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PROPOSAL OF THE GRACILARIALES ORD. NOV. (RHODOPHYTA) BASED ON AN ANALYSIS OF THE REPRODUCTIVE DEVELOPMENT OF *GRACILARIA VERRUCOSA*[1]

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ABSTRACT

The mode of division of vegetative cells, formation of spermatangial parent cells, initiation of the carpogonial branch apparatus, and formation of tetrasporangial initials are homologous developmental processes that are documented for the first time in the type species of the economically important family Gracilariaceae, Gracilaria verrucosa *(Hudson) Papenfuss from the British Isles.* G. verrucosa *is characterized by a supporting cell of intercalary origin that bears a 2-celled carpogonial branch flanked by two sterile branches, direct fusion of cells of sterile branches onto the carpogonium, formation of an extensive carpogonial fusion cell through the incorporation of additional gametophytic cells prior to gonimoblast initiation, gonimoblast initials produced from fusion cell lobes, schizogenous development of the cystocarp cavity, inner gonimoblast cells producing tubular nutritive cells that fuse with cells of the pericarp or floor of the cystocarp, absence of cytologically modified tissue in the floor of the cystocarp, and carposporangial initials produced in clusters or irregular chains. Spermatangial parent cells are generated in filaments from intercalary cortical cells that line an intercellular space forming a 'pit' or 'conceptacle'. Tetrasporangial initials are transformed from terminal cells derived through division of an outer cortical cell. Tetrasporangia are cruciately divided.*

The Gracilariaceae is removed from Gigartinales and transferred to the new order Gracilariales. Their closest living relatives appear to be agarophytes belonging to the Gelidiales and Ahnfeltiales.

Key index words: agarophytes; Gracilaria; *Gracilariaceae;* ***Gracilariales*** *ord. nov.; morphology; reproductive development; Rhodophyta; systematics; taxonomy*

The characterization and systematic position of the Gracilariaceae remain largely unresolved. Since some members are of economic importance as a source of the phycocolloid agar, it is especially critical that the family be well defined morphologically and well circumscribed taxonomically (Bird and McLachlan 1982). Despite a large body of literature on the morphology of species of the Gracilariaceae, conflicting interpretations continue to be maintained regarding the manner of formation of the male gametangia and the nature of the female reproductive system before and after fertilization.

In this study the developmental morphology of the male and female reproductive systems is described in the type species of *Gracilaria, G. verrucosa* (Hudson) Papenfuss. A new order, Gracilariales, is proposed and characterized based on the family Gracilariaceae.

HISTORICAL REVIEW

Greville (1830:123) erected *Gracilaria* to contain four species. No type was designated. Later, Schmitz (1889:443) lectotypified the genus with *Gracilaria confervoides* (Linnaeus) Greville based on *Fucus confervoides* Linnaeus (1763:162), a later homonym of *Fucus confervoides* Hudson (1762:474) [= *Rhodomela confervoides* (Hudson) Silva (1952:269)].

Silva (pers. comm.) informs us that the earliest available basionym applicable to *Gracilaria confervoides* is *Flagellaria confervoides* Stackhouse (1809:92). Accordingly, the generitype species is *Gracilaria confervoides* (Stackhouse) Greville. Papenfuss (1950:195) advocated that the earliest correct name was *Fucus verrucosus* Hudson (1762:470) and made the combination *Gracilaria verrucosa* (Hudson) Papenfuss. Dixon and Irvine (1977:210) chose Hudson's description as the lectotype of *G. verrucosa,* following a method of typification widely practiced at that time when type specimens were presumably lost (Irvine and Dixon 1982). However, under the *International Code of Botanical Nomenclature* recently adopted at the Berlin Congress (Greuter et al. 1988:art. 7.5), only a specimen or an illustration may serve as a lectotype. As we show in a separate paper (Fredericq and Hommersand 1989), two species belonging to different genera presently are confused under *Gracilaria verrucosa* in England and neighboring areas.

The name *Gracilaria verrucosa* (Hudson) Papenfuss has been somewhat indiscriminately applied to terete, irregularly branched specimens of *Gracilaria,* the species having been reported worldwide from warm temperate to tropical waters. Many extra-European records of *G. verrucosa* from outside the eastern Atlantic Ocean now are considered to be incorrect (e.g. Bird et al. 1982, Abbott 1985, Abbott et al. 1985, Chang and Xia 1985, Rodriguez de Rios 1986), and *G. verrucosa* may be confined to the northeastern Atlantic Ocean. Reports from outside this area require careful examination.

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MATERIALS AND METHODS

Among specimens of *Gracilaria verrucosa* examined are those from the United Kingdom: female plants, Criccieth, Gwynned, N. Wales, 10.I.85, W. E. Jones (NCU); female plants, Broad Haven near Haverford West, Pembrokeshire, S. Wales, 10.i.88, C. Maggs (NCU); Republic of Ireland: male plants, tidepool, Spanish Point, Blackhead, Co. Clare, 25.vii.87, M. Hommersand (NCU); female plants and tetrasporophytes, Blackhead, 12.vi.87, J. Brodie; female plants, rockpool, Blackhead, 12.viii.87, J. Brodie (NCU); and France: female plants, Roscoff, Finistère, 15.vii.27, H. Kylin (LD). Herbarium abbreviations follow Holmgren et al. (1981). For a complete list of specimens examined see Fredericq (1988).

Material used in developmental studies was fixed and preserved in 5–10% formalin/seawater. Transverse and longitudinal hand sections were stained with aceto-iron-hematoxylin-chloral hydrate (Wittmann 1965) and mounted in 50:50 Hoyer's mounting medium according to the procedure of Hommersand and Fredericq (1988).

RESULTS

Vegetative morphology. Thalli are erect, to 25 cm tall with up to three orders of branches, terete throughout, straw colored or purplish. Although a single main axis sometimes is distinguishable (Fig. 2, at right) usually there are several (Fig. 1) to few main axes arising from a small discoid holdfast (Fig. 2). Branching is irregularly subdichotomous (Fig. 1) to unilateral (Fig. 2). Second and third order branches taper toward their apices and are not constricted at their point of insertion. Occasionally, branches of all orders are beset with small proliferations (Fig. 2) and are sparingly branched in the distal regions.

Gracilaria verrucosa is pseudoparenchymatous throughout (Figs. 3, 4). A longitudinal section through the finest branchlet reveals a central axis and an apical cell that divides by an obliquely longitudinal concavo-convex septum (Fig. 5), followed by transverse division of the subapical cell, as seen in the neigboring cell files. Surface cortical cells divide in the same manner as the apical cell of the central axis (Fig. 6). Secondary pit-connections are formed between all but the most distal cells of cortical cell files, quickly obscuring the fundamental uniaxial growth pattern. A transverse section through the apex of a young branch shows a loosely compacted inner cortex and thin-walled medulla with large intercellular spaces (Fig. 3). The boundary between the cortex and medulla is indistinct except that cells in the outer two to three layers are conspicuously pigmented. The medulla is composed of enlarged, highly vacuolate, thick-walled cells (Fig. 4). Outer cortical cells are either uninucleate or multinucleate and are typically longer than broad (Figs. 3–6). Deciduous hair cells near the apex are scarce to abundant (Fig. 37). Lateral branches are initiated from surface cortical cells.

Female reproductive apparatus before fertilization. The female reproductive apparatus develops near the apex of a main axis or lateral branch and consists of a supporting cell that bears a 2-celled carpogonial branch flanked by a pair of sterile branches (Figs. 9–11). It is initiated from an outer, uninucleate cortical cell that becomes larger and more darkly staining than neighboring cells. Such a cell undergoes an oblique longitudinal division by a concavo-convex septum, followed by a second oblique division of the subapical cell to produce a basal intercalary cell that will subtend the carpogonial branch and two peripheral cells destined to produce the sterile branches (Fig. 7).

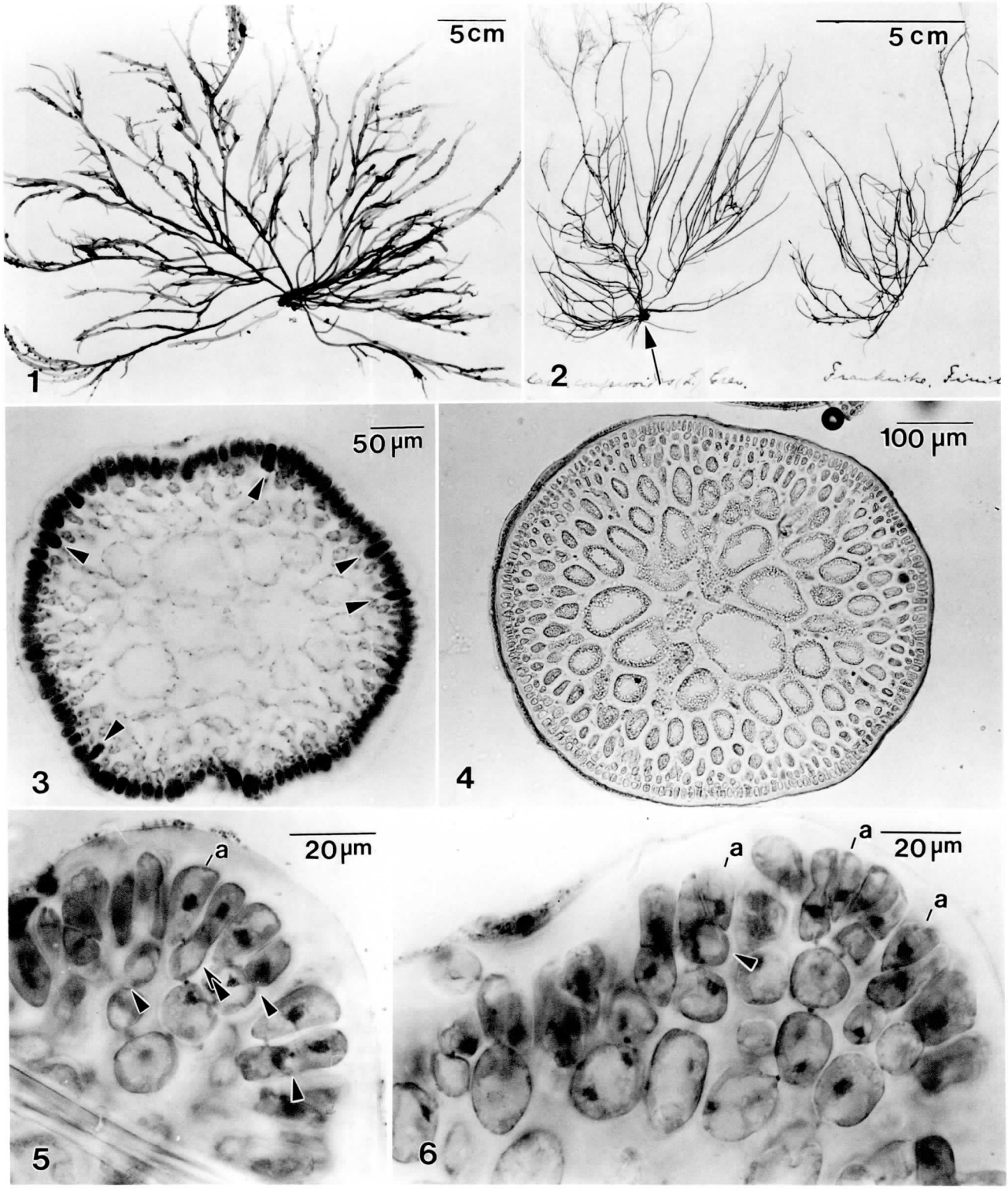
The uninucleate supporting cell cuts off a conical carpogonial branch initial towards the thallus surface (Fig. 8). The initial undergoes a single transverse division to produce a roundish carpogonium and a rectilinear hypogynous cell (Fig. 9). At maturity the supporting cell is a multinucleate, subspherical (Fig. 10) to wedge-shaped (Fig. 11) intercalary cell, distinguishable from surrounding cortical cells by its larger size. The carpogonium remains uninucleate, expands while becoming conical, and forms a straight trichogyne extending towards the thallus surface (Figs. 10, 11). The hypogynous cell becomes binucleate or occasionally multinucleate.

The two sterile branch initials divide transversely, forming a pair of unbranched sterile laterals that flank the carpogonial branch (Figs. 9–11). Cells of the sterile branches remain uninucleate up to the 3-celled stage.

The carpogonium never extends above the surface of the cortex, and the tip of the trichogyne was not seen to project beyond the surface of the outer cuticular layer. The tip of the trichogyne apparently collapses becoming funnel-shaped immediately after fertilization, and spermatia or bacteria accumulate in the trichogyne funnel in fertilized carpogonia that have aborted (Fig. 12, arrow). Aborted carpogonia are common in the vicinity of young cystocarps. They are shriveled, with collapsed cell contents and a degenerating nucleus (Fig. 12).

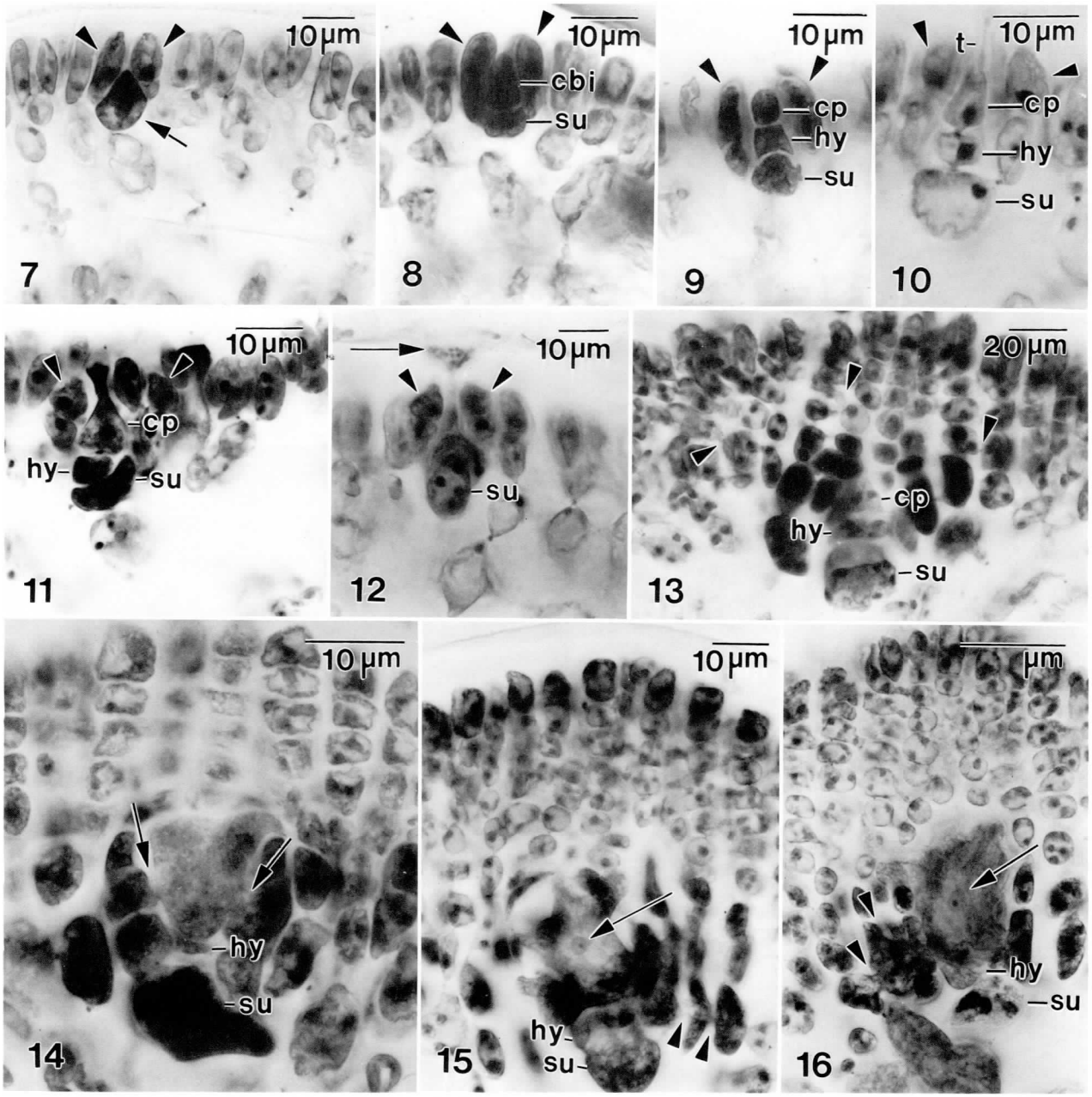
Early post-fertilization stages. Two nuclei, assumed to be the carpogonium and spermatium nuclei, were occasionally seen inside the carpogonium prior to their fusion (Fig. 13). Already at this stage, the two sterile groups and surrounding cortical filaments have started to grow forming files of cells that initiate the pericarp (Fig. 13). Secondary pit-connections are quickly established between neighboring files of pericarp cells, which become multinucleate (Figs. 13, 15, 16). The initially rectilinear shapes of the pericarp cells (Fig. 14) become more spherical in response to the abundant formation of secondary pit-connections (Figs. 15, 16).

Soon after fertilization, the lowermost three cells of the sterile branches flanking the carpogonial branch each fuse directly onto the expanding carpogonium, circumventing the hypogynous cell and forming a carpogonial fusion cell (Fig. 14). The original squarish shape of the sterile cells is lost as they are completely incorporated into the expanding fusion cell (Fig. 15). The conspicuous fertization nucleus sits in the center of the fusion cell (Figs. 15,



NOTE: Abbreviations used in figures: a = apical cell; cbi = carpogonial branch initial; cp = carpogonium; fu = fusion cell; hy = hypogynous cell; o = ostiolar region; su = supporting cell; t = trichogyne.

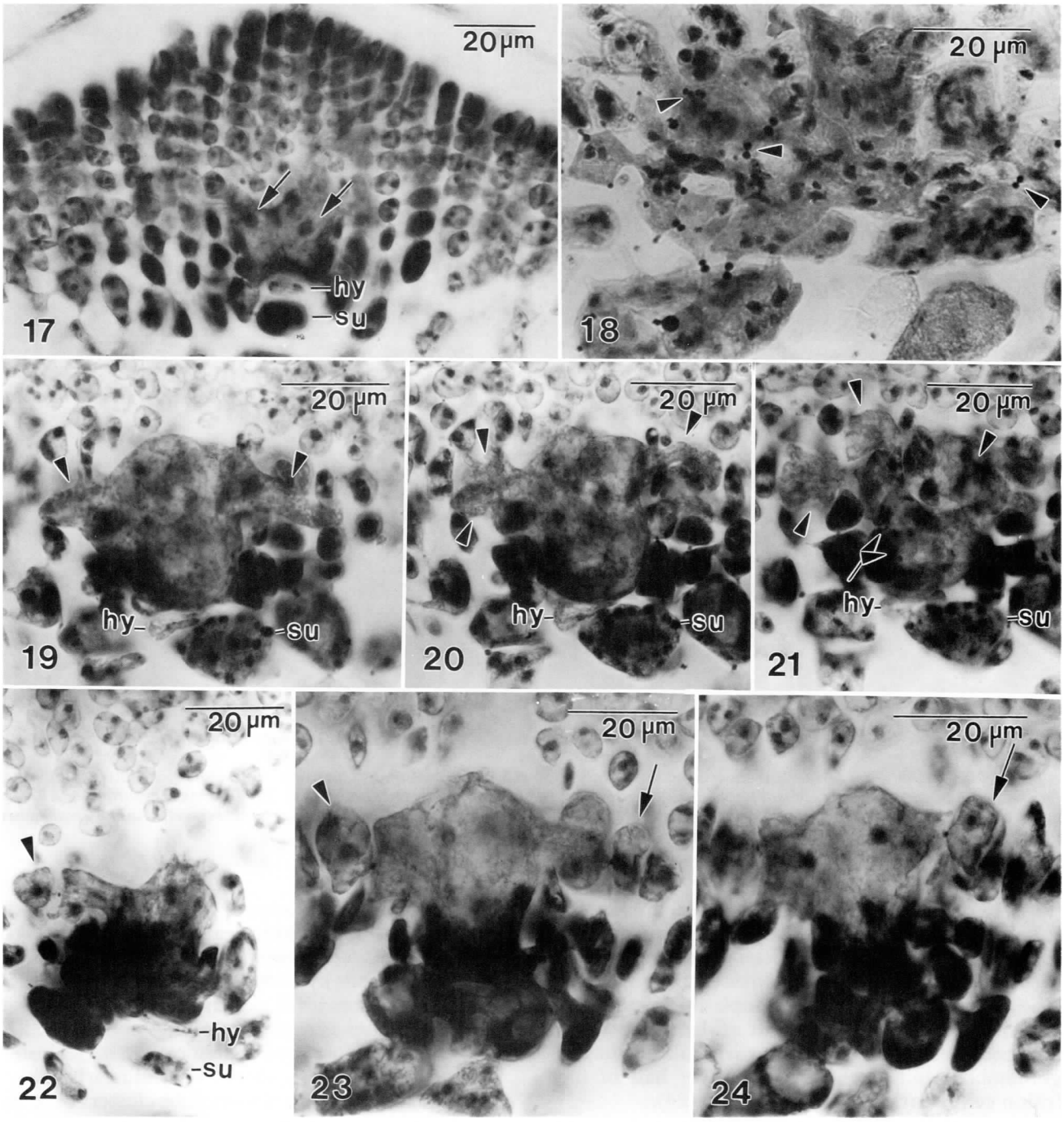
FIGS. 1–6. *Gracilaria verrucosa.* FIG. 1. Habit of wet cystocarpic specimen from N. Wales. FIG. 2. Habit of wet cystocarpic specimen from France. Arrow points to discoid holdfast. FIG. 3. Transverse section through second-order branch within 1 mm of apex; stained with hematoxylin. Arrowheads point to hair cell primordia (Ireland). FIG. 4. Transverse section through base of first order branch; stained with aniline blue (Ireland). FIG. 5. Longitudinal section through finest branchlet showing apical cell of central axis. Double arrowhead points to subapical cells prior and arrowheads after transverse division (Ireland). FIG. 6. Transverse section through cortex, showing apical cells of cortical cell files. Arrowhead points to transversely divided subapical cell (Ireland).



FIGS. 7–16. *Gracilaria verrucosa* from Ireland. FIG. 7. Outer cortex with supporting cell (arrow) and sterile branch initials (arrowheads). FIG. 8. Supporting cell with carpogonial branch initial and sterile branch initials (arrowheads). FIG. 9. Supporting cell with hypogynous cell, carpogonium and sterile branches (arrowheads). FIGS. 10, 11. Carpogonial branch apparatus consisting of supporting cell, sterile branches (arrowheads), hypogynous cell and carpogonium with trichogyne. FIG. 12. Degenerating carpogonium with trichogyne (arrow) not extending above cuticle, and uninucleate sterile cells (arrowheads). Note aggregation of spermatia or bacteria in trichogyne funnel. FIG. 13. Binucleate carpogonium. Abundant formation of secondary pit-connections (arrowheads) in incipient pericarp. FIG. 14. Sterile cells fused directly (arrows) onto enlarged carpogonium after fertilization. FIG. 15. Fusion cell with fertilization nucleus (arrow). Surrounding vegetative cells (arrowheads) are being incorporated into fusion cell. FIG. 16. Young fusion cell with fertilization nucleus (arrow) and progressive incorporation of vegetative cells (arrowheads).

16), whereas the nuclei of the hypogynous cell degenerate.

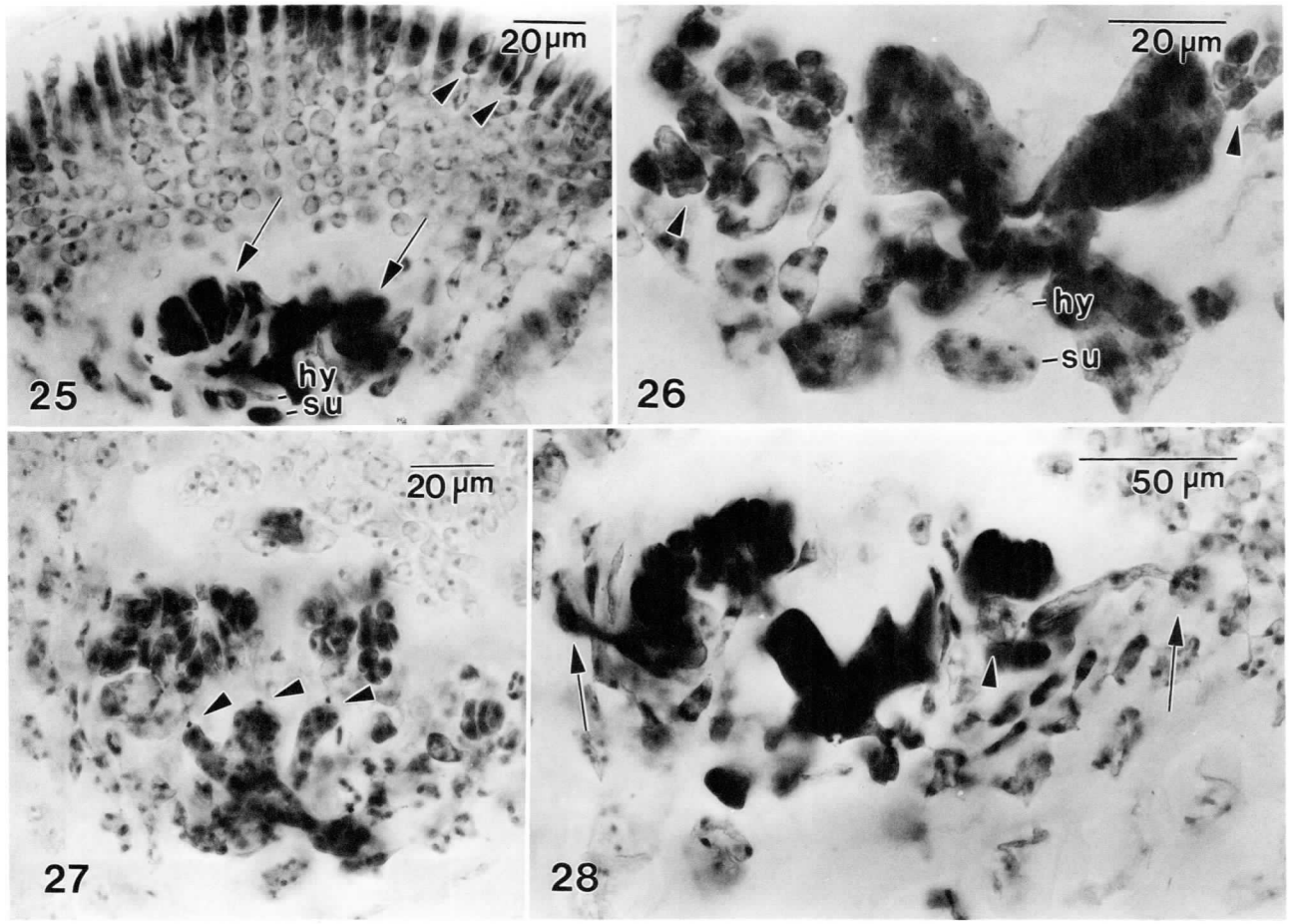
Fusion cell buildup in *Gracilaria verrucosa* is not limited to primary fusions of the sterile cells with the fertilized carpogonium. The lowermost sterile cells that have already fused onto the carpogonium incorporate multinucleate neighboring vegetative cells laterally for a distance of up to five cortical files (Figs. 15, 16). Fusions proceed around existing pit-connections, and the pit-connections may enlarge prior to their dissolution (Fig. 18). The fusion cell of *Gracilaria verrucosa* thus contains a mixture of



FIGS. 17–24. *Gracilaria verrucosa.* Figs. 17, 19–24: specimen from Ireland; Fig. 18: specimen from N. Wales. FIG. 17. Division of the zygote nucleus (arrows) inside fusion cell. FIG. 18. Progressive incorporation of vegetative cells into fusion cell around enlarged pit-connections (arrowheads). FIGS. 19–21. Three different focal planes showing fusion cell with lobed cytoplasmic extensions (arrowheads). FIG. 21. Nuclei in metaphase undergoing synchronous division (double arrows) inside fusion cell. FIG. 22. Uninucleate gonimoblast initial (arrowhead) cut off from lobe of fusion cell. FIGS. 23, 24. Gonimoblast initial (arrowhead) and gonimoblast file (arrow) seen in two different focal planes. Note that basal part of fusion cell stains darker than upper part.

diploid nuclei (derivatives of the fertilization nucleus) and haploid nuclei (nuclei from secondarily incorporated vegetative cells). Division of the fertilization nucleus occurs early (Fig. 17) when only few basal vegetative cells have been incorporated into the fusion cell or later after an extensive fusion cell has been established (Figs. 15, 16).

Gonimoblast development. Prior to gonimoblast initiation, the fusion cell forms several cytoplasmic projections (Figs. 19–21) and contains few diploid nuclei

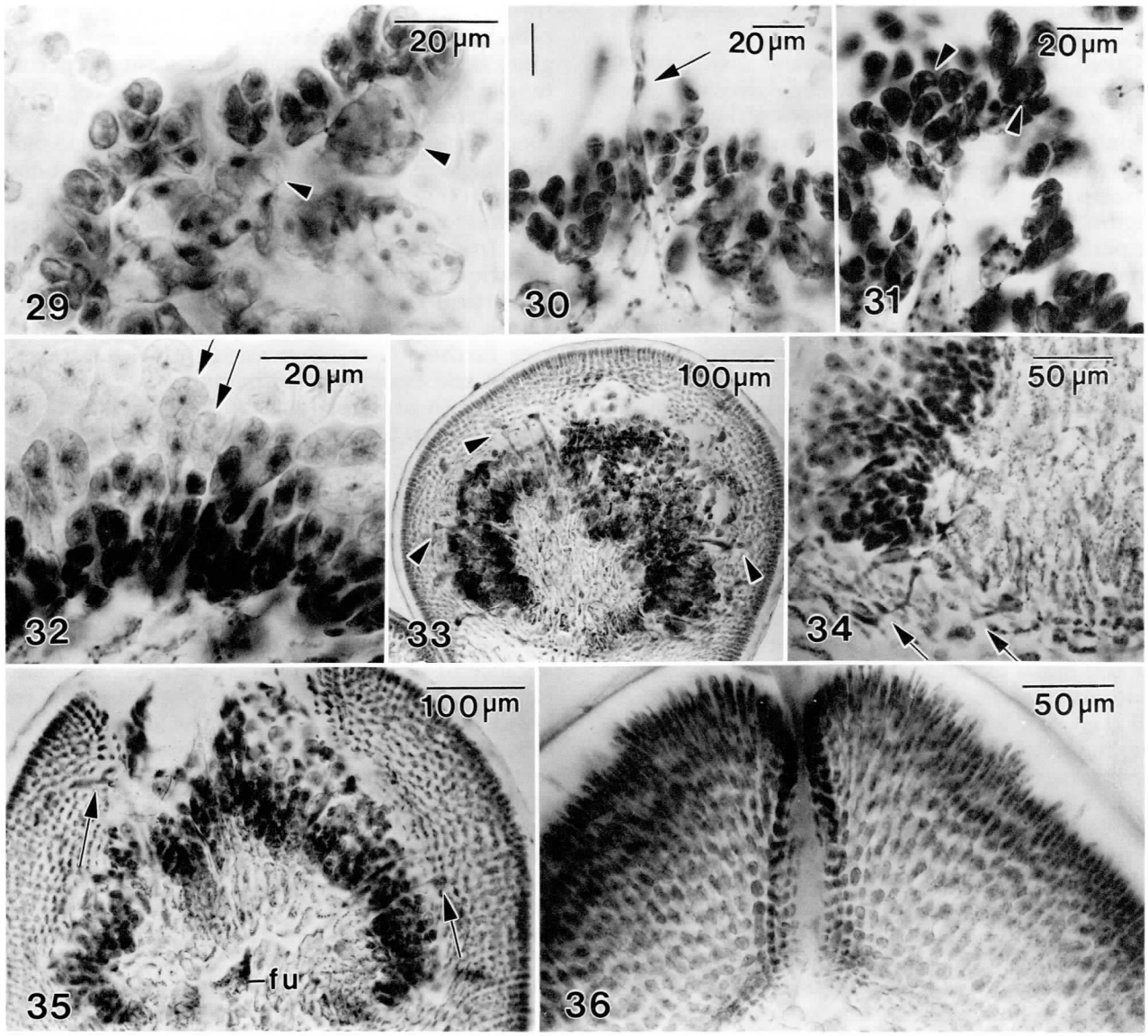


FIGS. 25–28. *Gracilaria verrucosa* from Ireland. FIG. 25. Fusion cell with gonimoblasts (arrows). Note oblique longitudinal division (arrowheads) of outer pericarp cells. FIG. 26. Small clusters of gonimoblast cells (arrowheads). FIG. 27. Dissected, multinucleate fusion cell with gonimoblast clusters borne on lobes (arrowheads). FIG. 28. Fusions (arrows) of tubular nutritive cells issuing from inner gonimoblast cell (arrowhead) with pericarp cells.

(Figs. 19–21). Occasionally many nuclei are seen in fusion cells that have not formed gonimoblast initials (Fig. 18). The diploid nuclei divide repeatedly, with one nucleus migrating into each projection. The terminal end of each cytoplasmic projection contains a single nucleus, is cut off by a cross-wall, and becomes a semicircular to irregularly shaped gonimoblast initial whereas the lower half of the fusion cell is darkly stained (Figs. 22–24).

At an early stage of gonimoblast formation, a cavity develops schizogenously distal to the fusion cell, probably through break-down of the primary pit-connections between pericarp cells and cells of the inner cortex (Fig. 25). The cystocarp cavity first develops as a low arch that extends laterally across only a few files of cells separating the pericarp from the floor of the cystocarp.

The first divisions of a gonimoblast initial are anticlinal and produce a short horizontal file up to three cells long that bends slightly downward (Figs. 23, 24). Each of the primary gonimoblast cells cuts off derivatives from its upper side and then expands rapidly, becoming multinucleate and vacuolate (Figs. 25–27). Repeated division of apical and subapical gonimoblast cells by concavo-convex septa (Fig. 26) typically produces small, darkly staining clusters that initially develop as separate gonimolobes (Figs. 27, 28). The innermost gonimoblast cells in each cluster become multinucleate, vacuolate, and nearly spherical (Fig. 29). Later, the gonimolobe clusters become confluent when secondary pit-connections are formed between their inner gonimoblast cells. Once linked by secondary pit-connections, the inner gonimoblast cells stretch and become vertically elongated, so that inner sterile tissues are sharply demarcated from the maturing zone of outer gonimoblast filaments (Figs. 33–35). The formation of young carposporangial filaments at first follows the pattern of early gonimoblast development based on concavo-convex divisions (Fig. 29). Terminal gonimoblast cells expand and produce clusters of carposporangial initials (Fig. 31). Finally, all carpo-



FIGS. 29–36. *Gracilaria verrucosa.* Figs. 29–32, 36: specimen from Ireland; Fig. 33: specimen from S. Wales; Figs. 34, 35: specimens from North Wales. FIG. 29. Vacuolate, subisodiametric, inner gonimoblast cells (arrowheads) bearing gonimoblast clusters. FIG. 30. Tubular nutritive cell (arrow) issuing from inner gonimoblast (Ireland). FIG. 31. Carposporangial initials dividing by concavo-convex septa (arrowheads). FIG. 32. Basipetal maturation of carposporangial chains (arrows) (Ireland). FIG. 33. Non-median section through cystocarp showing tubular nutritive cells fusing both with cells of pericarp (arrowheads) and cystocarp floor. FIG. 34. Non-median section. Detail of tubular nutritive cells fused to cells of cystocarp floor (arrows). FIG. 35. Median section through cystocarp showing remnants of fusion cell, central sterile tissue, and a few tubular nutritive cells (arrows). FIG. 36. Longitudinal section of ostiole.

sporangial initials are transformed into files of carposporangia that mature basipetally (Fig. 32) in irregular chains of up to six cells (Figs. 33–35).

As the carposporophyte matures, multinucleate, non-septate tubular nutritive cells subtended by inner gonimoblast cells (Figs. 28, 30) elongate toward the pericarp and fuse directly with one or more periclinally stretched pericarp cells (Figs. 28, 33–35) without forming conjunctor cells or secondary pit-connections. Tubular nutritive cells are scarce to abundant in any given cystocarp. A non-median transverse section through a mature cystocarp (Figs. 33, 34) reveals that tubular nutritive cells fuse with cells of both pericarp and floor of the cystocarp. In contrast, a median section shows them fusing exclusively with cells of the pericarp (Fig. 35). A distinct zone of cytologically modified cells in the floor of the cystocarp, corresponding to an inner pericarp, is never present in *G. verrucosa.* Inner sterile gonimoblast cells typically become elongated, thick-walled, and vacuolate in mature cystocarps. The fusion cell, the elongated inner gonimoblast cells, and

the tubular nutritive cells are often preserved, even in old herbarium specimens.

Outer pericarp cells that initially divide transversely undergo oblique longitudinal divisions at an early stage of gonimoblast formation (Fig. 25, arrowheads) becoming elongate and pointed, and continue to divide terminally to produce a dome-shaped pericarp (Fig. 36). An ostiolar region, seemingly the original trichogyne space, is formed in the center of the pericarp. Mature cystocarps may protrude in any direction from the surface of a branch and are not constricted at their bases (Fig. 1).

Development of spermatangia. Spermatangial maturation is acropetal, the youngest spermatangial patches occurring 2–5 mm behind branch tips (Fig. 37). The spermatangial parent cell initial is an intercalary cell that is pit-connected to one (Figs. 40–42) or two (Figs. 38, 39) outer cortical cells and is distinguished from surrounding vegetative cells by its darker staining properties and larger nucleus. It is formed from an outer cortical cell that undergoes an oblique longitudinal division by means of a concavo-convex septum, followed by oblique division of the subapical cell roughly perpendicular to the plane of the first. The small intercalary spermatangial parent cell initial expands rapidly until it reaches the same level at the branch surface as the outer cortical cell (Fig. 42), whereupon it undergoes an oblique longitudinal division (Figs. 43, 44) to produce two spermatangial parent cells. Depending on the number of initial divisions, either one (Fig. 43) or two (Fig. 44) spermatangial parent cells may be pit-connected to an outer cortical cell. The larger parent cell next undergoes an oblique longitudinal division, giving rise to three elongate spermatangial parent cells (Fig. 45). At the three-celled stage, each spermatangial parent cell cuts off one spermatangium distally by a concavo-convex septum (Figs. 46–49). Following initiation of a spermatangium, each of the peripheral parent cells protrudes distally (Fig. 48), and the protrusion is cut off to become a further spermatangial parent cell. Successive spermatangial parent cells are formed peripherally by oblique longitudinal divisions, resulting in a branched filament that fills the intercellular space between distal cortical cells (Fig. 50). Since secondary pit-connections are formed only sparingly between distal cortical cells, intercellular spaces are abundant and a branched spermatangial filament completely fills an intercellular space (Figs. 51–54), forming a cavity. Such a spermatangial configuration is commonly referred to as a 'pit' or 'conceptacle.' Cortical cells with pit-connections to the original spermatangial parent cell initial remain connected to it (Fig. 52). If the pit-connection to the bearing cell occupies a distal position, repeated division of the spermatangial mother cells proceeds from only one side.

Each of the spermatangial parent cells making up the branched, filamentous system generally cuts off a single spermatangium, and spermatangia are formed progressively as the spermatangial parent cells mature. Although usually only one spermatangium is produced at a time from a single spermatangial parent cell, occasionally two are cut off simultaneously (Fig. 53). Squash preparations of spermatangial parent cells lining a conceptacle show that they are linked exclusively by primary pit-connections (Fig. 55).

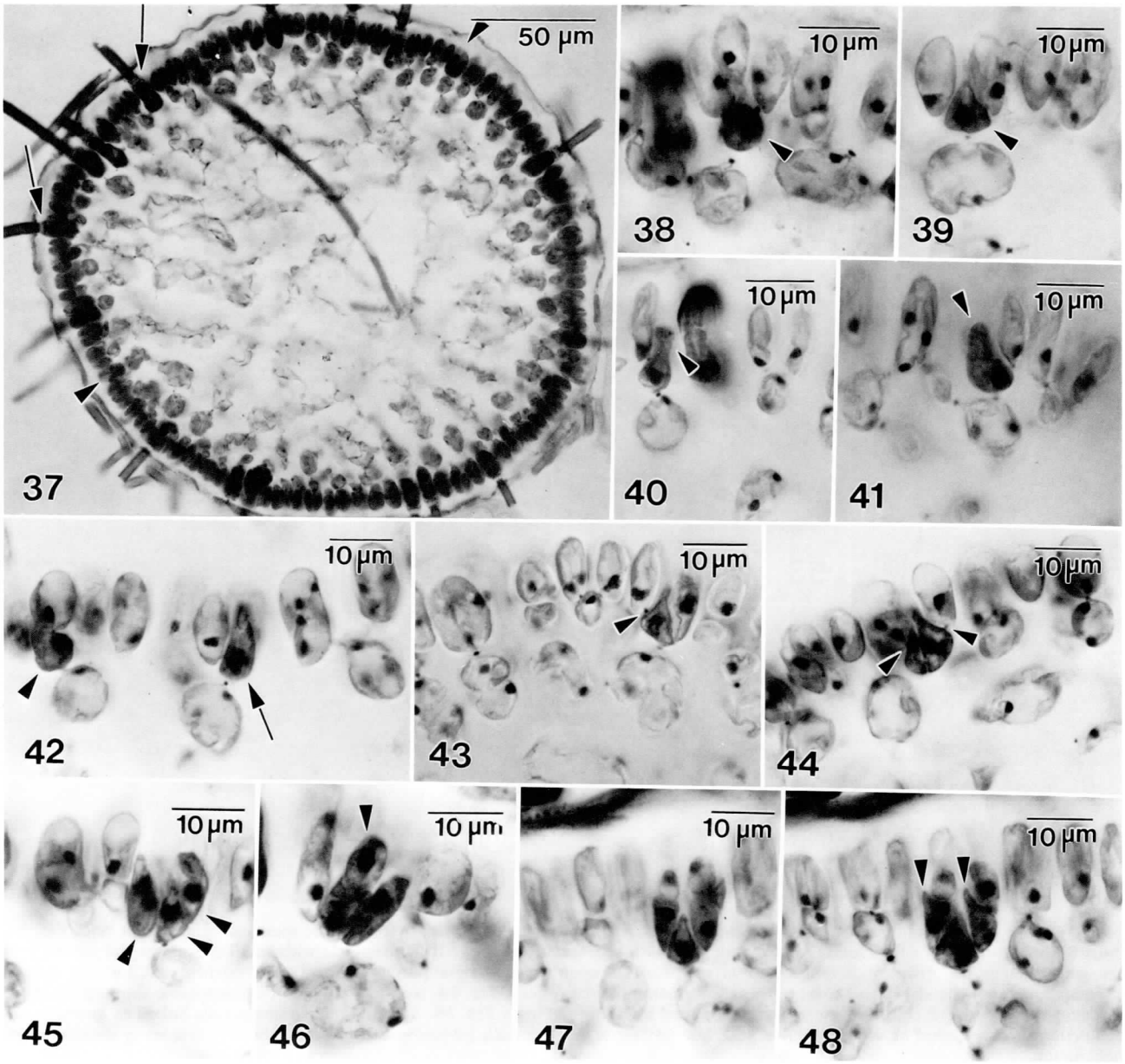
Tetrasporangia. Tetrasporangial initials differentiate from terminal cells (Fig. 56) derived through an oblique longitudinal division of a uninucleate outer cortical cell. The subapical cell divides transversely cutting off a sterile cortical cell that can divide further. A tetrasporangial initial expands in length and breadth and undergoes two successive divisions to produce four cruciately arranged tetraspores (Fig. 57).

DISCUSSION

The mode of division of vegetative cells, the formation of spermatangial parent cells, the initiation of the carpogonial branch apparatus, and the formation of tetrasporangial initials are homologous developmental processes that are documented here for the first time in *Gracilaria verrucosa.* In every instance an outer cortical cell divides along an oblique longitudinal plane by a concavo-convex septum, followed by an oblique or transverse division of the subapical cell. This homologous developmental pattern is diagnostic for the Gracilariaceae (Fredericq 1988).

Female reproductive system. Sjöstedt (1926) was the first to establish that the carpogonial branch is a 2-celled filament and to follow its development in *Gracilaria verrucosa* (as *G. confervoides*) and two other *Gracilaria* species. Kylin (1930), Dawson (1949), Oliveira (1969), Oza (1976), Edelstein et al. (1978), Yamamoto (1978) and Fredericq and Norris (1985) documented similar carpogonial branches in the same and other species from many different localities. Sjöstedt (1926) observed that the primary cortical cell cuts off one or two vegetative cells before the appearance of the carpogonial branch initial. Yamamoto (1978, Fig. 26) illustrated the carpogonial branch initial as a terminal cell cut off from an intercalary supporting cell. In both cases, cells flanking the carpogonial branch are regarded as being ordinary cortical cells. Our observations agree with those of Sjöstedt, except that we always see two lateral filament initials in *G. verrucosa,* and both are cytologically differentiated from vegetative cortical cells at the time they are cut off.

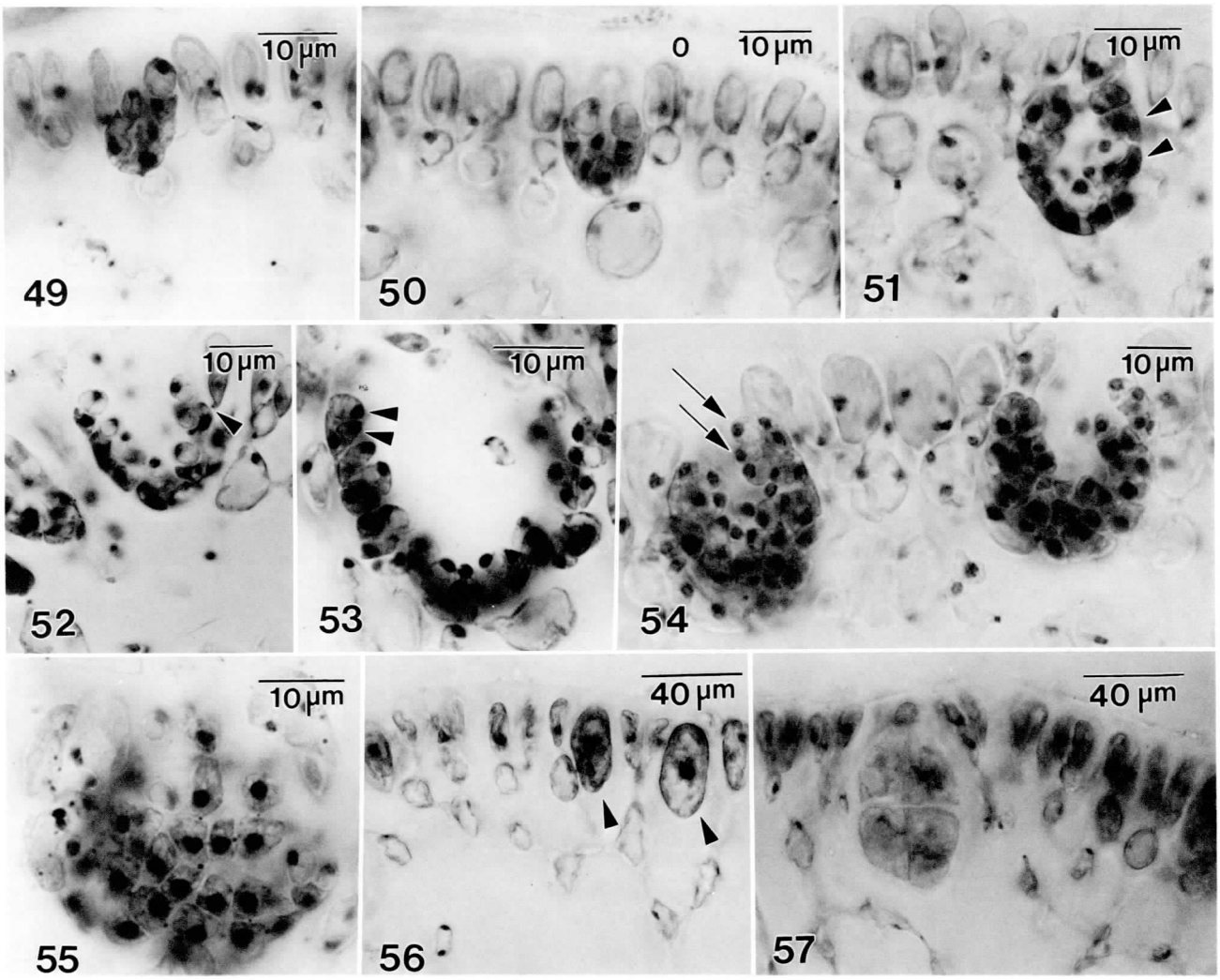
Four structures have been proposed as the site of gonimoblast initiation in the Gracilariaceae: (1) the carpogonium after its fusion with vegetative cortical cells (Sjöstedt 1926, Kylin 1930, Oliveira 1969, Delivopoulos and Tsekos 1983, Fredericq and Norris 1985), (2) the fusion product of the carpogonium, hypogynous cell and supporting cell after fusion with cortical cells (Oza 1976), (3) an auxiliary cell after



FIGS. 37–48. *Gracilaria verrucosa* from Ireland. FIG. 37. Transverse section close to apex of young spermatangial branch with spermatangial parent cell initials (arrowheads) and hair cells (arrows). FIGS. 38, 39. Intercalary, spermatangial parent cell initials (arrowhead). FIGS. 40, 41. Outward protrusion of spermatangial parent cell (arrowheads). FIG. 42. Spermatangial parent cell (arrowhead) extending to level of outer cortical cell (arrow). FIG. 43. Uninucleate spermatangial parent cells connected (arrowhead) to one outer cortical cell. FIG. 44. Same as in Fig. 43 but connected to two outer cortical cells (arrowheads). FIG. 45. Three spermatangial parent cells (arrowheads). FIG. 46. Spermatangium (arrowhead) cut off by concavo-convex wall from spermatangial parent cell. FIGS. 47, 48. Different focal planes, showing concavo-convex division of spermatangial parent cells to form spermatangia. Part of each spermatangial parent cell protrudes (Fig. 48, arrowheads) distally as a prelude to further divisions.

its fusion with cortical cells (Greig-Smith 1954, Yamamoto 1978), (4) the fusion product of the hypogynous cell, supporting cell and neighboring cells (Edelstein et al. 1978), and (5) the carpogonium after the lower cells of the flanking sterile filaments borne on the supporting cell have fused with it (this paper).

In some interpretations cells that fuse with the carpogonium have been interpreted as auxiliary cells, in others not. Kylin (1923) distinguished between auxiliary cells that issue gonimoblasts ('typical auxiliary cells') and those having a purely nutritive function ('nurse cells'). Papenfuss (1951) and Drew (1954) called the 'nurse cells' 'nutritive auxiliary cells' and referred to cells that combine both gonimoblast initiation and nutritive functions as 'generative auxiliary cells.' Drew further stipulated that an auxiliary cell be a 'specified' cell, ". . . a gametophytic cell in a specified position with which the carpogonium fus-



FIGS. 49–57. *Gracilaria verrucosa* from Ireland. FIG. 49. Spermatangia cut off singly from three spermatangial parent cells. FIG. 50. Short branched filament of spermatangial parent cells with spermatangia. FIG. 51. Conceptacle with small spermatangial parent cells (arrowheads). FIG. 52. Primary pit-connection (arrowhead) between spermatangial parent cell and outer cortical cell. FIG. 53. Pair of spermatangia (arrowheads) cut off from terminal spermatangial parent cell. FIG. 54. Non-median view of conceptacle showing filaments of spermatangial parent cells and spermatangia formed inwardly (arrows). FIG. 55. Spermatangial parent cells linked by primary pit-connections and oriented at various angles. FIG. 56. Tetrasporangial initials (arrowheads). FIG. 57. Mature, cruciately divided tetrasporangium.

es before the formation of gonimoblast, or with which a primary gonimoblast [= connecting filament) has fused." Hommersand and Fredericq (1989) proposed that an auxiliary cell functions as a site for introducing morphogenetic factors that either initiate gonimoblasts or transform their mode of development, and may also play a role in rejecting incompatible or disharmonious fertilizations. They referred to any cell that only provides nutriment to the developing gonimoblast as a 'nutritive cell.' Their broad use of the term 'nutritive cell' thus includes 'nurse cells' sensu Kylin (1923) and 'nutritive auxiliary cells' sensu Papenfuss (1951) and Drew (1954).

Sjöstedt (1926) commented that a connection certainly takes place between the carpogonium and richly protoplasmic vegetative cells in the environs, "but not with one predesigned in structure and nuclear conditions," and he concluded that no true auxiliary cell occurs in *Gracilaria.* At first, Kylin (1930) was ambivalent about the presence of auxiliary cells. Later, he stated that none are seen before fertilization (Kylin 1956). We have shown that gonimoblasts issue directly from the carpogonium only after the lower cells of the flanking sterile filaments borne on the supporting cell have fused onto the carpogonium. The direction of primary fusions is from the sterile cells toward the carpogonium and not vice versa, as Sjöstedt (1926) postulated. These cells are 'specified' in accordance with Drew's definition of an auxiliary cell; however, their nuclei are not specially modified and they appear to play only a nutritive role in the subsequent development of

the fusion cell. The cytoplasmic boundary between the expanding fertilized carpogonium and the fused, multinucleate sterile cells is maintained until gonimoblast initials have been cut off, and vegetative nuclei do not enter the central region occupied by the fertilization nucleus.

A special behavior pattern is seen in *Gracilaria* in which the carpogonium becomes isolated from surrounding gametophytic cells by the vacuolization and early degeneration of the hypogynous cell and its nuclei after fertilization. This separation appears to act as a physiological barrier that isolates the carpogonium conferring properties and capabilities on it similar to those of a generative auxiliary cell in other families of the Florideophycidae. We conclude that, instead of an auxiliary cell, a unique type of generative fusion cell is formed prior to gonimoblast initiation that includes the carpogonium containing the fertilization nucleus or its derivatives and any incorporated cells of the flanking sterile branches.

The primary fusion cell of *Gracilaria* expands in size secondarily through the incorporation of additional gametophytic cells in the floor of the cystocarp which take place around existing pit-connections. Such a 'secondary' fusion cell is present only in *Gracilaria* and *Polycavernosa* and is absent in other genera of the Gracilariaceae (Fredericq 1988). The gonimoblast is not incorporated into the fusion cell at any stage of its development, a feature that distinguishes it from fusion cells commonly found in the order Gigartinales (see Hommersand and Fredericq 1989).

Thuret and Bornet (1878) illustrated the elongated tubes in the cystocarp of *Gracilaria* which they thought originated either from the pericarp or from the gonimoblasts. Sjöstedt (1926) named these tubes 'nutrient tubular cells,' to which he ascribed a nutritive function. A new term, 'nutritive filaments', subsequently was coined by Dawson (1949:3). Kraft (1977b) proposed using a purely descriptive term, 'traversing filaments,' whereas Abbott et al. (1985) called them 'absorbing filaments.' In this study we call these structures 'tubular nutritive cells,' a minor variant of the term used by Sjöstedt (1926). They are actually not filaments at all, but elongate, multinucleate, non-septate cells issuing from inner gonimoblast cells that fuse with gametophytic cells, either in the outer pericarp or in the floor of the cystocarp.

Hommersand and Fredericq (1989) proposed that the growth of the red algal carposporophyte typically proceeds in stages, with periods of active development separated by periods of comparative inactivity during which developmental patterns undergo a change. We see that in *Gracilaria* both pre- and post-fertilization development can be divided into a series of eight events during which existing cells are transformed for carrying out specific roles in carposporophyte development: (1) transformation of a surface cortical cell and its division into an intercalary supporting cell and a pair of sterile branch initials, (2) development of a two-celled carpogonial branch and formation of a pair of flanking sterile filaments, (3) fertilization followed by modification of the hypogynous cell isolating the carpogonium and the simultaneous initiation of an external pericarp through the transverse division of superficial cortical cells, (4) fusion of nutrient-rich cells of the sterile filaments flanking the carpogonium onto the carpogonium to form a primary fusion cell, (5) secondary incorporation of surrounding gametophytic cells into an expanding fusion cell, (6) initiation of gonimoblasts from lobes of the nutriment-rich fusion cell accompanied by the schizogenous development of the cystocarp cavity, (7) growth of clusters of gonimoblast cells, and (8) depletion of nutriment from the fusion cell and the concomitant initiation of multinucleate tubular nutritive cells from inner gonimoblast cells that fuse onto cells of the pericarp or floor of the cystocarp at the same time that the carposporangia differentiate.

Gracilaria is the only non-parasitic genus of the Gracilariaceae that is characterized by the absence of an inner pericarp consisting of a prominent, cytologically modified nutritive tissue in the floor of the cystocarp. Conjunctor cells are not cut off from gonimoblast cells adjacent to the floor of the cystocarp nor do ordinary terminal gonimoblast cells fuse with gametophytic cells in the floor of the cystocarp as in *Gracilariopsis* (Fredericq and Hommersand 1989). Instead, in *Gracilaria,* the darkly staining gametophytic cells adjoining the fertilized carpogonium are incorporated into an expanding fusion cell early in gonimoblast development. Later, one-celled, multinucleate tubular nutritive cells are transformed from inner gonimoblast cells that fuse with cells of the pericarp, evidently as a means of supplying additional food material to the maturing carposporophyte.

Male reproductive system. Thuret *in* Lejolis (1863: 134) first described the organization of male reproductive structures, which he called antheridia, in deep conceptacles in *Gracilaria verrucosa* (as *G. confervoides*). Thuret and Bornet (1878, pl. 40) provided an illustration and commented that this configuration diverged from the usual Floridean antheridial type. The same authors reported that the antheridial parent cells in *G. armata* J. Agardh and in *G. bursa-pastoris* (S. G. Gmelin) Silva (as *G. compressa*) originated through repeated divisions of cortical cells, forming patches of various sizes on the thallus surface. Buffham (1893:294, pl. 13) illustrated and confirmed the observations of Thuret and Bornet for *G. verrucosa* (as *G. confervoides*). Dawson (1949) discovered three distinct types of spermatangial configuration at maturity in the Gracilariaceae and stressed their systematic value. Subsequently, Dawson (1961:291–292) documented that spermatangia could be organized either in deep or shallow, nonconfluent conceptacles or be distributed in a con-

tinuous superficial layer. In his monograph on Japanese Gracilariaceae, Ohmi (1958) stressed that spermatangial configuration was the prime character for distinguishing species. Later, Yamamoto (1975:113, Fig. 27) divided the genus into three subgenera based on the three different types of male organs: (1) *Gracilariella,* with *chorda*-type males: superficial spermatangia scattered continuously over the thallus surface, (2) *Textoriella,* with *textorii*-type males: spermatangia situated in shallow concave conceptacles within the cortex, and (3) *Gracilaria,* with *verrucosa*-type males: spermatangia arranged in deep conceptacles within the cortex. Recently, Reading and Schneider (1986) showed a continuum between the *textorii* and *verrucosa*-type male configuration in "*Gracilaria verrucosa*" from North Carolina.

Yamamoto (1978) viewed the spermatangial parent cell initial as being an outer cortical cell that may produce a branched system covering the inner surface of the conceptacle. Our results, in contrast, show that the spermatangial parent cell primordium is an intercalary cortical cell in *G. verrucosa.* Repeated division of this intercalary initial generates a branched filament composed of spermatangial parent cells that line an intercellular space, forming the male conceptacle.

Taxonomic Implications

Christensen (1987) rejected Silva's (1980) proposal to conserve the family name Gracilariaceae Nägeli (1847:240, as 'Gracilariae'), in as much as Article 69 of the Botanical Code (Voss et al. 1981) removed the stigma of illegitimacy for initially superfluous family names. Sjöstedt (1926) was unable to assign *Gracilaria* to any existing family and referred its taxonomic status to *Sedis incertae.* Kylin (1930) proposed the Gracilariaceae, a *nomen nudum,* without formal diagnosis to contain the genera *Gracilaria* and *Cordylecladia* J. Agardh (1852:702) after confirming Sjöstedt's (1926) observations on the unique post-fertilization features of *Gracilaria.* He placed the Gracilariaceae in the Nemastomatales (as Nemastomales) based on the presence of an auxiliary cell that is a segmental cell of an ordinary vegetative filament. Kylin (1932) subsequently transferred the Gracilariaceae, along with the other families of Nemastomatales, to the Gigartinales.

The most recent characterization of the family is found in Kylin (1956) which lists eight genera: *Gracilaria* Greville, *Corallopsis* Greville, *Melanthalia* Montagne, *Tylotus* J. Agardh, *Curdiea* Harvey, *Gelidiopsis* Schmitz, *Ceratodictyon* Zanardini and *Gracilariophila* Setchell et Wilson *in* Wilson. After Kylin's death, his wife, E. Kylin, added *Gracilariopsis* Dawson in the appendix (Kylin 1956:578). Since Kylin's analysis of the family, several generic revisions have been proposed. Dawson (1949) segregated *Gracilariopsis* from *Gracilaria* based primarily on the absence of 'nutritive filaments' (= tubular nutritive cells) in the cystocarp. Dawson (1949) excluded *Tylotus* from the Gracilariaceae based on the presence of zonate tetrasporangia in the type species, *T. obtusatus* (Sonder) J. Agardh. Later, Kraft (1977a) transferred *Tylotus* to the Dicranemaceae. Norris (1987) synonymized *Gelidiopsis* with *Ceratodictyon* based on vegetative similarities and transferred six species of *Gelidiopsis* to *Ceratodictyon,* which he retained in the Gracilariaceae. Kraft (pers. comm.) is investigating both of these genera and has indicated that they belong in the Rhodymeniales. Dawson (1954) reduced *Corallopsis* to taxonomic synonymy under *Gracilaria,* judging that the thallus constrictions characteristic of *Corallopsis* do not justify generic segregation. Chang and Xia (1963:120) erected *Polycavernosa,* which they distinguished primarily by the occurrence of spermatangia in multiple cavities and the presence of darkly staining 'nutritive filaments' originating from the bases of gonimoblasts.

The Gracilariaceae currently contains seven genera: 1) *Gracilaria* Greville 1830 [type: *G. confervoides* (Stackhouse) Grev. 1830:123; basionym: *Flagellaria confervoides* Stackhouse 1809:92], correct name: *Gracilaria verrucosa* (Hudson) Papenfuss; 2) *Gracilariopsis* Dawson 1949:40 [type: *Gracilariopsis sjoestedtii* (Kylin) Dawson 1949:40; basionym: *Gracilaria sjoestedtii* Kylin 1930:55; correct name: *G. lemaneiformis* (Bory) Dawson, Acleto et Foldvik 1964:59; basionym: *Gigartina lemaneiformis* Bory 1828:151]; 3) *Gracilariophila* Setchell et Wilson *in* Wilson 1910: 81 [type: *G. oryzoides* Setchell et Wilson *in* Wilson 1910:81]; 4) *Polycavernosa* Chang et Xia 1963:120 [type: *P. fastigiata* Chang et Xia 1963:120]; 5) *Curdiea* Harvey 1855:333 [type: *Curdiea laciniata* Harvey 1855:333, correct name *C. calophyllis* (Areschoug) Fredericq *ined.*; basionym *Gracilaria calophyllis* Areschoug 1854:350]; 6) *Melanthalia* Montagne 1843:296 [type: *M. obtusata* (Labillardière) J. Agardh 1852:614; basionym: *Fucus obtustatus* Labillardière 1806:111]; 7) *Congracilaria* Yamamoto 1986 [type: *C. babae* Yamamoto 1986:287].

All members of the Gracilariaceae are characterized by the following features of the female reproductive system: 1) a supporting cell of intercalary origin that bears a two-celled carpogonial branch flanked by two or more sterile branches, 2) direct fusion of cells of the sterile branches onto the persistent carpogonium, 3) isolation of the carpogonium upon degeneration of the hypogynous cell leading to the formation of a generative fusion cell that cuts off several gonimoblast initials, 4) formation of an ostiolate pericarp, 5) schizogenous development of the cystocarp cavity, and 6) secondary fusions of specialized, terminal gonimoblast cells with cells of gametophytic tissues. Diagnostic characters that separate genera include 1) the extent of incorporation of additional vegetative cells into the fusion cell, 2) the presence and nature of any nutritive tissues, 3) the specific character of any secondary fusions, and 4) the mode of gonimoblast maturation (Fredericq 1988).

The Gracilariaceae is currently placed in the Gig-

artinales Schmitz (1892:18), an order characterized by auxiliary cells present before fertilization, diploidization of the auxiliary cell by an external connection (connecting cell, connecting filament, or short tube), and pit-connections whose pit plugs lack plug caps (for a review see Kraft and Robins 1985, Gabrielson and Garbary 1986). In contrast, the Gracilariaceae is characterized by the absence of auxiliary cells and connecting cells or connecting filaments. Instead, the cells of the sterile branches borne on the supporting cell fuse with the carpogonium to form a generative fusion cell that cuts off multiple gonimoblast initials.

The Gracilariaceae are agarophytes that may contain significant amounts of agarose. In addition, some illustrations of pit plugs of *Gracilaria* suggest the presence of a single plug cap layer and a striated plug core similar to that present in the Gelidiaceae (Hommersand and Fredericq 1988).

Because of its unique combination of characters, we believe that the Gracilariaceae should not be retained in the Gigartinales. We therefore propose Gracilariales *ordo novus* to include the Gracilariaceae Nägeli 1847.

Diagnosis of Order

Gracilariales Fredericq et Hommersand, ord. nov.

Initium incrementi carposporarum et tetrasporarum similime et sequitur Dumontia*-typum germinationis. Incrementum apparenter uniaxiale cum rapido occulto axis centralis. Thalli pseudoparenchymati. Parietes cellularum et matrix intercellularis continentes agarum et alteras substantias propias agarophytorum. Synapses primae et secundae continentes obturamenta cum singuli strato capitularis. Gametophyti masculi ferentes uninucleata initia parentalia genita aut cellulis superficialibus et formantes stratum continuum superficiale, aut genita in filamenta e cellulis intercalaribus corticalibus et formantes depressiones conceptaculoides. Spermatangia genita singula e cellulis parentibus per divisionem transversalem aut per septa concavo-convexa. Gametophyti feminei praebentes extrinseca fila carpogonialia bicellulata tecta a latere duobus aut pluribus filis sterilibus genitis super cellulam supportantem originantem intercalarem. Filum carpogoniale constans ex cellula hypogyna binucleata ad multinucleatem et carpogonio distali cum trichogyna stricta, ephemera ad modice persistenti. Cellulae auxiliares et fila connectentia absentia. Nucleus diploideus remanens in carpogonio post fecundationem, cum conjunctione directa cellularum filorum sterilium intus carpogonium persistens, cum generatione cellulae fusionis genialis ex qua emergunt nonnulla initia gonimoblasti. Crypta crystocarpii hemisphaerica schizogenea formatur. Derivationes gonimoblasti formantes secundas fusiones cum cellulis telarum gametophytarum. Synapses secundae formantur inter cellulas interiores gonimoblasti, producentes hemisphaericam, multinucleatam telam gonimoblastam sterilem; cellulae exteriores gonimoblasti transformantur in catenas vel fasciculos carposporangiorum obovatorum ad sphaericum. Cystocarpia elevata, compacta, hemisphaerica, cum pericarpio ostiolato. Tetrasporangia vel bisporangia dispersa admodum sub superficie ramorum vel inclusa in nemathecio. Initia tetrasporangiorum transformata e cellulis apicalis derivatis divisione longitudinali cellularum corticis exterioris. Tetrasporangia cruciata divisa.*

Free living members typically exhibiting a *Polysiphonia*-type life history, with occasional mixed gametophytic and tetrasporophytic stages found on a single specimen. Thalli erect or recumbent; adelphoparasites forming hemispherical pustules. Early development of carpospores and tetraspores identical and following *Dumontia*-type germination.

Thalli of various forms, terete to flattened, fleshy to cartilaginous, unbranched or branched in various ways; main axes arising from a single discoid or coalesced holdfast. Growth apparently uniaxial with rapid concealment of central axis. Branches initiated through transformation of surface cells into apical initials of cortical files that divide by concavo-convex septa, followed by oblique or transverse division of the subterminal cell. Thalli pseudoparenchymatous throughout, consisting of a small-celled cortex of uninucleate or multinucleate cortical cells, and a central medulla of large, multinucleate to enucleate cells. Cuticle overlying entire cortical zone. Secondary pit-connections numerous, linking adjacent medullary and subterminal cortical cells. Cell walls and intercellular matrix containing agar and other polymers characteristic of agarophytes. Pit plugs with one cap layer.

Sexual plants typically dioecious. Male gametophytes bearing uninucleate spermatangial parent cells produced either from surface cortical cells and forming a superficial continuous layer or generated in filaments from intercalary cortical cells and forming conceptacle-like depressions. Spermatangia produced singly from spermatangial parent cells by transverse division or by concavo-convex septa.

Female gametophytes bearing outwardly directed two-celled carpogonial branches flanked by two or more sterile branches borne on a multinucleate supporting cell of intercalary origin. Carpogonial branch consisting of a binucleate to multinucleate hypogynous cell and a distal, uninucleate carpogonium with a straight, ephemeral to moderately persistent trichogyne. Auxiliary cells and connecting cells and filaments absent. Diploid nucleus remaining in carpogonium following fertilization; cells of sterile branches fusing directly onto persistent carpogonium, isolation of the carpogonium with degeneration of the hypogynous cell leading to formation of a generative fusion cell that cuts off multiple gonimoblast initials laterally and toward the thallus surface. Cystocarp cavity hemispherical, developing schizogenously. Gonimoblast derivatives establishing secondary fusions with cells of gametophytic tissues in the floor of the cystocarp, or in both the floor and overlying pericarp. Cells of the sterile inner gonimoblast tissue interconnected by secondary

pit-connections; outer gonimoblast cells transformed into chains or clusters of obovoid to subspherical carposporangia. Cystocarps protruding, compact, hemispherical, with a thick-walled ostiolate pericarp.

Meiosporangia (tetrasporangia) or bisporangia scattered just beneath the unmodified surface of branches or embedded within a nemathecium. Tetrasporangial initials transformed from apical cells derived through longitudinal division of outer cortical cells. Tetrasporangia broadly ellipsoid to spherical and cruciately divided into four uninucleate spores.

Typical family: Gracilariaceae.

Taxonomic Relationships of the Gracilariales

Gabrielson and Garbary (1986) postulated that the Gigartinales, as currently understood, is paraphyletic and probably also polyphyletic. The proposed separation of the Gracilariaceae from the Gigartinales sensu Kraft and Robins (1985) and its elevation to ordinal status helps to clarify the Gigartinales.

The closest living relatives of the Gracilariales are most likely found among extant agarophytes that may contain significant amounts of agarose; namely, the Gelidiales Kylin (1923) and Ahnfeltiales Maggs and Pueschel (1989). The Ahnfeltiales have pit plugs that lack both pit plug caps and cap membranes (Maggs and Pueschel 1989), whereas both the Gelidiales and Gracilariales appear to have pit plugs with one plug cap and cap membranes (see Hommersand and Fredericq 1988). In all three orders the spermatangial mother cell cuts off a single spermatangium, usually by a transverse division. Thalli are pseudoparenchymatous with secondary pit-connections abundant in all three orders. A carpogonial branch is either absent or atypical, as in the Gracilariales. The gonimoblast develops directly from the carpogonium and typical auxiliary cells are absent. Nutrition of the carposporophyte is supported by extensive fusions with gametophytic tissues in all three orders. These include special nutritive filaments (Gelidiales), outer cortical cells with the formation of an external nemathecial pustule (Ahnfeltiales), or cells in the floor of the cystocarp or pericarp with the formation of a schizogenous cystocarp cavity (Gracilariales). Although the Ahnfeltiales, Gelidiales and Gracilariales may be related ancestrally, they are sufficiently distinct to justify recognition of all three orders.

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Abbott, I. A. 1985. New species of *Gracilaria* Grev. (Gracilariaceae, Rhodophyta) from California and Hawaii. *In* Abbott, I. A. & Norris, J. N. [Eds.] *Taxonomy of Economic Seaweeds.* Sea Grant Program, University of California, La Jolla, pp. 115–21.

Abbott, I. A., Chiang, Y. M., Fredericq, S., Norris, J. N., Tsuda, R. T., Xia, B. & Yamamoto, H. 1985. The red alga *Gracilaria* Greville (Gracilariaceae, Gigartinales): Introduction. *In* Abbott, I. A. & Norris, J. N. [Eds.] *Taxonomy of Economic Seaweeds.* Sea Grant Program, University of California, La Jolla, pp. 67–8.

Agardh, J. G. 1852. *Species Genera et Ordines Algarum, . . .* C. W. K. Gleerup, Lund. Vol. 2(3), pp. [2] + 787–1291 [1139–58 omitted].

Areschoug, J. E. 1854. Phyceae novae et minus cognitae . . . *Uppsala Soc. Sci. Nova Acta* 3(1):328–72.

Bird, C. J. & McLachlan, J. 1982. Taxonomy of *Gracilaria*: taxonomic criteria in *Gracilaria* (Rhodophyta, Gigartinales). *Bot. Mar.* 25:557–62.

Bird, C. J., van der Meer, J. P. & McLachlan, J. 1982. A comment on *Gracilaria verrucosa* (Huds.) Papenf. (Rhodophyta:Gigartinales). *J. Mar. Biol. Assoc. U.K.* 62:453–9.

Bory de Saint-Vincent, J. B. G. M. 1827–1829. Vol. I, pts. 1–6: Cryptogamie. *In* Duperrey, L. I. [Ed.] *Voyage Autour du Monde, . . . La Coquille . . .* A. Bertrand, Paris. Pts. 1–2:[iii]–96 pp., [13] pls. Pts. 3–4:97–200, [13] pls. Pts. 5–6:201–301, [13] pls.

Buffham, T. H. 1893. On the antheridia, etc., of some Florideae. *Quekett Microsc. Club* (London), Ser. II 5(3):291–305.

Chang, C. F. & Xia, B. 1963. *Polycavernosa*, a new genus of the Gracilariaceae. *Stud. Mar. Sinica* 3:119–26, pls. 1–2.

——— 1985. On *Gracilaria asiatica* sp. nov. and *G. verrucosa* (Huds.) Papenfuss. *Oceanol. Limnol. Sin.* 16(3):175–80.

Christensen, T. 1987. Report of the Committee for Algae. *Taxon* 36:66–9.

Dawson, E. Y. 1949. Studies of northeast Pacific Gracilariaceae. *Allan Hancock Found. Publ. Occ. Pap.* 7:1–105.

——— 1954. Notes on tropical Pacific marine algae. *Bull. So. Calif. Acad. Sci.* 53:1–7.

——— 1961. Marine red algae of Pacific Mexico. Part 4. Gigartinales. *Pac. Naturalist* 2:191–343.

Dawson, E. Y., Acleto, C. & Foldvik, N. 1964. The seaweeds of Peru. *Nova Hedwigia* 13:[iii] + 111 pp., pls. 1–81.

Delivopoulos, S. G. & Tsekos, I. 1983. A light microscope study of carposporophyte development in *Gracilaria verrucosa* (Hudson) Papenfuss. *Ann. Bot.* 52:317–23.

Dixon, P. S. & Irvine, L. M. 1977. *Seaweeds of the British Isles.* Vol. 1. Rhodophyta. Part 1. Introduction, Nemaliales, Gigartinales. British Museum (Nat. Hist.), London, xi + 252 pp.

Drew, K. 1954. The organization and inter-relationships of the carposporophytes of living Florideae. *Phytomorphology* 4:55–69.

Edelstein, T., Chen, L. C. M. & McLachlan, J. 1978. Studies on *Gracilaria* (Gigartinales, Rhodophyta): reproductive structures. *J. Phycol.* 14:92–100.

Fredericq, S. 1988. Developmental morphology and systematics of the Gracilariaceae (Rhodophyta). Ph.D. thesis, University of North Carolina, Chapel Hill, 340 pp.

Fredericq, S. & Hommersand, M. H. 1989. Comparative morphology and taxonomic status of *Gracilariopsis* (Gracilariales, Rhodophyta). *J. Phycol.* 25:228–41.

Fredericq, S. & Norris, J. N. 1985. Morphological studies on some tropical species of *Gracilaria* Grev. (Gracilariaceae, Rhodophyta): taxonomic concepts based on reproductive morphology. *In* Abbott, I. A. & Norris, J. N. [Eds.] *Taxonomy of Economic Seaweeds.* California Sea Grant Program, University of California, La Jolla, pp. 137–55.

Gabrielson, P. W. & Garbary, D. 1986. Systematics of red algae. *CRC Crit. Rev. Pl. Sci.* 3:325–66.

Greig-Smith, E. 1954. Cytological observations on *Gracilaria multipartita. Brit. Phycol. J.* 2:4–5.

Greuter, W. et al. [Eds.] 1988. *International Code of Botanical Nomenclature* (Berlin Code). Koeltz Sc. Books.

Greville, R. K. 1830. *Algae Britannicae, . . .* MacLachlan & Stewart, Edinburgh, [iii] + lxxxviii + 218 pp., pls. 1–19.

Harvey, W. H. 1855. Short characters of some new genera and

species of algae discovered on the coast of the colony of Victoria, Australia. *Ann. Mag. Nat. Hist. (London)* 5:332–6, pl. viii.

Holmgren, P. K., Keuken, W. & Schofield, E. K. 1981. Index herbariorum. I. The herbaria of the world, 7th ed. *Regnum Veg.* 106:1–452.

Hommersand, M. H. & Fredericq, S. 1988. An investigation of cystocarp development in *Gelidium pteridifolium* with a revised description of the Gelidiales (Rhodophyta). *Phycologia* 27: 254–72.

——— 1989. Sexual reproduction and cystocarp development. *In* Cole, K. M. & Sheath, R. G. [Eds.] *Biology of the Red Algae.* Cambridge University Press, Cambridge. In press.

Hudson, W. 1762. *Flora Anglica* . . . [Ed. 1]. J. Nourse . . . et C. Moran, London, vii + [8] + 506 + [23] pp.

Irvine, L. M. & Dixon, P. S. 1982. The typification of Hudson's algae: a taxonomic and nomenclatural reappraisal. *Bull. Br. Mus. Nat. Hist.* (Bot.) 10(2):91–105.

Kraft, G. T. 1977a. Studies of marine algae in the lesser known families of the Gigartinales (Rhodophyta). II. The Dicranemaceae. *Aust. J. Bot.* 25:219–67.

——— 1977b. Transfer of the New Zealand red alga *Tylotus proliferus* (Gracilariaceae, Gigartinales) to the genus *Gracilaria*. *New Zealand J. Bot.* 15:495–502.

Kraft, G. T. & Robins, P. A. 1985. Is the order Cryptonemiales (Rhodophyta) defensible? *Phycologia* 24:67–77.

Kylin, H. 1923. Studien über die Entwicklungsgeschichte der Florideen. *K. svenska Vetensk.-Akad. Handl.* [ser.4], 63(11):1–139.

——— 1930. Über die Entwicklungsgeschichte der Florideen. *Lunds Univ. Årsskr.*, N.F. Avd. 2, 23(6):1–104.

——— 1932. Die Florideenordnung Gigartinales. *Lunds Univ. Årsskr.*, N.F. Avd. 2, 28(8):1–88.

——— 1956. *Die Gattungen der Rhodophyceen.* C. W. K. Gleerup, Lund, [XV] + 673 pp.

Labillardière, J. J. H. 1806. *De Novae Hollandiae Plantarum Specimen* . . . Vol. 2, pts. 15–27. Dominae Huzard, Paris, 130 pp., pls. 141–265.

Lejolis, A. 1863. *Liste des algues marines de Cherbourg,* . . . J. B. Baillière et Fils, Paris, 168 pp., 6 pls.

Linnaeus, C. 1763. *Species Plantarum,* 2nd ed., Vol. 2. L. Salvii, Holmiae, pp. 782–1684.

Maggs, C. A. & Pueschel, C. M. 1989. Morphology and development of *Ahnfeltia plicata* (Rhodophyta): proposal of Ahnfeltiales ord. nov. *J. Phycol.* 25:333–51.

Montagne, J. P. F. C. 1843. Quatrième centurie de plantes cellulaires . . . *Ann. Sci. Nat. Bot.* Sér. 2, 20:352–79.

Nägeli, C. W. 1847. Die neuern Algensysteme . . . *Neue Denkschr. allg. schweiz. ges. Naturwiss.* 9(2):[i] + 275 pp., pls. 1–10.

Norris, R. E. 1987. The systematic position of *Gelidiopsis* and *Ceratodictyon* (Gigartinales, Rhodophyceae), genera new to South Africa. *S. Afr. J. Bot.* 53:239–46.

Ohmi, H. 1958. The species of *Gracilaria* and *Gracilariopsis* from Japan and adjacent waters. *Mem. Fac. Fish., Hokkaido Univ.* 6: 1–66.

Oliveira, J. C. 1969. Recherches sur le développement et les organes reproducteurs des *Gracilaria* de la Manche. *Rev. de Scienc. Biol.* A(2):11–49.

Oza, M. 1976. Studies on Indian *Gracilaria*. II. The development of reproductive structures of *Gracilaria corticata*. *Bot. Mar.* 19:107–14.

Papenfuss, G. F. 1950. Review of the genera of algae described by Stackhouse. *Hydrobiologia* 2:181–208.

——— 1951. Problems in the classification of the marine algae. *Svensk Bot. Tidskr.* 45:4–11.

Reading, R. P. & Schneider, C. W. 1986. On the male conceptacles of two terete species of *Gracilaria* (Rhodophyta, Gigartinales) from North Carolina. *J. Phycol.* 22:395–98.

Rodriguez de Rios, N. 1986. Sobre la verdadera identidad de la llamada *Gracilaria verrucosa* (Hudson) Papenfuss en Venezuela. *Ernstia* 38:32–9.

Schmitz, F. 1889. Systematische übersicht der bisher bekannten Gattungen der Florideen. *Flora* 72:435–56, pl. 21.

——— 1892. [6. Klasse Rhodophyceae] 2. Unterklasse Florideae. *In* Engler, A., *Syllabus* . . . *Grosse Ausgabe.* Berlin, pp. 16–23.

Silva, P. C. 1952. A review of nomenclatural conservation in the algae from the point of view of the Type method. *Univ. Calif. Publ. Bot.* 25:241–323.

——— 1980. Names of classes and families of living algae with special reference to their use in the Index Nominum Genericorum (Plantarum). *Regnum Veg.* 103:1–156.

Sjöstedt, L. G. 1926. Floridean studies. *Acta Univ. Lund,* N.F. Avd. 2, 22(4):1–95.

Stackhouse, J. 1809. Tentamen marino-cryptogamicum, . . . *Mem. Soc. Imp. Naturalistes Moscou* 2:50–97, pls. 5–6.

Thuret, J. & Bornet, E. 1878. *Etudes Phycologiques* . . . *Analyses d'Algues Marines* . . . G. Masson, . . . , Paris, [v] + iii + 105 pp., pls. 1–51.

Voss, E. G. et al. [Eds.] 1981. *International Code of Botanical Nomenclature* (Sydney Code). Dr. W. Junk, The Hague, 445 pp.

Wilson, H. L. 1910. *Gracilariophila,* a new parasite on *Gracilaria confervoides*. *Univ. Calif. Publ. Bot.* 4:75–84, pls. 12–3.

Wittmann, W. 1965. Aceto-iron-haematoxylin-chloral hydrate for chromosome staining. *Stain Technol.* 40:161–4.

Yamamoto, H. 1975. The relationships between *Gracilariopsis* and *Gracilaria* from Japan. *Bull. Fac. Fish., Hokkaido Univ.* 26: 217–22.

——— 1978. Systematical and anatomical study of the genus *Gracilaria* in Japan. *Mem. Fac. Fish., Hokkaido Univ.* 25:97–152.

——— 1986. *Congracilaria babae* gen. et sp. nov. (Gracilariaceae), an adelphoparasite growing on *Gracilaria salicornia* of Japan. *Bull. Fac. Fish. Hokkaido Univ.* 37:281–90.