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Conservation Genetics and Species Recovery

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Recent advances in molecular genetics have proven to be extremely useful in efforts to conserve imperiled species. Genetics data are used to identify appropriate units of management (e.g., populations, metapopulations), effective sizes of breeding populations, population mixing rates, and other variables. These data help managers make decisions about which populations to preserve, whether to move individuals from one site to another, how to breed species most effectively in captivity, and even, in some cases, what taxonomic classification is most appropriate. Many U.S. Geological Survey (USGS) Science Centers and Cooperative Fish and Wildlife Research Units have developed capabilities in genetics research. The two case studies that follow illustrate how USGS geneticists are assisting managers in recovering species on the brink.

Key Largo Woodrat

The Key Largo woodrat (*Neotoma floridana smalli*) is confined to a single small population in less than 2,225 acres (900 hectares) of tropical hardwood forest in southern Florida. Listed as endangered since 1984, the survival of the Key Largo woodrat is threatened by development (it does not do well in urban areas) and predation by feral cats and other animals. Predictions of sea-level rise and storm surges associated with hurricanes suggest that climate change will also challenge the woodrats' survival in its specialized habitat on Key Largo.

The danger to the Key Largo woodrat is so high that managers and scientists settled on captive breeding as the most likely means of preventing extinction and promoting the subspecies' recovery. Geneticists at the Leetown Science Center in West Virginia, working in collaboration with the Fish and Wildlife Service's South Florida Ecological Services Field Office and Walt Disney World's Animal Kingdom, developed a captive breeding program that maximizes genetic diversity, thereby minimizing problems that may arise from inbreeding. A suite of genetic markers was identified and screened, which allowed genetic "fingerprinting" (genotyping) of over 140 wild Key Largo woodrats. The genotypes were then analyzed to identify appro-

appropriate paired matings for two captive colonies.

Determining the population size and genetic diversity of wild Key Largo woodrats has not been easy. With any endangered species, developing a non-lethal, minimally invasive method of obtaining DNA is highly desirable, so researchers created a method to extract DNA from woodrat scat, or feces. Because woodrats, like many wild mammals, are infested with ticks, a second method of obtaining DNA involves removing ticks to extract, amplify, and analyze the woodrat DNA contained in their blood meal.

Breeding this mammal in the controlled environment of the Disney Animal Kingdom facility has also presented challenges. Females may or may not accept

Key Largo woodrat



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The endangered fairy shrimp *Branchinecta sandiegonensis*.

the male rats that researchers select for them as genetically appropriate mates. On occasion, the females intimidate, injure, or kill the male, and males may injure each other.

Despite these difficulties, 18 offspring resulting from captive breeding have been produced, genotyped, and introduced into the captive breeding program. The unique genotype of each individual will be used to monitor any woodrats returned to the wild gene pool at Key Largo.

Fairy Shrimp in Southern California

Fairy shrimp are small crustaceans that inhabit seasonal wetlands such as

vernal pools, desert playas, and wet meadows. Because they are adapted to living in temporary wetlands, many species can complete the aquatic portion of their life cycle in a matter of days or weeks. During the dry season, embryos encapsulated in hard cysts lie dormant in pool sediments for months or even years (Figure 3). Of the approximately 50 species known in the U.S., five that occur in California are federally listed as endangered or threatened, mainly due to habitat loss. This is especially true in southern California, where over 98 percent of historical vernal pool habitat has been lost to urban development.

In southern California, the endangered San Diego fairy shrimp (*Branchinecta sandiegonensis*) and the threatened vernal pool fairy shrimp (*Branchinecta lynchi*) overlap in distribution with a more common species, the versatile fairy shrimp (*Branchinecta lindahli*). These three closely related species can be distinguished by appearance during their adult stage. However, adults are present only when pools have been filled for a sufficient length of time, which, in drought-prone southern California, can happen rarely or in some years not at all.

Developing a method to distinguish *Branchinecta* fairy shrimp species in their cyst stage provides two advantages. First, it allows listed fairy shrimp to be detected over a much wider time frame. Second, sampling cysts from vernal pools during the dormant dry season is less destructive to other species, particularly the specialized plants that characterize these sensitive habitats. (Southern California vernal pools are also home to five federally listed plant species.) Cyst identification may allow listed species to be detected on project sites earlier in the planning process, and cysts may be a better target for surveys and long-term monitoring at protected and restored sites.

Working closely with researchers at San Diego State University and University of San Diego, USGS geneticists at the Western Ecological Research

Obtaining blood from a Key Largo woodrat for DNA research.



Center's San Diego Field Station have developed a genetic assay that can be used to isolate DNA from fairy shrimp cysts and to screen the DNA to distinguish among species. This assay method provides a fast and accurate method of identifying *Branchinecta* cysts from southern California vernal pools. While it was designed to distinguish among the three *Branchinecta* species present in southern California, it could be modified to assay other regional *Branchinecta* species assemblages.

More generally, using genetic methods to differentiate among the fairy shrimp taxa may be appropriate when species that are similar in appearance occur in the same habitats or when physical differences are only apparent in particular life stages. Genetic identification can aid recovery efforts by allowing the presence or absence of listed species to be determined quickly, so that appropriate management actions can be applied in a timely manner.

These examples describe two very different applications of genetic information, yet both apply novel techniques to aspects of endangered species recovery. They represent just a small fraction of the genetic and molecular research ongoing within USGS research centers (http://biology.usgs.gov/genetics_genomics/conservation_genetics.html, and <http://fresc.usgs.gov/products/fs/fs-2006-3108.pdf>). The field of conservation genetics continues to evolve at a rapid pace. As data collection and analysis methods improve, we are progressing from single gene studies towards more genome-wide approaches, allowing both a deeper understanding of the processes that regulate genetic variation within species and a greater ability to conserve species in peril.

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A vernal pool located on the Miramar Marine Corps Air Base in San Diego County, CA, photographed during the wet and dry seasons.

