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Myxomycetes associated with bryophyte mats in the Mountain Lake area of southwestern Virginia

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Abstract: Samples of the bryophyte (but mostly mosses) mats found on soil, decaying logs, and the bases of trees in the Mountain Lake area of southwestern Virginia were collected and used to prepare a series of moist chamber cultures. Twenty-two of the 45 cultures (49%) yielded evidence (plasmodia or fruiting bodies) of myxomycetes, and six species were recorded. These data suggest that bryophyte mats are a microhabitat for myxomycetes in temperate deciduous forests, but both species richness and abundance of these organisms are relatively low.

Key words: ecology, moist chamber cultures, mosses, slime molds, temperate deciduous forests

Introduction

Myxomycetes, also called plasmodial slime molds or myxogastrids, are a group of amoeboid protists usually present and often abundant in terrestrial ecosystems, especially various types of forests. The myxomycete life cycle encompasses two very different trophic stages, one consisting of uninucleate amoebae, with or without flagella, and the other consisting of a distinctive multinucleate structure, the plasmodium (Martin et al. 1983). Under favorable conditions, the plasmodium gives rise to one or more fruiting bodies (also referred to as sporocarps) containing spores. Although the fruiting bodies produced by myxomycetes are somewhat suggestive of those produced by certain higher fungi, they are considerably smaller (usually no more than 1–2 mm tall) and totally different in structure.

The fruiting bodies of myxomycetes develop in the field under natural conditions and are typically found in association with such substrates as decaying wood and other types of dead organic matter. Fruiting bodies also appear on samples of dead organic matter (especially pieces of the dead outer bark of living trees and various types of plant debris when these are collected, returned to the laboratory and used to prepare what are known as moist chamber cultures [Stephenson and Stempen 1994]). In both cases, the fruiting bodies can be collected and placed in small pasteboard boxes for permanent storage. If properly curated, fruiting bodies remain suitable for study for many years.

Although it might be assumed that the substrate on which the fruiting bodies of a myxomycete develop is the same one that supported the trophic stages, the ability of the plasmodium to migrate some distance before fruiting has the potential of causing this not to be the case. Myxomycete plasmodia are known to migrate to the highest and driest part of a substrate prior to forming fruiting bodies. Therefore, it seems likely that the upper portions of a bryophyte mat provide a convenient exposed surface for fruiting. A number of species of myxomycetes are often found in association with bryophytes (Gray and Alexopoulos 1968). For example, Martin and Alexopoulos (1969) mentioned bryophytes (e.g., …usually associated with mosses….among mosses…(on) mosses and liverworts…mossy logs and stumps…often fruiting on mosses) in the habitat descriptions of some 49 different species of myxomycetes, and the occurrence of myxomycetes on mosses has been noted in a number of instances (e.g., Kaiser 1913, Farr 1979).

Stephenson and Studlar (1985) reported 170 collections of myxomycetes that had fruited on bryophytes. Fifty-two species of myxomycetes were represented among the 170 collections. More than half (55%) of the collections considered in their study were obtained in the general vicinity of the University of Virginia's Mountain Lake Biological Station in southwestern Virginia. The majority of the myxomycetes they recorded were relatively common species not known to be consistently associated with bryophytes. Consequently, the association of the two organisms seemed likely to be fortuitous and not ecologically significant.

However, the question might be asked as to whether there are some species of myxomycetes that are associated with the microhabitat represented by bryophyte mats. Stephenson and Rojas (2020) demonstrated that mosses can serve as spore traps for myxomycetes. They placed small-mesh nylon bags filled with autoclaved samples of mosses out in the field in Costa Rica and northwest Arkansas. The bags were suspended some distance above the ground and left in place for more than four months. When the bags were recollected, the samples of mosses were used to prepare a two series of moist chamber cultures. Thirty-eight of the 40 (95%) cultures placed out in Costa Rica were positive for myxomycetes, and 28 of 40 (70%) of cultures placed out in northwest Arkansas yielded evidence of myxomycetes. Numbers of species recorded were 14 and 3, respectively. The much lower yield for northwest Arkansas was attributed to the fact that the forest was rather xeric, presumably with few myxomycetes forming fruiting bodies under natural conditions in the field. In contrast, the forests at Mountain Lake would be regarded as relatively moist, and myxomycete fruiting bodies are often abundant as reported by Stephenson (1988).

However, the question still remains as to whether mosses represent a primary microhabitat for myxomycetes in forests. In the present study, bryophyte (but mostly mosses) mats were collected from the field, returned to the laboratory, and placed in moist chamber cultures to determine what myxomycetes might be present.

Materials and methods

This study was carried out in the Mountain Lake area (37°22'N, 80°37'W; elevation ca 1168 m) of Giles County in southwestern Virginia, which is in the Ridge and Valley physiographic province of the southern Appalachian Mountains (Fenneman 1938). Temperate deciduous forests are present at all of the localities where samples of bryophytes were collected, and these forests are largely dominated by various species of oaks (Stephenson 1988). Bryophyte mats are not uncommon and occur on the ground, on decaying logs (Figure 1), and on bark at the bases of some of the trees. *Thuidium delicatulum* (Hedw.) Schimp. is the dominant mat-forming moss in the Mountain Lake area, but other species encountered were *Dicranum scoparium* Hedw., *Leucobryum glaucum* (P. Beauv.) Lindb., *Polytrichum commune* Hedw., *Pleurozium schreberi* (Brid.) Mitt., and *Hypnum curvifolium* Hedw. Many of the mats consisted of a mixture of two or more species. Mosses were identified with the use of McKnight et al. (2013). Collection numbers given below are those of the first coauthor.

Figure 1. Fallen log covered with bryophyte mats.

Examples of the substrates mentioned above observed to have a bryophyte mat present were examined and a portion of the mat was collected. Each of the 15 samples collected consisted of enough material to prepare three moist chamber cultures, and all samples were returned to the laboratory for processing.

In the laboratory, the mat was divided into portions that would fit into a 90 mm plastic disposable Petri dish. Before being placed in a dish, any portion of the substrate (e.g., bark, decayed wood or soil) was removed to the extent possible. Once prepared, enough distilled water was added to each dish to cover most of the bryophytes. After approximately 24 hours, the pH of each dish was determined and recorded and then most of the water was poured off.

What was now a moist chamber culture was set aside out of direct sunlight and then examined with the aid of a dissecting microscope at weekly internals over a period of three months. Water was added as necessary to maintain moist conditions in the cultures. Any fruiting bodies of myxomycetes appearing in these cultures were collected or recorded. These were placed in small pasteboard boxes for permanent storage. Identifications were made with standard references for myxomycetes (e.g., Martin and Alexopoulos 1969). Nomenclature follows Lado (2005-2021).

Results

Twenty-two of the 45 (49%) moist chamber cultures yielded evidence (plasmodial or fruiting bodies) of myxomycetes. Fruiting bodies appeared in 15 cultures (33%) and only plasmodia were recorded from seven cultures (16%). Six species of myxomycetes were recorded. These were *Arcyria cinerea* (Bull.) Pers. (six specimens, including 34442 and 34446), *Licea operculata* (Wingdate) G. W. Martin (four

specimens, including 34440 and 34441), *Trichia subfusca* Rex (three specimens, including 34447), *Diderma effusum* (three specimens, including 34581 and 34606), *Cribraria microcarpa* (one specimen, 34588), and *Perichaena chrysosperma* (one specimen, 34581). All fruitings were small, consisting of one to a few fruiting bodies. Most appeared within the first two months after the cultures were prepared, with *L. operculata* the first species recorded.

Values of pH determined for the 44 cultures ranged from 4.4 to 8.3, with a mean value of 7.5. The latter value is higher than most substrates in the general study area for which the pH is known (Stephenson 1989).

Discussion

Bryophytes are not listed as the typical substrate for any the six species recorded in the present study, based on information in Martin and Alexopoulos (1969). The substrate for *Arcyria cinerea* is listed as "Dead wood, plant debris or the dung of herbivorous animals," for *Licea operculata* as "Bark, less common on leaves," for *Trichia subfusca* as "Dead wood," for *Perichaena chrysosperma* as "Dead wood, especially the inner bark of fallen branches; less commonly on leaves and dung of herbivorous animals," for *Cribraria microcarpa* as "Dead wood," and for *Diderma effusum* as "Dead leaves and stems of herbaceous plants; sometime wood or dung." However, *A. cinerea* has one of the broadest niches of any myxomycete and has been recorded from a wide range of substrates. As such, its occurrence on mosses would not be unexpected. There are a few records of *Diderma effusum* and *Trichia subfusca* on mosses, but *Cribraria microcarpa*, *Licea operculata*, and *Perichaena chrysosperma* have rarely been reported on this substrate (MyCoPortal 2021).

The fact that bryophytes produce antibacterial substances has long been known (McCleary et al. 1960). Kang et al. (2007) investigated the antibacterial properties of ten species of bryophytes, and nine of these showed antibacterial activities against Gram (+) bacteria. Some mosses such as *Hylocomium splendens* displayed a higher level of activity than most other mosses. The trophic stages of myxomycetes, in their natural habitats, feed mostly upon bacteria. In a bryophyte mat, it seems possible that the antibacterial properties of the bryophytes making up the mat could reduce the population levels of the bacteria present. If this was the case, the mat would not represent an especially suitable microhabitat for myxomycetes. Circumstantial evidence in support of such a hypothesis would include the relatively low species richness of the assemblage of myxomycetes associated with bryophyte mats, the fact that fruitings of myxomycetes were uniformly small, and *Arcyria cinerea* was the only species characterized by the production of what might be considered a relatively large fruiting body.

In summary, the data obtained in the present study indicate that bryophyte mats are a microhabitat for myxomycetes in temperate deciduous forests, although both species richness and abundance of these organisms are relatively low. Nevertheless, the fact that 49% of all moist chamber cultures prepared with samples of bryophytes yielded evidence of myxomycetes suggests that bryophytes are consistently associated with a microhabitat that represents a component of forest ecosystems that is not usually considered in surveys for this group of organisms.

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