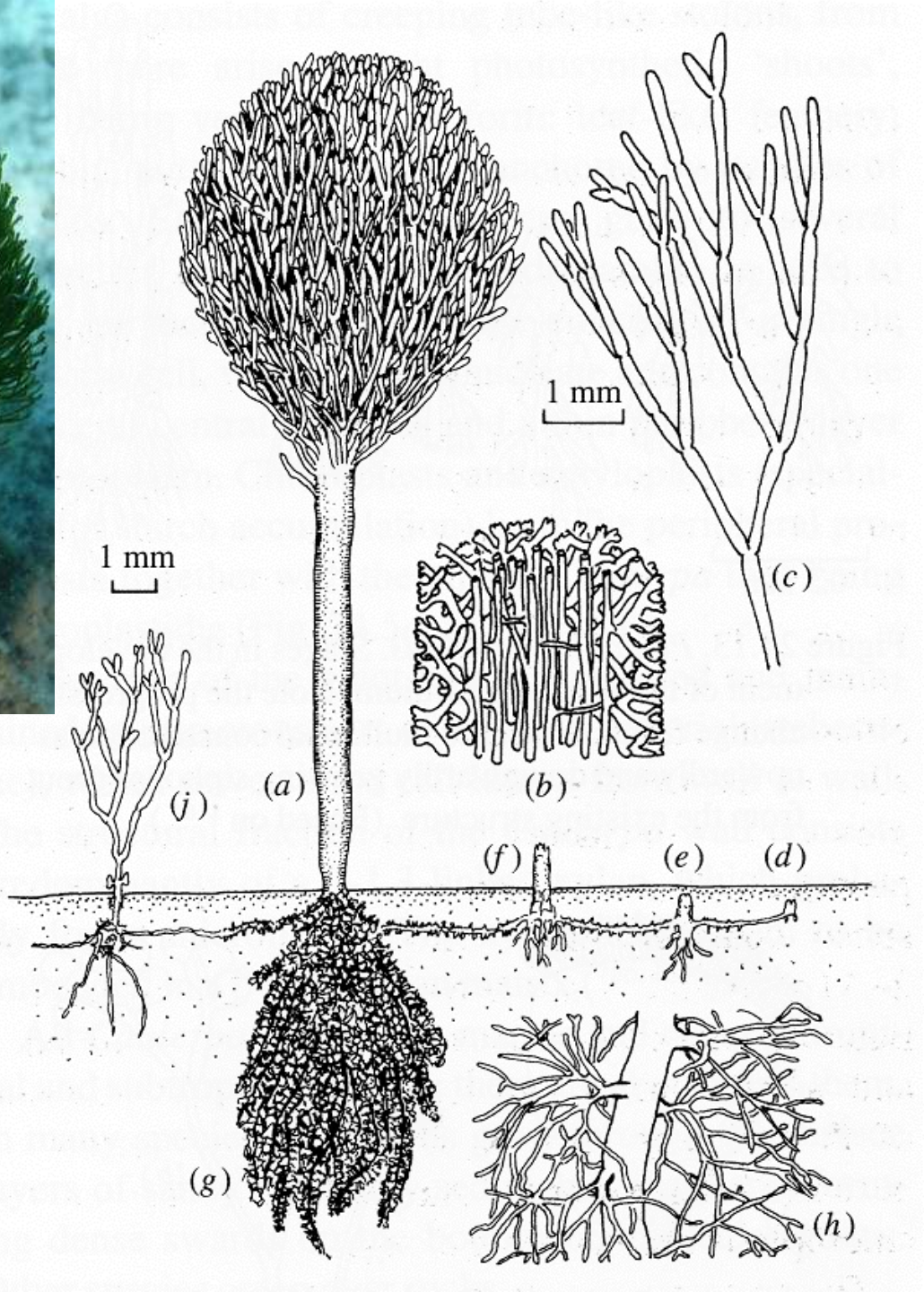


Genus: Udotea and Syringodium

Location: Puerto Rico

Udotea Panama reef

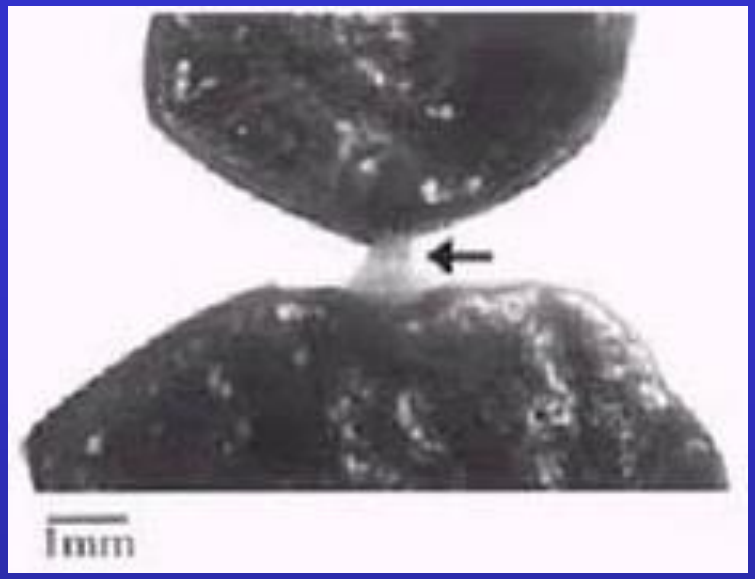
Neptune's Shaving Brush



Halimeda

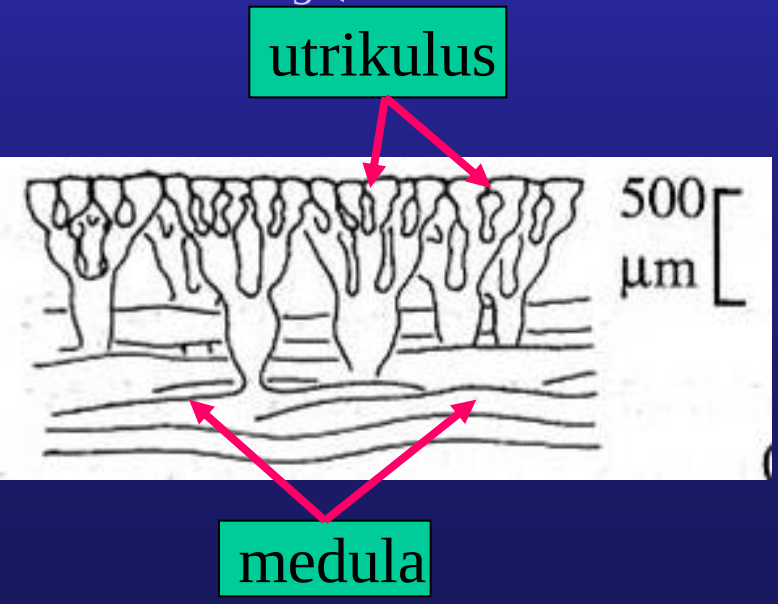
Halimeda tuna

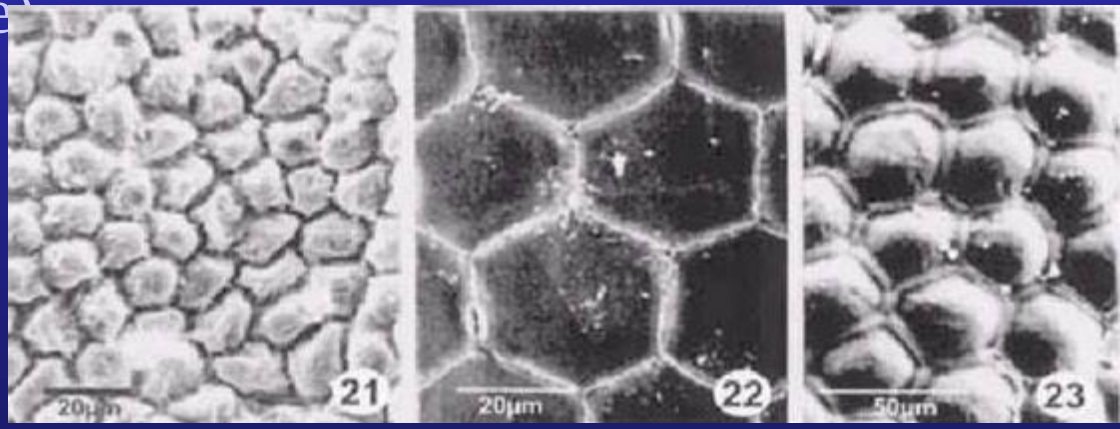
Halimeda cuneata



detailed view in SEM

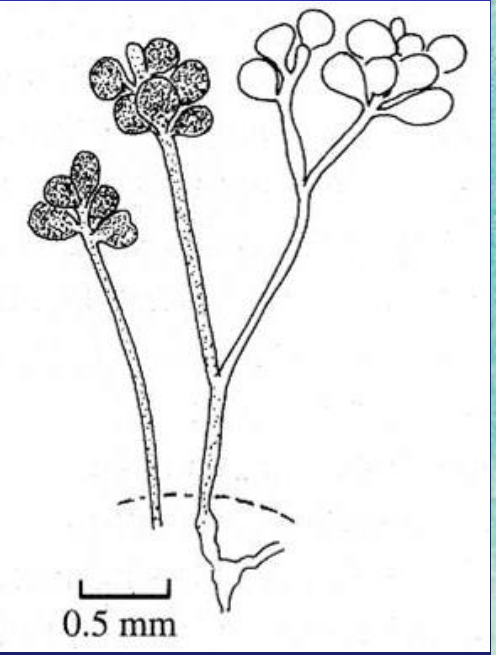
H. cuneata – nodulus není inkrustován $CaCO_3$ (calcium carbonate)

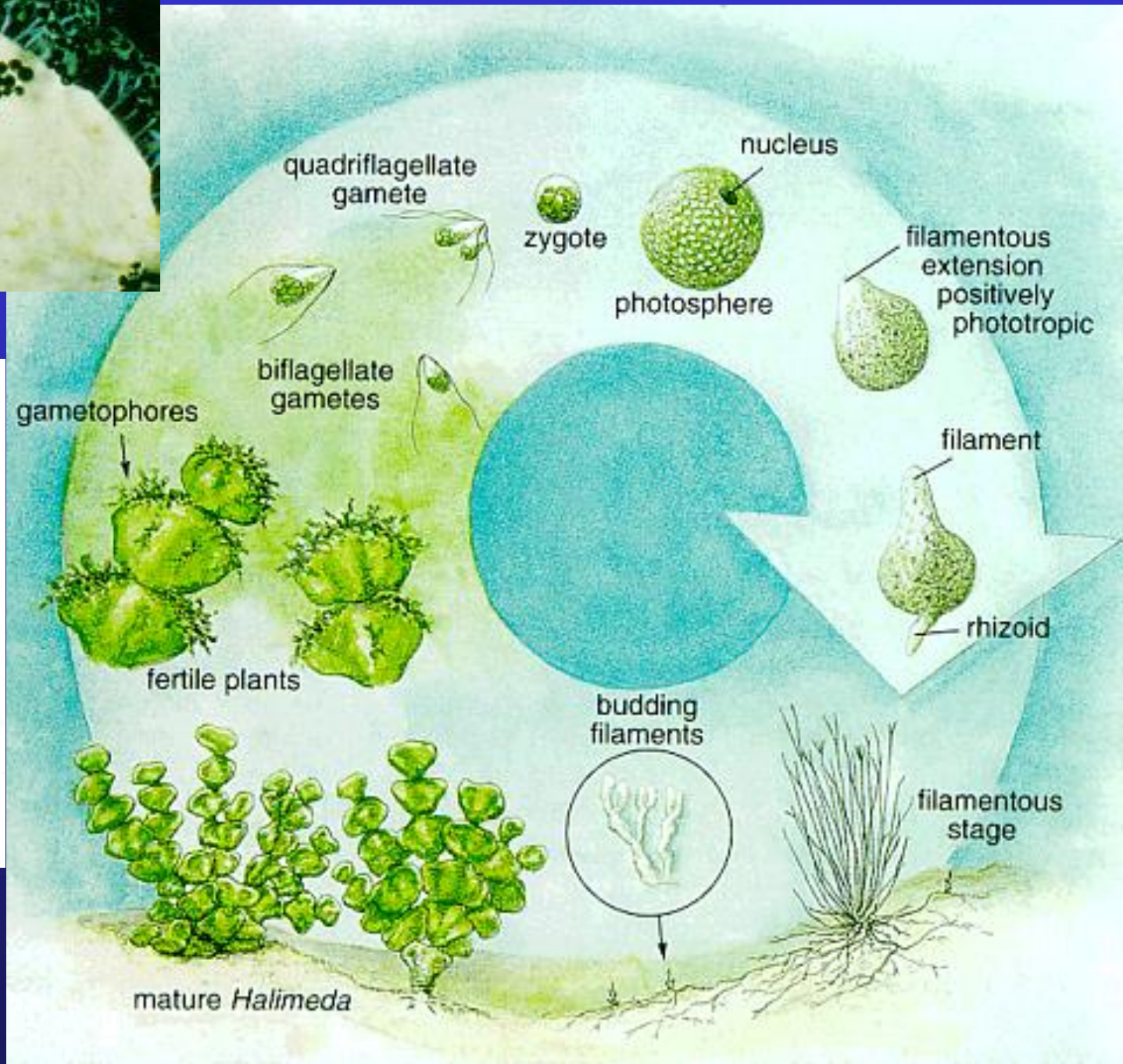




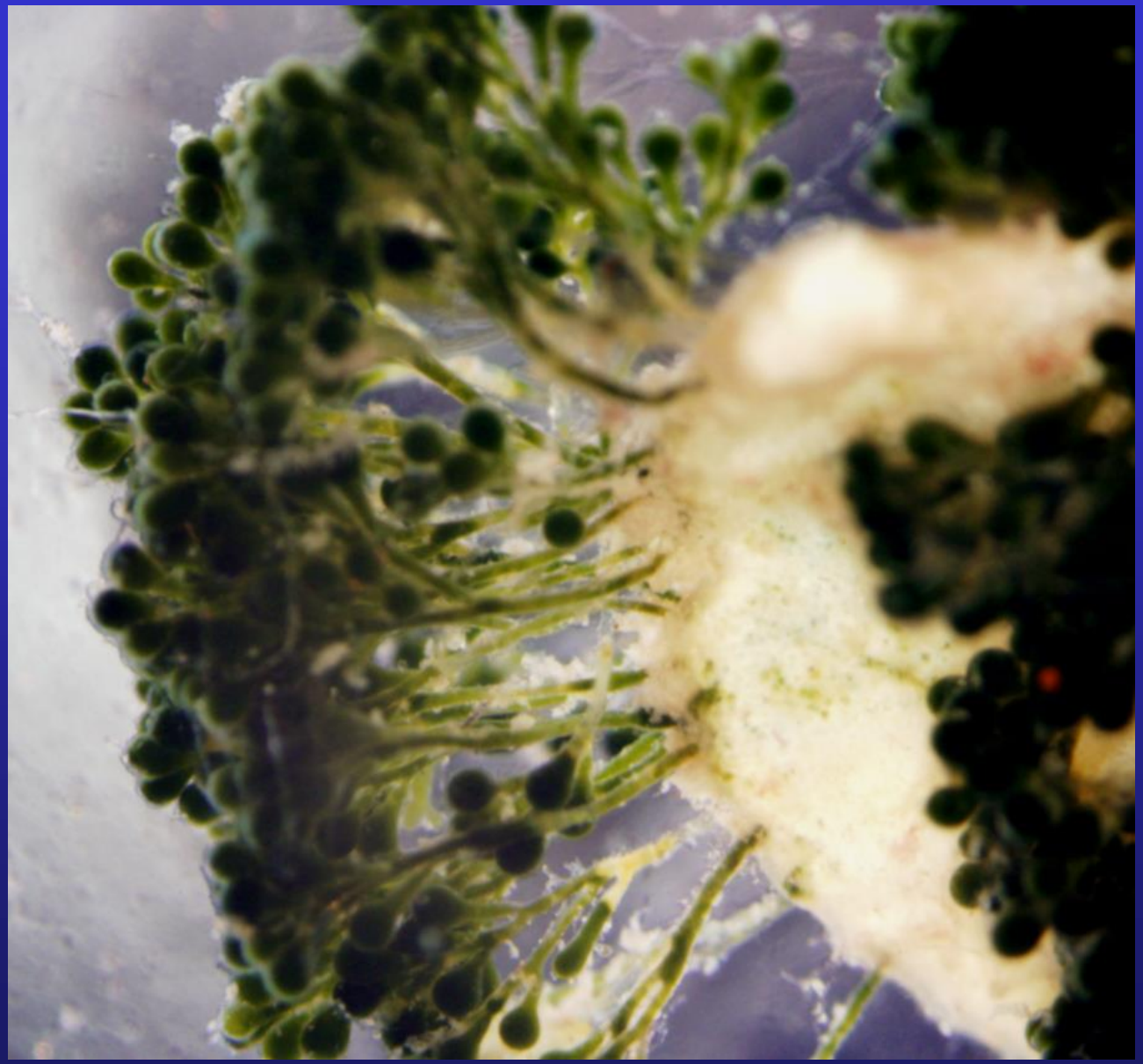
segment surface in different *Halimeda* species SEM

gametangia





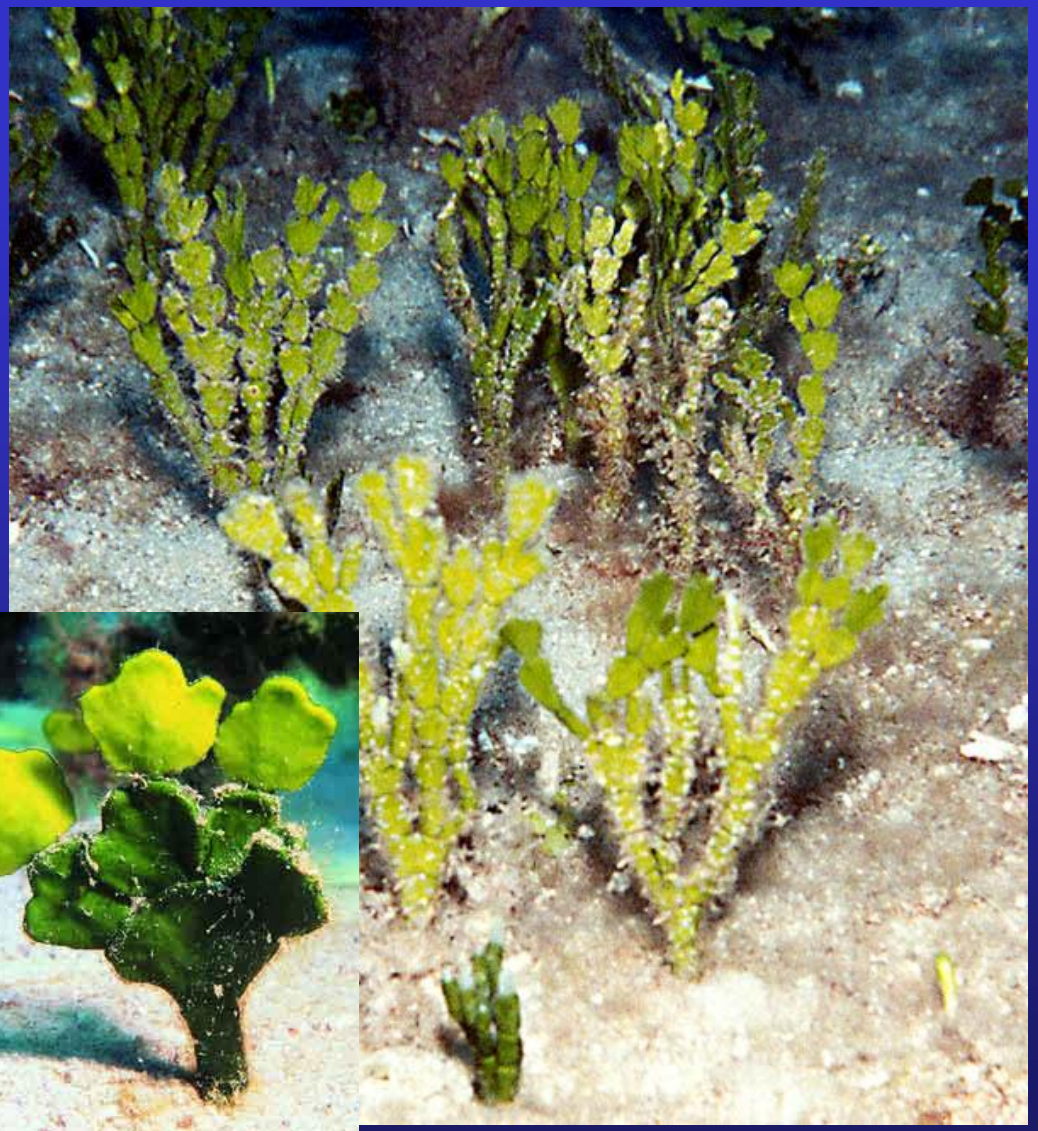
Halimeda



gametangia *Halimeda tuna* – Mediterranean Sea

Halimeda

H. discoidea



Halimdea incrassata

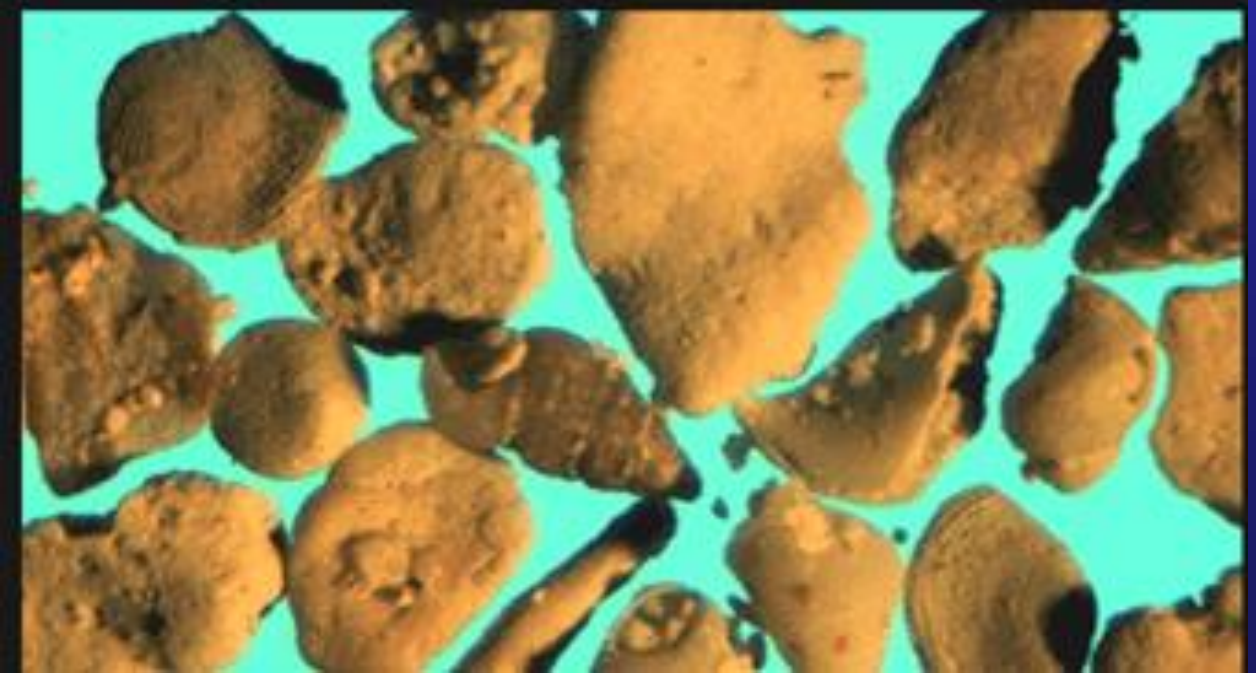
Halimeda

submarine turtlegrass beds

This Arrowhead Crab, *Huenia heraldica,* uses living leaves of Halimeda algae as a hat. Mimikry

Halimeda

tropic beach in Belize



The flattened grains are fragments from jointed calcareous plates of the green alga **Halimeda** (probably **H. opuntia**). This interesting green alga grows among the submarine turtlegrass beds and the luxuriant coral reefs of this region.

Fig. 1. Illustration of morphological variation in the species *Halimeda cuneata* (A–N), *H. discoidea* (O–Q) and *H. tuna* (R, S). (A, B) *H. cuneata* 1. (C, D) *H. cuneata* 2. (E, F) *H. cuneata* 3. (G, H) *H. cuneata* 4. (I, J) *H. cuneata* f. *digitata*. (K, L) *H. cuneata* f. *undulata*. (M, N) *H. cuneata* 7. (O) *H. discoidea* 1. (P) *H. discoidea* 2. (Q) *H. discoidea* 3. (R) *H. tuna* 1. (S) *H. tuna* 2.

Verbruggen et al. 2005

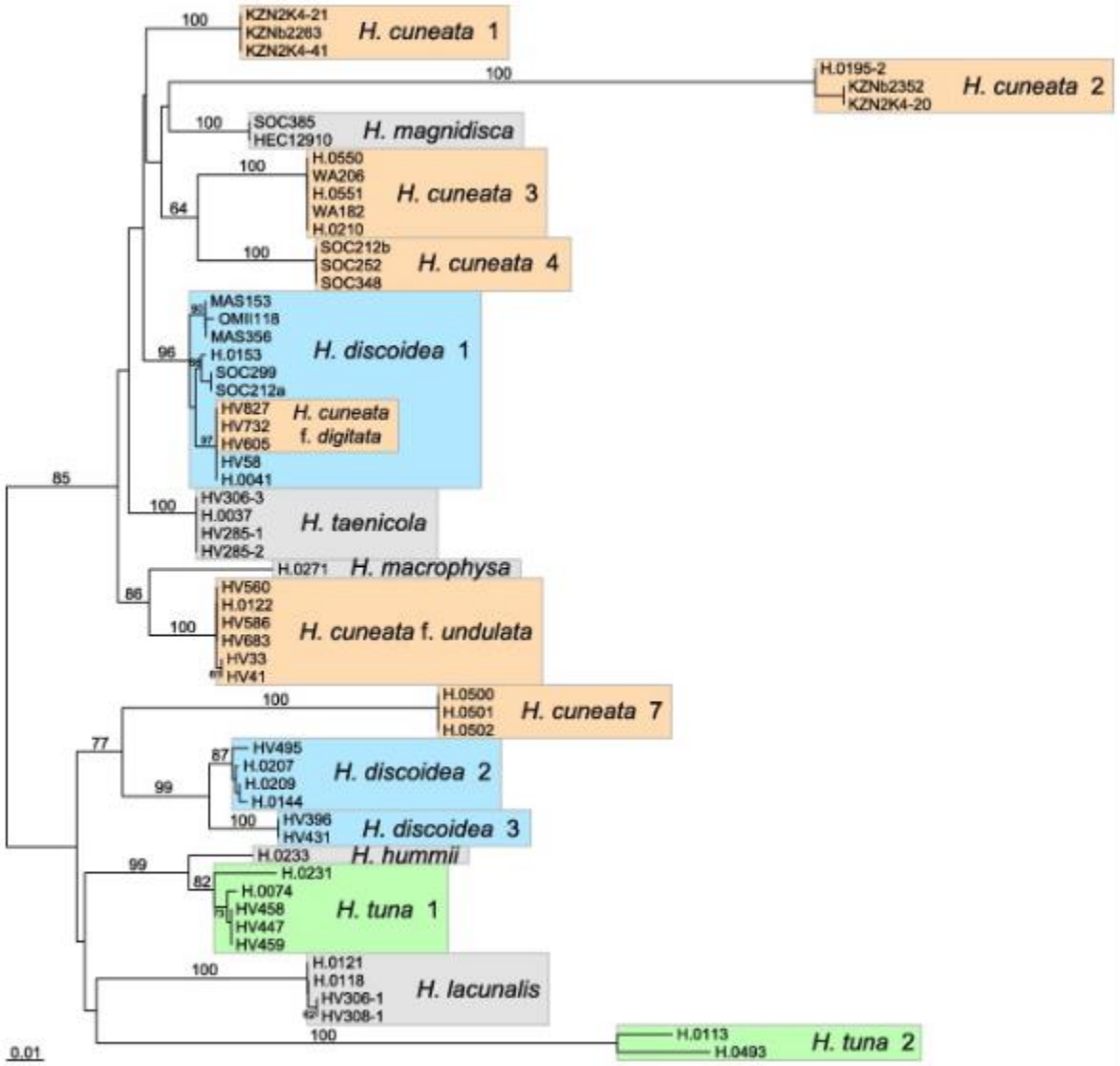
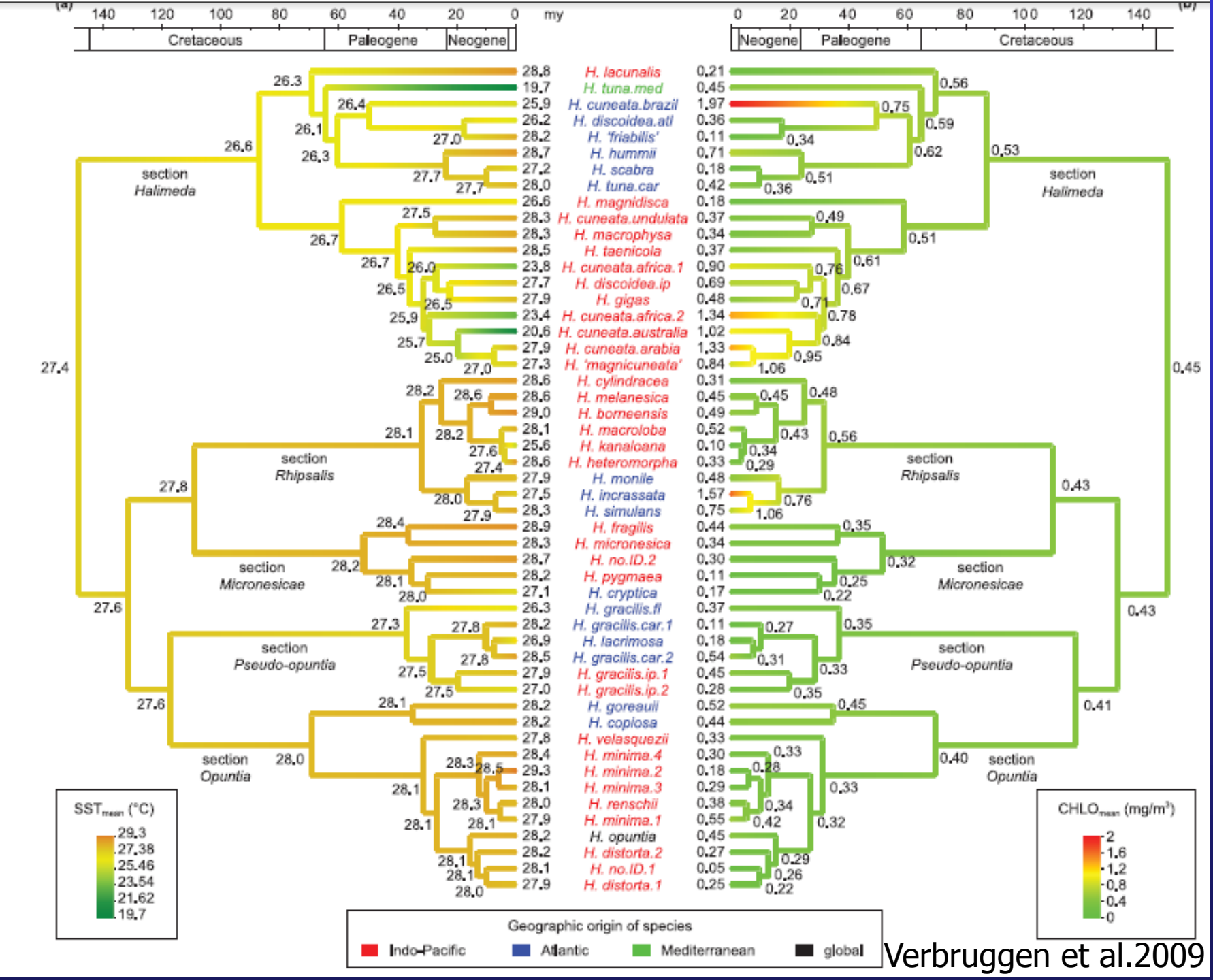
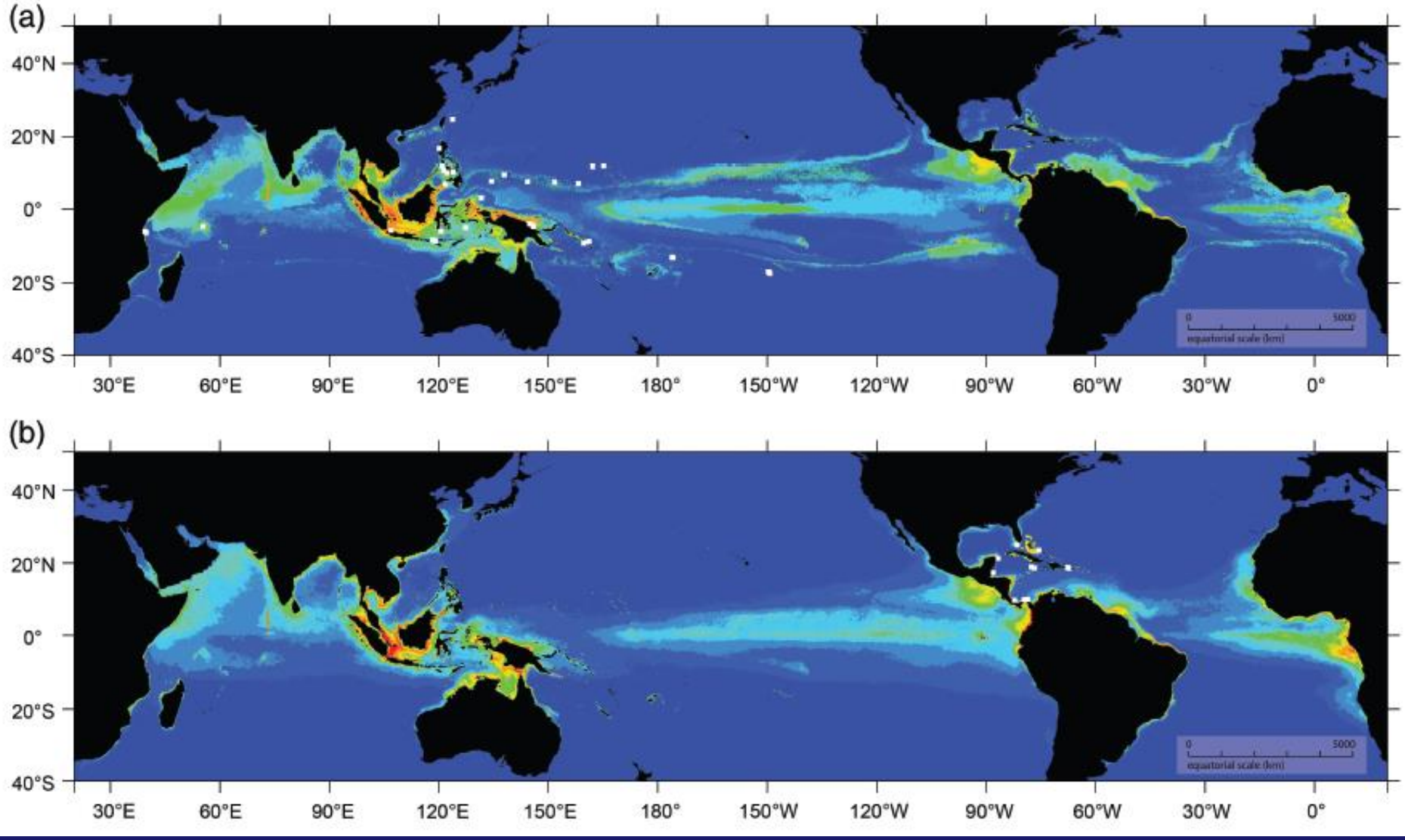


Fig. 3. Neighbor joining phylogram inferred from *rps*19–*rps*3 sequences of 60 *Halimeda* specimens (score = 1.13797). The outgroup was removed from the tree. Bootstrap proportions exceeding 50% are indicated at branches.



Verbruggen et al.2009

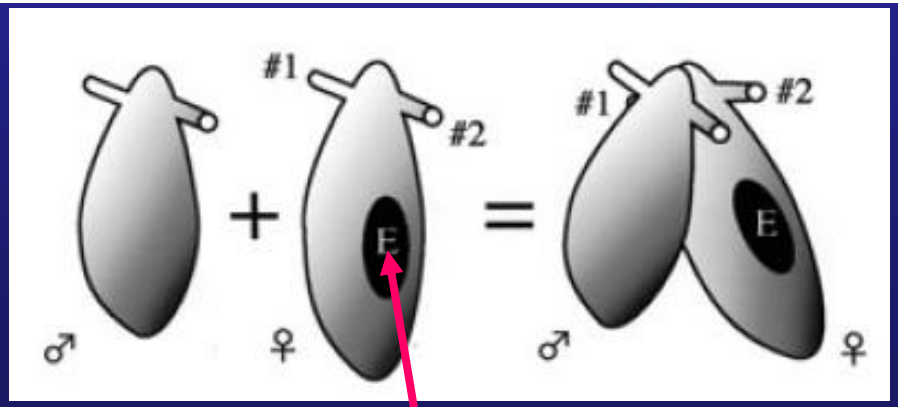


(a) Niche model of the exclusively Indo-Pacific species *Halimeda borneensis* indicating habitat suitability in some Atlantic regions. (b) Niche model of the exclusively Caribbean species *Halimeda simulans* predicting habitat suitability in several Indo-Pacific regions.

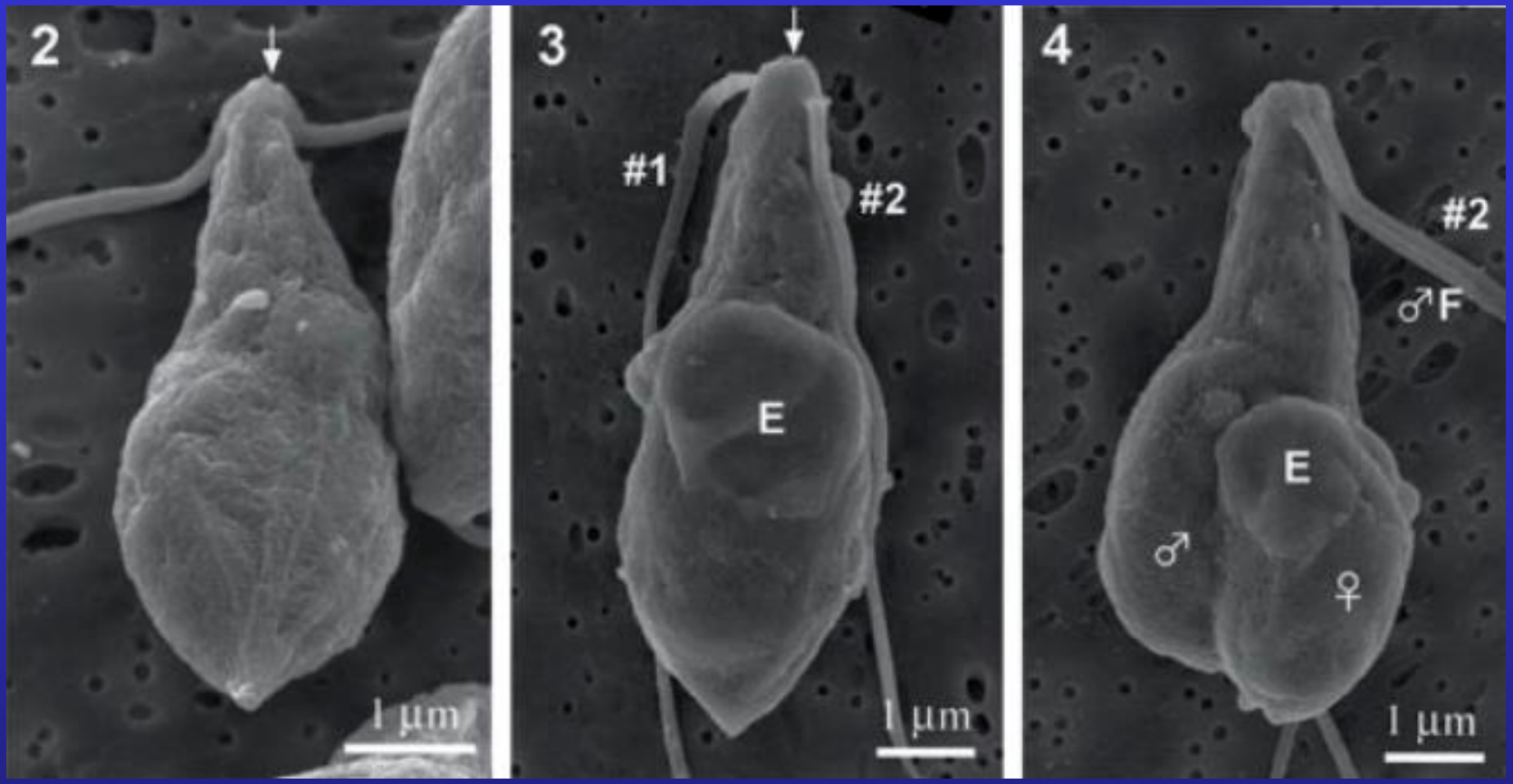
Caulerpa

uniaxial thallus

heteroplastidic



stigma

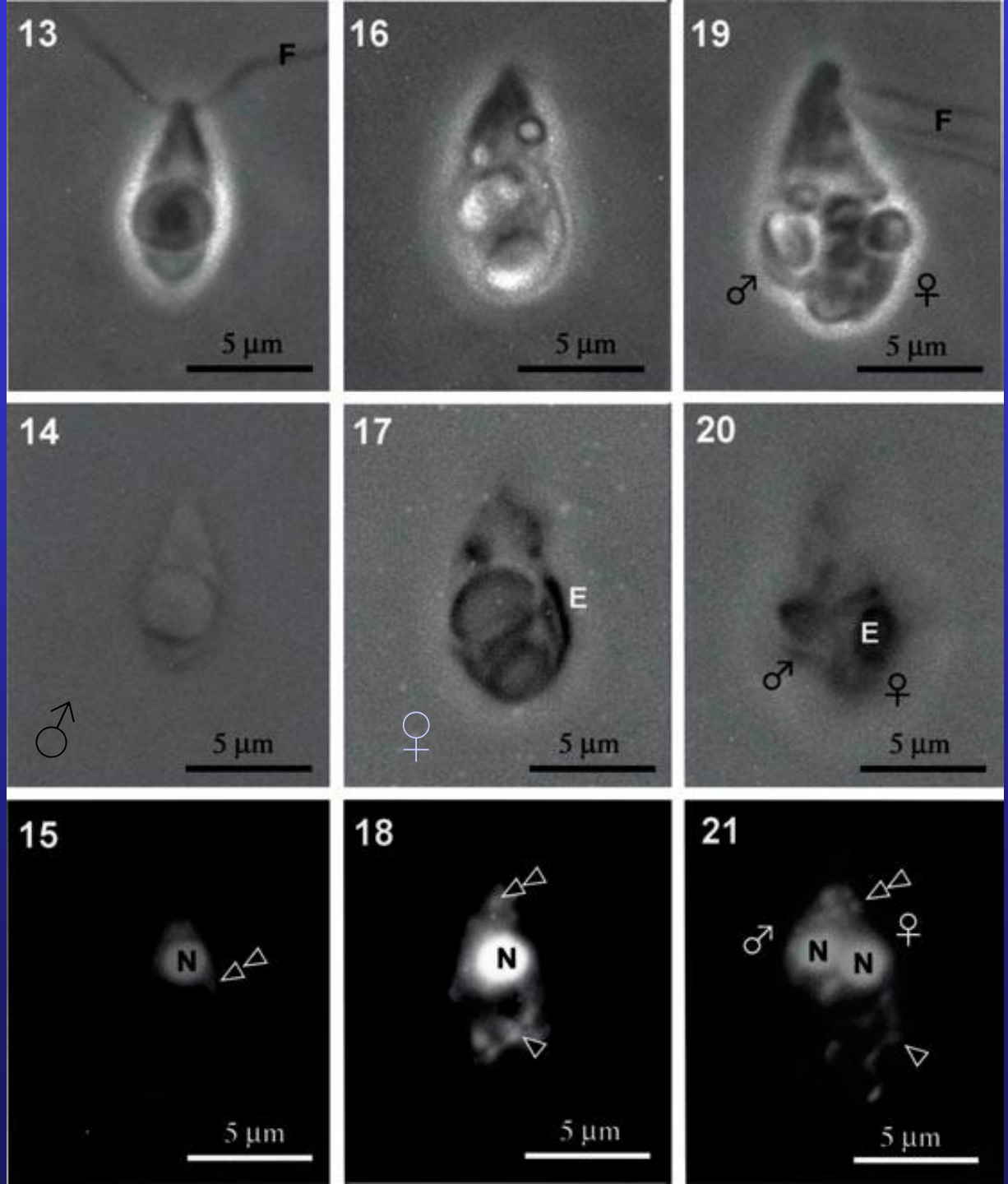


♂ ♀ planozygote

FE SEM gametes of *Caulerpa serrulata*

Miyamura et Nagumo, 2007

Inheritance pattern of chloroplast DNA is correlated with gamete types based on sex-specific arrangement of the cell fusion site in *Caulerpa*



Caulerpa taxifolia – „killer alga“ invasive species, succesfull along Mediterranean Sea shores and California

C. taxifolia ve Středozemním moři

https://www.youtube.com/watch?v=H0snJY3GHuU

https://www.youtube.com/watch?v=h-odcuD9BvA

https://www.youtube.com/watch?v=AajEW07qCGw

SCUBA DIVERS

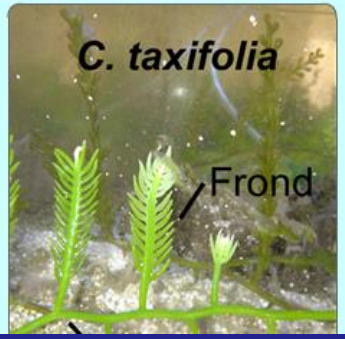
The help of SCUBA divers is critical to finding new infestations of *Caulerpa* in the wild. You can help by learning what *Caulerpa* looks like and keeping an eye out for it on your dives.

There are many species of *Caulerpa* that are used in the aquarium trade that could be released into the wild. Though many of them are tropical species, *Caulerpa taxifolia* and *Caulerpa racemosa* have both taken over large areas of seafloor in temperate waters due to their cold tolerance. Because *Caulerpa* is only native in the United States in Hawaii and some parts of Florida, divers should be on the lookout for all species of *Caulerpa*, which have many consistent features between them. All are bright green, have a runner by which it spreads, and upright fronds (Identification).

Caulerpa can grow on mud, sand or rock, and in shallow or deep water. It grows low to the bottom, typically six to 12 inches high. It can grow in calm or rough waters. Fragments of *Caulerpa* do not float and it is therefore not likely to be found on docks, boat hulls or in ballast water.

If you think you have found *Caulerpa* in the wild:

- Note as much information as possible (location, depth, bottom type, size of the patch).
- Record the location with a GPS if possible.
- Take a photo if possible.
- Collect a small piece and press it flat in newspaper (be careful - even a 1-mm piece broken off can start a new infestation).

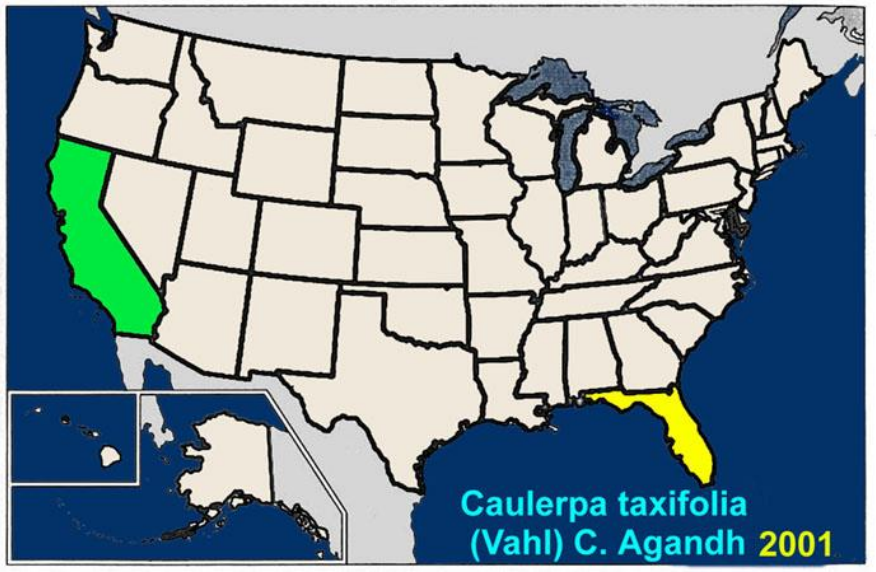


Report the sighting and it will be investigated.

Click here to report a sighting of *Caulerpa* in the wild or in a pet store.

Click here to learn how to identify *Caulerpa*.

Thank you for your help. New infestations will most likely be detected by the SCUBA diving ...unity. A map of surveys done for *Caulerpa* in California is available here.



jediný výrazný nepřítel *Elysia subornata* – sap sucking slag - hermaphrodite

Caulerpa **predators are being recruited to attack the invader.** ***Oxynoe*** **(left) has a partial shell to protect its reproductive organs and digestive glands, while** ***Elysia*** **(right) is shell-free. With each feeding on only 5 centimeters of frond per day, Meinesz estimates that efficient control would require more than 1,000 slugs per square meter of** ***Caulerpa.***

Chemical: Colonies of *C. taxifolia* that were discovered in Southern California were eradicated by covering and sealing them with PVC tarpaulins and injecting liquid chlorine underneath. Subsequent treatments at another location used solid chlorine formulations (Anderson & Keppner, 2001). Costs of the Southern Californian eradication were $US2.33 million from 2000-01 for control and monitoring (Carlton, 2001), with an ongoing annual surveillance cost of $US1.2 million until 2004 (Anderson, 2004).
Application of coarse sea salt at a concentration of ~50kg/m² has been used with moderate success in Australia, eradicating *C. taxifolia* from an area almost 5200 m² in one case, although in another case an area of 3000 m² showed a reduction in algal density but eradication was not achieved. The use of this method in the cooler months, when *C. taxifolia* naturally dies back, was recommended. Salting has so far only been succesfully used on soft sediments in water <6m in depth (Glasby *et. Al*, 2004).

Physical: Simply covering *C. Taxifolia* colonies with black PVC plastic was found to be reasonably successful in Croatia. A total area of 512 m² was treated, with either no or sporadic regrowth occurring after treatment (McEnnulty *et. Al*, 2001).
Manual removal by scuba divers was successful in eradicating a small patch of *C. Taxifolia*, around 3.4 m², in the French Mediterranean. The use of a suction pump to remove all fragments has also met with moderate success in other areas. Clearance rates for manual removal are from <1 m² to ~3 m² per diver per hour (McEnnulty *et.al*, 2001).

Biol Invasions (2015) 17:2717–2728
DOI 10.1007/s10530-015-0908-1

ORIGINAL PAPER

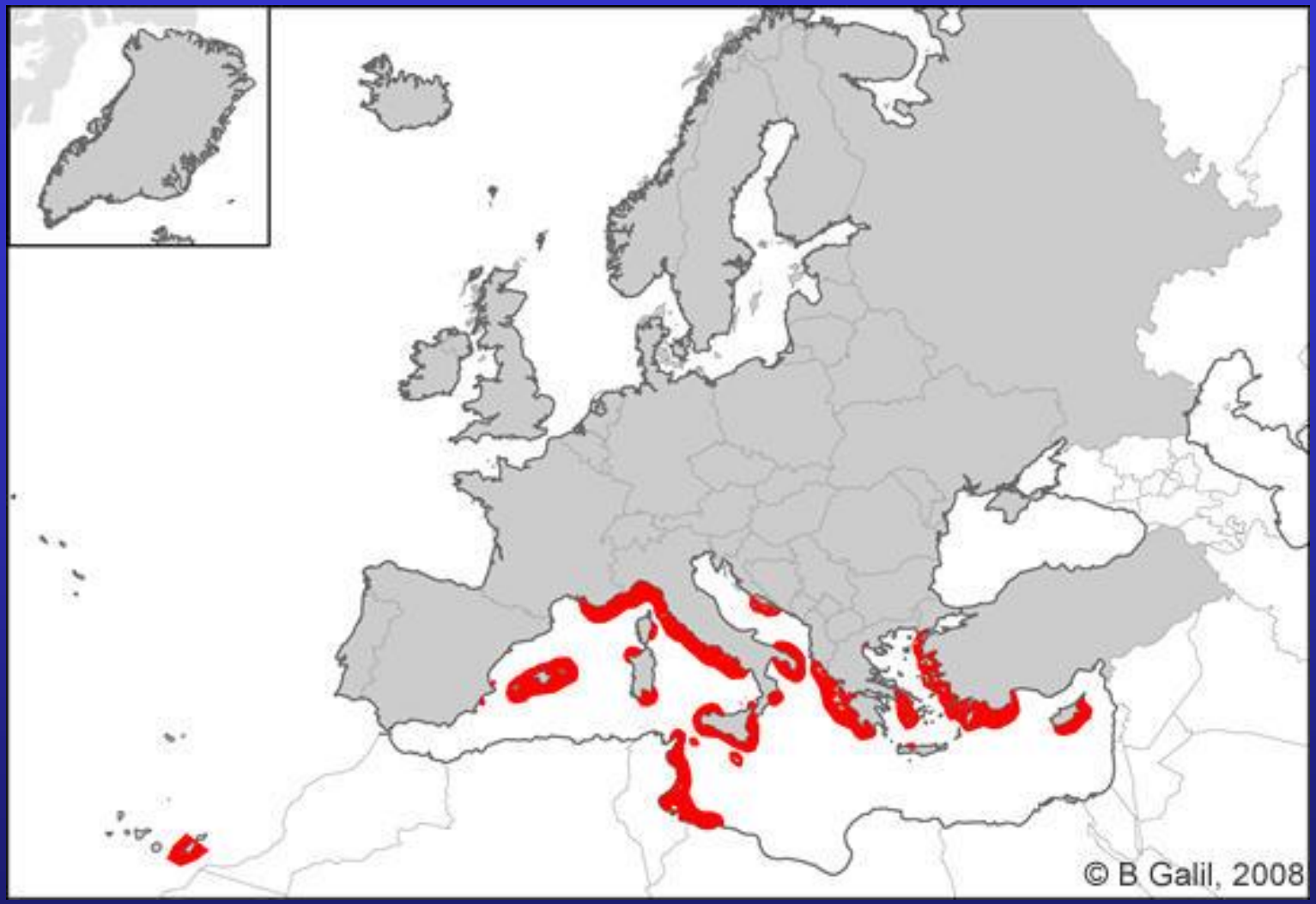
A tale of two invaders: divergent spreading kinetics of the alien green algae *Caulerpa taxifolia* and *Caulerpa cylindracea*

Monica Montefalcone · Carla Morri · Valeriano Parravicini · Carlo Nike Bianchi

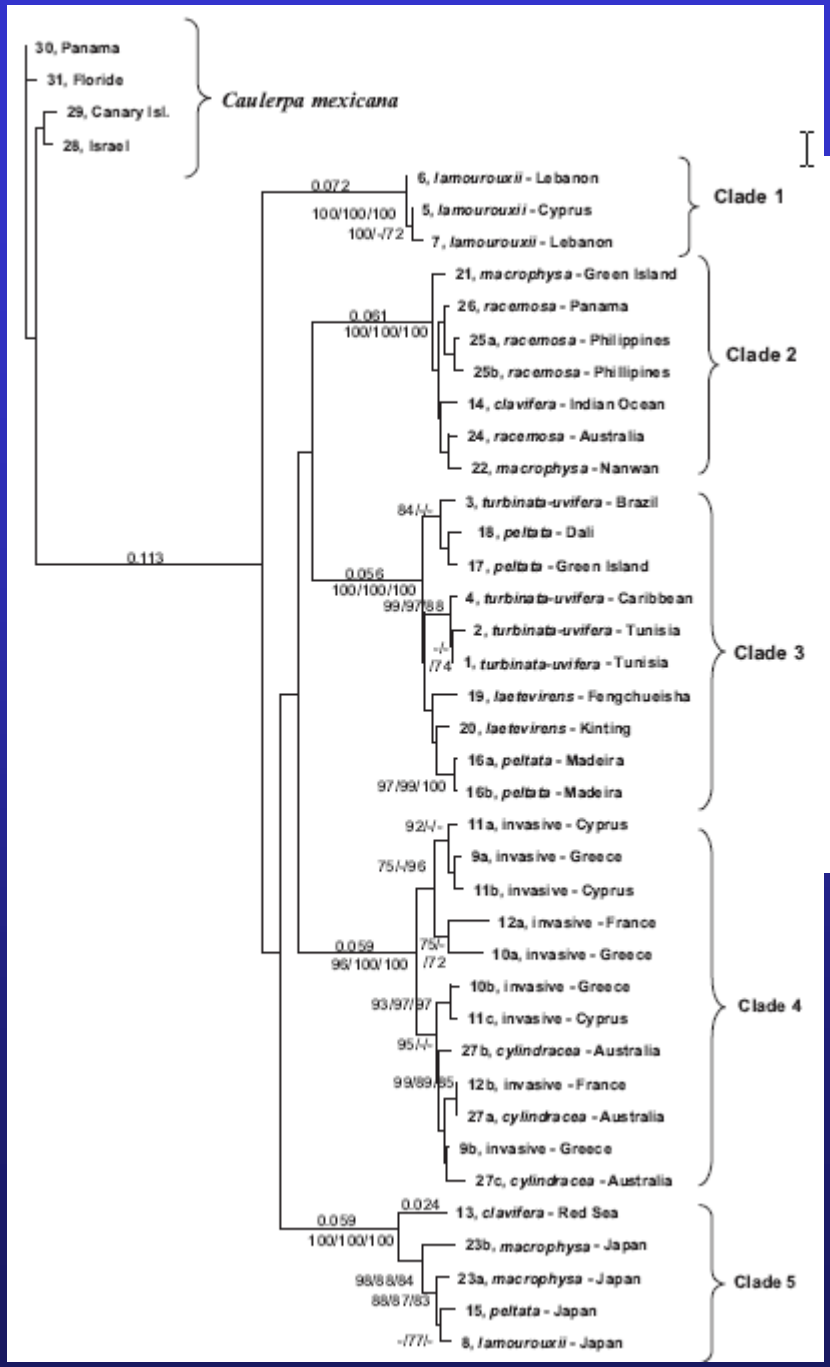
impressive expansion
phase from 1984 to 2000 but then,
its dispersal
rate showed lower than that
predicted and the species
did not persist in areas formerly
colonized

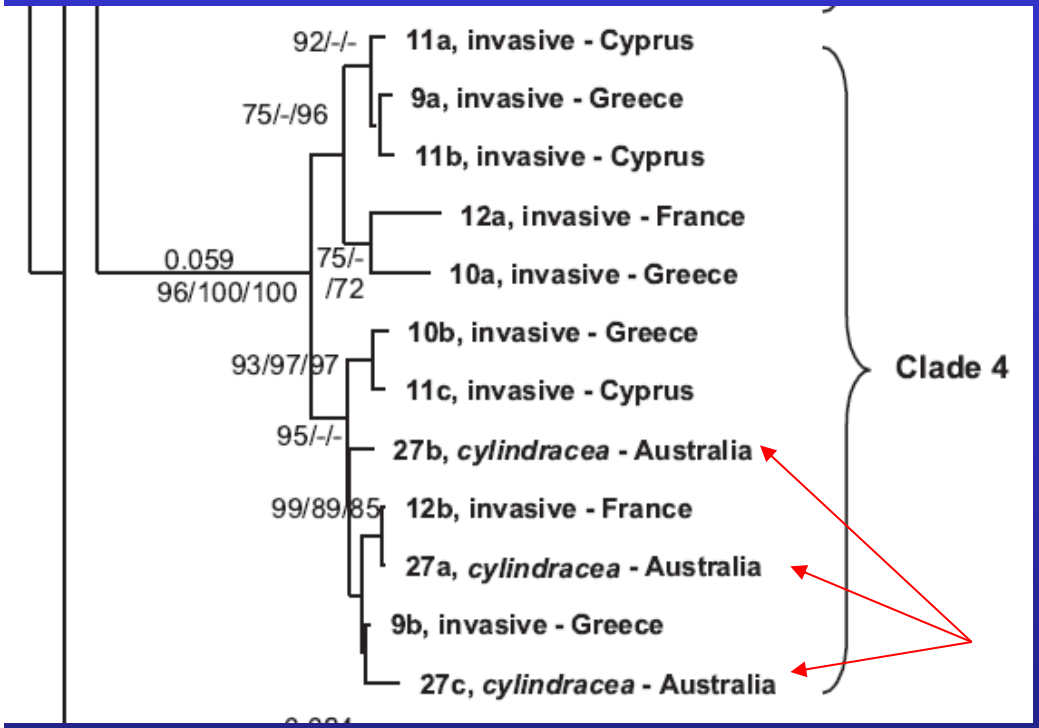
Invasion of *Caulerpa cylindracea*

first found in Libia
1990



invazní varieta - do r. 2002 11 států, všechny velké ostrovy, daleko rychlejší invaze než u *C. taxifolia*

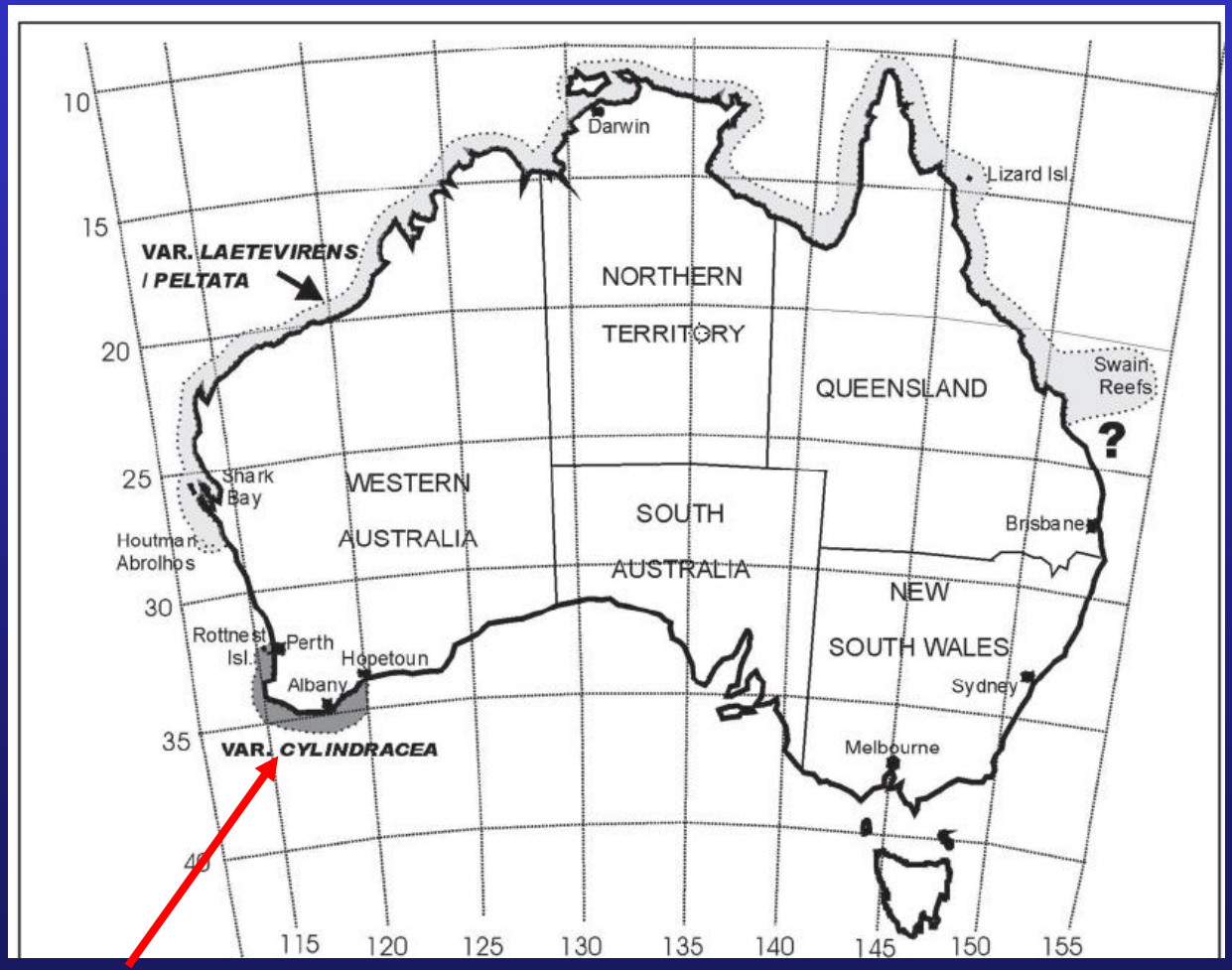




Neighbour-joining treeof ITS1, 5.8S and ITS2 sequences from a range of Caulerpa racemosa isolates, with C. mexicana as theoutgroup. NJ/MP/ML

Verlaque et al. 2003

invaze je výsledkem recentní introdukce, identifikovali „invazní varietu“ - totožná s *Caulerpa cylindracea* -endemit jihozápadní Austrálie



Caulerpa lentilifera

pěstována na Filipínách, v Japonsku – písčito-bahnitá dna mělkých chráněných zátok, antibakteriální a antifungální účinky (fukoidany) proti vysokému tlaku a revmatismu

Lato

Umi-budō served Okinawan style

Lato, the strange sea salad Coron, Philippines

Coron, Filipíny - kmen Tagbanua se živí pěstováním a prodejem *C. lentilifera*

http://www.lemanger.fr/en/index.php/lato-the-strange-sea-salad/?lang=en

The big baskets will travel to Manila by cargo. There, their commercial value will increase dramaticaly. In Coron, the buyers get the lato from the Tagbanua for 8 pesos a kilo. In Puerto Princesa, the capital of the province of Palawan, the prices are already as high as 100 pesos a kilo. In Manila, it’s even more.

Some of the lato will be sold here, at the Coron Town market.
Women will then prepare small bags of 1 kg of lato that will be sold 10 pesos, so about 15 euro cents.

Dasycladales

Základní charakteristika:

• stélka vždy sifonální, většina zástupců inkrustována $CaCO_3$

• strukturní polysacharidy BS – β-1,4 mannany (celulóza dominantní složkou gametangiálních cyst)

• gamety 2 bičíky, CCW orientace bičíkových bazí,

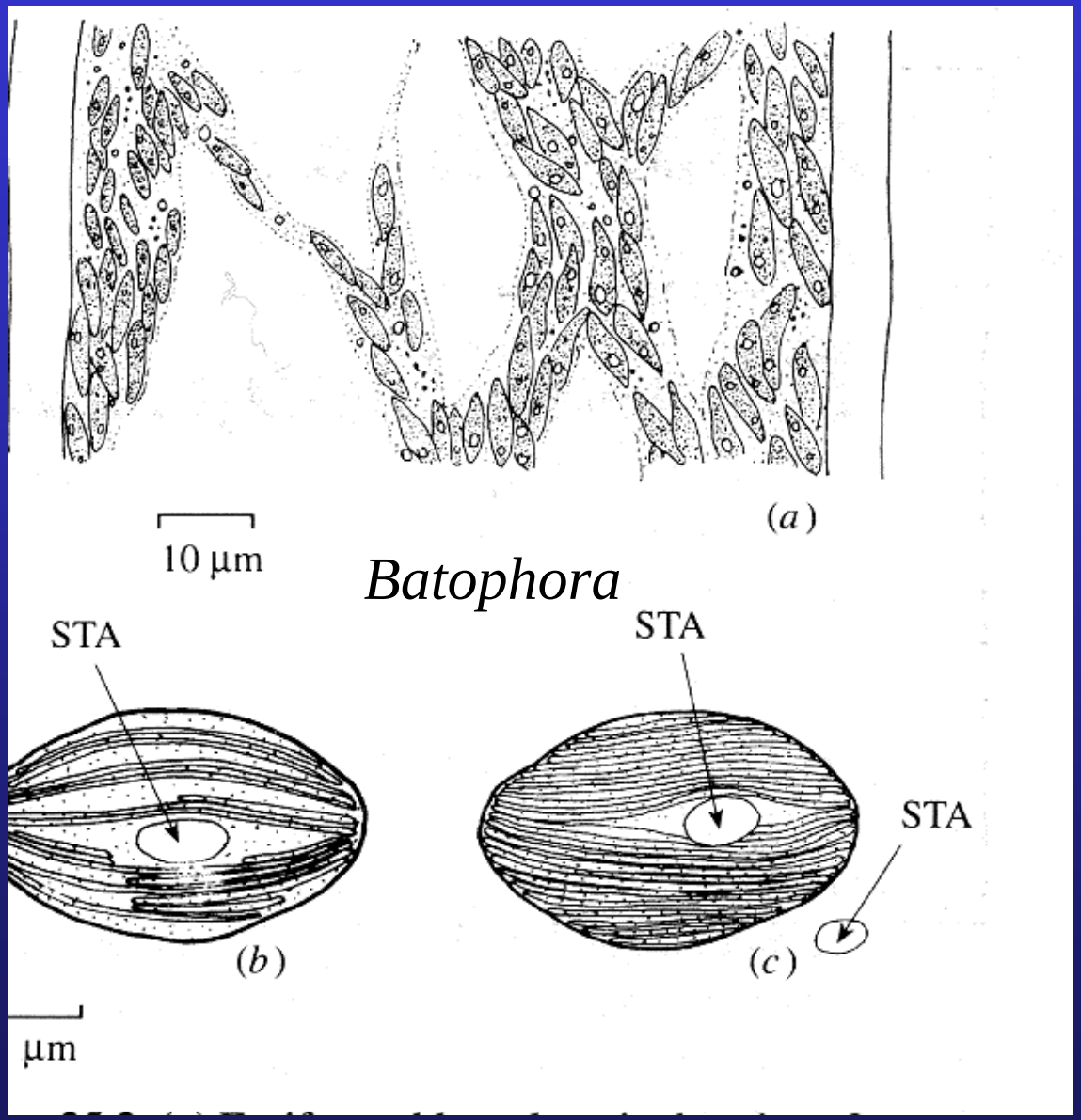
• mitóza uzavřená, perzistující telofázové vřeténko, jádro v telofázi – činkovitý tvar; mitóza acentrická

• četné chloroplasty v nástěnné vrstvě cytoplasmy, bez pyrenoidu, obsahují zrna fruktanu, zásobní polysacharidy i v cytoplasmě

• proudění cytoplasmy v podélné ose stélky, proudem cytoplasmy unášena jádra a chloroplasty

• haplontní životní cyklus (haploidní gametofyt, mikroskopická zygota s obrovským diploidním jádrem), izogamie

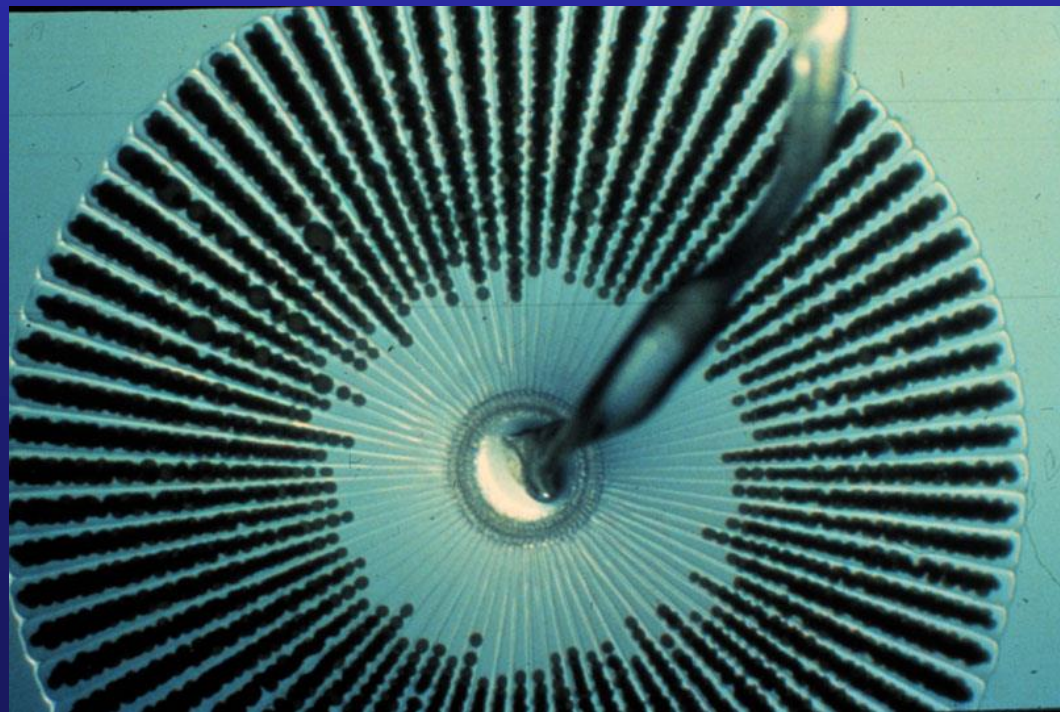
Chloroplasts



Acetabularia

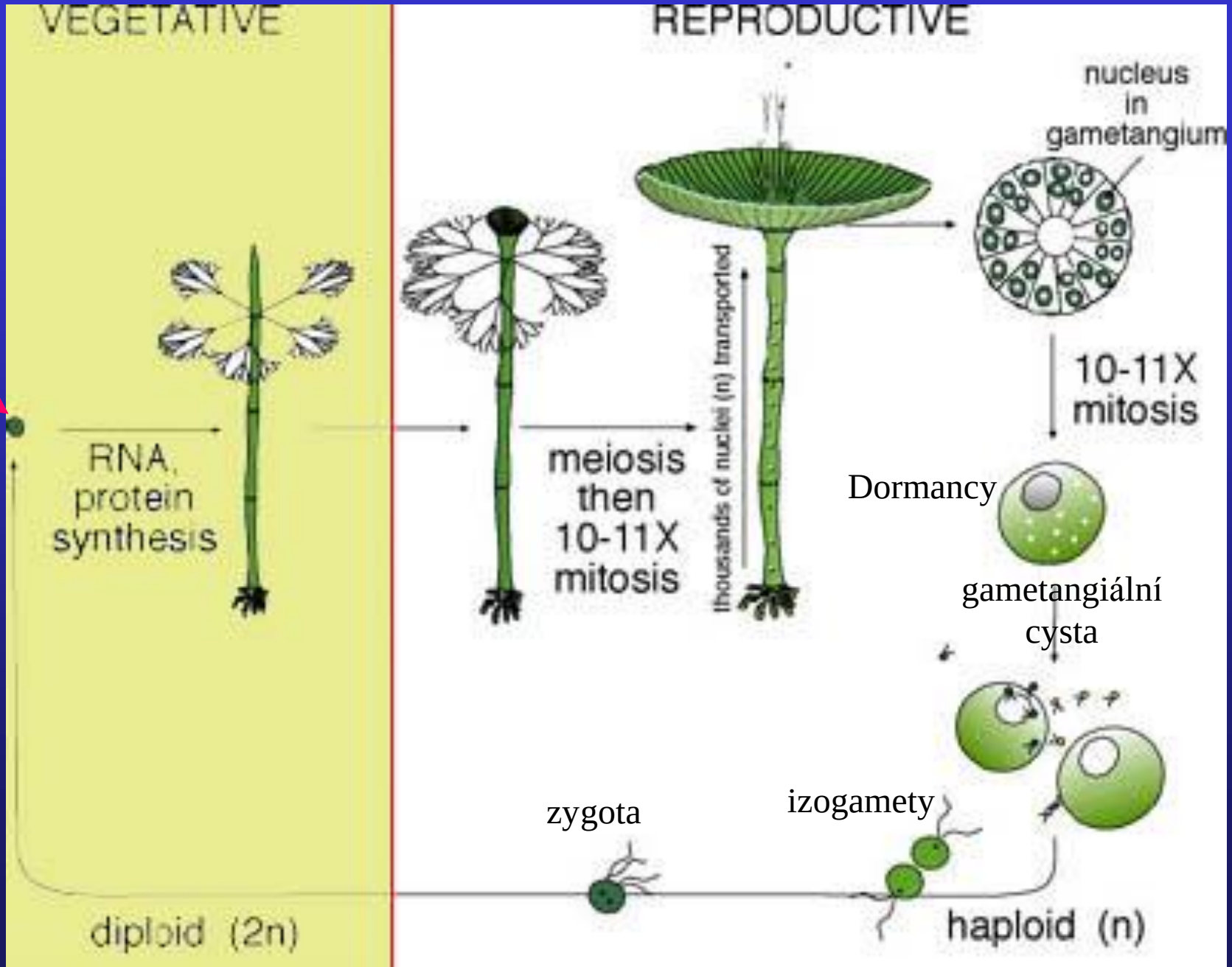
Cymopodia

Acetabularia



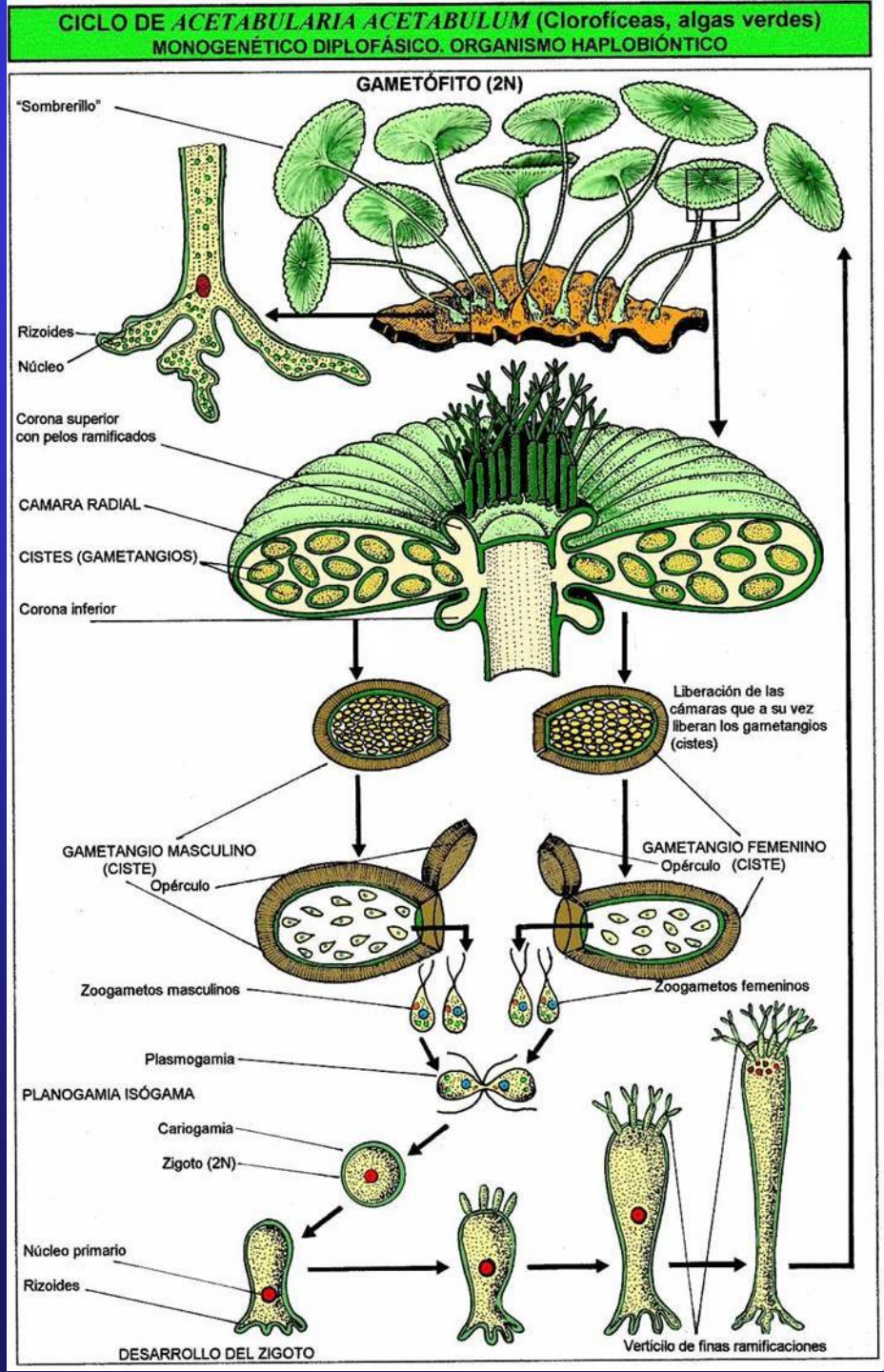
Acetabularia mediterranea displays two circadian rhythms: (*a*) in photosynthetic capacity and (*b*) in chloroplast shape. In the middle of the light period, the greatest volume of O_2 is evolved; at the same time, the “mature” chloroplasts have an elongated shape. In the middle of the dark period, the smallest volume of O_2 is evolved and the shape of the chloroplasts is more spherical.

Acetabularia

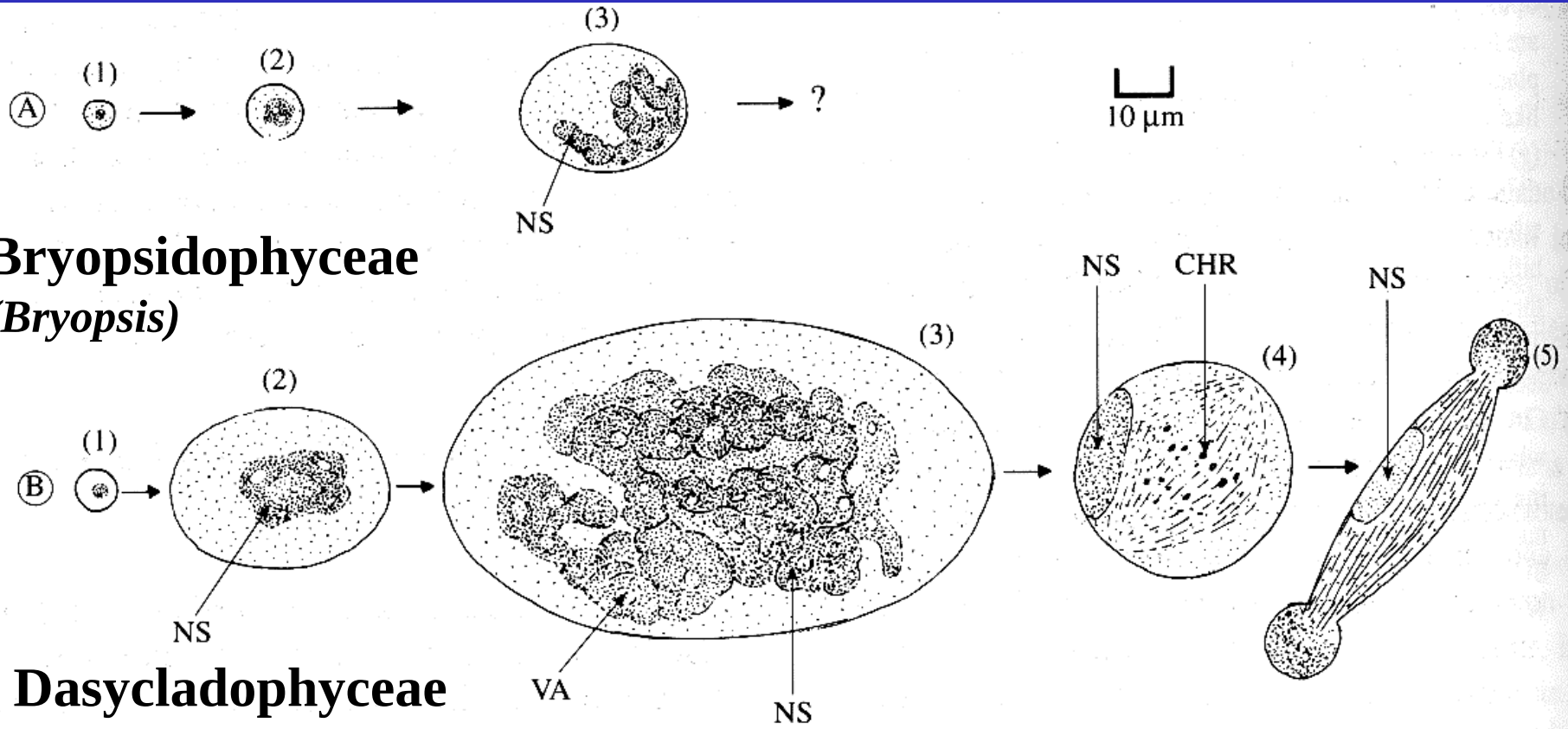


Acetabularia

whorls of hairs



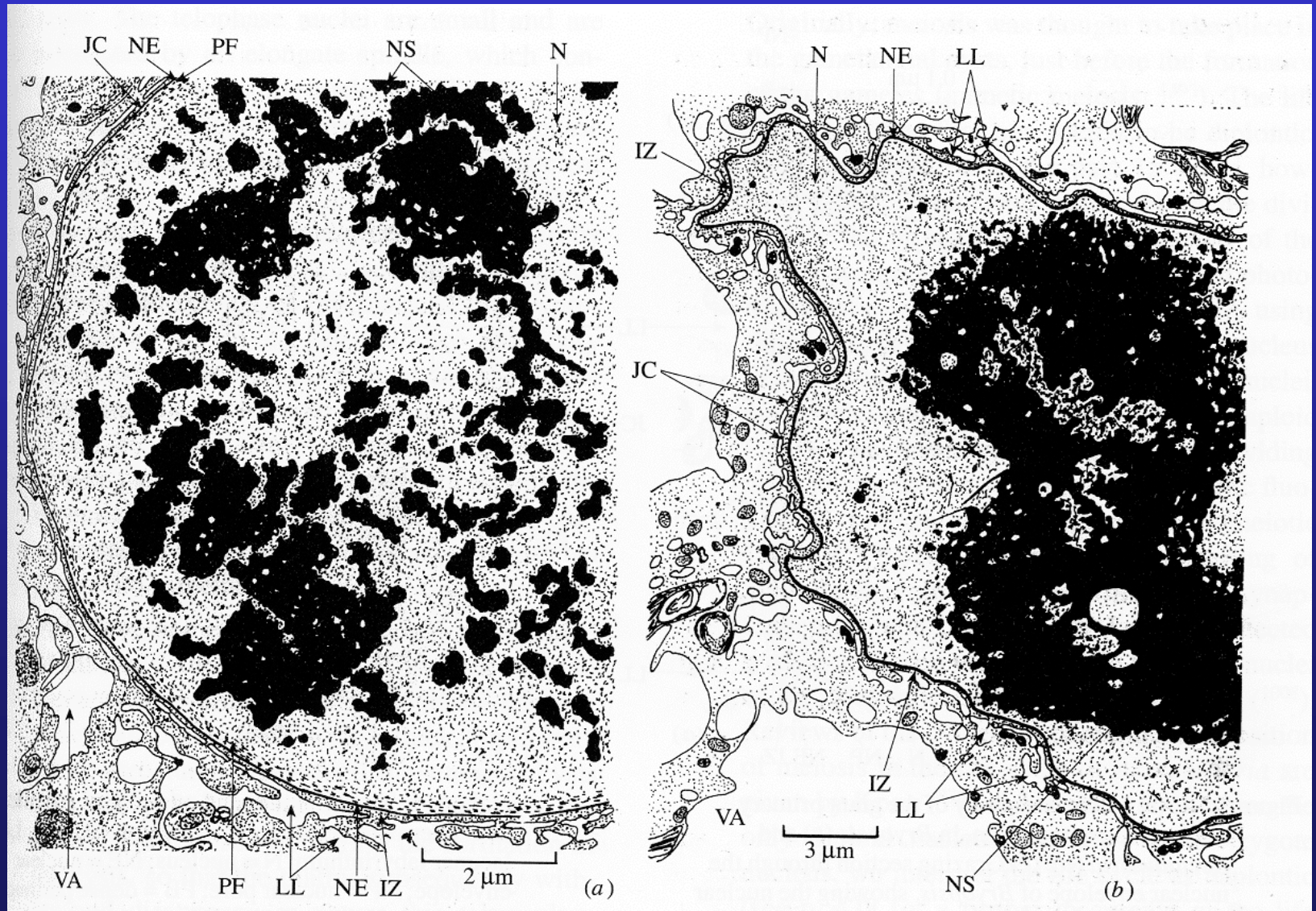
The giant primary nucleus Bryopsidales x Dasycladales



Ultrastructure of the primary nusleus

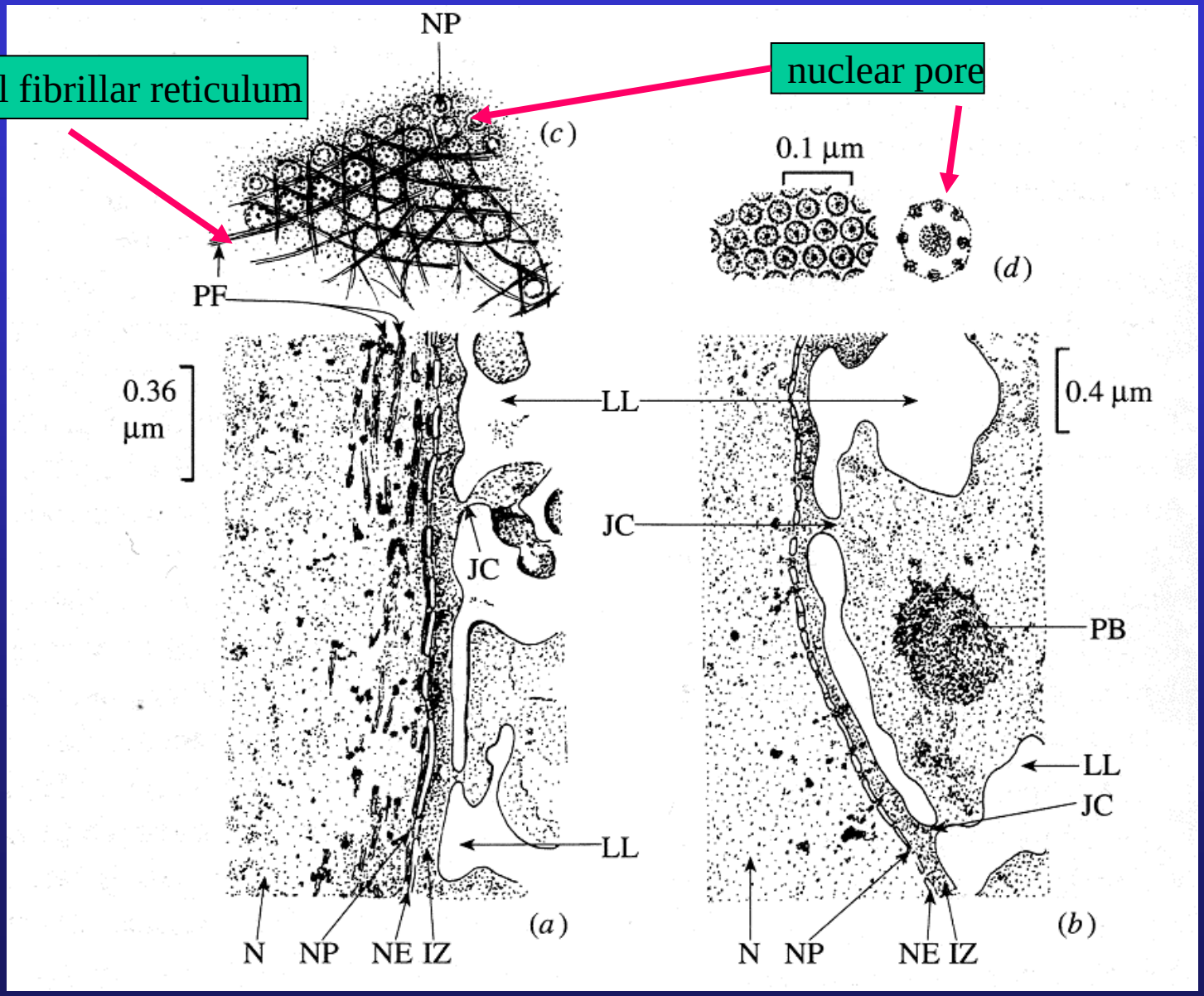
intermediate zone (IZ) lacunar labyrinthum (LL) junction channel (JC)

Bryopsis



Acetabularia

Ultrastruktura primárního jádra



Bryopsis

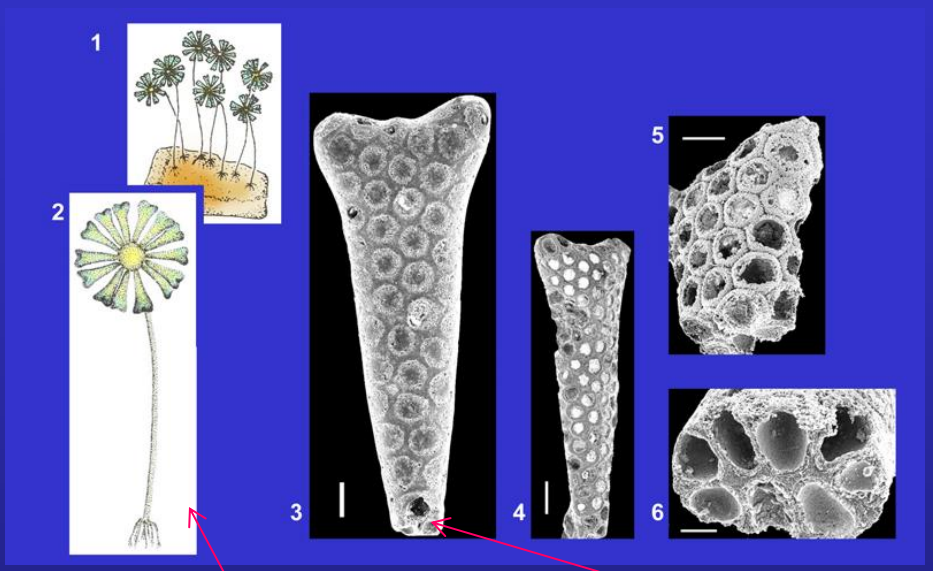
Acetabularia

Cenozoic Dasycladales (Eocene)

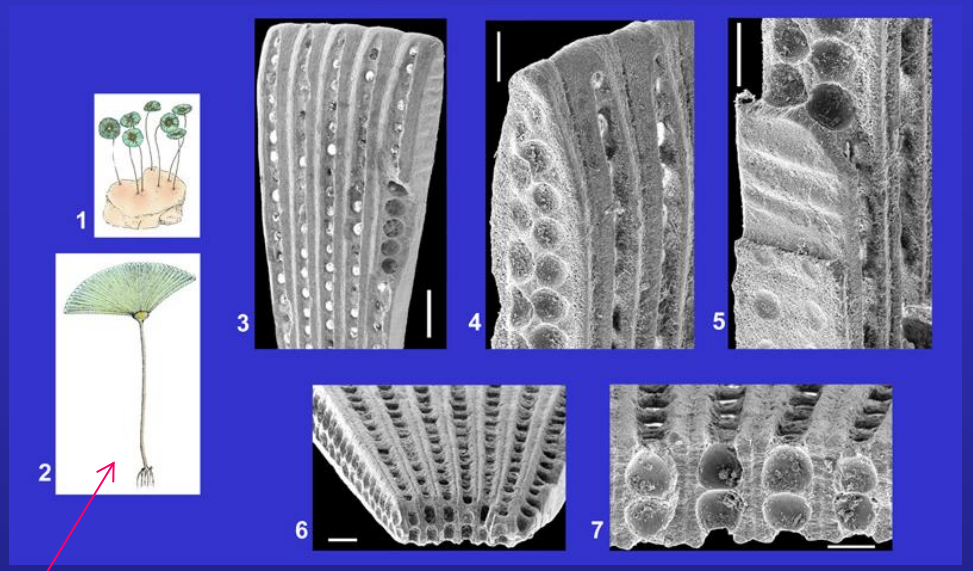
discovered in the Cenozoic strata of the French sedimentary basins

http://paleopolis.rediris.es/cg/CG2009_SP01/

Acicularia cornigera L. & J. MORELLET, 1922



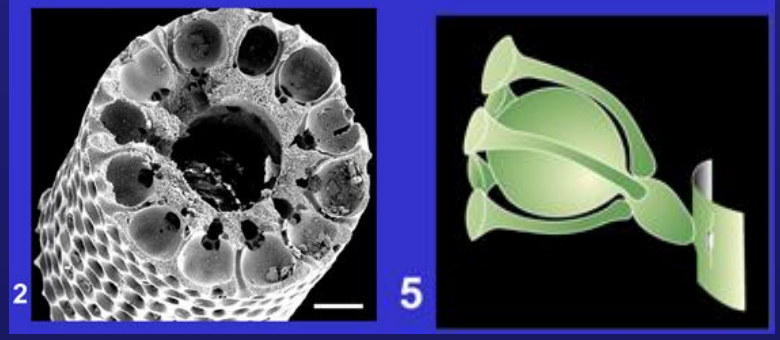
Acicularia costulata GÉNOT, 1987



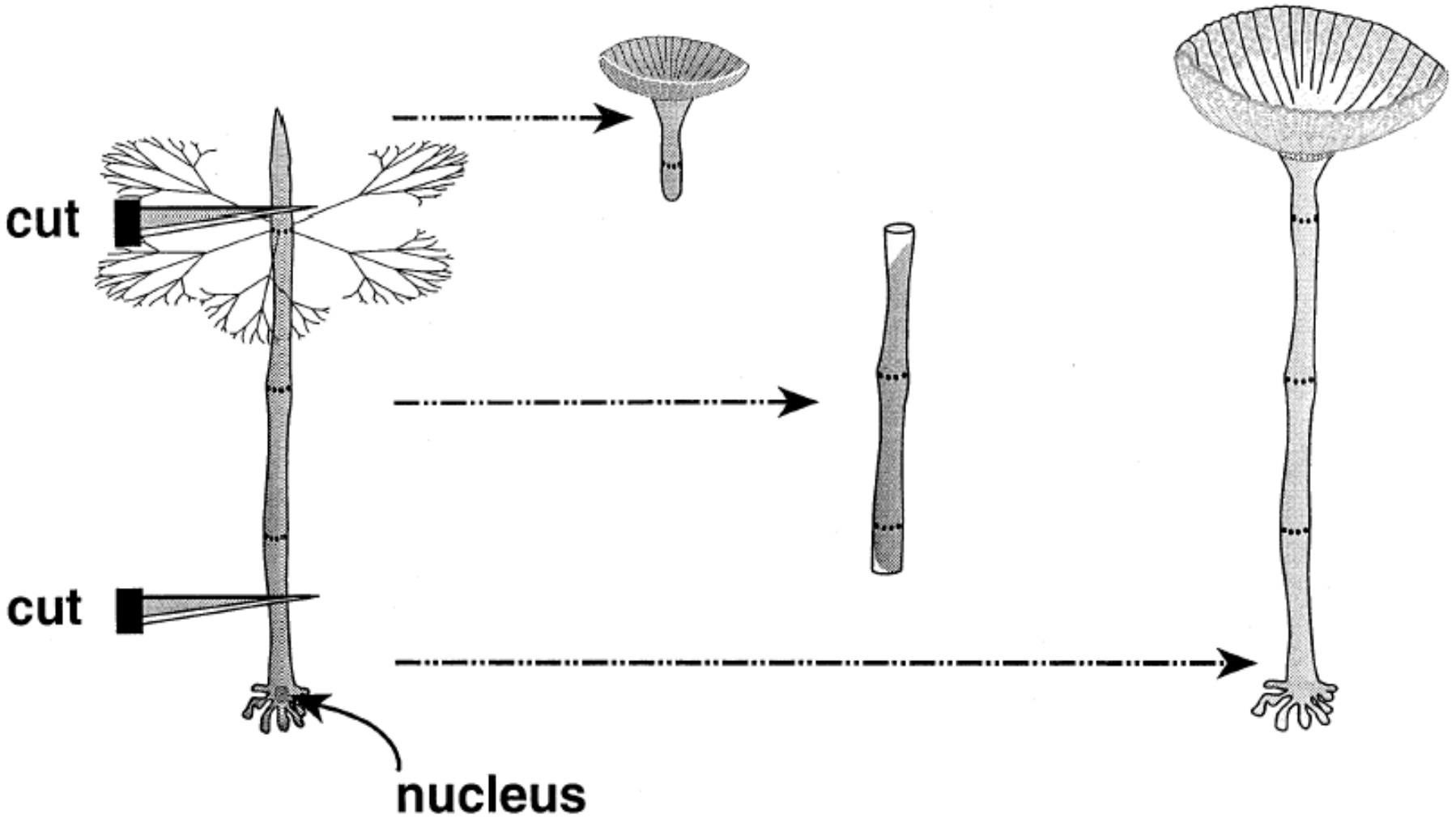
External view of a gametophore

Reconstruction of a living representative

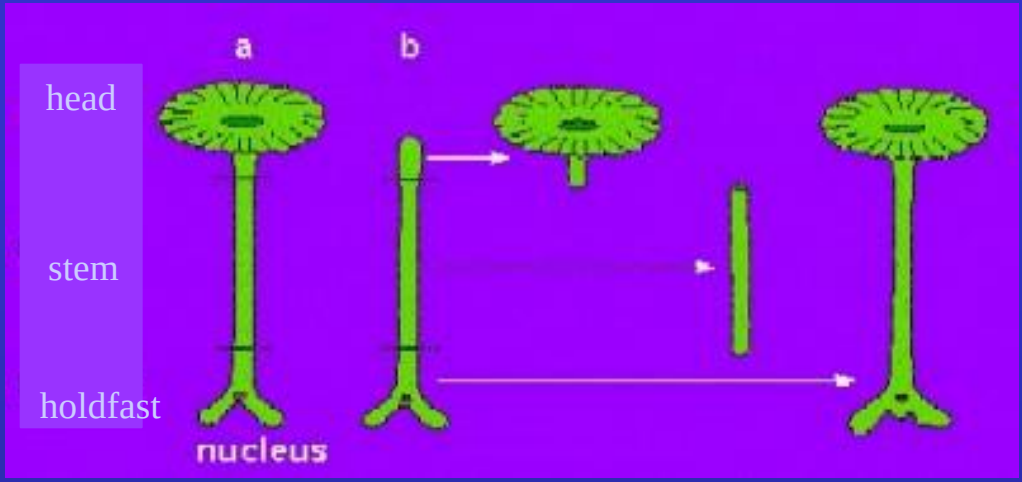
Cymopolia turgescens GÉNOT, 1987



Amputation experiments

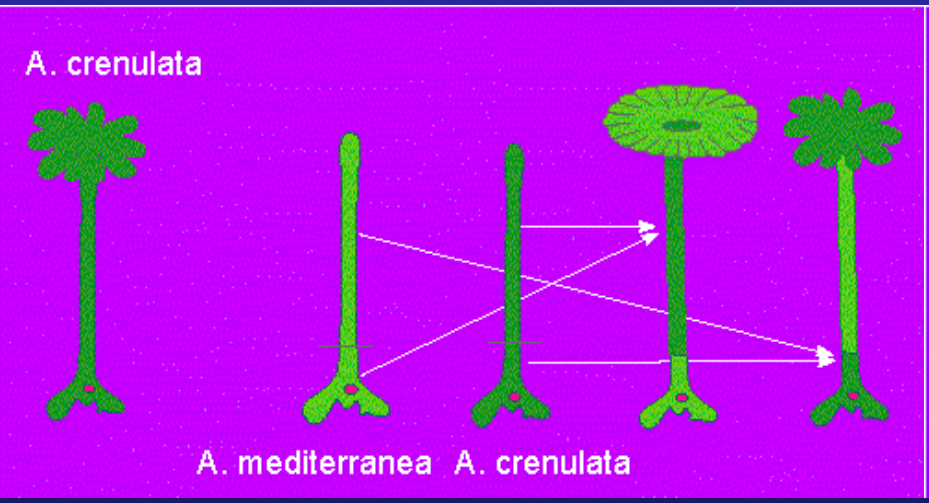


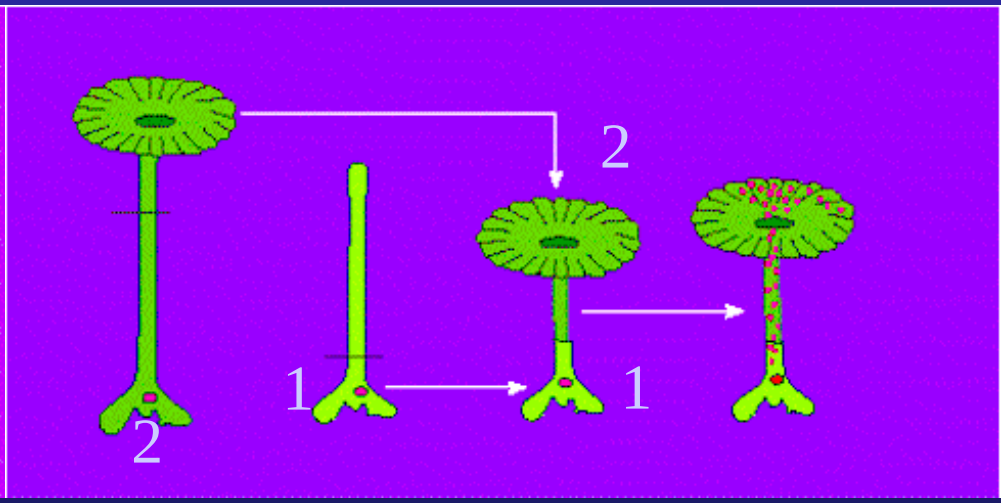
Hämmerlings reconstruction experiments in the 1930‘s



Regeneration of the head only occurs in the section carrying the nucleus --> thallus regeneration is under the control of the nucleus

Morphology of the head: stems of two different species with different head morphology are plugged on nucleus-carrying holdfast of two species; the new head corresponds to the species of the nucleus --> head morphology is controlled by the nucleus





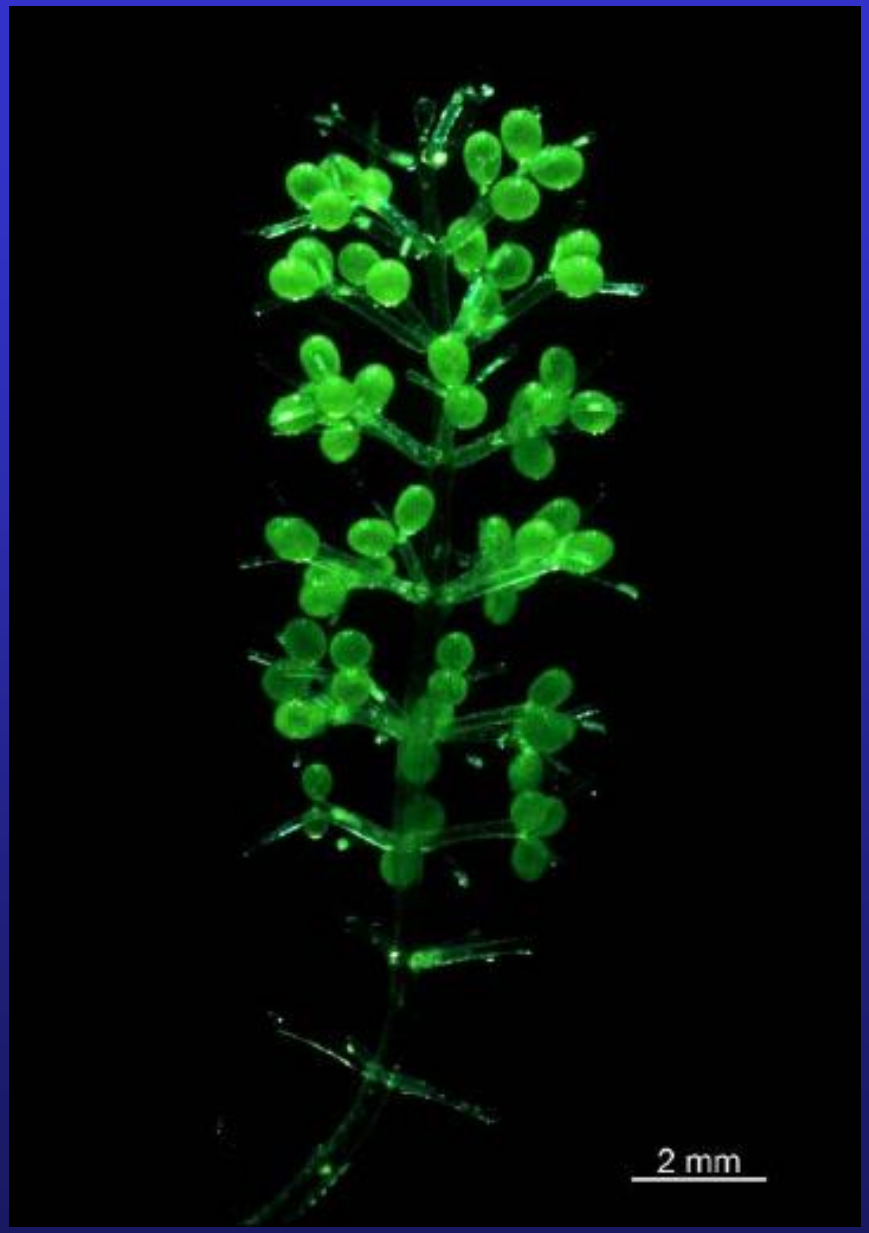
Gamete production: stem and head of species 2 are plugged on holdfast of species 1; micronuclei and gametes are formed within the new (species 2) head by the nucleus of species 1

shallow tropic lagunes

Batophora

many whorls of repeatedly branched laterals. Gametangia at nodes of the laterals. Thalli are not calcified. Shallow lagoones. More tolerant to salinuty changes.

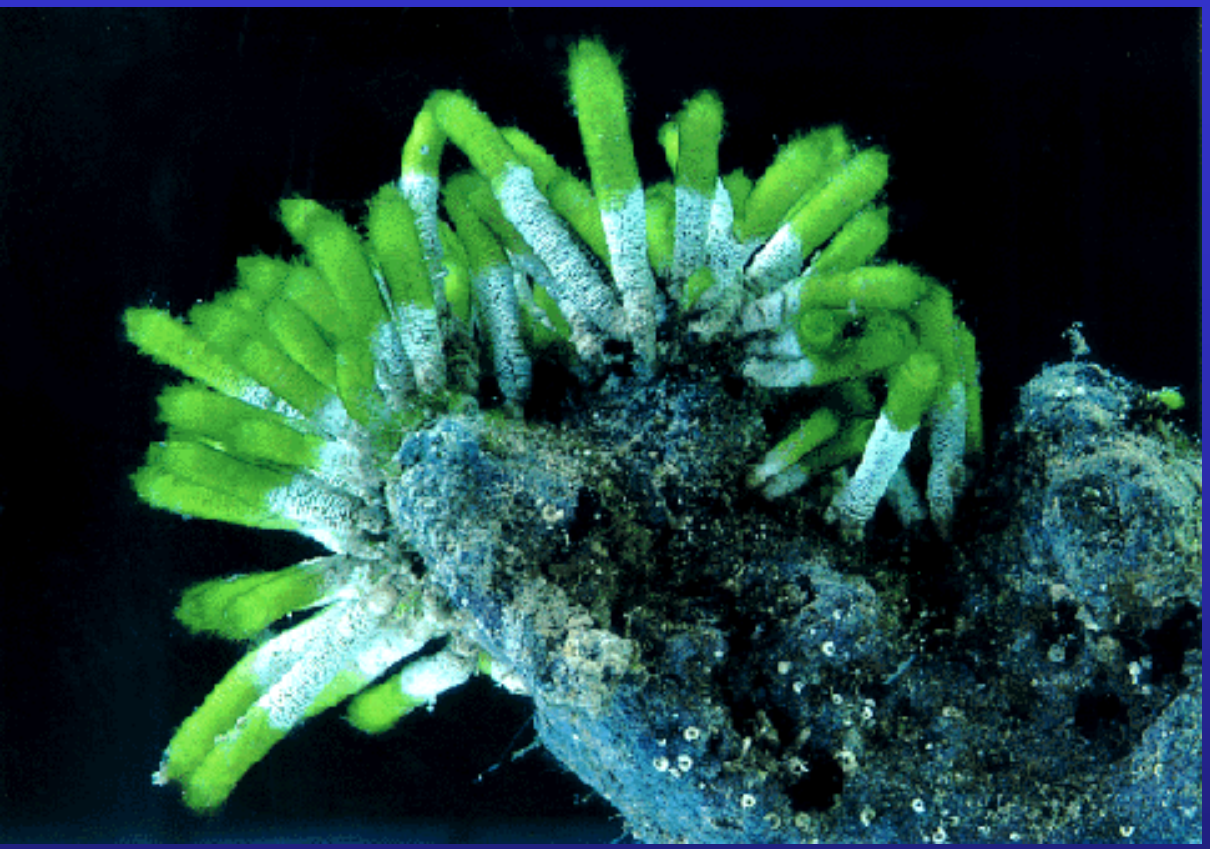
Batophora



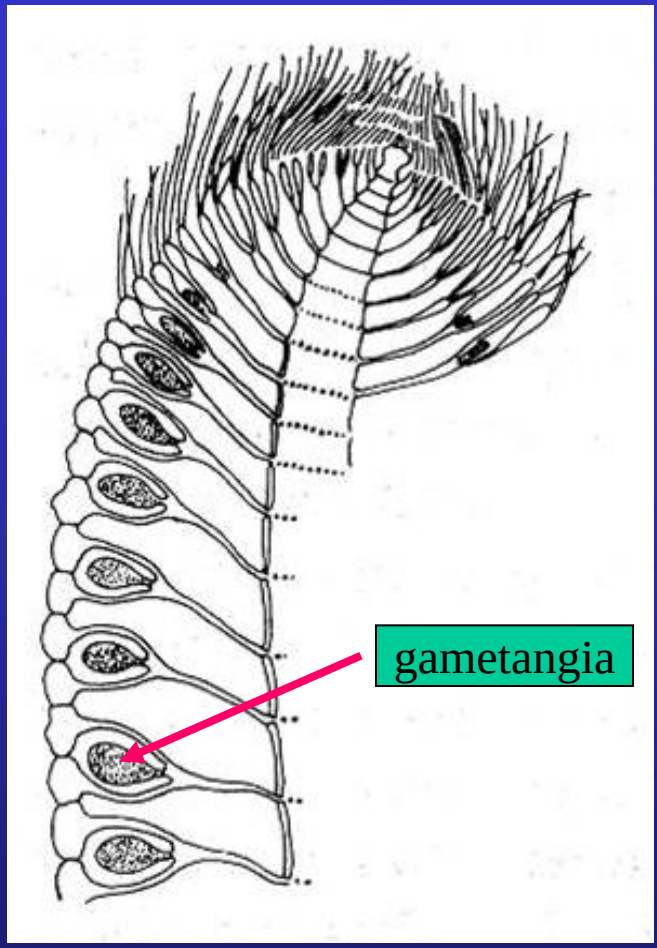
upper part of a gametophore-bearing cell

part of a field-collected cell with gametangia-bearing gametophores

Neomeris



Vertikální osa končí chomáčem světle zelených vlasovitých výběžků. Stélka silně inkrustována



Podélný řez vrcholem stélky

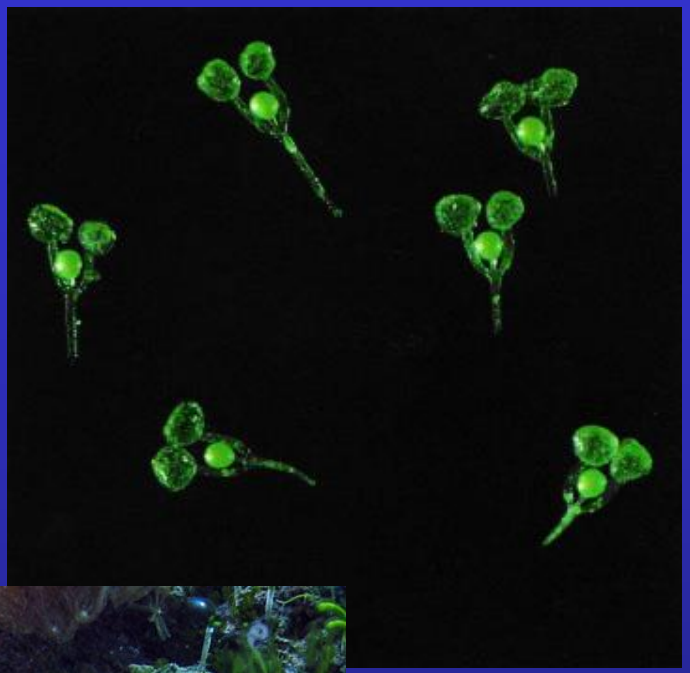
Photo-Atlas of living Dasycladales S. Berger, 2006

Neomeris

young cells

three cells

Neomeris

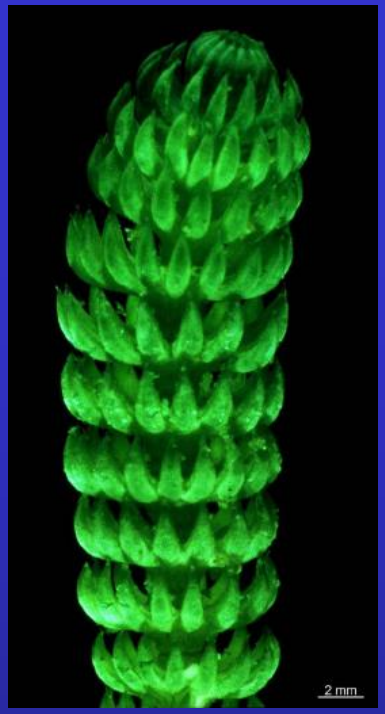


isolated laterals with gametophores

mature cell as it appears in its natural habitat

Halicoryne

Halicoryne wrightii



upper part of a cell with gametangia growing in its natural habitat

Dense coverage of Halicoryne spicata

Halicoryne

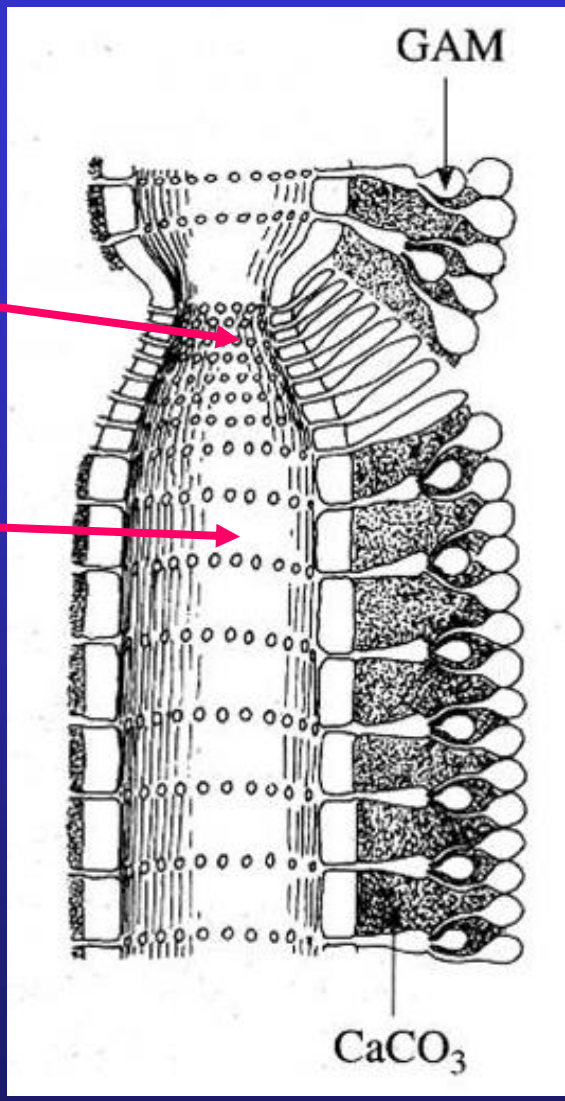
young cell

upper part of a cell, in part with gametangia

Cymopolia (Roman mythology – daughter of Neptune)

internodium

nodus



thalli have repeatedly branched construction. Branching occurs in a single plain.

Calcified segments vs. uncalcified joints

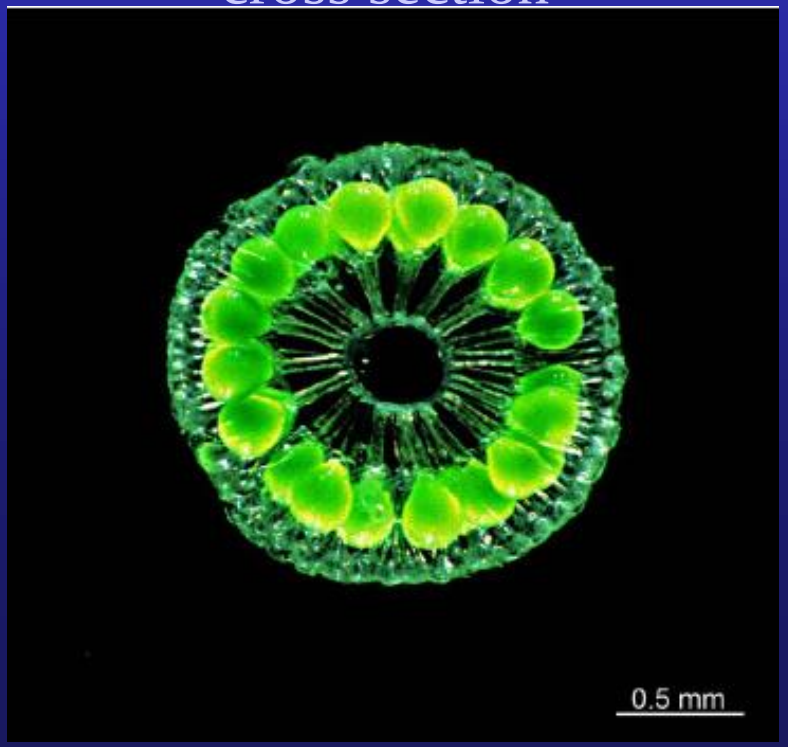
Cymopolia

Cymopolia van-bosseae

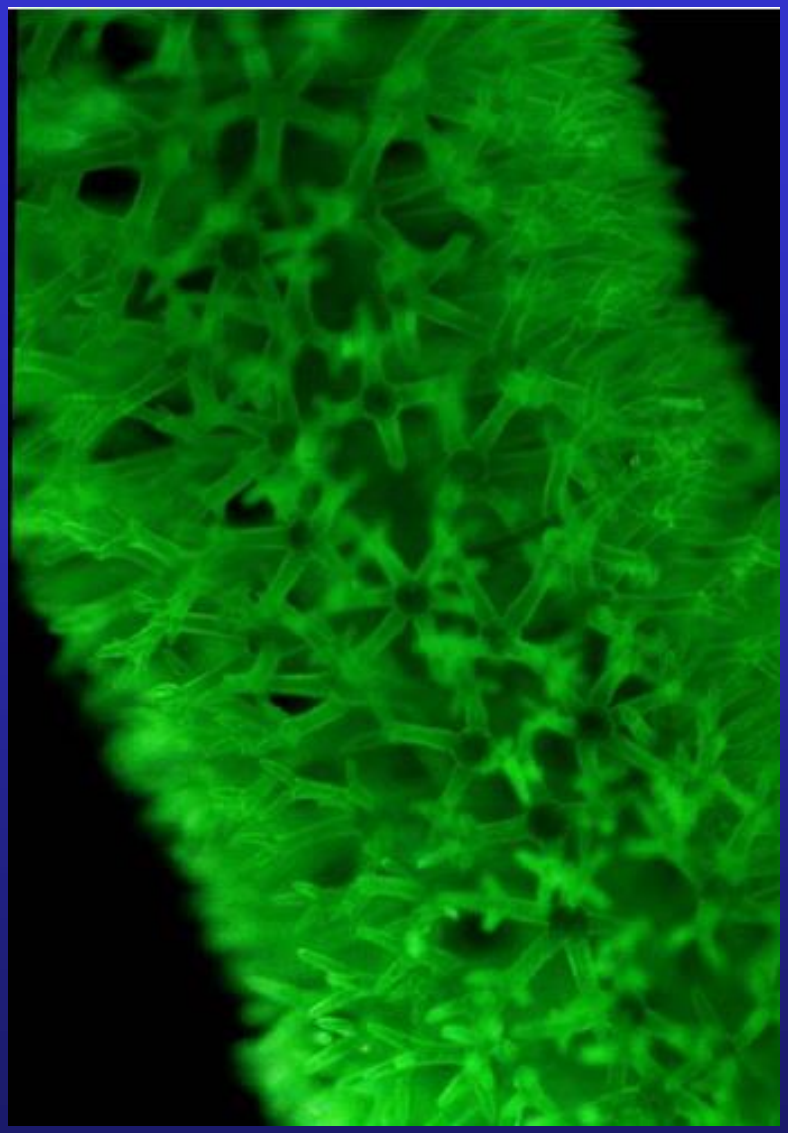
C. barbata

longitudinal section

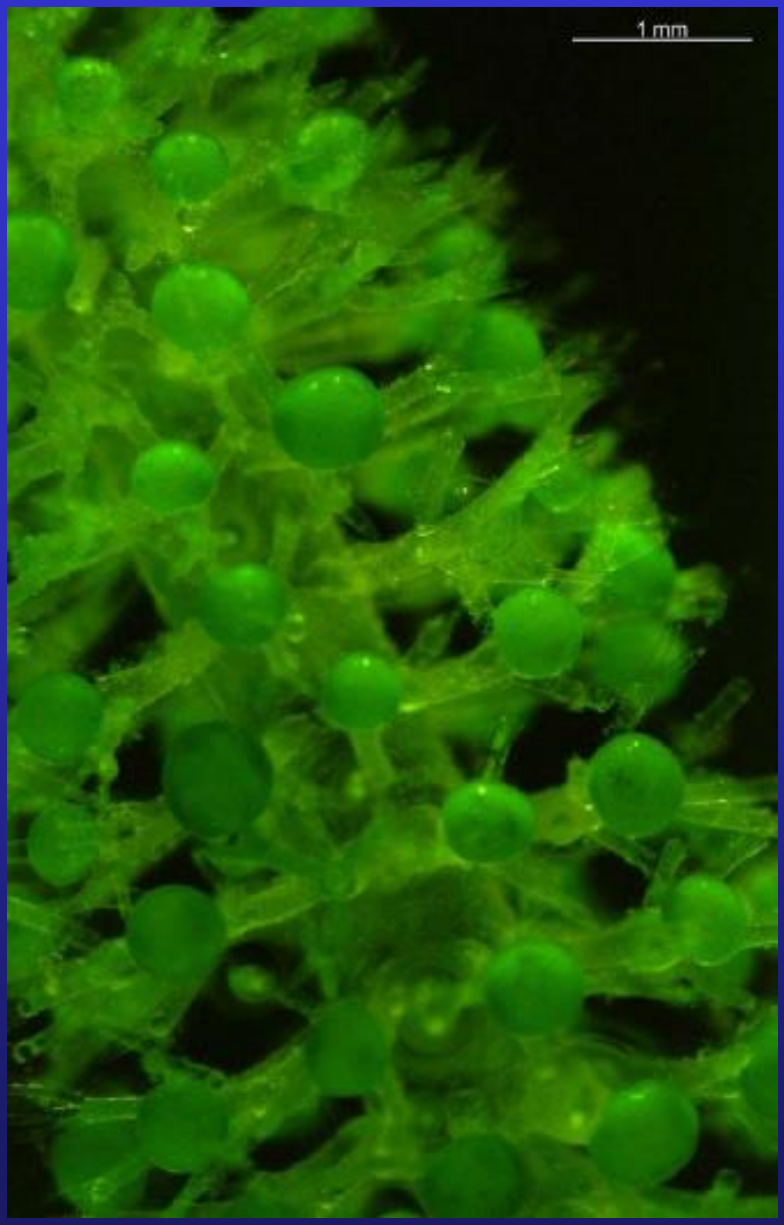
cross section



Dasycladus

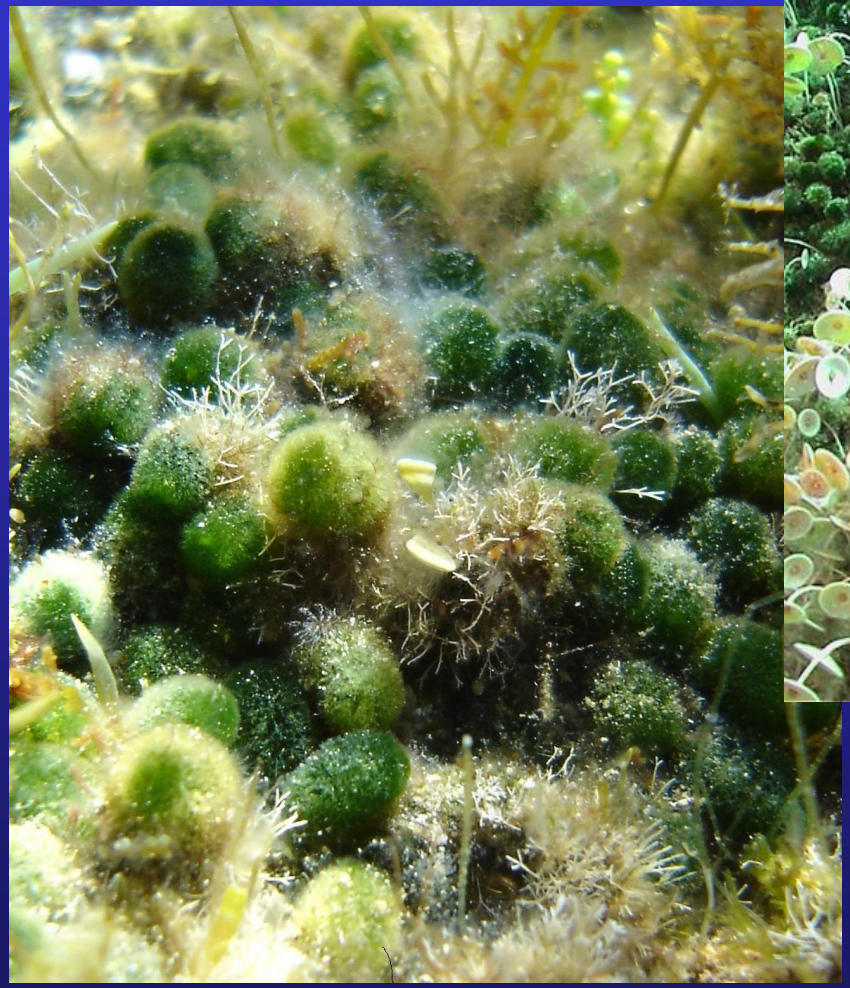


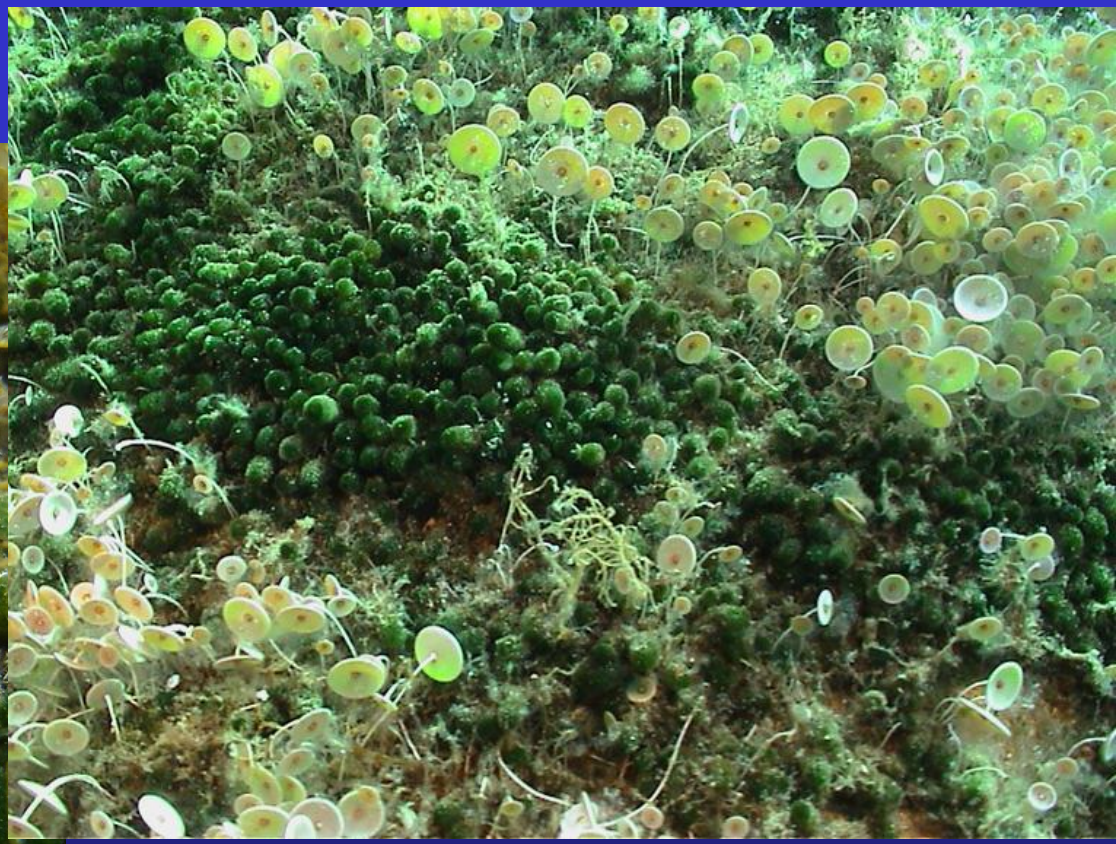
beginning of gametangia formation



part of a mature cell

Dasycladus





Dasycladus

Acetabularia

algaeBASE

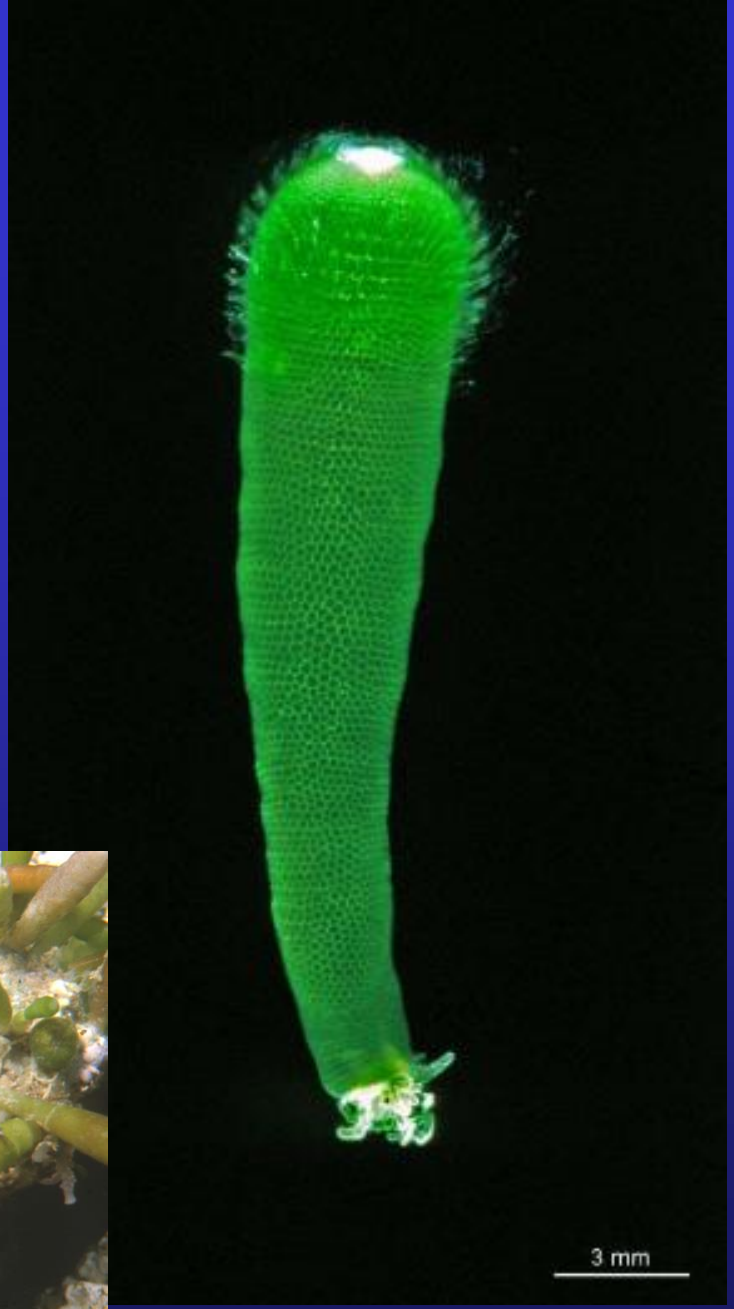
Bornetella sphaerica

Bornetella nitida

1 mm

Bornetella oligospora

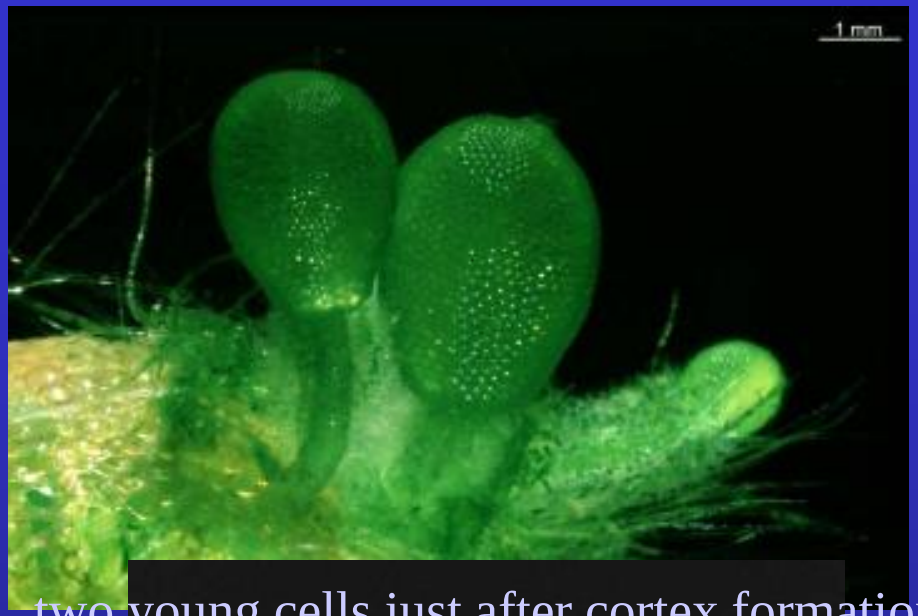
young cell



immature cell

Bornetella sphaerica

young cells

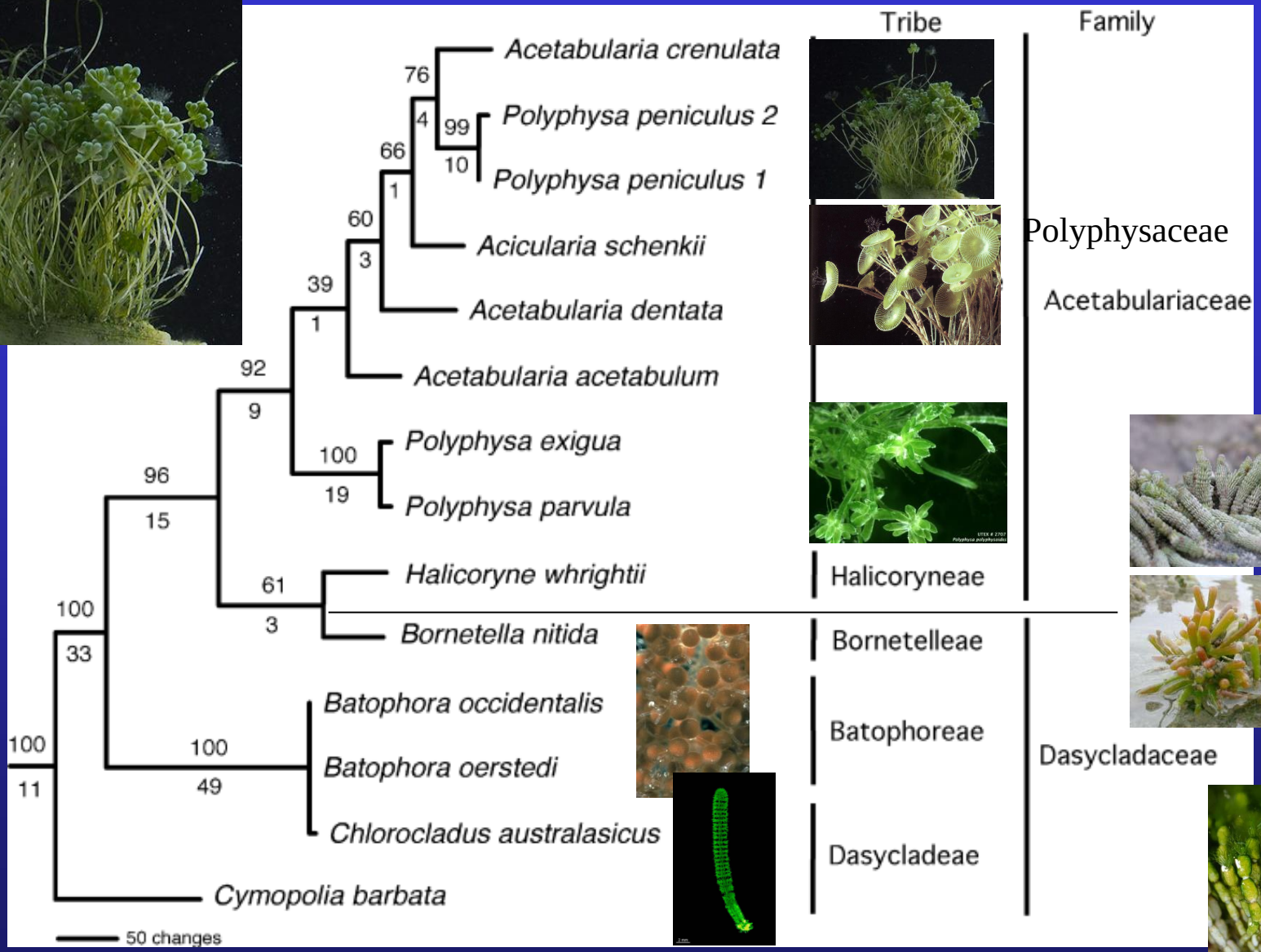


two young cells just after cortex formation

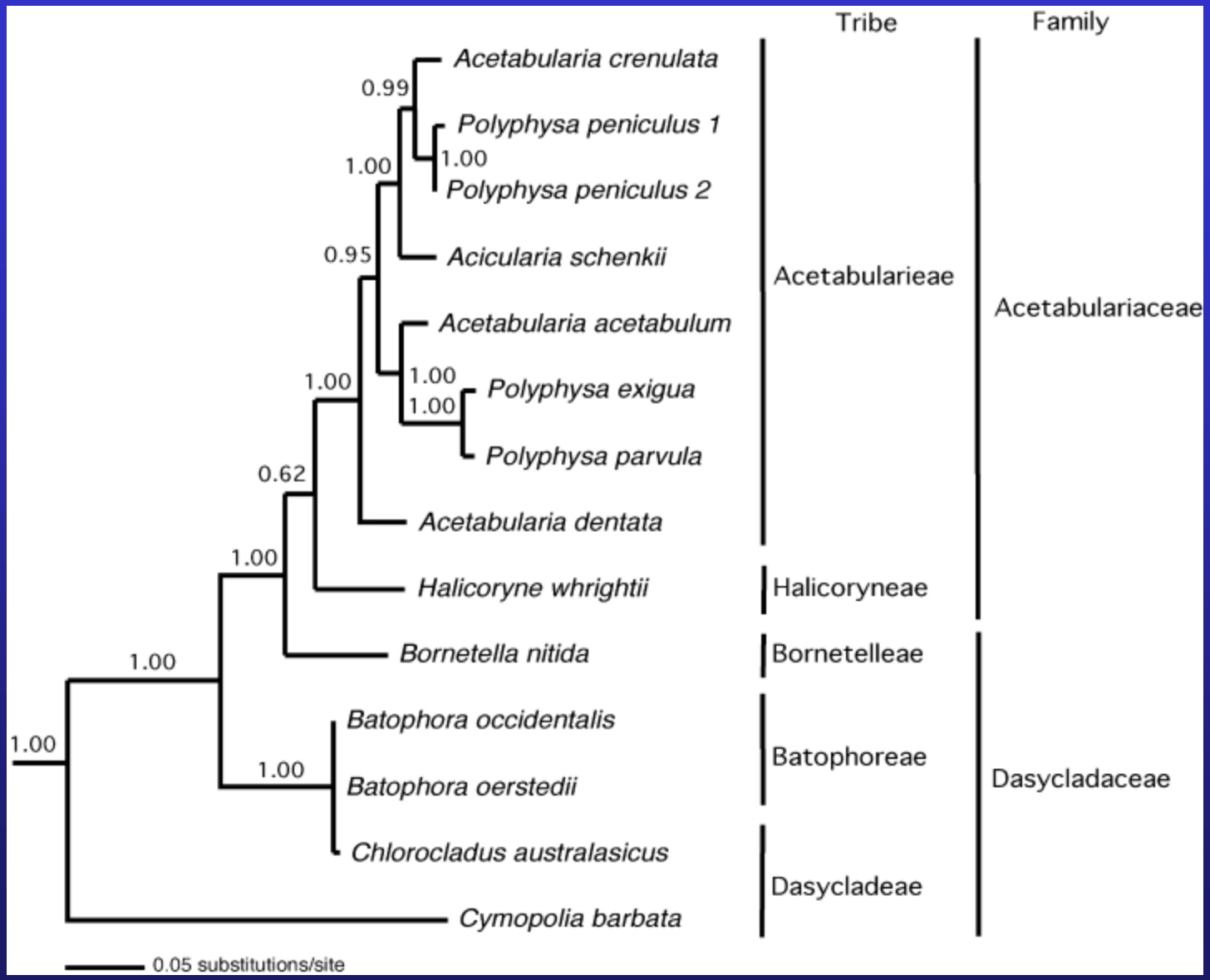
Bornetella sphaerica

longitudinal section through a cell with gametophores

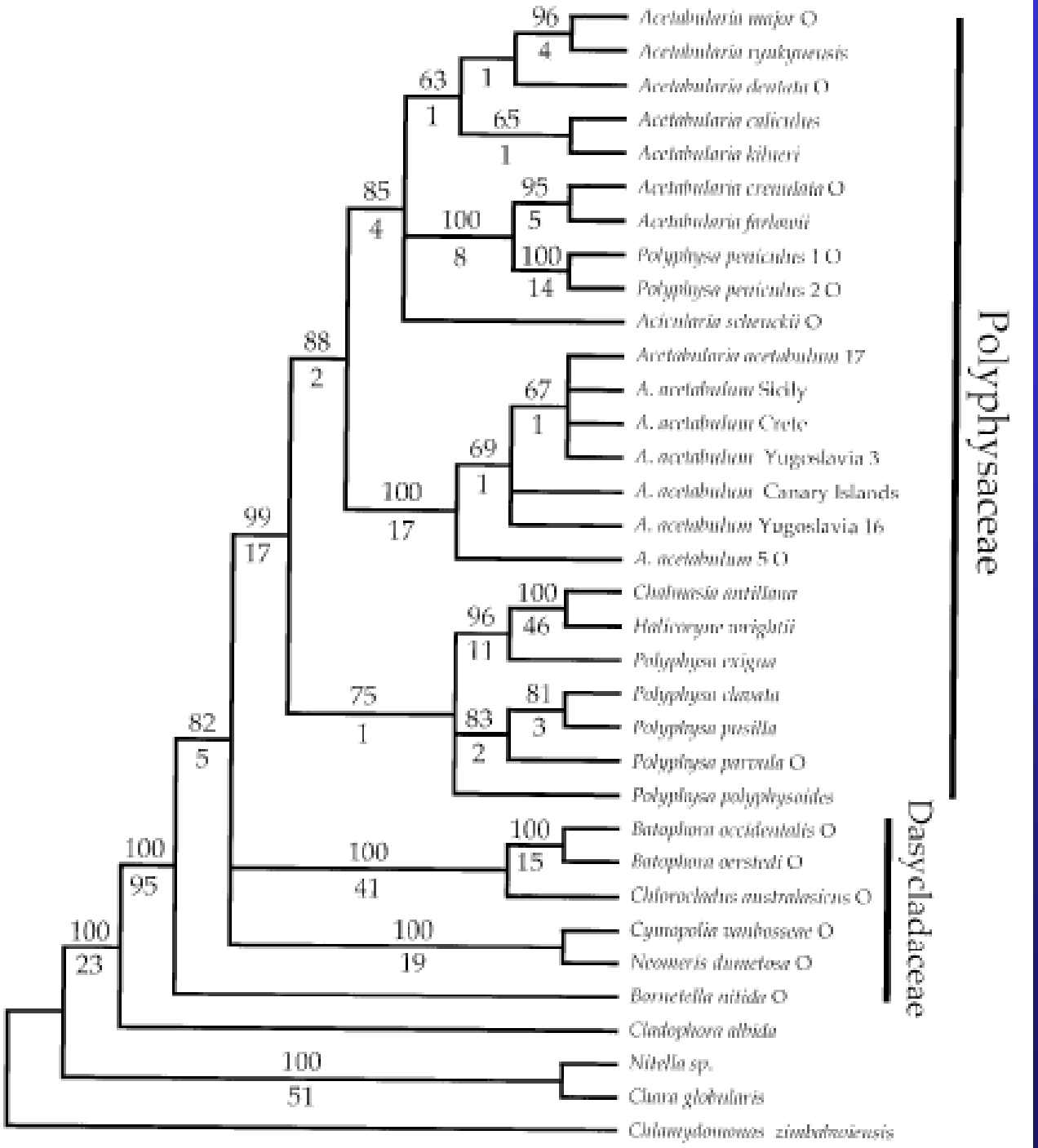
cross section through a cell with gametophores



One of two most parsimonious trees from unweighted MP analysis of *rbc*L sequence data (Zechman, 2003)



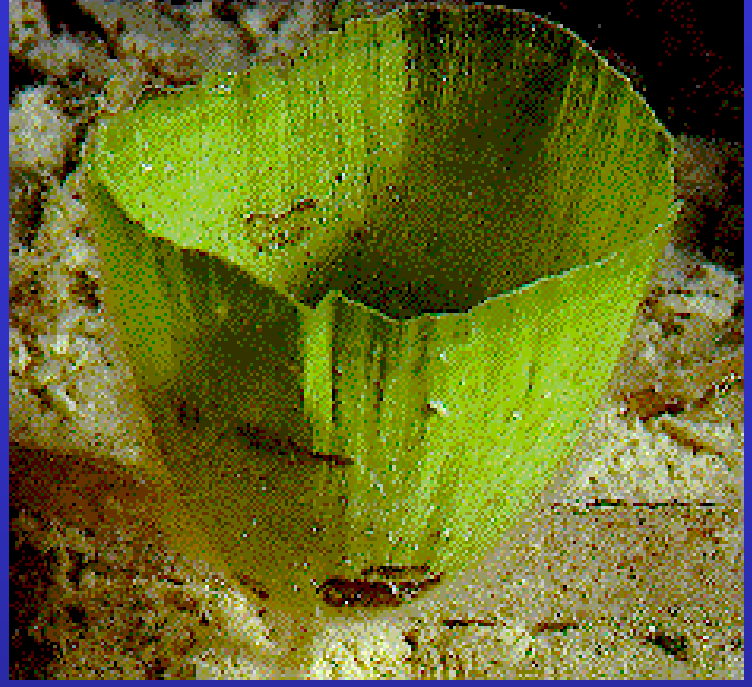
Tree topology resulting from Bayesian and ML analyses.



Strict consensus of 28 most parsimonious trees inferred from 18S rDNA

(Berger et al. 2003)

1



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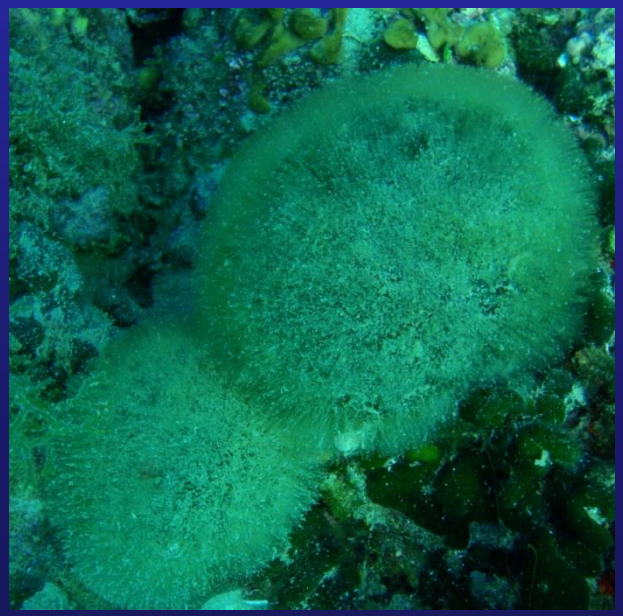

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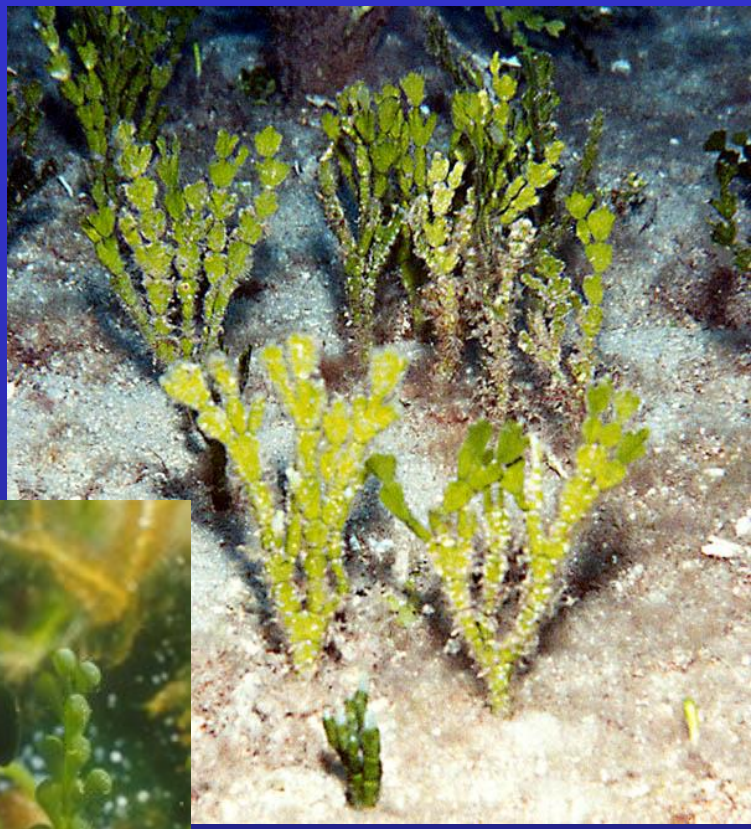

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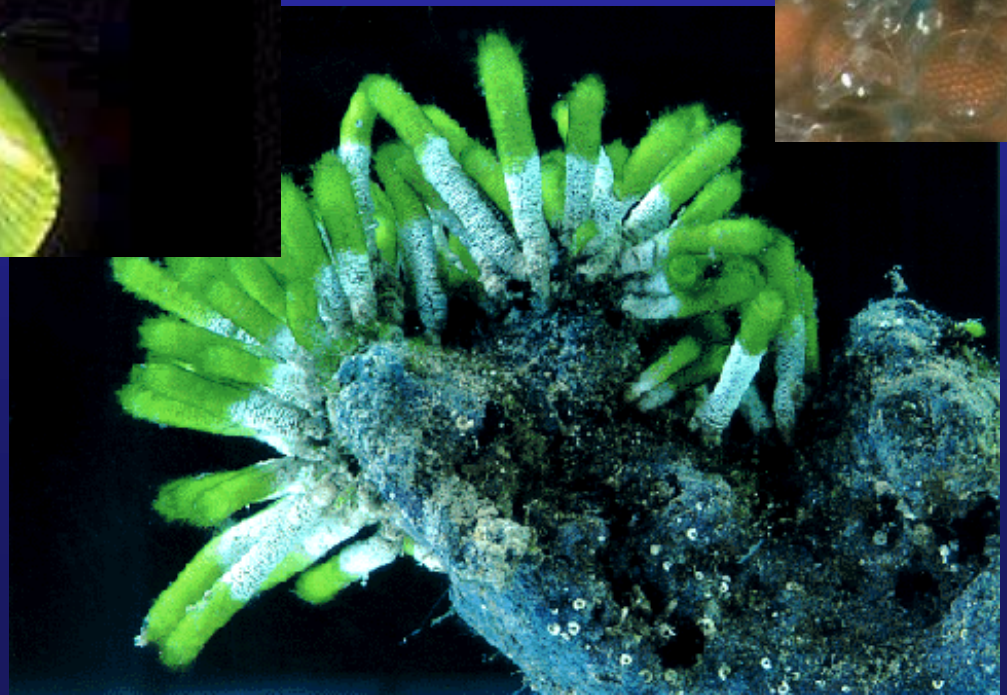

10

11

12