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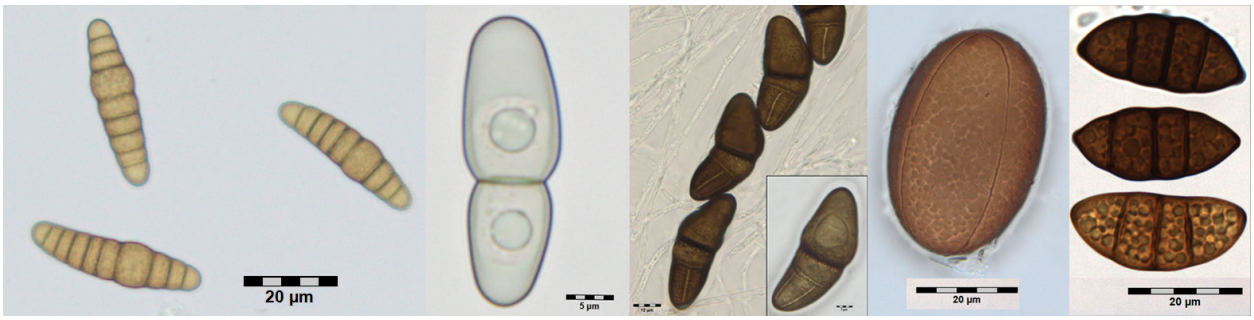
An Excursion to Collect Tropical Fungi in Gorongosa National Park, Mozambique, May 20 – June 17, 2016

By Jason Karakehian

On May 5, 2014 I was listening to a local radio talk show that featured Harvard biologist Edward O. Wilson and his new book with photographs by another Harvard entomologist, Piotr Naskrecki. In *A Window on Eternity: A Biologist's Walk Through Gorongosa National Park*, Wilson describes Gorongosa National Park (GNP) as having once been teeming with wildlife and tourists but now decimated by the Mozambican civil war that began in 1977 and ended in 1992. Occupying a region the size of the state of Rhode Island and situated in the middle of Mozambique in Sofala Province, the Park was a frequent battleground. The soldiers hunted any animal generally over 20 pounds for food, except the crocodiles that were too elusive to be caught and slaughtered. After the end of the civil war the Park remained unprotected and poaching continued. The result was a landscape wherein one particular visitor noted in 2004 that one could travel for a day in the Park without seeing any large animals other than birds (Wilson, 2014).

That visitor was Greg Carr, who began that year, in contract with the Mozambican government, to fund the restoration of the Park through his nonprofit, the Gregory C. Carr Foundation. The approach is holistic; at the time of our visit over 400 people from local communities were employed in the Park and in the surrounding communities in jobs ranging from construction



Ascospores of various fungi collected. From left to right: *Oedohysterium sinense*; *Ostreichnion* sp.; unknown Dothideomycete (Pleosporales); *Ascobolus bistisii*; *Rhytidhysteron rufulum*.

The collectors. From left to right: Teresa Iturriaga, Leif Ryvarden, Jason Karakehian, Meg Coates Palgrave.

and hospitality, community education, research, health and agriculture training to patrolling for poachers – becoming "fiscals," in Portuguese, or Park Rangers. The populations of animals, such as waterbuck, sable antelope, impala and kudu, are rapidly growing in the Park and tourists are returning. Elephant can be seen regularly, and the number of lions is increasing, but populations of other top predators, such as hyena and leopard, were virtually extirpated and have not yet made a comeback.

Wilson further described the inception of the Wilson Laboratory at the Park. It is one leg of the effort to restore the Park through establishing a destination location for scientists to conduct field work. It is modeled along the lines of the Smithsonian Tropical Research Institute on Barro Colorado Island in Panama. Biological surveys have commenced in the Park and reference collections of insects, reptiles, and vascular plants have been created. Keenly listening to Wilson I noted that he hadn't mentioned fungi! After searching the literature I learned that were no concerted efforts to study the fungi in Mozambique, and that a survey of fungi in the Park would be a first.[1] With the assistance of Professor Pfister and my colleagues in the Pfister Lab and elsewhere, I put together a proposal to conduct a survey of fungi in GNP under the aegis of the Farlow Herbarium. I obtained funding through the Ella Lyman Cabot Trust, a Boston-based foundation dedicated to making change in the world by supporting the projects proposed by individuals. The Associate Director of the E. O. Wilson Biodiversity Laboratory, who also happens to be Piotr Naskrecki, has his office conveniently next to the Harvard Herbaria in the Museum of Comparative Zoology. He met regularly with me and my colleague, Teresa Iturriaga, to discuss the project.

We arrived in the Park on May 20 after a brief visit to the herbarium at Eduardo Mondlane University. At the herbarium we examined a dozen or so specimens of undetermined lichenized fungi. This is the only collection of fungi in any institution in the country. Once in the Park we got to work visiting as many different habitats within the Park boundary as possible and collected micro- and macro-fungi. In addition to Teresa and I, who specialize in discomycetes or "cup-fungi," we were joined by Leif Ryvarden from Oslo, Norway who is a world expert on wood-decay fungi such as "crusts" and "polypores." Also joining us was botanist Meg Coates Palgrave from Zimbabwe who revised and updated *Trees of Southern Africa* by Keith Coates Palgrave, or "the bible" as it is known to the botanists of the region. Her familiarity with the Park and expert identifications of the region's trees, as well as her interpretations of the various landscapes that we visited, were invaluable to us. Whenever we left the fence-bound

1 However, I recently learned that mycological research in Gorongosa was conducted before our survey and was published in a recent paper by Rodríguez-Echeverría, et al., Arbuscular mycorrhizal fungi communities from tropical Africa reveal strong ecological structure. *New Phytologist*. 2016. doi: 10.1111/nph.14122

Various fungi collected at GNP. From top left to right: *Ascobolus bistisii* ascoma on impala dung; *Coccomyces* sp. on dead leaf of *Brachystegia boehmii*; *Coniodictyum chevalieri* on living fruits of *Ziziphus mucronata*; Oedohysterium sinense hysterothecia on rotten wood; *Schizophyllum commune*; unknown anamorphic fungus emerging from rotten fruit of the palm *Hyphaene coriacea*; *Acervus epispartius*; *Porostereum spadiceum*; *Marasmius haematocephalus*; *Geastrum* sp.; *Xylaria* sp. on rotten petioles of *Hyphaene coriacea*; *Flavodon flavus*; *Microporus quarrei*; *Trametes elegans*; *Stilbocrea* sp.

Chitengo base camp we were accompanied by an armed "fiscal" who would keep an eye out for animals as we walked through the bush.

Voucher specimens were examined and given an "MOZ" collection number, photographed, then dried and wrapped for return to our home institutions for final determinations and division into duplicates. Duplicate specimens will be deposited in the herbaria at the Wilson Lab in GNP and Eduardo Mondlane University, in the capitol Maputo, the Farlow Herbarium and the Botanical Museum in Oslo, Norway. Our collection data, including photographs and localities, will be added to the Park's biodiversity database.

Due to the effects of a particularly strong El Niño that began in early 2015, there was severe drought in mid-to-southern Mozambique. In fact, the summer rainy season, normally November through March, never materialized. Luckily for Mozambicans in the region, a little late season rain allowed for late planting of crops and helped initiate some fungal growth. Collecting proved difficult until a few regions of closed canopy forest were discovered not far from Chitengo base camp. Still, the diversity of fungi in fruit was limited; there were very few species of fleshy mushrooms or discomycetes to be found and many carbonaceous fructifications were senescent or dead. Despite these conditions we made 500 collections in the one month we were there.

The dry season meant roads were free of mud making travel easier and we visited some diverse landscapes and habitats in the Park. Notably, we visited a sand forest, a closed canopy dry forest, where we found no ferns or mosses and very little leaf litter on the loose, sandy forest floor due to the action of termites. Miombo woodlands, composed of *Brachystegia* and *Julbernardia* tree species were beautiful and interesting landscapes, but probably would have been more productive for fungi in the wet season. In the calcareous soils to the east of Chitengo, across the Urema River, we visited the southernmost region of the Cheringoma Plateau, noting epiphytic orchids, stag horn ferns and crustose lichens in great diversity, along with tsetse flies which were a nuisance but

Leif Ryvarden beside *Pseudopiptoporus devians* (Bres.) Ryvarden fruiting on living *Sclerocarya birrea*.

do not carry the sleeping sickness pathogen that affects humans. There we explored moist riverine habitat in shaded gorges, dambo grasslands that are periodically flooded, and dry miombo woodlands where we observed large fruitings of stipitate polypores like *Amauroderma* and *Humphreya eminii* that somewhat resembles stipitate forms of *Ganoderma*. *Amauroderma* is mycorrhizal and *Humphreya* is a root pathogen.

Immense active and abandoned termite mounds were found in great profusion throughout the Park. After perusing Roger Heim's beautifully illustrated 1977 treatise, *Termites et Champignons*, Teresa and I were eager to search these mycologically promising ecosystems for any associated fungi such as *Termitomyces* and *Xylaria* species, but found nothing in fruit. Encircling the butt of a living *Sclerocarya birrea* tree, Leif found *Pseudopiptoporus devians* (Bres.) Ryvarden, a polypore in a genus that he had erected in 1980 based only on the type collection made over one hundred years ago in 1913 in Zumba, Tete Province, Mozambique, far northwest of the Park near the Zimbabwean border. Since dung was in no short supply, it was easy to collect samples from a variety of her-

bivores such as elephant, impala, and warthog. I placed them in moist chambers to wait for fungi to appear. Over a period of 10 -14 days we were rewarded with pyrenomycetes such as *Delitschia* and *Sporormiella* species as well as discomycetes such as species of *Thecotheus*, *Pseudombrophilia* and *Ascobolus bistissi*, a species with large, wine-colored, ellipsoid spores with interlocking ridged ornamentations resembling the surface of a brain.

Intriguingly, in many of the mixed palm woodlands north of Chitengo, we noticed very few epiphytes on the trees. Mosses, lichens and liverworts were infrequently encountered and species diversity appeared to be low. We could see that these landscapes become arid in the dry winter season and were frequently burned. Furthermore, there was very little old downed wood or leaf litter. Any trunk or branch that had not been burned was rendered a fragile, hollow husk by the action of omnipresent termites. Termitaria dotted the landscape, the very large abandoned ones supporting shrubs and enormous specimens of trees that would normally be found in wetter habitats and in richer soils. On nearly every living tree in the landscape, the termites had built channels, sinuous networks like veins made of a delicate crust of sand, running up the trunks and branches to protect themselves from the sun and allowing them to forage in the crowns. Termites are integral players in the carbon cycle in these ecosystems and it is profound to contemplate just how much organic matter they move into their termitaria to support the fungi on which they feed.

On the final day of collecting, I had the opportunity to travel by helicopter to the moist habitat in the closed canopy forest of limestone gorges in the Cheringoma Plateau. In the air just north of Chitengo I could clearly see the many different landscapes of the Park such as the mixed palm/deciduous tree woodlands intermixed with small regions dominated by palm – called palmvelds – and the vast grassland that is the floodplain surrounding Lake Urema, with its contorted network of rivers and whole conventions of crocodiles on the banks, hurriedly splashing into the water after being startled by noise of our passing. In the distance is the leviathanic bulk of the great inselberg that is Mt. Gorongosa, an undulating dark gray silhouette that breaches the flat and featureless horizon. Finally, we approached the heavily forested slopes of the limestone gorges. The forest canopy here appeared thick and dense with varied textures and colors from the crowns of a diversity of tree species. I frequently spotted *Sterculia appendiculata*, the "Tall Star-chestnut" tree, that towered above the canopy line like creatures out of Tolkein's middle-earth: the "tree-folk" or *Ents*, who were shepherds of their forests. Even the Baobabs with their massive silver-gray trunks and limbs did not challenge them. It was not a little shock to my sense of proportion to perceive these trees from above, but then to slowly settle and gently land in the grass and to be small again amid their grandeur.

If one were to return to GNP searching for fungi it would certainly be best to go between December and March, during a normal rainy season. Due to the recent political tensions and conflict, we were unable to visit Mt. Gorongosa. There is a rain forest and particular plants such as *Widdringtonia nodiflora* (Cupressaceae), one of three gymnosperm species in the Park. All three are known only from that location. A survey of fungi in that region and of termite mounds specifically would be a worthwhile venue for another month's work. ■

Acknowledgements:
Thanks to Melinda Peterson, Danny Haelewaters, Teresa Iturriaga, Donald Pfister and Marc Stalmans, Director of Scientific Services at GNP, for reviewing this manuscript.

References

Wilson, E. O. and Naskrecki, P. (2014). A window on eternity: a biologist's walk through Gorongosa National Park. New York, NY. Simon & Schuster.

Greetings, Farewell Best Wishes

Lisa DeCesare has served as an archivist for the Botany Libraries for 19 years, and has been a regular contributor to this newsletter. Those who visited the Farlow have enjoyed meeting her, and benefited from her many informative and creative exhibits. Lisa has formed lasting friendships with many members of the Farlow community. We wish her the best of luck and great success in her future endeavors.

Melinda Peterson has joined our group as Research Lab Coordinator for Don Pfister and Chuck Davis. Melinda comes to us from the Law School. She is supporting the Friends of the Farlow and is also involved in various curatorial projects. When you are in the building please stop in Farlow room 1 and say hello! ■

FoF Book Sale Update

Judith Warnement regrets to report that due to book sale fatigue and a lackluster inventory, the book sale pick list was not generated this year. The income generated from the sale has also declined in the last couple of years, so we welcome your ideas, comments, and book donations! ■

2016 Annual Meeting

The 2016 annual meeting was held on Saturday, November 5th in the HUH Seminar Room at 22 Divinity Avenue, Cambridge.

Our annual lecture featured **Sarah Watkinson** from the Department of Plant Sciences at Oxford University. In "Plumbing the Forest Floor" Sarah discussed her work utilizing genomic approaches to understand the mechanisms of wood decay, particularly related to the brown rot fungi. She explained how nutrients flow through the networks of mycelial cords formed by woodland basidiomycetes (including common saprotrophic and ectomycorrhizal species) and the innovative methods being used to investigate their diverse foraging strategies, physiology and role in forest ecosystem nutrient dynamics. She also presented a video on amino acid flow by her colleague Mark Fricker. Employing this knowledge, Sarah has discovered and patented a non-toxic solution to target control of the dry rot fungus, a forest floor species by origin, which has found a unique and pestilential niche in our buildings.

A reception followed the lecture in the Farlow Library. ■

Visitors & Researchers

Walter Pfliegler, PhD, University of Debrecen, Hungary, May 1 – 31, 2016.

João Araújo, a graduate student from Pennsylvania State University, visited on August 17th. He annotated many of our *Cordyceps* and *Ophiocordyceps* specimens.

Zhang Zhaohui, a bryologist from Guizhou Normal University, Guiyang, China, September 23 – March 22, 2017. He is stdying mosses from limestone and karst areas.

Michal Gorczak, an EU intern and PhD student from Poland who is working on laboubeniales. He will be here until the end of December. ■

New Addition to the Farlow Archives

This past April, thanks to our very own eagle-eyed Jason Karakehian, we were alerted to the sale of some William Gilson Farlow (1844-1919) letters on eBay. We purchased the set of six letters from W. G. Farlow to Fred Carlton Stewart (1868-1946), Botanist and the Head of the Department of Botany of the New York Agricultural Experiment Station at Geneva.

Fred Carlton Stewart was born in Clymer, NY on February 13, 1868. He received his B. S. and M. S. degrees from Iowa State College, before assignment to the Geneva Station in New York, and newly created substation at Jamaica, Long Island in 1894. In his role as mycologist, he attacked the issue of plant disease control. After three years, he resigned from this position and entered graduate study at Cornell University; subsequently, he decided he could accomplish more through European travel and networking with prominent mycologists. In 1898, following a year-long sabbatical for additional training in plant pathology at Cornell and in Europe, Stewart became Botanist and Head of the Department of Botany of the New York Agricultural Experiment Station at Geneva. In 1923, the Station joined the auspices of Cornell University, where Stewart became a Professor of Botany. Despite his duties as a plant pathologist, he also published a handful of papers on edible wild mushrooms and mushroom cultivation. When he retired in 1936, he was granted the distinguished title of Professor Emeritus. Stewart was a pioneer in his work with potato diseases, and the author of many papers on plant pathology, mycology, and allied subjects.

These newly acquired letters, dating from 1894-1896, account for a period when Farlow began to withdraw from teaching, redirecting his students to Roland Thaxter at Harvard. In August of 1896, Stewart asked Farlow for advice about leaving his position at the New York Experimental Station, and how he might parley his qualifications to become a university professor. Farlow responded candidly with trepidation regarding the state of university funding and availability of positions in botany. *"...I must confess I feel considerable anxiety. The condition of money matters and the depression which must follow in university funds and the prospect of young men in getting good places in the near future have worried me a good deal."*

The Farlow and Thaxter archive collections hold original correspondences between Stewart, Farlow and Thaxter. The Farlow collection consists of thousands of letters – 135 bound volumes – the majority of which were written to Farlow and not by him. With this small acquisition, we can learn a considerable amount about the study and practice of botany at the end of the 19th Century from Farlow's perspective.

Please contact botref@oeb.harvard.edu if you have any questions about this, or any other, collection in our archives. ■

Upcoming 2017 Seminar: "Lichens, Biofilms and Stone"

From July 23-29, 2017, Judy Jacob and Michaela Schmull will present "Lichens, Bioflms and Stone" in the Eagle Hill Natural History Science Seminar Series. Lectures will include basic lichen morphology and species identification; biofilm morphology; the role of lichens and biofilms in the environment; basic geology; the history of stone quarrying, finishing, and construction; and the history and contemporary practices of preservation "treatments" for stone. In the laboratory and through various field excursions, students will examine and identify lichens, biofilms, and stones. Furthermore, they will examine the impact of surface manipulation of stone (cutting, polishing, chemical applications) and how these impacts may influence (or not) the growth of lichens and biofilms. Participants to represent a wide variety of disciplines and avocations; prior knowledge of lichens, biofilms, or stone will be useful but not necessary. ■

Teaching, Research & Other Activities

In May, **Don Pfister** joined mycologists from Brazil and Argentenia who taught a short course at the Parque Katalapi, a Biological Station of the Universidad de Concepcion, located near Puerto Montt in northern Patagonia. Don wrote an article, "Fungi and Forests: Shaping landscapes, cuisine and commerce," documenting some of the work being done on fungi in Patagonia. This is in the current issue of *ReVista,* a publication of Harvard's David Rockefeller Center for Latin American Studies. (For the full article, please visit http://revista.drclas.harvard.edu/book/fungi-and-forests-shaping-landscapes-cuisine-and-commerce-0).

Don also published "What a Painfully Interesting Subject: Charles Darwin's Studies of Potato Late Blight" in the October issue of *BioScience*. (The full article can be found at: http://bioscience.oxfordjournals.org/content/early/2016/10/07/biosci.biw114.short?rss=1).

OEB 54 The Biology of Fungi, is currently being offered for the Fall 2016 semester. The students organized the "Fungus Fair" in the Harvard Museum of Natural History on October 15th, a quasi-annual event that is open to the public, where students offer short lectures and demonstrations on various aspects of fungal biology. There was a mushroom display of fungi from the area and a series of tables with demonstrations ranging from yeasts to insect parasites, to coal and its relationship to fungal physiology.

Don was also involved in helping to plan the renovated **Glass Flowers** gallery. A video outlining the activities around the new installation was featured in a May 17th article in the Harvard Gazette: (http://news.harvard.edu/gazette/story/2016/05/putting-the-glass-flowers-in-new-light/0).

Graduate student **Danny Haelewaters** traveled to Panama in July to collect lady beetles in Davíd and bat flies in Gamboa. He presented his *Hesperomyces virescens* research at the 2016 Mycological Society of America Annual Meeting in

Berkeley, CA and at the International Congress of Entomology in Orlando, FL. While in Florida, he collected *Laboulbeniopsis termitarius* and visited the lab of Dr. Matthew E. Smith, a former postdoc in the Pfister Lab. Danny was also awarded a Mycological Society of America Graduate Research Fellowship, in addition to a short-term Research Fellowship from the Smithsonian Tropical Research Institute, for continuation of his work in Panama. Finally, Haelewaters is the primary author on a paper review about parasites of *Harmonia axyridis*, which was published online in *BioControl*. (Abstract available at http://link.springer.com/article/10.1007/s10526-016-9766-8).

Danny has also published the article "Expecting the Unexpected: Looking for Lady Beetles and Their Parasites" (http://link.springer.com/article/10.1007%2Fs10530-016-1077-6) and was co-author of "The Harlequin Ladybird, Harmonia axyridis: Global Perspectives on Invasion History and Ecology" in *Biological Invasions*.

Former undergraduate **Tristan Wang** has published two papers this year, resulting from his work at the Farlow Herbarium. One paper documents the new discovery of Laboulbeniales found on cockroaches in the basement of the Farlow; the second refers to a description of a new species of *Laboulbenia* from Thaxter's collection. Wang is currently a research intern working with Alabama A&M University. ■

New Discovery at the Arnold Arboretum

Work done at the Arnold Arboretum by former Post Doctoral Fellow **Rosanne Healy** and undergraduate **Hannah Zurier** on truffles garnered headlines in several publications.

Healy and Zurier were able to identify a previously unknown truffle associated with trees in the Arnold Arboretum. The truffle was named *Tuber arnoldiana*.

An article appeared in the June 24th *Boston Globe* called "Researchers discovered new types of truffles at Arnold Arboretum." (Available at http://www.bostonglobe.com/metro/2016/06/24/researchers-discovered-new-species-truffle-arboretum-but-they-didn-taste/C2JCzTHw9otFiRY4M0tt6N/story.html)

The September 16th *Harvard Gazette* had another article titled "At the Arboretum, a scientific swerve: Search for famed truffle sends undergrad on path of discovery." (http://news.harvard.edu/gazette/story/2016/09/at-the-arboretum-a-scientific-swerve/?utm_source=SilverpopMailing&utm_medium=email&utm_campaign=09.20.2016%20%281%29).

We continue on various digital projects. These include databasing and imaging the macroalgae and now the microfungi. These projects aim to make collections in US Herbaria and museums accessible to researchers and the public worldwide. In addition to those collections mentioned above, we have scanned and databased macrofungi, lichens and bryophytes. ■

Updates Around the Building

A considerable amount of work continues on the infrastructure of the Farlow building.

Window air conditioners on the first floor were replaced by high tech heat exchange units that have made the rooms much quieter and more uniformly cool.

The herbarium wing of the building has undergone some repair work intent on assuring that that part of the building is weather-tight. All of the window caulking was replaced, and the brownstones were repointed. Missing and broken slate shingles on the roof were replaced.

In addition, three new exterior LED light fixtures have been added to the building, ensuring that the pedestrian walkway and the exterior door are better illuminated than before.

Upgrades to internet cabling is underway. ■




2016 Clara Cummings Walk

Why was it that many of the participants in this year's Clara Cummings Walk barely made it to the site of the former Dover Union Iron Mill, and the view of Boston from Noanet Peak, two of the landmarks for which the Noanet Woodlands are known? Most of us were too busy doing field identifications of our favorite cryptograms, work that generated a list for the Trustees of Reservations that includes: 18 mosses, 2 hepatics, 31 fungi, 3 slime molds, and 61 species of lichens. The "citizen scientists" who generated this first list of cryptogams for the Noanet Woodlands were Deborah Cato, Zaac Chavez, George Davis, Karen Davis, Kenneth Fienberg, Martha Finta, Michelle Kennedy, Elizabeth Kneiper, Mike Prague, Hal Schaefer, Genevieve Tocci, and Tom Walker. Elizabeth Kneiper, the organizer and chief of the walk, was issued the permit for collecting material that needed either microscopic or chemical confirmation.

The list includes species names, authorities, common names and substrates. Common names can vary from field guide to field guide, and therefore are not universally included in a species list (but were submitted by participants and have been included here.

The 2016 Clara Cumming Walk at Noanet Woodlands, Dover MA, June 12, 2016.

The list was generated primarily from field identifications of species noted along the Caryl Park (Red Dot) and Summit (Yellow Dot) trails on June 12, 2016, and a quick preview walk. We hope to post this list and those from previous Clara Cummings walks on the FoF webpage. For a trail map of the Noanet Woodlands, please visit their website at www.thetrustees.org.

If there are any questions regarding this list, please contact Elizabeth Kneiper (Ekneiper@aol.com) ■

Parasites of Parasites: Blood-sucking Bat Flies Infected With Enigmatic Laboulbeniales fungi

By Walter Pfliegler, short-term Farlow Herbarium Research Rellow & Danny Haelewaters, graduate student.

Bats (Chiroptera) sustain highly diverse communities of invertebrate parasites. Two families of flies (Diptera: Nycteribiidae and Streblidae) and one family of bugs (Heteroptera: Polyctenidae) are exclusively ectoparasitic on bats, among a plethora of other insect and mite species. Some of these parasites – several genera of bat flies – are themselves parasitized by ectoparasitic fungi in the order Laboulbeniales (Ascomycota). They were initially described as acanthocephalan worms in Kolenati's article "Epizoa der Nycteribien" (1857), which illustrated how atypical Laboulbeniales fungi are in their morphology and lifestyle.

Roland Thaxter's pioneering and extensive efforts on Laboulbeniales diversity included descriptions of three genera of bat fly-specific species: *Arthrorhynchus* (3 species), *Gloeandromyces* (2), and *Nycteromyces* (1). Only a handful of distribution records are known regarding bat-fly associated Laboulbeniales. These hyperparasitic fungi are ecological and evolutionary enigmas that are rarely encountered by entomologists or mycologists. However, chiropterologists and parasitologists working with bats and bat flies occasionally encounter and easily recognize fly specimens infected with these fungi. The authors of this report were independently approached by enthusiastic and alert zoologists who observed these fungi on Eastern Hemisphere and Neotropical bat flies. This led to the collaboration of our research groups at Harvard University and the University of Debrecen, dealing with evolutionary and ecological aspects of these fungi. Over the past year and a half, in two collecting trips to Panama by Haelewaters and one research visit by Pfliegler to the Farlow Herbarium, 12,000 previously collected bat flies were screened for fungi. This has broadened our knowledge of bat fly-infecting Laboulbeniales.

The screening process involved determining the presence of the fungi on the fly specimens, recording the level of infection (number of thalli), the position(s) of infection (several members of the order are known to display position specificity regarding their infection, although this phenomenon has not been suggested for bat-fly-associated species), and making slide preparations of thalli for species identification. Thalli of highly infected flies were used for DNA isolation and amplification of nuclear ribosomal gene regions using molecular methods developed by our groups.

Arthrorhynchus nycteribiae infection in the form of a dark spot with thalli on the abdomen of a Penicillidia conspicua bat fly. Right: a single thallus.

The host flies included more than a dozen genera collected from several host bat species in Hungary, Romania, Ecuador, Honduras, Mexico, and Trinidad. We found all three genera that infect bat flies. *Nycteromyces* was described by Roland Thaxter and was only known from the holotype specimens. We managed to significantly extend the geographic range and known host species of this genus, and to generate ribosomal DNA sequences

Exceptionally dense infection of Arthrorhynchus nycteribiae thalli on the abdomen of a Penicillidia conspicua fly.

suitable for phylogenetic studies. The genus *Gloeandromyces* was also described by Thaxter over a hundred years ago, with two species that also had not been recorded since their first description. We sampled these two species from several genera of flies both from Central and South America, and discovered three new species. To properly characterize the new fungi and to prepare species descriptions, we compared our specimens to the original holotype slides of the two known species at the Farlow Herbarium. We updated morphological descriptions of these, as well as the new material, using differential interference contrast (DIC) optics and microphotography. Additionally, we managed to generate sequences from three species for phylogenetic studies. *Arthrorhynchus* was finally found on bat flies collected in Hungary and Romania.

Using our previously generated database of Laboulbeniales sequences, we determined the phylogenetic positions of bat fly infecting genera on the phylogenetic tree. Surprisingly, we found the species of three genera independent lineages. One of these lineages, *Gloeandromyces*, falls within the species-rich genus *Stigmatomyces*, which is specific to flies. The other two lineages (*Nycteromyces* and *Arthrorhynchus*) each have a sister relationship to genera that are associated with bugs (Heteroptera). We hypothesized that a host shift from bugs to bat flies has occurred at least twice, which may or may not have been associated with bat-associated bugs.

Although many of these fungi were described in the early 20th century, much about their biology is still unknown. In this project, we seek to unravel evolutionary patterns and ecological traits that may be related to parasitism by Laboulbeniales. These are exciting times; we are only at the cusp of new insights on the ecology, evolutionary driving forces and phylogeny of this diverse order of fungi. ■

One Trichobius joblongi bat fly infected by two different species of Gloeandromyces. Gloeandromyces streblae grows on the wings of this individual (left), while Gloeandromyces sp. nov. seems restricted to the abdomen (right).

Join us!

Receive the FOF Newsletter, notification of the annual book sale, discount on Farlow publications and services, invitations to the annual meeting and other events, and a special welcome when visiting the Farlow.

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