Myxomycete species concepts, monotypic genera, the fossil record, and additional examples of good taxonomic practice

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Conceptos de especies en mixomicetos, géneros monotípicos, el registro fósil y ejemplos adicionales de una buena práctica taxonómica

Resumen. Se destacan y amplían algunos de los principales elementos para una buena práctica taxonómica. Se revisan los conceptos de especie en los mixomicetos, a la vez que se discuten los géneros monotípicos, con ejemplos en *Badhamiopsis ainoae*, *Protophysarum phloiogenum* y *Trabrooksia applanata*. Se sugiere que las secuencias de ADN resolverán el rango taxonómico al que los géneros monotípicos deben de asignarse en la clasificación de los mixomicetos. Se evalúa y discute por primera vez la evidencia fósil de mixomicetos encontrada en ámbar. *Perichaena brevifila, P. microspora, P. pedata* y *P. syncarpon* habitan exclusivamente en la hojarasca y son un ejemplo como las diferencias ecológicas y los patrones de estacionalidad basados en las observaciones de campo registradas en los datos de las colecciones, pueden complementar las diferencias morfológicas para la separación de las distintas especies. El futuro de la Sistemática de los mixomicetos requiere un cambio de la taxonomía descriptiva a estudios de mayor profundidad basados en hipótesis para probar relaciones filogéneticas, patrones biogeográficos y restricciones de las especies a hábitats con características ecológicas especiales.
Palabras clave: *Badhamiopsis*, ecología, *Perichaena, Protophysarum*, estacionalidad, *Trabrooksia*.

Abstract. This paper highlights and expands on some of the major points of good taxonomic practice. Myxomycete species concepts are reviewed, monotypic genera are discussed and critiqued, and case study examples are given for *Badhamiopsis ainoae, Protophysarum phloiogenum,* and *Trabrooksia applanata*. Monotypic genera are suggested for DNA sequencing to resolve the correct taxonomic rank in myxomycete classification. Fossil evidence of myxomycetes found in amber is evaluated and discussed for the first time. *Perichaena* species represented by *P. brevifila, P. microspora, P. pedata,* and *P. syncarpon,* are restricted to leaf litter habitats and serve as examples of fruiting seasonality patterns and ecological differences based on detailed field observations recorded in collection data. This additional ecological information can supplement morphological differences in distinct species. The future of myxomycete systematics requires a shift from descriptive taxonomy to in-depth studies using hypotheses that test phylogenetic relationships, biogeographical patterns of distribution, and the restriction of species to habitats with special ecological characteristics.
Key words: *Badhamiopsis*, ecology, *Perichaena, Protophysarum*, seasonality, *Trabrooksia*.

Recibido 23 de junio 2006; aceptado 20 de junio 2007.
Received 23 June 2006; accepted 20 June 2007.

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REVISTA MEXICANA DE MICOLOGÍA 27: 9-19, 2008

ORIGINAL

Introduction

The number of myxomycete species recognized by Martin and Alexopoulos [23] was roughly 422 compared to the most recent estimates of 600 by Nannenga-Bremekamp [27], and 925 of Yamamoto [37]. Schnittler and Mitchell [31] cited 1,012 subgeneric taxa of myxomycetes described as valid, including 866 at the species level, and Lado [21] noted 900 legitimate names for accepted species. Lado's taxonomic database with 1,012 taxa could be subdivided into 446 taxa estimated to be common (more than 20 collections and reported from several localities) 258 to be rare (known from 2-20 collections and more than one locality), and 305 reported only from the type locality (one or a few collections) [31]. A figure by Schnittler and Mitchell [31] of the rate of species descriptions beginning about 1965 shows a dramatically increasing number of described new taxa per year, throughout the world.

What are the reasons for this "explosion" of many new species in the last 35 years? Certainly more myxomycologists were searching in more habitats, such as the canopy of living trees [17] and different areas of the world [32]. However, the increase seems to be present during the time when Nanennega-Bremekamp and many other non-academic taxonomists were working privately as individuals, coupled with a trend for the "splitting" of genera and species, resulted in too many dubious species based on single or less than four collections from a restricted locality. Unfortunately, many single collections only known from the type locality had too few fruiting bodies and resulted in species descriptions based on limited material, inadequate for comparisons with other taxa. Furthermore, type specimens were often retained in private herbaria and papers were published in regional and not international journals that undergo a rigorous merit review process. In most cases species were illustrated with line drawings that lack the fine structure details of scanning electron microscopy (SEM). Spore-to-spore cultivation and multiple collections were lacking, complicating accurate species descriptions, especially when considering the variability and limited number of fruiting body and morphological characters. This approach to taxonomy was followed in many countries, resulting in a proliferation of species.

The involvement of non-academic taxonomists working privately as individuals is possible because many myxomycete species produce fruiting bodies visible to the naked eye and materials needed for the collection and preservation of myxomycete fruiting bodies are readily available. Furthermore, identification of myxomycetes can be learned quickly because fruiting body terminology is relatively simple. Specimens can be examined using tools such as dissecting needles, handheld blowers, and hand lenses that are inexpensive to purchase or are handcrafted [34]. Slide preparations do not require complex chemicals, tap water is sufficient. Moist chamber bark cultures are simple to prepare using materials available at commercial stores. Bark cultures yield plasmodia, plasmodial tracks, and developing fruiting bodies easily observed at 10 to 100 times magnification [19]. Myxomycetes, much like the macrofungi (mushrooms), pique the curiosity of amateur collectors that eventually result in publication of new species without academic, professional taxonomists as collaborators. Indeed, myxomycete fruiting bodies often win photographic prizes at the annual North American Mycological Society meeting because of their striking beauty [19]. In comparison, related groups of organisms, the dictyostelids and protostelids, require laboratory culture methods and equipment not available to most amateurs so these groups were studied almost exclusively by professional, academic myxomycologists. Thus, the taxonomic confusion and number of synonyms for dictyostelids and protostelids were minimized, especially in view of the fact that culture techniques are required for their isolation and observation.

Many new myxomycete species have been discovered by both amateurs and professionals since the last world monograph published in 1969 [23]. Monographic publications are necessary to assess the validity of new species but have a long preparation time, requiring specimens on loan from herbaria (although electronic databases speed this process, when available) and examination of specimens from closely related species. In addition, accurate species descriptions are based on light microscopy and SEM quality photographs that require more time.

For example, to adequately monograph the genus *Cribraria* (which has more than 40 species that are often difficult to distinguish at the species level), it would probably take five to ten years. To avoid others from publishing first, authors are prone to publish hastily, compromising good taxonomic practice. The pressure to publish quickly is related to the prestige factor associated with describing a new species and not for work to clarify species concepts and solve taxonomic problems.

The myxomycete species problem and concepts as discussed by Clark [5] suggested that taxonomists become more familiar with the potential morphological variation due to geographical restriction of apomictic clonal lines and also acquire a better understanding of population, developmental, and reproductive biology. Genetic mating systems in *Didymium iridis* (Ditmar) Fr. and *Didymium squamulosum* (Alb. & Schwein.) Fr. produce species complexes consisting of related sibling species that are potential morphospecies often geographically isolated. His final summation is worth a direct quotation: "Therefore, it is suggested that taxonomists undertake the naming of new morphospecies with due care, and that they base their descriptions on reasonable number of sporophores (as many as possible) collected from a number of different areas (as widespread as possible), combined with a basic understanding of the population and developmental biology of the myxomycetes". This paper should be required reading for all myxomycete taxonomists.

Schnittler and Mitchell [31] suggested five criteria that authors of new species follow as a prelude to describing new species: 1. search literature published throughout the world for possible matching descriptions; 2. reference the new taxon with several specimens from more than one locality; 3. create exact descriptions based on SEM observations, color standardized with color charts, and spore-to-spore cultures to ascertain constancy of morphological characters; 4. compare morphologically similar species to make sure characters deviate in more than one character, such as clustered versus free spores; 5. give details of the habitat such as vegetation type, elevation, and localities. These criteria were previously discussed by Keller [16] in his Plenary Address at ICSEM2 held at Madrid, Spain under the following topical headings: importance of ecological field collections; importance of collecting; importance of type collections; importance of spore-to-spore cultivation; living cultures – a biological standard; importance of monographic works; importance of computerization of mycological collections; importance of DNA sequencing techniques. It is unfortunate that Schnittler and Mitchell [31] did not review, cite, or include these observations about good taxonomic practice [16].

The purpose of this paper will highlight and expand on some of the major points of "good taxonomic practice" previously noted with an additional caveat associated with habitats and seasonality patterns. Myxomycete species concepts will be reviewed, monotypic genera will be documented and critiqued, and case studies given to evaluate the past, present, and future application of methodologies that will enhance our understanding of taxa in the Myxomycetes [18].

Monotypic Genera – human artifact or real?

Monotypic genera are those genera that include only one species. Higher taxonomic ranks are human creations that give relative order to the species and represent hierarchies of

diverging characters [33]. Therefore, ranking taxa, including the creation of monotypic genera, is highly subjective. The problem is exacerbated by the practice of publishing new names in local, non-peer reviewed journals. Thus, new descriptions and changes can be proposed without a literature review and assessment by experts in the field. The current number of monotypic genera in the Myxomycetes was determined using three sources. In accordance with Martin and Alexopolous [23], monotypic genera represent 17 of 53 genera or 32%, according to Keller and Braun [19] monotypic genera represent 21 of 57 genera or 39%, and according to Lado [21] monotypic genera represent 14 of 59 genera or 24%. Species which represent monotypic genera according to each source are shown in Table 1.

Fossil Record

Monotypic genera may be valid if it is the last remaining species from a group that has otherwise gone extinct, however, this validation would require representative examples within the fossil record. Protozoa are known from the fossil record preserved in amber [10], however myxomycete preservation is rare. Fossilized myxomycetes are only known as preserved in amber, which is plant resin that hardened under the right conditions and over a long period of time. Considering the habitats myxomycetes occupied and their fragile structure, fossilization of fruiting bodies or spores in amber is the most likely and stage . To the best of our knowledge, no myxomycologist has evaluated fossilized myxomycete taxa represented by fruiting bodies in the fossil record and this represents the most current review.

Myxomycete fruiting bodies have been reported in Baltic amber [6, 8]. The first certain myxomycete in the fossil record was a species assigned to *Stemonitis splendens* Rostaf. in Baltic amber from the Tertiary, Eocene approximately 35 to 40 million years ago. Images (see [6] Figure 1-7, Plate 15) clearly show stalked sporangia with hypothallus and columella. Further images (see [6] Figure 6 and 7) represent a well preserved columella with capillitial attachments still retaining a surface net intact, typical of an extant species of *Stemonitis,* possibly *S. splendens.* The preservation is remarkable and there is no question that this is a species of *Stemonitis* and a myxomycete. Even the black (dark) color of the fruiting body structural parts (hypothallus, stalks, and capillitial threads) are still attached to the columella with branching patterns and a surface network intact as seen in optical section [6].

Arcyria sulcata Dörfelt & Schimdt is another example of a myxomycete species from fossilized Baltic amber that was provided with a Latin diagnosis and described as a new species. The paper title "The oldest fossil myxogastroid slime mould" is based on Baltic amber from a similar time-period (early Tertiary, Eocene) as the *Stemonitis* species. However, it is not clear which myxomycete species came first in the fossil record. The color illustrations (see [8] Figures 1-4) collectively show the stalk and capillitial threads attached at the base to the calyculus. The color of the fruiting body is not mentioned in the species description nor is the color evident in the amber. The capillitum appears as a coiled network of apparently elastic threads. Special light microscopic techniques (laser scanning-microscope) show the ornamentation (cogs and rings) on the capillitial threads typical of *Arcyria* species [8]. The general habit appears similar to the extant myxomycete species *Arcryia denudata* (L.) Wettst.**,** suggesting myxomycete fruiting bodies have changed little in 35 to 40 million years.

Protophysarum balticum Dörfelt & Schmidt was described as a new myxomycete species in Baltic amber from the Tertiary period. A Latin diagnosis was not included, only an English description. This specimen in amber clearly lacks the features of *Photophysarum phloiogenum* M. Blackw. & Alexop. based on the following morphological characters: the stalk is much larger and thicker, the basal part of the spore case is persistent, somewhat turbinate, with a thickened wall, sometimes only a cuplike base remains. Apparently the

Table 1. List of monotypic genera

Martin and Alexopolous (1969)
Arcyodes incarnata (Alb. & Schwein.) O. F. Cook
Barbeyella minutissima Meyl.
Brefeldia maxima (Fr.) Rostaf.
Calomyxa metallica (Berk.) Nieuwl.
Calonema aureum Morgan
*Cienkowskia reticulata** (Alb. & Schwein.) Rostaf.
Clastoderma debaryanum A. Blytt
Cornuvia serpula (Wigand) Rostaf.
Erionema aureum Penz.
Leocarpus fragilis (Dicks.) Rostaf.
Lindbladia tubulina Fr.
Listerella paradoxa E. Jahn.
Minakatella longifila G. Lister
Mucilago crustacea F. H. Wigg.
Physarella oblonga (Berk. & M. A. Curtis) Morgan
Prototrichia metallica (Berk.) Massee
Wilczekia evelinae Meyl.

Keller and Braun (1999)
Arcyodes incarnata (Alb. & Schwein.) O. F. Cook
Arcyriatella congregata Hochg. & Gottsb.
Badhamiopsis ainoae (Yamash.) T. E. Brooks & H. W.Keller
Barbeyella minutissima Meyl.
Brefeldia maxima (Fr.) Rostaf.
Calomyxa metallica (Berk.) Nieuwl.
Cornuvia serpula (Wigand) Rostaf.
Dictydiaethalium plumbeum (Schumach.) Rostaf.
Erionema aureum Penz.
Kelleromyxa fimicola (Dearn. & Bisby) Eliasson
Leocarpus fragilis (Dicks.) Rostaf.
Leptoderma iridescens G. Lister
Lindbladia tubulina Fr.
Listerella paradoxa E. Jahn.
Minakatella longifila G. Lister
Mucilago crustacea F. H. Wigg.
Physarella oblonga (Berk. & M. A. Curtis) Morgan
Protophysarum phloiogenum M. Blackw. & Alexop.
Prototrichia metallica (Berk.) Massee
Trabrooksia applanata H. W. Keller
*Willkommlangea reticulata** (Alb. & Schwein.) Kuntze

Lado NOMENMYX (2001)
Arcyriatella congregata Hochg. & Gottsb.
Barbeyella minutissiuma Meyl.
Brefeldia maxima (Fr.) Rostaf.
Cornuvia serpula (Wigand) Rostaf.
Kelleromyxa fimicola (Dearn. & Bisby) Eliasson
Leocarpus fragilis (Dicks.) Rostaf.
Lindbladia tubulina Fr.
Listerella paradoxa E. Jahn.
Minakatella longifila G. Lister
Mucilago crustacea F.H. Wigg.
Physarella oblonga (Berk. & M. A. Curtis) Morgan
Protophysarum phloiogenum M. Blackw & Alexop.
Prototrichia metallica (Berk.) Massee
*Willkommlangea reticulata** (Alb. & Schwein) Kuntze

* *Willkommlangea reticulata* and *Cienkowskia reticulata* are now recognized as synonyms.

surface or internal areas of the stalk and upper parts show no cellular detail, however, the filaments are more likely fungal in origin. Images (see [7] Figure 1, G) show the thickened margin is enrolled, unlike *Protophysarum* which has a delicate spherical spore case that is thin and fragile without a cuplike base. Myxomycete peridia and stalks are not cellular whereas lichens often have filaments or cells indicative of fungi. Dr. Thorsten Lumbsch, an expert lichenologist at the Chicago Field Museum, reviewed the text and illustrations of this paper [7]. He indicated that the specimen is not a myxomycete but a lichen in the calicioid group sometimes referred to as "stubble lichens". Stalked species of *Licea* sometimes resemble calicioid lichens (Keller, pers. obs.), however the spores of *Protophysarum balticum* appear as a solid mass whereas myxomycete species usually have a powdery spore mass. The spore mass in some myxomycete species may stay within the spore case when the top portion undergoes circumscissile dehiscence and the lid falls away from the peridium, as in the stalked species *Licea operculata* (Wingate) G.W. Martin. However, *Protophysarum balticum* more closely resmbles the calicioid lichen, *Chaenotheca* species, fossilized in amber and illustrated by Rikkinen [30] (see mature ascoma in [30] Figures 1 and 2).

In addition to fruiting bodies, other myxomycete life cycle stages have been preserved in amber, such as the plasmodium of a physaraceous species [35]. Even so, there are problems with the description of the fossilized plasmodium. The objects inside the amber are a continuous

section that is described as the veins of a phaneroplasmodium and vesicles that are described as part of a plasmodium that has budded off and sclerotized. Every part of the plasmodium appears as though it is a network of bubbles that Waggoner and Poinar [35] describe as spherules, and suggest is the result of initial plasmodial sclerotization. Inside each spherule, Waggoner and Poinar also state that there are typically 0 to 6 spherical nuclei that are 8 to 24 µm in diameter and classify the plasmodium in the Physarales. However, the Physarales is an order in which the nuclei are characteristically small, for example, the nuclei of *Physarum polycephalum* Schwein. are well documented [11] as globose to elliptical in shape and 2.5 to 7 µm in diameter. The "nuclei" described by Waggoner and Poinar are outside of the size range characteristic of the Physarales and outside of the size range expected for most myxomycetes. The images are unconvincing, especially the scattered bubbles and questionable "nuclei" that lack any organization or internal details suggesting a plasmodium or sclerotium. It is doubtful that the objects inside the amber are actually a myxomycete plasmodium based on the description by Waggoner and Poinar and the amorphous properties of a myxomycete plasmodium that would make the capture of a plasmodium by resin extremely unlikely. Arguments for the description of fossilized plasmodium would have been more convincing had the authors provided an image of a partially sclerotized plasmodium in vitro compared to that in the amber, provided reasoning for the difference in expected versus observed nuclei size, and consulted the expertise of a professional myxomycologist.

The validation of monotypic genera in the myxomycetes using the fossil record is highly problematic considering the improbability of fossilization and difficulty in assigning specimens to the correct genus. Fossilized specimens of both *Stemonitis splendens* and *Arcyria sulcata* appear to be valid, however, it is doubtful that *Protophysarum balticum* is even a myxomycete and more likely to be a calicioid lichen. However, if the fossilized species of *Protophysarum* was the extinct sister species to *Protophysarum phloiogenum*, according to some species concepts, the genus would no longer be considered monotypic and would be a good example where an extant species represents a monotypic genus.

Additional Methods of Good Taxonomic Practice

Phylogenetic Analysis

The description of a new species that represents a monotypic genus is compelling when experts agree the specimens differ from all other genera by more than one character. When a genus is based on only one character it is open to questionable interpretation. The authors suggest the use of DNA analysis as additional evidence to support monotypic genera [9]. Good candidates for genetic analysis are: *Arcyriatella congregata* Hochg. & Gottsb., *Erionema aureum* Penz., *Kelleromyxa fimicola* (Dearn. & Bisby) Eliasson, *Leocarpus fragilis* (Dicks.) Rostaf., *Lindbladia tubulina* Fr., *Listerella paradoxa* E. Jahn., *Minakatella longifila* G. Lister, *Physarella oblonga* (Berk. & M. A. Curtis) Morgan, *Protophysarum phloiogenum, Prototrichia metallica* (Berk.) Massee, and *Trabrooksia applanata* H. W. Keller (Table 1). A good example of the use of phylogenetic analysis is represented by the genus *Schenella. Schenella simplex* T. Macbr. was analyzed using molecular DNA sequencing and was found to be synonymous with the fungal gasteromycete (puffball) *Pyrenogaster* [9].

Ecology and Seasonal Fruiting

The ecology and seasonal occurrence should be recorded regularly when studying myxomycetes, since little is known about the phenology of myxomycetes. Although these are not characters used to classify myxomycetes, they may be supporting characters by which to group or split morphologically similar specimens. Therefore, collections should always thoroughly describe the habitat in detail,

Table 2. Seasonal occurrence of *Perichaena* species

	Months												
	J	F	M	A	M	J	J	A	S	O	N	D	Total
Perichaena brevifila	1		1						2	3	6		13
P. microspora						3	3	3					9
P. pedata						2	4	3			1		10
P. syncarpon	1					1	8	8	4	1	1		24

"specimen found on upper layer of decaying leaves under full cover of a bush in an urban landscape" rather than "found on leaves." Furthermore, the collection date of fruiting bodies should always be recorded. A good example of the use of detailed habitat description and seasonality of fruiting to support classification is in the monograph of *Perichaena* in a thesis by Keller [14]. The thesis recorded the seasonal occurrence of *Perichaena* species from shaded, decaying leaves or straw stacks. Detailed notes about the habitat showed that *P. brevifila* T. E. Brooks & H. W. Keller was found near the bottom of leaf litter and other species were found near the top of leaf litter, and *P. microspora* Penz. & Lister was collected only from Florida and Louisiana and is therefore considered a Southeastern species in the United States of America (U.S.A.)

Collections were made between 1930 and 1977 from various locations in the U.S.A.: *Perichaena brevifila* from Georgia, Kansas, and Virginia, *P. microspora* from Florida, Georgia, and Louisiana (collection data supplemented from labels provided by BPI), *P. pedata* (Lister & G. Lister) Lister ex E. Jahn from Illinois, Florida, and Kansas, and *P. syncarpon* T. E. Brooks from Iowa and Kansas. The collection dates of the four *Perichaena* species that only occurred on either shaded, decaying leaves or straw stacks were compared (Table 2). The occurrence of *P. brevifila* was mostly (>1 observation) in September, October, and November, whereas the species *P. microspora* and *P. pedata* occurred mostly in June, July, and August, and *P. syncarpon* occurred mostly in July, August, and September (Table 2). Seasonality of fruiting alone is not a defining characteristic of *Perichaena* species. However, these data add support to the recognition of four separate species. Details about the habitat may explain why there is a seasonal difference in occurrence of the species and why there are a few collections that are outliers. If the species of *Perichaena* require similar average temperatures for fruiting, the occurrence of *P. brevifila* at the bottom of the decaying leaf litter may explain the difference in seasonality. It takes longer for average temperatures at the bottom of a leaf litter to reach the same temperature at the top of leaf litter. The months where only one collection is recorded may also be explained by the habitat. Leaf cover may provide enough protection for the fruiting bodies to be preserved for months, which would explain the collection of *P. brevifila* in January and March, and the collection of *P. syncarpon* in January and *P. pedata* in November.

The assumption is that collectors regularly and equally collected during each month and from the same habitats. In general, myxomycete fruiting in the U.S.A. is most diverse and abundant in June, July, and August. Exceptions to this generality are known from California where the rainy months are in January and February. Indeed, those months correspond to the months where myxomycete fruiting is most diverse and abundant, lending support to the seasonality of myxomycetes based on precipitation and temperatures. Clearly, in the case of *Perichaena* species, notes about the ecology and seasonal occurrrence of fruiting bodies in the field add support to deliniation of species within the genus. The same could be true for other genera but more observations of seasonal occurrence and ecology must be recorded before that conclusion can be made.

Discussion of Selected Monotypic Genera

***Trabrooksia applanata*, a monotypic genus case study**

Trabrooksia applanata was described as a new monotypic genus in 1980 to honor Travis E. Brooks and to commemorate his collection of corticolous myxomycetes [15]. This taxon was first collected in 1962 by T. E. Brooks. Since then more than 45 collections are known from seven states in the U.S.A. and also the countries of Great Britain, Ireland, and Japan. It occurs on living trees such as *Acer negundo* L., *Fraxinus* sp., *Juniperus virginiana* L., *Podocarpus macrophylla* (Th.) Sw., *Populus balsamifera* L., and *Ulmus* sp. *Trabrooksia* has been recognized by authors in numerous publications [13, 19, 25, 26, 29]. A new variety, *T. applanata* var. *microspora* Y. Yamam., was described by Yamamoto [36] with slightly smaller spores (8-9 µm) based on a single collection. His description does not mention the iridescent peridium and the spore size when compared to 11-13 µm cited by Keller [15] that falls within a size range not unlike many other myxomycete species. The variety *microspora* should be considered a synonym of *T. applanata.*

In 1983 M. L. Farr updated and revised the Martin and Alexopoulos 1969 world monograph [24], including *Trabrooksia* in the key to the genera but noting the following: "*Trabrooksia applanata*, type and only species of the genus *Trabrooksia* H. W. Keller, is highly suggestive of a limeless form of *Didymium sturgisii* Hagelst, with which it was compared in the protologue [15]. It has not been grown in culture, nor tested for presence of elemental calcium. The absence of lime combined with the other traits preclude the classifying this slime mold in any other genus. Whether or not the absence of lime is an inherent or environmentally induced character is not quite certain as yet, but so far it has proved stable in numerous collections from various states. The genus is keyed here, at least for purposes of specimen identification." This commentary is provided here as a direct quotation because Martin *et al* [24] did not assign *Trabrooksia* as a synonym of *Didymium sturgisii*.

NOMENMYX is a nomenclatural treatment to give one correct name for every species [21]. The names compiled were based on scanning literature sources and not examination of holotype specimens or collections of each species. *Trabrooksia applanata* is listed as a synonym of *Didymium sturgisii* [21] and the source is Martin, Alexopoulos & Farr, Gen. Myxomycetes: 71 [24]. This nomenclatural determination is premature in view of the fact that there are numerous morphological characters that distinguish these two taxa. Additional specimens of both species have been compared morphologically along with detailed species descriptions by Brooks [4] (*Trabrooksia* p. 173-176, *Didymium sturgisii* p. 185-189); by Keller [15] (*Trabrooksia* p. 396-401); by Keller and Braun [19] (*Trabrooksia* p. 152, *Didymium sturgisii* p. 149-150).

Morphological comparison of *Didymium sturgisii* and *Trabrooksia applanata*

Fructifications are similar in shape, form, and size, usually as flattened to irregularly effused plasmodiocarps and less often as sessile sporangia. Color and peridial characteristics are distinctly different in the two taxa. The peridium in *Trabrooksia* is thin, membranous, transparent with a silvery to iridescent surface, rarely brownish because of reflected light on the internal spore mass, lacking structural calcium carbonate as seen with SEM (see [15] Figures 7-10) and with no effervescence in clear lactophenol; in *Didymium sturgisii* the peridium is a thin calcareous crust of crystals either aggregated with points protruding or sometimes free and stellate, color more grayish when lightly sprinkled with crystals, noncalcareous specimens were not seen. Many plasmodiocarps of *D. sturgisii* that are prematurely dried or aberrant have a calcareous peridium. Noncalcareous specimens of *D. sturgisii* should be expected since the presence or absence of calcium carbonate is a variable character in species of *Didymium*, as in other species of the Physarales. When fructifications prematurely dry in the field or in moist chamber cultures these environmental conditions often yield noncalcareous forms, however, none were seen in *D. sturgisii* [19].

The capillitial system is distinctly different in the two taxa. *Trabrooksia* has simple, subparallel, tubular threads vertically aligned (see [15] Figures 4, 5, 6 as seen with the light microscope and Figures 9, 11 with SEM) attached above to the peridium and below to the base of the plasmodiocarp. The capillitial threads rarely branch, lack calcium carbonate, anastomosing, and the violaceous color seen in many species of *Didymium*. The capillitial system is a constant character in all of the specimens examined. In contrast, *D. sturgisii* has trabeculae (calcareous pillars) often with broad funnel-shaped attachments to the upper peridial wall [12], extending and attached to the base or sometimes with ends truncated, unattached, and giving rise to branching, slender, capillitial threads mostly with violaceous colors. The capillitial system may be unassociated with the trabeculae and in some of the larger plasmodiocarps, may be simple, branched or anastomosing, with the extremities hyaline and attenuating into narrow attachments [4, 19]. The spores appear similar in size and ornamentation in both species but additional SEMs are needed of *D. sturgisii.*

Specimens examined of *D. sturgisii* were cited in Brooks [4] and Keller and Braun [19]. Additional collections examined: Spain, E. Guadalajara, Tamajon-Guadalajara, on bark of *Juniperus oxycedrus* L., 23 March 1980, collected by *C. Lado, MA-Fungi 17073*; E. Guadalajara, Tamajon-Erata Ntra. Sra. Enebral, on bark of *Juniperus thurifera* L., 22 October 1980, collected by *C. Lado, MA-Fungi 17177*. U.S.A., Iowa, East Okoboji, on decaying wood, 4 August 1933, *BPI 817869*; Colorado, Gilpin County, Perigo N Slope, dead aspen bark, moist chamber, wetted 21 July 9, 1979, harvested 21 August 1979, collected by *Chapman, BPI 817874*. Specimens of *Trabrooksia applanata* were examined and cited by Keller [15].

***Badhamiopsis ainoae*, a monotypic genus case study**

Badhamiopsis ainoae (Yamash.) T. E. Brooks & H. W. Keller is most similar in appearance and more easily confused with *Trabrooksia applanata*. The two taxa are common corticolous myxomycetes restricted to the bark surface of living trees and vines and sometimes appear in close proximity on the same piece of bark in the field and in moist chambers, especially on *Juniperus virginiana*. *Badhamiopsis* is clearly a physaraceous myxomycete, however, sometimes non-calcareous fruiting bodies fail to produce bubbles in clear lactophenol indicative of calcium carbonate. *Badhamiopsis* differs from all other genera in the Physaraceae in its absence of a capillitial network. The predominately effused plasmodiocarps and capillitial system were the basis for the recognition of a separate genus. The capillitium consists of tubular invaginations from the upper peridium, oriented as vertical, unbranched pillars, more or less spike-like, usually enclosing dense deposits of white calcareous granules (not crystalline), attenuating as short, slender, often bifurcate, hyaline, non-calcareous threads attached to the base of the plasmodiocarp [20]. The plasmodiocarps, when broken open, detach from the bottom where the delicate hyaline, non-calcareous, bifurcate threads break and the calcareous spike-like pillars remain attached like icicles to the upper peridium. When the spores are removed, this is a distinctive feature of the species. However, under certain environmental conditions on living trees in the field and in moist chamber, the vertical pillars are thin, hyaline, and non-calcareous, similar to *Trabrooksia*. Specimens examined were cited in Keller and Brooks [20] and Keller and Braun [19].

Two additional taxa assigned to *Badhamiopsis* merit commentary. *Badhamiopsis nucleata* H. Z. Li is stalked, globose with petaloid dehiscence exposing a branching, calcareous capillitium based on the Latin diagnosis. Four scanning electron micrographs attempt to illustrate what appears to be a stalked species of *Badhamia* or possibly a *Physarum*. However, the SEMs lack the high resolution to

show detailed fine structure of the fruiting body. Furthermore, the stalked habit, the branching capillitium, and larger spore size (15-18 µm in diameter) are characters that would exclude this taxon from the genus *Badhamiopsis* [22]. A single collection of *Badhamiopsis cavifera* Nann.-Brem. & Y. Yamamoto on moss from a living tree has a capillitium typical of a *Diderma* or *Didymiu*m. Two sessile, pulvinate sporangia are illustrated that apparently represent the basis for the description since no other fruiting bodies are mentioned in the text. Without the holotype available to examine and such a scanty specimen it is difficult to assess if this represents a new taxon. However, the line drawings clearly indicate that the habit, the color and calcareous peridium, the obvious didymiaceous capillitial threads, exclude this taxon from the genus *Badhamiopsis* [28].

***Protophysarum phloiogenum*, a monotypic case study**

Protophysarum phloiogenum is an example of a good monotypic genus [2]. At first glance, the specimen appeared to be a *Lamproderma* within the Stemonitaceae, however, four distinct characters contradicted that placement. First, the plasmodium was a phaneroplasmodium not an aphanoplasmodium; second, sporangial development is subhypothallic not epihypothallic; third, calcium carbonate was found associated with the peridium and capillitium; and finally, the spores germinated via splitting open rather than through a pore. The description is a good example of patience in taxonomic descriptions as the specimens were cultured spore-to-spore over a period of seven years, with variation noted only in spore size. Furthermore, additional work on the species has been published [1, 3] which lends further support for the species, including analysis of sporophore development and identification of the species from additional locations. Interestingly, the initial collections were made from the bark of living *Ulmus americana* L. in Colorado and later collections were made from the pith of dead saguaro, *Carnegiea gigantea* (Engelm.) Britt. & Rose, in Saguaro National Monument, Arizona.

Conclusions

Phylogenetic relationships based on molecular methodologies will highlight the taxonomy of Myxomycetes in the 21st Century. The criteria for species concepts will surely involve DNA analysis that focus on population diversity and the complexities of understanding population differentiation. Species concepts should include more data than just morphological differences with additional information from biogeographical patterns, habitat analysis based on field observations over time, and seasonality patterns in circumscribed geographical areas. Selected monotypic genera represent sources of DNA that will eludicate taxonomic ranks (orders, families, and genera) and a more correct assignment of problematic taxa such as *Protophysarum* and *Trabrooksia.* Monotypic genera may be the last representatives of taxa gone extinct but the paucity of a fossil record leaves many unanswered questions. Good taxonomic practice and monographic studies using modern methodologies are important because they lay the foundation for sound scientific research and allow the application of knowledge across a broad spectrum. The future of myxomycete systematics requires a shift from descriptive taxonomy to in-depth studies using hypotheses that test phylogenetic relationships, biogeographical patterns of distribution, and the restriction of species to habitats with special ecological characteristics. Only when myxomycologists collaborate will we reveal answers to the question: "What is a myxomycete species?" [18].

Acknowledgments

This research was financially supported in part by grants from the National Science Foundation, Biodiversity and Survey Program, DEB #0343447, National Geographic Committee for Research and Exploration #7272-02, and, Discover Life in America Award #2002-17. We thank Carrie M. Harrington for the English to Spanish translation.

References

1. Blackwell, M., 1974. A study of sporophore development in the myxomycete *Protophysarum phloiogenum.* Archives for Microbiology 99: 331-344.
2. Blackwell, M., C.J. Alexopoulos, 1975. Taxonomic Studies in the Myxomycetes. IV. *Protophysarum phloiogenum*, a new genus and species of Physaraceae. Mycologia 67: 32-37.
3. Blackwell, M., R.L. Gilbertson, 1984. Distribution and sporulation phenology of Myxomycetes in the Sonoran Desert of Arizona. Microbial Ecology 10: 369-377.
4. Brooks, T.E., 1967. A study of corticolous Myxomycetes. Ph.D. Dissertation, University of Kansas, Lawrence, United States of America.
5. Clark, J., 2000. The species problem in the Myxomycetes. Stapfia 73: 39-53.
6. Domke, W., 1952. Der erste sichere Fund eines Myxomycetes im Baltischen Berstein (*Stemonitis splendens* Rost. Fa. Succini fa. nov. Foss.). Mitteilungen aus dem Geologischen Staatsinstitut in Hamburg 21: 154-161.
7. Dörfelt, H., A.R. Schmidt, 2006. An archaic slime mould in Baltic amber. Palaeontology 49: 1013-1017.
8. Dörfelt, H., A.R. Schmidt, P. Ullmann, J.Wunderlich, 2003. The oldest fossil myxogastroid slime mould. Mycological Research 107: 123-126.
9. Estrada-Torres, A., T.W. Gaither, D.L. Miller, C. Lado, H.W. Keller, 2005. The myxomycete genus *Schenella*: morphological and DNA sequence evidence for synonymy with the gasteromycete genus *Pyrenogaster*. Mycologia 97: 139-149.
10. Frederick, L., 1990. Phylum plasmodial slime molds, class Myxomycota. *In:* Margulis, L., J.O. Corliss, M. Melkonian, and D.J. Chapman (eds.), Handbook of Protoctista. Jones and Bartlett Publishers, Boston. Pp. 467-483.
11. Gray, W.D., C.J. Alexopoulos, 1968. Biology of the Myxomycetes. New York: Ronald Press.
12. Hagelstein, R., 1944. The Mycetozoa of North America based upon the specimens in the Herbarium of the New York Botanical Garden. Mineola, New York.
13. Ing, B., 1999. The Myxomycetes of Britain and Ireland: an identification handbook. Richmond Publishing Company, Slough, England.
14. Keller, H.W., 1971. The genus *Perichaena* (Myxomycetes): a taxonomic and cultural study. Ph.D. Dissertation, University of Iowa, Iowa.
15. Keller, H.W., 1980. Corticolous Myxomycetes VIII: *Trabrooksia,* a new genus, 72: 395-403.
16. Keller, H.W., 1996. Invited plenary address, Biosystematics of Myxomycetes: a futuristic view. *In:* Lado, C. and J.C. Hernandez (eds.), Second International Congress on the systematics and ecology of Myxomycetes (ICSEM2), organized by Real Jardín Botánico, CSIC, Madrid, Spain, Abstract Volume: 23-37.
17. Keller, H.W., 2004. Tree canopy biodiversity and student research experiences in Great Smoky Mountains National Park. Systematics and geography of plants 74: 47-65.
18. Keller, H.W., 2005. What is a myxomycete species? *In*: Galindo, G., M.G. Santiago- Martínez, A. Montoya Esquivel, A. Estrada-Torres (eds.), Fifth International Congress on systematics and ecology of Myxomycetes. Abstracts of oral and poster presentations. Universidad Autónoma de Tlaxcala. 45.
19. Keller, H.W., K.L. Braun, 1999. Myxomycetes of Ohio: their systematics, biology and use in teaching. Ohio Biological Survey Bulletin New Series Vol. 13, Number 2, 1-182.
20. Keller, H.W., T.E. Brooks, 1976. Corticolous Myxomycetes IV: *Badhamiopsis,* a new genus for *Badhamia ainoae*, Mycologia 68: 834-841.
21. Lado, C., 2001. NOMENMYX. A nomenclatural taxabase of Myxomycetes. Cuadernos de Trabajo Flora Micological Iberica 16: 1-221.
22. Li Hui-zhong. 1989. Myxomycetes from China IV: Taxonomic study in the Myxomycetes of Shennongjia. *In:* Fungi and Lichens of Shennongjia, Mycological and Lichenological Expedition to Shenongjia, Academia Sinica. World Publishing Corp, Beijing. China. Pp. 1-78d.
23. Martin, G.W., C.J. Alexopoulos, 1969. The Myxomycetes. University of Iowa Press, Iowa.
24. Martin, G.W, C.J. Alexopoulos, M.L. Farr, 1983. The genera of Myxomycetes. University of Iowa Press, Iowa.
25. Mitchell, D.W., 2004. A key to the corticolous Myxomycota, Systematics and geography of plants 74: 261-285.
26. Mitchell, D.W., 2005. Myxomycetes, CD-ROM of illustrated keys and inventory of all world species. CD for MS Windows. DiKey, dichotomous keys to various groups of organsisms. Inventory and synoptic keys of all legitimately described myxomycete taxa. This is a private publication by the author and is regularly updated.
27. Nannenga-Bremekamp, N.E., 1991. A guide to temperate Myxomycetes, an English translation by A. Feest and Y. Burggraaf of De Nederlandse Myxomyceten. Biopress Ltd., Bristol, England.
28. Nannenga-Bremekamp, N.E, Y. Yamamoto. 1988. *Badhamiopsis cavifera* (Myxomycetes), a new species from Japan. Journal of Japanese Botany 63: 28-30.
29. Neubert, H., W. Nowotny, K. Baumann, 1996. Die Myxomyceten Deutschlands und des angtenzenden Alpenraumes unter besonderer Beruchsichtigung Osterreichs, Band. 2. Physarales. Karlheinz Baumann: Gomaringen.
30. Rikkinen, J. 2003. Calicioid lichens from European Tertiary amber. Mycologia 95: 1032-1036.
31. Schnittler, M., D.W. Mitchell, 2000. Species diversity in Myxomycetes based on the morphological species concept – a critical comparison. Stapfia 73: 55-62.
32. Stephenson, S.L, H. Stempen, 1994. Myxomycetes: a Handbook of Slime Molds. Timber Press, Portland, Oregon.
33. Stuessy, T.F., 1990. Plant Taxonomy. Columbia University Press, New York, New York.
34. Sundberg, W.J., H.W. Keller, 1996. Myxomycetes: some tools and tips on collection, care, and use of specimens. Inoculum 47 (4): 12-14.
35. Waggoner, B.M., G.O. Poinar, Jr., 1992. A fossil myxomycete plasmodium from Eocene-Oligocene amber of the Dominican Republic. Journal of Protozoology 39: 639-642.
36. Yamamoto, Y., 1994. Some new and rare records of Myxomycetes from Japan. Hikobia 11: 523-530.
37. Yamamoto, Y., 1998. The myxomycete biota of Japan. Shinjuku-ku, Tokyo, Toyo Shorin Publishing.