THE LIFE HISTORY AND DEVELOPMENT OF *MURRAYELLA PERICLADOS* (C. AGARDH) SCHMITZ (RHODOPHYTA, RHODOMELACEAE) IN CULTURE

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ABSTRACT — The life history of *Murrayella periclados* was examined in culture. It was observed to possess a typical *Polysiphonia*-type life history. Plants initially developed as monosiphonous filaments and subsequently produced polysiphonous axes which characterize typical *Murrayella* morphology. Gametophytic and tetrasporophytic plants matured in approximately 2 months. Both monoecious and dioecious gametophytes were observed in culture with spermatangia developing prior to the appearance of procarps in either case. Spermatangial plants are described for the genus for the first time.

RÉSUMÉ — Le cycle complet de développement de *Murrayella periclados* a été obtenu en culture; il est du type *Polysiphonia*. Les thalles se développent tout d'abord en filaments monosiphonés qui produisent ensuite des axes polysiphonés caractéristiques du g. *Murrayella*. Gamétophytes et tétrasporophytes sont fertiles après deux mois environ. Des gamétophytes monoïques et dioïques ont été observés en culture, les spermatocystes apparaissant dans chaque cas avant l'initiation des procarpes. Les gamétophytes mâles sont décrits pour la première fois dans ce genre. (traduit par la rédaction).

KEY WORDS : life history, development, culture, *Murrayella periclados*, Rhodomelaceae, Rhodophyta.

INTRODUCTION

Murrayella periclados (C. Agardh) Schmitz (1893) was originally described as a species of *Hutchinsia* by C. AGARDH (1828) from St. Croix, U.S. Virgin Islands, although its distribution includes the tropical Pacific (HOLLENBERG, 1968; KAPRAUN and BOWDEN, 1978; SCHNETTER and BULA MEYER, 1982). In the Caribbean *Murrayella* is a characteristic plant of mangrove swamps, seaside caverns and rocks in shallow quiet water. It is generally found in association with *Bostrychia*, *Caloglossa* and *Catenella* epiphytic on the aerial and prop roots of mangroves where it forms dark reddish brown, densely felted

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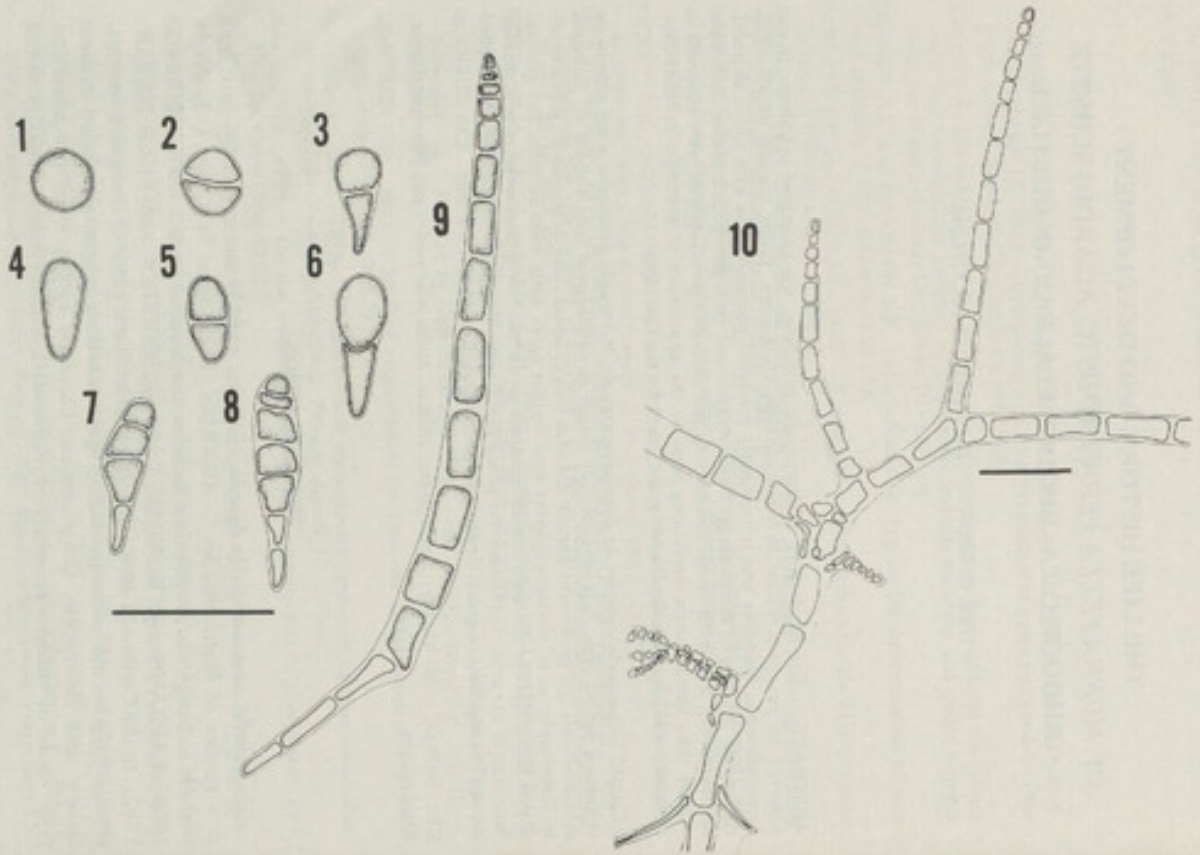


Figures 1-10 : Murrayella periclados sporelings. Figures 1-6 : zero to 48 hours. Figures 7 & 8 : four and five day old sporelings.

cushions (BØRGESEN, 1918). These plants comprise what POST (1955) referred to as the «Bostrychietum complex».

Details of *Murayella periclados* morphology have been provided by BØRGESEN (1918) and TAYLOR (1960). *Murrayella periclados* possesses both creeping and erect filaments which have four pericentral cells and lack a cortical layer. Growth is by means of an apical cell which cuts off flat discoid segments proximally. From these axial cells, branch initials are produced spirally before division to form pericentral cells; branching is thus exogenous. Pigmented monosiphonous ramelli, branched or unbranched and with or without a polysiphonous base, are alternately radially disposed on polysiphonous main axes (BØRGESEN, 1918).

Considerably less information is available concerning reproductive events in *Murrayella*. Gametophytic plants are not commonly collected, and spermatangial plants have been reported by KYLIN (1956) as being unknown. We report here the complete life history of *Murrayella periclados* in culture.

METHODS AND MATERIALS

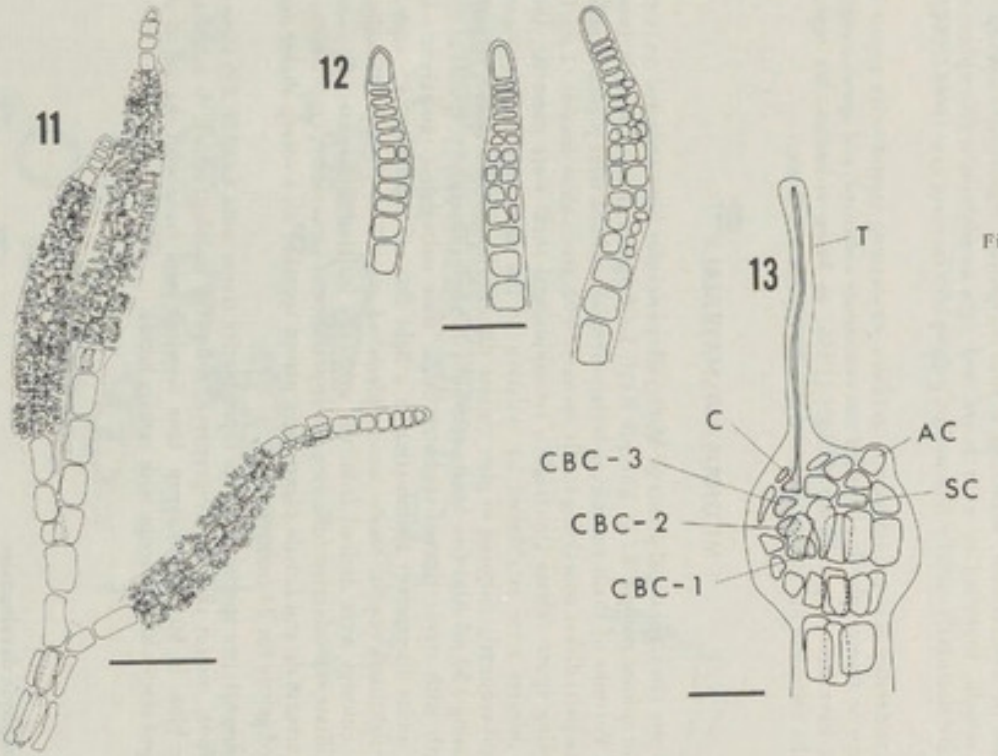
Mature tetrasporangial plants of *Murrayella periclados* were collected on roots of *Rhizophora mangle* L. at Enrique Reef, La Parguera, Puerto Rico in June, 1985. Portions of thalli containing tetrasporangial stichidia were placed in polystyrene petri dishes containing sterile seawater with germanium dioxide (5 mg/l). Following spore release (overnight), tetrasporangial thalli were removed. Unialgal cultures were established by isolation of the discharged spores. Plants were subsequently cultured in glass culture dishes (100 mm diameter x 80 mm) containing 150 ml Alga-Gro Media (Carolina Biological Supply) prepared at 1/4 strength with sterile filtered seawater. Algae were cultured at temperature of 25°C under fluorescent illumination at a light flux of 20µmol.m^{-2}s^{-1} with a 16:8 (light:dark) cycle. Media were replenished every 7 to 10 days. All subsequent cultures were derived from the originally isolated tetraspores. In order to facilitate reproduction following the appearance of gametophyte reproductive structures, male and female plants were placed together on a rotary shaker and agitated gently for 24 to 36 hours.

Representative specimens of all reproductive stages were fixed in 4 % formalin. Whole mount slides were prepared, following staining with 1 % acidified aniline blue, in 60 % Karo syrup. Line drawings were made with the aid of an Olympus research microscope with camera lucida.

RESULTS

Gametophyte development

Spores from wild *Murrayella periclados* tetrasporangia were spherical and measured 18 to 22 µm in diameter following release (fig. 1). Most became



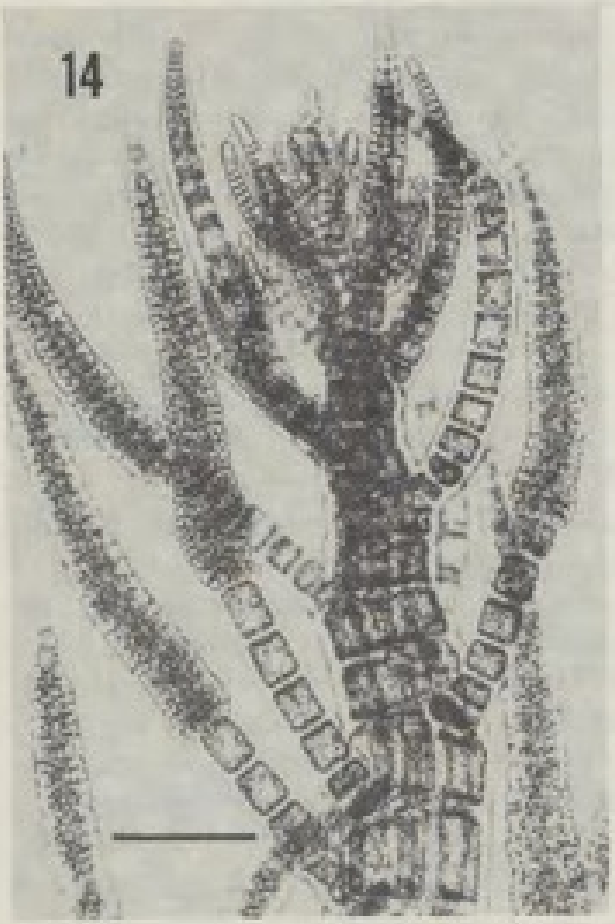
Figures 11-13 : *Murrayella periclados*. Figure 11 : three mature spermatangial branches. Scale : 100 µm. Figure 12: immature spermatangial branches. Scale : 50 µm. Figure 13: procarp showing 4-celled carpogonial branch arising from supporting cell. AC : apical cell; C: carpogonia; CBC : carpogonial branch cell; SC : supporting cell; T : trichogyne. Scale : 50 µm.

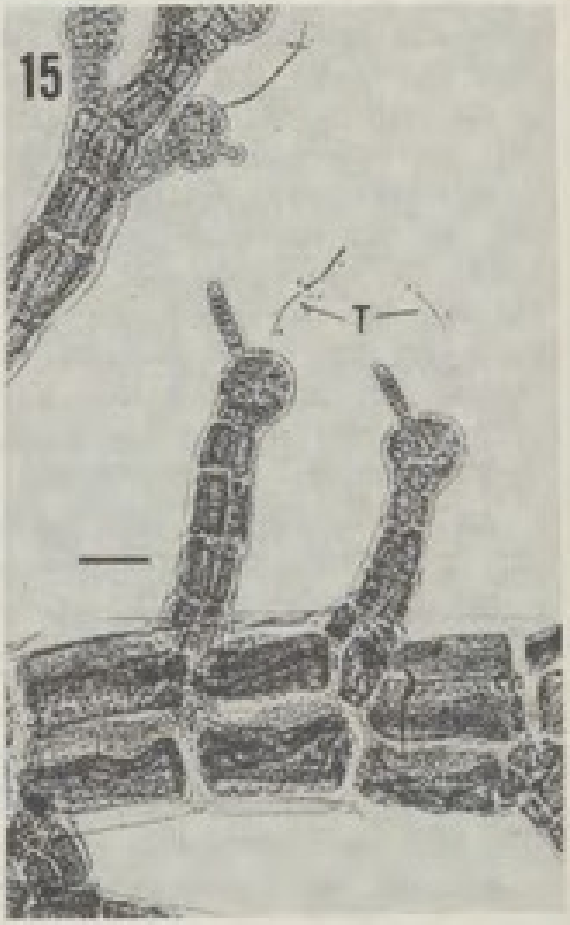
attached to the culture vessel bottom within 24 hours, although in some cases attachment was loose or plants failed to attach altogether. Within 12 to 24 hours after attachment, first indication of germination was evidenced by elongation of the spore (fig. 4). This elongation was followed by a transverse division that separates two nearly identical cells (fig. 5). Less commonly the first cleavage occurred without elongation of the germinating spore (fig. 2). The resultant two cells represented initials of a vegetative pole and a rhizoidal pole (CHEMIN, 1937). The initials developed in opposite directions contributing to the length of the uniseriate sporeling (figs. 3, 6-9). Early sporeling development was of the *Ceramium* type and is typical of the order Ceramiales (CHEMIN, 1937). The rhizoidal filament soon became less pigmented and by the fourth of fifth day following germination it developed lateral rhizoids. Few plants were observed to develop more than a single rhizoidal filament from the rhizoidal pole.

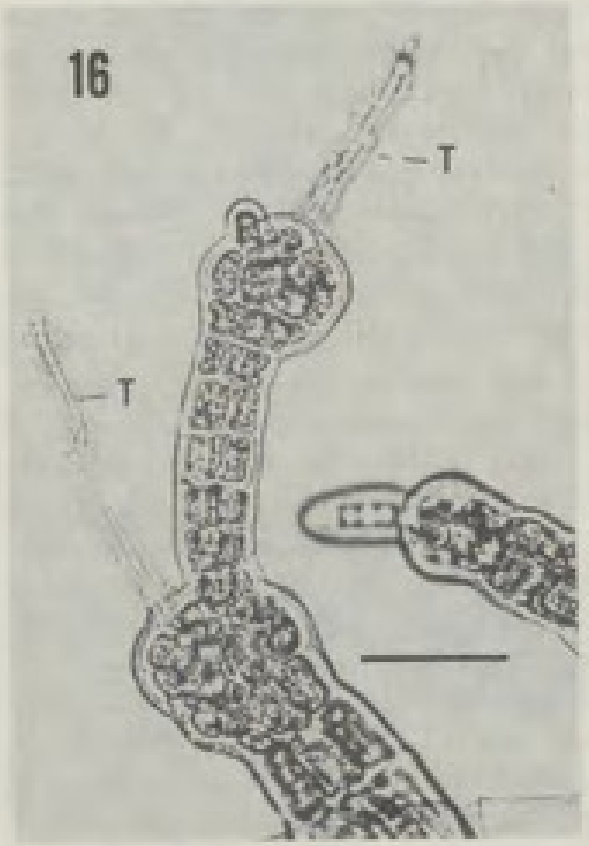
An erect, pigmented, uniseriate filament developed from the vegetative pole (figs. 7-9). The sporeling was generally four cells in length within 48 hours of germination. After 8 to 10 days, the primary filament initiated uniseriate laterals. At approximately 15 days, polysiphonous branches first developed (fig. 10). They were generally initiated from cells located near the base of the vegetative monosiphonous filament and were prostrate, creeping along the substrate. These polysiphonous axes subsequently produced erect polysiphonous branches. With continuous growth of the polysiphonous axes, the original monosiphonous filament became obscured.

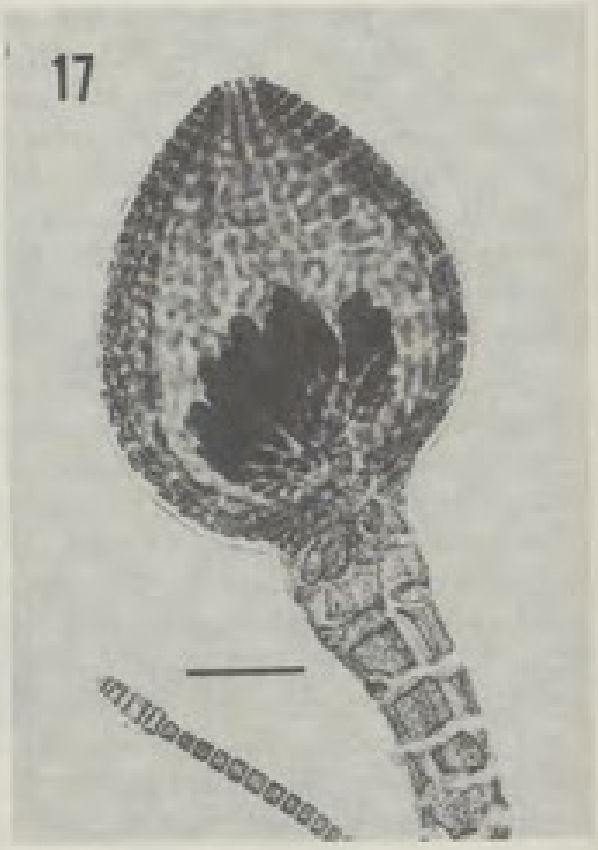
Following 40 to 50 days in culture, gametophytes became reproductive with male reproductive structures preceding the development of procarps. Gametophytes were mostly dioecious but approximately 20 % of the cultured plants were monoecious (fig. 20). Spermatangial clusters were elongate-cylindrical in shape and were concentrated, but not limited to, apical areas (fig. 14). Spermatangial branches generally terminated in a uniseriate file of cells (fig. 11). Mature and immature spermatangial branches are illustrated in figures 11 and 12. Axial cells of the spermatangial branch cut off pericentral cells that redivide to give rise to spermatangial mother cells. The mother cells produced either one or two spermatia. The mature structure measured from 240 to 400 µm in length and from 35 to 45 µm in diameter. Individual spermatia were 3 to 5 µm in diameter.

Procarps were consistently observed 8 days after the appearance of spermatangia on either dioecious or monoecious plants. Procarps were developed at the tips of fertile polysiphonous branches or very rarely at an intercalary position on polysiphonous laterals. The sub-apical segment of the fertile branch cut off five pericentral cells, one of them (the supporting cell) giving rise to the four-celled carpogonial branch. The supporting cell was differentiated from other pericentral cells by its frequently irregular shape. Cells of the carpogonial branch (fig. 13) were somewhat variable in shape and size. Carpogonial branch cell (CBC) -1 was generally rounded and measured 8 to 10 µm in diameter. CBC-2 was round or ovate and generally the same size as CBC-1. The hypogynous cell, CBC-3, was usually the smallest in the carpogonial branch and was irregular in shape, generally measuring 4 to 6 µm in diameter. The carpogonium was triangu-









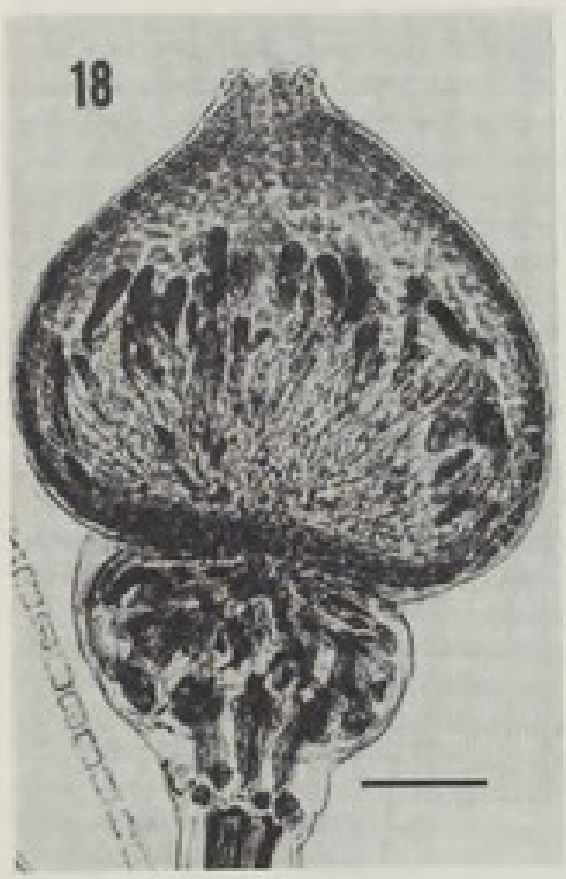
lar in shape and measured 6 to 7 µm at its broadest point. The carpogonium bore a trichogyne that when fully developed, extended to over 200 µm beyond the surface of the young pericarp (fig. 13). Trichogynes were not ephemeral structures in cultured *Murrayella periclados* as they persisted well into cystocarp maturation (fig. 19). The pericarp was present prior to the complete differentiation of the carpogonial branch making post fertilization events difficult to evaluate. The mature urceolate cystocarps (fig. 17) measured 340 to 520 µm in length and 300 to 510 in width at their broadest point.

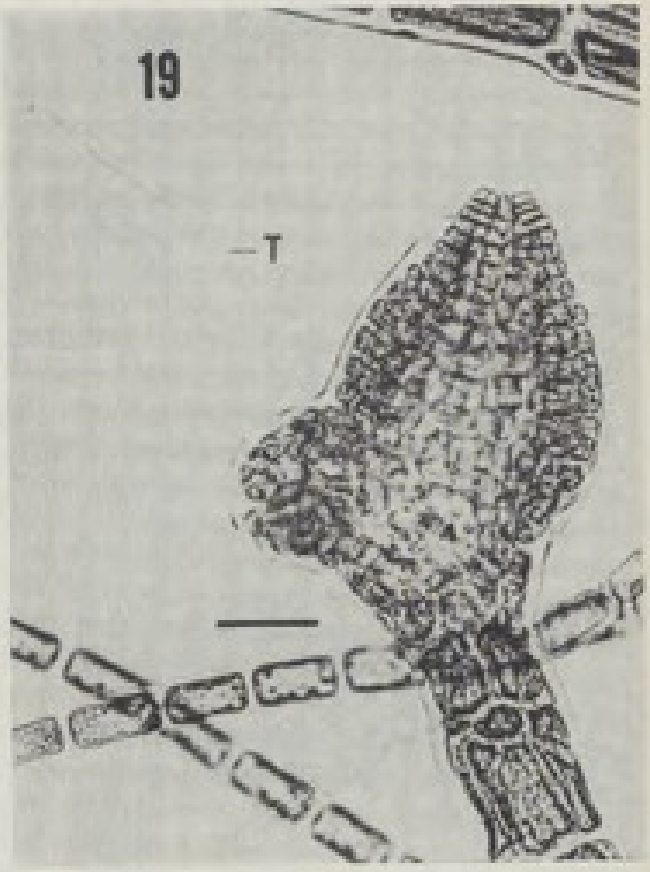
Several unusual reproductive features were noted which we have not observed in field-collected plants. With some regularity, supporting cells were observed to possess two carpogonial branches. While actual fertilization was not confirmed, the apparent result of fertilization of both carpogonia borne within the same procarp, is what we have designated «double cystocarps» (fig. 19). Despite the fact that fertilization was not observed (although numerous spermatia attached to trichogynes were regularly seen), it is assumed to have taken place as isolated female dioecious gametophyte plants did not develop cystocarps parthenogenetically. Frequently the apical cell of the female fertile branches continued development after formation of the pericarp (fig. 15). On rare occasions a second terminal cystocarp was borne on the same fertile branch (fig. 16). Unusual cystocarps with enlarged bases (fig. 18) were also commonly seen in cultured plants.

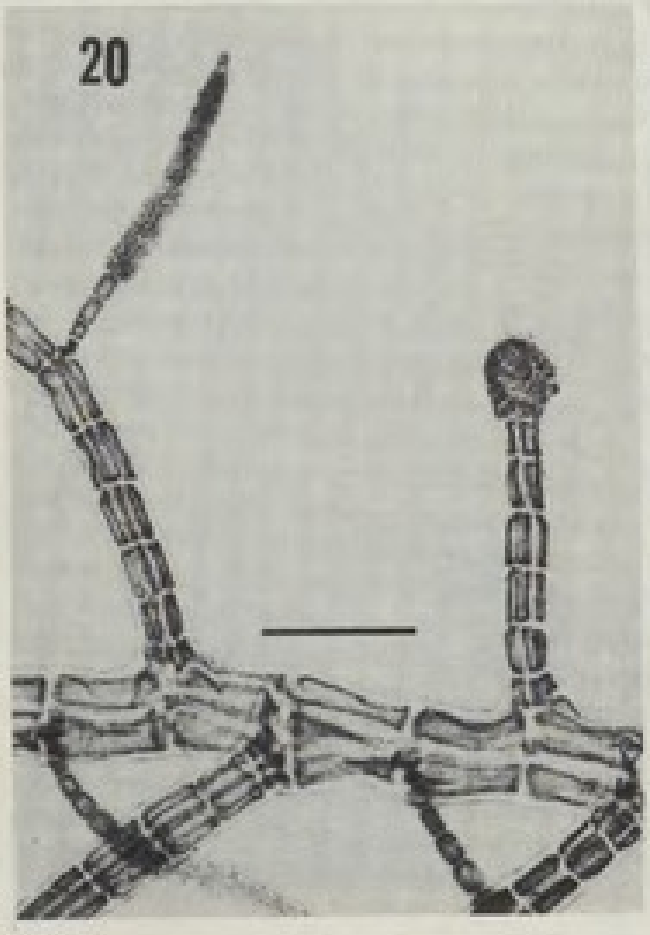
Sporophyte development

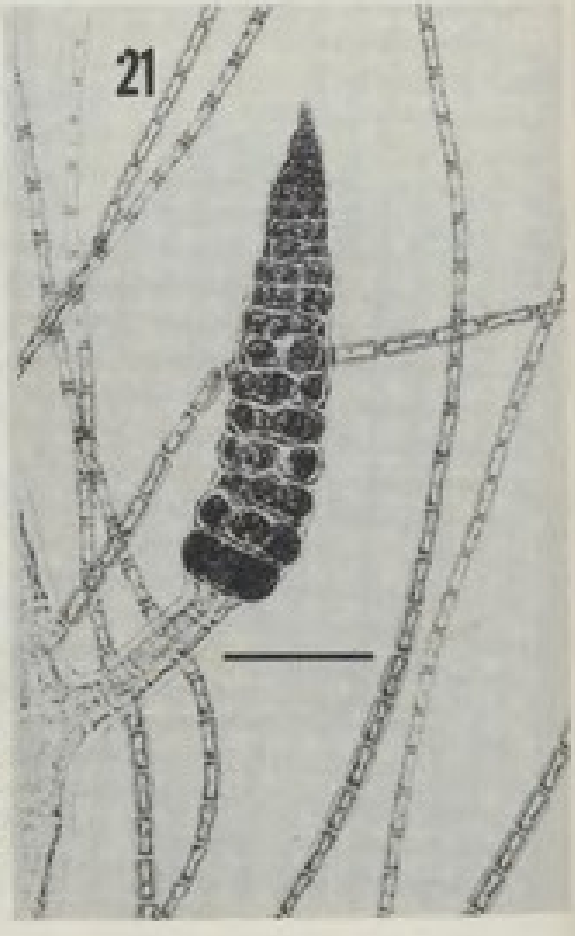
After liberation from the cystocarp, carpospores were spherical in shape and had a diameter of from 40 to 50 µm. Their attachment to the substratum and vegetative development were similar to that described for gametophyte sporelings above. Tetrasporophytic plants grew and matured to produce tetrasporangial stichidia within 60 to 75 days. Tetrasporangia were produced in nearly cylindrical stichidia borne terminally on distichously ramifying polysiphonous branchlets. The lower parts of these branchlets were provided with side branches as is the case for sterile branchlets. Each of the four pericentral cells in each fertile segment produced a tetrasporangium with three cover cells. Mature stichidia (fig. 20) measured 450 to 950 µm in length and 85 to 100 µm in diameter. Tetrasporangia measured an average of 50 µm.

Figures 14-17 : *Murrayella periclados*. Figure 14 : spermatangial clusters located at branch apex. Scale : 100 µm. Figure 15 : young cystocarps, terminal on fertile branches. Apical cells of fertile branches remain active, each producing a monosiphonous filament distally. Trichogynes (T) with attached spermatia are evident. Scale : 50 µm. Figure 16 : two young cystocarps on same fertile branch, resulting from continued growth of apical cell of fertile branch after formation of proximal cystocarp. Trichogynes with attached spermatia are evident. Scale : 50 µm. Figure 17 : normal maturing cystocarp. Scale : 100 µm.









DISCUSSION

The life history of *Murrayella periclados* in culture was a regular *Polysiphonia*-type. Plants retained their characteristic morphology throughout the course of culture observations. Time required for completion of the life history was approximately 4 months which is within the usual range of 2.5 to 6 months reported by WEST and NORRIS (1966) for completion of life histories in most Florideophycideae. It is likely that time for completion of life history could have been shortened by using light flux densities greater than 20 $\mu mol.m^{-2} s^{-1}$.

Life history periods of two months or less have been reported for *Callithamnion byssoides* Arnott (EDWARDS, 1969), *Champia parvula* (C. Agardh) Harvey (STEELE and THURSBY, 1981), and *Gelidium coulteri* Harvey (MACLER and WEST, unpubl.).

Spermatangia are described for *Murrayella* for the first time and are similar to those described for *Polysiphonia fastigiata* Greville by GRUBB (1924) in which pericentral cells cut off several spermatangial mother cells that in turn cut off one or two spermatia. An examination of dried specimens plus freshly collected *M. periclados* revealed a low incidence of plants identifiable as male or female. This is difficult to explain given the readiness with which gametophytic plants developed into functional reproductive units in culture. Spermatangia in some red algae may be short-lived entities which can thus be easily overlooked although the spermatangia of *M. periclados* in culture were generally persistent for months once initiated. It is common, however, for reproduction of all phases of most Ceramiales and other rhodophytes in culture to continue indefinitely (WEST, personal communication).

Procarps within the order Ceramiales generally contain a single carpogonial branch (HOMMERSAND, 1963). The not uncommon presence of two carpogonial branches in addition to the more regular presence of a single carpogonial branch produced by a single supporting cell was an unexpected finding. Two carpogonial branches borne by the same supporting cell are seen in some Delesseriaceae genera (HOMMERSAND, 1963; WYNNE, 1983) and in a few Ceramiaceae genera (HOMMERSAND, 1963).

Gametophytes of most Ceramiales are known to be dioecious (BOLD and WYNNE, 1985). The presence of both a monoecious and dioecious condition in *Murrayella periclados* was thus additionally unexpected. The degree to which this is naturally prevalent is unknown. *Polysiphonia flexicaulis* (Harvey) Collins and *Callithamnion baileyi* Harvey have been labelled as being either monoecious

Figures 18-21 : *Murrayella periclados*. Figure 18 : cystocarp with unusual enlargement at base. Scale : 100 µm. Figure 19 : «Double cystocarp», the apparent result of successful fertilization of two carpogonial branches borne on the same supporting cell. One trichogyne (T) is still evident. Scale : 50 µm. Figure 20 : monoecious plant. Scale : 200 µm. Figure 21 : tetrasporangial stichidia. Scale : 200 µm.

or dioecious (TAYLOR, 1957) although WHITTICK and WEST (1979) have reported the latter species to be monoecious. HASSINGER-HUIZINGA (1952) discussed the occasional formation of monoecious *Callithamnion corymbosum* (Smith) Lyngbye in culture. WEST (1970) additionally reported finding a monoecious strain of the otherwise dioecious *Rhodochorton purpureum* (Lightfoot) Rosenvinge based on culture studies. The need for further research into the phenomenon of monoecism in the rhodomelacean genus *Murrayella* is clearly indicated.

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