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Life history of *Erythrocladia subintegra* ROSENVINGE (Bangiales, Rhodophyceae) in culture

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The life history of *Erythrocladia subintegra* ROSENVINGE, used primarily as food of gastropods (such as abalones and top shells) in aquaculture farms, was studied under laboratory conditions. Reproducing by monospores, its life history was completed in 5-11 days at different temperature-daylength regimes.

Monospores produced asexually were liberated at 15°C SD and were collected on glass slides. They were grown on culture dishes containing modified PROVASOLI's enriched seawater (ESI) and incubated at different temperature-daylength conditions.

Irrespective of daylengths, onset of maturation and polystromatic formation of the disc-plantlets were faster at higher temperatures (20°-25°C) than in lower temperatures (10°-15°C) and morphological variations were also observed.

Key Index Words: *Bangiales*; Erythrocladia subintegra; *Erythropeltidaceae*; *life history*; *monospore.*

The family Erythropeltidaceae (Bangiales, Bangiophyceae, Rhodophyta) includes 6 genera (MURRAY *et al.*, 1972): *Erythrocladia* ROSENVINGE, *Erythropeltis* SCHMITZ, *Erythrotrichia* ARESCHOUG, *Porphyropsis* ROSENVINGE, *Smithora* HOLLENBERG, and *Membranella* HOLLENBERG and ABBOTT. Studies on this family are limited to descriptive morphology and little is known of their life histories. The genus *Erythrocladia*, consisting of 14 species (NICHOLS and LISSANT 1967) is cosmopolitan in distribution. *Erythrocladia subintegra* is widely distributed in the Atlantic, Indian and Pacific Oceans. In Japan, its presence is recorded from Hokkaido to Ryukyu (TANAKA 1952). Life history studies on *Erythrocladia* are scanty as in other members of the family. The developmental variations of monospore germlings in *Erythrocladia subintegra* using different substrates were studied by NICHOLS and LISSANT (1967) in Missouri.

Erythrocladia subintegra is a microscopic, purplish red plant which is roundish or elliptical on surface view and whose cells divide dichotomously toward the marginal portion forming a monostromatic disc with continuous margin.

Abalones (*Haliotis*) or top shells (*Batillus*) are known to feed principally on algae, which they scrape off the rock by means of radula. At the Sea Organism Culture Center in Obama, Fukui, *Erythrocladia subintegra* are grown on vinyl plates as primary food of young abalones. To determine qualitatively the effect of temperature-daylength conditions on their growth, the authors studied the life history of *Erythrocladia subintegra* for aquaculture purposes.

Materials and Methods

Algal specimens found attached to vinyl plates were collected at the Sea Organism Culture Center, Katsumi, Obama, Fukui (35°32′N and 135°43.5′E) on October 29, 1983.

Fertile microscopic plants bearing monosporangia were scraped and placed in 90 mm ×20 mm disposable plastic Petri dishes containing autoclaved seawater. Liberated monospores at 15°C Short Day (10 h light : 14 h dark) were collected on glass slides and grown in culture dishes containing modified PROVASOLI's enriched seawater medium (ESI, TATEWAKI 1966). Newly released monospores were incubated at different temperature-daylength conditions : 25°C LD, 20°C LD and SD, 15°C LD and SD and 10°C SD; where LD (Long Day) is (a) 16 h light : 8 h dark or (b) 14 h light : 10 h dark ; SD (Short Day) is 10 h light : 14 h dark.

Monospores and developing plantlets grown on glass slides were permanently stained using HEIDENHAIN's iron haematoxylin-orange G (JENSEN 1962) to observe their respective nuclei.

Results

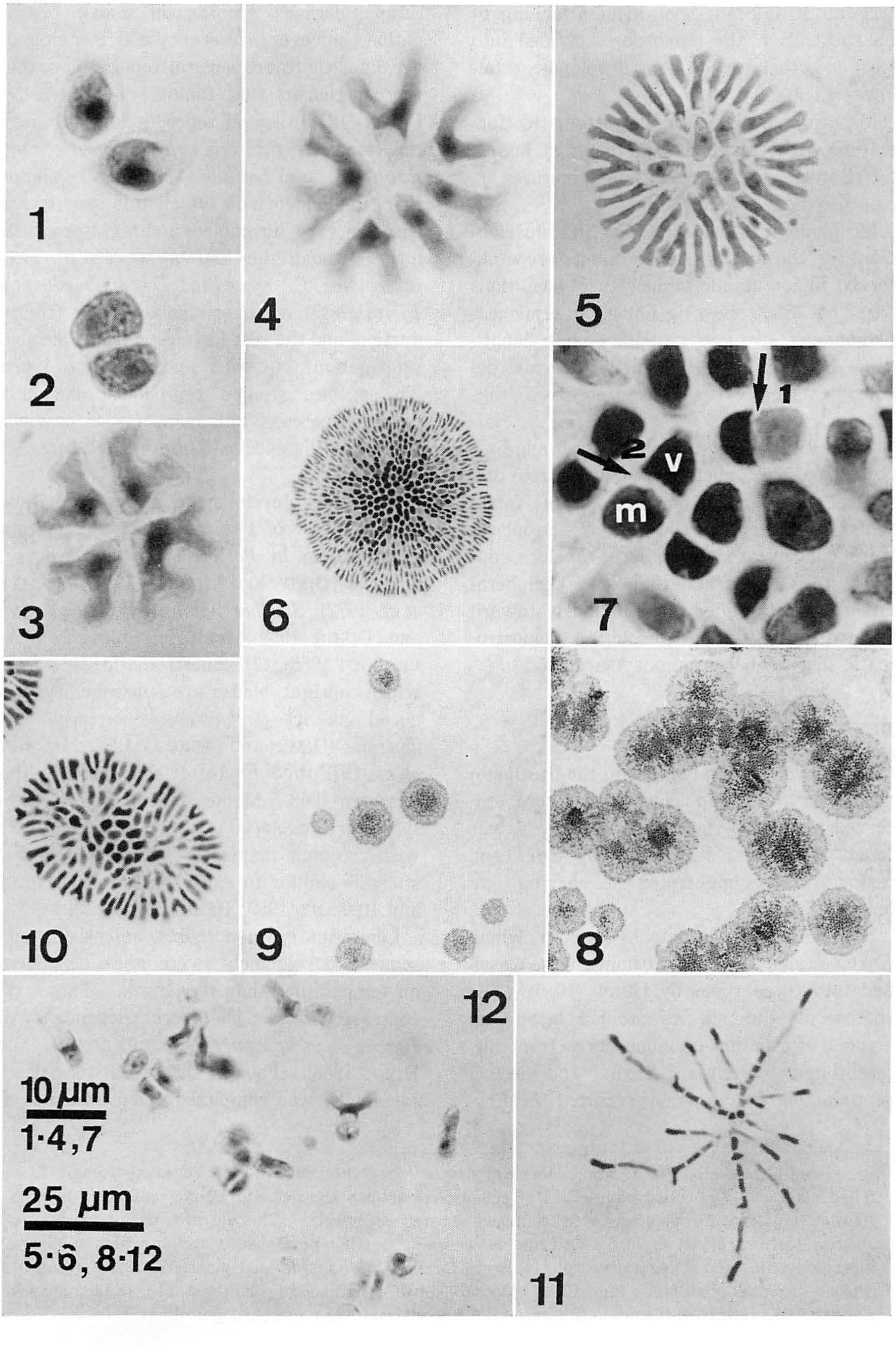
Monospores measuring 4.0–10.0 μm in diameter were released from monosporangium at 15°C SD. They were spherical in shape, containing a parietal, cup-shaped chloroplast and a pyrenoid (Fig. 1).

Settled monospores at 15°C SD underwent nuclear division followed by chloroplast division which resulted in the formation of 2-celled plantlets measuring 7.5 μm in diameter after 1–2 days (Fig. 2). A nucleus was found positioned in the center of each cell. Successive bipartitions or simultaneous cleavages occurred as a result of cytoplasmic divisions. Further divisions of the plantlet took place with peripheral lobing and subsequent dichotomous divisions of closely associated filaments, thus 4-celled plantlets, measuring 15.0–22.0 μm in diameter were formed (Fig. 3). The plantlet became a monostromatic circular disk of adherent branching filaments, measuring 30.5 μm in diameter after 4 days (Fig. 4). With further growth, the plantlet formed two layers (37.5–52.5 μm in diameter) after 5 days (Fig. 5). The mature plantlets which were centrally polystromatic consisting of 6–14 layers measuring 53.0–99.0 μm in diameter (Fig. 6). Each vegetative cell centrally located in a polystromatic disc divided into 2 unequal portions (Fig. 7, arrow 1). The larger cell remained initially vegetative (Fig. 7, v by arrow 2) until further division, while the smaller cell developed into a sporangium (Fig. 7, m by arrow 2) from which a monospore was released. Monospores were released from the sporangium on the 7th day of culture. Adjacent plants having polystromatic areas at their center anastomosed with each other, thus the typical circular disc characteristic of *Erythrocladia subintegra* was obscured (Fig. 8).

When matured plantlets bearing monosporangia were inoculated at 25°C LD, 20°C LD & SD, 15°C LD & SD and 10°C SD, monospores were released within two days in all temperature-daylength conditions ex-

Table 1. Temperature-daylength conditions and period of time (days) for the formation of 2 to 7-celled plantlets, monostromatic plantlets and polystromatic plants and for the release of monospores in *Erythrocladia subintegra*.

Temperature-daylength conditions	Formation of 2 to 7 celled plantlets (days)	Formation of monostromatic plantlets (days)	Formation of polystromatic plantlets (days)	Release of monospore (days)
25°C LD (b)	2	3	4	5
20°C LD (a)	2	3	5	6
20°C SD	2	4	5	6
15°C LD (b)	4	4	5	7
15°C SD	5	6	7	8
10°C SD	5	7	9	11


1
2
3
4
5
6
7
1
2
v
m
8
9
10
11
12
10 µm
1·4,7
25 µm
5·6, 8·12

cept at 10°C SD. Soon after attaching to the substratum, the monospores divided into two and their subsequent development followed (Table 1).

Maturity as well as polystromatic formation of the plantlet were faster at higher temperatures than in lower temperatures irrespective of daylengths.

Morphological variations in the development of the monospore germlings were observed under higher temperature conditions (20° and 25°C) showing oblong to branched-filamentous plantlet ca. 100 μm in length (Figs 10–11). Likewise, early abnormal development of monospores was observed (Fig. 12).

Stained with iron-haematoxylin-orange G, the dark blue nuclei of monospores and young developing plantlets were found centrally located in the cell. Chloroplasts were concentrated around the nucleus, appearing to be darker than the peripheral areas. Cells divided dichotomously toward the marginal portion forming a monostromatic disc with continuous margin.

Discussion

The developmental stages in the life history of *Erythrocladia subintegra* were observed under laboratory conditions. It was found to complete its life history under each temperature regime tested at varying day lengths.

Spores in the Bangiophyceae to which *Erythrocladia subintegra* belongs were classified into three types by DREW (1956). The spores of the species studied belong to Type 1—formation of monospores from differentiated sporangia. Sexual and asexual reproductions have been reported in Erythropeltidaceae; records on sexual propagation, however, are scanty and incomplete. To date, there are several reports on sexual reproduction in this family. As cited by DIXON (1973), sexual reproduction has been reported in *Erythrocladia, Erythropeltis, Erythrotrichia*, and *Smithora*, although he claims that "the evidence on which this reports are based is very incomplete and highly conflicting". Among the known species of *Erythrocladia, E. recondita, E. vagabunda* and *E. insignis* have questionable sexual reproduction, and the rest have established asexual propagation (NICHOLS and LISSANT 1967). The species studied reproduced asexually by monospores. This supports a previous study on this species (NICHOLS and LISSANT 1967).

Monospore development in Erythropeltidaceae is either of two ways: (1) filamentous germling—as in *Porphyropsis coccinea* var. *dawsonii* (DIXON and MURRAY 1981, MURRAY *et al.* 1972), *Smithora naiadum* (RICHARDSON and DIXON 1969), and *Erythrotrichia* (HEEREBOUT 1968); (2) monostromatic disc—from which upright blades are subsequently produced as in *Porphyropsis coccinea* var. *dawsonii* (DIXON and MURRAY 1981, MURRAY *et al.* 1972) and *Erythrotrichia carnea* (HEEREBOUT 1968). Monospores of *Erythrocladia subintegra* developed into monostromatic discs with creeping filaments. The result of our study is similar to earlier reports (NICHOLS and LISSANT 1967, HEERBOUT 1968).

Liberation of monospores and their subsequent development were more dependent on temperature than daylength. This is in contrast with the photoperiod dependency of *Bangia fuscopurpurea* (RICHARDSON and DIXON 1968). Though the life history of *E. subintegra* was completed under all tempera-

Figs 1–9 Developmental stages of *Erythrocladia subintegra*. 1. Settled monospores. 2. 2-celled plantlet. 3. 4-celled plantlet. 4. 6-celled circular disc plantlet. 5. Young centrally 2-layered plantlet. 6. Mature plantlet with centrally located sporangia. 7. Central part of a mature plantlet (arrow 1: just divided two cells and arrow 2: further developed stage at which a monosporangium (m) and a vegetative cell (v) were formed. 8. Anastomosed plantlets. 9. A group of typical disc-like plantlets. Figs 10–12 Morphological variations of plantlets. 10. Oblong-shaped plantlet. 11. Branched-filamentous plantlet. 12. Abnormal one-two celled germlings.

tures tested, the onset of maturation varied. Higher temperatures, irrespective of day-lengths favored earlier development of monostromatic plantlets. Likewise, plantlets became polystromatic at the center earlier at higher temperatures than in lower ones.

Morphological variations of the plantlets were more frequently observed at higher temperatures. A previous study (NICHOLS and LISSANT 1967) showed that developmental variations in *E. subintegra* depended on (1) size of monospores, such that small monospores developed into filamentous plantlets, while large ones grew from elliptical to irregular disc-type plantlets, and (2) type of substrates. In our study, we confirmed the influence of temperature on morphological variations, but the influence of different types of substrates was not ascertained. Anastomosis was common between adjacent and maturing plantlets. This observation confirms earlier study on this species (NICHOLS and LISSANT 1967).

Early development of characteristic monostromatic circular disc and maturity of *Erythrocladia subintegra* were favored at higher temperatures (20-25°C). The typical shape of this species was retained for several days so long as the temperature was maintained at 15°C. For studies on life history and taxonomy, maintenance of cultures at lower temperatures is preferable, but for purposes of food production for gastropods such as abalones and top shells, higher temperature is more practical. The accessibility of food for gastropods in aquaculture is thus sustained.

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アニシア Q. フルタドーポンセ・梅崎　勇: イソハナビ *Erythrocladia subintegra* (紅藻類ウシケノリ目）の生活史の研究

アワビ・サザエ等の幼貝の飼料となっている紅藻類ウシケノリ目のイソハナビを実験室内で培養して，その生活史を研究した。本種は単胞子によって生殖し，10°-25°C 長日及び短日条件下で5～11日で生活史を完了する。また，日長に関係なく高温（20～25°C）の方が低温条件（10～15°C）よりは藻体の生長及び単胞子の形成と放出が早くなる。このように，単胞子によって短期間に増殖がくり返されるので貝類の初期飼料として有用な藻であるといえる。(606 京都市左京区北白川追分町　京都大学農学研究科熱帯農学専攻水産資源学研究室)