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A FUNGUS IN *ANOMIA SIMPLEX* SHELL

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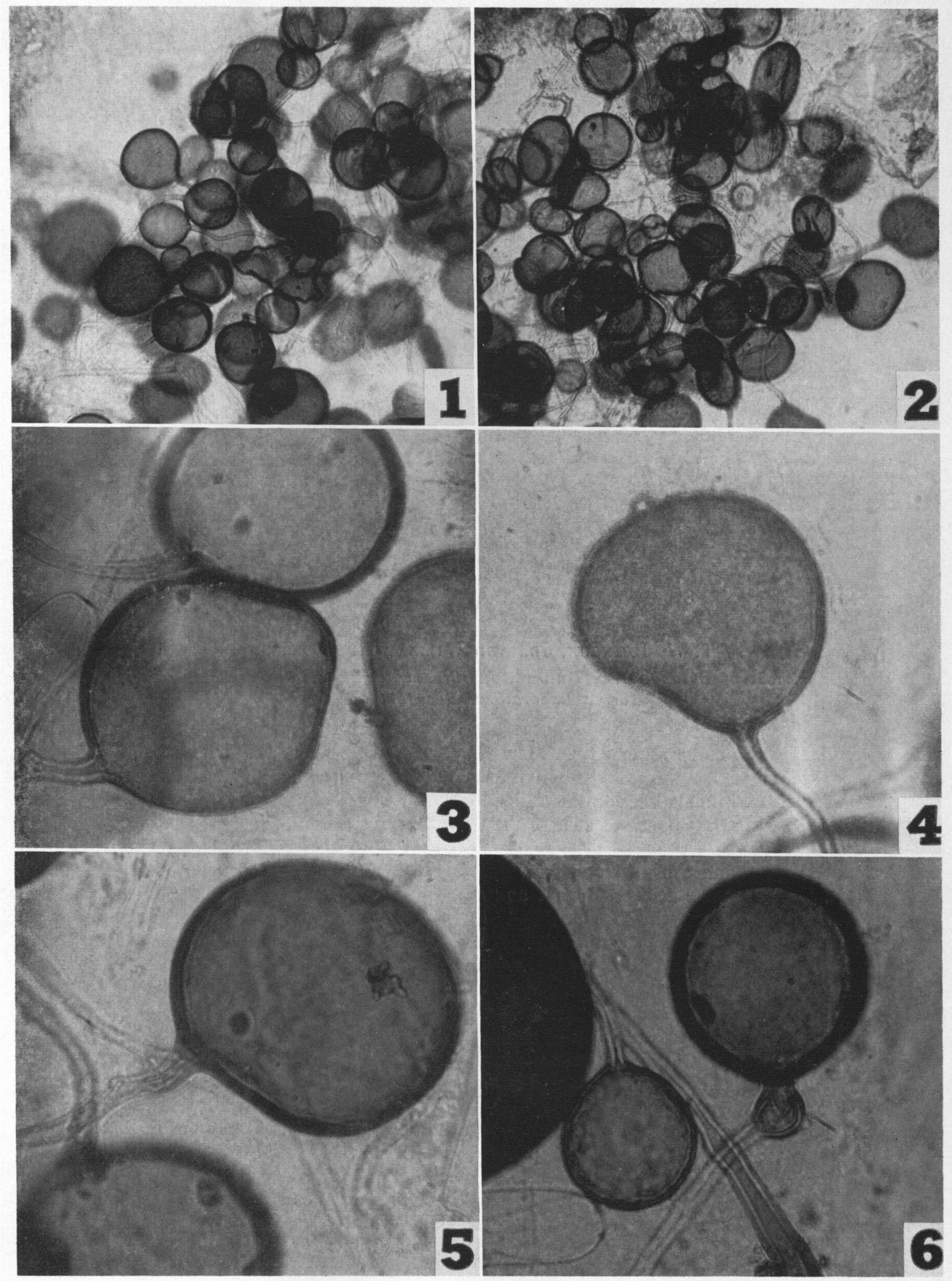
As early as 1860, Kölliker reported ". . . lower forms of plants . . ." in such nonliving accretions of marine animals as mollusk shells, fish scales, and sponge spicules. These he thought to be fungi. Stirrup (1872) also observed filaments or tubules in gastropod shells, among other calcareous materials. Bornet and Flahault (1889) described two "fungi" under the names *Ostracoblabe implexa* and *Lythopythium gangliiforme* in shells of living and dead Mollusca. More recent are the report by Bonar (1936) of *Didymella conchae* in shells of *Acmaea* species and other animals and Korringa's account (1951) of "shell disease" in *Ostrea edulis,* thought to be caused by a fungus. These and other records of "fungi" in calcareous materials have been reviewed and summarized by Johnson and Sparrow (1961).

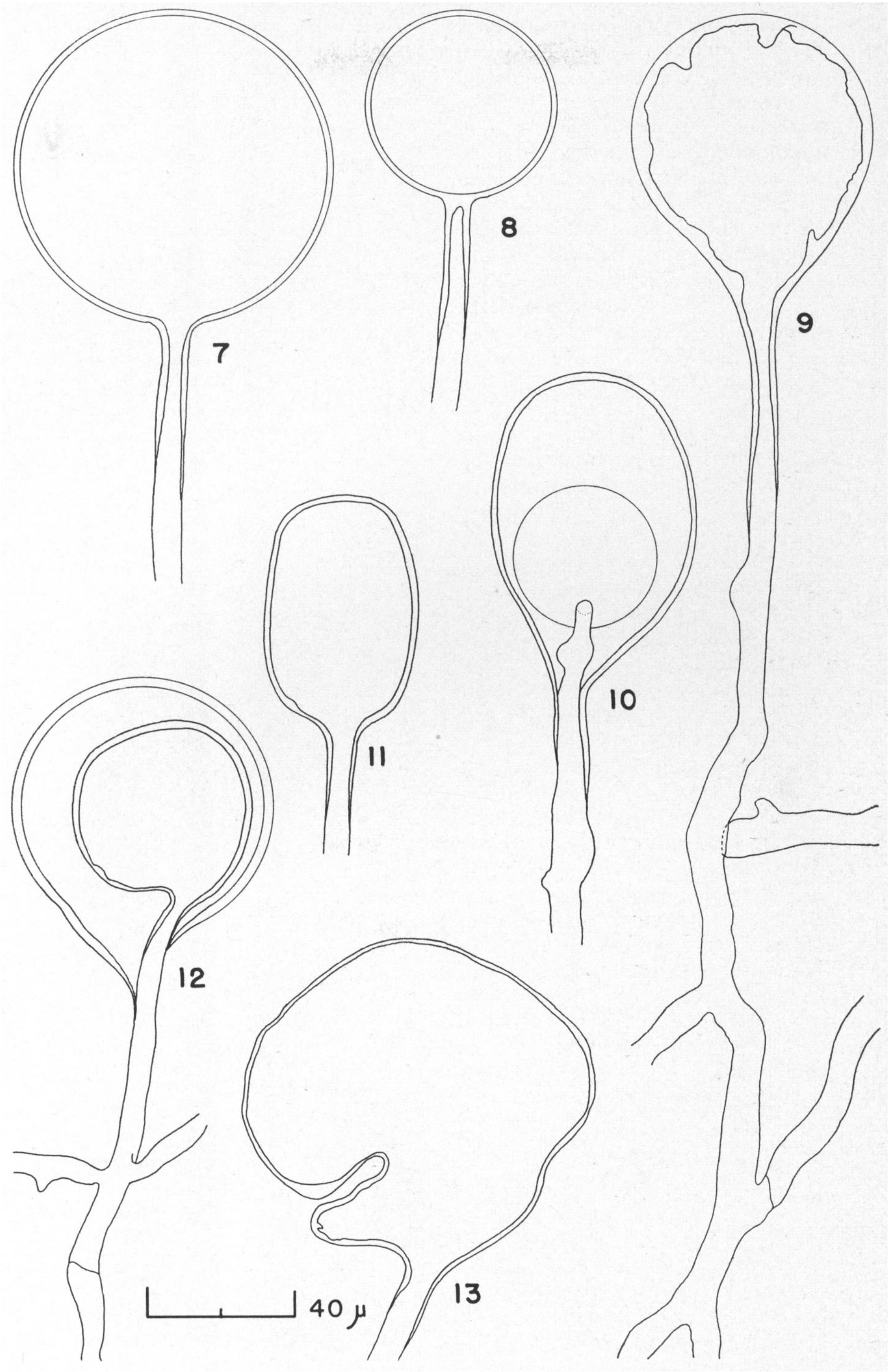
Porter and Zebrowski (1937) published an account of "lime-loving" fungi in shell fragments from Cambrian deposits as well as more recent ones. Although they write of fungi in beach sands, their observations were on calcareous fragments in these sands. In 1936, Zebrowski named several of the "fungi" he collected, and assigned these new taxa to the Cladochytriaceae. Among the collections cited by him were shell fragments from the beach areas in the vicinity of Beaufort, North Carolina. *Dodgella inconstans* and *D. priscus* were two such species.

In the summer of 1961, the junior author began an extensive survey of shell fragments from various sandy beaches in the vicinity of the Duke University Marine Laboratory at Beaufort in an attempt to confirm, if possible, the observations made earlier by Zebrowski. We established the premise that if some of Zebrowski's fungi could again be examined, it might be possible to obtain convincing evidence whether the structures shown by him were or were not fungal. We encountered considerable difficulty initially in applying the methodology for study as outlined by Porter and Zebrowski (1937). By modifying their techniques, however, we were able to find, in shell fragments and in unbroken but cast-off shell, several of the reported fungi as well as additional ones. An account of one specimen is given in this paper.

The fungus occurred in a cast-off shell of *Anomia simplex* d'Orbigny (smooth jingle shell) from the Pivers Island beach, North Carolina. Studies on other shell fragments showed the



FIGS. 1–6. Fungus in *Anomia* shell. FIGS. 1, 2, 5, 6. General aspect of chlamydospores. FIG. 3. Chlamydospore with protoplasmic reticulum. FIG. 4. Papillate chlamydospore. Figures 1 and 2, approximately X100; others, approximately X430.



FIGS. 7–13. Fungus in *Anomia* shell. FIG. 7. Spherical chlamydospore. FIG. 8. Spherical chlamydospore with basal septum. FIG. 9. Obpyriform chlamydospore and portion of subtending hypha; inner wall surface irregular. FIGS. 10, 12. Internal proliferation. FIG. 11. Dolioform chlamydospore. FIG. 13. Lobed chlamydospore.

need for partial decalcification before direct microscopic examination could be satisfactory. The jingle shell was therefore broken into small pieces and submerged for a few minutes in dilute HCl. Sufficiently transluscent particles were then washed and mounted on slides in Hoyer's mounting medium.

The fungus in *Anomia* shell consists of intra- and extramatrical hyphae and extramatrical chlamydospore-like bodies (Figs. 1, 2). Hyphae are abundant, intermittently irregular in outline (Fig. 9), and sparingly branched. Near their junction with the chlamydospores the hyphae are yellow to yellow-brown because of their thickened walls (Figs. 3, 7). The intramatrical hyphae are hyaline. Both pseudosepta and septa appear, very sparingly, in the hyaline portions of the mycelium. Hyphal diameter averages 6.5 μ in the intramatrical portions and 8 μ at the base of the terminal bodies.

Spherical chlamydospores predominate, although oval, pyriform, obpyriform, asymmetrical and cylindrical examples, as well as some that are quite irregular, are also produced (Fig. 13). No intercalary ones were observed. Color, similarly, is variable, although a medium brown predominates. Some chlamydospores, possibly immature, are pale yellow or yellow; a few are dark brown. The wall is either uniformly thickened (Figs. 7, 8) or irregularly so (Fig. 9). In some of the terminal bodies, the junction of the hypha with the body itself is without a septum (Figs. 11–13), but in others a thick septum (Fig. 8) is evident. A very few chlamydospores are provided with one or two papillae (Fig. 4). Internal proliferation (Figs. 10, 12) is not common, at least in our specimens. In the medium-brown chlamydospores particularly, there is a very faint configuration suggesting a fine reticulum (Fig. 3). This is probably the cytoplasm, for in bodies without a basal septum it is contiguous with the hyphal cytoplasm. The chlamydospores are quite variable in diameter, ranging from 51 to 151 μ. They, as well as the hyphae, react positively to the usual cellulose test.

There is no evidence of additional reproductive bodies such as sporangia, zygospores, or conidia. Sporelike bodies are not present in the chlamydospores we have examined. How the organism develops, sporulates, and is disseminated, therefore, remains unsolved.

The affinities of this fungus to known genera are obscure. The general morphology of the inflated terminal bodies, however, is strikingly reminiscent of chlamydospores of certain *Endogone* species (Thaxter, 1922, and others). Principal points of similarity are color, the thickened chlamydospore wall, and the thick-walled nature of the hyphae near the juncture with this cell.

Endogone species characteristically produce a loose or firm sclerotium-like body in which the reproductive units occur. Furthermore, it seems generally accepted that *Endogone* fructifications contain either sporangia, zygospores, or chlamydospores, but never more than one of these types in any "sclerotium." Our material seems to fit well into the latter of these two features of *Endogone,* but not into the former. We have not observed sclerotium-like structures, although the hyphae and chlamydospores do form a dense, restricted mass on the shell. This fungal mat could hardly qualify as a sclerotium. It is reasonable to suspect, however, that under nonterricolous conditions a sclerotium may be absent. The fungus may be assigned tentatively to *Endogone,* but final judgment in this respect must await collections of further samples and culture studies of them.

Summary

A chlamydospore-producing fungus occurring in the shell of *Anomia simplex* is described but not named. Affinities of the organism to *Endogone* are discussed briefly.

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MIKTONISCUS GRAYI, A NEW SPECIES OF TERRESTRIAL ISOPOD CRUSTACEA FROM NORTH CAROLINA

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The genus *Miktoniscus* was first described by Kesselyak in 1930 and redescribed by Vandel in 1950. The genus is part of the subfamily Trichoniscinae of the family Trichoniscidae. Some of the biogeographical facts relevant to the genus are reviewed by Vandel (1960). *Miktoniscus grayi,* n. sp., is apparently endemic to North America, as are *M. halophilus* Blake, 1931, from New England, *M. medcofi* Van Name, 1940, from Illinois, *M. humus* Muliak, 1942, from Louisiana, and *M. racovitzai* Vandel, 1950, from Virginia. All of the species are endemic to eastern and midwest United States.

At present time *Miktoniscus grayi,* n. sp., has been found only in two places in North Carolina. It has been found extensively near Durham in the Duke Forest in places where there are hardwood logs in the proper stage of decay, especially the New Hope Creek lowland. The species has also been taken in abundance under the bark of a log, in association with *Armadillidium vulgare,* near Creedmoor Lake about 15 miles north of Durham. Gravid females are found from April to September, and there are about seven eggs or embryos per female.

The species is found in deciduous hardwood logs in the decay under the bark, and in the adjoining leaf litter in the warmer part of the year when there is enough moisture. Oak logs yield the largest number of animals. The species is also found in decayed logs, in the moist leaf litter of springs and along the banks of shaded brooks which do not dry during the summer. A description of *Miktoniscus grayi,* n. sp., follows:

The body is about one-third as wide as it is long, and the second antennae, head, thoracic tergites, and posterior borders of the first three abdominal tergites are covered with pointed tubercles. The tubercles are arranged so as to suggest rows or patterns which, upon closer examination, do not exist except for a fairly distinct row of small tubercles on the posterior border of each tergite. The body from the anterior view is elliptically arched, resembling half a cylinder. The frontal outline of the head, as seen from above, lacks any indication of lateral lobes, but the lateral view reveals small, rather acutely pointed lobes beneath the moderately large, single, well-pigmented ocellus. The second antenna, if extended posteriorly, would terminate near the third thoracic segment. Large spines are found on the distal part of the fifth peduncular segment of the antenna, and a large spine also continues straight from the fourth segment near the base of the fifth segment. Spines also extend ventrally from the second peduncular segment. The fifth peduncular segment is slightly longer than the flagellum, which has four articles, the last of which has a few setae on the tip. The triarticulate first antennae are tipped by several short setae.

The head is moderately large and not deeply set in the first thoracic segment. The posteriolateral edges of the first two thoracic segments point anteriorly, and the posterior border of the third segment is almost at a right angle to the anterior-posterior axis. The posteriolateral borders of thoracic segments 4 to 7 are progressively more pointed to segment 7, which has the longest and most acute points. These overlap the first three segments of the abdomen. The abdomen is abruptly narrower than the thorax, but not so abruptly narrower as that of other described species of the genus. The lateral edges of the first two abdominal segments are not visible, and the posteriolateral edges of the last three abdominal segments are acutely pointed in dorsal aspect. The lateral

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