Myxomycetes isolated from submerged plant material collected in the Big Thicket National Preserve, Texas

Winsett KE[1*] and Stephenson SL[2]

[1]Department of Biology, 8600 University Boulevard, University of Southern Indiana, Evansville, Indiana, 47712 USA katherine.winsett@gmail.com
[2]Department of Biological Sciences, 1 University of Arkansas, Fayetteville, Arkansas, 72701 USA slsteph@uark.edu

Winsett KE, Stephenson SL 2013 – Myxomycetes isolated from submerged plant material collected in the Big Thicket National Preserve, Texas. Mycosphere 4(2), 227–231, Doi 10.5943/mycosphere/4/2/5

Samples of submerged plant material obtained from streams and other bodies of water at 11 different collecting sites in management units of the Big Thicket National Preserve in Texas were used to prepare 90 moist chamber cultures for the isolation of myxomycetes. These cultures were maintained in the laboratory for 10 weeks, during which they yielded 14 species representing 12 genera. This total included five species (*Licea belmontiana*, *Craterium concinnum*, *Diachea bulbillosa*, *Oligonema schweinitzii*, and *Physarum echinosporum*) not recorded previously for the Big Thicket National Preserve and one species (*D. bulbillosa*) new for the state of Texas. Although usually not included in biodiversity surveys for myxomycetes, our data suggest that submerged plant material may support a few species that may be missed if only terrestrial habitats are considered.

Key words – All Taxa Biodiversity Inventory – aquatic myxomycetes – biodiversity – moist chamber culture – state record

Article Information
Received 11 February 2013
Accepted 3 March 2013
Published online 23 March 2013
*Corresponding author: Katherine Winsett – e-mail – katherine.winsett@gmail.com

Introduction

Myxomycetes (plasmodial slime molds or myxogastrids) are generally known from terrestrial habitats, where they are associated with various types of dead and decaying plant material (Stephenson and Stempen 1994). However, there have been a number of reports of these organisms from aquatic habitats. For example, Ward (1886) described the occurrence of the fruiting bodies of *Didymium difforme* on the completely submerged roots of water hyacinth. Later, Parker (1946) demonstrated that it was possible to culture the plasmodia of three different species of myxomycetes while these were submerged in water, although she did not observe the formation of fruiting bodies under these conditions. Kappel and Anken (1992) noted the presence of a plasmodium on the inside of an aquarium, something that has been observed by the second author on several occasions. Gottsberger and Nannenga-Bremekamp (1971) actually described what they considered to be an aquatic myxomycete (*Didymium aquatile*) from Brazil as a species new to science. To our knowledge, there have been only two studies

Table 1 Collecting sites in the Big Thicket National Preserve.

Site ID	Preserve Unit	Lat/Long	Type of plant community*	Type of aquatic habitat
H-2	Beaumont	30.23795/ -94.10513	Floodplain hardwood forest	cypress-tupelo swamp
BSU-1	Big Sandy Creek	30.61359/ -94.67659	Floodplain hardwood forest	stream
BEAVERSLIDE	Big Sandy Creek	30.57584/ -94.64314	Flooded cypress-tupelo	pond
B-CR	Big Sandy Creek	30.57725/ -94.64452	Floodplain hardwood forest	stream
CL-1	Canyonlands	30.74714/ -94.15192	Lower slope hardwood pine forest	stream
CL-2	Canyonlands	30.74794/ -94.15141	Lower slope hardwood pine forest	pool near spring
CL-N	Canyonlands	30.75009/ -94.14631	Cypress-tupelo swamp forest	stream
CN-B	Canyonlands	30.71804/ -94.13658	Lower slope hardwood pine forest	pond
CN-D	Canyonlands	30.71935/ -94.13921	Lower slope hardwood pine forest	stream
SEGNO	Menard Creek Corridor	30.57008/ -94.70122	Floodplain hardwood forest	stream
LR	Lance Rosier	30.26285/ -94.51449	Palmetto-hardwood flatland forest	pond

*Plant communities are based upon information in Marks and Harcombe (1981), Brown et al. (2006a, b), Watson (2006), Brown et al. (2009).

that attempted to survey the myxomycetes associated with aquatic habitats. Shearer and Crane (1986) recorded 13 species from submerged, decayed plant substrates and balsa wood baits from swamps in southern Illinois, whereas Lindley et al. (2007) recovered four species from dead plant parts collected from below the surface of the water in five small ponds in northwestern Arkansas and northeastern Oklahoma. The objective of the research reported herein was to carry out a more intensive survey of the myxomycetes associated with aquatic habitats. The survey was carried out in the context of an inventory of these organisms in the Big Thicket National Preserve in Texas.

General Study Area

This study was carried out in the Big Thicket National Preserve (NP), which encompasses a large portion of the remaining area historically referred to as the biological Big Thicket. The Big Thicket region may have once covered a total area of nearly 1.5 million ha but is now highly impacted by human activities, including oil and gas exploration and the development of commercial forests (Gunter 1993, Diggs et al. 2006, Watson 2006). The Big Thicket NP includes just over 40,000 ha divided among 15 management units spread across seven counties.

The Big Thicket is located within the West Gulf Coastal Plain in southeastern Texas and is characterized by an exceedingly diverse series of different biological habitats formed as a result of the co-occurrence in a single area of multiple ecosystems, including elements of the eastern hardwood forests, central North American grasslands, subtropical coastal plains, and southeastern swamps (Diggs et al. 2006, Watson 2006). This portion of southeastern Texas has a warm, humid, subtropical climate with generally higher amounts of rainfall than most other areas of Texas. This results in the presence of a number of wetland habitats, including upland wet pine savannahs, tupelo-cypress swamps, and wetland baygalls (Marks and Harcombe 1981, Diggs et al. 2006, MacRoberts and MacRoberts 2008).

The plant material examined in this study was collected from 11 study sites representing five Preserve units (Table 1). All material was collected from freshwater habitats within the Preserve. Six of these were shallow streams, three sites were at the edges of shallow ponds, one site was a pool near the spring that feeds a freshwater stream, and one site was at the edge of a cypress-tupelo swamp (Figure 1).

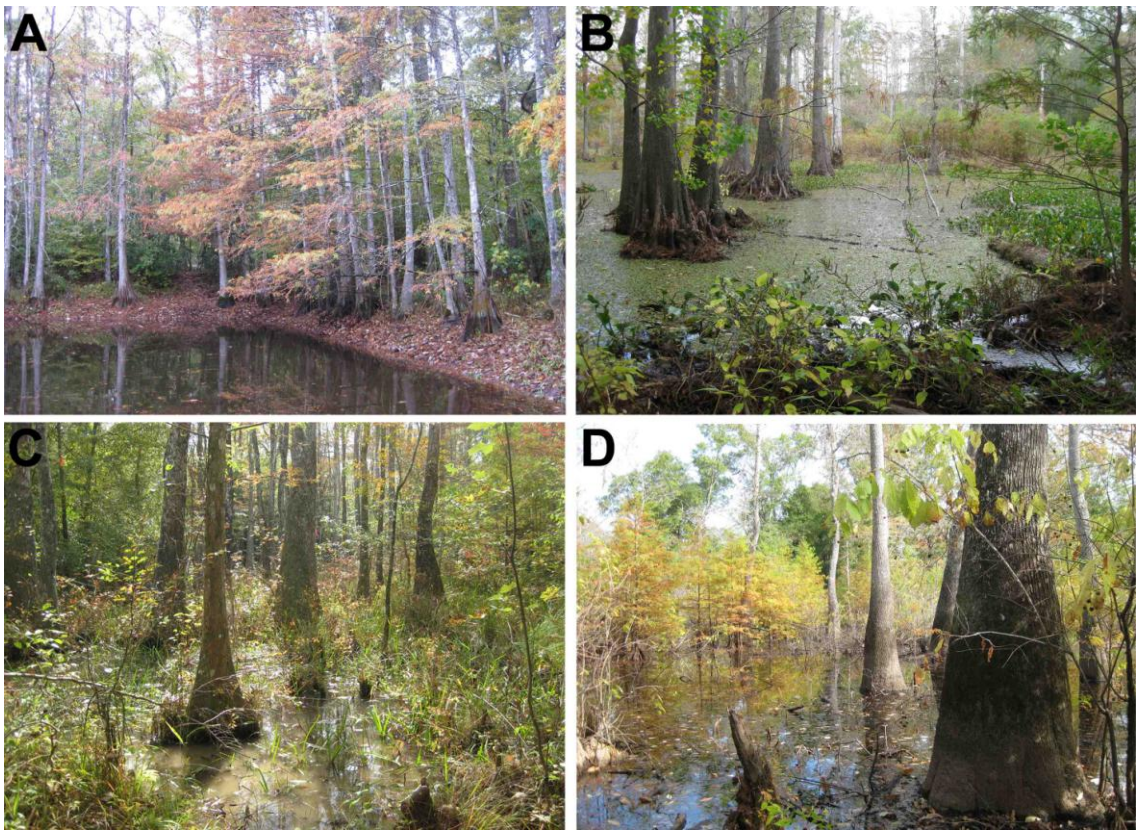

Fig. 1 – a. Pond at Beaverslide trail. b. Beaver pond in Canyonlands Unit c. cypress-tupelo swamp in northern end of Canyonlands Unit d. cypress-tupelo swamp in Houseman Tract, Beaumont Unit.

Materials and Methods

During the period of November 8-13 in 2010, 34 samples of dead and decaying plant material from freshwater streams and pools within the Big Thicket National Preserve were collected and placed in plastic bags. Twenty-nine samples consisted of plant material completely submerged in fresh water, and five samples consisted of material floating on the water surface or only partially submerged. Though collected in plastic in the field, the sample material in each was transferred to a paper bag and allowed to air dry as soon as they were transported out of the field and to the research station. Later, this dried material was processed using the moist chamber culture technique as described by Stephenson and Stempen (1994).

Ninety moist chamber cultures were prepared. Three replicate cultures were prepared from most of the samples, but only one or two cultures were prepared for a few samples because the amount of material available was limited. Each moist chamber culture consisted of a disposable, sterile plastic Petri dish. Enough sample material was placed in the Petri dish to form a single layer over a disk of filter paper placed in the bottom of the dish. The Petri dish was filled with sterile deionized water to cover the sample material and left standing to soak for approximately 24 hours. After this period of time, the pH of the water remaining in each dish was determined with the use of a portable pH meter. Excess water was then poured from the dish, and the latter stored on a shelf out of direct sunlight and at room temperature.

Moist chamber cultures were maintained for a period of approximately 10 weeks and checked weekly for any evidence (plasmodia or fruiting bodies) of myxomycetes. Deionized water was added as necessary to keep the sample material moist without having any standing water in the dish. Mature fruiting bodies and a portion of the substrate upon which they occurred were removed and preserved for permanent storage by mounting, using white glue, the substrate on slips of acid-free cardstock paper, which was then placed in small pasteboard pill boxes. All fruiting bodies of the same species obtained from the same moist chamber culture were considered to

Table 2 Occurrence of myxomycetes on plant material collected from aquatic habitats. The number of records from completely submerged material is given in parentheses.

Species	Number of records	pH range	Mean pH
Arcyria cinerea (Bull.) Pers	2 (2)	5.9-6.2	6.1
Craterium concinnum Rex	1 (1)	4.3	–
Diachea bulbillosa (Berk. & Broome) Lister	1 (1)	6.8	–
Diachea leucopodia (Bull.) Rostaf.	3 (2)	4.8-5.8	5.5
Diderma effusum (Schwein.) Morgan	23 (20)	4.3-7.5	6.4
Didymium minus (Lister) Morgan	2 (2)	5.9-6.2	6.1
Didymium squamulosum (Alb. & Schwein.) Fr	1 (1)	6.1	–
Lamproderma scintillins (Berk. & Broome) Morgan	10 (10)	5.3-7.6	6.7
Licea belmontiana Nann.-Bremek	1 (1)	5.9	–
Metatrichia vesparia (Batsch) Nann.-Bremek. ex G.W. Martin & Alexop.	1 (1)	6.8	–
Oligonema schweinitzii (Berk.) G.W. Martin	1 (0)	4.8	–
Perichaena depressa Lib.	1 (1)	6.1-6.7	6.4
Physarum echinosporum Lister	1 (1)	5.7	–
Stemonitis fusca var. *nigrescens* (Rex) Torrend	2 (2)	4.8-5.8	5.5

represent one record or collection. All collections were deposited in the mycological herbarium (UARKM) of the University of Arkansas. Nomenclature used herein essentially follows Lado (2005-2012).

The occurrence of a particular species in at least one of the replicate moist chamber cultures prepared for each sample was considered to represent a single record. In some, instances, the same species appeared in all of the replicate cultures prepared from a given sample. The mean pH for each species was derived from the values recorded for all of the moist chamber cultures in which the species occurred, including the replicate cultures from the same sample.

Results

Of the 90 moist chamber cultures prepared with samples of material obtained from aquatic habitats, 80% produced some evidence of myxomycetes. Only two of the 34 samples (a sample of *Riccardia multifida* and a sample of a submerged grass collected from a stream) produced no evidence of myxomycetes. The positive cultures yielded a total of 14 species representing 12 different genera, although one species (*Oligonema schweinitzii*) was recovered from a sample consisting of plant material that had not been completely submerged.

The two most common species recorded were *Diderma effusum* (23 collections) and *Lamproderma scintillans* (10 collections). The former was recorded from all 11 collecting sites, whereas *L. scintillans* was recorded from only three collecting sites, all of which were within the Canyonlands Unit. More than half (8) of the species isolated from aquatic habitats were represented by only a single record, with three others represented by two or three records.

Values of pH determined for moist chamber cultures ranged from 4.3 to 7.5, with a mean value of 6.4. Because of the limited number of records for most species, their distributional relationships with respect to pH could not be evaluated. However, it can be noted that the two most common species were recorded over a wide range of pH conditions (Table 2).

Discussion

The 14 species recovered included five examples (*Licea belmontiana, Craterium concinnum, Diachea bulbillosa, Oligonema schweinitzii*, and *Physarum echinosporum*) not recorded previously for the Big Thicket National Preserve and one species (*Diachea bulbillosa*) that was a new record for the state of Texas (Winsett and Stephenson 2012). Consequently, our data suggest that the majority of myxomycetes associated with aquatic habitats are not particularly common. Interestingly, *Lamproderma scintillins* was last collected in 1971 (by C. J. Alexopoulos) and

was not recorded by the first author during the course of intensive surveys of terrestrial habitats carried out between 2007 and 2010. As such, its relative abundance in aquatic habitats is surprising.

As already noted, five of the species we recorded from aquatic habitats had not been collected previously in the Preserve despite a significant collecting effort that extended over four years (Winsett and Stephenson 2012). Although usually not included in biodiversity surveys for myxomycetes, our data suggest that submerged plant material may support species that could be missed if only terrestrial habitats are considered.

Acknowledgements

This work was carried out in cooperation with and funding from the Big Thicket Association. Additional funding was provided by the National Science Foundation (grant DEB-0316284 to the University of Arkansas), and the University of Arkansas Department of Biological Sciences. Scientific and logistical support from the personnel at the Big Thicket National Preserve and the staff at the Big Thicket Field Research Station was invaluable for the success of this project. We are grateful to the contributors to the field survey effort including Dale Kruse, Paul Roling, and Sophia Sweeney.

References

Brown LE, MacRoberts BR, MacRoberts MH, Harcombe PA, Pruess WW, Elsik IS, Jones SD. 2006a – Annotated checklist of the vascular flora of the Lance Rosier Unit of the Big Thicket National Preserve, Hardin County, Texas. Sida, Bot. Misc. 22, 1175–1189.

Brown LE, MacRoberts BR, MacRoberts MH, Harcombe PA, Pruess WW, Elsik IS, Jones SD. 2006b – Annotated checklist of the vascular flora of the Big Sandy Creek Unit, Big Thicket National Preserve, Texas. Sida, Bot. Misc. 22, 705–723.

Brown LE, MacRoberts BR, MacRoberts MH, Pruess WW. 2009 – Annotated checklist of the vascular flora of the Menard Creek Corridor Unit of the Big Thicket National Preserve, Polk, Liberty, and Hardin Counties, Texas. J. Bot. Res. Inst. Texas 3, 443–455.

Diggs GM, Lipscomb BL, Reed MD, O'Kennon RJ. 2006 – Illustrated flora of east Texas. Sida, Bot. Misc. 26, 1–1594.

Gottsberger G, Nannenga-Bremekamp NE. 1971 – A new species of *Didymium* from Brazil. Koninkl Nederl Akad Van Wetenschappen Ser C 74, 264–268.

Gunter PAY. 1993 – The Big Thicket: An Ecological Reevaluation. University of North Texas Press, Denton TX. 243 pp.

Kappel T, Anken RH. 1992 – An aquarium myxomycete: *Didymium nigripes*. Mycologist 6, 106–107.

Lado C. 2005-2012 – An on line nomenclatural information system of Eumycetozoa. http://www.nomen.eumycetozoa.com. Accessed on 15 September 2012.

Lindley LA, Stephenson SL, Spiegel FW. 2007 – Protostelids and myxomycetes isolated from aquatic habitats. Mycologia 99, 504–509.

MacRoberts MH, MacRoberts BR. 2008 – The Big Thicket of Texas as floristically unique habitat. J. Bot. Res. Inst. Texas 2, 665–671.

Marks PL, Harcombe PA. 1981 – Forest vegetation of the Big Thicket, southeast Texas. Ecol. Monogr. 51, 287–305.

Parker H. 1946 – Studies on the nutrition of some aquatic myxomycetes. J. Elisha Mitchell Soc: 62, 232–247.

Shearer CA, Crane JK. 1986 – Illinois fungi XII. Fungi and myxomycetes from wood and leaves submerged in southern Illinois swamps. Mycotaxon 25, 527–538.

Stephenson SL, Stempen H. 1994 – Myxomycetes: a Handbook of Slime Molds. Timber Press, Portland, Oregon.

Ward MH. 1886 – The morphology and physiology of an aquatic myxomycete. Quarter J. Microscop Sci 24, 64–86.

Winsett KE, Stephenson SL. 2012 – An annotated checklist of the myxomycetes of the Big Thicket National Preserve, Texas. J. Bot. Res. Inst. Texas 6, 287–298.