OBSERVATIONS ON THE ALGAE GROWING ON THE POND TORTOISES, WITH SPECIAL REFERENCE TO BASICLADIA CRASSA HOFFMANN & TILDEN^{*}

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With 41 Text-figures

Introduction

On one day of December, 1951, a vial containing some fresh-water algae was sent up to the writer by Dr. T. TOKIOKA of the Seto Marine Biological Laboratory. The algae were gathered from the shells of living tortoises, bred in a porcelain basin at the quadrangle of Mr. S. KITAJIMA in the City of Tanabe, Wakayama Prefecture. Examining the material microscopically, the writer found that the algal mass consisted for the most part of a species of *Basicladia*, a filamentous green alga of *Cladophoraceae*. Several species of other algal genera were also found growing epiphytically on its filaments, and some minute forms, moreover, found associated with them. In the course of the study of the species of *Basicladia*, the writer noticed the material was not sufficient for the detailed investigation. Thereupon he took an opportunity of visiting the place in March, 1952, and collected more complete samples.

Algae growing on living animals have attracted considerable attention of phycologists on account of their peculiar habitat, and a good number of publications on the subject has been appeared in the world. But being little known for the algae growing on the shells of pond tortoises in Japan, the writer wishes to report a preliminary note on this occasion. He intends to publish more satisfactory results in future after ample collections are made from various parts of our country. The present study, based upon the small collection, may be somewhat imperfect. This paper, however, dealing principally with *Basicladia* is the first particular information of the sort in Japan.

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T. TOKIOKA, who has been kindly enough to give him the opportunity for this study and publication. The writer owes to Mr. S. KITAJIMA for the material.

In Japan, an old tortoise with its shell overgrown with water plants is commonly called "Mino game." In China it is known as "Lü-mao kwei," which means the "green haired tortoise." In Europe and America it is also wellknown as "moss back turtle." Of course, the so called "hairs" or "moss" are filaments of some green algae growing on the carapace of the animal. Where there is a rich growth of green filaments, the animal presents an attractive appearance. Japanese as well as Chinese consider such a tortoise as an auspicious animal, and it has often been treated in classics. The "green haired tortoises" are occasionally kept by the gardners and sometimes they have a good sale.

"Mino-game" are popular among Japanese people and the animals may probably have a fairly wide distribution in the country, but any scientific investigation has hardly been executed by phycologists of Japan. Once, Dr. H. NAKANO (1919) gave a brief account on the green algae growing on the shells of pond tortoises, but he did not perform any actual study. And later in 1920, Dr. K. YENDO gave a brief description of *Chaetomorpha Chelonum* Collins var. *japonica* YENDO from Prov. Iyo. This variety belongs probably to *Basicladia crassa*. According to Mr. S. KITAJIMA, the late K. MINAKATA noticed the occurrence of the filamentous green alga on the pond tortoises bred at his garden, but he was too prudent to publish any report.

Several workers of Europe and America recorded the algae growing on the shells of living turtles. Collins (1909) described for the first time on an alga collected from the carapace of a snapping turtle and he referred it to the *Chaelomorpha*. But HOFFMANN and TILDEN collected an alga from the shells of living snapping turtle, *Chelydra serpentina*, in Minnesota, and after careful examination they noticed some fundamental characteristics separating it from *Chaelomorpha*. The species of *Chaelomorpha* possess unbranched erect filaments, while the alga under consideration has a heterotrichous habit, i.e. the little branched erect filaments arising from creeping threads. Thus they established a new genus *Basicladia*, giving descriptions of the two species, *B. crassa* and *B. chelonum*. Collins' *Chaetomorpha chelonum* was identified as *B. chelonum*.

Later in 1935, C. C. WANG furnished an information on the occurrence of *Basicladia crassa* and *Cladophora glomerata*(L.) KUTZ. var. *nana* WANG in China. According to this report, the former species has a wide distribution, but the latter has been found to be rare. In addition to these filamentous green algae, a

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prostrate form on the carapace has been known. It is *Ulvella involvens* (SAVI) SCHMIDLE, a species of *Chae'ophoraceae*, recorded from European and American fresh-water turtles.

Observations

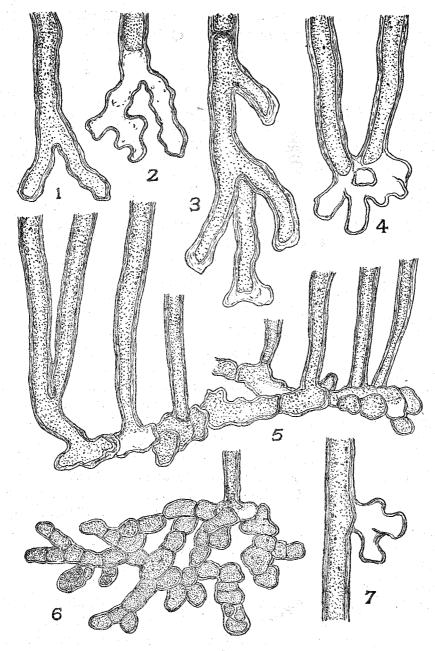
The present alga was collected from the shells of the pond tortoises, *Clemmys japonica*. The animal is known as "Ishi-game" in Japan, and in its younger age it is often called "Zeni-game." The present tortoises are about 10 cm in length. The green filaments are forming tufts and densely covering the dorsal surface of the animals. The alga has been identified as *Basicladia crassa* HOFFMANN & TILDEN. It generally agrees with the original description and that of WANG. Although some differences may be found in detailed structures, they may be attributed to the different conditions of life.

The alga has an interesting habitat, that is the host animals enjoy an amphibious life and the alga together is under allied conditions. This alga is occasionally exposed to the atmospheric air according to the behaviour of the animals. The exposure may be short or long in time, but is never periodical as is in the case of marine algae of tidal zone. It is an encouraging problem to make clear up how the alga can stand the indefinite dryness. The alga does not excrete gelatinous material on its surface and cannot be protected from desiccation by means of gelatinous sheaths like some subaerial algae. The thick stratified membranes, however, probably serve as some protection. It is interesting characteristics that most species of *Cladophoraceae* have cells thick-walled but without gelatinous sheaths, and inhabit in waters well aerated.

The fact that a number of epizoic algae can afford to grow on definite hosts is commonly known. Now for the substratum of *Basicladia*, it has been known to be strictly limited to the shells of living tortoises. According to C. C. WANG, she failed to find the same species of *Basicladia* on any other animal or on some other materials from places where the tortoises lived. As for the special selection of substratum in the species of *Basicladia*, the writer has devoted attention and found in the present investigation an occasional exception to the generally accepted assertion. The present alga grows densely covering almost all the dorsal surface of the animal, and, what is more, it thrives also on the part of basin-wall to a considerable extent. The present tortoises with their algal community have been brought from the garden of the late K. MINAKATA and have been kept in a porcelain basin filled with clear water drawn from a well. The algal tufts on the basin-wall may have their origin in the algal growth on the tortoises. Some detached filamentsmay have attached to grow on the wall, but perhaps the anchoring of liberated.

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Figs. 1-7. 1-4, irregularly lobed outgrowths of the attaching basal cells; 5, prostrate filaments from which arising several basal cells of the upright filaments, the left one showing the characteristic basal branching; 6, basal creeping filaments, composed of short cells; 7, a part of one cell in the median portion of the upright filament with a tobed rhizoid-like outgrowth. All $\times 500$.

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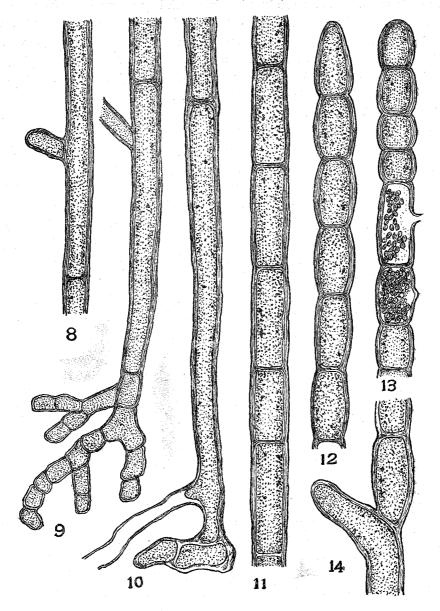
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swarmers is more probable origin of this special occurrence. After careful examination, the algae attached to these different substrata, the shells and the basin-wall, have been proved to be the same plant. The density of algal growth on the part of the tortoises has been more vigorous than on the part of the basin-wall, but the highest elongation of individual filaments has been attained in the latter. *Basicladia crassa* is permanently attached to the shells of living tortoises or occasionally as in this case grows on some other substrata, usually forming dense tufts. It is bright or dark green in colour, somewhat rigid in feeling. According to Mr. S. KITAJIMA, the upright filaments attain to the flourishing condition in cold season, becoming short in warm and hot seasons. The growth relation, however, is probably otherwise in wild state.

As mentioned above, Basicladia is heterotrichous, composed of creeping filaments and upright filaments arising from them. Attachment is effected by rhizoidal outgrowths of the basal cells or by a number of creeping irregularly septate filaments, branched or unbranched (Figs. 1-6, 9, 10). They are often intricated together to form a dense continuous expansion on the shells, and are often coralloid in appearance. The rhizoidal outgrowths of the basal cells are usually irregularly branched, rarely unbranced; colourless or often with chloroplast extended from the basal cells. The rhizoidal outgrowths are occasionally found in the cells of upright filaments (Fig. 7). The cells of creeping filaments are irregular in shape and in size, and may be empty or filled with protoplasts. Some of them may divide into a number of short cells, which contain chloroplast and filled with protoplasm (Fig. 8). These cells may probably be a kind of storage-cells, and may have abilities of propagation and perennation hereafter. The function of these cells has hot yet been confirmed in this study, but at least the presence of this sort of cells is the first record for Basicladia.

The upright erect filaments are straight, cylindrical. The regular but little branching at the basal cells was found by HOFFMANN and TILDEN, and upon this basis they separated *Basicladia* from *Chaetomorpha* (Fig. 5). Lateral branching are occasional, mostly arising from the cells of lower portions of erect filaments and rarely in the upper (Figs. 8, 9, 14). The upright filaments are not or usually more or less constricted at the cross walls, especially in the upper portion. They are somewhat tapering or becoming rather broad toward the apex.

The cells are usually cylindrical, but often varying in shape as well as in size according to the position in the filaments or to the degree of maturity. In younger stage, cells generally cylindrical in shape, and have almost the same size throughout the whole length of the filaments.



Figs. 8-14. 8, a part of a filament with a short lateral branch; 9, lower portion of a single filament with creeping branched filaments; 10, characteristically elongate basal cell with its outgrowth; 11, the median portion of the same filament, showing cylindrical cells; 12, the apical portion of the same showing somewhat short and broad cells; 13, the apical portion of a filament, showing several subglobose cells and assumed zoosporangia; 14, a part of a filament: with a lateral branch. All $\times 500$.

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In mature filaments, cells of the lower portions are also cylindrical, but those of the upper portions are often short and broad, barrel shaped to subglobose, frequently arranged in a moniliform series. Basal cells are elongate, often very long; apical cells are short or long, cylindrical or ellipsodial to ovoid, with rounded or somewhat pointed apex.

The cell membranes are thick and stratified distinctly. In younger plants they are, of course, somewhat thin and slightly stratified, but in older they become thick and about 3 to 6 stratifications are visible under microscope. The surface of membrane is rather smooth in younger state, but somewhat roughened at maturity, especially in the lower portions of upright filaments.

The fructiferous cells, presumably zoosporangia, are usually formed in the upper portions of the filaments, single or in series. They are cylindrical to almost globular (Fig. 13). In regard to the characters of the living zoospores, the writer cannot say any particular. But they must be liberated through an aperture near the middle of lateral wall of the sporangia. Several empty sporangia are occasionally observed in the material.

Various measurements of the present alga are as follows: The whole length of the upright filaments 2-5 cm.; basal cells $32-120 \ \mu$ in diameter, $415-3175 \ \mu$ in length, up to 30 or more diameters long; following cells $37-125 \ \mu \times 140-450 \ \mu$, 2-8 diameters long; apical cells $30-95 \ \mu \times 60-275 \ \mu$, 1.5-3 diameters long; sporangia $63-127 \ \mu \times 70-215 \ \mu$, 1-15 diameters long; cells in younger filaments $20-25 \ \mu$ in diameter, $110-186 \ \mu$ in length, up to 8 diameters long.

As for the position of the alga in classification system, it has generally been thought to be a genus belonging to the family *Cladophoraceae*. But the heterotrichous habit of *Basicladia* has not been observed in other genera of the family, and thereupon its inclusion among the same family is a question to be solved. Considering from the cell structure, some affinity with *Cladophora* and *Chaelomorpha* is undoubtful. FRITSCH, F. E. (1935) treated in his excellent volume the genus *Basicladia* briefly at the end of the chapter of *Cladophorales*.

Now some mention must be made of a number of fresh-water algae epiphytic on or accompanied with *Basicladia crassa*. The absence of gelatinous sheaths in *Basicladia* makes the outer surface of the filaments a favourable substratum for the growth of epiphytes. The epiphytic members are mostly filamentous green algae such as *Ulothrix tenerrima* Kürz., *U. aequalis* Kürz., *Stigeoclonium tenue* (Ac.) Kürz. and *Oedogonium* sp. One species of *Xenococcus* (*Cyanophyceae*) is also found. These epiphytes often densely cloth the old filaments of *Basicladia crassa*. A certain species of minute dimensions belonging to several groups of algae are found growing on or intermingled

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with the Basicladia and these epiphytes. Some of them are as follows: Chroococcus minor (KUTZ.) NÄGELI, Oscillatoria brevis KUTZ., Navicula cincta (EHRB.) KUTZ., Gomphonema parvulum (KUTZ.) GRUN., Scenedesmus obliquus (TURP.) KUTZ., S. quadricauda (TURP.) BRÉB. f. typicus CHODAT, Cosmarium angulosum BRÉB.

Summary

An alga growing on the shells of the pond tortoises, *Clemmys japonica*, is for the first time recorded in Japan. The host animals have been bred in a porcelain basin at a quadrangle in Tanabe City, Wakayama Pref. After examination, the alga has been determined as *Basicladia crassa* HOFFMANN & TILDEN. Morphological observations on the alga are described in detail.

The alga has hitherto been known to grow only on the pond tortoises, but the present investigation has shown the possibility of its occurrence on other substratum. As for the prostrate filaments, the writer found a kind of storage-cells, filled with chloroplast and other materials.

Although the alga has cells quite resembling those of *Chaelomorpha* and *Cladophora*, its inclusion among the family *Cladophoraceae* is a problem to be solved, considering the heterotrichous habit. It must be also an interesting subject to give a full account of the peculiar amphibious habitat and to follow up its life-cycle.

Several species of the algae growing on or accompanied with *Basic'adia* crassa are briefly mentioned.

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