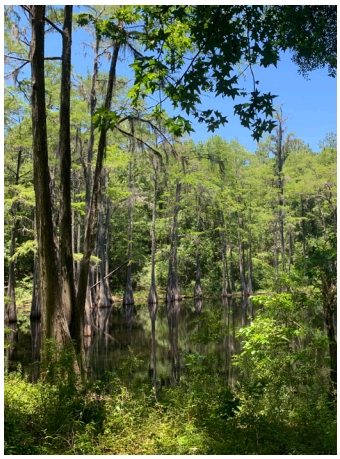
MUSEUM OF NATURAL HISTORY



2020 Annual Report

Auburn University Museum of Natural History

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Auburn University Museum of Natural History

The mission of the Auburn University Museum of Natural History is to document, understand, and preserve biodiversity in order to educate people of all ages about our planet's rich natural history. Our vision is to emerge as the primary repository for all natural history collections currently maintained at Auburn University and to function as a center of excellence for biodiversity research, education, and outreach. We will capitalize on strengths of the biodiversity heritage collections in our care and the vast organismal knowledge base of the curators and staff to establish a gateway through which all segments of society can come discover the natural sciences and appreciate the relevance of biodiversity to human health and quality of life. We will preserve and document the rich natural heritage of Alabama while concurrently creating opportunities for students and teachers from regional schools, the general public, students at Auburn University, and researchers to explore our planet's biodiversity. We seek to inspire an appreciation of nature and the environment so that we might better conserve it for future generations.

Location

AU Museum of Natural History
Biodiversity Learning Center
381 Mell Street
Auburn Unversity

Alabama Natural Heritage Program®
1090 South Donahue Drive
Auburn University, AL 36849

Fax:
AUMNH: (334) 844-9234
ALNHP: (334) 844-5017

Websites

Auburn University
Museum of Natural History:
aumnh.org

www.facebook.com/AUMNH

AL Natural Heritage Program®:
http://www.auburn.edu/cosam/natural_history_museum/alnhp/

Affiliated Websites

NatureServe
www.natureserve.org

From the Director:

Like all institutions, the Auburn University Museum of Natural History was affected by COVID-19. Because we are not a public museum, we have weathered the time far better than a lot of other museums have. Our hearts go out to all of the museum professionals that have lost their jobs or have been furloughed because of this disease. Still, we had to make the transition to work from home and the scuttling of most of our usual outreach programs. Perhaps the hardest hit was that we had to postpone the opening of the museum's first official display for our dinosaur egg. With any luck, we will be able to officially unveil this display in the fall. Despite all of these setbacks, we found new ways to interact with the public.

Outreach and Education Director Toni Bruner was able to come up with a few programs that allowed us to stay relevant. In particular, a lot of us filmed spotlight videos that focused on certain species in the collections or areas out in the field. Toni also filmed segments for an Alabama Mountains to the Gulf program. This program generally brought teachers out into the field to interact with scientists, but this year it went virtual. Toni also obtained funding for making a mobile museum that was completed in 2021, and it is already starting to go on the road.

A lot of the collection managers were able to focus on getting more specimens into the computer database, something that works well from home. Because Auburn University never fully shut down, everyone was still able to get in and process specimens to be placed into the collections and to maintain what we have.

We were even able to get out into the field. The members of the Alabama Natural Heritage Program kept up their busy field schedules studying rare and endangered biota of the southeastern US. We did a lot of field work for a grant to survey reference wetlands for the US Environmental Protection Agency. This grant is to go to some of the best surviving wetlands in the state and determine what species occur there. That way, researchers know what to expect in wetlands of each major type so that we can better understand changes to these important resources as humans encroach upon them. We also completed research on the museum-wide survey of Redstone Arsenal which included a survey of the effects of DDT on small mammals on the base.

2021 is proceeding much the way that 2020 did, but we are slowly opening back up. As more people become vaccinated, the risk of COVID-19 infection goes down and places like ours can offer more programs. This world-wide tragedy has hopefully taught us that we are all in this together. This is not unlike the natural world that we are dependent on and that the museum is tasked with studying. We are reliant on our ecosystems, and the world is a better place when we all can work together to keep as much of nature flourishing as possible.

Jonathan W. Armbruster
Director, Auburn University Museum of Natural History

Primary funders (in alphabetical order)

Alabama Department of Conservation and Natural Resources, Division of Wildlife & Freshwater Fisheries
- Green Salamander modeling and survey
- Mississippi gopher frog survey
- Alabama red-bellied turtle nesting in Alligator nests study
- Indigo snake monitoring and captive breeding
- Green salamander habitat modeling and site verification
- Genetic and Habitat Analyses to Support Recovery Efforts for the Flattened Musk Turtle
- White Fringeless Orchid Modeling
- Habitat modeling of Relict Trillium in Georgia and Alabama
- Statewide Status Assessment of Pondberry
- Whorled sunflower habitat modeling and site verification

Alabama Department of Conservation and Natural Resources, State Lands Division
- Environmental Science and Art - AUMNH Outreach at Wehle

Department of Defense
- Arnold Air Force Base gopher frog survey

Louisiana Department of Wildlife and Fisheries
- Occurrence of Western Chicken Turtle in Louisiana

National Science Foundation
- CAREER: Testing alternative routes of adaptive phenotype-environment matching across heterogenous landscapes in wild populations
- IOS: Effects of electron transport dysfunction on carotenoid ketolation
- Collaborative Research: Red Carotenoids as Signals of Respiratory Chain Function
- Collaborative Research: Documenting marine biodiversity through digitization of invertebrate collections
- DDIG: Copepod Mate Choice
- EPSCoR: Genome to fitness - an analysis of the stress response in *Peromyscus*
- CAREER: Effects of mitohormesis on reproduction and longevity

The Nature Conservancy
- Waterdog and musk turtle eDNA survey

U.S. Army Garrison - Redstone
- Planning Level Survey of Redstone Arsenal for At-risk Species and Ecologically Significant Communities
- Non-native Invasive Plant Species Study

U.S. Environmental Protection Agency
- Establishment of Wetland Reference Sites in Alabama

U.S. Fish and Wildlife Service
- Flattened Musk Turtle and Black Warrior Waterdog Population Study

U.S. Forest Service
- eDNA analysis for Bankhead National Forest

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AUMNH COLLECTIONS

John D. Freeman Herbarium

Accessions/Acquisitions/Exchanges/Loans

The year 2020 hit hard with the COVID-19 pandemic and disrupted the lives and livelihoods of people throughout the world. The Freeman Herbarium was likewise brought to a standstill for several months this year so overall productivity dipped this year. Despite this the herbarium continues to be active and grew this year to house over 82,000 specimens of vascular plants, mosses, liverworts, lichens and fungi from all over the world. Exchange and gift specimens were down this year. We received only 150 lichen specimens on exchange but were able to catch up on some backlog and send out 537 plants and 54 lichens to six other institutions. The multi-year effort to finish accessioning over 2100 sheets of grapevines (the genus *Vitis*) is nearing completion with only several hundred specimens left to georeference. Auburn will have one of the best collections of *Vitis* in the southeastern United States.

Digitization/Database Development

Over 3,000 specimens were glued, processed and added to the Specify database during 2020. These include vascular plants (2,485), mosses (1) and lichens (574). Our vascular plants, lichens, bryophytes and fungi, are searchable online at the AUMNH website (aumnh.org/research-collections/plants/search-plant-database/). Our Alabama vascular plants are searchable on the Alabama Plant Atlas website (www.floraofalabama.org), Morphbank (www.morphbank.net) and SERNEC portal (www.sernecportal.org). Many of our lichen specimens may be searched at the Consortium of North American Lichen Herbaria (www.lichenportal.org/cnalh/) and our bryophyte specimens are on the Consortium of North American Bryophyte Herbaria (https://bryophyteportal.org/

Curtis Hansen and students pressing plants during Curious Curators Camp

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Teaching, Students, & Volunteers

We had a couple of volunteers early in the year before the university shut down, and then again late in 2020 when the campus began to open up again. Volunteers play a critical role in the overall success of managing the Freeman Herbarium. In 2020, we had volunteer help from Colton Seals, and undergraduates Brannan Cliver, Callie McCann, and Areta Dickerson. They all did a job gluing specimens and working through our backlog. Chris Taylor, a grad student, nearly finished the Chewacla State Park Flora project and continued repairing/mounting the historic St. Bernard Collection. Systematic Botany continues to be successfully taught every spring semester with students gaining knowledge and experience in plant

Research & Collections Related Activities

Graduate student Adekola Ouoyemi is doing research with Dr. Leslie Goertzen on the origin of *Eleusine* (Poaceae, Goose grass) in Africa and species distributions throughout the World. He's using herbarium loans and specimen data online to understand the native range of the different species. In addition, he is examining the morphological distinctions among the species.

Marc Johnson, also a graduate student with Dr. Goertzen, is doing a bio-inventory of vascular plant species in the Yates Lake Wildlife Management Area, Elmore County, AL, including collecting specimens for the Freeman Herbarium and using museum resources to identify plants. Marc received museum RA funding this summer to add hundreds of voucher specimens from the Yates Lake project to the herbarium An additional aspect of Marc's dissertation research involves the use of digital herbarium records to build habitat models for the endangered Relict Trillium, *Trillium reliquum*.

During this shortened field season, Curtis Hansen collected specimens from Redstone Arsenal (Madison Co., AL), Cheaha State Park, and Bibb County, Alabama. He has coauthored papers on lichen floristics, a new population of a rare lichen species, and the Chewacla State Park Flora. He also made great strides in processing the historic St. Bernard collections.

The Freeman Herbarium houses historic plant collections from the St. Bernard Herbarium, formerly located in Cullman Co., AL and that were transferred to the herbarium in 1995. These historic and important collections are now getting more attention by being repaired and prepared for incorporation into the main collection.

PhD Student Marc Johnson pressing plants for his research

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Ichthyology Collection

The fish collection continued to grow at a reasonable rate in 2020. In all, the collection added 5,657 voucher specimens in 779 lots. The majority of the newly cataloged material came from older backlog collections made by AUMNH personnel and from Dr. Elise Irwin's lab in the School of Fisheries and Allied Aquaculture. Additionally, we accessioned material collected by natural resources personnel in Florida and Georgia. A significant portion of remote work time was spent georeferencing older collections in the database. As a result, most of the fish collection's Georgia and Alabama holdings now have latitude and longitude associated with their localities. These data add to the value of those collections for researchers and conservation officers searching for distribution records of species in the collection.

The fish collection continued to receive a high volume of loan requests from researchers around the world leading up to the March shutdown. In 2020, the fish collection sent out eight loans the majority of which were domestic. Most of the international loan requests received had to be postponed until 2021 due to difficulties shipping and tracking packages internationally.

The fish collection had four graduate students and multiple undergraduates work on the collections this year. One graduate student, Malorie Hayes, completed her dissertation work, which included research on fishes from three different continents. We additionally completed research on the museum-wide EPA and Redstone survey projects. Altogether, 29 papers were published that cited AUMNH specimens or records.

Ancistrus kellerae, a newly described species of catfish from the Guiana Shield

Yaluwak primus, a newly described genus and species from the Brazilian-Guiana Shield

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Herpetological Collections

The Division of Herpetology continues its efforts to uphold and advance the museum's mission to document, study and educate the public about the biodiversity of Alabama and the world. Division staff, students and associates continue to conduct and promote collection growth and curation, participate in collections-based research, and in facilitate the dissemination of information through scholarly publication and museum outreach.

Accessions/Acquisitions/Exchanges/Loans

Due to the pandemic and the associated restrictions for travel and research, the year was a slow one for growth in the collections. However, there were some contributions as just under 200 amphibian and reptile specimens were accessioned into the various herpetological collections representing a collection growth of close to 0.44%. These specimens included adults and larvae, along with their associated tissues. Additionally, 400 lots of larval amphibians and many hours of frog call recordings were collected as part of the museum's EPA wetland project. Many of the specimens accessioned in 2020 were from graduate student research projects and included brown anoles (*Norops sagrei*) and Eastern Fence Lizards (*Sceloporus undulatus*).

There was also considerable growth in our frozen tissue collection with over 250 tissues and DNA extracts added. These include tissues from our accessioned specimens, as well as some from animals that were photo vouchered and others from animals that were not collected.

Last, we also added over 20 digital vouchers to our photo/audio/video voucher collection. These virtual vouchers help us fill in gaps in our knowledge of Alabama's amphibians and reptiles as well provide important records of sensitive and threatened species.

Although the pandemic did limit museum operations and access, for much of 2020, the herpetology collections continued to be utilized by researchers in 2020. During the year, a total of 5 loans were processed and more than 13 data and tissue requests were processed. We were also happy to host 4 visiting scientists, which utilized the museum's specimens to further their research.

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Digitization/Database Development

The work of digitizing and serving the herpetological databases online continues. The majority of the wet collections are available online through the museum's website. The frozen tissue collection has been organized and is now digitized. It is now being georeferenced, updated, and imported into Specify. Additionally, we continue also to add to and maintain a series of ecological databases that are available online through our website. Last year also saw the beginnings of efforts to photograph the museums specimens. These photographs will be added to our Specify database and be made available online, allowing researchers and the public to see each individual specimen as they search the database.

Teaching, Students, & Volunteers

The Museum's herpetological scientific and teaching collections were also extensively utilized by undergraduate and graduate courses for class use, class projects and individual research. The availability of room 251 as a lab instructional area has allowed several courses to teach their labs within the museum. These include Vertebrate Biodiversity, Herpetology. Not only did students utilized the herpetological teaching collections for lab, several student class projects were based out of the herpetological collections.

A crucial component of our museum family is the group of dedicated and talented volunteers and interns that work with us at the museum. In 2020, the pandemic limited our ability to utilize volunteers in the museum collections, but their efforts were critical to the maintenance and upkeep of the education/outreach live animal collection. Their efforts are invaluable to the mission of the museum, and we look forward to returning to our normal volunteer capacity in the Fall of 2021.

<u>Volunteers</u>
Morgan Hancock
Allison Sharp
Brooke Newell
Sydney Spurlock
Lorrianny Martinez

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Citizen Science

The museum continues to participate in citizen science programs that allow Alabamians to participate in the collection of real scientific data and add to our knowledge base on the calling phenology of our state's frog species as well as the geographic distribution of our herpetofaunal diversity. The AUMNH is home to a chapter of FrogWatch USA, a nation-wide citizen science program where volunteers monitor frog call activity to help conserve amphibians and wetlands. Over the last two years, several volunteer workshops have been held throughout the state. The museum is also home to the Alabama Herp Atlas Project (AHAP), a citizen science program where citizens can send in photo, audio or video documentation of any amphibian or reptile species. These records are curated and added to both our photo voucher catalog our geographic distribution maps for those species. As describe above, over 22 vouchers were accessioned in 2020, several of which represented county records.

Live Animal Collection

Continuing our long tradition, the museum's live animal ambassadors continue to be an indispensable resource for both inhouse events and outreach programs. Additionally, our live animals are utilized by other campus programs and departments, further increasing both the impact of the museum collections and the visibility of the museum. As with everything else in 2020, the number of outreach programs was affected by the pandemic. In all, live animals were utilized in only a few events in 2020 but they are ready and anxious to interact with Alabamians throughout the state in 2021.

Collections manager David Laurencio at the Sustainability Picnic

Research & Collections Related Activities

Herpetology personnel continued producing original collections-based and collections-related research. In 2020, no less than 40 publications were produced related to the herpetological collections were published by museum staff and students. These papers showcase the breadth of research being conducted at the AUMNH.

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Ornithological Collections

Accessions/Acquisitions/Exchanges/Loans

The AUMNH ornithology collection is comprised of approximately 4000 specimens, primarily from the southeastern United States, but also with specimens from throughout North America and a few specimens from Central America, Europe, and Oceania. Approximately half of all specimens are study skins. The collection has approximately 2000 tissue specimens, and about 200 loose feather specimens—among the largest loose feather collections in the world. To complement its research collections, the museum houses a separate avian teaching collection with approximately 300 specimens used in courses such as Ornithology and Natural History of the Vertebrates. This teaching collection was prepared almost entirely by students taking Ornithology.

55 new bird skins were added over the last year. We also added five nests and tissues from 40 birds.

The ornithology collection welcomed one visiting researcher: Dr. Greg Jackson, from Birmingham.

Teaching, Students, & Volunteers

Even during the COVID-19 pandemic the museum had one standout volunteer, Sophie Hirsh. Under Hill's direction, Sophie prepared 55 new bird skins and collected tissue from many of these specimens.

The Museum's Ornithology research and teaching collections were also extensively utilized by undergraduate and graduate courses. These collections were utilized to teach both Vertebrate Biodiversity and Ornithology. Nick Justyn, doctoral student under Hill, used the bird collection extensively in a comparative study of mechanisms of color production.

Digitization/Database Development

The ornithological collections are housed in Specify and are available online. There is more data available for each bird however. These data are located on the hand written specimen tags which are affixed the foot of each specimen. The process of digitizing all remaining data located on the specimen tags continues.

Research & Collections Activities

The avian research collection was used extensively by Nick Justyn in a study of mechanisms of color production. That study will be a chapter in Justyn's dissertation and will be published in a journal.

Mammal Collection

Accessions/Acquisitions/Exchanges/Loans

The AUMNH mammal collection is comprised of just over 5750 specimens, primarily from east-central Alabama. The collection has a focus on insectivores, bats, rodents and carnivores and consists of traditional skin and skull preparations with numerous taxidermy mounts, completed skeletons, fluid-preserved specimens and frozen tissues. Museum specimens are accompanied by standard measurements, such as tail length, mass, and total length, along with information about the collection site and date. To complement its research collections, the museum houses a separate teaching collection used in courses such as Mammalogy and Natural History of the Vertebrates. Although the pandemic did limit use of the collection in 2020, the collection received two data requests.

Teaching, Students, & Volunteers

The Museum's Mammalogy research and teaching collections were also extensively utilized by undergraduate and graduate courses. These collections were utilized to teach both Vertebrate Biodiversity and Mammalogy. Throughout the year, mammal specimens used to teach species identification, skeletal specimens were utilized for a lab assignment, and each lab section toured the museums to learned about the importance of and utilization of the natural history collections.

To aid with research and teaching of Mammalogy in the pandemic, COSAM acquired 3D scanner. Utilizing the scanner, a database representing most of the mammals occurring in Alabama was generated. The database included newly scanned skulls, skulls that were already online, and photographs of skins in our collection.

In fall 2020, the use of this database replaced hands-on use of the educational collections in Mammalogy.

Digitization/Database Development

As with all of the museum collections, the mammal database is in the process of moving over to the Specify platform.

Outreach

The mammal collection (and the curator) were used to provide instruction during the Boy Scout's Merit Badge University.

Mammal skulls

Vertebrate Paleontology Collection

Accessions/Acquisitions/Exchanges/Loans

The vertebrate paleontology collections at Auburn University include close to 2,500 specimens. The collection focuses on the state of Alabama, but also includes significant material from other portions of the southeastern United States. The Vertebrate Paleontology Collection contains Mesozoic and Cenozoic material, both terrestrial and marine, primarily from the Cretaceous period. This includes terrestrial dinosaurs as well as marine groups such as Plesiosaurs and Mososaurs. It also contains important collections of terrestrial mammals form the Cenozoic Era.

Teaching, Students, & Volunteers

Our vertebrate paleontology volunteers continued to provide incredible help in organizing the vertebrate paleontological collection. Through their continuing efforts, the collection is being more accessible and organized and the fossils' housing is being improved.

Volunteers
Claire Wilson
Skye Walker

Invertebrate Paleontology Collection

Accessions/Acquisitions/Exchanges/Loans

The museum's invertebrate paleontological collections were first curated in 2016. The museum is home to a small collection of over 120 invertebrate fossils.

Digitization/Database Development

The invertebrate paleontology database is digitized and awaits preparation and transfer to the Specify platform.

Entomology Collection

Accessions/Acquisitions/Exchanges/Loans

The entomology collection stayed busy during 2020 cleaning up the database and entering new specimen records. We had one loan with 71 specimens and six data requests. Approximately 5,000 insects were pinned. We added two additional county records for the Southeastern Blueberry Bee, Habropoda laboriosa. Additionally, we recorded only the third Alabama specimen of a large mason bee, *Osmia chalybea.* This is a thistle specialist, yet was collected on blackberry!

Digitization/Database Development

To date, we have almost 211,000 insects and 11,000 arachnids and myriapods digitized. The data are available through iDigBio: http://ipt.idigbio.org/resource?r=aum-entomology. They are also published on GBIF and SCAN Bugs.

Collections Manager, Melissa Callahan, teaching students about insects

Teaching, Students, & Volunteers

We are thankful for all of the students working and volunteering in the entomology collection this year. They worked diligently prepping specimens, digitizing, and helping with outreach events!

Alan Jeon

Chloe Kaczvinsky

Charles Stephen

Jordan Sykes

Melissa taught the Auburn Montessori School about arachnids and insects in February 2020. Due to COVID-19, we shifted our events to include a video tour of the entomology collection, and specimen spotlights on social media.

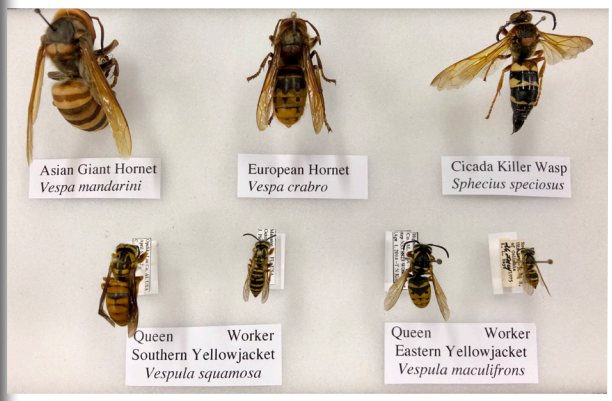


Photo for social media showing the most common insects confused with the Murder Hornet.

Invertebrate Collection

Accessions/Acquisitions/Exchanges/Loans

The Invertebrate Collection saw continued growth in 2020, adding over 2200 lots to the collection. The specimens included representatives of all major invertebrate taxa. Most of the specimens came from Dr. Halanych's previous Antarctic research cruises. Additionally, other projects in the lab of Dr. Nathan Whelan have contributed pleurocerid specimens.

Digitization/Database Development

We digitized over 2200 lots in 2020, as the invertebrate collection is continually digitized as it is accessioned. Over 200 lots were geo-referenced, and all the information is added to our Specify database. Each specimen is given a barcode for more efficient tracking. The data should soon be available online at the AUMNH website, and shared with GBIF, iDigBio, InvertEBase, as well as other online sources. To date, we have over 21,000 molluscan invertebrates and over 10,000 non-molluscan invertebrates digitized.

Research & Collections Related Activities

Expedition to the Antarctic – RVIB Nathaniel B. Palmer. – Collections of Marine Antarctic Invertebrates.

The Curator of Marine Invertebrates, Dr. Kenneth M. Halanych and the Marine and Aquatic Invertebrates Collections Manager, Nusrat Noor embarked on a 14-week expedition to the Antarctic from September to December of 2020 with a team of 20 scientists from multiple institutions including, Auburn University, University of Alabama, Central Michigan University, and University of Alaska Anchorage. They sampled marine invertebrates living in some of the most remote oceans in the world. The specimens are currently on their way back to the States so they can be accessioned into the museum and used for research and educational purposes.

Teaching, Students, & Volunteers

The invertebrate collection has benefitted from the amazing students and volunteers who have worked on specimen collection, upkeep, accessioning, digitization, and outreach. We are grateful for all of their contributions.

Hannah Meede (Undergraduate, DBS)

Caitlyn Redak (Graduate, DBS)

AUMNH Research Projects

Redstone Arsenal Planning Level Surveys

Redstone Arsenal contracted with ALNHP and AUMNH to conduct planning-level surveys to document the precise number of species of conservation concern on the installation. Surveys focused on areas designated as Ecologically Sensitive Areas but also included other areas of suitable habitat for rare species. Surveys completed in 2020 and focused on caves, fishes, and plants.

Insect and arachnid surveys were completed in 2019, although much of the lab work to identify specimens and add them to museum collections was completed in 2020. Cave fauna collected throughout the subterranean surveys were also identified and added to the collections. Several additional fish surveys were completed in 2020 and more individuals of the state protected Tuscumbia darter *Etheostoma tuscumbia* were caught, identified, and released. This species is a Tennessee River endemic, and it appears to be doing well in several spring and spring-fed systems on the Arsenal.

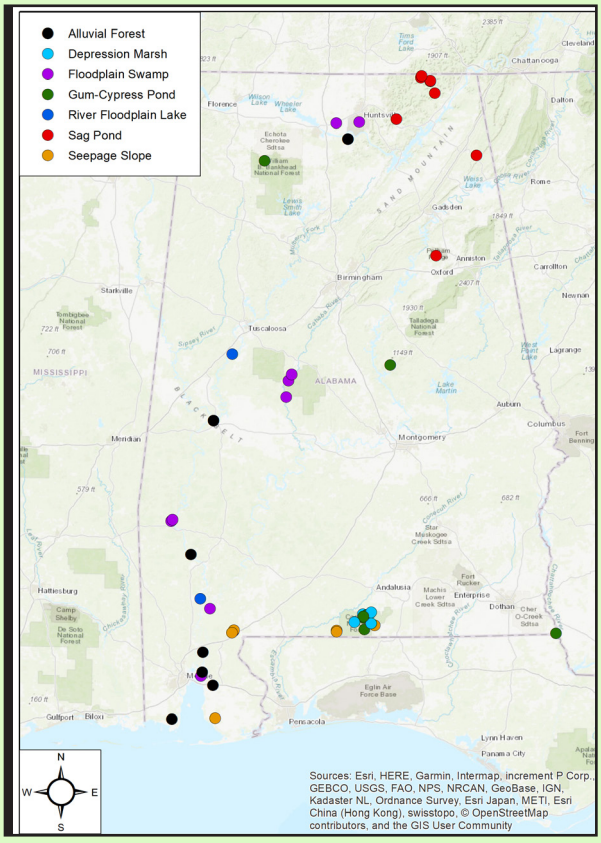
Additional plant surveys were also completed in 2020 for a more species-specific inventory. Numerous specimens were collected for incorporation into the herbarium's collections as well. Photo-vouchers were taken of even more plant specimens by Dr. Les Goertzen and those records will also be included in the final report.

The rare plant and natural community component of the Redstone Biological Assessment was finalized and included as part of a comprehensive report submitted to the military in May 2021. The study consisted of two components: 1) updating existing and documenting new occurrences of taxa monitored by ALNHP; and 2) classifying and documenting occurrences of significant ecological associations. Seventeen species of rare plants represented by 19 occurrences were documented, including 10 previously unrecorded taxa. Augmenting the rare plant surveys, documentation of ecological communities entailed gathering data on vegetation structure and species diversity followed by cross-walking field information with the National Vegetation Classification ecological system framework to determine a proper identity of a plant association. Five existing occurrences of natural communities were updated and 21 new records were generated as part of the study.

All field work for this project concluded in 2020 and a draft of the final report was submitted to the Department of Defense at Redstone Arsenal in May 2021.

Reference Wetlands Study

In 2018, the Environmental Protection Agency awarded AUMNH/ALNHP a grant to conduct a reference wetland study. The Museum has partnered with Troy University to accomplish the study, whose primary goal is to enhance recognition and protection of wetlands throughout Alabama by establishing permanent wetland reference sites across the state using an Ecological Integrity Assessment framework. This framework developed by NatureServe and the Natural Heritage Network, was designed to support planning and management for the conservation of wetlands and other natural communities through quantifying ecological integrity based on metrics of biotic and abiotic condition, size, and landscape context. Reference wetland sites will provide a standard against which to measure the condition of similar wetland types – a starting point for establishing desired future conditions to inform land management and conservation efforts. Wetland integrity will be evaluated focusing on intensive field assessments of flora and fauna. These assessments will include collecting data to support a floristic quality assessment and indices of biotic integrity for faunal taxa.



Map of high quality wetlands from which reference sites will be selected.

The final product is anticipated to complement and strengthen the state's ability to implement a comprehensive water quality monitoring and wetlands assessment program by providing baseline data to fill information gaps. The collection of vegetation data has begun, and will be part of the deliverables that include GIS data and maps, a database of completed field assessment forms, and hard and electronic copy of final report summarizing the project results. This information can be used for setting conservation priorities, identifying restoration strategies, and monitoring the effectiveness of conservation actions.

Much of the field survey work for this project was completed in 2020, including many fish, bird, macroinvertebrate, herpetological, and plant surveys. The project is anticipated to be completed by December 2021.

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ALABAMA NATURAL HERITAGE PROGRAM[SM]

The mission of the Alabama Natural Heritage Program[SM] (ALNHP) is to provide the best available scientific information on the biological diversity of Alabama to guide conservation action and promote sound stewardship practices. ALNHP is administered by the Auburn University Muesum of Natural History, Department of Biological Science. Established by The Nature Conservancy in 1989, it is one of a network of such programs across the United States, Canada, and Latin America, collectively known as the Natural Heritage Network (NHN). As a member of the NHN, ALNHP is represented by its membership organization NatureServe. NatureServe works to aggregate data from individual Network Programs and is dedicated to the furtherance of the Network and the application of Heritage data to biodiversity conservation.

Natural Heritage Programs have three broad functions:

- to collect information on the status and distribution of species and natural communities,
- to manage this information in a standardized way, and
- to disseminate this information to a wide array of users.

Natural Heritage Programs use a standardized information management system to track biodiversity data including taxonomy, distribution, population trends, condition, and viability. ALNHP provides the following services: biodiversity data management, inventory, biological monitoring, conservation planning, Geographic Information System services, and land management expertise.

NatureServe
A Network Connecting Science With Conservation

NatureServe is a non-profit conservation organization that provides the scientific information and tools needed to help guide effective conservation action.

NatureServe represents an international network of biological inventories - known as natural heritage programs or conservation data centers - operating in all 50 U.S. states, Canada, Latin America and the Caribbean. NatureServe and its network of natural heritage programs are the leading source for information about rare and endangered species and threatened ecosystems.Together we not only collect and manage detailed local information on plants, animals, and ecosystems, but develop information products, data management tools, and conservation services to help meet local, national, and global conservation needs. The objective scientific information about species and ecosystems developed by NatureServe is used by all sectors of society - conservation groups, government agencies, corporations, academia, and the public - to make informed decisions about managing our natural resources.

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Project Summaries

Black Warrior waterdog and flattened musk turtle eDNA survey of Locust Fork

The Locust Fork of the Upper Black Warrior River basin supports a suite of federally listed aquatic species including snails, fish, and mussels, plus an amphibian and reptile. The U.S. Fish and Wildlife Service, Alabama Department of Conservation and Natural Resources, and Geological Survey of Alabama have prioritized watersheds in the state to focus management and conservation actions for aquatic species restoration and recovery. These watersheds have been designated Strategic Habitat Units (SHUs) and the Locust Fork is one of the recognized SHUs.

The Black Warrior waterdog (*Necturus alabamensis*) and the flattened musk turtle (*Sternotherus depressus*) are endemic to the Upper Black Warrior River, are ecologically linked by habitat, and are federally listed. Based on recent survey work using eDNA and conventional sampling outside of the streams within the Bankhead National Forest, the Locust Fork appears to have the best remaining populations of these species. This project, as part of a larger project funded by the National Fish and Wildlife Foundation to The Nature Conservancy, is also a collaboration with the Black WArrior Riverkeeper, and Tangles Bank Conservation. The objective is to survey the entire free-flowing reach of the Locust Fork for both species using eDNA. Results from these surveys will be used in a larger landscape and water quality analysis to provide guidance for habitat restoration and watershed conservation.

So far, water samples have been taken along the Locust Fork with a spacing interval of 1.5 km for eDNA analysis during warm season and cool season to encompass the activity period of both species. Total warm season sample stream distance was 175 km and total cool season stream distance was 193 km. Lab analysis has been completed for the warm season samples with 13 Flattened Musk Turtle site detections; cool season samples have not been analyzed. Ultimate lab analysis will be with meta-barcoding on a suite of imperiled species of the river to include the target species, fish, and mussels.

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Occurrence of Western Chicken Turtle in Louisiana

The Western Chicken Turtle (*Deirochelys reticularia miaria*), is distributed west of the Mississippi River in Arkansas, Louisiana, Missouri, and Texas. Records for the Chicken Turtle in Louisiana are distributed through much of the state exclusive of the toe east of the Mississippi River; subspecies east of the Mississippi River is the nominate D. r. reticularia. In the Louisiana Wildlife Action Plan this Western Chicken Turtle is in the Tier I category, those species most in need of immediate conservation action. Chicken Turtles inhabit wetlands and use surrounding upland habitats for nesting and overwintering. Threats reported in others states include conversion of bottomland hardwood and cypress swamps to agriculture (Trauth et al. 2004; Buhlmann et al. 2008).

In this project, we help the state address survey needs for the Western Chicken Turtle in Louisiana as laid out by the State Wildlife Action Plan (SWAP). We expect to provide baseline data on the presence and habitat preference of the Chicken Turtle and demonstrate the use of environmental DNA (eDNA) as a means to monitor the species in the future. Objectives are 1) to determine current occurrence and distribution of the Western Chicken Turtle in Louisiana; 2) demonstrate the use of environmental DNA (eDNA) as a monitoring tool for Western Chicken Turtle and 3) identify potentially suitable habitat through species distribution modeling and quantify anthropogenic threats to available habitat.

The Western Chicken Turtle has been reported from approximately 60 historic localities. These historic localities have been examined with most having been severely altered, degraded, or destroyed. The next step in potential pond location was to search for ponds using GIS on large tracts of public land (US Forest Service, Wildlife Management Areas) and contact resource managers regarding potential ponds, additionally historic Southern Crawfish Frog localities were included to expand upon the potential pond list. This approach produced a list of approximately 45 ponds. About 2/3 of these ponds have been sampled for eDNA during spring and summer seasons. Once eDNA analysis has been completed any ponds with positive detection with be turtle trapped in the upcoming year.

Chicken Turtle

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We will conduct site visits to selected localities based upon accessibility, collect water samples for eDNA analysis, and sample for Western Chicken Turtles with visual wading and trapping. Trap sets will be baited hoop nets, with or without leads, box traps, and crawfish traps depending upon water depth and wetland type being sampled. All species of turtles captured will be sexed, weighed, measured, marginally notched for unique identification, and a tissue sample taken. Chicken Turtles are active from March through September (Buhlmann et al. 2008; Carr and Tolson 2017) therefore we will target our eDNA collections during this time as activity of the target species may increase eDNA detection probability (de Souza et al. 2016).

Geospatial modeling and analysis approaches will be used to identify likely areas of occurrence and predict future loss of habitat in Louisiana due to land use change. Ryberg et al. (2017) used Maximum Entropy modeling (Maxent; Phillips et al. 2006; Phillips and Dudik 2008), a type of species distribution modeling (SDM) to map potentially suitable habitats for the Western Chicken Turtle across its range. Spatial scale, sampling bias, data inputs, and settings can all affect SDM model results (Elith et al. 2011; Merow et al. 2013), therefore we will build a boosted regression tree SDM (Death 2007) to identify agreement with Maxent. Our surveys will provide a level of validation of the SDM results. We will follow the methods outlined by Ryberg et al. (2017) to quantify current and future threats to Western Chicken Turtle habitat in Louisiana. Sites will be surveyed more than once using multiple techniques, therefore we will also be able to estimate detectability and model occupancy (Nichols et al. 2008; Peterman et al. 2013).

In 2020, initial site visits occurred in Louisiana to identify suitable sites for eDNA sampling and turtle trapping. An initial habitat suitability model was also constructed for Louisiana using the methods of Ryberg et al. (2017). Additional models and future threats analyses will be completed in 2021 and 2022.

Based on our trapping and eDNA, and habitat model results we will provide an update on the current predicted distribution of the Western Chicken Turtle, and information on turtle species assemblages affiliated with Western Chicken Turtle wetlands.

This project has been amended to include eDNA analysis for the Southern Crawfish Frog and Tiger Salamander which utilize similar and at times the same pond type as the Western Chicken Turtle. Project will continue through September 2022.

Southern Crawfish Frog

Green Salamander Habitat Suitability and Status Survey

The green salamander (*Aneides aeneus*) in Alabama is distributed across the Appalachian and Cumberland Plateaus where it occupies rocky bluffs and outcrops within hardwood forests. Intact forest cover provides shade, moderates temperatures, and maintains humid conditions favorable to the terrestrial salamander. Removal or disturbance of forest cover and/or alteration of rocky habitat disrupt the micro-climate and this is thought to be a factor in population decline (Mount 1975; Cline 2004; Elliott 2008).

Photo: J.D. Wilson

The Center for Biological Diversity petitioned the US Fish and Wildlife Service to evaluate the species' status in 2012. Information on the status of the species in Alabama is needed by the Alabama Department of Conservation and Natural Resources and the USFWS to guide decisions pertaining to conservation and listing. The combined approach of using species-specific habitat models to guide survey efforts has led to the discovery of new occurrences or populations for some threatened salamander species (Apodaca et al. 2012; Peterman et al. 2013). Our use of these methods may be effective for more thoroughly assessing the status and range of green salamander in Alabama.

The objectives of the study were to (1) develop a habitat suitability model for the green salamander (Aneides aeneus) in Alabama, and (2) use the results to guide surveys to field truth and collect data on the salamander to better understand the status and conservation needs for the species.

A preliminary habitat suitability model was developed to guide survey efforts. We gathered 184 presence locations to train and test this model. Data were from a variety of sources including museum collections, iNaturalist observations, Alabama Natural Heritage Program data, and personal observations from collaborating scientists. Environmental predictors were selected based on known habitat preferences and included land use, geology, soil characteristics, topography, and hydrology.

All environmental predictor data had a 30-meter resolution or were resampled to a 30-meter resolution. These layers were all processed and clipped to the modeling extent, North Alabama, using ArcGIS Pro 2.4.1 (ESRI, Redlands, CA). The Maximum Entropy algorithm (MaxEnt; Phillips et al. 2006) has been shown to perform well, particularly when modeling rare species with few presence locations and no true absence data (Elith et al. 2011). Therefore, these layers, along with the input presence data were processed using the Maximum Entropy (MaxEnt) algorithm.

Model outputs include a probability surface where cell values range from 0 (low probability of occurrence) to 1 (high probability of occurrence). After the model was developed, the probability surface and survey data were added to ArcGIS Pro version 2.4 (Figure 1).

The distribution of habitat suitability probabilities for sites with green

salamanders absent vs. present. Due to unequal sample sizes, a Wilcoxon Rank-Sum Test was used to determine whether probabilities of habitat suitability were significantly different between sample sites where green salamanders were absent vs. where they were present. All maps were constructed in ArcGIS Pro version 2.4 (ESRI; Redlands, CA) and all statistical analyses were conducted in RStudio version 1.1.456.

Survey sites were then selected across a range of low-high probabilities with most sites falling into the medium and high categories. Since public lands are more easily accessible, land ownership, and site access in terms of proximity to roads were also considered during site selection.

Model performance was high (AUC=0.927) and variables that contributed most to the model included mean temperature, geology, amount of forest, and amount of agriculture.

Of the 138 unique sites that were surveyed, green salamanders were detected at 34 of the sites, and no green salamanders were detected at the remaining 104 sites. The boxplot shows that sites with green salamanders present generally had higher probabilities of habitat suitability compared to locations where they were absent (Figure 2). When comparing modeled habitat suitability probabilities for these sites, the mean probability for sites with green salamanders present (mean = 0.58) was significantly higher than for sites without green salamanders (mean = 0.45; $P < 0.01$).

After surveys were completed, the additional presence data were combined with the original 184 locations, resulting in 245 presence locations. The same settings and environmental data were used to create a new MaxEnt model.

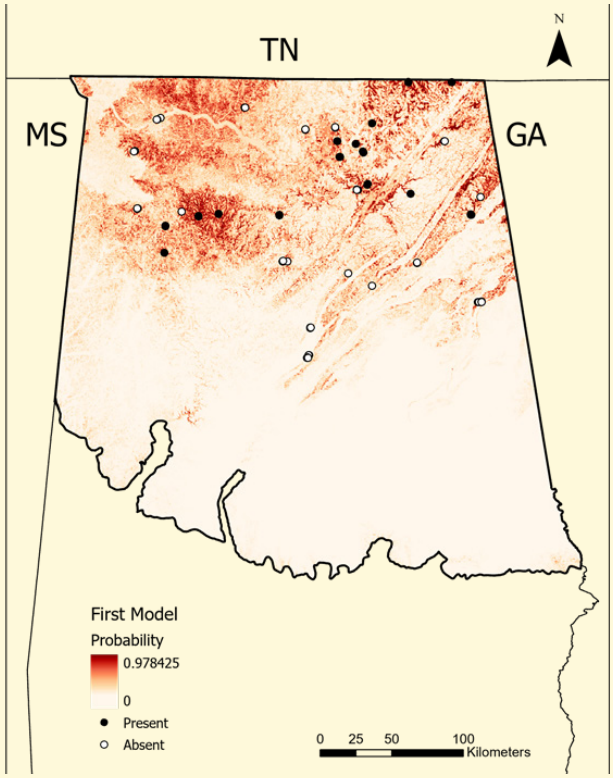

Figure 1. Habitat suitability map for the Green Salamander (Aneides aeneus) in Alabama.

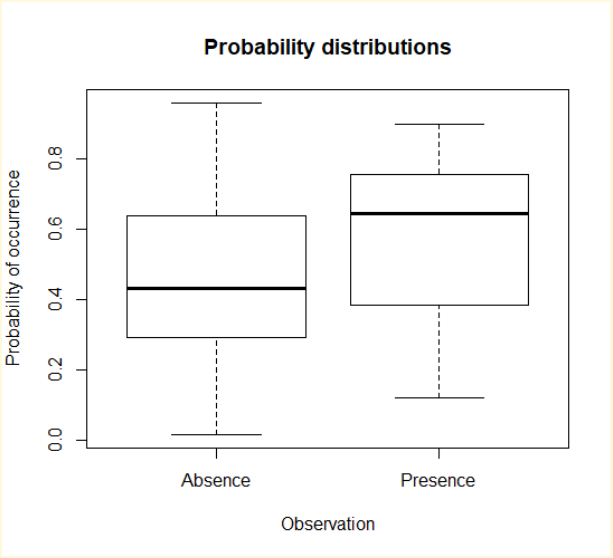

Figure 2. Probability of habitat suitability comparing presence and absence of the Green Salamander.

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Thirty-two to 34 sites were surveyed; 32 sites for habitat, tree data, and green salamander occupancy plus two additional for green salamander occupancy only. Seven species of salamanders (*Aneides aeneus, Desmognathus conanti, Eurycea cirrigera, Eurycea longicauda, Eurycea lucifuga, Plethodon glutinosus, Pseudotriton ruber*) were observed during the surveys. Salamanders (any species) were observed at 30 sites, green salamanders were observed at 15 sites.

A total of 549 individual salamanders of all species were tallied during the surveys. Green salamander was the second most observed species per site and total number (0.44 relative frequency) after the slimy salamander (*Plethodon glutinosus)* (0.74 relative frequency). Slimy salamander was, numerically, the most frequently observed species with the green salamander second in numbers observed (0.30 and 0.18 relative frequency, respectively).

Sites sampled were based on habitat model suitability results and included those within the known range of the green salamander plus sites outlying the known range. Sites sampled were within the Appalachian Plateau, Ridge and Valley, and Tennessee Valley Physiographic Units. Based on the green salamander map in Mount (1975) and Physiographic Unit map in Guyer (2015) Appalachian Plateau and Ridge and Valley are the primary and secondary physiographic units of importance regarding the distribution of the species in Alabama (Table 1).

Counties with multiple physiographic units and green salamander occurrences, green salamander localities lie within the Appalachian Plateau. Counties lacking records (Mount 1975) within Appalachian Plateau physiographic unit are Jefferson, Blount, Cullman, Fayette, Walker and represent a gap in green salamander distribution in Alabama. Major surface geology associated with green salamander observations were limestone, sandstone, and shale and associated with the Appalachian Plateau physiographic unit.

Table 1. Physiographic Units and counties overlying range of green salamander in Alabama.

Physiographic Unit	County	*A. aeneus* records
Appalachian Plateau	Blount, Cullman	no
Appalachian Plateau	Cullman	no
Appalachian Plateau (Ridge & Valley)	DeKalb	yes*
Appalachian Plateau (Ridge & Valley)	Jefferson	no
Appalachian Plateau	Jackson, Marshall, Winston	yes
Appalachian Plateau (TN Valley)	Colbert, Madison	yes*
Appalachian Plateau (Fall Line Hills, Moulton Valley)	Franklin, Lawrence	yes*
Ridge & Valley	Calhoun, Etowah	no
Ridge &Valley	Cherokee	yes
Tennessee Valley	Limestone	no
Ridge & Valley (Appalachian Plateau)	St. Clair	yes*

*In Appalachian Plateau

Data collected at each site was categorized by rock crevice, tree species, and salamander survey. Sites with rock outcroppings were chosen to be sampled and data taken at each sample location included date, latitude and longitude, and time of survey if searching for salamanders.

Rock crevice data collected at each sample site included rock face orientation (aspect in degrees), crevice distance above ground (cm), crevice maximum interior height (cm), crevice maximum width (cm), and crevice maximum depth (cm) (Table 2). Corresponding tree data was taken within a 10m x 10m plot at each crevice sample site and included species and diameter at breast height (dbh), plus distance to the rock face (Table 3). Salamander surveys were conducted at each rock crevice-tree data sample location and data collected included beginning and ending time, species identification of each salamander observed and age class of each green salamander observed, substrate occupied by salamander, if concealed within a crevice or exposed, surface temperature (C), air temperature (C), wind speed (mph), relative humidity (%), and dew point.

Average aspect, direction of rock face, for sites without green salamander observations was 194o, or approximately south. Average aspect for sites with green salamander observations was 157 degrees, or approximately southeast. Sites with green salamanders had crevices with average height, width, and depth greater than sites lacking green salamander observations (Table 2).

No significant differences were found with average number of tree species per plot, average number of trees per plot, average DBH of trees, or average distance of trees to rock face between plots sampled at sites with and without green salamander occupancy (Table 3).

Table 2. Rock face crevice data for sites with and without green salamander observations.

Green salamander occupancy	Average no. crevic-es per plot	Average aspect of rock face (degrees)	Average distance above ground of crevice (cm)	Average maximum crevice height (cm)	Average maximum crevice width (cm)	Average maximum crevice depth (cm)
No	28.8	194	91.2	8.5	38.2	11.6
Yes	25.9	157	98.9	10.9	44.8	14.8

Table 3. Adjacent forest cover data for sites with and without green salamander observations.

Green salamander occupancy	No. total tree taxa across plots	Average no. tree species per plot	Average no. trees per plot	Average DBH of trees (cm)	Average tree dis-tance to rock face (m)
No	34	8.1	27.7	19.8	4.4
Yes	26	7.5	27.7	21.7	4.6

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Escambia Map Turtle

There are more than 400 "at-risk" species in the southeast US currently petitioned for federal listing under the Endangered Species Act (ESA). Both Alabama and Florida contain more of these at-risk species than any other southeastern US state (NatureServe Central Databases January 2012). In response to a mega-petition filed by the Center for Biological Diversity to evaluate the need for listing of these approximate 400 aquatic species, the USFWS Southeast Region has implemented an at-risk species conservation strategy to work proactively with public and private partners to conserve these species over the next decade with the goal of eliminating the need to list these species under the ESA (http://www.fws.gov/southeast/candidateconservation/). One of the thirteen reptile species in the petition is the Escambia map turtle (*Graptemys ernsti*), a freshwater species found only in Alabama and Florida in the Conecuh-Escambia, Yellow, and Choctawhatchee-Pea rivers. Current information on the status of the Escambia map turtle in Alabama and Florida is limited; it is a State Wildlife Action Plan Priority 2 species, thus up-to-date information is needed for ADCNR in formulating conservation strategies for this species and for the USFWS decision regarding listing. The objective of the study is to obtain information on the current distribution and abundance of the Escambia map turtle in Alabama and assess the conservation status of the species.

Map turtles as a group are noted for their drainage-specific endemism and well-developed basking behavior. Abundance and distribution data on basking map turtles are easily gathered with visual survey methods. Suitable riverine stretches, those with an abundance of dead wood for basking, were previously sampled in 2000 and 2002 with supplemental data collected in 2012. Data on turtles were collected through basking surveys by identifying turtles; all observed individuals were identified to species, and when possible, sex, and age class. Distinguishing sexual characters between male and female include 1) females achieving a much larger size, 285 mm carapace length, males ca. 170 mm carapace length; 2) females with conspicuously large head; 3) males with long and enlarged tail. Basking turtles of large size and enlarged head were categorized as "female," turtles of smaller size with a noticeably large tail were categorized as "male," turtles without distinguishable characters were categorized as "unclassifiable," these individuals may have been immature female, immature male, or adult male in which the tail could not be observed. Data were recorded per river kilometer, date, and river stretch to allow for a metric of turtles/river kilometer to be calculated for each sampling session.

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In this study, selected river stretches in the Conecuh River from the 2000 surveys have been resurveyed, additionally sections not surveyed in 2000 were included in 2018. Surveys were conducted from either canoe or a 14' flat bottom boat equipped with a 25 hp outboard motor. Water depth was the primary determinant in the mode of transportation. Use of a motorized boat allowed both up- and downstream surveys while with a canoe only downstream surveys could be performed. Image stabilization binoculars were used to spot, identify, and sex basking turtles. Survey end points and survey length were recorded with a GPS. In smaller rivers, those with channel width of less than 75 m, both banks could be scanned for turtles. In large river channels, over 75 m wide, only one bank per survey effort could be sampled effectively.

During 2018, all surveys were conducted in the Conecuh River in Covington and Escambia counties. All surveys took place below Point A dam downstream to Brewton except for one section in the flowing reach above Gantt Lake. Surveys in 2000 also included Conecuh River, Sepulga River and Patsaliga Creek (Conecuh River drainage) and Yellow River (Figure 1).

In 2000, total survey distance in the Conecuh River was 45.7 river kilometers with an average of 12.3 Graptemys ernsti/ river km. The total survey distance in the Conecuh River in 2018 was 139.3 river km with an average of 7.9 Graptemys ernsti/river km. During both survey years, G. ernsti was the most commonly seen turtle in the Conecuh River. G. ernsti were observed at a higher density (12.3 vs. 7.9), in 2000 than in 2018. Underlying cause for difference in observed numbers is unknown at this time. River conditions in 2000 were at normal summer flow,while in 2018 water levels were higher. In 2018, many normally exposed bask sites were inundated forcing turtles to use basking structures that were often within thickets of riparian trees. In both survey years, females were observed more often than males or individuals that could not be assigned to an age/size class. Smaller individuals are more difficult to observe than larger females in the occluded bask site conditions. Additional surveys for Escambia map turtle will be conducted in 2019.

This project is being funded by the Alabama Department of Conservation and Natural Resources.

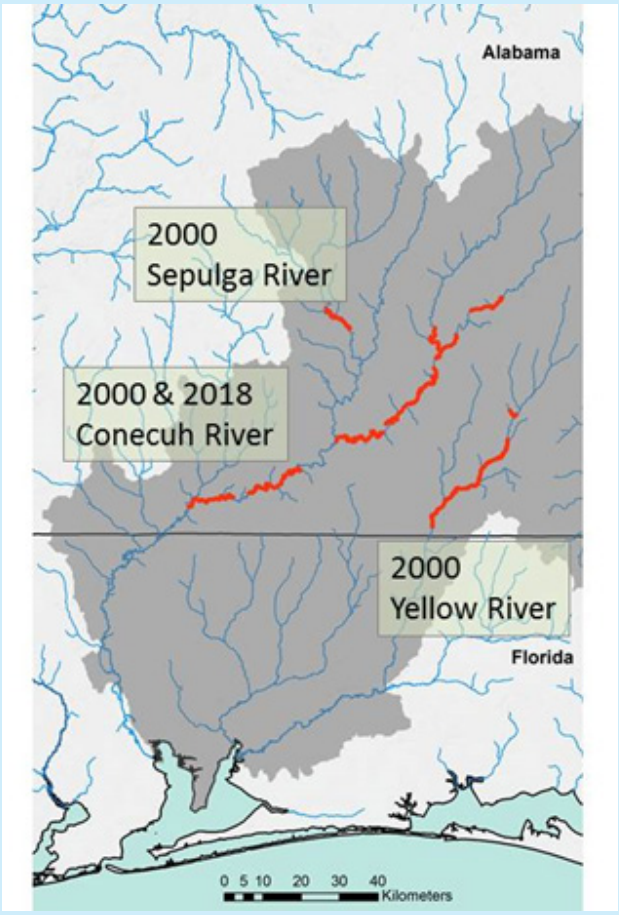

Figure 1. Surveyed river reaches.

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Investigation of Alabama Red-bellied Turtle Nesting in American Alligator Nests

The Alabama red-bellied turtle (*Pseudemys alabamensis*) is a federally endangered freshwater turtle inhabiting the lower rivers of coastal Alabama and Mississippi, a region with dense American alligator populations. Although listed under the U.S. Endangered Species Act the Alabama red-bellied turtle remains poorly studied (Lovich and Ennen 2013) particularly with respect to nesting and hatchling survival. Most of the information available on nesting and nest success is from studies and observations by Nelson (2009) in Alabama and Floyd (1995) in Mississippi. Nelson et al. (2009) studied Alabama red-bellied turtle nesting activity at four sites in the Mobile-Tensaw Delta, the spoil deposit on the north end of Gravine Island, and the Mobile Bay Causeway, Big Island, and Meaher State Park. In Mississippi nest sites have been described from three locations (Floyd 1995). In both states all nest sites described and studied were anthropogenically altered, thus a data gap exists in reporting nesting in natural settings by the Alabama red-bellied turtle.

In 2018, 40 alligator nests were located in the lower reaches of MTD (C. Murray pers, comm.). Alligator nest construction and oviposition in the Mobile-Tensaw Delta (MTD) occurs in July and hatching occurs in early September (C. Murray pers. comm.). Nelson et al. (2009) reported nesting activity of the Alabama red-bellied turtle from early May to September, peaking in June to July which coincides with alligator nesting activity. Murray reported that alligator nests in the MTD are constructed some distance from open water, and Alabama red-bellied turtle nest distance from water has been reported as 30-123 m (Nelson et al. 2009), and up to 100 m (Floyd 1995), distances from water that overlap that for alligator nests.

In Florida, the closely related Florida red-bellied turtle (*Pseudemys nelsoni*) nests both in well-drained soils near freshwater habitats and alligator nests (Goodwin and Marion 1977; Deitz and Jackson 1979; Kushlan and Kushlan 1980; Hunt 1987; Enge et al. 2000). Godwin (2002) suggested that a natural nest site for the Alabama red-bellied turtle is alligator nests but this possible nesting medium has yet to be investigated.

Murray (pers. comm.) states that the alligator population in MTD > 3,000 individuals thus the availability of alligator nests as nest habitat is present. Godwin (unpubl. data) has observed long, shallow scars on carapaces and plastrons of female Alabama red-bellied turtles suggesting that alligator-turtle

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interactions are not unusual and may be occurring at alligator nesting locations.

The need to be addressed in this study is to examine alligator nests as natural nesting substrate for the Alabama red-bellied turtle and, if found, study the success of Alabama red-bellied turtle nests.

The objectives of the study are to investigate the use of alligator nests by Alabama red-bellied turtle as a natural nest site and, if used, the hatching success of turtle nests within alligator nests.

Since the majority of known nest sites of the Alabama red-bellied turtle are anthropogenically-altered locations, natural nest sites need to be located and studied. One expected result is to document alligator nests as natural nest sites for the Alabama red-bellied turtle and determine nest success. This work benefits the natural history and conservation of the Alabama red-bellied turtle by providing critical nesting data, a direct metric of population persistence at the life history stage most susceptible to mortality. Information for resource managers and direction for future research relevant to conservation of the Alabama red-bellied turtle will be provided (e.g. sex ratio studies of hatchling turtles, satellite telemetry of female turtles to locate nest sites, etc.).

This project will be conducted in collaboration with Dr. Chris Murray (Southeastern Louisiana University). An initial alligator nest survey will be made in early July 2021 as part of an on-going study on alligator demography in the MTD carried out by Murray and ADCNR. Pertaining to the turtle study described herein, data collected on each alligator nest will include: date, latitude and longitude, distance from water, female in attendance, nest width, and nest height. Vegetation of alligator nests will be carefully excavated to search for the presence of turtle eggs without disrupting the viability of alligator eggs (protocol from Murray et al. 2013).

A second survey of the previously located alligator nests will be made in September 2021 to collect data on turtle nest condition, success, and turtle species identification. Other turtle species syntopic with the Alabama red-bellied turtle include river cooter (*Pseudemys concinna*), slider (*Trachemys scripta*), Florida softshell (*Apalone ferox*), alligator snapping turtle (*Macrochelys temminckii)* and common snapping turtle (C*helydra serpentina*) and identification will be either examination of hatchling(s) or with genetic analysis of eggshell remains. If hatched or depredated, all eggshells will be collected for a genetic analysis. Genetic samples will be provided to the Tangled Bank Conservation lab for analysis to assign species affiliations of hatched or depredated nests.

This project, funded by ADCNR, is underway and will be performed in collaboration with Dr. Chris Murray from Southeastern Louisiana University.

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Habitat Modeling and Site Verification for the Whorled Sunflower in Alabama

Whorled sunflower is an endemic of the southeastern U.S. recently listed as threatened under the Endangered Species Act, an action sanctioned by the U.S. Fish and Wildlife Service based on the species' small number of occurrences, a low reproductive capacity, and various threats that include habitat degradation. During 2020 the ALNHP and AUMNH implemented a habitat suitability modeling study across Alabama in an effort to locate additional occurrences of the species. While no new occurrences were discovered, several sites containing promising habitat, including prairies and herb-dominated wetlands, were observed.

The objective of the project was to promote conservation of the whorled sunflower in Alabama by determining the rate of success in locating new occurrences of the taxon based on species occupancy models. The study also focused on evaluating environmental conditions and assessing reproductive potential (e.g., census of reproductively active plants) in extant occurrences, thus enabling land managers and conservationists to prioritize sites for long-term protection. The study was sponsored by ADCNR

Range-wide Status Assessment of Ravine Sedge (*Carex impressinervia*)

Ravine sedge is a globally imperiled species currently known from less than 25 occurrences in four southeastern states. The plant prefers forested ravines often just upslope of drainage courses. An herbaceous evergreen perennial, the species was described in 1987 by Charles Bryson, Robert Kral, and James Manhart based on specimens collected near Centreville in Bibb County, Alabama. Because of a low number of occurrences, the U.S. Fish and Wildlife Service has commissioned the AUMNH/ALNHP to update existing records for known occurrences across the range of the species, assessing habitat integrity and threats that will enable land managers to develop appropriate conservation strategies. The final report will be completed in 2022.

Photo by Keith Bradley

Eastern Indigo Snake

The Eastern Indigo Snake was once an important species, and apex predator, of the longleaf pine ecosystem of south Alabama; it is now presumed extirpated from the state with the exception of the released snakes in Conecuh National Forest. The return of the Eastern Indigo Snake as an ecological element of the longleaf pine and associated ecosystems of southern Alabama is being done through a reintroduction of the species. Captive breeding is the most efficient route to acquire the number of young snakes needed for this endeavor, and a concerted release effort at one site is in progress for the establishment of a viable population.

Conecuh National Forest (CNF) is situated within the historic range of the Eastern Indigo Snake and has been selected as the initial reintroduction site for several reasons: (1) the U.S. Forest Service has undertaken a progressive longleaf restoration project; (2) CNF possesses the habitat heterogeneity needed for Eastern Indigo Snakes, which includes the presence of gopher tortoises; (3) and CNF is well placed in the GCPEP Apalachicola-Blackwater River State Forest-Conecuh National Forest-Eglin Air Force Base corridor.

Our objective is to establish a self-sustaining population of Eastern Indigo Snakes at Conecuh National Forest by introducing 30 snakes/year to reach 300 snakes released. Therefore, we are collecting data through monitoring to assess survivorship, sex ratio, and, ultimately, demographics of the population of indigo snakes. We collect monitoring data by walking transects within suitable habitat and inspecting refuges (i.e. gopher tortoise burrows) in the fall and winter (October-March) when snakes are reproductively active.

Twenty-two snakes were released at on 7 May 2020. Sex ratio of released snakes was equal with 11 males and 11 females. All snakes were hatched in 2018, thus approximately 1 2/3 years old. Six male snakes and five female snakes had been housed at the Welaka National Fish Hatchery (WNFH) during the previous year. Five male snakes and six female snakes were reared at the OCIC. All snakes were captive hatched at OCIC and held until approximately one year of age before transport to the WNFH for housing. Snakes were released at or in natural refugia, i.e. gopher tortoise burrow.

Male snout-vent lengths (SVL) ranged from 1166 to 1381 mm with an average of 1259 mm. Female SVLs ranged from 1185 to 1362 mm with an average of 1262 mm. Male mass ranged from 976 to 1535 g with an average of 1244 g. Female mass ranged from 1029 to 1536 with an average of 1227 g.

To assess survivorship, sex ratio, and, ultimately, demographics of the population of Eastern Indigo Snakes we

are conducting pedestrian surveys for indigo snakes and sampling vertebrate assemblages (amphibian, reptile, bird, small mammal) with drift fence arrays.

We have continued to use drift fence arrays and box traps to trap Eastern Indigo Snakes in the area from January-October in each year of the project. Another important monitoring technique involves walking transects within suitable habitat and inspecting refuges (i.e. gopher tortoise burrows) in the winter (January-April) when snakes are reproductively active. Transect search effort will be standardized by search time, distance travelled and person-hours.

Eighteen drift fence arrays were open from 10 March to 26 September to trap Eastern Indigo Snakes. Drift fence traps were operational for a total of 144 to 189 out of a potential of 204 days, or 73 to 93% of time available. Traps were operational for a total of 2,964 trap-days. While Eastern Indigo Snakes (9 capture events of 8 individuals) were captured in drift fences the result was quite low, 0.003 per trap-day, but this provided data on growth and movement that otherwise would have been lacking. Thirteen species of snakes were captured in drift fences. Eastern Indigo Snakes were captured at a higher frequency than five of the 13 species (Table 1).

Standardized visual searches were conducted between 10 December 2019 and 14 March 2020 at 12 discrete sites. Sites were sampled from one to 44 times for a total of 84 surveys and 148.7 man-hours (Table 2). Site selection was either the original release site, Nellie Pond release site, or clusters of gopher tortoise burrows. With each survey location, date, time, observer(s), refuge type, and evidence of activity was recorded. The type of refuge (tortoise burrow, armadillo burrow, or stump hole) encountered was tallied. Snake evidence categories were snake, snake track on a tortoise burrow apron, or shed skin. Eastern Indigo Snakes were observed 18 times, all at Nellie Pond. Other snake species observed were Black Racer (Coluber constrictor) and Florida Pinesnake (Pituophis melanoleucus). All snakes encountered were captured, identified, and measured.

Six individual snakes were recaptured for a total of 18* recapture events. Curiously, all snakes captured were females. Five snakes recaptured were from the 2019 release and two were from the 2018 release. One snake was recaptured seven times, one five times, three recaptured twice, and one recaptured once. (Snakes recaptured more than once were generally NOT handled after the first capture in order to minimize disturbance.) (Table 3).

Snakes when recaptured were scanned for a PIT tag, weighed, and measured. Male snakes had an average growth rate of -3.71 g/day and female snakes 0.87 g/day. Male snakes had an average growth rate of 2.57 mm/day for SVL and female snakes an average growth rate of 1.41 mm/day. The average distance between capture points for male snakes was 0. 5 km and for female snakes 0.47 km.

A new method for monitoring using trail cameras was also introduced. Ten Reconyx HyperFire PC800 cameras were placed on trees facing Gopher Tortoise burrows at Nellie Pond. The cameras were assigned randomly to different burrows every seven (7) days. As motion capture is insufficient for capturing images of smaller or slow-moving animals, cameras were programmed for time-lapse,

Table 1. Drift fence snake captures from 10 March to 26 September 2020.

Species	Total captured	CPUE	Relative frequency
Black Racer (*Coluber constrictor*)	57	0.019	0.46
Eastern Coachwhip (*Coluber flagellum*)	13	0.004	0.10
Florida Pinesnake (*Melanoleucus lodingi*)	12	0.004	0.10
Copperhead (*Agkistrodon contortrix*)	11	0.004	0.09
Eastern Indigo Snake (*Drymarchon couperi*)	9	0.003	0.07
Florida Cottonmouth (*Agkistrodon piscivorous*)	5	0.002	0.04
Eastern Diamond-backed Rattlesnake (*Crotalus adamanteus*)	5	0.002	0.04
Gray Ratsnake (*Pantherophis obsoletus*)	5	0.002	0.04
Banded Watersnake (*Nerodia fasciata*)	4	0.001	0.03
Eastern Mud Snake (*Farancia abacura*)	2	0.001	0.02

taking pictures every 30 seconds from sunrise to sunset. Batteries and SD cards were replaced every 7 days, and all photos were inventoried on a computer. Monitoring in this manner allowed for daily, all-day monitoring during the snakes' active periods, filling the gaps when pedestrian surveys were not feasible or not conducted. They also allow for un-intrusive monitoring of the species. The only limitation of this technique is the inability to identify individual snakes; however, this information can be supplemented by other techniques. Cameras were placed between 22 and 25 Nov 2019 and from 19 Jan to 14 Apr 2020. The cameras yielded approximately 1.17 million photos taken, of which 1,916 featured one or more indigo snakes (0.02%). Of 111 burrows observed, indigo snake activity was recorded at eight of them. It took anywhere from 1 – 2 hours to go through a week's worth of photos at each burrow, for an approximate total of 250 person hours. Typically one snake was observed per burrow, but up to four individual indigo snakes were observed at one burrow. Mating behavior was also observed at the same burrow on 30 Jan, for a total of 7 frames, or 3.5 minutes. Snakes were observed anywhere from 1 frame (30 seconds) to 193 frames (1 hour, 36 minutes, 30 seconds), although that was rare. While it is impossible to discern individuals on the cameras, it is estimated that seven individuals were observed on camera. Snakes typically were observed basking near their burrows or slithering off-screen and later returning. If observed more than once, they often emerged and re-entered the burrow around the same time each day.

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To document the effect upon the prey base by the introduction of the Eastern Indigo Snake. Benefits from this study will aid in formulating predictions of changes in amphibian, reptile, and small mammal communities at additional release sites within CNF, and in selecting future release sites of Eastern Indigo Snakes at other locations outside CNF.

Eleven species of frogs and toads, two species of salamanders, five species of lizards, 13 species of snakes, nine species of birds, and, and eight species of mammals were captured. (Table 2).

This project is funded with a State Wildlife Grant through the Alabama Department of Conservation and Natural Resources.

Table 2. Total captures from drift fence results by category for taxa, individuals per taxa, and relative frequency of captures for each category.

Category	Taxa	Total Captures	Relative Frequency
Anurans	10	145	0.34
Salamanders	2	4	0.01
Lizards	3	93	0.06
Snakes	11	125	0.29
Turtles	2	5	0.01
Birds	6	25	0.06
Mammals	7	93	0.22
Total	41	424	

Statewide Status Assessment of Pondberry (*Lindera melissifolia*)

Pondberry (*Lindera melissifolia*) is one of several southeastern Coastal Plain plant species that has become globally imperiled as an artifact of adverse modifications of its habitats: margins of naturally occurring ponds and depressions embedded in a matrix of pine and mixed pine-hardwood uplands. The taxon is a diecious clonal species currently represented by approximately 25 extant occurrences across seven southeastern states, several of which are small consisting of only either male or female individuals, severely limiting reproduction potential. The small size of many occurrences, restricted reproduction capabilities, and its inherently relatively narrow ecological niche serve as a testament to the conservation need the species now faces. This study focuses on surveying for new occurrences in Alabama and providing a general ecological assessment of sites visited. The project is sponsored through Section 6 funding with a completion date of December 2021.

Gopher Frog Environmental DNA (eDNA) Survey at Arnold Air Force Base

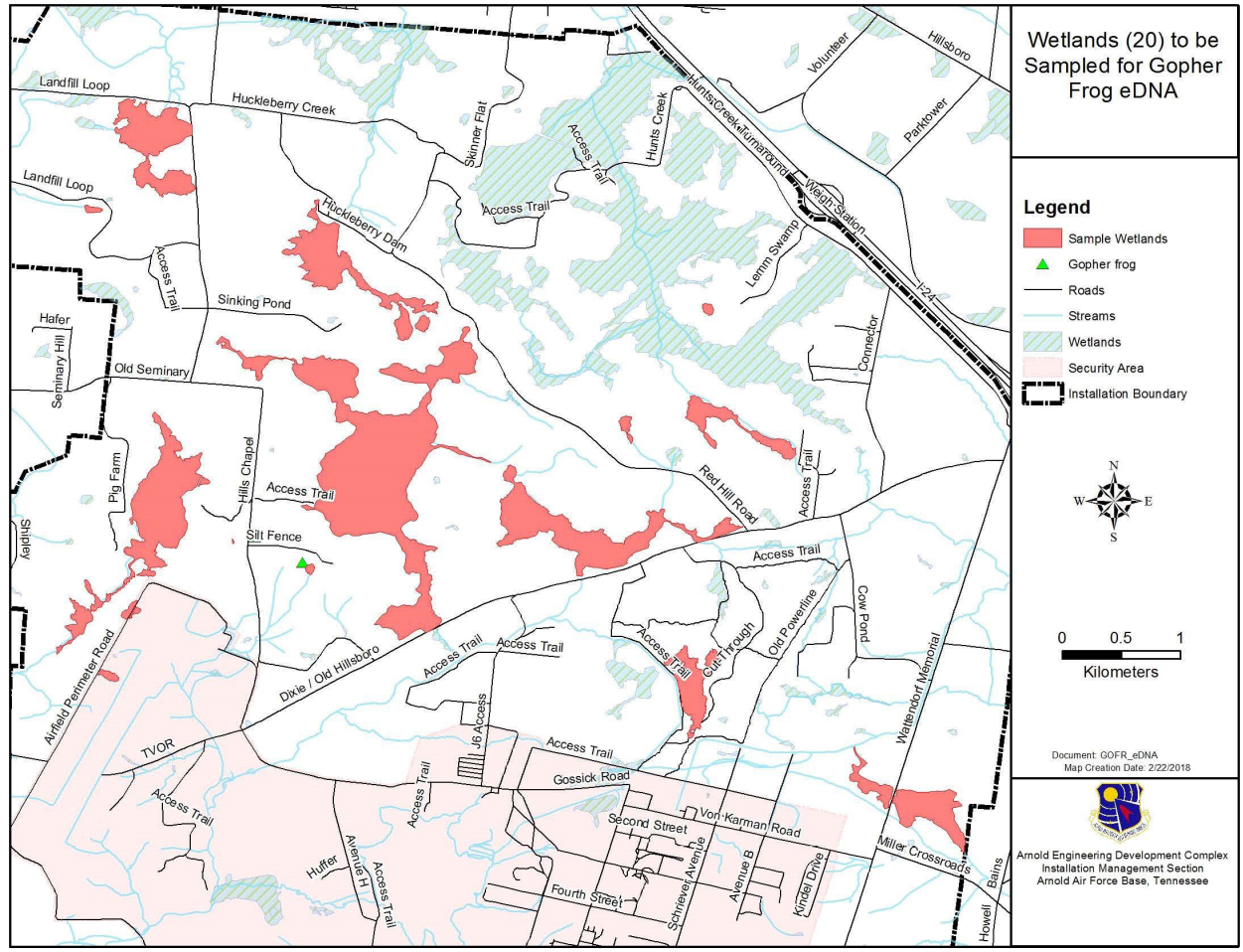
The gopher frog (*Lithobates capito*), formerly *Rana capito*, is under review for listing by the U.S. Fish and Wildlife Service. Primarily an inhabitant of the Coastal Plain, the gopher frog is a winter breeding species using ephemeral ponds that typically dry during summer months and fill with fall and winter rains. Metamorphic frogs exit ponds in early summer. Outside of the Coastal Plain, one historic population was documented in the Ridge and Valley of Alabama plus a second approximately 100 km north at Arnold Air Force Base (AFB) in Tennessee. The presence of the gopher frog at Arnold AFB is an anomaly, yet disjunct populations outside of the Coastal Plain have been reported, for example one in Shelby Co., AL. Two individuals, one a gravid female, were collected at Arnold AFB in the 1990s, yet no breeding ponds have been located despite intensive sampling efforts which have included night-time and automated aural surveys for calling males, egg mass and tadpole surveys, and drift fence trapping.

Environmental DNA (eDNA) is an effective detection tool for rare species that can be used when conventional sampling fails. In Conecuh National Forest in south Alabama eDNA has been used to detect gopher frogs at known and undocumented breeding sites. In the ponds at Arnold Air Force Base we will be using eDNA to sample for gopher frog presence. Approximately 20 wetlands on Arnold AFB have been identified as potential breeding sites for the gopher frog and from this list 12 have been selected as ponds to be sampled.

The gopher frog, if present, likely occurs in low density on Arnold AFB based on the collection of only two adult specimens, and presumably the adult breeding and tadpole developmental period is similar to that of other gopher frog populations from January to May. Sample collection during the activity period of the target species is critical to the detection of species with eDNA. We began water sample collections in December and continued through May when tadpoles were expected to be in the ponds and eDNA concentrations to be elevated. Approximalte 180 samples were collected over the winter and spring. All water samples were negative for gopher frog eDNA.

The second goal of this project is to determine which population the two gopher frogs captured from Arnold AFB in the late 1990s are nearest to genetically. Because these frogs have been preserved in formalin, new challenges must be overcome to successfully extract DNA from their tissues. Once that is done, we will compare the ultraconserved elements of the Arnold AFB gopher frogs to corresponding DNA segments from gopher frog tissues collected from southeastern US populations.

Photo by Kevin Enge



Map of 20 wetlands on Arnold Air Force Base in Tennessee that will be sampled for Gopher Frog eDNA

Genetic analysis of the formalin preserved specimens from Arnold Air Force Base was initiated. DNA was successfully extracted from the specimens and DNA samples from other gopher frog populations were obtained for comparative purposes. The impact of COVID-19 delayed progress with the lab analysis.

This project is funded through the Department of Defense.

Mississippi Gopher Frog

The Mississippi gopher frog (*Lithobates (Rana) sevosa*), is currently known to have extant populations only in southeastern Mississippi, yet historically ranged from southern Louisiana to southwest Alabama. This gopher frog is a state protected Priority 1 species, federally endangered, and likely historically ranged into Alabama in Mobile and Washington counties (Mount 1975; Bailey and Means 2004). One recovery criterion is to have a metapopulation in eastern Mississippi or Alabama (USFWS 2015), therefore, surveys are needed to determine if extant populations of the Mississippi gopher frog are present in Alabama. If so, conservation and management actions should be implemented for the species. Objectives of the study are to perform a habitat modeling analysis to guide search efforts for potential pond and upland habitat and to survey ephemeral ponds in Mobile and Washington counties for the presence of the Mississippi gopher frog.

The Mississippi gopher frog is an inhabitant of longleaf pine sandhills ecosystem, often associated with the presence of the gopher tortoise, and breeds in fishless, isolated ponds. Potential ponds will be visited and surveyed for adults, calling males, and with eDNA. The use of GIS will be the first step to locate potential ponds. Potential ponds will likely be natural depression ponds surrounded with suitable gopher tortoise soils and/or known gopher tortoise localities. Once ponds are identified landowners will be contacted for permission to sample for the presence of the Mississippi gopher frog.

Surveys will be during the breeding period and done with visual surveys for adult frogs and egg masses. Ponds will be surveyed a minimum of two times with water samples being collected for an environmental DNA (eDNA) analysis. From each pond a minimum of three liters of water will be collected with water collections spatially distributed to provide good representation of the pond. Water will be filtered through a cellulose nitrate filter with a pore size of 1.0µm to 5.0µm and stored in a vial with the DNA buffering agent CTAB. Upon completion of filtering, the filter will be removed and placed in a vial containing CTAB. In the lab, DNA analysis, using qPCR protocol, will be performed.

Captive rearing of the Mississippi Gopher Frog has been done at the Sandhill Crane National Wildlife Refuge in Jackson County, MS. Water samples were obtained in May 2014 from cattle tanks occupied by late stage and metamorphosing L. sevosus, filtered, and stored in CTAB. These samples were collected to provide positive L. sevosus eDNA controls and tested positive for eDNA detection indicating that molecular primers using the ND2 primer from McKee et al. (2015) are effective to detect L. sevosus DNA.

The number of presence locations used to train a habitat suitability model can have substantial effects on model output (Sofaer et al. 2019). Since the Dusky Gopher Frog *Lithobates sevosus* is so rare, presence locations used for the model includes observations of both L. sevosus and its congener, Gopher Frog Lithobates capito (*L. sevosus* data provided by the U.S. Fish and Wildlife service and the Louisiana Natural Heritage Program). These two species have very similar life histories and habitat requirements and preferences (USFWS 2015).

Environmental predictors were selected based on known habitat preferences, and included land use, soil characteristics, topography, hydrologic features, and precipitation.

We used MaxEnt software version 3.4.1 for the gopher frog due to our lack of absence data, and documentation of superior performance for similar modeling scenarios (Elith et al. 2011). Model outputs include a probability surface where cell values range from 0 (low probability of occurrence) to 1 (high probability of occurrence). After the model was developed, the probability surface was added to ArcGIS Pro version 2.4 in order to further examine areas of higher probability (ESRI; Redlands, CA).

Model performance was very high, as the training AUC was 0.988 and the test data AUC was 0.994. Longleaf pine forest availability, average March precipitation (30-year normal), and percentage of clay in the soil (0-100cm depth) were the most important predictor variables (Table 1). Probability values in the preliminary model ranged from 0 to 0.964 (Figure 1).

Figure 1. Preliminary habitat suitability model for the Mississippi Gopher Frog (*Lithobates sevosus*).

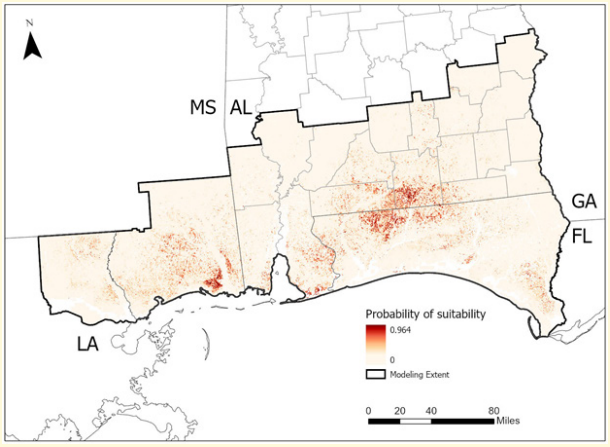


Table 1. Estimates of the relative contributions of the environmental predictor variables to the MaxEnt models.

Variable	Percent contribution	Permutation importance
Longleaf Pine	32	18.6
Avg Precipitation - March	21.4	13
Percent Clay in Soil	10.7	28.9
Dist to Breeding Pond	9.3	4.4
Canopy Cover	5.7	6
Dist to Developed Land	5.1	9.2
Flooding Frequency	4	8.5
Drainage Class	3.8	1.1
Avg Precipitation- January	3	1.2
Avg Precipitation-February	2.8	3.2
Relative Elevation	0.9	1.1
Soil Slope	0.8	4
Ponding Frequency	0.3	0.2
Depth to Water Table	0.2	0.4
Percent Sand in Soil	0	0

A preliminary field survey was conducted early March in Mobile and Washington counties to assess accessibility of potential sites. Sites selected were six ponds listed as HIGH from Bailey (2009

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All ponds are located on private lands and were not accessed at this time. An additional potential pond, also on private land, was found in southern Washington County. More potential ponds have been identified by examining the habitat suitability model (Figures 2, 3). Public county records were searched for landowner information but no landowners were contacted due to normal services having been disrupted by COVID. Discussions are underway with Alabama Department of Conservation and Natural Resources, U.S. Fish and Wildlife Service, Longleaf Alliance, and other groups seeking information for additional potential ponds and landowner access.

Figure 2. Highlighted area in Mobile County with high probability of Mississippi Gopher Frog habitat. Arrow points to the specific site that may support a suitable pond.

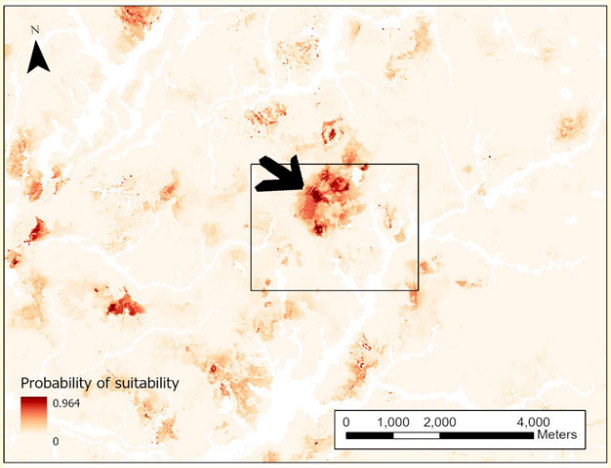

Figure 3. Aerial view of an area in Mobile County with high probability of Mississippi Gopher Frog habitat. Arrow points to the specific site with a potential pond.

Habitat Modeling and Site Verification for the White Fringeless Orchid in Alabama

The white fringeless orchid is one of many southern Appalachian plant species that are listed under the Endangered Species Act, for which suitable habitat is likely plentiful but remains unverified. The species assumes a relatively broad but sporadic distribution across the mountainous region of seven southeastern states. The plant was listed as a federally threatened species in October 2016 based on a small number of occurrences, a low reproductive capacity, and various threats that include habitat degradation (alteration, fragmentation, succession, and forest management practices) as well as direct damage to individual plants. During 2020 the ALNHP and AUMNH implemented a habitat suitability modeling study across Alabama in an effort to locate additional occurrences of the species. Four new occurrences were discovered with several sites supporting optimal conditions, but which no plants were found.

The objective of the project was to promote conservation of the white fringeless orchid in Alabama by determining the rate of success in locating new occurrences of the taxon based on species occupancy models. The study also focused on evaluating environmental conditions and assessing reproductive potential (e.g., census of reproductively active plants) in extant occurrences, thus enabling land managers and conservationists to prioritize sites for long-term protection. The study was sponsored by ADCNR through Section 6 funding.

A paper will be submitted for potential publication in the peer-reviewed journal Southeastern Naturalist highlighting the outcome of the study.

Noxious weed mapping and monitoring at Redstone Arsenal

In 2020 the Alabama Natural Heritage Program partnered with Redstone Arsenal to conduct a non-native invasive species mapping and monitoring initiative to foster a greater understanding how invasive plants are impacting natural areas on the installation. The goal of the proposed study will be to assist RSA natural resource managers in preparing successful treatment plans to control weed infestations. Through mapping and monitoring efforts, resource managers will also acquire a greater understanding in predicting those areas potentially subject to weed invasion, understand the biology of the invasion process and determine means by which weeds spread, and to assess the biological and economic impact of weed invasion. Three parcels have been selected where infestations were identified and marked in the field using GPS technology, and delineated as either points or polygons depending on size. Survey data has begun to be generated into maps denoting the identity, distribution, and size of each infestation through use of symbols to denote percent cover and codes for indicating weed species. Monitoring has also begun, which entails repetitive surveys to track weed populations over time. Permanent plots have been placed within select infestations where a 50-meter transect has been established, supplemented with quadrats at 3-meter intervals to calculate the frequency and density of a given species within each plot. The study is expected to be completed by the end of 2022.

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Flattened Musk Turtle and Black Warrior Waterdog Population Study

The flattened musk turtle (*Sternotherus depressus*) and the Black Warrior waterdog (*Necturus alabamensis*) are two listed species endemic to the Cumberland Plateau physiographic province of the upper Black Warrior River watershed in Alabama, a designated strategic habitat unit (SHU). The flattened musk turtle was listed as threatened in 1987 and the Black Warrior waterdog was listed as endangered in 2018. In light of a continued decline in the flattened musk turtle's and Black Warrior waterdog's status, an analysis of conservation actions and alternatives was identified as a critical need and a working group was established to facilitate such an endeavor by involving State, Federal, academic, and non-governmental organization participation.

Discussions between the Alabama Department of Conservation and Natural Resources, Auburn University, Turtle Survival Alliance, and US Fish and Wildlife Service began to move forward to identify key needs and conservation actions. Building a captive population for propagation purposes was identified as a priority, however before such an activity can be initiated, a controlled propagation plan will need to be developed. This would require the assurance that stable, wild populations will not be negatively impacted if individuals are removed, either temporarily or permanently.

The best remaining populations of the flattened musk turtle and Black Warrior waterdog reside in Sipsey Fork and Brushy Creek in the Bankhead National Forest. Therefore, the demographics of these populations need to be understood to help inform conservation alternative analyses, status assessments, and guide captive propagation efforts if deemed appropriate. The need is to conduct a mark-recapture study on populations of these two species to determine their population stabilities.

This project presents a prime opportunity to educate a new generation of conservation professionals. By coordinating through local conservation associations, we will identify and hire student interns to assist in field work in order to nurture their passion in conservation and develop the expertise needed to begin their career in the field. The component will ensure the next generation of conservation leaders are from backgrounds that represent the intrinsic diversity of the public we serve.

Objectives of the study are to assess the population status of the flattened musk turtle and Black Warrior waterdog in Sipsey Fork and Brushy Creek. This study will initiate a long-term mark-recapture study and provide needed data toward assessing conservation alternatives and decision making for both species, facilitate the development of a controlled propagation plan, and provide requisite information to conduct a species status assessment for the flattened musk turtle. This project will educate a new generation of professionals through a student internship program focused on traditionally underrepresented groups that will promote diverse conservation community that is able to meet the challenges of the future. Expected results are to provide data and analyses on the population demographics and status of the flattened musk turtle and Black Warrior waterdog in Bankhead National Forest and develop baseline data for longer-term mark-recapture studies.

This project is in the early stages of field collection of data and is funded through the US Fish and Wildlife Service.

Genetic & Habitat Analyses to Support Recovery Efforts for Flattened Musk Turtle

The flattened musk turtle (*Sternotherus depressus*) is a small, aquatic, freshwater turtle endemic to the Upper Black Warrior River drainage of northwestern Alabama (Figure 1).

Since receiving a federal listing status of threatened in 1987, studies indicate it has continued to decline throughout the range in spite of federal protection. Studies performed after federal listing have shown that the flattened musk turtle has been extirpated from approximately 56% of previously occupied sites, about 37% of historic populations have declined, with only 7% populations stable and in relatively pristine habitat.

The plight of the flattened musk turtle has come to the attention of the Turtle Conservation Coalition. In their recent publication of "Turtles in Trouble the World's 25+ Most Endangered Tortoises and Freshwater Turtles – 2018," the flattened musk turtle is one of the three species of the United States included on the list. Listed as Priority 2 - "High Conservation Concern" in the Alabama State Wildlife Action Plan (AL SWAP, p. 16) and "threatened" under the federal Endangered Species Act, this project will begin to address answers needed to establish an assurance colony to prevent extirpations and preserve genetic diversity.

The Flattened Musk Turtle USFWS 5-year review and Species Status Assessment will be completed concurrently with this project; which will inform and augment those processes. The long-term need is to establish an assurance colony to preserve genetic diversity and provide a source of turtles for augmentation and reintroductions in streams that have shown improved water and habitat quality following restoration efforts. Prior to engaging in actions to build an assurance colony, protocols need to be developed. Genetic and habitat analysis is needed to guide this process.

Objectives are to develop a habitat model of the 2nd, 3rd, and 4th orders to identify sites with quality flattened musk turtle habitat and sties where habitat restoration or protection is needed for future recovery efforts, identify areas with threat and areas with potential for population reintroduction, conduct field visits to assess sites, collect tissue samples from across the range to analyze genetic variability across the range and within populations.

The process of habitat modeling will be stepwise across the four levels of habitat selection with initial habitat analyses conducted at the 1st order of habitat selection (Johnson, 1980), across the geographic range of the flattened musk turtle. Subsequent 2nd, 3rd, and 4th order (population, patch, and microhabitat scales; Johnson, 1980) habitat analyses will be conducted utilizing field-based habitat surveys to validate site quality at sites determined to be suitable by 1st order habitat models. Importantly, this process will apprise habitat suitability throughout the geographic range of the flattened musk turtle as well as verify the suitability of favorable sites across all four levels of habitat selection.

Flattened musk turtles will be captured from throughout the range to collect tissue samples for genetic representation study; all individuals captured will be returned to the wild once capture data and tissue sample have been collected. Samples will be acquired from the major sub-watersheds of the Upper Black Warrior, Sipsey Fork, Brushy Creek, North River, Locust Fork, and Mulberry Fork.

When available previously collected DNA samples will be utilized, where lacking field collection of turtles will be done. Capture data for each individual will include date and specific locality (latitude and longitude, stream of capture), morphological measurements, sex if an adult, and a tissue sample. Field collections will be monitored through number of localities sampled for turtles, with an accumulation of search effort (i.e. by days, or person-hours), and the number of turtles handled during each search effort. A metric of catch-per-unit-effort (CPUE) will be used to monitor effectiveness of sampling within each sub-watershed. Capture techniques will be by wading and hand capture or trapping following approved USFWS protocol.

To estimate overall genetic diversity of, and the relatedness among sampled individuals, we will collect DNA sequence data from tens of thousands of restriction-site-associated (RAD) loci (Graham et al. 2015, Hoffberg et al. 2016) from across their genomes. These RAD sequence loci have been shown to provide more information about (and thus more accurate and precise estimates of) relatedness and genetic diversity compared to other genetic markers, like microsatellites (Thrasher et al. 2018).

Results expected are a habitat suitability model to be used by resource managers to guide watershed and habitat improvements, to identify stream reaches with suitable habitat and water conditions to be searched for flattened musk turtles or evaluated for flattened musk turtle reintroduction sites, if the species is absent. Genetic study will provide information on genetic variability and isolation between populations, estimate how many individuals, minimally, may be needed to establish an assurance colony and from which populations and aid in determining assurance colony minimum requirements to maintain genetic representation similar to wild populations.

The project is funded by a State Wildlife Grant through ADCNR.

Database Development

Biotics Biodiversity Database

ALNHP maintains a comprehensive database on the location and conservation status of species and ecological communities in Alabama. Biotics 5 is an integrated, web-enabled platform for tabular and spatial data management that centralizes the data and software hosting in a shared "cloud" environment. The database is maintained by NatureServe using a software-as-a-service delivery model. Biotics 5 provides a common data management platform for members of the NatureServe network to achieve and maintain a unified taxonomy and consistent application of our shared data standards and methodology. Biotics 5 provides the framework for managing taxonomic and biological data on elements of biodiversity and mapping known locations for elements of concern.

The Biotics database is supported by funding through our inventory and conservation planning projects. Although building and improving the database has always been a primary goal of the program, securing funding to support this important program area remains a challenge. ALNHP is currently tracking 1,654 rare plant and animal taxa (Fig. 1). There are 1,100 individual occurrences of these species documented in Biotics, with the majority of the Element Occurrences (EO) being for vascular plants or aquatic species (Fig. 2).

Since March 2008, we have been working on improving our database compliance with the Benchmark Data Content Standards (BDCS) for natural heritage data. This past year's efforts focused on Updating the State Wildlife Action Plan status to match the latest SGCN list revisions and last observation date. Another focal area for database improvement was addressing the data backlog. In 2020, this effort resulted in the creation of 367 new occurrence records with many other occurrence records updated. We will continue working to improve the database with the goal of meeting all BDCS goals and reducing the backlog. The focus in the coming years will be reducing the data backlog, continued review of Benchmark Data Content Standards, and QC of EO Rank, EO Rank Date, and Survey Date.

One of the important tasks each heritage program performs is the regular compilation of a Rare Species Inventory List for the state that ranks each element tracked by the program based on the number and quality of occurrences. Our latest revised Alabama Inventory List was published November 2020, with the list distributed to cooperators and other interested parties and posted to the ALNHP website.

Data Requests

In 2020, ALNHP has responded to 44 requests for data or information from academia, conservation non-profits, government agencies, NatureServe, other Heritage Network members, or cooperating partners. The number of requests was smaller than in 2019 because in late 2019, species lists by county were made available on our website, which fulfilled the needs of many requestors from the previous year.

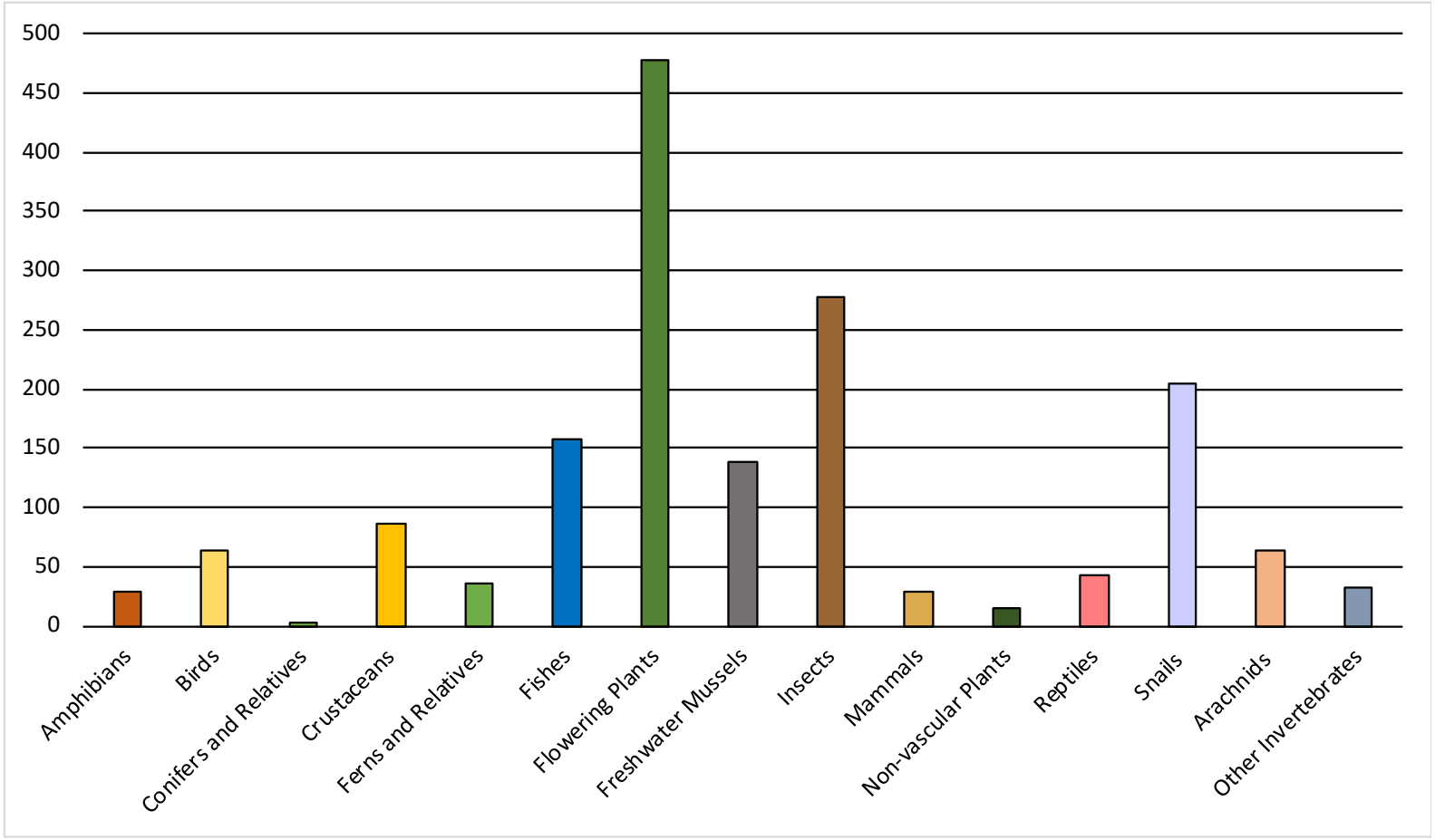


Figure 1. Number of rare plant and animal species tracked by ALNHP (total 1,654).

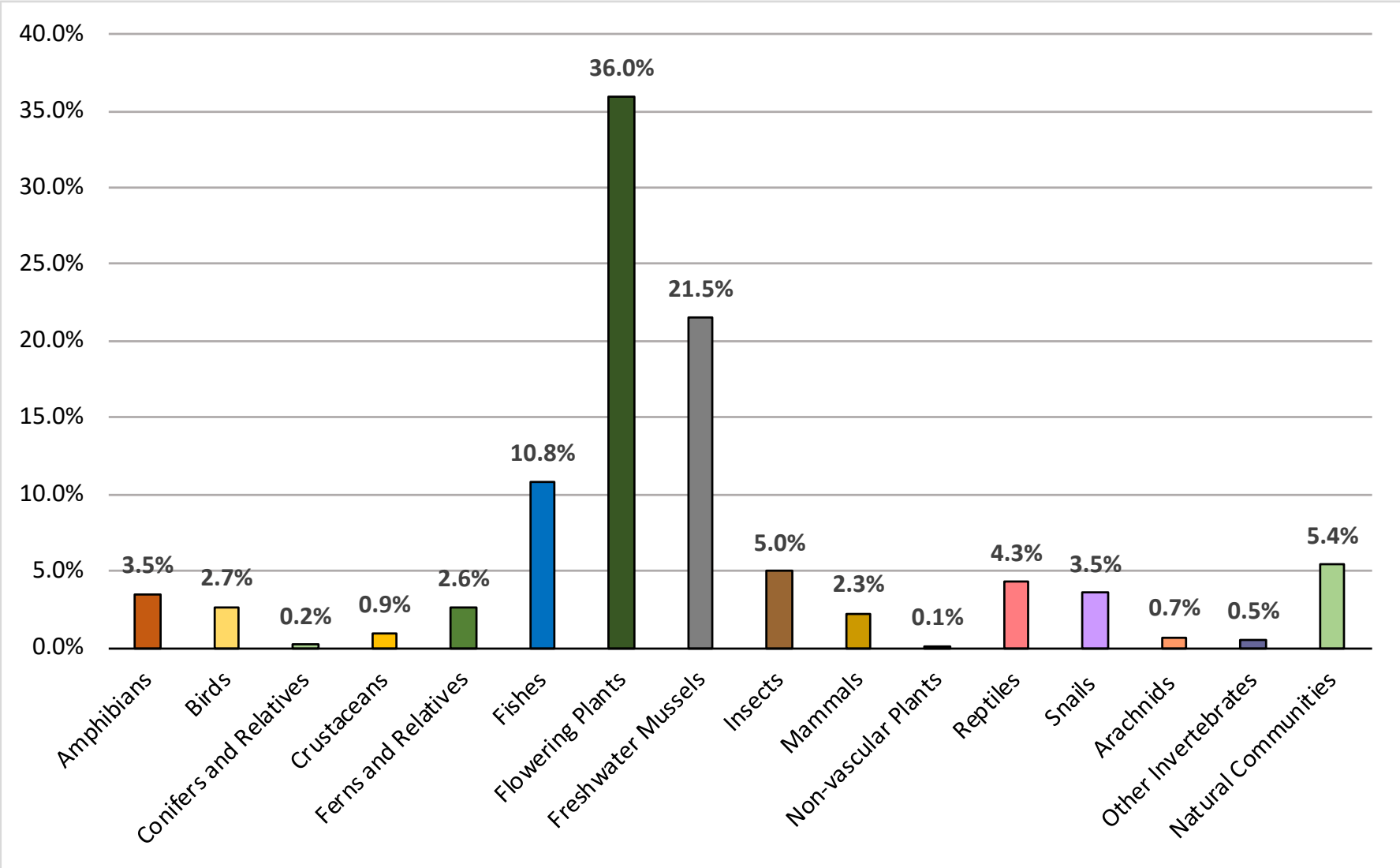


Figure 2. Percentage of 10,406 Element Occurrences in Biotics by major taxonomic group.

OUTREACH

The Auburn University Museum of Natural History (AUMNH) is committed to serving Auburn, the state of Alabama and the southeast region by conducting a variety of Outreach Programs. These programs typically range from monthly public tours to presentations at Alabama State Parks. Highlighting the research and education aspects of the Museum's work, outreach promotes conservation, awareness, and enthusiasm for the natural world around us.

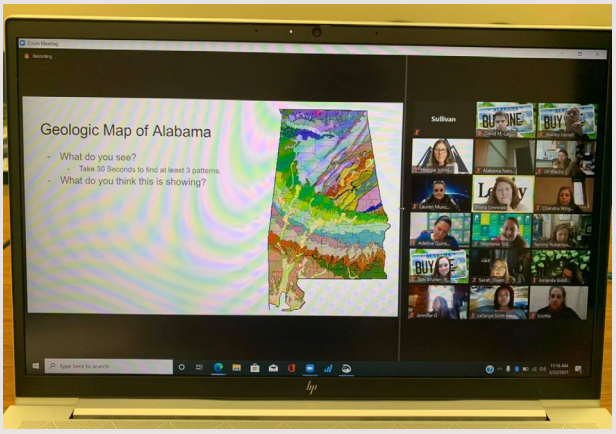
The year 2020 proved challenging for outreach, as the COVID-19 pandemic forced us to cancel all of our traditional programs from early March through the end of the year. This meant we only had two Wehle field days in early March for 38 7th grade students. From January-March, the Museum led nine tours for various school groups and university classes, and our outreach coordinator, Toni Bruner, gave special presentations at six events reaching more than 400 students and educators.

When the university switched to modified operations in early March, the AUMNH boosted its online presence by creating videos and specimen highlights to share with the public. Content was posted three times a week for most of 2020 and some of our videos were viewed more than 1,000 times! Content was shared on our Facebook, Instagram, Twitter, and Youtube accounts.

Mountains to the Gulf, a week-long hands-on crash course in Alabama's geography, ecology, and natural history for educators, was not feasible for 2020 because of the pandemic. Outreach Coordinator Toni Bruner, recognizing the importance of and demand for this program, worked with Legacy Partners in Environmental Education to create a virtual Mountains to the Gulf program. Toni traveled around the state in the summer and fall of 2020 to meet with experts at all of the typical stops and film videos to document the amazing features of the following sites:

- Cahaba Lilies
- Solon Dixon Center
- Dauphin Island
- AU Museum of Natural History
- Wetumpka Impact Crater
- Wehle Nature Center
- Coosa River
- Minkin Fossil Site
- Wild South, Bankhead National Forest
- Camp McDowell, Nauvoo
- Cheaha State Park
- Tumbling Rock Cave
- Little River Canyon
- DeSoto State Park & AU Bear research Project

The program debuted in early 2021 with a full class of participants!



Screenshot of Virtual Mountains to the Gulf Program in early 2021.

OUTREACH

Toni also received a RC&D grant to create a "Mobile Museum". The grant allowed us to wrap our outreach van and purchase natural history exhibits that can be transported and set up at schools and educational events.

As COVID-19 safety restrictions began to ease in the fall, The AUMNH partnered with the COSAM Outreach office to organize an outdoor "Night with the Museum". For this evening event, families were seated together and socially-distanced from one another. Each family was given an observation kit with specimens to look at and learn about as museum staff members presented. Staff also walked around with live animals to show attendees.

Observation stations for attendees to view during staff presentations.

Museum staff showing specimens to "Night with the Museum" attendees.

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OUTREACH

A number of additional activities were still able to take place in 2020, many virtually but some in-person once COVID-19 restrictions eased. These events included

- Presenting at the Cook Museum in Decatur, Alabama
- Virtual classes for schools with corresponding activities dropped off at the schools
- Junior Mad Scientist at Opelika AMSTI
- Working with the Jule Collins Musem on a partnered exhibit for 2021
- Installing the Museum's Dinosaur Egg Exhibit, although the unveiling was postponed

The museum also revamped its apparel store on the AU Marketplace by imporving the aesthetics of the store, and offering new products. The museum began selling Nature-Inspired earrings, now offers reusable water bottles, and also began a membership program. The museum also launched a new and improved website!

Toni has also been active in outreach across the state by participating as a board member for Legacy, the EEAA, and the Indigo Snake Board. She has also partnered with many organizations to lead and coordinate workshops and field trips.

Outreach coordinator, Toni Bruner, at the "Night with the Museum" event

SIGNIFICANT DISCOVERIES

Significant Botanical Discoveries

Thirty-one lichens and lichenicolous taxa were reported new to Alabama in a publication by Hansen et al. (2020) which documented the lichens collected during the 26th annual Tuckerman Workshop.

A new, larger population of the rare lichen species, *Phaephyscia leana,* discovered near Redstone Arsenal where the first state record was recorded (Kelso & Hansen 2020).

Seventy new county records are reported in from the published vascular flora of Chewacla State Park, Auburn, AL (Taylor et al. 2020).

Significant Zoological Discoveries

Adding to the incredible diversity of amphibians found in the state, Craig Guyer, our Curator Emeritus of Herpetology, and colleagues described two new species of salamander found in the southern drainages of Alabama. One of the species, the Escambia Waterdog (*Necturus mounti*), was named after our former curator of Herpetology, Dr. Robert Mount.

Our ornithology curator, Dr. Geoffrey Hill was part of a team that discovered a gene the underlies sexual dichromatism in birds. This is a major discovery in the field of color genetics. This discovery was published in the journal Science (citation below).

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PUBLICATIONS & PRESENTATIONS

Asterisks (*) denote Auburn University student authors or presenters.

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Lawson, K.M., J.L. Ridgway, A.T Mueller, J.Faulkner, and R.Calfee. 2020. Semiautomated process for enumeration of fishes from recreational-grade side-scan sonar imagery. North American Journal of Fisheries Management 40:75-83.

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Project Reports

ALNHP

Godwin, J., and K.M. Lawson. 2020. Green salamander habitat modeling and survey. Final report submitted to the Alabama Department of Conservation and Natural Resources, Division of Wildlife and Freshwater Fisheries, Montgomery, Alabama. Auburn University, Alabama. 18 pp.

Godwin, James. 2020. Escambia Map Turtle (Graptemys ernsti) Status Survey.

Report submitted to the Alabama Department of Conservation and Natural Resources, Division of Wildlife and Freshwater Fisheries, Montgomery, Alabama. Alabama Natural Heritage Program®, Auburn University, Alabama. 13 pages.

Godwin, James. 2020. Mississippi Gopher Frog (Lithobates sevosus) Survey. Report submitted to the Alabama Department of Conservation and Natural Resources, Division of Wildlife and Freshwater Fisheries, Montgomery, Alabama. Alabama Natural Heritage Program®, Auburn University, Alabama. 11 pages.

Godwin, James. 2020. Eastern Indigo Snake Reintroduction in Conecuh National Forest: Future Release Site Selection and Impact on Prey Species. Report submitted to the Alabama Department of Conservation and Natural Resources, Division of Wildlife and Freshwater Fisheries, Montgomery, Alabama. Alabama Natural Heritage Program®, Auburn University, Alabama. 10 pages.

Hermann, S.M., K.M. Lawson, and C. Moore. 2020. Template for selecting gopher tortoise conservation areas. Final report submitted to the Alabama Department of Conservation and Natural Resources, Division of Wildlife and Freshwater Fisheries, Montgomery, Alabama. Auburn University, Alabama. 39 pp.

Schotz, A. 2020. Range-wide status assessment of ***Phlox pulchra*** (Wherry) Wherry (Polemoniaceae), Wherry's Phlox. Alabama Natural Heritage Program, Auburn University, Alabama. Unpublished report for the United States Fish and Wildlife Service. 40 pp., including 5 Appendices.

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Meeting Presentations

Herpetology

Anamza, T.* and J. R. Oaks (2020). Improving estimation of shared evolutionary events. Conference poster. Southeastern Population Ecology and Evolutionary Genetics (SEPEEG) Conference. Auburn, Alabama.

Anchalee Aowphol, *Attapol Rujirawan, *Natee Ampai, and Perry L. Wood, Jr. (2020). Uncovering diversity of Karst dwelling geckos in Thailand: Insights from morphological and molecular evidence. The 9th World Congress of Herpetology, Dunedin, New Zealand. Conference Poster.

Benedict, C. A.*, L. L. Grismer, J. R. Oaks, and P. L. Wood, Jr. (2020). Phylogenetic placement of Burmese tree frogs in the genus ***Polypedates***. Conference poster. Annual Meeting of the Society for Integrative & Comparative Biology. Austin, Texas.

Cobb, K. A.* and J. R. Oaks (2020). Comparing linked versus unlinked character models for species-tree inference. Conference poster. Meeting of the Society of Systematic Biologists. Gainesville, Florida.

Duc Le Minh, Jesse Grismer, L. Lee Grismer, and Perry L. Wood, Jr. (2020). Independent evolution of habitat preferences in 238 species of ***Cyrtodactylus*** Gray, 1827. The 9th World Congress of Herpetology, Dunedin, New Zealand.

Klabacka, R. L.*, H. Parry, J. Yap, R. Cook, T.* Herron, L. M. Horne, J.* Maldonado, G. Alvarez, A. Kavazis, J. R. Oaks, M. Fujita, J. Johnson, and T. S. Schwartz (2020). The powerhouse of asexual cost? Endurance and mitochondrial efficiency in parthenogenetic whiptail lizards (genus Aspidoscelis). Conference talk. 9th World Congress of Herpetology. University of Otago, Dunedin, New Zealand.

Natee Ampai*, Attapol Rujirawan*, Perry L. Wood, Jr., Brian L. Stuart, and Anchalee Aowphol. (2020). Integrative taxonomy of the rock gecko ***Cnemaspis siamensis*** species complex (Squamata: Gekkonidae) in southern Thailand. The 9th World Congress of Herpetology, Dunedin, New Zealand.

Wood, Jr., P. L., J. A. McGuire, L. L. Grismer, L. J. Welton, J. R. Oaks, and R. M. Brown (2020). Phylogenomics of the Green Crested Lizards (Bronchocela): with insights on spatial and temporal evolution. Conference poster. Meeting of the Society of Systematic Biologists. Gainesville, Florida.

Ornithology

Heine, K. B., Justyn, N. M., Hill, G. E., Tucker, V. L., Jung, D., Pollock, B., and Hood, W. R. Modeling Mitochondrial Behavior and Morphology from TEM Micrographs of Copepod Myocytes Following Ultraviolet Irradiation. The Society for Integrative and Comparative Biology, Austin, TX. January 2020.

Justyn, N. M., Weaver, R. J., and Hill, G. E. Exploring the Coloration Mechanisms of the Painted Bunting (***Passerina ciris***). The Alabama Ornithological Society, Auburn, AL. January 2020. Poster presentation.

Justyn, N. M., Heine, K. B., Peteya, J. A., Hood, W. R., Shawkey, M. D., Wang, B., and Hill, G. E. Persistence of Red Coloration in the Eyespots of Copepods (***Tigriopus californicus***) on Carotenoid-free Diets. The Society for Integrative and Comparative Biology, Austin, TX. January 2020.

Powers, M.J., Weaver, R.J., Heine, K.B., Hill, G.E. First clutch size is a reliable proxy for reproductive success in the marine copepod, ***Tigriopus californicus***. Society of Integrative and Comparative Biology 2020.

Powers, M. J., Castaneda, N. J., Hill, G. E. Comparing the frequency of Rock Pigeon plumage pattern morphs in urban and rural environments using citizen-science data. Alabama Ornithological Society Winter Meeting 2020.

Powers, M. J., Heine, B. K., Wilson, A.E., Hill, G.E. A Meta-analysis of Patterns of Mate Choice in Copepods. Society of Integrative and Comparative Biology Annual Meeting 2020.

Mammalogy

Andreasen, V. A.*, Yap, K. N.*, Yamada, K., Williams, A.*, Zikeli, S.*, Kavazis, A. N., Hood, W. R. (2020). The impact of maternal corticosterone on offspring morphology and mitochondrial physiology. Society for Integrative and Comparative Biology, Austin, TX

Favorit, V. R., A. N. Kavazis, W.R. Hood, P. Villamediana, A.L. Skibiel. (2020). Mitochondrial function in the liver and skeletal muscle of mid-lactation dairy cattle. American Dairy Science Association. Poster Presentation.

Heine, K. B.*, Justyn, N. M., Hill, G. E., Tucker, V. L.*, Jung, D. *, Pollock, B.*, Hood W.R. (2020). Modeling Mitochondrial Behavior and Morphology from TEM Micrographs of Copepod Myocytes Following Ultraviolet Irradiation. Society for Integrative and Comparative Biology, Austin, TX

Hood, W. R. (2020). Life history, condition dependency, and mitochondrial performance. Society for Integrative and Comparative Biology, Virtual.

Justyn, N. M., Heine, K. B.*, Peteya, J. A., Hood, W. R., Shawkey, M. D., Wang, B., Hill, G. E. (2020). Persistence of Carotenoids in the Red Eyespots of Copepods (***Tigriopus californicus***) on Carotenoid-free Diets. Society for Integrative and Comparative Biology, Austin, TX

Niitepõld K.*, H. A. Parry, A. G. Appel, J. de Roode, A. N. Kavazis, W. R. Hood. (2020). Of monarchs and mitochondria: Effects of starvation and infection on flight physiology in the monarch butterfly. International Congress of Entomology, Helsinki, Finland

Parry, H. A., Yap, K. N.*, Gladden, L. B., Hill, G. E., Hood, W. R., Kavazis, A. N. (2020). MitoMobile Validation: Taking a Molecular Physiology Lab to the Field. Society for Integrative and Comparative Biology, Austin, TX

Yamada, K. Y. H.*, Zikeli, S. L.*, Yap, K. N.*, Zhang, Y., Kiaris, H., Kavazis, A. N., Hood, W. R. (2020). The relationship between the unfolded protein response and mitochondrial performance in deer mice maintained in a natural context. Society for Integrative and Comparative Biology, Austin, TX

Yap K. N.*, K. Y. H. Yamada*, S. L. Zikeli*, Y. Zhang, Y. Zhang, A. N. Kavazis, L. B. Gladden, M. D. Roberts, H. Kiaris, W. R. Hood. (2020). Individual variation in cellular unfolded protein response, respiratory capacity, and stress tolerance in deer mice (***Peromyscus maniculatus***). Society for Integrative and Comparative Biology, Virtual.

Zikeli, S.*, Yamada, K.*, Yap, K.*, Zhang Y., Kiaris, H., Hood, W. R. (2020). Shy and Stressed? Correlations Between Corticosterone Level, Unfolded Protein Response, and Animal Personality. Society for Integrative and Comparative Biology, Austin, TX. Poster Presentation.

ALNHP

Schotz, A. 2019. Trilliums of Alabama and their Conservation. Trillium Workshop, Mt. Cuba Center, Delaware; Oct. 4-6.

Funded Projects

Funding Source	Project Name	Responsible Party	Amount	Status
ADCNR	Indigo Snake monitoring	Godwin and Warner	$250,973	Ongoing
ADCNR	Genetic and Habitat Analyses to Support Recovery Efforts for Flattened Musk Turtle	Godwin and Armbruster	$159,048	Ongoing
ADCNR	Mississippi gopher frog	Godwin and Armbruster	$27,403	Ongoing
Department of Defense	Arnold Air Force Base gopher frog	Godwin and Armbruster	$47,500	Ongoing
Louisiana Department of Wildlife and Fisheries	Occurrences of western chicken turtle in Louisiana	Godwin and Oaks	$136,436	Ongoing
The Nature Conservancy	Waterdog and Musk Turtle eDNA Survey 2018-20	Godwin and Armbruster	$60,191	Ongoing
ADCNR	Habitat improvement and population expansion of green pitcher plant at Desoto State Park, AL	Schotz and Hansen	$23,557	Ongoing
USFWS	Bog Spicebush Status Assessment	Schotz	$25,000	Ongoing
USFWS	Wherry's Phlox Status Assessment	Schotz	$30,000	Ongoing
ADCNR	White Fringeless Orchid Modeling	Schotz and Hansen	$39,000	Ongoing
EPA	Reference Wetland Study	Armbruster et al.	$229,452	Ongoing
U.S. Army Garrison	Redstone Arsenal Biological Inventory	Armbruster et al.	$100,000	Ongoing
NSF	Compactorized Shelving for the Wet Collections of AUMNH	Armbruster et al.	$195,450	Ongoing
NSF	Aquatic refuge and recovery in the face of drought	Armbruster et al.	$170,986	Ongoing
NSF	Collaborative Research: Red carotenoids as signals of respiratory chain function	Hill	$480,000	Ongoing
PAIR (internal)	Auburn Mitomobile	Hill	$600,000	Ongoing
NSF	DDIG: copepod mate choice	Hill/Weaver	$22,000	Ongoing
ADCNR	Predictive habitat modeling of geographic distribution for the federaly endangered Relict Trillium (*Trillium reliquum*) in Georgia and Alabama	Goertzen and Johnson	$22,000	Ongoing

Funded Projects

NSF	Testing alternative routes of adaptive phenotype-environment matching across heterogeneous landscapes in wild populations	Warner	$1,160,000	Ongoing
NSF	Effects of electron transport dysfunction on carotenoid ketolation	Hill	$550,000	Ongoing
NSF	Supplement CAREER Proposal	Hood and Williams	$55,000	Ongoing
NSF	Genome to Fitness: An analysis of the stress response in *Peromyscus*	Hood	$1,348,649	Ongoing
NSF	Effects of mitohormesis on reproduction and longevity	Hood	$1,032,465	Ongoing
NSF	Collaborative Research: Documenting marine biodiversity through digitization of invertebrate collections	Halanych	$34,019	Ongoing
AL Soybean Producers Assoc	Machine learning soybean diseases	Hardy	$10,000	Ongoing
ADCNR	Investigation of Alabama Red-bellied Turtle Nesting in Alligator Nests	Godwin and Armbruster	$30,062	Ongoing
USFWS	Flattened Musk Turtle and Black Warrior Waterdog Populations Study	Godwin and Armbruster	$85,852	Ongoing