

## *Leachiella pacifica* Kugrens (Choreocolacaceae, Rhodophyceae), new to Japan

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A parasitic red alga was found growing on the thalli of *Polysiphonia* sp. and *Pterosiphonia bipinnata* (Postels et Ruprecht) Falkenberg collected from Nemuro, eastern Hokkaido, Japan. We identified this red alga as *Leachiella pacifica* Kugrens based on the results of morphological observations. The thallus of this alga consisted of an external pustule and internal penetrating filaments. The pustule was small, spherical, and milk-white in color without pigmentation. Internal filaments were uniseriate, frequently branched, and penetrated into host tissue to form secondary pit-connections with host cells. Gametophytes and tetrasporophyte were similar in vegetative morphology. *L. pacifica* is similar to *Choreocolax polysiphoniae* Reinsch, reported from the Atlantic Ocean, in vegetative structure, host specificity and carposogonial branch system. But *Leachiella* differs in the structure of the cystocarp, which is the most important feature to distinguish genera in the family Choreocolacaceae. *L. pacifica* has gonimoblasts which radiate from central large fusion cell and cut off terminally carposporangia. On the other hand, *C. polysiphoniae* is provided with gonimoblasts that construct a conceptacle-like structure and cut off carposporangia inwardly.

*Key Index Words:* alloparasite—Choreocolacaceae—Choreocolax polysiphoniae—Crytonemiales—*Leachiella pacifica*—morphology—parasitic red algae—Rhodophyceae.

A parasitic red alga, *Leachiella pacifica* Kugrens is a common species on Pacific coasts of America. The alga was first reported as *Choreocolax polysiphoniae* Reinsch because of the similarities of the vegetative morphology and the procarp structure (Kugrens and West 1972, Goff and Coleman 1984). Kugrens and West (1972) studied the ultrastructure of this plant. Goff and Coleman (1984) examined nuclear events during carposporogenesis, spermatangium formation, post-fertilization and tetrasporogenesis. Kugrens (1982) established the genus *Leachiella* emphasizing its cystocarp structure.

On the Japanese coast, the Choreocolacaceae was represented by only *Gelidiocolax mammillata* Fan et Papenfuss (Yoshida 1977). This is the second report on

the family Choreocolacaceae in Japan.

### Material and Method

The plants were collected from Nemuro peninsula, east Hokkaido, Japan, from April, 1987 to October 1988. The plants were growing exclusively on the thalli of *Polysiphonia* sp. and *Pterosiphonia bipinnata* (Postels et Ruprecht) Falkenberg which were growing in the littoral zone or were drifting ashore. Collected material was kept alive at a low temperature or was fixed with formalin sea water, and carried back to the laboratory. Preparations for microscopic observations were made by cutting the material with a razor blade or freezing microtome, or smashing tissue on the slide-glass after treatment with 10% sodium hydroxide solution for 12–24 hours and following the wash with distilled water for 12–24 hours, stained with aniline blue solution and mounted in glycerol

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sea water.

### Observation

Mature plants of *L. pacifica* were collected in every season from April 1987 to October

1988 (Table 1). The fresh thalli were milk-white in color without noticeable pigmentation (Fig. 1A). Color of the thalli became brown with formalin sea water treatment. The thalli consisted of external pustule and internal filaments penetrating through the host

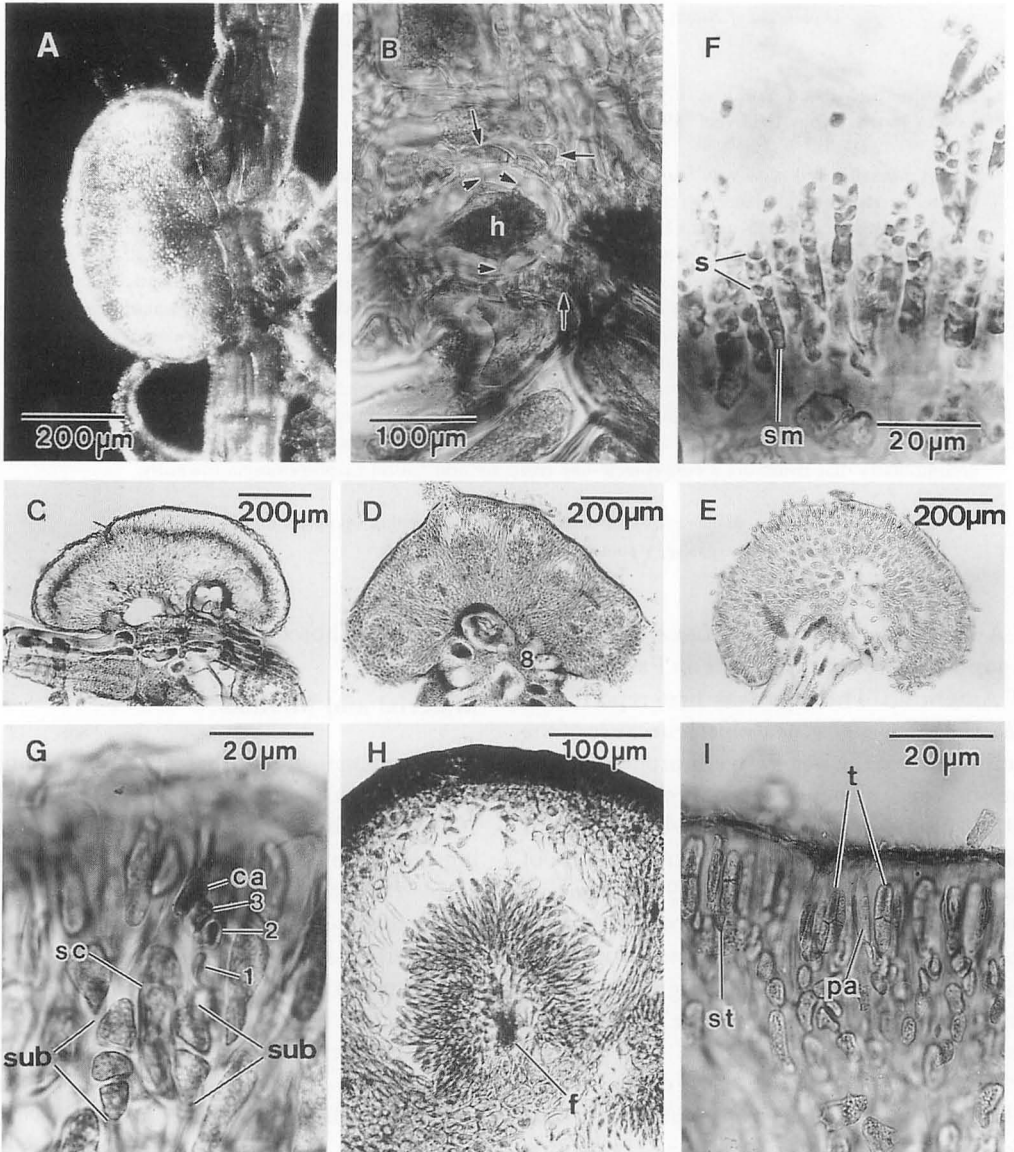


Fig. 1. *Leachiella pacifica* Kugrens. A. Thallus growing on the *Polysiphonia* sp. B. Penetrating filaments (arrows) secondary pit-connections (arrow heads) with host cells. C. Male gametophyte. D. Female gametophyte with mature carposporophyte. E. Tetrasporophyte. F. Spermatangia formed in cortical layer of male gametophyte. G. Mature carpogonial branch. H. Cystocarp. I. Tetrasporangia: ca, carpogonium; f, fusion cell; h, host cell; pa, paraphysis; s, spermatangia; sc, supporting cell; sm, spermatangial mother cell; st, stalk cell; sub, subsidiary cell; t, tetrasporangium; 1-3 first, second and third cell of carpogonial branch.

Table 1. *Leachiella pacifica* Kugrens. The months when the mature specimens were collected in Nemuro peninsula during study period, from 1987 to 1988.

Generation	month											
	Jan.	Feb.	Mar.	Apr.	May	Jun.	Jul.	Aug.	Sep.	Oct.	Nov.	Dec.
Male Gametophyte						+		+				+
Female Gametophyte		+		+		+		+				+
Tetrasporophyte		+		+		+		+				+

tissue. The pustules were spherical or hemispherical in shape (Fig. 1A). They were usually smaller than 1 mm in diameter and mature plants were sometimes up to 1.6 mm in diameter. Medullary cells of pustules were ellipsoidal or cylindrical, and were up to 80  $\mu\text{m}$  long, 40  $\mu\text{m}$  broad. The cells became smaller outwardly. Cortical cells were spherical or obovoid in shape, measuring 8-15  $\mu\text{m}$  long, 5-8  $\mu\text{m}$  broad.

The penetrating filaments were uniseriate and frequently branched. Cells of the filament were cylindrical or ellipsoidal, 6-17  $\mu\text{m}$  broad, 1-4 times as long as breadth. They formed secondary pit-connections with host cells (Fig. 1B). Vegetative structure of male

and female gametophytes and tetrasporophyte were similar in morphology (Fig. 1C-D).

Mature male gametophytes were covered with particularly thick cuticle. They formed spermatangia on entire cortical layer of their pustules (Fig. 1C). Cortical cells transformed into elongated spermatangial mother cell, which cut off the spermatangia by oblique walls. Several (usually 4 to 7) spermatangia were alternately arranged in chain (Fig. 1F, Fig. 2A). They were angular at first, and were 3-5  $\mu\text{m}$  in diameter.

Numerous procarps were formed in cortical layer of a female gametophyte pustule. They were provided with a 4-celled carpogonial

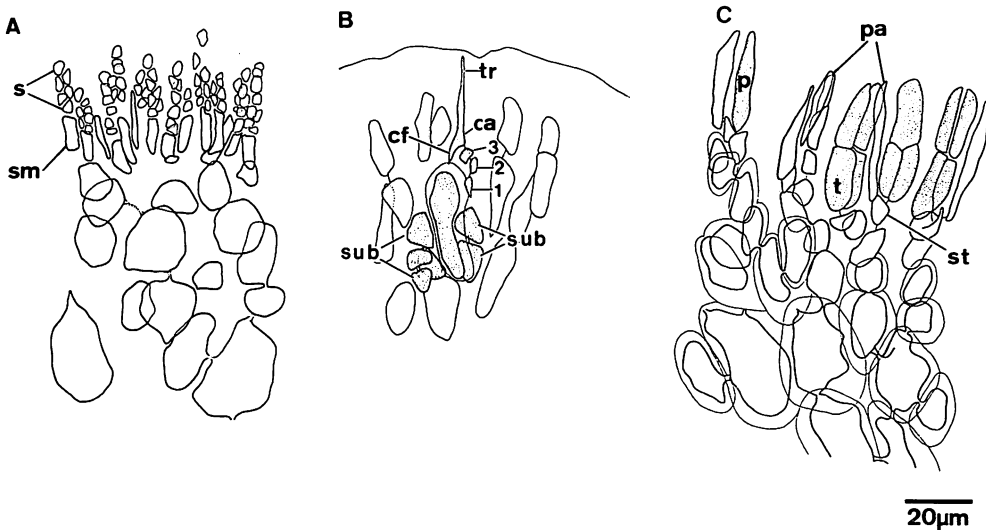


Fig. 2. *Leachiella pacifica*. A. Spermatangia. B. Carpogonial branch, connecting with supporting cell. C. Tetrasporangia formed with cruciately divided. ca, carpogonium; cf, connecting filament; p, tetrasporangial primordium; pa, paraphysis; s, spermatangium; sm, spermatangial mother cell; st, stalk cell; sub, subsidiary cells; t, tetrasporangium; tr, trichogyne; 1-3 first, second and third cell of carpogonial branch.

branch and a few groups of subsidiarily cells (Fig. 1G, Fig. 2B). An initial cell of a carpogonial branch was cut off from upper side of a supporting cell and develops into a carpogonial branch. A carpogonium was conical, and extended a trichogyne to the surface of pustule. Two or three groups of subsidiarily cells were formed along a supporting cell. Number of subsidiarily cells in a group was indefinite (Fig. 1G, Fig. 2B). After fertilization, the carpogonium extended a connecting filament to the supporting cell (Fig. 2B). The supporting cell fused with subsidiarily cells after connecting with a fertilized carpogonium. They developed into a large and irregular shaped fusion cell. The fusion cell was up to 110  $\mu\text{m}$  long, 80  $\mu\text{m}$  broad. The fusion cells gave rise to gonimoblasts radiating from them. Gonimoblasts cut off carposporangia terminally. Carposporangia were waterdrop-shaped and were about 20  $\mu\text{m}$  long, 8  $\mu\text{m}$  broad. Spherical or hemispherical carposporophytes were up to 300  $\mu\text{m}$  in diameter, were surrounded by elongated vegetative cells of gametophyte forming pericarp (Fig. 1H). An inconspicuous ostiole was present in the pericarp over the carposporophyte.

Mature tetrasporophytes produced tetrasporangia in the cortical layer of pustules (Fig. 1E). The cortical cells elongated and divided into two cells. An upper cell of them became a tetrasporangial primordium. It divided cruciately and formed four tetraspores. Tetrasporangia were long-ellipsoidal and 20–40  $\mu\text{m}$  long, 10–20  $\mu\text{m}$  broad. Another cells surrounding tetrasporangia continuously divided and formed paraphyses which were laterally arranged along the tetrasporangia. (Fig. 1I, Fig. 2C).

## Discussion

Sturch (1926) established the family Choreocolacaceae based on *Choreocolax polysiphoniae* growing in Atlantic Ocean. Nemuro plants were in agreement with his descriptions of the Choreocolacaceae in the following points. The plant was

holoparasitic without pigmentation. It consisted of small spherical or hemispherical external colorless pustule, and of filaments penetrating through the host tissue. Supporting cells were laterally provided with a carpogonial branch. Therefore this parasitic red alga belonged to the family Choreocolacaceae.

In the Choreocolacaceae, three genera are recognized from northern Hemisphere: *Choreocolax* Reinsch (Sturch 1926), *Harveyella* Schmitz et Reinke (Sturch 1899, 1924, Goff and Cole 1975) and *Leachiella* Kugrens (Kugrens 1982, Goff and Coleman 1984). They are provided with four-celled carpogonial branch, and are distinguished by difference of cystocarp structure (Sturch 1926, Goff and Cole 1975). In *Choreocolax*, gonimoblasts elongate and form conceptacle-like cystocarps. They cut off carposporangia inwardly (Sturch 1926). *Harveyella* has diskshaped cystocarps consisting of gonimoblasts extending through the female gametophyte tissue and surrounded by elongated vegetative cells of gametophyte (Sturch 1924, Goff and Cole 1975). The structure of cystocarp of Nemuro plant was different from these two genera. In Nemuro plant, a large fusion cell gave rise radiated gonimoblasts around forming spherical or hemispherical carposporophytes. These were surrounded by a pericarp consisting of elongated vegetative cells of the gametophyte. An ostiole formed in the pericarp, over the carposporophyte. These features of cystocarp quite agreed with those of *Leachiella pacifica* Kugrens (1982).

The genus *Leachiella* was represented by only one species *L. pacifica* (Kugrens 1982). This species had been identified as *Choreocolax polysiphoniae*, because of its similarities of vegetative and procarp structures and host specificity; *L. pacifica* and *C. polysiphoniae* grew parasitic on *Polysiphonia* spp. (Sturch 1924, Kugrens et West 1973, Goff and Coleman 1984). Kugrens (1982) distinguished *L. pacifica* from *C. polysiphoniae* by the following features, six-celled carpogonial branch, carposporophyte structure and tetrahedral

tetraspore division. On the other hand, Goff and Coleman (1984) proved that carpogonial branch was four-celled, supporting cell functioning as an auxiliary cell and ostioles present. These features were different from descriptions of Kugrens (1982). They did not accept the genus *Leachiella* because of insufficiency of observations of *C. polysiphoniae* from Atlantic Ocean (Goff and Coleman 1984). Our observations agreed with the description of Goff and Coleman (1984).

We accepted the genus *Leachiella*, and identified Nemuro parasitic red alga as *L. pacifica*, because cystocarp structure was the most important features to distinguish genera in the family Choreocolacaceae, and there are clear differences between *Leachiella* and *Choreocolax*.

We noticed certain differences between the plants growing at Pacific coast of America and Nemuro plants. American plants are always smaller than 1 mm in size, and have tetrasporangia divided tetrahedrally (Kugrens 1982, Goff and Coleman 1984). Nemuro plants were sometimes larger than 1 mm in diameter, and tetrasporangia were divided cruciately.

Our plants had one or two tetrasporangial paraphyses. And the alga formed four to seven spermatangia which were cut off by alternating oblique divisions, and were arranged in chain. These features were similar to *Harveyella mirabilis* (Reinsch) Schmitz et Reinke growing in both Pacific and Atlantic Oceans (Sturch 1899, 1924), and were absent in *C. polysiphoniae*. Thus it seems that *L. pacifica* is nearer relative of *H. mirabilis* than of *C. polysiphoniae*.

Taxonomic placement of the family Choreocolacaceae has not been clear (Kugrens 1982, Goff and Cole 1975). Kylin (1956) placed the Choreocolacaceae in the Cryptonemiales based on the procarp structure and post-fertilization events of the Choreocolacaceae that were similar to those of the Kallymeniaceae, one of the families of Cryptonemiales (Kylin 1956). Thus we followed Kylin's opinion and regarded Choreocolacaceae as a member of Cryptonemiales.

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*Leachiella pacifica* Kugrens（コレオコラククス科，カクレイト目）

北海道東部根室半島沿岸でイトグサ (*Polysiphonia* sp.) およびイトヤナギ (*Pterosiphonia bipinnata*) の藻体上に寄生している紅藻を採集した。この寄生藻は、枕状の外生部と宿主組織内に貫入する内生的な糸状体部分より成っていた。外生部は、球形か半球形で乳白色を呈し、直径は最大 1.6 mm に達した。宿主体内に貫入する糸状体は単列で、宿主細胞と二次的な原形質連絡を形成していた。雌雄の配偶体と四分孢子体は同じ形態をとり、イトグサ型生活環を持つ。栄養体、造果枝の形態からコレオコラククス科に属し、4細胞性のプロカルプを備え、大型の融合細胞から造胞糸を周囲に放射状に生じ、球形の果孢子体を形成することから、アメリカ大陸太平洋岸より報告されている *Leachiella pacifica* Kugrens（コレオコラククス科，カクレイト目）と同定した。和名はハネグサヤドリとしたい。(060 札幌市北区北10条西8丁目 北海道大学理学部植物学教室；\*現所属 222 横浜市港北区新横浜3-6-12 日総第12ビルディング 日本エヌ・ユー・エス株式会社)