Morel Mushrooms, Macrofungi, Corticolous Myxomycetes, And American Elm Trees

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Abstract:

Ulmus americana (American elm) occurs in Texas and mostly has escaped the pathogenic fungus Ophiostoma ulmi and O. novo-ulmi, the causative agents of Dutch elm disease. The American elm is a beautiful ornamental shade tree that was planted in cities and towns throughout the Midwest as a monoculture, facilitating the spread of Dutch elm disease by the elm bark beetle insect vector and by root grafts. The association between dying elm trees and morel mushrooms is described from Texas to Iowa and Ohio. Described here is the early history of moist chamber cultures using American elm tree trunk bark collected in the 1930s at

the University of Iowa. American elm cultures recently led to the discovery of new species of Mycena, corticioid crust fungi represented by species of Dendrothele, and myxomycete life cycle stages. Many corticolous myxomycete species in the genus Licea are small in the 100-200 µm range, easily overlooked in moist chamber bark cultures from living trees, and often are species new to science, for example, Licea iridescens recently described. Photographic images of rare and undescribed tiny myxomycete species are shown using multi-focus imaging and computerized stacking giving greater depth of field and three-dimensional detail. The importance of myxomycete stalks is discussed especially in regard to internal

structure and external slime sheaths.

Key Words:

Licea species, moist chamber bark cultures, *Morchella* species, *Ophiostoma ulmi*, slime molds, *Ulmus americana*.

Introduction

The demise of American elm trees begins in my hometown Peabody, Kansas in the 1950s, and continued into the 1960s and 70s at the University of Iowa. Although the pathogen for Dutch elm disease had been around long before then, the devastation and removal of American elm trees was visually most dramatic in Iowa City when I was a Ph.D. student studying myxomycetes with Professor George W. Martin at the University of Iowa.



A merican elm were widely seen on the campus and lined the streets in Iowa City. More than 2,000 American elms once grew on the University of Iowa campus but were eventually ravaged by the Dutch elm fungal pathogen so that only two survived. Apparently one tree that survived was located on the campus Pentacrest. It was a towering giant estimated to be about 15 feet in circumference, 93 feet in height, and supposedly planted in the 1880s, and recognized as a state champion tree (Snee, 2012).

myxomycetes were abundant, especially *Perichaena chrysosperma*, *P. depressa*, and *Licea biforis* on the underside of the bark separating from the woody tree trunk. This was a massive fruiting of *Licea biforis* represented by thousands of date-shaped, orangish sporangia with their distinctive longitudinal slit-like dehiscence and bright yellow spore mass. (Fig. 2).

American elms and morel mushrooms

Morel hunters in Iowa, Kansas, Missouri, and Nebraska (especially in

Fig. 2. Photo by Edward D. Forrester. *Licea biforis* sessile sporangia.



American elm were most often planted as a monoculture close together as they lined both street sides in Iowa City. The mycology laboratory in the Department of Botany at University of Iowa was the testing center for isolating and confirming infections of *Ophiostoma ulmi* for American elm in Iowa City (1967–1971). When one tree became infected with *O. ulmi* (then referred to as *Ceratiocystis ulmi*), all nearby trees also became infected through root grafts, so that every tree on the block had to be cut down.

A group of dead elm trees was found near Hawkeye Drive Apartments where married students and our family lived on the outskirts of Iowa City. These dead trees were still standing; their outer layers of bark were separating from the wood or had already fallen to the ground (Fig. 1). Morels were never observed around these trees but the river bottoms of western Missouri and surrounding bluffs of eastern Kansas) will find helpful tips for where and when to locate and how to collect morel mushrooms in Thompson (1994). Morels are early spring mushrooms in Texas and occur from early March in the Hill Country around Austin northward to the Dallas-Fort Worth area. In Texas, cedar groves of mostly mountain cedar (Juniperus ashei), and less often eastern red cedar (Juniperus virginiana), occur in areas that yield productive fruiting bodies of the "yellow morel" (Morchella americana), formerly called M. esculenta (see Bunyard, 2013, for discussion of name changes). An interesting report of morels (Morchella americana) was described for the first time at River Legacy Parks in Arlington, Texas, March 21, 1998, growing near a cedar elm (Ulmus crassifolia). A few days later in the same location, the giant "thickfooted morel" (*Morchella* "*crassipes*") was found (12–14 inches in height with a stalk 5 inches in length and 2.4 inches in width). This "species" is now considered simply an older developmental phase of *M. americana* and not a distinct species (Keller, 1998).

Morels assigned to *Morchella americana* have been found by David Lewis, March 12, 2005 in Edwards County near Barksdale as part of the famous Edwards Plateau and Hill Country of Texas. This is rugged terrain of small ravines and canyons with scattered mixed juniper-oak woods. Another site farther north at Lake Whitney in Bosque County abundant *Morchella americana* fruit bodies were collected on March 24, 2001 in oak-juniper woods. March is the best collecting time in South-Central to North Texas for morel mushrooms.

Midwestern morel collection sites are productive in early March beginning in Texas and continuing northward until early June in northern Minnesota. April and May are the prime times to collect morels in Kansas, Missouri, Iowa, and Ohio, states where I (HWK) have collected morels.

Collection times depend upon adequate soil moisture with temperatures between 50–55F and when day and night-time temperatures vary between 55-70F. This usually means about the time red bud trees (Cercis canadensis) and lilac shrubs (Syringa *vulgaris*) start to bloom and ground plants like mayapples (Podophyllum *peltatum*) cover the forest floor. The tree canopy is more open with sunlight reaching ground sites because the deciduous trees have not yet leafed out. Warmer earlier spring temperatures may also mean earlier morel mushrooms. Old abandoned apple orchards (Malus sp.) with some dying trees are also productive sites for morels. However, the spectacular "bushels of fruiting bodies" of Morchella americana that appear in season around dying American elm trees were the best habitats according to Thompson (1994).

In the 1950s and 1960s American elm trees were dying by the thousands of Dutch elm disease in cities and towns throughout the Midwestern states and this created ideal habitats for morel mushroom hunters who flocked to their favorite sites to find morels. However, as



Thompson notes, American elms were most productive in the first year of dying and even the second year after they died, but once the bark had sloughed off, the morels no longer were found (Fig. 3). Thompson (1994) describes collecting 1,500 pounds a day in the 1970s. Unfortunately, stands of American elm today are hard to find, although scattered trees in some areas are still productive. Thompson's continuous field observations of the changing color phases of grey to yellow Morchella americana suggested these were color phases of the same species that was later confirmed by genomic studies (Fig. 4). Fewer American elms resulted in also locating dying cottonwood trees and stumps as another source of morels. Thompson was often collecting in state parks, and at that time it was legal, but today in many areas it is illegal, so make sure you follow state and federal laws.

Willie May (WM) from Missouri has been collecting morel mushrooms since 1991 (Fig. 5). Every spring in the months of April and May he forays in Missouri, Illinois, Iowa and Wisconsin, with his first priority spotting groves of dying elms. Healthy live American elm trees are not usually the best source of morel mushrooms. Indeed, if the tree begins dying in the fall, usually the next spring morels are not found. Typically, American elm trees should be completely dead before morels



Fig. 4. Photo by Walt Sturgeon, Morchella americana near dead American elm tree, May 4, 2015, Columbiana County, Ohio.





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are induced to develop and then are limited to only a couple of years (WM, personal observation).

The two best tree species in the Midwest associated with morel mushroom occurrences are American elms, Ulmus americana, (probably the most productive and most frequent source of morels) and white ash, Fraxinus americana, (probably the second best tree, Fig. 6) and other species in both genera of these trees. Ash trees are dying by the millions from emerald ash borer beetle, however, dying or dead ash trees apparently do not create morel mushroom collecting opportunities. Unfortunately, the fate of ash trees appears similar to that of American elms with massive tree die-offs except in isolated habitats. Some habitats may not have these tree species so look for alternative tree species such as tulip tree (Liriodendron *tulipifera*); cottonwoods (*Populus*) spp.), oaks (Quercus spp.), sycamores (Platanus occidentalis), black locusts (Robinia pseudoacacia), black cherry (Prunus serotina), among others. Although these aforementioned morel collecting sites more frequently are the sources of morels, they also may appear unpredictably in unexpected places (Hemmes and Stallman, 2020).

Morel mushrooms are not the only fungi associated with elm trees. This spring be on the lookout for other macrofungi—some fruiting directly from the bark of elms. And if you want to better your chances at seeing some fungi (and slime molds), why not try to culture them indoors. Just bring in some bark from live trees, set up in a moist chamber, and watch for fruitings.

Previous studies on tree canopy corticolous macrofungi

Macrofungi are known to fruit from the bark of many kinds of trees. Previously, three fleshy macrofungi represented by Lentaria byssiseda, Mycetinis opacus, and Mycena supina were found on the trunk bark or lateral branches of living trees in the Great Smoky Mountains National Park (GSMNP) (Keller, 2004; Keller et al., 2009). More than 500 individual trees that included 52 taxa were climbed using the double rope climbing method and sampled for myxomycetes and macrofungi (Kilgore et al., 2008). This occurred each summer over an eight-year-period (2000-2008). The basidiomycete macrofungi observed and collected certainly was a scanty harvest from large healthy living trees in a temperate and old growth deciduous forest.

Fleshy macrofungi appear to be rare on tree trunks and in the upper canopy (Keller et al., 2009). In addition, the recent discovery of a novel species *Mycena ulmi* from an American elm tree in the Fort Worth Botanic Garden is another example of an apparent cryptic hidden species (Perry et al., 2020). At least six field trips failed to detect any basidiomes of *Mycena ulmi* on the trunk bark surface but moist chamber cultures yielded 12 collections all developed in tree bark moist chamber cultures. Some field trips were made to the American elm tree site following rainy periods and cloudy weather of several days but mycelium or basidiomes were not observed. This suggests that the discovery of fleshy macrofungi on the trunk bark of living trees may be a potential source of hidden biodiversity given the use of moist chamber cultures.

Another example of macrofungi on living healthy trees is the genus *Dendrothele* referred to commonly as white crust corticioid fungi. An ongoing study of these fungi in nature parks, Fort Worth residential areas, at the Botanical Research Institute of Texas, Fort Worth Botanic Garden in Fort Worth and in River Legacy Park, Arlington, Texas, Tarrant County, has yielded more than 100 collections of mostly Dendrothele jacobi on American elms (27 trees) and cedar elms (32 trees) (Bordelon et al., 2020). Cedar elms predominate on the eastern side of the BRIT campus near University Drive (15 trees) with *Dendrothele jacobi*. Healthy Juniperus virginiana (eastern red cedar) predominate (17 trees) at Tandy Hills Natural Area in Fort Worth with Dendrothele nivosa specifically associated with these trees (Fig. 7).



Cortoicolous myxomycetes and American elm trees

Gilbert and Martin (1933) generally are given credit for introducing the moist chamber culture technique using bark from living trees. Many tiny myxomycete fruiting bodies were discovered on wetted bark samples in glass Petri dishes taken from different living tree species, including American elm, referred to as "elm" in their paper. Gilbert was a student of G. W. Martin working in the Mycology Laboratory at the University of Iowa (Keller, 2012). Moist chamber bark cultures from living trees yielded 30 myxomycete species; however, in some cases, the source of the tree species was not given (Gilbert and Martin, 1933). Noteworthy observations included the fast-forming fruiting bodies, some on the second or third day after wetting and others after five or six days. Myxomycete species cultivated from elm bark were *Comatricha laxa*, *C. fimbriata, Enerthenema papillatum,* Clastoderma debaryanum var. emperitorium, Cribraria minutissima, C. violacea, Licea biforis, L. tenera, *Hymenobolina parasitica* (currently as *Licea*), *Margarita metallica* (currently as Calomyxa), Ophiotheca wrightii (currently as Perichaena chrysosperma), Arcyria cinerea, and Hemitrichia minor. Echinostelium minutum is listed as common on frondose and coniferous trees but no specific tree was named. These species are considered corticolous myxomycetes that develop, grow and form fruiting bodies on the bark surface of living trees and woody vines (Keller and Braun, 1969; Everhart and Keller, 2008; Keller et al., 2008; Keller et.al., 2009). Most of these corticolous myxomycetes have been collected in the field on bark of living trees especially Juniperus virginiana and Ulmus americana (Keller and Braun, 1999; Keller et.al, 2009); Parker and Keller, 2003).

The majority of myxomycete fruiting bodies develop along the bark edges "edge effect" and in deeper crevices and pockets where these areas stay wetter longer and dry out more slowly than on the bark surface. Corticolous myxomycete species diversity appears highest in rank order: eastern red cedar, American elm, and red maple (*Acer rubrum*).

In another paper, Gilbert (1934) described three corticolous myxomycete species new to science: *Macbrideola scintillans* and *M. decapillata* (*Macbrideola* is a commemorative generic name for Thomas H. Macbride, a world famous myxomycetologist and former President of the University of Iowa) and *Hymenobolina pedicellata* (currently in the genus *Licea*). All three species form tiny plasmodia, each producing one tiny stalked sporangium on tree trunk bark of living trees.

Corticolous myxomycete species diversity on bark of living trees

A series of papers documented the occurrence of myxomycete fruiting bodies and other life forms, including

Order Echinosteliales	Clastoderma debaryanum var. emperatorium+, C. microcarpum*, Echinostelium arboreum+, E. coelocephalum+, E. minutum+
Order Liceales	Cribraria minutissima+, C. violacea+, Licea biforis*+, L. denudescens*+, L. inconspicua*, L. iridescens+, L. kleistobolus*+, L. marginata*, L. nannengae+, L. parasitica*+, L. pedicellata+, L. perexigua+, L. scyphoides*+, L. a spiny-spored undescribed new species+; L. an orange- operculate new species+
Order Physarales	Badhamia affinis*, Badhamiopsis ainoae*+, Diderma chondrioderma*, D. corrugatum*, Didymium clavus*+, D. orthonemata*, Physarum crateriforme*+, Trabrooksia applanata*
Order Stemonitales	Comatricha fimbriata+, C. laxa*+, Enerthenema papillatum+, Macbrideola cornea+, M. decapillata+, M. martinii+, M. scintillans+
Order Trichiales	Arcyria cinerea*+, Calomyxa metallica*+, Dianema (clustered spores) +, Minakatella longifila*, Perichaena chrysosperma*+, P. depressa*+, P. minor+, 42 total species

TABLE 1. Myxomycete species list from American elm trees (taxa arranged alphabetically by genus and species). Corticolous myxomycete species listed from multiple living American elm trees based on field collections (*) and moist chamber bark cultures (+) arranged by taxonomic orders and alphabetically by genus and species (Gilbert and Martin, 1933; Gilbert, 1934; Martin and Alexopoulos, 1969; Keller and Braun, 1999; and HWK collection books). These collections were compiled over a period of about 70 years, and along with those from eastern red cedar, represent the two species of trees where the majority of corticolous myxomycete species occur (Keller et al., 2009).

ferns, fungi, lichens, liverworts, mosses, myxobacteria, and tardigrades, on the trunk bark of living trees and woody vines high in the tree canopy in GSMNP (Snell and Keller, 2003; Snell et al., 2003; Keller et al., 2003, Davison and Keller, 2004; Keller, 2004; Keller et al., 2004; Keller, 2005; Keller, et al., 2009; Kilgore et al., 2008; Keller and Barfield, 2017). Elm trees were not included in these tree canopy studies because they were not present in sufficient numbers and size.

Canopy corticolous myxomycete species diversity on living trees usually followed a higher pH pattern nearer 7.0 to a lower pH of 4.0 and were rank ordered by number of species as follows: eastern red cedar 50, red maple 49, white oak 41, tulip tree 39, white ash 31, white pine 24 (Snell and Keller, 2003; Keller, 2004; Keller et al., 2009). These same tree species coincidentally also may be associated with morels. American elms also had approximately 42 species of corticolous myxomycetes (see Table 1) and ranked in the top four tree species. Many of the smaller myxomycete species in the genera *Echinostelium* and *Licea* are found on the trunk bark of living trees (Keller and Brooks, 1976; Keller and Brooks, 1977; Keller and Braun, 1999; Marshall and Keller, 2018; Keller and Marshall, 2019).

Methodology

Field collections and sampling of Ulmus americana *trees*

American elm trees selected for this study were the largest at each of the study sites; minimum trunk diameter of 60 cm at dbh, diameter at breast height=1.5 meters) and total height (18 to 30 meters). Nine American elm trees were sampled at Oliver Nature Park, two at the Fort Worth Botanic Garden, and five at the Fort Worth Nature Center and Refuge for a total of 16 trees. *Dendrothele* specimens were made on giant American elm trees in residential areas of Fort Worth, from area nature parks, and the FWBG campus.

A heavy bladed knife was used to pry tree bark samples avoiding any damage to underlying tissues. Pieces of bark were collected from all sides of the trunk at approximately 1.5 to 1.8 meters until enough samples per tree were gathered to half-fill a paper bag (ca 1000 cm³). Every bag was labeled with the identifying tree number, collection date, and site location. Most bark samples were collected during the summer months of June, July, and August and were placed in moist chamber cultures within two weeks. Field collections of *Dendrothele* were made throughout the year whenever weather conditions permitted.

Moist chamber bark cultures

Moist chamber culture preparation techniques were described in part previously (Keller et al., 2008; Keller and Marshall, 2019; Kilgore et al., 2009; Perry et al., 2020; Scarborough et al., 2009). Consult other articles that describe inexpensive ways to prepare moist chamber tree bark cultures (Keller et al., 2008). This study followed a moist chamber culture protocol described herein. Two moist chamber cultures per tree were prepared using the American elm trunk bark samples to document the corticolous myxomycetes present. Each sterile, plastic Petri dish (150 x 25 mm) was lined with one sterilized P8-creped filter paper fitted to cover the bottom. Bark samples were randomly selected from a paper bag and placed in the Petri dish bark outer-side up so that the bark pieces were close, but not overlapping or touching. Each Petri dish was labeled with the tree number, collection site, and date wetted on the lid and bottom side. 30 mL of pH 7 deionized sterile water was added around the bark to ensure thorough and even wetting. After 24 hours any excess water was decanted. Cultures were placed near a window exposed to indirect natural light and ambient room light and remained in the same spot unless they were being examined under a dissecting microscope. Incubation occurred with light and dark cycles of June to July

while remaining at room temperature at approximately 22–25C. On day two and day six after wetting and every two to four days thereafter for four weeks the plates were examined under a Nikon dissecting microscope with a MKII Fiber Optic light source at magnifications of 50x to 150x. Each dish was examined systematically from side to side after the lid was removed to ensure that each piece of bark was examined. However, removing the lid was minimized to avoid exposure due to aerial contamination, especially by *Trichoderma*, a commonly encountered green mold.

Location and preservation of myxomycete fruiting bodies

Moist chamber cultures of tree bark often have tiny scattered sporangia of Echinostelium and Licea species (less than 0.1 mm). Their size and scattered habit make it difficult to find these sporangia, therefore to save time, insect pins (size 1 with nylon head stainless steel needle, 40 mm long and 0.35 in diameter) and (silver flat-head 1-inch needles) were used to pin point fruiting body locations. When moist chambers were re-examined pins facilitate making microscope slides for identifications. However, these pins may become dislodged when preparing permanent collections and type specimens on dried tree bark glued in boxes. Pins also may interfere with photographic techniques and must be removed when orienting the specimen under the microscope.

The presence of myxomycete fruiting bodies on dried tree bark were pinpointed using a white paint permanent waterproof liquid opaque marker with an extra fine (0.7 mm) tip. A white dot was placed equidistant on either side of the fruiting body and close enough in a straight line so that both the white dots and specimen are in the field of view simultaneously. This technique should prove valuable in marking the location of myxomycete fruiting bodies in type collections especially when their presence is obscured by look-alike life forms. All observations of plasmodia and fruiting bodies of date and time were recorded in a notebook. Identifications were made using dichotomous keys and illustrations in Martin and Alexopoulos (1969). Pieces of dried bark with fruiting bodies of myxomycete sporangia were removed and glued to the bottom of

a standard collecting box 4.5 x 10.5 x 2 cm. Labels included species name. state, county, collection locality, habitat, UTM coordinates, bark collection date, wet date, harvest date, collector's name (legit) and accession number, person identifier (fide) were affixed to the box top. A Nikon Alphaphot light compound microscope was used to examine slides and make measurements using a calibrated ocular reticule to identify myxomycete species.

Microscope glass slides (75 mm x 25 mm x 1 mm thick) with a frosted end were used to make mounts of myxomycete structures for identification. Small pins attached to wooden handles were used to remove fruiting bodies from the bark. A droplet of clear lactophenol was placed in the center of the microscope slide, sporangia were added, and a square cover slip (22 x 22 mm and 0.16 mm thick) was gradually lowered at an inclined angle over the specimen to avoid creating air bubbles. Permanent slides were made for future use by sealing the edge of the cover slip with ZUT a resiniferous slideringing compound. Identification labels were written in lead pencil on the frosted end of the slide covered with clear tape to prevent future smudging. Slides were wrapped with lens paper and placed in properly labeled collection boxes.

Photography using light and scanning electron microscopes

An Olympus BH2 microscope with EPI Illumination and ultra-long working distance objectives of 5X, 10X and 20X in combination with 10X eyepieces was used to observe myxomycete sporangia. Photographic images were made with a Sony A6000 and a NFK 2.5X relay lens at 10 mega-pixel settings. Multiple image focal stacking was used for both microscopic slides and habit photographs. Each image stack varied between 10 to 75 individual images merged using Helicon Focus software version 6.8.0. Image stacking increased depth of field so that the entire sporangium (stalk and spore case) were in sharp focus. Images were optimized using Adobe Photoshop CC 19.0.

SEM specimens were mounted on 12.5 mm stubs with conductive tape by excising one of the sporangia including a small amount of the substrate then mounted on a piece of double stick



Fig. 8. Photo by Harold W. Keller, labels by Courtney M. Kilgore.

tape which is attached to a small (1x2") card stock. The card was inverted and the sporangia pressed to the top, tapped, removed and the procedure repeated until the desired amount of the specimen had adhered to the stub. A small paint brush was used to distribute spores across the stub adhesive surface. The stub was coated with gold using a Hummer VII sputtering machine to a thickness of 3 nm. A new genomics facility at BRIT includes a Hitachi SU 3500 high resolution SEM which was used to observe and photograph specimens. Images were recorded in a reduced contrast mode and processed in Photoshop to retain highlight information and extract the maximum detail available.

Importance of myxomycete fruiting bodies

More information concerning the importance of myxomycete fruiting bodies can be found in Keller and Everhart (2010). This article published in FUNGI has been cited 2,933 times from June 2016-March 2021 as posted on the Univ. of Nebraska Digital Commons. The morphological characters used in monograph species descriptions and keys to identify myxomycete species are associated with the fruiting bodies (Martin and Alexopoulos, 1969). The presence or absence of a peridium, capillitium, surface net, columella, calcareous structures, and stalks serve to distinguish the orders, families, genera, and species (Fig. 8). This labeled stalked

sporangium of *Macbrideola declinata* illustrates the structural parts when it was described as a species new to science by Brooks and Keller (Eliasson et al., 1988). The ornamentation and color of the spores, provides additional morphological characters, sometimes only seen with the higher magnification 1,000x under oil immersion objective of a light microscope or with the scanning electron microscope.

Moreover, one example of missing morphological characters in myxomycete species descriptions is the presence of an external stalk sheath (Figs. 9a and 9b) and internal presence or absence of stalk fibers. These morphological stalk characters are helpful when assigning taxa to genera such as Comatricha, Lamproderma (Meriderma), Macbrideola, and Stemonitis. Comatricha and Lamproderma have fibrous stalks characterized by an interlaced network of mostly dark fibers as in Comatricha laxa when transmitted light passes through the stalk (Fig. 9c) and when the stalk is embedded, sectioned, and stained with methylene blue (Fig. 9d). These fibers



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Fig. 9a. Photo by Edward D. Forrester. *Comatricha species* with slime sheath at base.



Fig. 9b. Photo by Edward D. Forrester. *Lamproderma sauteri* stalked sporangium with plasmodial slime sheath at base.

are sometimes difficult to see because the stalk is too opaque and impossible to discern their presence. However, if the stalk is broken near the base the fibers can be seen often protruding at the break as in figure for *Lamproderma cribrarioides* (Fig. 9e). *Stemonitis* species have stalks that are hollow (Fig. 9f). One note: the preferred term used here for myxomycete fruiting bodies is stalk and the preferred term for fleshy fungi is stipe.

Observations of selected corticolous myxomycetes, arranged alphabetically by genus and species

Carlos Lado (2021) maintains a running updated total of myxomycete taxa as part of an online nomenclatural



Fig. 9c. *Comatricha laxa*, note internal strands of fibers.



Fig. 9d. *Comatricha laxa*, note upper portion of sporangium with spores and stalk fibers stained blue.



Fig. 9e. Photo by Edward D. Forrester. Lamproderma cribrarioides broken stalk at base with protruding fibers and sheath along edge.

information system of Eumycetozoa (myxomycetes). The most recent data for myxomycete taxa is as follows: the number of accepted species of myxomycetes (not subspecies, varieties



Fig. 9f. Cross section of *Stemonitis* hollow stalk (center) stained with methylene blue.



Fig. 9g. Macbrideola scintillans.



Fig. 10. Photo by Edward D. Forrester. *Echinostelium arboreum* stalked sporangium.

or forms) is about 1,100 species mainly because the figure has grown in recent years. The actual number varies between 1060–1095 as some species are under review due to their validity.

Echinostelium arboreum is a rare corticolous myxomycete that represents a new record for the state of Texas (Fig. 10). Currently there are 18 valid *Echinostelium* species recorded by Lado





(2021), however, *E. arboreum* is the only species with a persistent shiny peridium. Although the stalked sporangia are small (100–120 μ m total height, 40–50 μ m in diameter) the combination of a shiny persistent peridium and golden color facilitate locating and identifying this species with a dissecting microscope (Keller and Brooks, 1976). SEM photographic images that show

morphological details of this species are illustrated by Moreno et al. (2001). This species was found multiple times on *Ulmus americana* trunk bark in moist chamber cultures at Oliver Nature Park in Mansfield, Tarrant County.

Licea biforis is a common species with a distinctive fusiform or date shape found on the trunk bark of living or standing dead trees especially *Ulmus* *americana* and *Malus* apple trees (Keller & Braun, 1999). The sessile sporangia or short plasmodiocarps have a longitudinal slit that opens like a clam exposing the bright yellow spore mass as in Fig. 2. The fruiting bodies are often present in great numbers which facilitates location and easy identification. Although the color is variable, the orangish color shown here is more typical.





Fig. 13. Photo by Edward D. Forrester. Crystals with yellow bands.



Licea iridescens is a species recently new to science that developed in moist chamber cultures with spectacular iridescent sporangia often associated with crystals of unknown origin and irregular shapes (Fig. 12) (Keller and Marshall, 2019). The sporangial sides sparkle with bluish, pinkish, and reddish colors when the peridium is highlighted by an external incident directed optical light source illuminating the surface. A dark apical patch of debris forms first in early developmental stages. Crystal fields on the tree trunk bark cover the bark surface brightly shining with an optical light source (Fig. 13). These crystals vary in size, are irregular in shape, and are often associated with Licea *iridescens* and apparently never have been described before in the published literature. These crystals were newly discovered on numerous American elm trunk tree bark surfaces and are part of an ongoing study to determine their composition, origin, and function. Collections: holotype: BRIT 478988.

Licea pedicellata has stalked black sporangia that occur abundantly on the trunk bark of living *Ulmus americana* and *Juniperus virginiana* trees. It is



possible to observe this species in the field with a 20X widefield hand lens. because of the larger size (total height 200–500 μm and 75–175 μm in diameter). Species descriptions vary with reference to the peridium and dehiscence patterns. The photograph shown here fits the species description in Keller and Braun 1999: "... the spore case often dries and the wrinkled peridial surface gives the appearance of plates and sutures." This photograph (Fig. 14) shows a smooth peridial outer surface, a furrowed stalk typical of most fresh mature specimens in moist chambers, and a crystal at the stalk base. Another study of stalked Liceas by Wrigley de Basanta and Lado (2005) has SEMs of this species but unfortunately does not confirm everything in their species description "...dehiscence into irregular platelets 20-30 µm wide ... "The dried sporangia displayed with SEM do not show this dehiscence pattern.

Licea pseudoconica is a distinctive and diminutive species (70–120 µm in height and 55–90 μ m in diameter) that has a prominent whitish cone of gelatinous debris apically giving the general appearance of a snow-capped mountain. It was described as a species new to science by Keller and Brooks (1977) but also discussed from collections made in Florida (Keller 1973). Abundant sessile sporangia often occur along bark edges. This edge affect facilitates locating and identifying sporangia highlighting the conical shape in profile. Developing early immature sporangial stages clearly show the apical cone of debris as a transparent mass separate from the underlying

spores (Fig. 15). This cone-shaped mass develops first before the spores form. In addition, the spores appear to develop first in the apical region. The mature sporefilled sporangial body proper is more spherical. These moist chamber trunk bark collections of Licea pseduoconica on Ulmus americana represent new records for the state of Texas.

Licea perexigua is a small sessile to short stalked corticolous myxomycete which is more difficult to identify because the spherical sporangium $(40-110 \ \mu m \ in \ diameter)$ resembles other objects on the tree bark surface. This species is a new record for the state of Texas.

Licea undescribed new species. This rare taxon was found at Oliver Nature Park, Tarrant County, Texas on the trunk bark surface of a living Ulmus americana tree in moist chamber cultures (Keller and Marshall 2019). The genus Licea has about 75 valid species (Lado, 2021). This taxon has one morphological character not found in any other species of myxomycete: a transparent, single, thin, colorless, peridium lacking ornamentation (Fig. 16). In addition, the black spores have a type of ornamentation consisting of long spines and a spore wall of uniform thickness not found in any other species in the genus Licea (Fig. 17). The sessile sporangia are difficult to detect and select because of their tiny size and their association with Nostoc ball developmental stages (a cyanophycean alga) that can be confused with sporangia. Light and SEM photographic images clearly highlight these morphological characters.

Licea undescribed new species. This taxon was rare with only a few sessile sporangia which were distinct because of the burnt orange color with a recessed operculum with raised edges and a silvery surface. The circular lid actually popped off on several sporangia exposing the yellow spores underneath. More specimens are needed before this

taxon can be described as a species new to science (Fig. 11).

Macbrideola cornea is common and widely distributed wherever *Ulmus americana* and *Juniperus virginiana* trees are found, especially in the latter case, when these trees are planted in cemeteries throughout the Midwest and Southeast. This is a new record for Tarrant County, Texas. The stalked sporangia (0.6–2.5 mm in total height and 0.12–0.3 mm in diameter) are widely scattered, usually solitary, with branching capillitial threads arising from the apex of the columella.

Macbrideola scintillans is a new record for Tarrant County, Texas but is apparently widely distributed throughout the world. It appears widely scattered and usually solitary in moist chamber trunk bark cultures appearing most frequently on Ulmus americana and Juniperus virginiana living trees in North America. A detailed species description, LMs, SEMs, and photographic illustrations highlight this species in Eliasson et al. (1988). In addition, Keller and Braun (1999) discuss this species with excerpts quoted here: "The sporangia are 50–150 μm in diameter and 125–300 μ m in total height. The silvery, persistent peridium looks very much like tinfoil and usually remains attached to the apex of the columella." The LM photographic image shown here (Fig. 9g) has a portion of the hollow stalk as a white slit in optical section. Furthermore, air bubbles are also sometimes trapped in the stalk providing additional evidence that

the stalk is hollow. The thin peridium remains as a conspicuous collar at the juncture of the columella and stalk.

Epilogue

American elms are more than beautiful ornamental shade trees. Joyce Kilmer's poem "Trees" (1913) reminds us how special a tree is in part because a nest of robins may live there. But American elm trees are far more than ornamental objects, they are part of our human experience as well as many other hidden life forms that live there. Urban trees improve the streetscape in many ways including: visual appeal and brilliant autumn golden-yellow fall leaf colors for American elms; improve air quality by absorbing CO₂, deplete ozone, and reduce particulate matter and noise abatement; provide shade from solar radiation lowering energy consumption in residential areas; serve as home and shelter for wildlife including nesting and roosting birds, squirrels, and other fauna; reduce storm water runoff and protection from wind; increase property real estate values; these are some of the value-added benefits of trees. How many of you remember the tree in your yard that you climbed during your youth or built a tree house or played hide and seek? One of the side effects of dead or dying elms in rural environments is the appearance of the highly prized morel mushroom in the spring months considered a delicacy by mycophiles.

Acknowledgments

All photos by the author, unless

otherwise noted. SEMs presented in this publication were generated with support from the George C. and Sue W. Sumner Molecular and Structural Laboratory at the Botanical Research Institute of Texas. Bob O'Kennon assisted with the collection of American elm bark. Vanessa Marshall prepared moist chamber bark cultures, made permanent slides, and boxed and labeled specimens. Edward D. Forrester took LM photographs and assisted with measurement of sporangia. Billy G. Stone assisted with microscopic slide photography and SEM preparation and photographs. Karen Nakasone identified all of the Dendrothele specimens. The author thanks the University of Florida Graduate School for a Postdoctoral Fellowship that supported his research on myxomycete stalks. The author thanks W. May and B. Bunyard for much valuable morel information and tips. This paper is dedicated to Professor Dr. Thomas W. Gaither, who was a Ph.D. student with HWK at the University of Iowa and co-author of myxomycete publications, outstanding botany teacher and field botanist, and a member of the famous FRIENDSHIP 9 from Rock Hill, South Carolina.

References Cited

Bordelon, A., H.W. Keller, and R.J. O'Kennon. 2020. Rediscovery of *Dentrothele* (white crust fungus) in North Texas: the importance of urban tree preservation. Texas Plant Conservation Conference, Botanical Research Institute of Texas, Session 1, Floristics. Bunyard, B.A. 2013. Morels: the name game. FUNGI 6(1): 27–32.

Davison, P.G., and H.W. Keller. 2004. Vertical distribution of liverworts within the forest canopy in the Southern Appalachians: contributions to the All Taxa Biodiversity Inventory of the Great Smoky Mountains National Park. *Evansia* 21: 79–87.

Eliasson, U.H., H.W. Keller, and J.A. Hutchison. 1988. Myxomycetes from Arkansas. *Mycotaxon* 32: 375–398.

Everhart, S.E., and H.W. Keller. 2008. Life history strategies of corticolous myxomycetes: the life cycle, fruiting bodies, plasmodial types, and taxonomic orders. *The International Journal of Fungal Diversity* 29: 1–16.

Gilbert, H.C., and G.W. Martin. 1933. Myxomycetes found on the bark of living trees. University of Iowa Studies in Natural History, Papers on Iowa fungi IV. 15(3): 3–8.

Gilbert, H.C. 1934. Three new species of Myxomycetes. University of Iowa Studies in Natural History. Contributions from the Botanical Laboratories 16: 153–159.

Hemmes, D.E., and J. Stallman. 2020. Morels in a bird's nest. FUNGI 12(4): 41–43.

Keller, H.W. 1973. Myxomycetes from the Everglades National Park and adjacent areas, I. *The Ohio Journal of Science* 73(6): 364–369.

Keller, H.W. 1998. Morels discovered at River Legacy Parks. *Living Science Natural News* 5(3): 2.

Keller, H.W. 2004. Tree canopy biodiversity: student research experiences in Great Smoky Mountains National Park. *Systematics and Geography of Plants* 74: 47–65.

Keller, H.W. 2005. Undergraduate research field experiences: tree canopy biodiversity in Great Smoky Mountains National Park and Pertle Springs, Warrensburg, Missouri. *Council on Undergraduate Research Quarterly* 25(4): 162–168. (Invited Paper).

Keller, H.W. 2012. Myxomycete history and taxonomy: highlights from the past, present, and future. *Mycotaxon* 122: 369–387.

Keller, H.W., and K.M. Barfield. 2017. The Great Smoky Mountains National Park: The People's Park. FUNGI 10(2): 44–64.

Keller, H.W., and K.L. Braun. 1999. Myxomycetes of Ohio: their systematics, biology and use in teaching. *Ohio Biological Survey Bulletin New Series*13(2): 1–182.

Keller, H.W., and T.E. Brooks. 1976. Corticolous myxomycetes V: observations on the genus *Echinostelium*. *Mycologia* 68: 1204–1220.

Keller, H.W., and T.E. Brooks. 1977. Corticolous myxomycetes VII: contribution toward a monograph of *Licea*, five new species. *Mycologia* 69: 667–684.

Keller, H.W., P. Davison, C. Haufler, and D.B. Lesmeister. 2003. *Polypodium appalachianum*: an unusual tree canopy epiphytic fern in the Great Smoky Mountains National Park. *American Fern Journal* 93: 36–41.

Keller, H.W., S.E. Everhart, M. Skrabal, and C.M. Kilgore. 2009. Tree canopy biodiversity in temperate forests: exploring islands in the sky. *Southeastern Biology* 56(1): 52–74. Invited paper.

Keller, H.W., and S.E. Everhart. 2010. Importance of myxomycetes in biological research and teaching. FUNGI 3(1): 29–43.

Keller, H.W., C.M. Kilgore, S.E. Everhart, G.J. Carmack, C.D. Crabtree, and A.R. Scarborough. 2008. Myxomycete plasmodia and fruiting bodies: unusual occurrences and user friendly study techniques. FUNGI 1(1): 24–37.

Keller, H.W., and V.M. Marshall. 2019. A new iridescent corticolous myxomycete species (*Licea*: Liceaceae: Liceales) and crystals on American elm tree bark in Texas, U.S.A. *Journal* of the Botanical Research Institute of Texas 13(2): 367–386.

Keller, H.W., M. Skrabal, U.H. Eliasson, and T.W. Gaither. 2004. Tree canopy biodiversity in the Great Smoky Mountains National Park: ecological and developmental observations of a new myxomycete species of *Diachea*. *Mycologia* 96: 537–547.

Kilgore, C.M., H.W. Keller, and J.S. Ely. 2009. Aerial reproductive structures on vascular plants as a microhabitat for myxomycetes. *Mycologia* 101: 303–317.

Kilgore, C.M., H.W. Keller, S.E. Everhart, A.R. Scarborough, K.L. Snell, M.S. Skrabal, C. Pottorff, and J.S. Ely. 2008. Research and student experiences using the doubled rope climbing method. *Journal of the Botanical Research Institute of Texas* 2(2): 1309–1336.

- Lado, C. 2005-2020. An on line nomenclatural information system of Eumycetozoa. Real Jardín Botánico, CSIC. Madrid, Spain. http://www. nomen.eumycetozoa.com (consulted January, 2021).
- Marshall, V.M., and H.W. Keller. 2018. Myxomycetes on American elms surviving Dutch elm disease in Texas. Texas Plant Conservation Conference, Botanical Research Institute of Texas, p. 19.
- Martin, G.W., and C.J. Alexopoulos. 1969. *The Myxomycetes*. University of Iowa Press, Iowa City; 561 p.
- Moreno, G., C. Illana and M. Lizárraga. 2001. SEM studies of the Myxomycetes from the Peninsula of Baja California (Mexico), III. Additions. *Annals of Botany Fennici* 38: 225–247.
- Parker, E.E., and H.W. Keller. 2003.
 Correlation of pH with assemblages of corticolous myxomycetes in Big Oak
 Tree State Park. *Journal of the McNair Central Achievers Program*, University of Central Missouri. Vol. XII
 (Issue 1): 4–8.
- Perry, B.A., H.W. Keller, E.D. Forrester, and B.G. Stone. 2020. A new corticolous species of *Mycena* section *viscipelles* (Basidiomycota), Agaricales) from the bark of a living American elm tree in Texas, U.S.A. *Journal of the Botanical Research Institute of Texas*. 14 (2): 167–185.
- Scarborough, A.R., H.W. Keller, and J.S. Ely. 2009. Species assemblages of tree canopy myxomycetes related to pH. *Castanea* 74(2): 93–104.
- Snee, T. 2012. The last elm, the tree Dutch elm disease would leave behind. The University of Iowa, *Iowa Now*.
- Snell, K.L., and H.W. Keller. 2003. Vertical distribution and assemblages of corticolous myxomycetes on five tree species in the Great Smoky Mountains National Park. *Mycologia* 95: 565–576.
- Snell K.L., H.W. Keller, and U.H. Eliasson. 2003. Tree canopy myxomycetes and new records from ground sites in the Great Smoky Mountains National Park. *Castanea* 68: 97–108.
- Thompson, V.V. 1994. *Morel: a lifetime of pursuit.* Pamphlet published by the Missouri Mycologial Society; 36 p.
- Wrigley de Basanta, D., and C. Lado. 2005. A taxonomic evaluation of the stipitate *Licea* species. *Fungal Diversity* 20: 261–314.