

BOTANICAL GAZETTE

FEBRUARY, 1896.

Contributions from the Cryptogamic Laboratory of Harvard University. XXX.

New or peculiar aquatic fungi. 3. *Blastocladia*.

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WITH PLATE III.

In the two preceding notes on aquatic fungi reference has several times been made to a paper by Reinsch¹ in which, among other interesting forms, he describes the curious genus *Blastocladia*; including the single species *B. Pringsheimii*, which, as far as the writer is aware, has not been again observed. The genus has since been wholly ignored by systematists or placed among the doubtful Saprolegniæ. Fischer in his *Phycomycetes*² includes it among the genera of this group which are doubtful or to be excluded, while Schroeter in his more recent revision³ finds a place for it among the Leptomitaceæ; on the ground that, although lacking the segmentation so characteristic of this order, it corresponds in general habit to the species of the genus *Rhipidium*, as was formerly pointed out by Cornu.⁴ The uncertainties which have surrounded it have been in part due to a lack of exact knowledge concerning it, and in part to the truly anomalous character of the plant itself, which occupies a distinctly isolated position among other aquatic *Phycomycetes*.

Notwithstanding the fact that it has remained unknown for twenty years since its original discovery, it has proved in the writer's experience to be one of the more common aquatic fungi, occurring in almost every locality where it has been

¹Pringsheim's Jahrb. f. wiss. Bot. 11: 291. 1876.

²Rabenh. Kryptogamenfl. 14: 367. 1892.

³Engler & Prantl Nat. Pflanzenfam. 93: 103. 1893. [I Th., 1 Abth.].

⁴Bull. Bot. Soc. de France 24: 227. 1877.

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sought for, both in the vicinity of Cambridge and of Kittery Point, Maine. The present note therefore is based on the examination of very abundant material from widely separated localities, which illustrates by an almost unbroken series the wide range of variation for which this plant is remarkable.

In general terms the fungus may be described as consisting of a highly developed unicellular main axis ("Hauptstamm") more or less clearly differentiated, which may remain simple or become distally variously branched, and is attached by copious rhizoids to the substratum on which it grows. The branching of this axis may be sub-umbellate or irregularly dichotomous, while the branches themselves may be in turn several times more or less irregularly branched, varying greatly in size, habit and appearance. The axis if it is simple, or its ultimate branches when it is otherwise, become as a rule more or less abruptly swollen distally into often well developed heads from the surface of which are produced, terminally or sub-terminally, the organs of reproduction, as well as certain peculiar sterile branchlets which will be mentioned subsequently. In some branched forms, however, this terminal swelling is not noticeable, the reproductive organs being borne singly at the tips of short ultimate branchlets (fig. 6). Apart from these organs which are separated from it by septa, the plant consists, as has been just mentioned, of a single cell, in the contents of which certain spherical oily masses, very variable in size, are usually conspicuous (figs. 1 and 3). These masses, which are sometimes wholly absent, were described by Reinsch⁵ as independent cells endogenously formed, at first free in the protoplasmic contents and later giving rise to the reproductive organs. After fixing themselves to the inner surface of the wall of the terminal enlargement of the axis already mentioned, they were said to burst through to the outside and develop into sporangia, "oospores" or "antheridia?" according to circumstances. In fresh material these bodies resemble homogeneous refractive oil globules; but when stained they seem to consist of a coherent mass of coarse granules, having the appearance represented in fig. 3. In his examination of these bodies, however, the writer has seen nothing which would indicate the

⁵ "Diese Zellchen legen sich an die innere Schlauchwandung an, durchbrechen — von der Berührungsstelle mit der Schlauchwandung auswachsend — die Membran des Schlauches und kommen auf der Aussenseite der Schlauchwandung als kleine Hökerschen zum Vorschein." — Reinsch l. c. 292.

truth of this, in itself improbable, supposition of Reinsch. Although they are somewhat peculiar from the fact that they stain readily, and permanently retain their form and characteristic appearance after the plant has been crushed and its contents scattered in the surrounding medium, there seems no ground for the belief that they are anything more than masses of fatty protoplasm, as would naturally be inferred from the fact that they may be present or absent according to the character of the nutrient substratum. The variation in size of such masses in a given individual is usually extreme, some appearing as mere minute granules, while others nearly fill the terminal heads. In no instance, moreover, has the writer seen one which seemed in any way to connect itself with the formation of one of the buds which develop into reproductive organs.

The zoosporangia first make their appearance as papillæ formed at or near the extremity of the axis or of its branches, which are soon cut off by septa as independent cells (fig. 4, at the left) and soon increase in size, assuming the form characteristic of the mature sporangia. The latter vary very greatly in shape and size, so that any one who chanced to find the extremes of variation without knowledge of intermediate forms would hardly hesitate to separate such varieties as distinct species; especially in view of the fact that variations in the sporangia are often associated with differences almost as extreme in the form and size of the resting spores as well as in the general habit of the main axis. From slender elongate nearly cylindrical zoosporangia (fig. 2) to much shorter and stouter (fig. 3) or even broadly oval forms, every gradation may be found; but in a given individual there is usually a general uniformity in their size and shape. They are formed in considerable numbers on a given tip, usually at its summit; but sometimes also in small numbers laterally below it, and when mature the contents divide into a very large number of zoospores, while a thickened papilla of dehiscence is formed at the apex (fig. 10). Finally this papilla, as it begins to deliquesce, is pushed off by the mass of zoospores within, and the latter make their escape one by one, swarming almost immediately. The empty sporangium wall eventually disappears, leaving a circular scar where it was attached, and the fertile surface of exhausted plants is thickly studded with such marks.

The zoospores are peculiar in appearance and can readily be distinguished from any similar bodies known to the writer by the characteristic disposition of their contents. In general form they are oval or elliptical and are, at least in many cases, biciliate; the two cilia arising side by side from the smaller end of the spore. In some instances it has been found impossible to make out more than a single cilium even after the zoospore was stained; but the presence of two (fig. 11) has been determined definitely in so many cases that this number may be considered as typical. The nucleus is very large and sub-triangular in outline, its base connected with that of the cilia by a fine strand of granular protoplasm. In front of the nucleus lies a broad and distinct mass of granular protoplasm while small groups of granules occur here and there around it in the otherwise nearly clear cytoplasm. In general appearance they are not unlike the zoospore of *Gonapodya*, but may always be distinguished by the form and position of the nucleus and the evident connection of the latter with the base of the cilia. As the fungus develops, growing as it almost invariably does in tufts, it forms the center of a dense mass of bacteria which finally choke the sporangia completely; so that as a rule only those first formed are able to discharge their contents. As a result the zoospores commonly die without escaping; the remains of their large nuclei just mentioned filling the sporangia, as is indicated in the central sporangium of fig. 3. These dead nuclei were taken by Reinsch for the zoospores themselves and are represented in his plate XVI, fig. 8.

Associated with the sporangia are often found numerous slender filaments which arise as buds in a fashion exactly similar to that by which the former are produced. They are very slender, simple or irregularly branched, without septa, and often greatly exceed the sporangia in length (fig. 2). In a majority of cases, however, they are wholly absent or undeveloped (fig. 1 at the right) and they seem to bear no definite relation either to the sporangia or to the resting conidia. They seem to be quite sterile and although Reinsch suggests that they may be antheridia they are probably without definite function, and are certainly not male organs.

The resting spores, already referred to, and described by Reinsch as doubtful oospores, do not as a rule make their appearance until some time after the sporangia have been de-

veloped; in other words the former are characteristic of the earlier, the latter of the later conditions of the plant. In origin these spores correspond in all respects to the sporangia; arising as buds from the surface of the swollen extremities of the axis from which they are soon separated by a septum. As they mature they usually assume a more or less oval form, become very thick walled and when ripe fall readily from their attachment, leaving a circular scar. The mature spore has two walls, an outer, thin and even, and an inner, thick and curiously modified, so as to present the appearance represented in fig. 12, when viewed in optical section. Whether the characteristic markings figured are really pits, as they appear to be, can hardly be definitely determined without an examination of an absolute section of the wall, and such a section has not been obtained. The surface view of these "pits" is represented by the circular outlines shown in fig. 13. Several large oil globules are usually present in the contents of these resting spores and all attempts to induce them to germinate have proved unsuccessful, although cultures have been continued for more than a month. In form they vary almost as much as the sporangia; being in some cases quite spherical, with a small papilla of attachment as in figs. 8 and 9, in others nearly oval with a broad base as in fig. 7, or long pyriform as in fig. 7. There is, however, a notable tendency in a given plant to produce resting spores of a given form even if, as in fig. 8, this form is an unusual one.

As far as can be determined these spores are wholly non-sexual in origin, and the most careful examination has failed to show the presence of anything which could be by any chance interpreted as an antheridium. The absence of any such organ naturally suggested the possibility of the existence of motile antherozoids similar to those found in *Monoblepharis*; but there is certainly only one kind of sporangium, and even if antherozoids in reality existed, it would be, in almost all cases, quite impossible for them to perform their office from the fact that, by the time the resting spores begin to form, the whole plant is, as has been already mentioned, completely enveloped in a solid mass of bacteria and other organisms which would effectually prevent access to the oogonium, if it were such, by any body dependent on cilia for its motility. The outer wall of the spore, moreover, is always continuous, showing no signs of any perforation or receptive spot.

Among the material collected in one locality near Kittery Point, numerous plants were found bearing peculiar sporangia, proportionately much larger and more nearly oval than the ordinary forms, many of which contained large well defined rounded masses of protoplasm which at first suggested the presence of oospheres; but further examination showed them to be early conditions of the resting spores of a species of *Rozella* parasitic on the *Blastocladia*, which, when mature, became spherical, thick walled, and echinulate.

The position of the genus among related forms is very doubtful, and although in habit it resembles *Rhipidium*, while its zoospores recall those of *Gonapodya*, there is no reason for believing that it is at all closely related to either of these genera. Its resting spores are in some respects comparable to the deciduous resting conidia of certain species of *Pythium*, especially in the new form described below, and for lack of any more satisfactory disposition, it may be placed provisionally among the *Pythiaceæ*; the only alternative being apparently the erection of a special family for its reception.

In addition to the material of *B. Pringsheimii* obtained, a second and much less well defined species was found in company with it in a single locality at Kittery Point. In this form the resting spores are less highly differentiated, though produced in greater abundance, and resemble thick walled sporangia, slightly modified in form. There can however be little doubt of the correctness of the generic reference of the plant, which may be characterized as follows:

***Blastocladia ramosa*, nov. sp.**—Main axis nearly cylindrical, attached to the substratum by rhizoidal divisions at its base; above copiously branched, irregularly or sub-dichotomously, the branchlets producing terminally and sub-terminally sporangia and resting spores. Sporangia broadly oval, bluntly pointed, $30 \times 15 \mu$. Resting conidia terminally bluntly rounded gradually narrower towards the truncate base, about $30 \times 11 \mu$. The whole plant 260 to 600μ high, its main axis 14 to 20μ in diameter.—On submerged twigs, Kittery Point, Maine. Plate III, figs. 14–16.

This species has been found in only one locality growing with *B. Pringsheimii* and other forms in a small pool in a sphagnum bog, and occurred in such small quantity that I was unable to observe the escape of its zoospores; since, in

the material examined, the zoosporangia were comparatively rare. In all cases the resting spores were formed in great profusion and seemed constant in form though somewhat variable in size. Their walls, though usually distinctly thickened, are never as conspicuously so as in the larger species, and might very properly be spoken of as conidia; since, in many instances, even after they are detached, their walls do not appear to be much thicker than those of the sporangia. I have never seen an instance, however, in which one seemed to have discharged its contents like the normal zoosporangia. The species is an insignificant one, and would not have been described without further observation, had it not possessed a certain interest in connection with its more highly developed ally.

For convenience of reference a description of *B. Pringsheimii* is appended, no measurements of this species having been previously published.

BLASTOCLADIA PRINGSHEIMII Reinsch.—Main axis simple or several times successively branched sub-dichotomously sub-umbellately or irregularly, the free extremities usually but not always distally swollen into more or less well defined terminal heads. Reproductive organs, sporangia and non-sexual resting spores, produced terminally and sub-terminally and often associated with slender sterile filaments branched or simple and similarly produced. Sporangia long cylindrical to long oval, more commonly more or less pod-shaped, producing very numerous biciliate zoospores. Resting spores formed like the sporangia, spherical, oval, or long piriform, the wall much thickened and pitted. The whole plant 200 to 750μ in height (exclusive of rhizoids), the larger terminal heads $100-150\mu$ in diameter. Main axis $30-90\mu$ in diameter. Zoosporangia $150 \times 25\mu$ ($50 \times 25 - 225 \times 18\mu$). Zoospores about $7 \times 5\mu$. Resting spores $50 \times 30-75 \times 44\mu$.—On apples and other decaying vegetable matter in ponds and ditches. Vicinity of Cambridge, Mass., and Kittery Point, Maine. On decaying apples in water, Germany (Reinsch). Plate III, figs. 1-13.

Cambridge, Mass.

EXPLANATION OF PLATE III.

Blastocladia Pringsheimii Reinsch.

Fig. 1. Single plant of large size showing rhizoids; sub-dichotomously branching axis with fatty bodies in its contents; sporangia, many of which are empty (two discharging zoospores); young resting spores and two sterile branchlets.

Fig. 2. An axis bearing two heads with long cylindrical sporangia, some of them empty, and long sterile branchlets.

Fig. 3. An older simple plant with larger fatty masses and mature resting spores; four of the sporangia empty the other two filled with the nuclei of dead zoospores.

Fig. 4. Mature resting spore *in situ*; at the left a young resting spore just separated as a bud from the surface of the head.

Fig. 5. Mature resting spore, surface view.

Fig. 6. Small plant; the habit and the form of the resting spores unusual.

Fig. 7. A branch of the same enlarged, with single terminal resting spore.

Fig. 8. A plant irregularly branched with spherical resting spores.

Fig. 9. Two resting spores of the same, that at the right seen in optical section.

Fig. 10. Sporangium just before the discharge of zoospores showing papilla.

Fig. 11. A single zoospore.

Fig. 12. Optical section of wall of resting spore.

Fig. 13. Surface view of a portion of wall of resting spore.

Blastocladia ramosa Thaxter.

Fig. 14. General habit of plant.

Fig. 15. A terminal branch with two sporangia (one of them empty) and six resting spores.

Fig. 16. Two resting spores showing relative thickness of wall.

* * * NOTE. The original figures were drawn with the following approximate magnifications in diameters, and have been reduced about one-third by photo-lithography. Figs. 1, 2, 3, 6, 8, 14, $\times 90$. Figs. 4, 5, 7, 9, 15, 16, $\times 420$. Figs. 11, 12, 13, $\times 925$.



Thaxter, Roland. 1896. "New or Peculiar Aquatic Fungi. 3. Blastocladia." *Botanical gazette* 21(2), 45–52. <https://doi.org/10.1086/327299>.

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DOI: <https://doi.org/10.1086/327299>

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