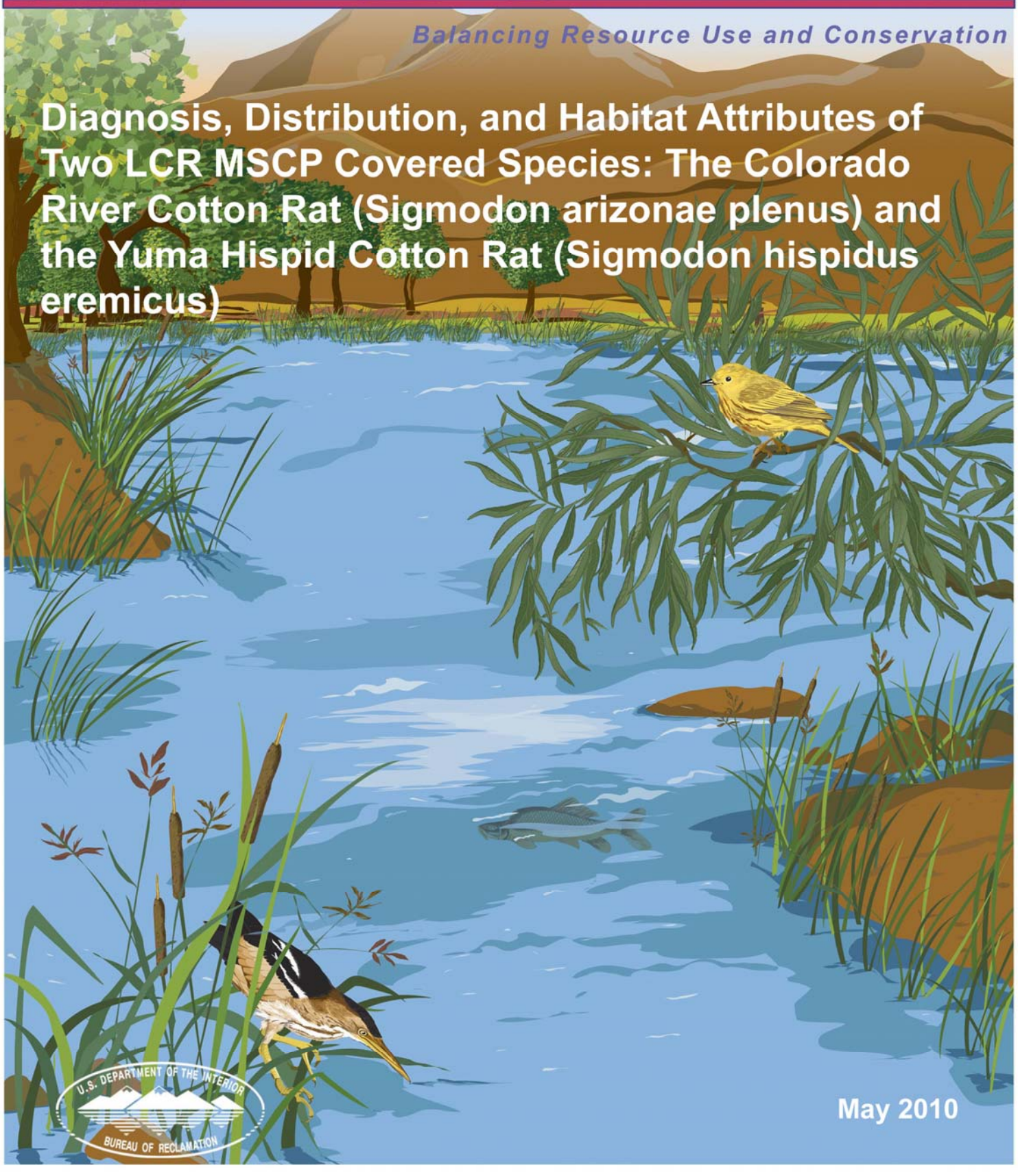
Lower Colorado River Multi-Species Conservation Program

Balancing Resource Use and Conservation

Diagnosis, Distribution, and Habitat Attributes of Two LCR MSCP Covered Species: The Colorado River Cotton Rat (Sigmodon arizonae plenus) and the Yuma Hispid Cotton Rat (Sigmodon hispidus eremicus)



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May 2010

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Diagnosis, Distribution, and Habitat Attributes of Two LCR MSCP Covered Species: The Colorado River Cotton Rat (*Sigmodon arizonae plenus*) and the Yuma Hispid Cotton Rat (*Sigmodon hispidus eremicus*)

Prepared by Sean Neiswenter, Wildlife Group

Lower Colorado River
Multi-Species Conservation Program
Bureau of Reclamation
Lower Colorado Region
Boulder City, Nevada
http://www.lcrmscp.gov

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Introduction

Two cotton rats in the genus *Sigmodon*, the Yuma hispid cotton rat (*Sigmodon hispidus eremicus*) and the Colorado River cotton rat (*Sigmodon arizonae plenus*), occur along the Lower Colorado River and are covered species under the Lower Colorado River Multi-Species Conservation Program (LCR MSCP). Reclamation biologists have established the existence of populations of each species, but diagnosis of species-level taxonomy is not considered reliable in the field.

Three investigators from the University of Nevada, Las Vegas are working in conjunction with the Bureau of Reclamation on an integrated project that addresses three focal needs of the LCR MSCP to enhance the likelihood of successful habitat identification, preservation, and restoration to sustain viable populations of Yuma hispid and Colorado River cotton rats. We are developing: 1) a method of species-level taxonomic diagnosis from field-trapped and released specimens, 2) an assessment of current species distributions (both current and potentially in contrast to distributions about a century ago) within the area covered by the LCR MSCP, and 3) an understanding of the population structure within each of the two species in order to properly choose populations to draw upon for relocation efforts. We are using a molecular genetic approach — specifically, DNA sequencing of a portion of the mitochondrial DNA (mtDNA) control region — to achieve these goals.

Methods

Trapping was conducted between October 2007 and March 2009. Specific sites were located by helicopter, boat, and auto during 24 days of surveying for optimal *Sigmodon* habitat along the LCR. Trapping effort varied depending on the amount of habitat available and was generally concentrated in areas that had habitat consistent with known *Sigmodon* preferences, including *Phragmites*, Johnsongrass, and other grassy substrates that form thick cover (Cameron and Spencer 1981). We set 50-250 Sherman live traps baited with a mixture of oatmeal, peanut butter, and vanilla beginning in the early evening. Traps were checked after sunrise and when present, up to 15 individuals of *Sigmodon* were processed for genetic tissue. Representative vouchers were collected from each population and will be deposited in an American Society of Mammalogists accredited museum; all other individuals were ear clipped and released at the site of capture.

We also collected representatives of each species from other areas (away from the LCR) within each species' distribution. This was done to identify the putative population from which the LCR populations have originated and which, if any, populations are still exchanging genes. We collected individuals of *S. hispidus* from southwestern New Mexico and eastern Arizona and *S. arizonae* from southeastern Arizona and Mexico. We have also acquired samples from the Museum of Southwestern Biology.

Total genomic DNA was extracted from tissue using a Qiagen DNeasy tissue kit following manufacturer protocols. A fragment of mitochondrial DNA including the control region has been amplified via polymerase chain reaction (PCR) and purified. A portion of the control region of the mitochondrial genome was sequenced because it is known to be extremely variable in other mammals and therefore is expected to provide the variation necessary for a study of population genetics. A total of 420 base pairs of the mitochondrial control region have been aligned with Sequencher 4.7 (Gene Codes Corporation). For comparison to previous literature that used a different mtDNA gene, we also sequenced cytochrome oxidase *b* (cyt b) from representative individuals of *S. hispidus*.

Results

Collection Localities

The distribution of successful and unsuccessful trapping localities along the LCR and their relation to historic localities for *S. a. plenus* and *S. h. eremicus* are shown in figures 1 and 2, respectively. We sampled 15 general localities that span the lower Colorado River (LCR), over the course of 21 nights. Of the sampled sites along the LCR, *Sigmodon* have been captured at only seven sites during the two years of trapping. Three sites produced confirmed samples of *S. h. eremicus* and four sites produced *S. a. plenus* (Figure 1). Qualitative trap success for *S. a. plenus* was generally high when the species was present. Fourteen individuals were collected in one night of trapping from an accretion bench near the Palo Verde Ecological Reserve (PVER) on the California side of the river, and nine were sampled in three nights of trapping about a mile north of this site at a second set of accretion benches on the Arizona side. The Cibola Nature Trail site also appears to have a high population density; in one night 14 individuals were trapped. While Pintail Slough produced confirmed *S. a. plenus*, only two individuals were captured after two nights of trapping. Captures of *S. h. eremicus* required much more effort (in trap nights). In most cases several nights of trapping were required to either document presence or collect multiple samples for the genetic analysis. In no case along the LCR were they as readily trapped as *S. a. plenus*. Near Laguna Dam on the Arizona side of the LCR, four individuals were collected during two nights of trapping. Around the Imperial Ponds in the Imperial National Wildlife Refuge, four nights of trapping resulted in three individuals, and two nights near Holtville, California in the Imperial Valley resulted in four individuals.

Sequencing

Approximate localities of samples used in the genetic analysis are shown in Figure 3. A portion of the mtDNA control region, 429 base pairs long, was sequenced from 56 *S. arizonae* and 28 *S. hispidus*. Phylogenetic results for both species are summarized in Figure 4. Sequencing recovered seven control region haplotypes from *S. arizonae*, three of which three occur on the LCR, two that appear to be unique to the LCR, and one that

is common throughout Arizona (Figure 5). A single haplotype was recovered from 14 individuals from the Cibola Nature Trails and 2 individuals from Pintail Slough. The two sites near PVER, one on the California side and one on the Arizona side of the LCR, have a different dominant haplotype than the other localities, and a single individual from the Arizona side has a haplotype that is predominantly found near Phoenix but is widespread throughout Arizona. The LCR haplotypes of *S. arizonae* are approximately 1% divergent from the closest sampled populations in central Arizona. Sequencing of *S. hispidus* along the LCR and in the westernmost portion of the range in New Mexico and eastern Arizona resulted in three control region haplotypes: the LCR is fixed for one haplotype and the other populations contain a combination of the other two haplotypes. The genetic distance between the LCR populations and the nearest populations of *S. hispidus* in Arizona is approximately 0.6% (Figure 4).

Discussion

Species Distribution and Taxonomic Diagnosis

In most cases *Sigmodon* were collected in the same general vicinity as an historic locality, suggesting a fairly consistent geographic range throughout the last century or more. For example, early last century Grinnell (1914) collected *Sigmodon* from "a few miles below Palo Verde" and in the current study *S. a. plenus* were captured at the Cibola Nature Trail restoration site approximately five miles south of Palo Verde. While Grinnell (1914) suggested *Sigmodon* probably did not occur above Ehrenburg, a population was known from southern Nevada in the early 20th century, although the species is currently thought to be extirpated in this area (Bradley 1965). The current study found *S. a. plenus* as far north as Pintail Slough, Arizona, although more sampling in southern Nevada (an area we are beginning to focus on) may document populations from the historic northern distribution. For example, samples of *S. arizonae* housed in the Santa Barbara Museum of Natural History were collected approximately 9 km south of the Nevada locality in 1996. *Sigmodon hispidus* was collected from three localities: Laguna Dam, Imperial Ponds, and near Holtville, California in the Imperial Valley, all of which are consistent with historic collection localities.

The two species have not been found sympatrically (occurring together) along the river in any study to date or in the intervening areas of approximately 47 km between the northernmost known populations of *S. h. eremicus* and southernmost *S. a. plenus*. Blood (1990) stated *S. a. plenus* and *S. h. eremicus* were distributed north and south of the Palo Verde Mountains. Our sampling along the LCR revealed *S. a. plenus* directly east of the Palo Verde Mountains in Cibola NWR. Furthermore, the Trigo Mountains, just south of Cibola NWR, span the Colorado River in a northeast to southwest direction in Arizona and contact the Chocolate Mountains, which form the northern border of the Imperial Valley in California. This area of the river is fairly narrow and steep, spans approximately 30-40 km, and appears to lack habitat typical of *Sigmodon*. Based on these findings, we suggest that the two species are allopatric along the LCR and refine the

barrier between the two species along the LCR as the Trigo Mountains. Therefore, any *Sigmodon* found north of the Trigo Mountains and Chocolate Mountains should be considered *S. a. plenus* and any *Sigmodon* south of those mountain ranges including the Imperial Valley of California should be considered *S. h. eremicus* for the purpose of field identification; however, we suggest molecular diagnosis whenever possible.

Population Structure

All localities sampled for *S. h. eremicus* appear to be fixed for a single control region haplotype, with several possible explanations for this finding. First, we may not have sampled enough localities or individuals to pick up any other haplotypes. *Sigmodon h. eremicus* was not locally common anywhere we trapped. Typical trap success was one or two individuals from each trapping session of 100 trap nights per session. Furthermore, success at a given locality was not consistent between trapping occasions; when a presence was detected at a site we were often unable to capture another *S. hispidus* during subsequent sampling occasions. *Sigmodon hispidus* might maintain low population densities because of a lack of preferred habitat or the LCR population may have been in a bust cycle during our 2-year sampling effort. Additionally, the LCR populations may have been founded by an extremely small population, either because original colonization included only a few individuals, or the population cycles typical for the species effectively results in a founder effect. The lack of diversity along the LCR could be caused by the extreme population cycles that *Sigmodon* typically experience, where periodic boom-bust cycles reduce the local population to small numbers of individuals that then repopulate the area. This would result in most individuals having a very similar genetic makeup.

We were unable to sample any *S. hispidus* from the Yuma area; however, sequences from this area are available from published literature (Peppers and Bradley 2000) that allow comparison to this project. The sequences are a different mitochondrial gene, so we sequenced representatives from the LCR population to compare with the Yuma samples previously published. The previously published sequences are the same as those obtained from our samples of *S. h. eremicus*. This is further evidence that there is minimal variation in mtDNA for LCR populations of *S. hispidus*. Because of the lack of genetic variation in the mtDNA genome of *S. h. eremicus*, relocation efforts probably need not worry about where the individuals come from along the LCR; however, we recommend samples from the closest possible population be used to limit any unforeseen genetic consequences and to mimic, as much as possible, natural dispersal in this species.

Sigmodon arizonae plenus has two unique control region haplotypes along the LCR, which differ by a single base pair. The third haplotype is a common haplotype distributed across Arizona but was only found in one individual on the LCR despite intensive sampling. This haplotype may represent a recent dispersal from the main distribution of *S. arizonae* (either natural or anthropogenic) or it could represent incomplete lineage sorting following isolation of the LCR populations. These two hypotheses are difficult if not impossible to distinguish with mtDNA alone. The population of *S. a. plenus* along the LCR has a unique karyotype (2n = 24, Zimmerman 1970) and mtDNA sequence

divergence is consistent with subspecies level diversification in *Sigmodon* (e.g. Peppers and Bradley 2000), reinforcing the subspecific status of the LCR population. Relocation efforts have several large source populations to draw from and as suggested for *S. h. eremicus*, individuals should be taken from the closest population to maintain the genetic structure of this population. However, relocation is probably not necessary for either of the *Sigmodon* species as there are populations distributed across the LCR and this species appears to readily colonize newly developed habitat (e.g. Cibola Nature Trails).

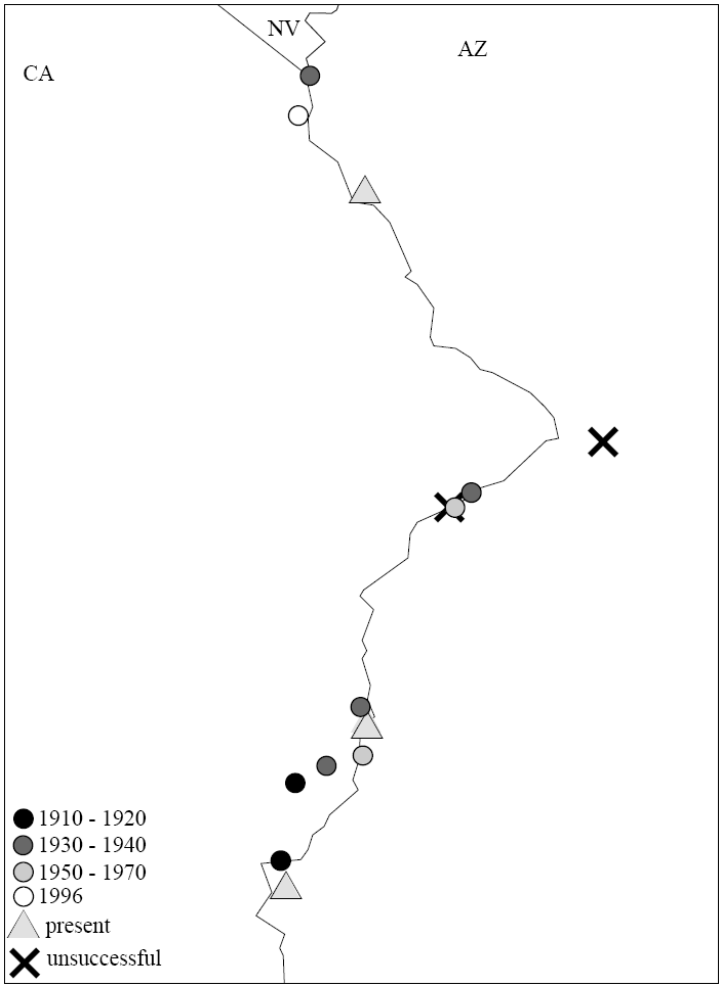

Figure 1. Historic and present collection localities of *Sigmodon arizonae plenus* depicting the known distribution of this species throughout the last century. For clarity, areas where *S. a. plenus* were unsuccessfully trapped during the present study are only shown for general regions away from successful localities (i.e. not all sites sampled during this study are shown).

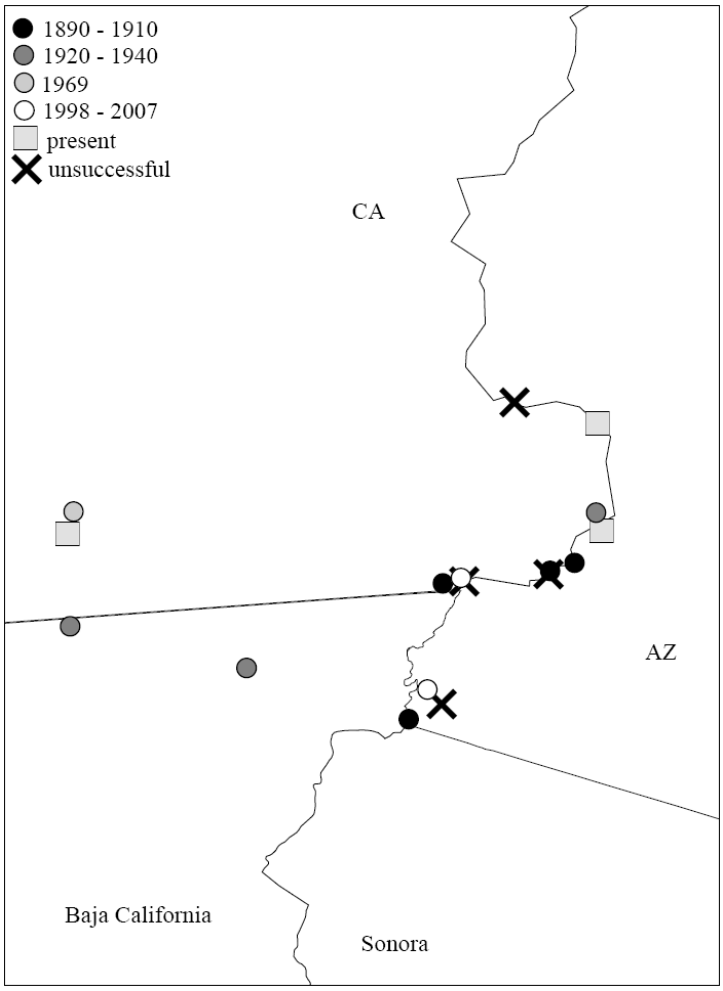

Figure 2. Historic and present collection localities of *Sigmodon hispidus eremicus* depicting the known distribution of this species throughout the last century. For clarity, areas where *S. h. eremicus* were unsuccessfully trapped during the present study are only shown for general regions away from successful localities (i.e. not all sites sampled during this study are shown).

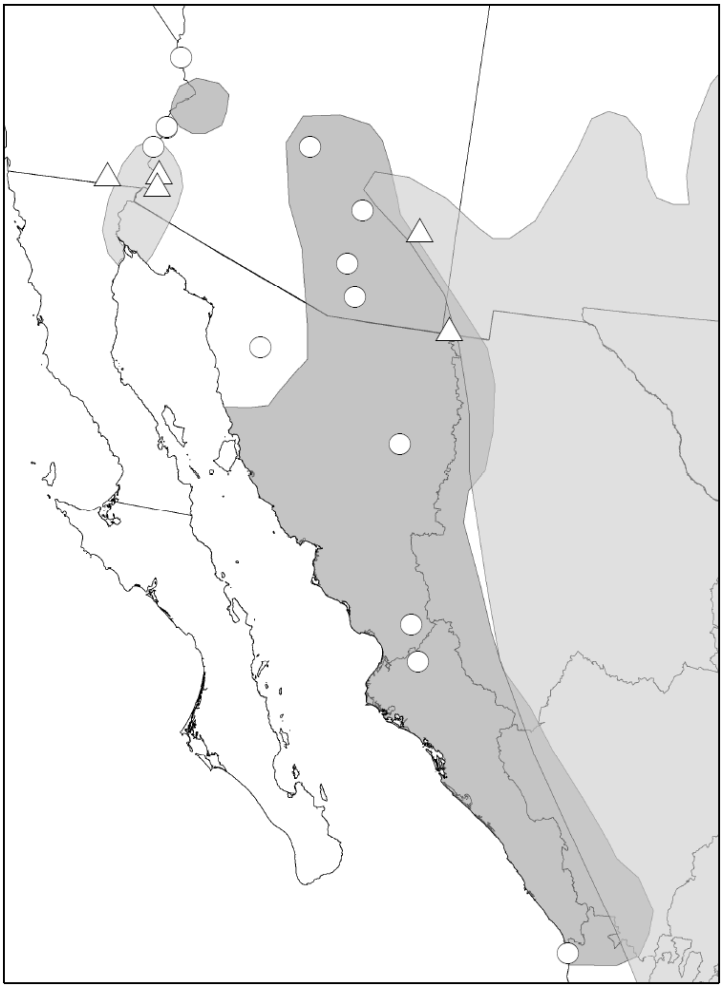

Figure 3. Geographic distribution of *Sigmodon arizonae* (dark grey) and the western portion of *S. hispidus* (light grey). Circles and triangles represent collection localities for *S. arizonae* and *S. hispidus*, respectively. Some symbols represent more than one geographically proximate locality. Geographic distributions from Patterson et al. (2007).

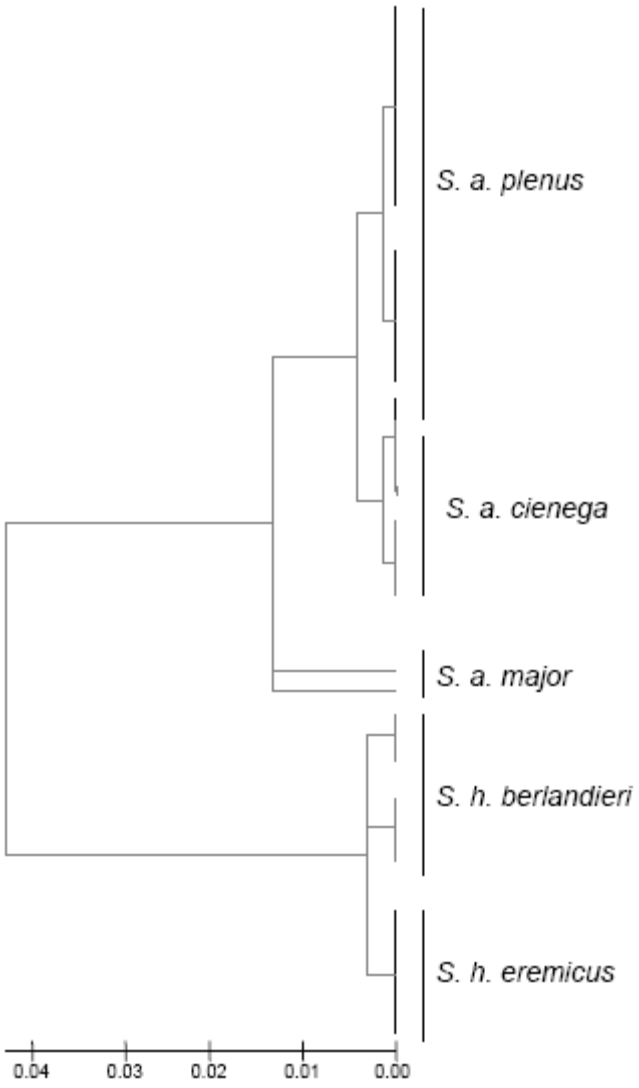

Figure 4. Neighbor joining tree of mitochondrial control region haplotypes from several subspecies of *Sigmodon arizonae* and *S. hispidus*. LCR haplotypes are displayed on the tree in black while other haplotypes are in gray. Scale shows uncorrected p-distance.

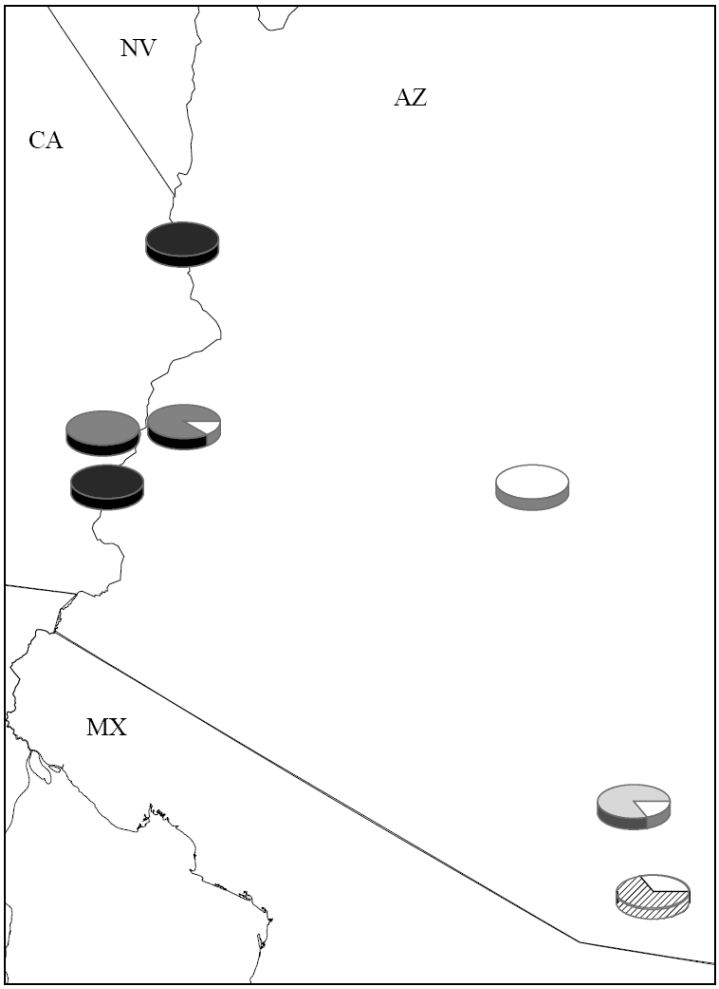

Figure 5. *Sigmodon arizonae* haplotype distribution in Arizona and California.

Literature Cited

Blood, B.R. 1990. Taxonomy and distribution of *Sigmodon* in California. Bulletin of the Southern California Academy of Sciences 89:86-93.

Bradley, W.G. 1965. Status of the cotton rat in Nevada. Journal of Mammalogy 47:349-350.

Cameron, G.N., and S.R. Spencer. 1981. Sigmodon hispidus. Mammal. Species No. 158. 9 pp.

Grinnell, J. 1914. An account of the mammals and birds of the Lower Colorado Valley with especial reference to the distributional problems represented. University of California Publications in Zoology 12:51-294.

Patterson, B.D., G. Ceballos, W. Sechrest, M.F. Tognelli, T. Brooks, L. Luna, P. Ortega, I. Salazar, and B.E. Young. 2007. Digital Distribution Maps of the Mammals of the Western Hemisphere, Version 3.0. NatureServe, Arlington, Virginia, USA.

Peppers, L.L., and R.D. Bradley. 2000. Cryptic species in *Sigmodon hispidus*: evidence from DNA sequences. Journal of Mammalogy 81:332-343.