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## ALGOLOGICAL NOTES

### I. CHLOROCHYTRIUM GLOEOPHILUM BOHLIN

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In August, 1909, there was collected at Gosnold Pond, Cuttyhunk, Mass., *Rivularia Bornetiana* Setch., which grows abundantly there on *Chara* and attached to stems of sedges. Upon examination the gelatinous matrix of the *Rivularia* was found to be more or less filled with a unicellular grass-green alga, which apparently failed to fit the published descriptions of any known genus, and living material was obtained in the hope that the life history of this plant might be determined. For the past seven years material has been collected at various times during June, July, and August, and impure cultures maintained through the greater part of this time. It is believed, therefore, that the information derived from this rather prolonged study comprises as complete a knowledge as may be expected of the alga growing under the conditions stated. As will be seen, the affinities of the plant would lead one to expect some kind of a motile spore or gamete, and one reason for continuing the investigation over so many years was the belief that such a spore existed. None has been found, however, and while ciliated spores may be discovered later, it hardly seems probable.

It early became evident that the published description most nearly corresponding with the form found in Gosnold Pond was *Chlorochytrium gloeophilum* growing in colonies of *Rivularia*, described by Bohlin<sup>1</sup> from preserved material from Paraguay. An exact copy of all the information furnished concerning this plant follows:

"1. *Chlorochytrium gloeophilum* n. sp. Tab. 1, Fig. 53, 54.  
Chl. cellulis ovato-oblongis, membrana hyalina, in uno vel utroque polo incrassata. In coloniis Rivulariarum nidulans.

<sup>1</sup> Bohlin, K. Die Algen der ersten Regnell'schen Expedition. I. Protococoiden. K. Svenska Vet.-Akad., Bihang till Handl. III. 23<sup>r</sup>: 28. pl. 1. f. 53. 1897.

Long. cell. 20–35, lat. cell. 8–18  $\mu$ .

Paraguay (86).

Möglicherweise könnte diese kleine Alge zu der Gattung *Kentrosphaera* Borzi gehören. Die Gestalt der Chromatophoren war nicht völlig zu erkennen. Die Wandverdickung des einen Zellendes und die Lebensweise sprechen nicht gegen eine solche Ansicht."

Presumably upon the basis of this last statement, since there is no indication of his having seen the plant, Brunnthaler<sup>1</sup> removes the species to *Centrosphaera*, publishing it as *Kentrosphaera gloeophila* (Bohlin) Brunnthaler, with the original description except that the measurements are given as "20–25  $\mu$  breit, 20–30  $\mu$  lang." This is apparently an error, since the figure published is copied from Bohlin's and is twice as long as wide. The habitat is likewise erroneously given as *R. nidulans*. As will be seen from the above original description, what Bohlin actually said was "in coloniis Rivulariarum nidulans," thus furnishing a most interesting example of the way in which names creep into the literature, as well as emphasizing the necessity of referring to the original in order to get accurate information.

The cell of what may be regarded as the normal vegetative condition of the alga found growing in *R. Bornetiana* measures from 25 to 30  $\mu$  in diameter, and is practically a perfect sphere. Later on, at the time of spore formation or because of the very considerable thickening of the wall and excrescences which may be formed, the dimensions vary very considerably, cells 50 $\times$ 25  $\mu$ , 70 $\times$ 35  $\mu$ , and in one instance 88 $\times$ 40  $\mu$  having been observed. In the latter case, however, exclusive of the thickened wall, the measurements were 64 $\times$ 25  $\mu$ .

There is a single chloroplast which lies close to the wall and usually lines the entire cell (pl. 18, figs. 2–4). Occasionally the chloroplast is incomplete, producing a light spot of varying size (pl. 18, fig. 1.) One large and prominent pyrenoid is always present, and a single nucleus, either centrally or laterally placed, may be made out by staining. At no time,

<sup>1</sup> Brunnthaler, J. Protococcales. In Pascher's Die Süßwasser-Flora Deutschlands, Österreichs und der Schweiz, Heft 6: 67–68. f. 5–6. 1915.

either in the vegetative cell or sporangium, is there the slightest indication of the radial arrangement of the chloroplast which is supposed to be characteristic of *Centrosphaera* and which Bohlin apparently considered necessary before placing his Paraguay plant in this genus. On this point Borzi<sup>1</sup> says:

“La cavità cellulare é ripiena di un protoplasma abbondante di chlorofilla differenziata in numerosi cordoni cilindrici, ora dritti, ora leggermente sinuosi, elegantemente disposti a raggio intorno al centro della cellula, dal quale si allontanano un po' lasciandovi scoperta un'area circolare scolorata.”

Brunnthaler<sup>2</sup> in his characterization of *Centrosphaera* states of the chloroplast: “Chromatophor grün oder gelblich-grün, wandständig, aus zahlreichen Körnern oder bandförmigen Strahlen bestehend, welche gegen das Zentrum der Zelle gerichtet sind und die Mitte freilassen.” This is obviously an expansion of the genus for the purpose of admitting *C. gloeophila*, the chloroplast of which shows clearly in Bohlin's figure (although somewhat plasmolized) that it in no way approaches the arrangement specified by Borzi, but is made up of numerous granules. One might well question the propriety of such a procedure, particularly since plants were not available for examination. I am accordingly not inclined to accept this disposition of Bohlin's plant or of the one collected at Cuttyhunk—which are undoubtedly the same thing—particularly since the present tendency, according to West,<sup>3</sup> is to combine under *Chlorochytrium* a number of genera such as *Endosphaera*, *Scotinosphaera*, *Chlorocystis*, and *Stomatochytrium*, the distinguishing characteristics of which are trivial or uncertain. The name should therefore stand, in my opinion, as *Chlorochytrium gloeophilum* Bohlin (*Centrosphaera gloeophilum* (Bohlin) Brunnthaler).

After the vegetative cell of *C. gloeophilum* matures, the wall almost invariably thickens until it is from 5 to 10  $\mu$  thick. This is independent of the peculiar excrescences or irregular outgrowths which may be more than half the length of the

<sup>1</sup> Borzi, A. Studi Algologici 1 : 90. 1883.

<sup>2</sup> Loc. cit.

<sup>3</sup> West, G. S. Algae 1 : 212. Cambridge, 1916.

cell itself. While these localized growths of the wall are usually external (pl. 18, figs. 2-4), they may likewise be internal (pl. 18, fig. 6), and although both the wall and these growths usually show a characteristic lamellate structure they may be entirely homogeneous.

Calcium oxalate crystals were infrequently observed within the cell (pl. 18, fig. 14).

The only type of reproduction observed was by aplanospores, which are freely formed throughout the growing season. These may be produced in cells which are circular in outline, but usually the sporangium is formed from a cell which is considerably longer than broad and on the wall of which a distinct excrescence has formed. The aplanospores are produced by successive division (pl. 18, figs. 7-11), and usually number from 32 to 64 in each sporangium. They are practically spherical and measure about  $4 \mu$  in diameter. By the time the aplanospores are completely formed there is frequently produced a distinct opening in the sporangium wall quite large enough to permit the escape of the spores. This opening may occur at any place in the wall but has occasionally been observed at the end of a tubular extension of the cell (pl. 18, fig. 12). Generally it is a distinct pore produced by the dissolution of the wall at that point but at times a considerable portion of the wall may be cut out and turned back in an irregular manner, suggesting somewhat the method of spore liberation in *Chlorocystis* (pl. 18, fig. 13). In spite of this provision for the escape of the spores, they rarely take advantage of it—in fact any aplanospores which leave the sporangium through the opening provided appear to have done so entirely by accident.

Usually the spores remain clustered together in about the position in which they were formed. As they increase in size the old sporangium wall disintegrates, and the new plants are gradually distributed through the gelatinous matrix of the *Rivularia* by the formation of new filaments of the blue-green and the action of such forces in the water as would be calculated to break up the original arrangement. The very definite provision for a means of escape for the spores sug-

gests that possibly *Chlorochytrium gloeophilum* originally possessed a motile spore, but that owing to the habitat adopted, in which a ciliated spore would be unable to swim, the cilia were lost. It was originally assumed that the plants at some stage in their existence left the *Rivularia*, and that ciliated spores would afford an easy means of again establishing themselves in the gelatinous colonies. This does not seem to be the case, however. The *Chlorochytrium* cells apparently never give up their endophytic habit, and new colonies of *Rivularia* are infected from aplanospores contained in the gelatin surrounding the young filaments which increase with the development of the *Rivularia* colony.

With the idea that possibly *C. gloeophilum* might occur in other species of *Rivularia* the following forms were examined from exsiccati: Rabenhorst, 295, *R. minor*; 355, *R. pygmaea*; 416, *R. minuta*; 648, *R. angulosa*; 743, *R. minuta*; 793, *R. Sprengeliana*; 931, *R. angulosa*; 932, *R. Lyngbyana*; 975, *R. Lenticula*; 976, *R. durissima*; 1095, *R. minuta*; 1125, *R. Sprengeliana*; 1452, *R. insignis*; 2184, *R. villosa*; 2540, *R. fluitans*; 2563, *R. terebralis*. Collins, Holden, and Setchell, 357, *R. atra*; 358, *R. Biasoletiana*; 260, *R. nitida*; 508, *R. compacta*; 860, *R. Biasoletiana*; 1015, *R. polyotis*. Tilden, 166, *R. Biasoletiana*; 289, *R. haematites*; 570, *R. Biasoletiana*; 571, *R. nitida*. In no case was there the slightest indication of the presence of the endophyte.

Specimens of the following included in the Missouri Botanical Garden Herbarium were also examined, but with negative results: *Rivularia nitida*, *R. bullata*, *R. fluitans*, *R. atra*, as well as several undetermined species. In view of the fact that none of these specimens showed the presence of *C. gloeophilum*, it is interesting to note that the specimen of *R. Borneiana* from Watch Hill Pond, Watch Hill, R. I., distributed as No. 157 in Collins, Holden, and Setchell's 'Phycotheca,' contained an abundance of grass-green cells within the gelatinous matrix, which was easily recognized as *C. gloeophilum*. Hence at the only two localities thus far noted in the United States for *R. Borneiana*, *C. gloeophilum* is found growing within it and apparently in no other species.

Bohlin did not give the species of *Rivularia* in which his plant was found and there is no way of telling whether it was *R. Bornetiana* or not.

A set of *C. gloeophilum* has been prepared from the Collins, Holden, and Setchell 'Phycotheca' and will be distributed during the year 1918.

## EXPLANATION OF PLATE

## PLATE 18

All figures are reproduced from camera drawings  $\times 580$ .

Fig. 1. Typical cell with chloroplast only partially lining the wall.

Figs. 2-5. Various examples of irregularities in thickened wall.

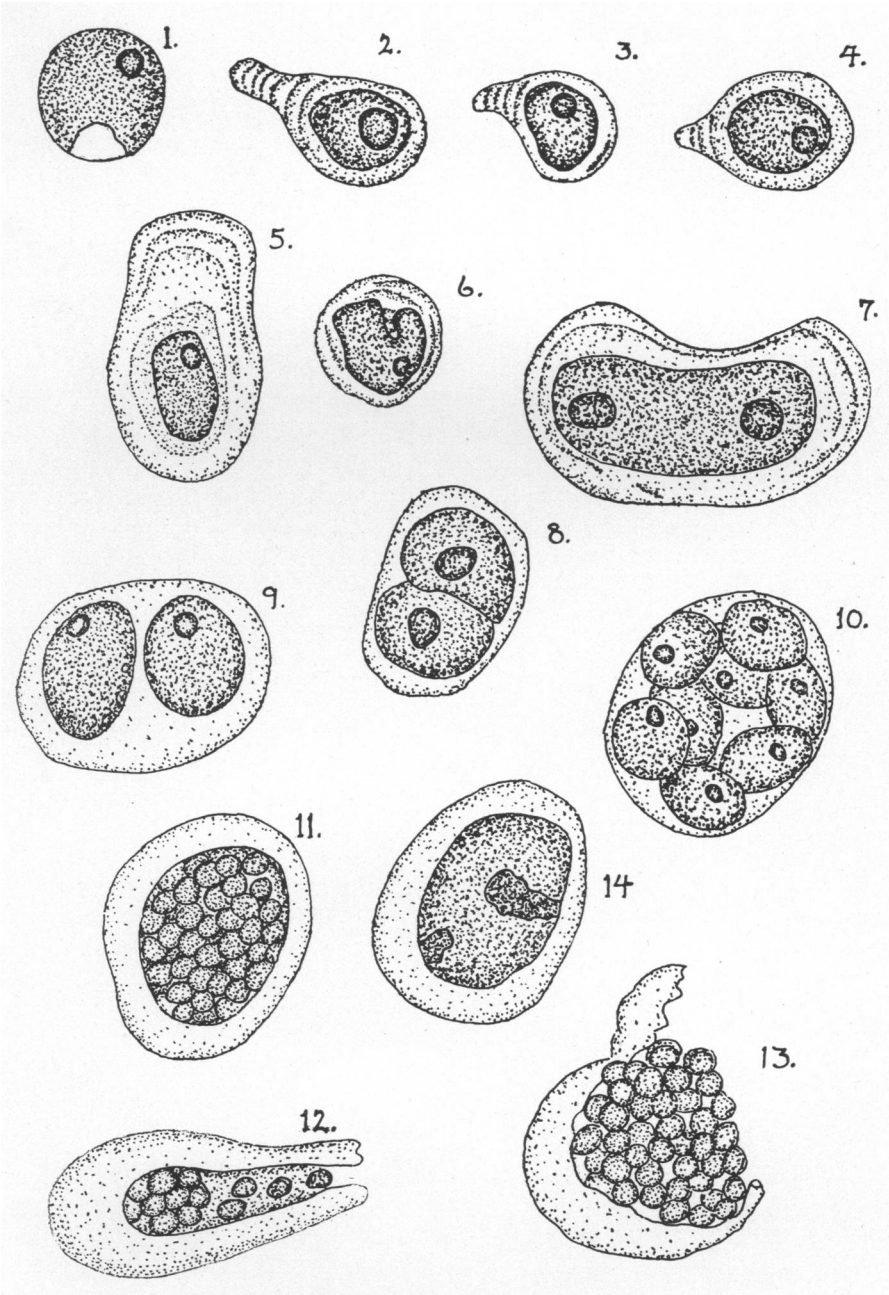
Fig. 6. Internal thickening of wall penetrating cell.

Figs. 7-11. Successive stages in the formation of spores.

Figs. 12-13. Means of spore liberation.

Fig. 14. Calcium oxalate crystals formed in cell.





MOORE—CHLOROCHYTRIUM GLOEOPHILUM