

Intellectual merit. The Museum of Comparative Zoology (MCZ) proposes to enhance the Southwest Collections of Arthropods Network (SCAN) Thematic Collection Network (TCN) by expanding and upgrading the representation of ants (a SCAN focal group) from the southwestern USA. The MCZ will develop a complete online image library of Formicidae from the southwestern states (AZ, UT, CO, NM). This will facilitate identification of ants, especially those in the collections of the SCAN TCN. It will include images of MCZ type specimens and annotated images of exemplar specimens from the MCZ collection for all species known to occur in the region. All types will be databased, geo-referenced, imaged and barcoded. All MCZ specimens will be barcoded and exemplars for each species imaged. To facilitate identifications, key morphological characters essential for species diagnosis will be illustrated along with brief explanatory comments. This field guide approach will create an effective interface for the general public as well as the scientific community. The result will greatly improve the ability of the SCAN TCN member institutions to correctly identify ant specimens in their collections to genus and species. The MCZ will also supplement the image library with occurrence data for approximately 90,000 terrestrial ants collected over the last 20-25 years in Arizona, New Mexico, Utah, Colorado and Texas, and this will facilitate the creation of distribution maps for each taxon. The MCZ has a world-class collection of ants from the region, representing about 90% of the approximately 422 species known from the four corners states (AZ, CO, UT, NM), and 50% of the species are represented by type material. More than 1,500,000 specimens of historical importance are present in the MCZ, and the collection data from these specimens provide information about species distributions going back to the early 1900's. Digital collection records for 90,000 ants will greatly add to the 60,000 specimen records of ants that the SCAN project is already scheduled to produce, thus contributing to the long term goal of using occurrence data for ecological modeling to gain a better understanding of the effects of climate change in the region.

Broader impacts. The image library and enlarged database of collection records produced by this research will be a resource available to SCAN and to the general public, and the creation of these resources will involve the training of graduate students, undergraduates and volunteers in museum databasing and curation. This research will enhance our understanding of biodiversity in the American Southwest, and help guide conservation strategies in the face of climate change. Ants are of wide interest to the general public, and the activities described here will be featured in public presentations and outreach. In particular, the broader impacts of this project will focus on the Navajo Nation, a sovereign region comprising over 30,000 square miles located at the heart of the target area. With support from the Christensen Fund, researchers from the MCZ have studied the ant fauna of the Navajo Reservation for the last four summers (<http://www.navajonature.org/>). In collaboration with the Dine Environmental Institute and with permits from the Navajo Nation Fish and Wildlife, and working with Navajo scientists and student interns, more than 15,000 ants have already been collected from about 80 different localities on the reservation. Specimens collected from this study are among those whose records will be incorporated into the SCAN database. With help from the Encyclopedia of Life (EOL), these data will be used to develop Navajo-friendly educational resources, including a downloadable field guide to the ants of the Nation.

Project Scope

In 2012, the National Science Foundation (NSF) funded a proposal from the Southwest Collections of Arthropods Network (SCAN) as a Thematic Collection Network (TCN) that would bring together resources from 10 small to large-sized arthropod collections in the American Southwest. An important long-term goal of SCAN is to create a virtual collection network that will facilitate specimen digitization and data integration that can later be used in ecological monitoring and species distribution/climate change modeling. In particular, SCAN is focusing its efforts on ground-dwelling arthropods such as ants, selected beetle families, grasshoppers and spiders because these taxa are taxonomically diverse, sensitive to temporal and spatial environmental changes and among the most commonly monitored terrestrial arthropods.

The Museum of Comparative Zoology at Harvard proposes to serve as a Partner to Existing Network (PEN) for SCAN by contributing expertise in the identification and digitization of ants from the American Southwest. SCAN is currently targeting 35 insect families and all ground-dwelling arachnid families. In order to achieve their goals, the participants in the SCAN TCN will need to be able to identify their taxa, many of which are poorly known and/or not well characterized in field guides. Four insect families are designated as the highest priority for digitization: Formicidae, Acrididae, Tenebrionidae, and Carabidae. Of these, Formicidae is the only family that does not have a dedicated taxonomist to aid in annotations. The MCZ can help to fill that gap.

The expertise in ant identification available at the MCZ, combined with its large collection of more than 1.5 million identified ants can be leveraged to enhance the overall quality of the data that will be generated by the SCAN project. Over the years, myrmecologists such as William Morton Wheeler, William Steele Creighton, William L. Brown, Jr., Edward O. Wilson, and George and Jeannette Wheeler have deposited more than 100,000 ant specimens from the American Southwest in the MCZ. At least 60,000 specimens represent recent collections by Stefan Cover, Gary Alpert, and William MacKay. Thousands of additional ants collected by graduate students, ecologists seeking determinations and others have been deposited in the MCZ over the last several decades.

All MCZ specimens will be barcoded and exemplars for each species imaged. On the broadest possible level, by providing a comprehensive, identification-oriented image library of ants of the southwestern USA, and by adding a total of 90,000 high quality occurrence records to the SCAN database, this PEN project will facilitate the identification of ant specimens at SCAN TCN partner collections, enhance the quality of TCN partner collections ant collection records and add a significant number of ant occurrence records from the MCZ to the SCAN database.

Project Goals

1. The MCZ will contribute high-resolution images of 217 MCZ ant types. This represents more than 50% of the 422 ant species occurring within the scope of the project. All ant types will be imaged in frontal, lateral and dorsal view along with label data. Label data will also be

imaged and databased. Each specimen will receive a unique barcode and locality data will be geo-referenced.

2. The MCZ will annotate images of 422 exemplar ant species for a shared image library. Detailed morphological character states critical to identification at the species level will be imaged and annotated for a subset of about 200 species so that a field guide to the ants of the Southwest is user-friendly. Frontal, lateral and dorsal images of many of these species are already posted on AntWeb (<http://www.antweb.org/>) and are available for use in this project.
3. Occurrence data for 15,000 digitized ants within the Navajo Nation and 30,000 digitized ants south of the Mogollon Rim in Arizona will be prioritized as a contribution toward the shared SCAN catalog. An additional 45,000 ant specimens from the Southwest present in the MCZ ant collection will be incorporated as they are digitized and imaged.
4. Undescribed species will be identified to morpho-species and along with their images, occurrence data will be incorporated into the larger project.
5. A significant public outreach component of this grant will be the production of a Navajo field guide to the ants of the Navajo Reservation. This field guide will include sections on identification, biology, distribution and ecology. Annotated images, keys and maps will enhance the guide.

MCZ ant collections from the Southwest

North America has approximately 668 extant species and subspecies of ants, and roughly 422 of these species occur in the Southwest region covered by the SCAN project. The MCZ ant collection holds 217 type specimens of these 422 species, or a little more than 50% of the total. In addition, the MCZ has a larger collection of voucher specimens from the Southwest thus demonstrating both breadth and depth. Breadth is due to the historical nature of this collection of ants, including collections from many of the pioneer myrmecologists of the day including Wheeler, Mann, Brown, Wilson and Cover. Depth comes from the many voucher specimens deposited into the MCZ ant collection from numerous research associates and other scientists working on North American ants. By maintaining an active curatorial staff over the last 100 years, the MCZ has amassed a significant occurrence data set for ants of the Southwest.

Also present in the MCZ collection are numerous specimens of undescribed terrestrial ant species. These specimens and others new to science will be discovered during the process of ant identifications among the 10 SCAN institutions. The MCZ will designate morpho-species code names for those species new to science and make their images and data available to the larger scientific community. It is often said that many new species are languishing, undiscovered, in museum collections, and this project creates an opportunity to alleviate this bottleneck.

The MCZ ant collection holds more than 100,000 mounted, labeled and identified ants from the American Southwest collected since the early 1900's. Our goal is to database most of them (an estimated 90,000) as part of the SCAN initiative. These ants include material that provides comprehensive occurrence data for many rare and/or highly localized species.

The Southwest material can be organized into two groups of specimens:

- Ants with unique data recorded in a spreadsheet. About 45,000 specimens (including those from the Navajo Nation) will be prioritized for digitization because data from their labels have already been entered into an electronic database, and local experts have identified the ants themselves to the species level.

- Ants with bulk (lot) data in a spreadsheet. A second group of 45,000 specimens have been mounted, labeled and identified to species. For the majority of these (~ 40,000), collection data have already been parsed into a spreadsheet format by MCZ staff for each collection event (or lot). Data capture for individual ants will take longer. The remaining ~5,000 are older specimens of historical interest whose labels are often telepathically brief and difficult to read, and most are lacking GPS coordinates and detailed habitat information that is typical of more recent specimen labels. Nevertheless, the historical depth of this older material will allow for capture of comprehensive occurrence data for rare or highly localized species. This is unusually valuable for investigating long term changes in species distributions, and is well worth the additional effort required in databasing.

Digitizing Efforts to Date

The MCZ insect type collection is one of the largest in North America and currently preserves the primary types of 28,588 species representing 29 orders, 565 families, and 7578 genera. The MCZ was among the first museums to post images and data for primary type specimens held in the entomology collections (<http://insects.oeb.harvard.edu/mcz/index.htm>). The last NSF Collections grant awarded to the Entomology Department in the MCZ was “Adding high resolution digital images to the online database of insect primary types in the Museum of Comparative Zoology” (NSF DBI 0237505, PI B.D. Farrell, \$430,000, 2003-2006). Although 16,716 (~58%) of the primary types were imaged as part of this project, these did not include any of the ant types.

A protocol to database, image and barcode the ant types has been developed, and 21 types have already been processed, although none of these are from the American Southwest. Ant specimens are imaged and barcoded, and label data are captured in MCZbase. MCZbase (Arctos) integrates collection data and images into the larger museum community. Although Arctos is widely used, no other SCAN museum currently uses it, and thus it will be important to develop a seamless workflow that adds Arctos in with Specify and Symbiota. For SCAN, Symbiota is the connector that interfaces with EOL, GBIF, etc.

Processing specimens

A general workflow for digitizing ants from the Southwest in the MCZ collection is shown in Table 1. More specific descriptions for each of the two groups of specimens described above are as follows.

- Ants with unique data recorded in a spreadsheet. Pinned ants can be identified in the spreadsheet by searching for "collection date + species name". Each pinned specimen has a unique line in the spreadsheet, with only a few pins containing more than one ant. So, separate

entries for each pinned ant do not need to be added to the spreadsheet at the work station when barcoding the ants.

• Ants with bulk (lot) data in a spreadsheet. Ants can be identified in each unit tray by a characteristic lot number printed in the lower right corner of the top label. Each lot number is a unique identifier for a species collected at a single location. A lot may comprise several individuals on a single pin, or several pinned individuals, or a single specimen, but each lot is a single species, from a specific locality. Specimen notes do not identify how many pins with ants are in a given lot. Thus separate entries for each pin need to be added to the spreadsheet at the work station when barcoding the ants.

Table 1 Summarized workflow for digitizing ants from the American Southwest - steps followed by the supervisor and assistant during the processing of each specimen.

Supervisor	Data capture by Curatorial assistant
Select drawer unit trays for processing	Record unique identifier from a nest series on label, count # of pins and ants on a pin
Color code relevant drawers as trainee becomes experienced	Copy data into additional # lines in spreadsheet compatible with MCZbase
Review data capture spreadsheet for quality control	Select ant and scan unique barcode into spreadsheet
Send spreadsheet to MCZbase for bulk uploading	Repeat for each pinned specimen from a nest series
	Return ant to unit tray designated for barcoded specimens
	Repeat for all specimens

Estimated time and cost to database each ant

- Spread sheet recorded ants and databased ants should take approximately ninety seconds each to enter the barcode number into the spreadsheet once the spreadsheets are prepared. We estimate that 90,000 ants x 90 seconds each = 2,250 hours
- Uploading all the records onto MCZ base using the bulk loader will take additional time (two weeks), but this can be done concurrently.
- Approximately 217 type specimens and 200 other exemplar ant specimens are to be imaged and databased. Ant types are imaged with three standard views plus diagnostic close-ups for taxonomically important characters. Pin labels are also imaged, specimens are barcoded. All types are geo-referenced. From our experience, each type specimen requires about 30 minutes to process. Thus the time required to image 217 ant types is 110 hours. The time to take close-up diagnostic images of 200 exemplar species is about 20 minutes per specimen for a total of 70 hours.

Using the estimated numbers above, and basing the digitization rate per specimen on the section of the grant devoted to data capture, we estimate that 90,000 individual ant specimens can be uploaded into SCAN during the two year grant period. An estimate of the cost per specimen for digitization is \$.73 each. This is itemized in greater detail in the Budget Justification.

Imaging types and exemplar species

Important morphological character states that are essential for diagnosis at both genus and species level identification will be imaged along with notation of corresponding explanatory text. This field guide approach creates an effective interface for the general public as well as the scientific community. Frequently, minute details such as number of teeth on the masticatory margin of the mandibles, number and shape of tibial spines, pilosity on legs or antennae and subtle differences in the base of the scape are essential in making a correct species level identification for ants.

Image capture for each specimen is via a standard set of views. For ants, frontal, lateral and dorsal images are taken in conjunction with the specimen label. Additional images are taken in consultation with an ant systematist to capture diagnostic characters for each taxon when required (**Figure 1**). Neutral gray is used for a background, and the specimen is cleaned first if possible. A metric scale bar with millimeter markings is included in every image. A Leica MZ16 stereomicroscope with a Leica dome light is coupled to a Leica DC420 digital video camera and synchronized to create source images. Helicon Focus is used to compile source images into a single in-focus image (montage) with an enlarged depth of field. Further processing of images is done with Adobe Photoshop and may include adjusting image size, lighting and sharpening.

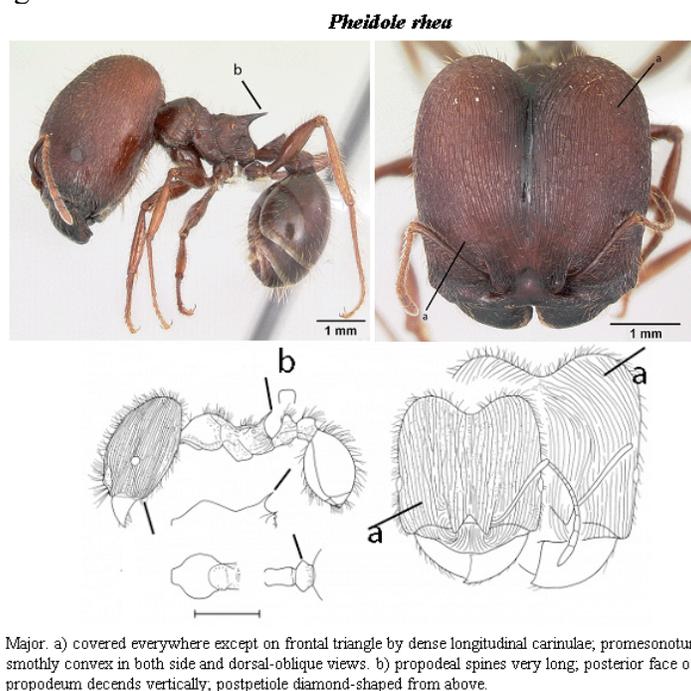


Figure 1. Lateral and frontal view of *Pheidole rhea*. Demonstration of character states important for identification at the species level.

Imaging protocol currently in use for MCZ ant types will be followed. Metadata for each image including photographer and date will be noted. A Leica MZ16 stereomicroscope with an attached Leica DF 420 video camera is used to capture images. Processing of source images into a single high resolution JPG montage is done using Helicon Focus. A second imaging system may be available within one year of this project improving the timeline for completion. Experience has shown that a straight four-hour period at a microscope taking images is a practical limit for a person engaged in this task. This is a primary reason for extending the grant period for two years. It allows rest between sessions at the computer and the microscope, improving efficiency and reducing human errors.

Each image can be uploaded into an Image Library or on-line website that provides additional information that will assist in correct species level identification. Not only annotated diagnostic characters illustrated with images, but also biology, distribution, ecology, taxonomic references and related material can be synthesized into species pages.

The final processed image is saved as a .tif file (archival format) for the MCZbase. The file is named according to the MCZ Archiving Digital Images protocol to facilitate use in museum-wide database. From the final, processed .tif file, .jpg copies are created for use on the MCZbase website. Specifically a 1000-pixel-wide image is the standard. Careful metadata records for all images are recorded including original file name, date the image was taken, name of individual who captured the image, and archiving destination on a shared drive. Metadata for digital images is a best-practice standard, so a Digital image Metadata template is provided to facilitate consistency and migration to MCZbase.

Cyberinfrastructure

Collection records will be incorporated at the MCZ node into MCZbase (Arctos), and this software will be linked to Specify 6 and MySQL database engine. MZCbase website is on-line at <http://mczbase.mcz.harvard.edu/>. Filtered Push will be integrated with Symbiota, Specify and iDigBio. (Morris *et al*, 2009; Wang *et al*, 2009; Dou *et al*, 2011).

MCZ Organization

Professor Naomi Pierce (Principal Investigator) will be responsible for overall management and financial oversight of this project. As Curator in the Entomology Department, she will oversee the implementation of the digitization described in the proposal, coordinate with members of the SCAN TCN, and give presentations to the scientific and general public, including education outreach involving the Navajo Nation.

Professor James Hanken (Senior Personnel) will provide the project with domain expertise in both biodiversity science and natural-science-collections management. He will oversee the Encyclopedia of Life (EOL) Learning + Education staff based at the Museum of Comparative Zoology, who will conduct the educational and outreach activities specified in the proposal in association with representatives of the Navajo Nation.

Linda Ford (Director of Collections Operations, MCZ) will be responsible for supervising database entry procedures including parsing of data and barcode entry.

Stefan Cover (Curatorial Assistant in the Ant Room) will be responsible for preparing specimens for imaging and consulting on ant identification and systematics.

Gary Alpert (Associate in Entomology in the Ant Room) will be responsible for supervising imaging of types and voucher specimens. He will provide coordination among SCAN museums with regard to cataloging and imaging activities, and will assist with outreach activities with the Navajo Nation.

Curatorial staff supported by this grant will be responsible for data capture and imaging.

MCZ Timeline

Table 2. List of major tasks and timeline with sequence for completion.

TIMELINE	Summer Year 1	Fall-Spring Year 1	Summer Year 2	Fall - Spring Year 2	Summer Year 2
MCZ coordination with SCAN meetings					
Digitize 90,000 specimens					
Image 217 Type Species					
MCZbase integration					
Image 200 exemplar specimens and annotate					
Data sharing (GBIF, MorphBank, EOL)					
Assessment and evaluation					

Training Plan for Participants and Students

The work described in this proposal provides excellent training opportunities for students and interns eager to learn museum curatorial skills. Work tasks involve two main pathways: 1) data capture, barcoding and database processing and 2) specimen imaging, metadata transfer and uploading. The Entomology Department of the MCZ has a lively student and intern program for participation in MCZ museum activities and research projects. For example, an ongoing project to database the Lepidoptera Collection in the MCZ has been extremely popular, and in the four years since its conception has trained and/or employed 67 students, work-study students, volunteers and interns.

The training involved in this project will also occur at several levels. The curatorial assistant funded by this grant will train student interns, but will also receive training by working closely with curatorial supervisory staff in the Entomology Department and MCZ Collections Operations to learn bioinformatics and imaging processing skills. Weekly departmental meetings will improve communications with all participants and insure coordinated and smooth operations. Finally, the outreach program involving Encyclopedia of Life will engage students, interns and staff in preparing materials for outreach activities.

Broader Impacts/Public Outreach

The broader impact activities for this proposal will be achieved through leveraging the expertise, tools, resources and audiences of our partner, the Encyclopedia of Life. The Encyclopedia of Life (EOL; <http://eol.org/>) has established significant educational and outreach programs and engages large audiences of scientists, educators and the general public around biodiversity-related topics. Information and materials from the project will be widely accessible to the general public through the free EOL website.

With financial support from the Christensen Fund, researchers from the MCZ have been studying the biodiversity of ants on the Navajo Reservation for the last four summers (<http://www.navajonature.org/>). Working in collaboration with the Dine Environmental Institute and with permits from the Navajo Nation Fish and Wildlife, more than 15,000 ants have been collected from about 80 different localities. Encompassing more than 30,000 square miles and providing significant drainage for the Colorado River, the Navajo Nation is home to numerous described and undescribed species of ants. Integrating the specimen data from these collections with the SCAN project will significantly broaden the impact of this project, and the occurrence data for these specimens will contribute greatly to ecological and distributional information on ants for the TCN network.

The specific broader impact activities of this grant will target the Navajo communities where this project takes place. Using tools developed by EOL, we propose to provide a field guide to the ants of the Navajo Nation that can be used as an educational as well as a scientific tool for environmental impact studies. Reaching these traditionally underserved communities will be accomplished by working through the existing and strong relationships established during previous projects during the last 4 years. We have several Navajo scientists working with us who will assist in the coordination and writing of public outreach documents. The broader impact activities will take place in the end of Year 1 and throughout Year 2 and will focus on Outreach, Capacity building and Dissemination.

At the end of Year 1, EOL staff will begin the outreach portion of the grant. They will work with the PIs to identify community individuals who are interested in being involved with the project. Between 5 and 10 local people will be involved in all aspects of the broader impacts activities. These might include community leaders, scientists, informal and formal science educators and students. Working with the local community, EOL and the PIs will create and implement an outreach plan that includes information about the NSF PEN project, its goals and the broader impact activities and objectives. This plan will be disseminated on the EOL website

under Model Projects (<http://education.eol.org/ideas/model-projects>) and in other appropriate formats (e.g., print version) determined by the Navajo community.

Capacity building activities, including a webinar and workshop, will take place in the first half of Year 2. These activities are aimed at understanding obstacles or parameters related to implementing the outreach and education work while also providing or enhancing the abilities that will allow local people to achieve successful results related to the NSF PEN project. EOL staff will organize a webinar with the community to introduce them to learning, education and outreach opportunities through EOL and to begin laying the groundwork for a follow-up, in-person workshop.

The capacity building activities will provide an introduction to and training on the use of biodiversity resources (Wilson 1988) and tools that can be managed and used by a spectrum of learners from young school children to nature enthusiasts to scientists. The focus of the activities will be to use the information generated by this grant, although the tools and resources can be applied to other biodiversity learning projects beyond this specific activity. The tools and resources will directly utilize the digitized Navajo field guide to ants made available through this grant. MCZbase is already an EOL Content Partner and as of September 2012 has provided 18,241 data objects for 4,098 taxa. These data objects and any new ones made available through the grant will be easily accessible. Examples of the EOL resources and tools that will be used include:

- EOL taxon pages (Ants - <http://eol.org/pages/699/overview>) that include authoritative text, images, video, sound.
- EOL Collections (http://eol.org/info/discover_collections) are a feature that allows any registered user to make a collection of data objects (i.e., taxon pages, images, video, etc) and annotate them for their own interests or projects.
- Customized field guides (<http://fieldguides.eol.org/>) draw upon information in EOL, but are in an editable format so users can add their own information and observations to personalize their field guide. Field guides can be printed in different formats.
- EOL-iNaturalist observational projects (<http://eol.org/info/336>) provide a web-based and/or mobile platform to make, record and share biodiversity observations based on a customized EOL Collection. Participants can see their collective effort toward documenting local biodiversity.

In the last part of Year 2, EOL will focus on dissemination of the project outcomes by posting and/or distributing them through local websites and media venues as well as through EOL. EOL has 2.3 million unique, international visitors annually. Project information will be disseminated in the EOL newsletter (70,000+ international readers), the EOL Blog and social media including Facebook (10,400 Likes) and Twitter (8,600 followers). The EOL Model Project section of the website will highlight the outcomes and links to project-specific Collections, field guides and EOL-iNaturalists projects.

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