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A Preliminary Survey of the Filamentous Phycomycetes Found in the Embarras River, Coles County, Illinois

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A PRELIMINARY SURVEY OF THE FILAMENTOUS PHYCOMYCETES FOUND IN THE EMBARRAS RIVER, COLES COUNTY, ILLINOIS.

(TITLE)

BY

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THESIS

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INTRODUCTION

The site of the projected Lincoln Reservoir dam will be across the Embarras River and its flood plain, 6½ miles south of Charleston, Illinois and about a mile southeast of Lincoln Log Cabin State Park. The resulting lake will extend northward from this dam and will encompass the river and the present site of Lake Charleston. The stated purpose of the reservoir is to supply water, to aid in flood control, and to support fish and wildlife conservation.

The completed reservoir will have three "pools", a permanent pool, a seasonal pool, and a maximum flood pool. The permanent pool will have a surface area of approximately 4,310 acres. It will have an elevation of 584 feet above sea level, and will be maintained only in the winter, when demand upon it will be least. The seasonal pool will have a surface area of 6,760 acres and will be 16 feet higher than the present lake; it will have an elevation of 596 feet to 600 feet above sea level. This level will be maintained during the summer months through October, when the demand will be greater. The maximum flood pool, with an elevation of 629 feet, will be approached only under conditions of heavy flooding, or about once every 80 years. The maximum flood pool would have a surface area of 21,250 acres, and extend nearly 60 miles northward from the dam site (Browning, 1970).

Until recently, there has been a paucity of information concerning the phycomycetous flora of the above mentioned area. Beneke (1948), in his monographic treatment of the Illinois species of the Saprolegniales,

made several collections from the Wabash water shed, including three samples from the upper Embarras River. In 1968, a survey of the aquatic biota of Coles County and adjacent areas was initiated under a research proposal funded by The Federal Water Pollution Control Administration. In this connection and supported in part by a University Research Grant, an extensive series of collections have been made from streams and lakes within Coles County (W. W. Scott, personal communication). However, to date, little information has been obtained pertinent to the specific identity of phycomycetous fungi from the southern section of the Embarras River, from Lake Charleston to the Cumberland County line. As it is now planned, the completed Lincoln Reservoir will inundate this part of the river, and all of Lake Charleston.

As the reservoir dam becomes operable, and the permanent and seasonal pools begin to stabilize, they will exert an effect on the newly submerged and adjacent lands. The impounded water will cover areas that are now wooded, grazed or cultivated. Until stability occurs, this water is expected to undergo constant biological and chemical changes. Present populations of plants and animals may undergo changes concommitant with the environmental changes. It is quite probable that a series of ecological succession patterns may take place.

In order that future population have relevance as far as discerning the effects of environmental changes, it is desirable to establish a population for the affected area prior to those changes. Such a study would make it possible to determine whether population and morphological variations of the fungi have occured, and if so, to what degree. It is to this end that the present work is aimed.

MATERIALS AND METHODS

Soil and water samples were collected in clean previously unused three to five ounce plastic screw cap bottles. The samples were collected from three zones at each collecting site. Each sample was labeled with the site number, the zone, and the date of collection. All samples were placed into culture within three hours of their collection.

In the laboratory, the soil or water was deposited in deep Petri dishes and the dishes filled to the three-quarter level with sterile distilled water. Each culture was then baited with halves of boiled hemp seed and pieces of boiled snakeskin. This technique, commonly used for the isolation of filamentous aquatic fungi, involves "sinking" the bait into the culture medium. There are two methods generally used for baiting collected soil and water samples. These involve either floating the bait on the surface of the medium, or submerging it in the medium, so that it comes in contact with the soil. The latter method was utilized here because it was believed that direct association of bait and soil would increase the chances of zoospore encystment and subsequent germination. Boiling the bait serves a dual function; it reduces the microbial population on the surface of the bait, and drives off much of the air, allowing the bait to be submerged in the medium.

Hemp seed has long been used as a substratum for the culture of aquatic fungi because it supports growth for long periods of time without drastically altering the nature of the culture medium, and because the achenes apparently inhibit non-fungal microbial growth (Harvey, 1925).

Snakeskin was used as a source of chitin for the isolation of those species which do not ordinarily grow upon hemp seed.

The "gross" cultures were incubated at room temperature for 48 hours, then examined for evidence of fungal growth. Those baits containing phycomycetous hyphae were transferred to fresh culture dishes and rebaited with the same type of substratum. The gross culture was also rebaited and later examined for the presence of more slowly growing species. These were later isolated in the same manner as the above. After all evidence of fungal growth had been obtained from the original culture, the excess water was decanted, and the plates stored for possible future reference.

For accurate species identification of phycomycetous fungi, it is desirable to establish unifungal cultures. Since two or more distinct isolates may develop on a single bait, it is often necessary to separate them into different cultures. The techniques used depends upon the genera involved. In cultures containing fungi which readily discharged their zoospores, the spores were collected by micropippette at the time of their discharge and were transferred to slide cultures of Potato Dextrose Agar (PDA). The slide cultures were prepared in the following manner: a small square of PDA was placed onto the surface of a glass slide; the zoospores were pipetted onto the surface of this medium and were covered with a glass cover slip, and the entire slide was placed on a bent glass rod in a Petri dish. A filter paper saturated with water was positioned in the bottom of the dish in order to maintain a relatively high humidity within the culture chamber. This culture originating from the germinating zoospore could then be removed and microscopically examined at will. To reduce the possibilities of bacterial contamination, aseptic conditions were maintained throughout the procedure, and all glassware were autoclaved

previous to use. Single germinating zoospores were aseptically removed to Petri dishes of the same medium. When the resulting mycelial mass was large enough to handle easily, a portion of the periphery containing hyphal tips was exised and transferred to a Petri dish containing water and the appropriate bait.

For those isolates which did not discharge their zoospores in the usual manner, a similar technique was employed. Isolates of Dictyuchus, for example, often produce abundant deciduous zoosporangia. These were treated, in their entirity, in the manner described above, as were the individual zoospores of more typical genera. In cases where no reproductive structures were apparent, single hyphae were removed and treated similarly.

In the case of mixed cultures containing Allomyces, the entire bait was removed and placed on filter paper to dry. After several days, sections of the paper containing the dry meiosporangia were placed into water culture and baited. The period of desiccation destroys other fungal hyphae, but fails to harm the thick walled meiosporangia.

The methods used in identifying each isolate largely follow those described by Scott (1961). Generic identification was based primarily upon vegetative morphology and upon asexual reproductive characteristics (i.e., the morphology of the zoosporangia, their position on the hyphae, the nature of zoospore discharge and the activity of the zoospores following discharge). To facilitate identification, only actively growing and reproducing isolates were used. It has been suggested by Dick (1963) that the morphology of several genera of Saprolegniales is altered by slight variations in pH. To prevent this, it was necessary to maintain a proper ratio of water to mycelial growth. Fresh bait and water were periodically added to each dish to prevent major pH changes from occuring and to maintain a dilute medium, thus retarding the growth of contaminating microorganisms.

Several monographic studies were consulted in making specific identification for a number of genera. In each case, the criteria established by the author were strictly adhered to in identifying the Embarras River isolates; namely, for Aphanomyces, A Monograph of the Genus Aphanomyces (Scott, 1961); for Achlya, The Genus Achlya, (Johnson, 1956); for Saprolegnia, The Genus Saprolegnia, (Seymour, 1970); and for Pythium, The Taxonomy, Host Range, and Geographical Distribution of the Genus Pythium, (Middleton, 1943). More recent publications by these and by additional authors were referred to for the identification of species described since the publication of the above monographs. Reference to these papers is made under the species concerned. The classical works of Sparrow (1960) and Coker (1923) were used to determine distribution of species and taxonomic criteria not available in the papers mentioned above.

After the identification of each isolate was accomplished, the isolate was transferred to a fresh culture dish and refrigerated at 10°C. The only further maintenance necessary was an occasional replacement of evaporated water and the addition of fresh bait.

TAXONOMIC CRITERIA

The importance of utilizing consistant morphological characteristics for the identification of phycomycetous fungi must be emphasized. To date, physiological studies are too limited to be reliably used in determining generic or specific identifications for most phycomycetous fungi (Unestam, 1966). Therefore, the assignment of an isolate to a specific taxon relies heavily upon certain major characteristics, or combination of characteristics. The relevance of morphological variation within a genus must also be considered when determining the significance of a characteristic, and consequently the taxonomic position of each isolate. Many morphological characteristics and much of the terminology used in describing these fungi are also in common usage in other areas of botanical science. The general understanding and widespread use of such terms precludes the necessity of including them here. However, certain terms are unique to the study of phycomycetous fungi and will be defined herein in the context of their application. Those terms having more restricted application or only occasional use will be defined as they are used.

Definition of Terms

Chlamydospore (gemma): a modified section of a hypha or hyphal cell, which becomes walled off from the rest of the hypha; it behaves as a resting spore.

Zoosporangial Renewal:

Basipetalous Succession: secondary zoosporangia are formed successively beneath the primary zoosporangium; zoospore release is accomplished through lateral exit pores. (Pl. I,g)

Cymose Branching: repeated branching of a hyphal tip, resembling a cyme. (Pl. I,e)

Internal Proliferation: the development of a secondary zoosporangium within the discharged primary zoosporangium; common within the genus Saprolegnia. (Pl. I,h)

Sympodial Branching: lateral branching of the hypha below the basal septum of a delimited zoosporangium. (Pl. I,f)

Types of Zoospore Discharge:

Achlyoid: primary zoospores encyst immediately as they emerge from the zoosporangium, forming a hollow sphere, which usually does, but may not adhere to the zoosporangial apex; typical of the genus Achlya. (Pl. I,b)

Traustothecoid: zoospores encyst within the zoosporangium, and are released by the delequesence of the zoosporangial wall; typical of the genus Thraustotheca. (Pl. I,d)

Dictyoid: encystment of the primary zoospores within the zoosporangium; secondary zoospores germinate following exit through lateral exit papillae; the empty cyst membranes may remain following discharge; typical of the genus Dictyuchus. (Pl. I,c)

Saprolegnioid: primary zoospores emerge singly, and encyst at a point removed from the zoosporangium; typical of the genus Saprolegnia. (Pl. I,a)

Primary Zoospores: pyriform zoospores bearing two dissimilar apical flagella of equal length; the tinsel type flagellum is directed forward and the whiplash type flagellum is directed backward.

Secondary Zoospore: reniform biflagellate zoospores, the flagella of which are also dimorphic, but attached laterally; the secondary

zoospore emerges from the primary cyst, swims for a time, then also encysts, ultimately germinating into a vegetative hypha.

Antheridial Origin:

Androgynous: the antheridial branch arises from the oogonial stalk, below the delimiting septum. (Pl. II,c)

Dielinous: the oogonium and its attendant antheridium arise from different hyphae. (Pl. II,a)

Hypogynous: antheridial branch laking, but with the antheridial cell abstricted as a part of the oogonial stalk immediately below the oogonium. (Pl. II,d)

Monoclinous: the oogonium and its attendant antheridium arise from the same hypha. (Pl. II,b)

Oospore Types:

Centric: with one or two peripheral layers of small oil droplets completely surrounding the ooplasm. (Pl. II,e)

Eccentric: with one large oil droplet on one side of the oospore, not completely surrounding the ooplasm. (Pl. II,f,g)

Subcentric: with one or more layers of oil droplets on one side of the oospore. (Pl. II,h)

DESCRIPTION OF THE AREA

The Embarras River originates in Champaign County, Illinois, and flows through portions of Douglas, Coles, Cumberland, Jasper, Crawford, and Lawrence Counties. The river terminates in Lawrence County, where it flows into the Wabash River. The river has an overall length of approximately 125 miles, and drains an area of 2,410 square miles. The elevation above sea level at the source is 730 feet and is 400 feet at the mouth (Horton, 1914).

The section of the river between the Riverview Dam, below Lake Charleston, in Coles County, and the Cumberland County line is, for the most part a slowly moving stream, and is approximately 15 to 20 miles long. It is fed by two main tributaries, Kickapoo Creek and Indian Creek, and a number of smaller "seasonal" streams.

Most of the areas bordering the river are farmed. Much of the land directly adjacent to the river itself is under cultivation or is used to raise cattle and hogs. A small amount of this land is wooded. The river is subject to the effluent from these areas, from city sewage treatment plants, and from local industries. The data included in Table I concerns the physical aspects of this sector of the Embarras River. They are included strictly to aid in defining the present environment and for the purpose of comparing possible changes in the river should a later population study be attempted following the completion of the Lincoln Reservoir. The data was compiled between September, 1969, and March, 1970, in connection with the above mentioned FWPCA research grant.

The soil and water samples were taken from 14 stations along both banks of this section of the river. The stations were chosen for their overall accessibility and their immediate proximity to a maintained roadway. These criteria were established in order to assure that additional samples could be collected from the same area should it become necessary.

As previously mentioned, three zones were sampled at each station. These zones are: (a) the water itself, at a depth of approximately 6 inches; (b) the "flood zone"; and (c) the dry soil immediately above the flood zone. The flood zone is that section of bank that is alternately submerged and exposed by changing water levels.

The two terrestrial zones were sampled for several reasons: the deposition of spores or vegetative mycelia from the river onto an otherwise exposed area might occur by flooding following heavy rains or spring thaws, or be carried from the river by amphibians or birds (Dick, 1961). This would result in the establishment of a temporary aquatic population at that site. Conversely, runoff from adjacent farmland and roadways might tend to wash terrestrial species into the river. Therefore, it was felt that sampling at all of the zones at a given time might aid in explaining the presence of such ecological anomalies, should they occur. In the same publication, Dick suggests that although there is a relatively constant flora in any given area, certain species seem to exhibit seasonal dominance, with the greatest overall frequency in autumn and spring. Therefore, samples were collected in October, December, March, and June, under the assumption that a single sampling might not accurately record the true nature of the total population of the river.

Specific descriptions of the areas sampled are listed below. Each has been arbitrarily assigned a letter in order to facilitate discussion.

In cases where the samples were taken from closely related areas, a letter-number combination was employed. A map showing the location of these areas is shown on Figure 1.

STATION G: NE ¼, Sec. 25, T12N-R9E.

Samples were collected from directly beneath the Route 130 bridge. The immediate embankment is devoid of green plants and is covered with rocks and cement fill material. The river bottom is composed of sand, gravel, and rocks. River dimensions average 2 feet in depth by 25 feet in width.

STATION G-1: NE ¼, Sec. 25, T12N-R9E.

Samples were collected from a sand bar below the embankment. The surrounding area is woody, and is surrounded by cultivated fields. The river bottom is sand and gravel. River dimensions average 2-3 feet in depth by 20 feet in width.

STATION G-2: SW ¼, Sec. 25, T12N-R9E.

Samples were collected approximately 50 yards downstream from Station G-1. The surrounding area is wooded and partly cultivated. The embankment is very eroded. The river bottom is sand, gravel, and silt. River dimensions average 2 feet in depth by 20 feet in width.

STATION G-3: NW ¼, Sec. 36, T12N-R9E.

Samples were collected approximately 50 yards downstream from Station G-2. The surrounding area, slope, and bottom composition are similar. River dimensions average 1½ feet in depth by 25 feet in width.

STATION G-4: NW ¼, Sec. 36, T12N-R9E.

Samples were collected approximately 100 yards downstream from Station G-3. Surrounding area, slope, and bottom composition are similar. River dimensions average 2 feet in depth by 25 feet in width.

STATION G-5: NW ¼, Sec. 36, T12N-R9E.

Samples were collected approximately 50 yards downstream from Station G-4, at a point where a runoff pipe empties into the river. The pipe drains an adjacent cornfield. The embankment and slope are similar to Station G-4. River bottom is sand and gravel with much more silt than the previous sampling stations. River dimensions average 2-3 feet in depth by 25 feet in width.

STATION G-6: SW ¼, Sec. 36, T12N-R9E.

This station is located about 10 yards downstream from Station G-5. The surrounding area is identical with that of the above station. The embankment is terraced, and is sandy and eroded. The river bottom is sand, gravel, and silt. River dimensions average 2 feet in depth and 25 feet in width.

STATION H-2: SE ¼, Sec. 36, T12N-R9E.

Samples were collected about 20 yards upstream from the mouth of Kickapoo Creek. The embankment slopes steeply. The surrounding area is predominantly wooded. The river bottom is sand and gravel. The river dimensions average 2½ feet in depth and 20 feet in width.

STATION H-1: SE ¼, Sec. 35, T12N-R9E.

Samples were collected at the mouth of Kickapoo Creek. The surrounding area is similar to the previous station. The river bottom is sand, gravel, and silt. The river dimensions average 1½ feet in depth by 20 feet in width.

STATION H: SE ¼, Sec. 35, T12N-R9E.

Samples were collected about 20 yards downstream from the mouth of Kickapoo Creek. The embankment slopes gently, and is composed of a fine sand and mud mixture. The surrounding area is similar to Station H. The river dimensions average 2 feet in depth by 20 feet in width.

STATION I: SW ¼, Sec. 2, T11N-R9E.

This area is locally known as "Walker's Ford". The embankment slopes gently and is terraced to the river's edge, where it drops more sharply. The surrounding area is wooded. The river bottom is sand and gravel. River dimensions average 3 feet in depth by 40 feet in width.

STATION J: NW ¼, Sec. 11, T11N-R9E.

Samples were collected on the west bank of the river, below Walker's Ford. The embankment drops sharply 3 or 4 feet. The surrounding area is predominantly wooded and cultivated fields. The river bottom is sand and silt. The river dimensions average 3 feet in depth by 30 feet in width.

STATION K: NW ¼, Sec. 23, T11N-R9E.

This station is the most southerly of the sampling stations. The banks are composed of a heavy black mud that averages 18 inches in depth. The river bottom is of similar composition. The river dimensions average 2 feet in depth by 20 feet in width.

RESULTS AND DISCUSSION

The following systematic account describes a total of 7 genera representative of the orders Blastocladiales, Saprolegniales and Peronosporales. All were isolated from the collecting stations previously described.

Generic descriptions were taken from the earlier cited monographic studies. In some cases the original description has been abbreviated in order to achieve greater clarity. Specific descriptions were made by observing the Embarras River isolates. Any major morphological discrepancies from the original species description is also discussed.

ALLOMYCES E. J. Butler

Ann. Bot. London, 25: 1027. 1911

Thallus consisting of a cylindrical more or less differentiated trunklike basal cell which gives rise distally to cylindrical dichotomously, subdichotomously, or sympodially branched, blunt-tipped successively more slender pseudoseptate hyphae of indefinite extent on which are borne the reproductive organs, contents often alveolately or reticulately vacuolate, anchored to the substratum by a system of endobiotic branched strongly tapering rhizoids; asexual plant bearing terminally, sympodially, or in basipetal succession thin-walled zoosporangia which discharge their fully formed posteriorly uniflagellate zoospores through one or more pores produced upon the deliquescence of prominent papillae, and persistent or deciduous resting spores with a thick brown punctate outer wall and a thin inner one, the resting spores upon germination

producing either (a) posteriorly uniflagellate planonts which give rise directly to new asexual plants or to sexual plants, or (b) posteriorly flagellate planonts which immediately encyst, each of the cysts forming endogenously four isogamous uniflagellate gametes which, after emerging from the cyst through a pore, fuse in pairs, the zygote germinating to form asexual plants; sexual plant similar to the sporophyte, monoecious, bearing male and female gametangia terminally in pairs or alternating in basipetal succession, gametes anisogamous, posteriorly uniflagellate, the small male always pigmented, the larger female colorless, the planozygote posteriorly biflagellate and germinating to form the asexual plant. From Sparrow (1961).

Allomyces anomalous

These isolates produce only darkly pigmented meiosporangia. Repeated attempts to produce a sexual cycle have failed. Therefore, these isolated are at present assumed to be A. anomalous, a tentative species designed to include those isolates which apparently lack sexual reproduction. (Sparrow, 1960).

Distribution:

Station H; water zone; October; isolated on hemp seed.

Station G-6; soil zone; October; isolated on hemp seed.

Station I; flood zone; March; isolated on hemp seed.

Station H-1; flood zone; March; isolated on hemp seed.

Station J; flood zone; June; isolated on hemp seed.

Allomyces arbuscula E. J. Butler

Sporophyte hyphae long, abundantly branched. Branching is dichotomous or sympodial. Mitosporangia single or catenulate; ovoid or ellipsoidal,

ends generally rounded; average size 45u-75u by 35u-45u; forming up to 4 discharge papillae. Meiosporangia abundant, ovoid, 30u-60u by 20u-40u; exospore thick, dark red to brown in color; punctate. Gametophyte thallus similar; gametangia strictly terminal in younger thalli, catenulate in older isolates. Colorless female gametangia large, averaging 50u by 20u; exie papillae lateral and terminal. Male gametangia are hypogenous; smaller than the female gametangia, averaging 7u cy 15u; contents pigmented, usually yellow brown to red. Female gametes ovoid, colorless; approximately 10u by 5u; male gametes ovoid to spherical, averaging 7u by 4u.

Distribution:

Station G; water zone; October; isolated on hemp seed.

Station G-3; water zone; October; isolated on hemp seed.

Station G-3; soil zone; October; isolated on hemp seed.

Station G-2; soil zone; October; isolated on hemp seed.

ACHLYA C. G. Nees

Nova Acta Acad. Leop. Carol., 11: 514. 1823

Thalli monoecious or dioecious. Hyphae stout or slender, more or less branched, straight or flexuous, gradually tapering from base to apex; variable in length. Gemmae, when present, formed by segmentation of the hyphae; variable in size and shape; functioning as zoosporangia or germinating by one or more slender hyphae which usually terminate in a small zoosporangium. Zoosporangia filiform, fusiform, naviculate, or clavate; renewed sympodially or by basipetalous development and cymose branching. Zoospores usually dimorphic; primary ones on discharge encysting at once to form a more or less spherical, hollow mass at the zoosporangial orifice and occasionally provided with two quickly evanescent,

apically attached flagella during emergence; secondary zoospores reniform, laterally biflagellate, germinating after encystment by a slender hypha; in a few species, aplanoid, dictyoid, or thraustothecoid discharge also present. Oogonia borne variously, being lateral on stalks of variable length, terminal, intercalary, or sessile; in some species, the immature ones proliferating; variously shaped, predominantly spherical or pyriform. Oogonial walls with or without ornamentations; pitted or unpitted. Antheridial branches diclinous, monoclinous, androgynous, or exigynous, in one species hypogynous; in a few species lacking. Antheridial cells predominantly tubular and clavate; laterally or apically appressed to the oogonial wall or attached by finger-like projections; fertilization tubes usually present. Oospheres generally maturing. Oospores one to many; centric, subcentric, or eccentric; germination, when present, usually accomplished by a slender germ tube ultimately terminating in a zoosporangium. From Johnson (1956).

Achlya conspicua Coker

Mycelium extensive; hyphae stout; abundantly branched at apices. Gemmae abundant, filiform or irregular; single or catenulate; may function as zoosporangia when mature. Zoosporangia very numerous, filiform or naviculate; averaging 300u by 30u sympodial renewal, occasionally cymose. Zoospore discharge achlyoid. Oogonia moderately abundant; laterally arranged; spherical; approximately 70u-80u in diameter. Oogonial wall smooth; pitted only at point of antheridial attachment. Oospheres rarely mature. Oospores eccentric; spherical; do not fill the oogonium; numerous, up to 30 per oogonium, averaging 5u to 15u in diameter.

Distribution:

Station H-1; water zone; June; isolated on hemp seed.

Station H; water zone; March; isolated on hemp seed.

Station H; water zone; June; isolated on hemp seed.

Station H; water zone; October; isolated on hemp seed.

Discussion:

The characteristics of this isolate agree in most respects with those described by Beneke (1948). The major differences are in zoosporangial size; Beneke's isolates averaged 39u by 327u, while the Embarras River isolates averaged 30u by 300u.

Achlya prolifera (Nees) DeBary

Diffuse mycelium, moderately branched. Principal hyphae stout, often as much as 118u at the base. Gemmae present; numerous, usually intercalary, rarely terminal. Primary zoosporangia terminal, secondary ones formed sympodially; length varies, up to 800u, averaging 40u in diameter. Zoospore emergence is achlyoid, secondary zoospore emergence may be lacking; in situ germination has been observed in some isolates. Oogonia abundant; terminal or laterally arranged; rarely intercalary; stalked, rarely sessile; pyriform or spherical; average diameter 50u-80u. Oogonial wall smooth with numerous pits. Oogonial stalks ½-2 times the diameter of the oogonia in length; straight, rarely curved. Antheridia with numerous branches; diclinous, often wrapping around the oogonia; laterially appressed. Oospheres maturing. Oospores eccentric, usually spherical; variable in number; usually more than two.

Distribution:

Station G-2; soil zone; October; isolated on hemp seed.

Station J; soil zone; October; isolated on hemp seed.

Discussion:

Beneke (1948) does not include in situ zoospore germination in his description of this species; however, Johnson, (1956), does suggest very

rare aplanoid zoospore discharge. In his monograph, Johnson examined Beneke's isolates; therefore, it may be assumed that this characteristic is indeed valid for the species, and may be a function of cultural conditions.

Achlya diffusa Harvey ex Johnson

Moderately extensive mycelium; diffuse; principle hyphae slender, averaging 50u at the base; few branches. Abundant gemmae, irregularly shaped; terminal or intercalary; single or catenulate arrangement; may germinate by hyphae or function as zoosporangia. Zoosporangia numerous, filiform or clavate; averaging 570u in length by 25u in diameter; zoosporangial renewal sympodial, occasionally cymose; basipetalous succession also reported by Johnson (1956). Zoospore discharge achlyoid. Oogonia abundant, shape variable: spherical, pyriform, or obovate; located laterally, or more frequently terminal or intercalary. Oogonial wall pitting variable in size and frequency; smaller oogonia may be unpitted. Oogonial stalks ½-4 times the diameter of the oogonium in length; stout, straight, or recurved. Antheridial branches persistant, occasionally branched; attached by projections. Oospheres mature, but soon abort. Oospores eccentric, spherical, usually not filling the oogonium; number depends upon size of the oogonium, ranging from 1 to 18 in the isolates studied. Laterally attached, subglobose, or irregularly shaped swellings are present along the oogonia-bearing hyphae; 50u-200u in diameter; pits or cospheres not observed; not seen to germinate.

Distribution:

Station G-1; soil zone; October; isolated on hemp seed.

Discussion:

Since its isolation by Harvey (1942) and its validation by Johnson (1956), this species has been only rarely described. The Embarras River

isolate agrees fundamentally with Johnson's description of a Michigan isolate; however, that organism was isolated from water, while the Illinois isolate was consistantly recovered from soil. This species has not been previously recorded in Illinois.

Achlya klebsiana Pieters

Hyphae diffuse, slender; principal hyphae averaging 75u in length, moderately branched. Gemmae fairly abundant, more numerous in older cultures; when mature may function as zoosporangia, or germinate into small zoosporangia-bearing hyphae. Primary zoosporangia abundant, straight or slightly bent; average size 45u-670u; however, some may be much smaller. Zoosporangial discharge is achlyoid; zoospores occasionally encyst within the zoosporangium; in situ germination not observed in this isolate. Oogonia quite numerous; stalked; laterally arranged on the hyphae; spherical or pyriform, averaging 40u-60u in diameter; stalks about 1-4 times the diameter of the oogonium: straight, occasionally bent. Oogonial wall smooth, pitted only under point of antheridial attachment; occasionally unpitted; if pitted, pits are obscured. Oospheres mature. Oospores eccentric, spherical, filling the oogonium, 2 to 6 or 8 per oogonium. Antheridia diclinous, rarely monoclinous; occasionally branched.

Distribution:

Station G-2; water zone; October; isolated on hemp seed.

Station G-4; water zone; October; isolated on hemp seed.

Station G-4; water zone; December; isolated on hemp seed.

Station G-6; water zone; December; isolated on hemp seed.

Discussion:

Although the average size of the zoosporangia in the above isolates were somewhat smaller than those described by Beneke (1948), their overall

size range was within the extremes listed in that monograph. The distinguishing feature of this organism is the occurrence of oogonial pits only under the point of antheridial attachment, and the extreme rarity of monoclinous antheridial branches.

Achlya americana Humphrey

Mycelium extensive, not dense; principal hyphae stout, others more slender; sparingly branched. Gemmae not abundant; shape frequently irregular; single; usually terminal; occasionally intercalary; may or may not function as zoosporangia. Zoosporangia fairly abundant; fusiform; long, 250u-800u by 20u-30u in width; sympodial renewal. Zoosporangial discharge is achlyoid; zoospore cluster persists. Oogonia quite numerous; laterally arranged; spherical to pyriform in shape; 50u-60u in diameter. Oogonial wall smooth, thin; pits present. Oogonial stalks ½-2½ times the diameter of the oogonium in length; stout, usually straight, rarely curved. Antheridia monoclinous, rarely diclinous; attached by projections. Oospheres mature. Oospores eccentric, spherical; fill the oogonium, fairly numerous; average 6-12 per oogonium, 21u-46u in diameter.

Distribution:

Station G-2; soil zone; October; isolated on hemp seed.

Station G-2; soil zone; December; isolated on hemp seed.

Discussion:

The distinguishing features of this species are the numerous pits on the oogonia and the large number of oospores per oogonium. Beneke (1948) characterizes his isolates as being androgynous and occasionally monoclinous; however, Johnson (1956) describes the species as having antheridia of monoclinous origin, and does not refer to androgynous antheridia.

Achlya flagellata Coker

Mycelium moderately extensive; principal hyphae sparingly branched, more so at the hyphal apex. Gemmae abundant; shape varies, pyriform or irregular, terminal or intercalary; catenulate or singly arranged; tend to disarticulate; may function as zoosporangia when mature, discharging zoospores through lateral exit papillae. Zoosporangia abundant; naviculate, fusiform; straight or occasionally curved; average dimensions 400u-500u by 30u-50u; renewal primarily sympodial, occasionally by basipetalous succession. Zoospore discharge achlyoid; spore cluster not persistant at tip of sporangium. Oogonia abundant; stalked; usually lateral, infrequently terminal; spherical to pyriform. Oogonial wall pitted, or pitted only under point of antheridial attachment; walls smooth. Stalk length averages 3 times the diameter of the oogonium; stout, straight, rarely curved. Antheridia diclinous, rarely monoclinous. Oospheres not maturing. Oospores eccentric, spherical, not filling the oogonium; average 3-8 per oogonium, 20u-26u in diameter.

Distribution:

Station G-1; flood zone; October; isolated on hemp seed.

Station G-6; water zone; October; isolated on hemp seed.

Station G-1; water zone; October; isolated on hemp seed.

Discussion:

The above isolates exhibit strictly the characteristics of A. flagellata as described by Coker (1923), and do not appear to have the characteristics of A. imperfecta, e.g. maturing oospheres and androgynous antheridia. Johnson (1956) suggests that because of overlapping morphological characteristics, that these species be regarded as variants of a single species.

Achlya orion Coker and Couch

Extensive mycelium; principal hyphae slender, profusely branched. Gemmae may or may not be present; filiform or irregular; intercalary and terminal. Abundant zoosporangia, usually clavate; straight, occasionally curved; average dimensions 220u by 30u; sympodial renewal. Zoospores discharge is achlyoid. Oogonia abundant; lateral; spherical, 22u-88u in diameter; occasionally proliferating. Oogonial wall smooth; pitted only under point of antheridial attachment. Oogonial stalks 1-10 times the diameter of the oogonium; usually bent, seldom straight. Oospores eccentric; spherical; 1-10 per oogonium; generally 22u-33u in diameter. Antheridial branches persist; androgynous and monoclinous; attached by projections.

Distribution:

Station I; soil zone; October; isolated on hemp seed.

Discussion:

Beneke (1948) bases his identification on the examination of a single isolate; although he does not list the bent oogonial stalks as an important characteristic, the remainder of his description agrees with that of Johnson (1956), and with the Embarras River isolates. Johnson examined Beneke's isolate and except for the emphasis placed upon the curved oogonial stalks, his description is in agreement with Beneke's.

Achlya sp. Isolate No. 10-24-69 (G-1)

Hyphae slender, 33u to 44u in diameter. Two week old colony diameter 1.75cm to 2.0cm. Gemmae not abundant in younger cultures; more numerous in older (2-3 week) colonies; filiform, occasionally intercalary; germination not observed in these cultures. Zoosporangia not abundant; a few exhibit small lateral exit papillae; clavate, filiform; straight, not curved; 44u-250u by 22u, most average 104u by 22u. Sympodial renewal. Zoospore

discharge strictly achlyoid. Individual encysted zoospores average 10.4u in diameter. Zoospore cluster not always peraistant at apex. Oogonia very numerous; laterally arranged; stalked; spherical to pyriform; 26.0u to 46.8u in diameter, averaging 37.5u. Wall pitting is absent. Wall ornamentations rounded to truncate, occasionally slightly bulbous at the tip; very abundant; size ranging from 7.7u by 7.7u to 10.4u by 10.4u. Stalks 1 to 3 times the diameter of the oogonium in length; rarely smaller; stout, curved, occasionally straight. Antheridia laterally appressed; androgynous in origin or often monoclinous, with the antheridia originating on the hyphae very close to the oogonial stalk. Simple or more often highly branched. Oospheres mature. Oospores eccentric; 1 to 5 per oogonium, usually 3 or 4, rarely more than 5, even in larger oogonia; spherical; 26.5u to 32.1u in diameter, averaging 28.7u, filling the oogonium.

Distribution:

Station G-1; soil zone; October; isolated on hemp seed.

Station G-3; soil zone; October; isolated on hemp seed.

Station G-6; soil zone; October; isolated on hemp seed.

Discussion:

This organism was at first thought to be an isolate of Achlya recurva, and bears a number of similarities to that species. No other species of Achlya bears the similar striking papillate oogonial wall projections. Species differentiation in Achlya is based strongly upon sexual morphology, with asexual morphology being of secondary importance. The similarities and differences between these species are as follows:

A. recurva bears oogonia which are also laterally arranged, and which are spherical in shape. However, the diameter of the oogonia range from 28u to 64u, with the majority of the oogonia being 35u to 50u in diameter.

This is consistantly larger than the oogonia of the above described isolates. The ornamentations on A. recurva are described as being truncate, and having an average length of 12u to 19u. The Embarras River isolates are not consistantly truncate, and are more often rounded or bulbous at the tip; their size ranges from 7.7u to 10.4u. The oogonial stalks of both are essentially similar, with the stalks of the Illinois isolate being occasionally more highly recurved. The antheridial branches of A. recurva are described as being simple, and only sparcely branched, and as being androgynous, and only rarely monoclinous in origin, while those of the Illinois isolates are often very profusely branched, and are quite often of monoclinous origin. The oospores of both are spherical and eccentric. The oospores of A. recurva range from 14u to 38u in diameter, averaging from 21u to 23u in diameter. The oospores of the Embarras River isolate ranged in size from 26.5u to 32.1u, averaging 28.7u in diameter. A. recurva is described as having 1 to 14 oospores per oogonium, averaging 4 to 8 per oogonium. The Illinois organisms ranged from 1 to 5 per oogonium, averaging 3 to 4 per oogonium. There were very rarely more than 5 per oogonium.

It is evident that both organisms bear a number of close similarities, especially in respect to the nature of the mature oospores and the overall morphology of the oogonium. However, these were not considered sufficient to warrant classifying these isolates as A. recurva, per se. There are several morphological descrepancies distinct enough to consider the two as separate species. Asexual structures are not always similar. Zoosporangia of the isolates were consistantly smaller, and lacked basipetalous succession. Gemmae were more numerous than those described for A. recurva. The Embarras River organisms were isolated only from soil samples, whereas A. recurva has been recovered from soil and water.

Therefore, it is felt at this time, in the absence of a more detailed investigation, that the organisms isolated from the Embarras River are not sufficiently similar to the described species Achlya recurva to warrant classifying them together. It is recognized that cultural differences may have some bearing on morphological characteristics, but not enough to contribute to the pronounced differences observed. Descriptions of Achlya recurva are taken from Johnson (1956).

APHANOMYCES DeBary

Jahrb. Wiss. Bot., 2: 178, pls. 19-21. 1860

Hyphae delicate, rarely course, then not exceeding 20u in diameter, hyaline light-brown, sparingly branched or much-branched and contorted; zoosporangia filamentous, of variable length, isodiametric or rarely tapering toward the apex, formed from undifferentiated vegetative hyphae, not proliferating internally, rarely with short side-branches; primary zoospores borne in a single row in the zoosporangia, encysting upon emergence at the orifice as in Achlya or, rarely, swimming prior to encystment as in Leptolegnia; primary zoospore cysts spherical, discharge poroid, papillate or, rarely schistose; secondary zoospores reniform, laterally biflagellate; oogonium terminal on short or long branches, smooth-walled or wall irregularly roughened or ornamented with hyaline or dark, contents homogeneous, finely granular, with or without a conspicuous oil globule; antheridia one to several, long-cylindrical, clavate, or short-tuberous; antheridial branches simple or branched, diclinous, monoclinous, or androgynous in origin; fertilization tubes present or sometimes not visible; specialized gemmae lacking; oospore germination rarely observed, generally by the formation of a long, branched germ-tube.

Saprophytic on plant and animal debris, in soil or water; parasitic on protozoans, rotifers, crustaceans, algae, other phycomycetous fungi, or on the roots of phanerogamous seedlings. Modified from Scott (1961).

Aphanomyces laevis DeBary

Slender branched hyphae; averaging 11u at the base. Gemmae not observed in these isolates. Zoosporangia abundant, not differentiated from subtending hyphae; zoospores typically in a single file, apical emergence is followed by encystment in a sphere at the zoosporangial apex, similar to that exhibited by the genus Achlya. Abundant oogonia; terminal, stalked; spherical or subspherical; averaging 22u in diameter; walls smooth, thin; pits not observed. Single thick walled oospore, usually averaging 17u in diameter; eccentric. Large antheridial branches numerous, persistant; often entwined about the oogonial stalks; androgynous, occasionally diclinous.

Distribution:

Station G-1; soil zone; October; isolated on snakeskin.

Station G; soil zone; October; isolated on snakeskin.

Station H; soil zone; October; isolated on snakeskin.

Station I; soil zone; October; isolated on snakeskin.

Station K; soil zone; March; isolated on snakeskin.

Station G-2; soil zone; June; isolated on snakeskin.

Station G-3; soil zone; June; isolated on snakeskin.

Station G-3; soil zone; June; isolated on snakeskin.

Station J; soil zone; June; isolated on snakeskin.

Discussion:

This common species of Aphanomyces was isolated numerous times from several locations. It agrees with the descriptions given by Beneke (1948)

and by Scott (1961). Investigators have described the parasitic nature of this species (Scott, 1961 and Coker, 1923) but a parasitic isolate has not as yet been collected in the Embarras River.

DICTYUCHUS Leitgeb

Bot. Zeit. 26: 503. 1868; Jahrb. Wiss.

Bot. 7: 357. 1869

Mycelium vigorous, although usually tardy in development, reaching up to 2-3.5cm in diameter on hemp seed; hyphae branched, up to 100u in diameter at the base, straight at first, becoming quite zigzag in older cultures through continued formation of sporangia from the base of which the hypha continues growth. Sporangia formed in great abundance, at first around the outer margin of the culture, later scattered over the entire surface. Primary sporangia long, cylindrical, often thicker in the distal half. Sporangia and spores in the D. monosporus group frequently break away from the hyphae and float on the surface of the water; most of the hyphae become sporangia, sometimes including oogonial stalks and antheridial branches. Sporangial wall persistant, forming a "true-net" in the D. monosporus group, quickly disappearing in the "false-net" group, although the spores cling together to retain the sporangial shape. Spores encysting within sporangia, later emerging through individual openings to the outside in the reniform, laterally biflagellate form, leaving the empty cysts in the form of a true or false-net. In some species the spores in the primary sporangia are discharged as in Achlya, forming a cluster at the sporangial mouth. Gemmae in most species lacking, although abundant in one species. Oogonia, when present, spherical, unpitted, and usually with one eccentric egg. Plants heterothallic, homothallic, parthenogenetic or apparently sexually sterile.

Dictyuchus monosporus Leitgeb

Extensive mycelium; principal hyphae avereging 1.5-2.0cm on hemp seed; moderately branched. Zoosporangia abundant; primary zoosporangia terminal, secondary zooeporangia formed by cymose branching. Zoosporangia average 220u by 22u, but often range from 400u to under 110u in length. Mature zoosporangia often deciduous. Zoospores typically encyst within the zoosporangium; they may be discharged in the manner typical of Dictyuchus, leaving a "true net" type of empty zoosporangium, or have been observed to germinate in situ. Oogonia bear a single oospore; the mature, eccentric oospores are 22u-66u in diameter; spherical; walls smooth, lacking pits. Antheridia diclinous, one to several per oogonium, persistant.

Distribution:

Station H; water zone; October; isolated on hemp seed.

Station G-1; water zone; October; isolated on hemp seed.

Station I; water zone; October; isolated on hemp seed.

Station G-1; water zone; December; isolated on hemp seed.

Station J; water zone; June; isolated on hemp seed.

Station K; water zone; June; isolated on hemp seed.

Discussion:

The above isolates agree closely with the description of D. monosporus given by Beneke (1948). Both organisms exhibit the characteristic "true net" type of empty zoosporangia.

Dictyuchus sp.

Mycelium delicate; diffuse; two week old colony size averaging 3.5-4.0cm. Principal hyphae unbranched, or branched near hyphal tip; hyphae average 33u in diameter. Zoosporangial size varies, ranging from 165u to 880u in length; average size 440u by 33u; clavate, much broader at

zoosporangial apex; renewed by basipetalous succession, occasionally sympodially. Empty zoosporangia are "true net". Zoospore discharge typically dictyoid, although in situ germination is fairly common. No sexual structures were observed in these isolates.

Distribution:

Station G-5; water zone; October; isolated on hemp seed.

Station G-6; water zone; October; isolated on hemp seed.

Discussion:

Both of these isolates appeared to exhibit identical morphological features, and on the basis of the available information, are assumed to be the same species. Beneke (1948) lists only one species as having a "true net" type of zoosporangium. His description of D. monosporus, agrees in many respects to the above described isolate; however, D. monosporus has zoosporangia which are characteristically smaller (112u-412u), and which are renewed by cymose branching. Cymose branching was not observed in the isolates examined. D. monosporus is a dioecious organism, and may not exhibit oogonia in every instance. No attempt was made to mate these isolates with a known strain of D. monosporus because it was felt that the morphological differences were great enough to rule out the possibility of being that organism.

PYTHIUM Pringsheim

Jahrb. Wiss. Bot., 1:304. 1858

Mycelium well developed, consisting of much-branched hyphae, occasionally bearing appressoria, sometimes forming tangled complexes, irregular toruloid elements, and chlamydospores; zoosporangium either entirely filamentous and undifferentiated from the vegetative hyphae, simple or branched, acrogenous or intercalary, or consisting of a series of basal complex

lobulations and a filamentous discharge tube, or a well defined sphaeroidal structure sharply distinct from its supporting hypha and acrogenous, intercalary or laterally sessile, with an emission tube of variable length, sometimes internally proliferous; zoospores somewhat reniform, each containing a single vacuole and with two oppositely directed flagella of approximately equal length. Zoospores are expelled from the sporangium as an undifferentiated mass into a delicate vesicle produced by the tip of the discharge tube where cleavage and maturation takes place, capable of repeated emergence before finally encysting and germinating: plants probably always monoecious; oogonia terminal or intercalary, spherical or subspherical when terminal, ellipsoidal to limoniform when intercalary, smooth-walled, or variously echinulated, for the most part forming a single egg with or without conspicuous periplasm; antheridia none or one to several, hypogynous, monoclinous or diclinous, allantoid, clavate, globose, suborbicular or trumpet-shaped, terminal or intercalary, borne on a short or long stalk, or sessile, usually one to four (may be lacking or if present up to twenty-five) to an oogonium, forming a distinct fertilization tube; oospores usually borne singly within the oogonium, plerotic or aplerotic, wall smooth or reticulate, thin or inspissate, the granular protoplasm usually bearing a conspicuous reserve globule and a lateral refringent body, upon germination forming one or several germ tubes, or zoospores.

Saprophytic and parasitic on plant and animal material in water and soil. Modified from Sparrow (1961).

Pythium rostratum Butler

Relatively large hyphae, up to about 10u in diameter. Zoosporangia spherical or oval; terminal or intercalary, averaging 22u in diameter.

Zoospores not abundant; average 11u in diameter; formed within a vesical. Oogonia spherical to subspherical; often intercalary, occasionally terminal; averaging 22u in diameter. Oospores are pleurotic (filling the oogonium). Antheridia monoclinous, occasionally hypogynous. Oospore single, thick walled, averaging 15u to 20u in diameter.

Distribution:

Station H; soil zone: October; isolated on hemp seed.

Discussion:

The morphology of the above isolate is identical in most respects to that species described by Middleton (1943). The principal difference is the scarcity of the "crooked necked" antheridia mentioned by that author.

Pythium pulchrum von Minden

Slender hyphae, well under 10u in diameter. Zoosporangia spherical or pyriform, terminal or intercalary, occasionally catenulate; averaging 22u-44u in diameter. Oogonia spherical, terminal or intercalary, averaging 33u in diameter. Antheridia hypogynous or monoclinous, usually one or two per oogonium. Oospore thick walled; single; averaging 22u in diameter; eccentric.

Distribution:

Station O; soil zone; October: isolated on snakeskin.

Station G; soil zone; June; isolated on snakeskin.

Discussion:

The major difference between the Embarras River isolates and the organism described by Middleton (1943) is that the Illinois isolates do not consistantly exhibit the highly refractive body present in the oospore with the oil droplet; however, this inconsistancy was not considered to be of great importance, since environmental conditions can often cause slight variations in certain morphological characteristics.

SAPROLEGNIA C. G. Nees

Nova Acta Acad. Leop.-Carol., 11, 513. 1823

Thalli monoecious. Hyphae stout or delicate, branched or unbranched, straight of flexuous, gradually tapering from base to apex; variable in length and diameter. Gemmae when present, formed by segmentation of the hyphae; variable in size and shape; functioning as zoosporangia or oogonia or germinating by one or more slender hyphae. Zoosporangia filiform, cylindrical, clavate, or irregular; terminal at first; renewed by internal proliferation, sympodially or by basipetalous development and cymose branching. Zoospores dimorphic; primary zoospores usually pyriform with two subapical flagella; swimming away from the zoosporangial orifice upon discharge, soon afterward encysting; primary cyst germinating by secondary reniform, laterally biflagellate zoospores; encysted secondary cysts germinating by a slender hypha or by secondary zoospore; polyplanetic; in a few species, aplanoid or dictyoid discharge also present. Oogonia born laterally on stalks of variable length, terminally or in an intercalary fashion, or sessile; variously shaped, predominantly spherical or pyriform, oogonial walls with or without ornamentations; pitted or unpitted. Oospheres generally maturing. Oospores one to many; centric, subcentric, eccentric or subeccentric; variable in size; spherical or ellipsoidal. Antheridial branches, when present, diclinous, monoclinous, androgynous, or hypogynous. Antheridial cells predominantly tubular and clavate; laterally or apically appressed to the oogonial wall or attached by finger-like projections; fertilization tubes usually present. Oospores germinating by hyphae or slender germ tube terminating in a zoosporangium.

Saprolegnia anisospora DeBary

Principle hyphae slender; moderately branched. Gemmae abundant; usually pyriform or irregular; catenulate, occasionally terminal; may function as

zoosporangia when mature. Abundant zoosporangia; clavate, curved; 127u by 24u; renewed by cymose branching and internal proliferation; zoospore discharge saprolegnioid. Stalked oogonia abundant; lateral, terminal and intercalary; spherical or pyriform; 45u-70u in diameter. Oogonial wall smooth; pitted under point of antheridial attachment; occasional pits may be obscure. Oogonial stalks short, straight or rarely bent. Oospores eccentric, generally 2-8 per oogonium, occasionally more in larger oogonia; usually filling the oogonium; 15u-22u in diameter. Antheridial branches diclinous, rarely monoclinous; numerous; persistant.

Distribution:

Station H; soil zone; June; isolated on hemp seed.

Station H-1; soil zone; June; isolated on hemp seed.

Discussion:

Beneke (1968) does not refer to a species of Saprolegnia exhibiting eccentric oospores. The above isolate exhibits this type of oospore and an occasional subcentric oospore; Seymour (1970) describes this mixed condition in six of the 14 isolates of S. anisospora examined by him.

This species has not been previously recorded in Illinois.

Saprolegnia ferax (Gruith) Thuret

Mycelium extensive; principle hyphae stout, moderately branched. Gemmae present or absent; pyriform or irregular; terminal or intercalary; usually single, occasionally catenulate; function as zoosporangia when mature. Zoosporangia very abundant, clavate and filiform; straight or bent; averaging 36u by 325u; renewed by internal proliferation, occasionally by cymose branching; zoospore discharge saprolegnioid; oogonia abundant; may form only in older cultures; terminal or intercalary; occasionally lateral; spherical or pyriform; 55u-66u in diameter; oogonial wall smooth,

conspicuously pitted; oogonial stalks straight, only rarely bent. Oospores centric; spherical; usually germinate into slender hyphae bearing a small terminal zoosporangium. Antheridial branches may be lacking; monoclinous or androgynous; apically or laterally appressed.

Distribution:

Station H-2; water zone; March; isolated on hemp seed.

Station K; water zone; March; isolated on hemp seed.

Station J; water zone; June; isolated on hemp seed.

Station K; water zone; June; isolated on hemp seed.

Discussion:

S. ferax is described as being the most abundant species of Saprolegnia, and is listed as the type species for the genus. The great degree of variety exhibited by this ubiquitous genus has given rise to the S. ferax "complex" which compromises S. mixta, S. monoica and S. ferax, per se. The overlapping characteristic of these species has given rise to much controversy as to the validity of having three distinct species. The morphology of the above isolates more closely agrees with the description of S. ferax as given by Seymour (1970), and is not considered to be either of the other two genera.

Saprolegnia turfosa (Minden) Gaumann

Mycelium dense; principle hyphae stout, sparingly branched. Gemmae absent or rare; pyriform or irregular when present; usually single and terminal. Zoosporangia not abundant; filiform; averaging 25u by 300u; renewed by internal proliferation. Zoospore discharge saprolegnioid. Oogenial stalked, abundant; usually lateral, occasionally terminal, rarely intercalary; oogonial wall smooth; small papillae rarely present. Pits conspicuous, numerous. Oogonial stalks short, usually no longer than

the diameter of the oogonium, usually shorter; straight. Antheridial branches monoclinous, occasionally androgynous. Oospheres maturing. Oospores centric; spherical; filling the oogonium; 9-11 per oogonium; 22u-26u in diameter.

Distribution:

Station G; soil zone; June; isolated on hemp seed.

Station G-1; soil zone; June; isolated on hemp seed.

Station G-2; soil zone; June; isolated on hemp seed.

Discussion:

Of the 14 isolates examined by Seymour (1970), only one formed the papillae on some oogonia. This was also true of the Embarras River isolates in that the small projections were only very rarely observed.

Saprolegnia sp.

Mycelium diffuse, extensive; sparcely branched, many hyphae unbranched. Principle hyphae average 1.5cm in length and 30u-40u in diameter. Primary zoosporangia terminal, averaging 550u by 44u in size; straight or slightly curved; often branched, with each branch bearing an apical exit pore; secondary zoosporangia are formed by internal proliferation. Gemmae are abundant; filiform, clavate, or irregular, occasionally pyriform, often branched; terminal or intercalary; germination not observed; oospore formation not observed. Sexual structures not observed in these isolates.

Distribution:

Station I; water zone; March; isolated on hemp seed.

Station I; water zone; June; isolated on hemp seed.

Discussion:

Beneke (1948) described Saprolegnia parasitica as being the only Illinois isolate not bearing oogonia under natural conditions, when grown on hemp seed. He further characterizes this species as having hyphae

whose medial diameter is 23u-38u with abundant irregularly shaped gemmae; and numerous internally proliferating zoosporangia, with the average dimensions of 210u-525u by 31u-56u; the similarity of asexual structures would suggest a relationship between this isolate and S. parasitica, but this cannot be proven until the Embarras River isolate can be shown to be parasitic to fish, and only then exhibit identical sexual characteristics.

THRAUSTOTHECA Humphrey

Trans. Am. Phil. Soc. II, 17: 131. 1893.

Hyphae for the most part stout, branching. Zoosporangia clavate to subcylindrical, frequently irregular, proliferating from below the old ones. Zoospores always, or in most cases, encysting within the zoosporangium after formation, more or less angular forms, then later, swelling and escaping by an irregular rupture or disintegration of the sporangial wall; not escaping immediately by an apical papilla except in the achlyoid primary zoosporangia of one species. Oospores one to several in each oogonium, eccentric. Antheridia present. From Beneke (1948).

Thraustotheca primoachlya

Dense mycelium; hyphae averaging 11u-33u in diameter; profusely branched. Gemmae not observed in this isolate. Zoosporangia abundant. Primary zoosporangia terminal, oospore discharge achlyoid; secondary zoosporangia laterally arranged, zoospore discharge thraustothecoid.

Distribution:

Station G-2; flood zone; October; isolated on hemp seed.

Station G-3; flood zone; October; isolated on hemp seed.

Station H; flood zone; October; isolated on hemp seed.

Station H-1; flood zone; October; isolated on hemp seed.

Concluding Remarks:

Of the 156 samples collected, 71 viable isolates of filamentous Phycomycetes were recovered. The genera identified were Achlya (7 species), Allomyces (2 species), Aphanomyces (1 species), Dictyuchus (2 species), Pythium (2 species), Saprolegnia (4 species), and Thraustotheca (1 species).

Of the three zones sampled, the "soil" was by far the most productive, yielding 41 isolates; the "water" zone yielded 21 isolates, and the "flood" zone yielded 9 isolates. A detailed vertical distribution study was not attempted at this time; therefore, no accurate correlation can be made between the zone and the organisms found therein.

From the wide choice of baiting material, only two types were utilized, and only one sampling technique was employed. It is logical to assume that a wider variety of baiting material and collecting techniques would yield species of filamentous Phycomycetes in addition to those described above.

Many cultures failed to produce evidence of fungal mycelia. This phenomenon may have a number of explanations. As previously mentioned, the baits utilized may not have been appropriate for certain species. The presence of contaminating microorganisms such as protozoans, bacteria, algae, and other fungi may have had an inhibitory effect on the growth of the Phycomycetes by altering the nature of the culture medium (e.g., changing the pH, or reducing the availability of organic and inorganic nutrients). Another possibility to consider would be the failure to collect viable phycomycetous spores or mycelia when the samples were taken. The composition of the culture medium based on sterile water could be sufficiently different from that of the river water to prevent the growth of fungi that might have otherwise been present in the collected sample. Finally, the Embarras River is the recipient of the polluting effluents

from a variety of sources. Such material dissolved in the river could possibly create an environment which is unable to support fungal mycelia.

The distribution of the phycomycetous fungi found in the Embarras River seems to vary seasonally, with the greater abundance of organisms recovered in October and June. From these results it can be tentatively concluded that certain species do exhibit seasonal periodicity. However, additional collections should be made in order to confirm this assumption. The relative abundance of fungi in October and June may be explained by the fact that during the early summer there is a greater supply of available nutrients, and higher optimum temperatures, and in early fall there is an abundance of naturally occurring host material (e.g., decaying organic matter such as leaves and the exoskeletons of summer insects), as well as favorable temperature.

The Chytridiales and Hyphochytridiales were excluded from this study. A more complete population study should include these organisms, and their distribution should be determined prior to the completion of the reservoir dam.

SUMMARY

Over a period of ten months, a collection of soil and water samples has been taken from the southerly sector of that portion of the Embarras River which flows through Coles County, Illinois. A total of 13 collecting stations was selected, and three zones at each station were sampled. Collections were made in October, December, March, and June. Every effort was made to consistently collect samples from the same locality at the time of each collection.

Filamentous Phycomycetes were found at every station, and in each zone. The distribution varied seasonally, with the greater abundance of isolates recovered in October and June. Of the three zones, the soil zone was by far the most productive; the water and flood zones yielded substantially fewer organisms.

Twenty identified species and two non-fruiting isolates representing 7 genera are described. Achlya diffusa and Saprolegnia anisospora are listed as possible new records for the state of Illinois. One isolate is suggested as a possible new species of Achlya, and a comparative morphological study of this isolate with a well defined isolate of A. recurva is described. Achlya conspicua appears to be endemic for one locality.

It is evident from the above results that filamentous Phycomycetes are ubiquitous for this portion of the Embarras River and its adjacent areas. Any changes in this population should become evident as the Lincoln Reservoir is completed.

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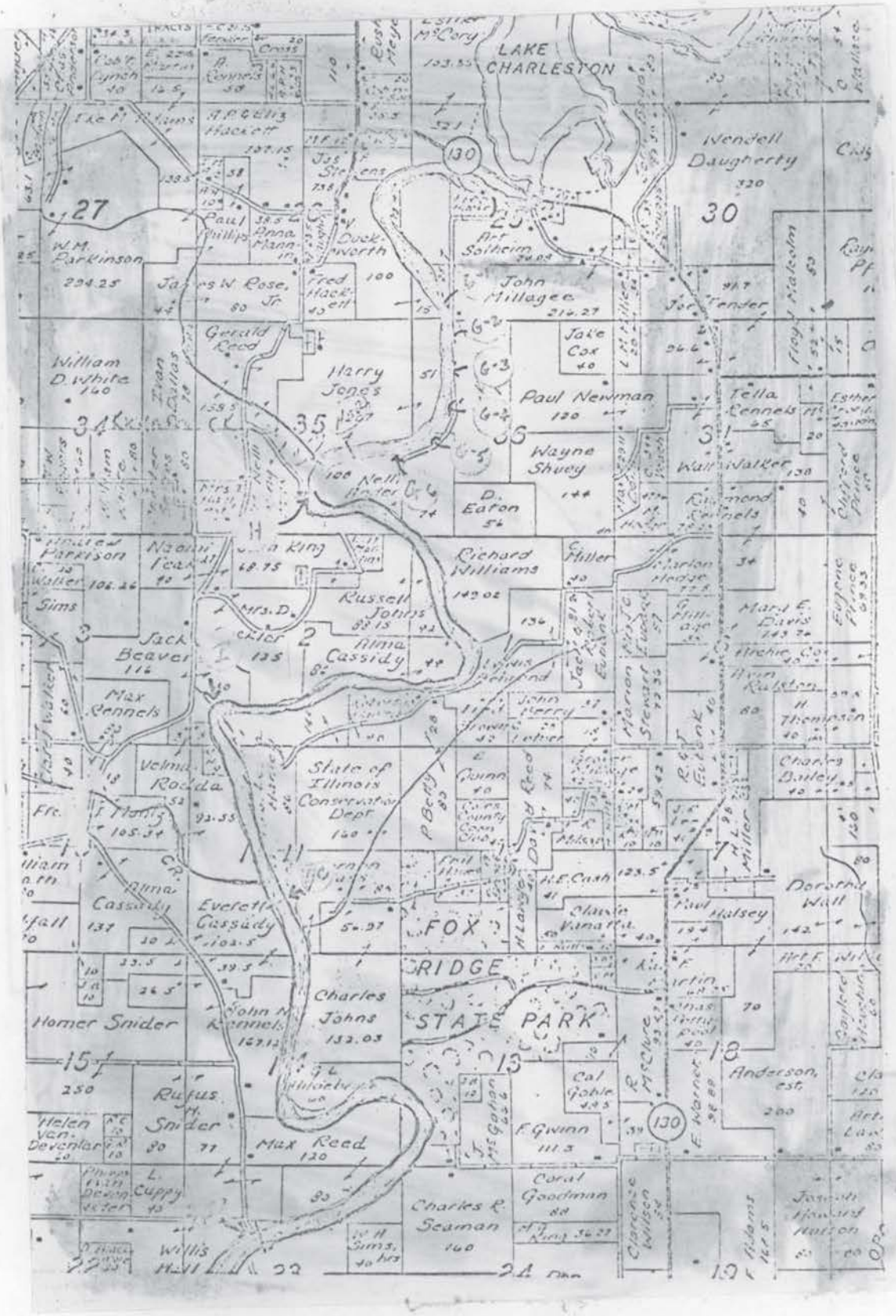


The Embarras River, Coles County, Illinois

Figure 1

PLATE II

Sexual Morphology

a. Diclinous antheridial origin

b. Monoclinous antheridial origin

c. Androgynous antheridial origin

d. Hypogynous antheridial origin

e. Centric oospore

f. Eccentric oospores

g. Eccentric oospores

h. Subcentric oospore

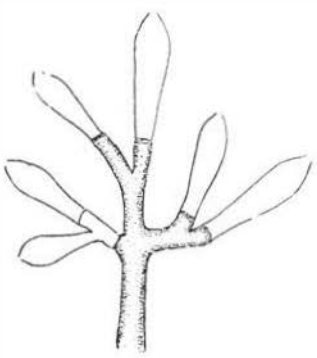
The Embarras River, Coles County, Illinois:

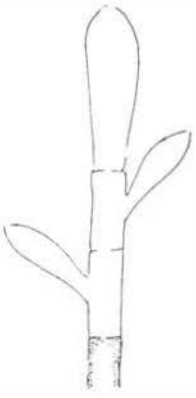
Physical and Chemical Ranges

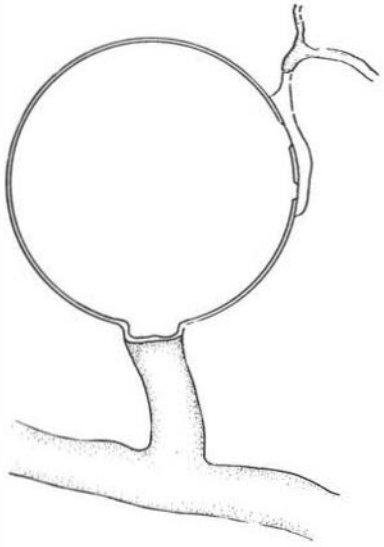
Factor	Range	
	September 1969	March 1970
Water temp. (°C)	21°	4°
Air temp. (°C)	25°	17°
Dissolved O_2 (ppm)	12.0	7.0
Turbidity (JTU)*	60.0	,5.0
pH	8.7	7.5
NO_3 (ppm)	88.00	0.00
NO_2 (ppm)	5.36	0.00
CO_2 (ppm)	+	40.00
Hardness (ppm)	490.00	240.00
PO_4 (ppm)	14.00	0.25

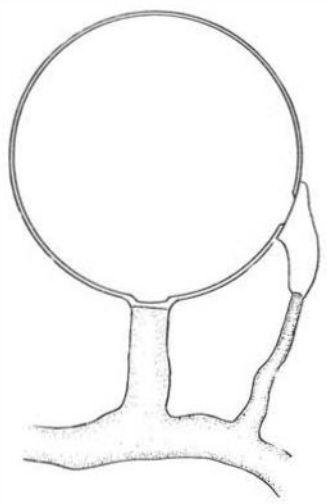
* Jackson Turbidity Units

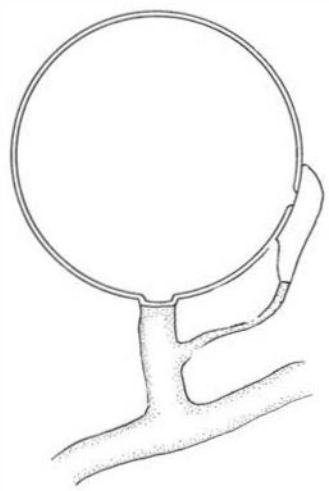
+ No Reading Available

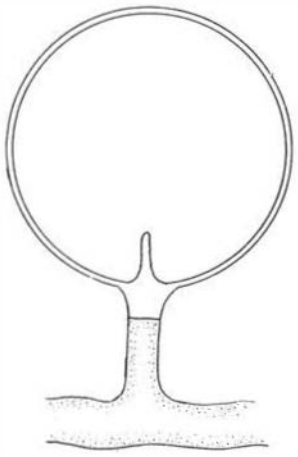












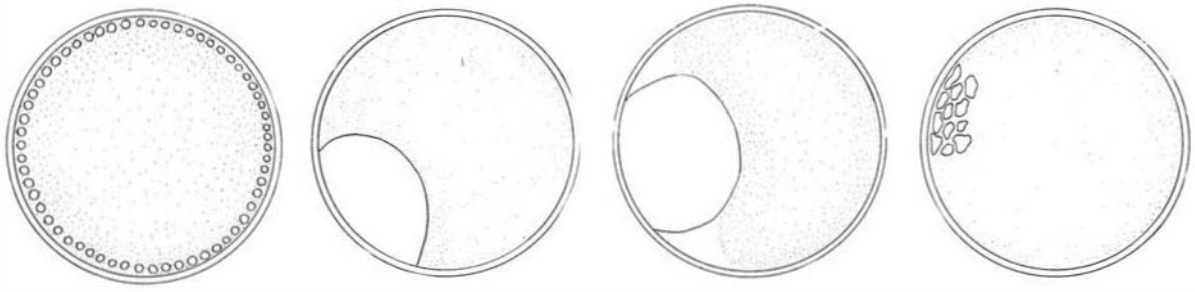


PLATE III

Achlya sp. (10-24-69 G-1)

Sexual and Asexual Morphology

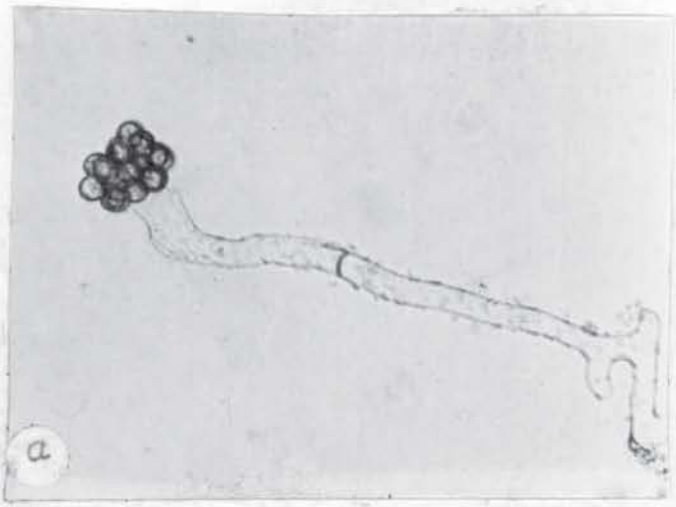
a. Zoosporangium with encysted zoospore cluster. Note: rare curved zoosporangium. 300X

b. Chlamydospore (gemma). 600X

c. Mature oogonium with attending antheridia. Note: recurved oogonial stalk. 625X

d. Mature oogonium. Note: eccentric oospores. 500X

e. Enlargement of oogonial ornamentations. 1.250X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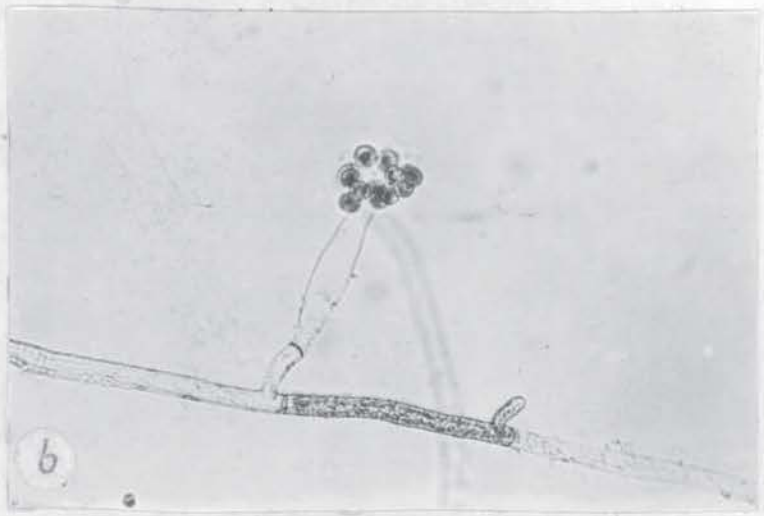
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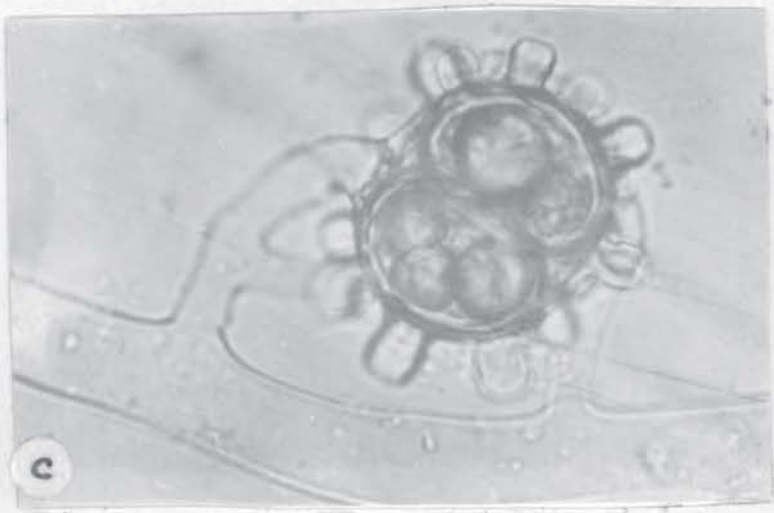
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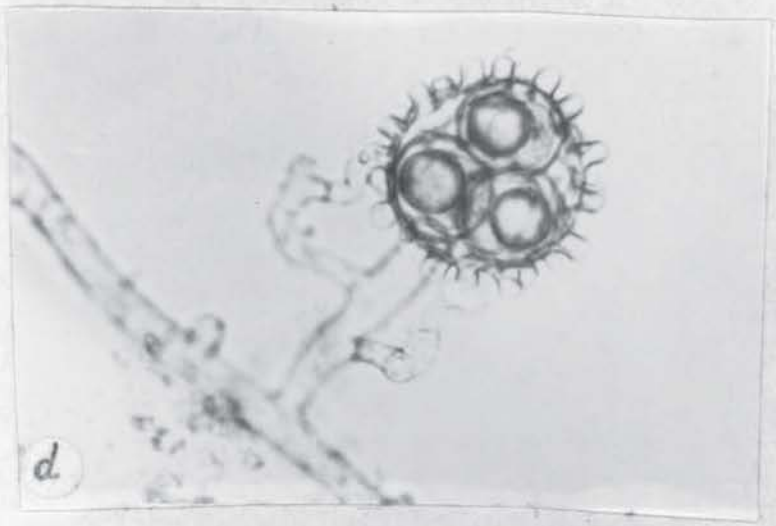
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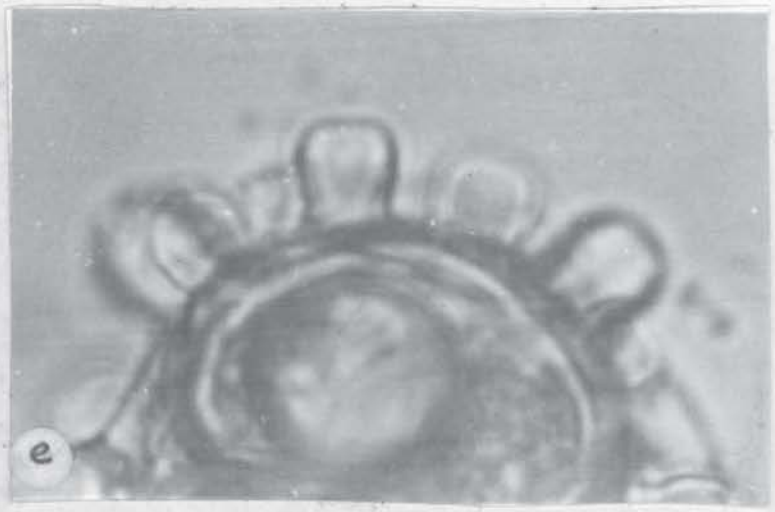
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