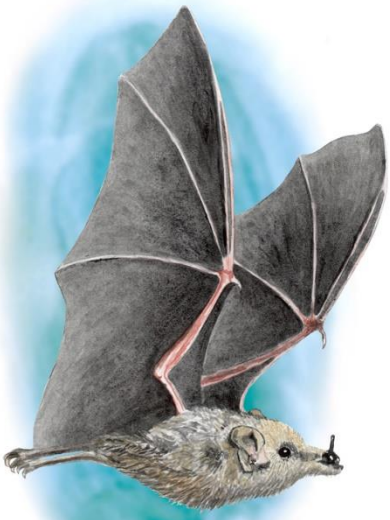
Pima County Ecological Monitoring Program

2021 Proposed Bat Monitoring Protocol and 2018 Monitoring Report



Mexican long-tongued bat
(*Cheronycteris mexicana*)

Townsend's big-eared bat
(*Corynorhinus townsendii)*

Lesser long-nosed bat
(*Leptonycteris yerbabuenae*)

California leaf-nosed bat
(*Macrotus californicus*)

Prepared by Pima County Office of Sustainability and Conservation Staff:
Jeff M. Gicklhorn
Ian W. Murray

Recommended Citation:
Gicklhorn, J.M. & I.W. Murray. 2020. 2021 Proposed Bat Monitoring Protocol and 2018 Monitoring Report. Ecological Monitoring Program, Pima County Multi-species Conservation Plan. Report to the U.S. Fish and Wildlife Service, Tucson, AZ.

Contents

Background & Objectives 1
Monitored bat species 2
Mexican long-tongued bat (*Choeronycteris mexicana*) 2
Pale Townsend's big-eared bat (*Corynorhinus townsendii pallescens*) 3
California leaf-nosed bat (*Macrotus californicus*) 4
Lesser long-nosed bat (*Leptonycteris yerbabuenae*) 4
Monitoring Sites 6
2018 Completed Monitoring Sites 8
Karen's Cave 8
Mine Site #3 9
Mine Site #2 10
Colossal Cave 11
Arkenstone Cave 12
Mine Site #4 13
Mine Site #1 Complex 14
Mine Site #6 15
Mine Site #5 Complex 16
Mine Site #7 17
2021 Recommended Monitoring Sites 18
Colossal Cave 20
Mine Site #1 20
Mine Site #2 20
Mine Site #3 – discretionary site 21
Mine Site #5 – discretionary site 21
New monitoring sites for the 2021 effort 22
Literature Cited 24
Appendix A: 2018 Final Monitoring Report A-1
Appendix B: Field Survey Protocols B-1
Appendix C: Field Data Forms C-1
Appendix D: Bat species observations on Pima County properties D-1

List of Figures

Figure 1. Bat monitoring sites on Pima County conservation lands. Long-term sites denote those sites surveyed in 2018 that were recommended to be surveyed again during the 2021 bat monitoring effort. 7

Figure 2. Stabilized entrance to Karen's Cave. 8

Figure 3. Camera setup at Mine Site #3. 9

Figure 4. Mine Site #3 shaft. 9

Figure 5. Mine Site #2 with locking gate, installed in 2007. 10

Figure 6. Bat Tunnel entrance, Colossal Cave, outfitted with harp trap. 11

Figure 7. Mine Site #4, one adit visible. 13

Figure 8. Mine Site #1 adit (one of four monitored in this effort). 14

Figure 9. Mine Site #6 adit entrance. 15

Figure 10. Mine Site #6 adit (one of two surveyed). 16

Figure 11. Mine Site #7 adit. 17

List of Tables

Table 1. Bat species documented at monitoring sites on Pima County preserve lands in 2018, and 2021 monitoring site recommendations. Some site names have been censored due to species sensitivity... 19

Abstract

Pima County's Multi-species Conservation Plan (MSCP) and Ecological Monitoring Program outline commitments that Pima County has made to monitor four species of cave and mine-roosting bat species covered under the MSCP - California leaf-nosed bats (*Macrotus californicus;* MACA), lesser long-nosed bats (*Leptonycteris yerbabuenae;* LEYE), Mexican long-tongued bats (*Choeronycteris mexicana;* CHME), and Pale Townsend's big-eared bats (*Corynorhinus townsendii pallescens;* COTO). Using available data on prior bat species surveys and the County's database of known cave and abandoned mine features, County staff chose ten sites to include in a bat monitoring effort in 2018. Pima County worked with Tucson Audubon and Sandy Wolf of Bat Research and Consulting to implement a variety of monitoring techniques at these features to assess bat species occupancy, site condition, and relevant management recommendations. We documented CHME roosting at two sites, both of which are likely maternity roosts. We documented MACA using two sites and included a winter roost exit count for one of the sites, which is the most important known local roost for this species. We report on COTO using two sites, including a maternity roost. This monitoring effort also yielded a number of recommendations for sound management, including highlighting two priority projects to potentially install and repair already existing bat-friendly gating at two sites. Pima County is continuing to build on its inventory and understanding of subterranean features that are used by bats and it is not unexpected that monitored sites may change as the County gains new information, or as bat use of particular sites changes over time. Moving forward, final recommendations identified here include changing the suite of sites to be monitored in subsequent rounds of bat monitoring to better address the four species of interest here. Pima County will continue to work with the USFWS to develop a strong bat monitoring program, including substituting some sites monitored in this effort for a proposed region-wide census of soil-piping cavities, caves, and abandoned mines in Cienega Creek Natural Preserve and Colossal Cave Mountain Park targeted at gaining a better understanding of CHME status in these preserves and the conditions and availability of roosts for this species.

Acknowledgements

We would sincerely like to thank the Tucson Audubon Society and Sandy Wolf (Bat Research and Consulting) for their efforts in inventorying all identified monitoring sites in 2018 and developing the survey specific methodology for the four bat species covered in this document. We are also particularly indebted to Pima County Natural Resources, Parks and Recreation staff Iris Rodden and Don Carter for inventorying hundreds of abandoned mine features and for their efforts that have led to a much better understanding of what bat species occur where, and for their ongoing stewardship of Pima County's bat resources. Brian Powell provided guidance and contributed substantially to field efforts. We also thank Colossal Cave Mountain Park staff for accommodating access and monitoring at Colossal Cave. Keith Hughes, BLM provided permits to access and monitor mine sites on Pima County's BLM lease lands. We thank Shawn Lowery, AZGFD, for helpful review and comments.

Background & Objectives

Pima County's Multi-species Conservation Plan (MSCP) covers six species of bats of conservation concern: 1) the Mexican long-tongued bat (*Choeronycteris mexicana*, CHME), 2) Pale Townsend's big-eared bat (*Corynorhinus townsendii*, COTO*),* 3) California leaf-nosed bat (*Macrotus californicus*, MACA), 4) Lesser long-nosed bat (*Leptonycteris yerbabuenae*, LEYE), 5) the Western red bat (*Lasiurus blossevillii*), and 6) the Western yellow bat (*Lasiurus xanthinus*). The first four species are subterranean obligates, meaning that they require subterranean caves and mine features to roost, while the last two species are tree-roosting obligates. The County's MSCP ensures that the County remains in compliance with its Section 10 incidental take permit that it has been issued from the U.S. Fish and Wildlife Service (USFWS).

As part of the MSCP, Pima County has agreed to monitor occupancy and site condition for the first four bat species (i.e., the cave and mine dwelling covered bat species) for ten sites combined, on County conservation lands as one part of Pima County's Ecological Monitoring Program (Pima County 2016). This protocol covers the various approaches that will be used to monitor caves, mines, and adits for occupancy of the four subterranean obligate bat species during the 30-year life of the MSCP. The individual bat monitoring protocols included here (i.e., live capture, internal mine surveys, exit counts, etc.) as well as the first round of bat monitoring results have been provided and carried out by bat biologist Sandy Wolf, working through Tucson Audubon.

Pima County conservation lands have numerous known caves and abandoned mine features, many of which provide potential habitat for the subterranean obligate bat species. These features range from naturally occurring caves to abandoned mine features to eroded soil piping crevices and cavities. The number of sites available for bat monitoring changes as new information becomes available concerning previously documented sites, or as entirely new sites are inventoried. Numerous factors can affect bat species occupancy of these sites, including feature type, season, surrounding habitat, structure, and integrity and volume of human visitation. Prior to development of this protocol Pima County staff spent much time reviewing records and inventorying cave and mine features to assess what information was available concerning bat occupancy status and safety considerations for many of these features. Though this inventory now includes hundreds of features, the County continues to document additional features as well as gather information on how bats use already known features.

Availability of sites also varies according to land ownership. Many of these locations are on lands that the County holds the grazing lease for, but that are owned by the Arizona State Land Department or U.S. Bureau of Land Management, rather than County-owned fee lands. All of the sites covered in this monitoring effort were either located on lands that Pima County owns, or if on leased lands, were on lands leased from the BLM. Additionally, many features represent a safety hazard to humans due to their dangerous and instable nature. Pima County continues to document these features, and after assessments may install warning signage, fencing, or gating for site on those lands that the County owns and manages. Where possible we recommend that any mines that are assessed follow a process compatible with that of the Arizona State Mine Inspector's office evaluation, and that these data are

included in relevant State databases. Other land managers such as the BLM and Arizona State Mine Inspector's office also inventory and manage abandoned mines to reduce risk to human health and protect bat roosts and may close or install bat-friendly gating on some mines and has funded prior efforts to census abandoned mine features and make recommendations for site closure or gating on BLM lands in the Altar Valley. For example, the BLM has installed steel cable net across some features on Rancho Seco. Ongoing abandoned mine assessments by Pima County as well as other land managers that are meant to mitigate the risks that these features pose to human health may also secondarily provide important information on bat species use, which could be leverage by the County's monitoring program.

Pima County's bat monitoring objective is to monitor the occupancy, site condition, and associated management needs of a subset of roosts used by the Mexican long-tongued, Lesser long-nosed, California leaf-nosed, and Pale Townsend's big-eared bats on Pima County open space lands. Monitored roosts will be those roosts that have particular importance to the local population of these four bat species. The County will also work towards documenting all of the known roosts sites for these species that are located on County lands, though not all will be chosen for additional monitoring efforts. Where possible and relevant, results from these inventory and monitoring efforts will be used to guide the management (i.e., gating) and/or restoration of key features that will benefit these and other bat species.

Monitored bat species

Mexican long-tongued bat (*Choeronycteris mexicana*)

The Mexican long-tongued bat is a nectarivorous bat that occurs in southernmost California and Texas (very few records that likely represent extralimital individuals), as well as southern Arizona and southwestern New Mexico extending south throughout large portions of Central America. Individuals range in size from 81 to 103 mm and weigh from 10 to 25 g (Arroyo-Cabrales 1987). CHME tend to roost in small colonies (usually < 10 bats) and individuals do not cluster tightly while roosting (Arroyo-Cabrales et al. 1987; Cryan and Bogan 2003). Most documented roosts in southeast Arizona occur near water and riparian vegetation located more broadly within desert grassland or Madrean evergreen woodland plant communities (Cryan and Bogan 2003). Females and males roost separately, and evidence suggests that only the females migrate north into southern Arizona and southwestern New Mexico to form maternity colonies, while adult males do not range into the United States (Cryan and Bogan 2003). Consequently, most roosts documented being used by this species in Pima County between May and July are likely to be maternity roosts. CHME will often roost within portions of mines or caves where there is some penetration of light either because the feature itself is shallow, or due to roosting closer to the entrance of a mine or cave.

This species produces a single pup between June to July, and in southern Arizona typically migrates south into Mexico between October and November, returning in early May, following the flowering cycles of their preferred food sources, agave, yucca, and columnar cacti (Hevly 1979; Carter and Peachey 1996). In eastern Pima County, Carter and Peachey (1996) found that the majority of pollen in bat guano samples collected between May and early September was from saguaro cactus and Schott's

agave, while October samples yielded primarily Palmer's agave pollen. This work also documented pollen from *Caesalpinia pulcherrima* and *Agave americana* in CHME guano, both common landscape plants in the Tucson area. Through stomach content analyses, Hevly (1979) showed that CHME largely fed on *Agave* sp. and columnar cactus pollen, but that relative to LEYA, CHME contained a greater diversity of pollen types. In CHME collected during summer and early fall in Arizona, this included pollen from plants such as *Yucca* sp., sotol, Chenpodiaceae, *Amaranthus* sp., and pollen from grasses. The latter three types of plants are wind pollinated and how pollen from these sources was ingested is not known. During a population study at Colossal Cave Mountain Park and Cienega Creek Natural Preserve (1994-1996), no CHME were noted in any of the monitored roosts later than November 13 (Carter and Peachey 1996). However, other observations, indicate some individuals may overwinter in this area.

CHME readily feeds from hummingbird feeders, and the AZGFD conduct citizen science-based monitoring of this species and the lesser long-nosed bat by recording the seasonal timing of hummingbird feeder use in both species (https://www.azgfd.com/wildlife/backyard-bats/). Mexican long-tongued bats have been found in roost sites located on the following County open space lands: Buehman Canyon, A7 Ranch, Cienega Creek Natural Preserve, and Colossal Cave Mountain Park (Appendix D). The species is considered a species of concern by BLM and the U.S. Forest Service.

Pale Townsend's big-eared bat (*Corynorhinus townsendii pallescens*)

The pale Townsend's big-eared bat is one of five subspecies of Townsend's big-eared bat that occurs broadly throughout the western United States into western Canada and central Mexico. All Arizona populations of this bat are considered to be this subspecies (AZGFD 2003). COTO is an insectivorous bat feeding heavily upon moths and tends to roost from open ceilings, such that guano located in open areas of a mine (i.e., not at the sides of a feature against the wall) in circular patches may be from this species (AZGFD 2003). Individuals range in size from 90 to 112 mm total body length weigh between 5 to 13 g (Kunz et al. 1982; Pierson et al. 1999). In southern Arizona this species does not migrate (though they occupy different roosts seasonally), but rather they hibernate in abandoned mines or caves with stable and cool temperatures. COTO generally overwinter singly or in small groups, whereas during summer females may form maternity colonies of 12 – 200 bats; males roost apart from females and evidence suggests they are mostly solitary (Kunz 1982; AZGFD 2003). Key conservation needs include protecting and minimizing disturbance of sites identified as hosting small maternity colonies by closing these sites to human access.

On Pima County conservation lands, COTO have been found in roost sites on Rancho Seco, M Diamond Ranch, Cienega Creek Natural Preserve, Diamond Bell Ranch, Marley Ranch, Old Hayhook Ranch, Oracle Ridge, and Colossal Cave Mountain Park (Appendix D). This subspecies is not federally listed, but is currently protected in several other western states. There are two other subspecies of Townsend's big-eared bat that are federally listed as endangered, the Ozark and the Virginia big-eared bats.

California leaf-nosed bat (*Macrotus californicus*)

The California leaf-nosed bat is an insectivorous bat known to occur from the Mojave, Sonoran, and Colorado deserts of the American southwest (southern California, Nevada, and Arizona) and western Mexico. Individuals range in size from 85 to 108 mm total body length and in weight from 12 to 20 g (Anderson 1969). Individuals are largely sedentary (though they do move between roosts as their seasonal thermoregulatory needs dictate) and are active year-round, relying on the ability to access winter roosts that are relatively warm. MACA frequently roosts near the roost opening, where there is some degree of illumination. MACA prey on relatively large arthropods such as grasshoppers and caterpillars, primarily gleaning prey from the ground or vegetation, and feeds on substantial numbers of diurnally active insects (AZGFD 2001). In some portions of its range, it also feeds on cactus fruit (Tuttle 1998). This species often forages in riparian and xeric-riparian environments (i.e., desert washes), and has been photographed at Mission Garden (Tucson, AZ) where they feed on caterpillars and other arthropods gleaned from the Garden's orchard. During the winter MACA requires thermally stable roosts that allow access to warm temperatures. During summer, males and females roost separately, while the sexes roost together during the winter (AZGFD 2001). Some roosts may be occupied year round, particularly those roosts that offer thermally stable and warm winter temperatures. Key needs include a more complete understanding and protection of key winter and summer roosts. Summer censuses may help located smaller roosts within a geographic area, that can then be managed as a unique population cluster.

Across Pima County's preserves, MACA roosts are known from Tucson Mountain Park and Rancho Seco, though they likely occur more broadly (Appendix D). Historically this species also occupied Colossal Cave, though its current status there is not known. This species is a federal species of concern and a wildlife species of special concern in Arizona.

Lesser long-nosed bat (*Leptonycteris yerbabuenae*)

The lesser long-nosed bat is a nectarivorous bat known to occur from southern Arizona and New Mexico (encompassing the northern tip of its overall distribution) into western Mexico and Central America. Individuals can be quite large and range in size from 74 - 90 mm in total body length and 350 to 360 mm in wingspan and weigh from 15 to 29 g (Arita 1999; Cole and Wilson 2006). Individuals undertake a long seasonal migration in order to follow the flowering cycle of their primary food sources, columnar cacti and agave. Large numbers of female LEYE arrive in southwestern Arizona by late April to have their pups and feed on abundant blooming saguaro cacti, leaving these roosts by mid-July, many of them moving east to feed on blooming agave in southeastern Arizona (Cole and Wilson 2006). LEYE is primarily present in eastern Pima County during late summer and early fall, largely feeding on flowering *Agave palmeri*. Most LEYE depart for Mexico by fall after the Agave are done flowering.

There are no known sites on Pima County preserves where this species roosts, though LEYE is known to forage on County lands, such as Sands Ranch and Cienega Creek Natural Preserve. Should a roost be located on Pima County preserve lands, the County will be responsible for annual monitoring of that site (using an exit count protocol) in coordination with AZGFD and USFWS. Before an exhaust system

caused colony abandonment in the 1960s, Colossal Cave hosted a maternity roost of this species. In recent years, LEYE has been captured during bat mist netting surveys at Colossal Cave Mountain Park, Agua Caliente Park, and Cienega Creek Natural Preserve. This species was listed as endangered by USFWS in 1988, however it was delisted in 2018 due to recovery.

Monitoring Sites

Pima County's conservation lands are broad and contain many abandoned mines, caves, and other features that may be used by roosting bats. Though Pima County has documented hundreds of abandoned mines across its lands, the sheer breadth of County lands means that we do not yet have a complete inventory of all features and how they are used by covered bat species (as well as non-covered bat species). Furthermore, given that bats may only seasonally occupy given features based on their life cycle needs, or may abandon or re-occupy particular roosts over time, a one-time survey of a particular feature may not accurately identify what species are potentially roosting there, or how important it may be.

Pima County has committed to monitoring 10 sites for bat occupancy and site condition for the four covered subterranean obligate bat species (combined), every three years. (In the event that a LEYE roost is found on County lands, that roost will be monitored annually.) Sites monitored in 2018 span the range of County conservation lands (both fee and leased lands), from the Las Guijas Mountains southwest of Tucson, to the Santa Catalina and Rincon Mountains to the east (Fig. 1). All selected sites were known to have bat occupancy, however the current species composition, population status, and relative importance (from a biological perspective) were not known for all sites. Below is a brief description of the location and general condition of each monitoring site. Some specific site information is omitted to reduce the likelihood of disturbance to these sensitive species.

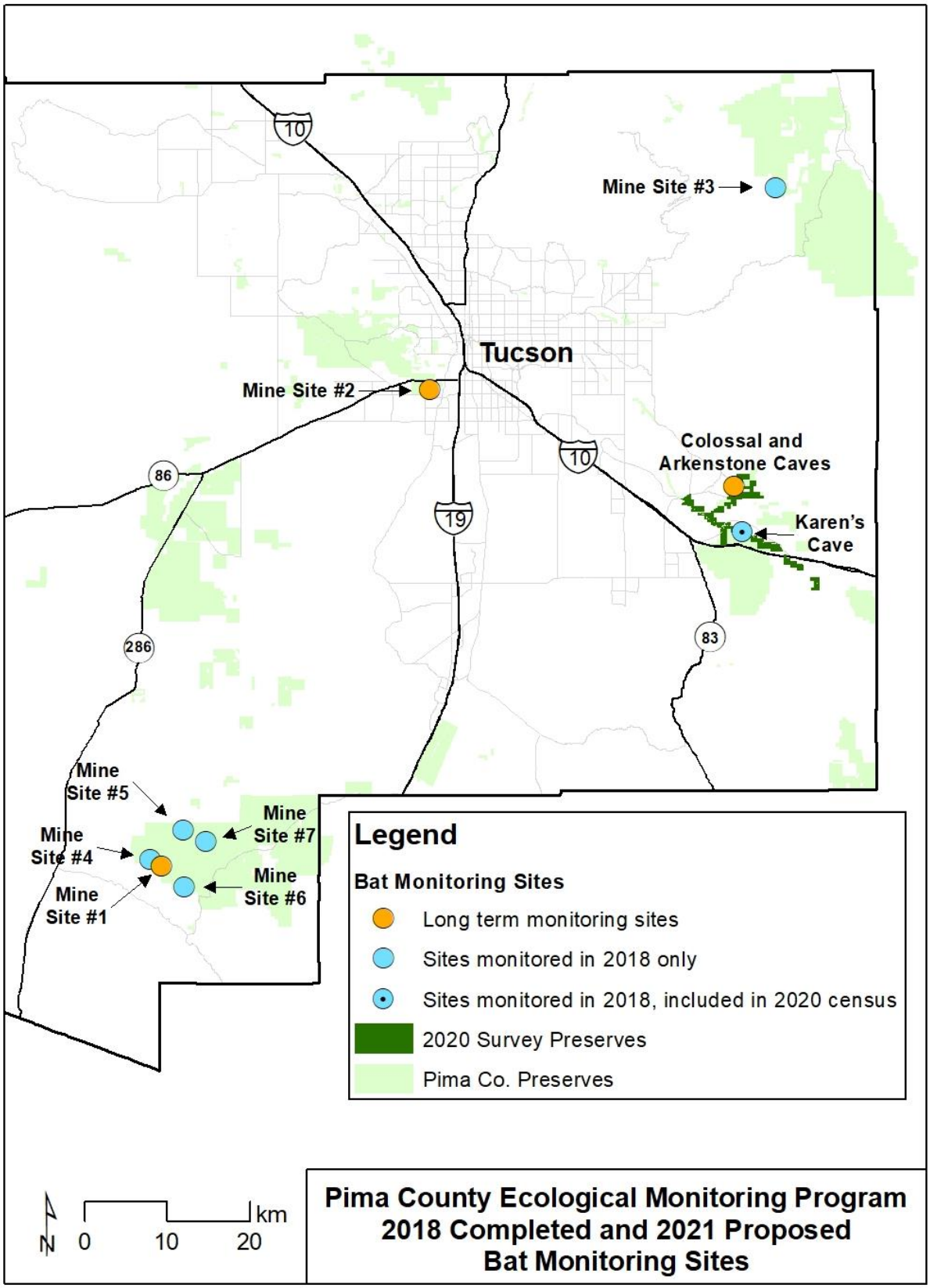


Figure 1. Bat monitoring sites on Pima County conservation lands. Long-term sites denote those sites surveyed in 2018 that were recommended to be surveyed again during the 2021 bat monitoring effort.

2018 Completed Monitoring Sites

We identified 10 sites to include in the bat monitoring completed in 2018. Some of these sites included up to four adjacent and discrete features or subterranean openings that were surveyed during each site visit. The following section provides a general overview of the physical characteristics, ownership, and the basis and/or prior understanding of the potential bat species known or suspected to be using each site.

Karen's Cave

Karen's Cave is a stabilized natural soil-piping feature within the Cienega Creek Natural Preserve. During 2013, both openings to the eroded soil cavern were stabilized with steel culverts in a USFWS Partners for Fish and Wildlife-funded project by Pima County Natural Resources, Parks and Recreation. The interior of the feature has several rooms and passages including the largest room at approximately 20 feet long, 15 feet wide, and 10 feet high at its highest point. Additionally, there are several interior chambers that are not accessible to humans due to small openings that then lead into larger rooms that bats could also roost in. This eroded soil cavity is known to have been a CHME maternity colony (with up to 4 adult and 4 juvenile CHME observed roosting inside) extending as far back as 1996 (Carter and Peachey 1996) as well as being used in the past by small numbers of hibernating COTO.

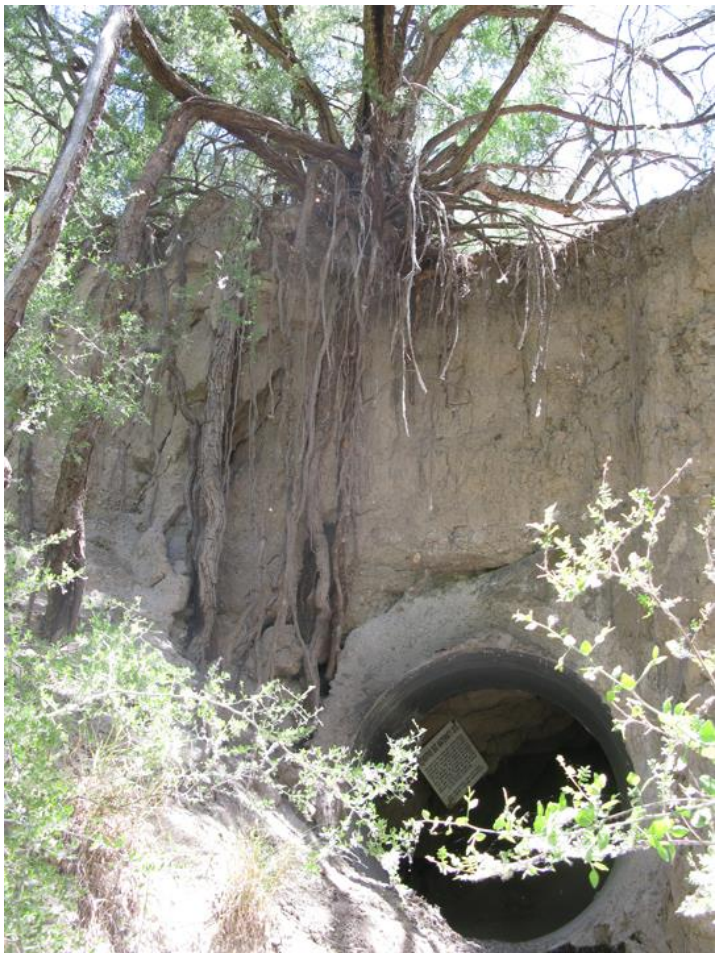


Figure 2. Stabilized entrance to Karen's Cave.

Mine Site #3

Mine site #3 is a mine site located in the Buehman Canyon Natural Preserve on the east side of the Santa Catalina Mountains. The Pima County RFCD owns the surface rights to the land, but the mineral rights are privately held. The site has one main deep, vertical shaft (ca. 8 feet by 12 feet). The shaft is a vertical hole extending downwards to an unknown depth and is fenced by barbed wire and chain link fencing (Figs. 3 & 4). Information provided in a 2017 letter of intent to acquire the mineral claims suggests that in the early 1900s a 130 foot deep shaft with 3,900 feet of drifts over two levels was dug. The site is remote and not readily accessed. Work from this effort as well as from acoustic monitoring in 2016 by Iris Rodden, Pima County Natural Resources, Parks and Recreation suggests that this is a large (> 1000 individuals) summer roost (bachelor or maternity roost) of cave myotis. Due to the sheer numbers of cave myotis recorded exiting on the 2 June 2018 monitoring date, it is not realistic to assess whether smaller numbers of other species may have also been present.

Figure 3. Camera setup at Mine Site #3.

Figure 4. Mine Site #3 shaft.

Mine Site #2

Mine Site #2 is a mine site located at the southern end of the Tucson Mountains within the County's Tucson Mountain Park. California leaf-nosed bats have been known to use this site for many years. In response to vandals entering the site and killing bats, Pima County Natural Resources, Parks and Recreation installed a bat compatible gate to this mine in 2001, that was further updated in 2007 (Wolf and Dalton 2007). Between 2009 and 2011 unknown persons dug out the mine roof just enough above the gate that a 15 inch high gap allows a person to climb over. This site has multiple features with one deep, gated adit (pictured below), several shallow adits, and multiple nearby prospects (Fig. 5). Only the large, gated adit is known to be regularly used by bats. According to prior surveys, the adjacent features are not frequently used, but these other features were not examined during this monitoring session. California leaf-nosed bats are known to roost in the main mine year round, with peak occupancy during the winter months, making this mine a locally important winter roost. There have been a number of monitoring efforts counting bats at this site, particularly during winter, such that future monitoring efforts here could be used to evaluate potential population trends for this site. Furthermore, this site is located near the urban-wild interface, has a high potential for human visitation and disturbance, and the previously installed gating requires further improvements due to vandalism and breaching of the bat gate which can now allow some human ingress.

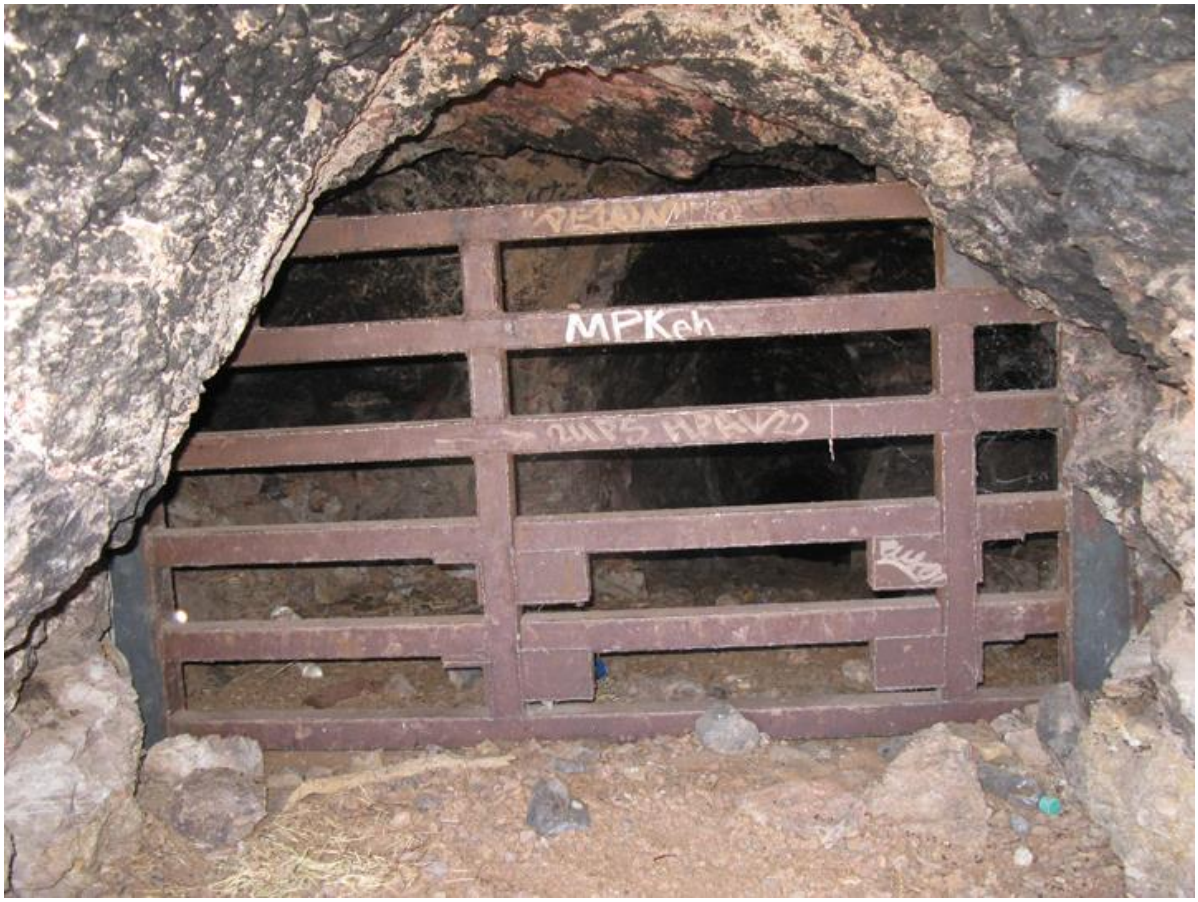


Figure 5. Mine Site #2 with locking gate, installed in 2007.

Colossal Cave

Colossal Cave is a large and complex 'dry' limestone cave located on the west side of the Rincon Mountains within Pima County's Colossal Cave Mountain Park. Estimates suggest that there are about 3 km of passages in this cavern (Brod 1987). This is a large cave system and is known to have at least three entrances within the mountain park (main cave entrance, bat-room tunnel entrance, and Bandit's Escape entrance). Beginning in the 1930s, the Civilian Conservation Corps developed the cave as a tourist experience, including enlarging the main entrance and some passage ways and building walkways and bridges (Brod 1987). At one time Colossal Cave hosted a maternity colony of lesser long-nosed bats (~ 3000 – 5000 bats), but the colony was abandoned (other bat species also largely abandoned the site at this time) after an exhaust fan was installed in 1966. This fan not only altered thermal conditions necessary for the maternity colony (i.e., creating drafts), but also blocked the entrance that this species used to access the cave (Petryszyn 1989). The location of the exhaust fan was changed in 1989 to make the site more compatible to use by bats. Colossal Cave is currently used by Townsend's big-eared bats (2018 capture results) and in the past has been confirmed as a maternity colony for this species (2011 survey data from Sandy Wolf). Pallid bats and cave myotis also use the cave. Lesser long-nosed bats are not known to be using the site, though the possibility remains that LEYE may again re-occupy the site. The cave currently has a commercial cave tour, visitor's center, and other recreational facilities and activities that are all operated by an outside company.

Figure 6. Bat Tunnel entrance, Colossal Cave, outfitted with harp trap.

Arkenstone Cave

Arkenstone Cave is a 'wet' limestone cave located on the west side of the Rincon Mountains within Pima County's Colossal Cave Mountain Park (as a wet cave, strict adherence to White Nose Syndrome protocols are especially important). Discovered in the 1960s, this cave has a single 0.18 m^2 horizontal entrance that is recessed into an entrance pit and is gated. This site is off-limits to the public and therefore has little to no human impact. Prior work has shown that this was a large maternity colony for fringed myotis (a species that today occurs at higher elevations in the Rincon Mountains) during the late Pleistocene, when conditions were cooler and wetter in the immediate area of the cave (Czaplewski and Peachey 2003). Current patterns of bat occupancy of this site are not well known, but earlier surveys documented CHME using this site (Carter and Peachey 1996). Muchmore and Pape (1999) described a new species of troglobytic pseudoscorpion that is only known from this cave highlighting the biological importance of this particular cave.

No site photos of Arkenstone Cave

Mine Site #4

Mine Site #4 is a remote mine site in the Las Guijas Mountains on the County's Rancho Seco property (located on County-owned land). This site consists of two shafts and an adit, all of which are fenced within the same barbed wire fence. Survey work by Don Carter (Pima County Natural Resources, Parks and Recreation) shows that these features are interconnected underground in a complicated and deep network of drifts and winzes. There are many other shafts and adits in the general area as well, many on BLM lands which the County holds in its Rancho Seco grazing lease. Monitoring efforts led by Natural Resources, Parks and Recreation have shown that this feature is an important cave myotis maternity colony, and that smaller numbers of species such as Townsend's big-eared bats also use this mine as a day roost during some times of the year.

Figure 7. Mine Site #4, one adit visible.

Mine Site #1 Complex

Mine Site #1 Complex is a series of shafts and adits in the Las Guijas Mountains on BLM land that Pima County holds as a grazing lease as part of its Rancho Seco property. Some or all of these features include private patented claims. We identified four nearby shafts (some of which may be interconnected) to survey in this monitoring effort based on findings reported by a BLM-funded Bat Conservation International survey effort in 2014-2015. This past survey effort canvassed seven mine features within about 220 m of one another and located a substantial maternity roost of Townsend's big-eared bats in one of them. The monitoring reported here located what is likely the same maternity colony of COTO in a nearby adit, and COTO probably use all of the features during some part of the year. Analysis of the DNA in sampled bat guano also showed use of at least two of these adits by cave myotis and big brown bats.

Figure 8. Mine Site #1 adit (one of four monitored in this effort).

Mine Site #6

Mine Site #6 is a series of shafts and adits in the Las Guijas Mountains on the County's Rancho Seco property. These features are on lands that the BLM owns and that Pima County holds the grazing lease on. Bat Conservation International's BLM-funded surveys included many adits and shafts around Mine Site #6, including the particular adit identified for Pima County's monitoring effort, picture below, an adit about 54 m long with at least one stope near the end. The mineral ownership is not known for this feature, but it may be a private claim. During a 2014 internal survey, one cave myotis was observed in the adit shown below, as well as guano from cave myotis and Townsend's big-eared bats. The surveyors concluded that this feature was likely a cave myotis day roost, and a COTO night roost.

Figure 9. Mine Site #6 adit entrance.

Mine Site #5 Complex

Mine Site #5 Complex is a series of shafts and adits between the Cerro Colorado Mountains and the Las Guijas Mountains. These features are on lands that Pima County holds the grazing lease for as part of Rancho Seco, and that are owned by the Bureau of Land Management. The BLM also likely holds the mineral rights to these features, but this has not been confirmed. Bat Conservation International surveyed at least 15 shafts and adits in this area, and two of these, one pictured below (situated about 200 m apart), were chosen for Pima County's monitoring efforts because MACA had been observed using both features as day roosts during the earlier 2014 surveys that BCI completed. Both sites were occupied by MACA during September surveys reported here.

The openings for both selected features are small, and internal surveys show that there has been a substantial amount of collapse through a large part of the length of one of the features. For example, both monitoring reported here, as well as the prior monitoring done by BCI shows that one adit is a relatively shallow feature showing signs that collapsing rock has blocked much of its former length. The other feature is about 58 m long, though in the monitoring reported here the surveyors were not able to completely survey its length due to its use as a rattlesnake hibernacula in winter and the presence of MACA closer to the entrance during the September survey. Though this second adit is only 'moderately' deep, there would be benefit to ascertaining if MACA uses this feature in the winter. Both sites are fenced with barbed wire and warning signs. During the County's 2018 monitoring effort, an additional three adjacent adits were noted, but not surveyed (two were covered with steel cable netting, and one was occupied by an active beehive, precluding entry).

Figure 10. Mine Site #6 adit (one of two surveyed).

Mine Site #7

Mine Site #7 is a series of shafts and adits in a complex of rocky hills west of the Cerro Colorado Mountains on the County's Rancho Seco property. These features occur on BLM lands. Bat Conservation International surveyed many of these features, one of which was selected for monitoring during Pima County's 2018 efforts. This adit is about 30 meters long and has two stopes. The ownership of the mineral rights at this feature is not known. The prior survey during 2014 noted guano from cave myotis, MACA, and COTO, though no bats were observed. Consequently, surveyors posited that this was a night roost.

Figure 11. Mine Site #7 adit.

2021 Recommended Monitoring Sites

County and USFWS staff have previously discussed the fact that the specific suite of County bat monitoring sites may change over time due to differences in how the covered bat species may use particular sites, as well as due to improvements in our inventory of potential bat roosts across County lands and the species that may use them. Results from the first round of bat monitoring in 2018 showed that some sites either showed little evidence of use by covered species, or were sites that may have been used by covered species but were primarily roosts for other species of bat not included on the MSCP and that would be logistically difficult to monitor and differentiate small numbers of covered bat species from thousands of individuals of other species (Table 1). Monitoring efforts targeted at three of the 'sites'- Mine Site #5, Colossal Cave, and Mine Site #1 - also required substantially more effort to monitor on each visit because these sites include 2, 3, and 4 discrete openings that need to be assessed for bat occupancy and exit counts.

Subsequently, Pima County recommends that only a subset of the sites monitored in 2018 continue to be monitored during the second round of bat monitoring to be done in 2021 (Table 1). These sites are Mine Site #2, Colossal Cave, and Mine Site #1 adits. We also recommend that Pima County's next round of bat monitoring include updated site assessments and bat occupancy status of all of the known soil piping features in Cienega Creek Natural Preserve (14 known in 1996), as well as status and bat occupancy of the nine cave, crevice, and mine features assessed by Carter and Peachey during their 1996 CHME monitoring effort in Colossal Cave Mountain Park (Carter and Peachey 1996)(Table 1). Pending any additional available discretionary resources, we also tentatively identify Mine Site #5 adits and Mine Site #3 as features that are of lesser priority, but that could also benefit from targeted surveys at particular times of the year to gather additional information concerning bat use of those sites. These sites would not be included in the list of committed monitoring sites for Pima County's next round of bat monitoring. We also do not recommend monitoring Mine Site #6 adit, Mine Site #7 adit, Mine Site #4, and would only re-visit Arkenstone Cave and Karen's Cave if snap shot status assessments of all CHME roosts in Cienega Creek Natural Park and/or Colossal Cave Mountain Park were undertaken (Table 1).

Table 1. Bat species documented at monitoring sites on Pima County preserve lands in 2018, and 2021 monitoring site recommendations. Some site names have been censored due to species sensitivity.

Site Name	Covered Species	Non-Covered Species	Site Substitution	Justification
2018 Monitoring Results				
Karen's Cave	Mexican long-tongued bat	N/A	No	Monitor as part of census of all known CHME roosts in Cienega Creek Natural Preserve
Mine site #1	Townsend's big-eared bat	Cave myotis Big brown bat	No	Reliable maternity colony
Arkenstone Cave	Mexican long-tongued bat	N/A	No	Monitor as part of census of all known CHME roosts in Colossal Cave Mountain Park
Colossal Cave	Townsend's big-eared bat	Cave myotis	No	Recent maternity colony for COTO; accessible site
Mine site #2	California leaf-nosed bat	N/A	No	Important winter roost
Mine site #3	N/A	Cave myotis*	Yes	Large roost of probable cave myotis; inaccessible
Mine site #4	N/A	Cave myotis*	Yes	Large roost of cave myotis; inaccessible
Mine Complex #5	California leaf-nosed bat	Cave myotis*	Yes	Small numbers of MACA; unstable site structure
Mine site #6	N/A	Cave myotis	Yes	No recent evidence for covered species use; frequent visitation
Mine site #7	N/A	N/A	Yes	Beehive – no internal survey
Proposed Substitutions				
Cienega Creek CHME roosts	Mexican long-tongued bat	Any other species	In lieu of dropped sites	Assess condition and occupancy of ~14 soil piping features
Colossal Cave CHME roosts	Mexican long-tongued bat	Any other species	In lieu of dropped sites	Assess condition and occupancy of ~9 rock and mine features

*Bats were not captured at these sites, but based on acoustic and trapping (at mine site #4) data collected in previous years by Pima County Natural Resources, Parks and Recreation staff, as well as exit behavior observed in this monitoring, the majority of bats observed at these sites are assumed to be cave myotis.

Colossal Cave

There are three entrances that bats in this cave could exit from and recording video at all of them for exit counts, as well as analyzing the video is a substantial undertaking such that a complete monitoring effort at this site is equal to that of at least two smaller sites. We will discuss with USFWS biologists whether considering a complete and accurate monitoring round at Colossal Cave Mountain Park may be considered equal to monitoring two sites given the complexity of this site.

Monitoring method

1. Capture emerging bats (bat room tunnel entrance) in late July to assess use as a Townsend's big-eared bat maternity colony.
2. Concurrent video emergence counts of the three exits to assess bat abundance and exit patterns as well as cave tour operation impacts
3. Work with Colossal Cave Mountain Park staff (i.e., cave tour leaders, management staff) to receive important observations of bat presence or behavior as well as emphasize bat friendly procedures, lighting, and tour reroutes where feasible. Assess impacts of management and tour-related actions on bat species

Additional opportunities given available resources or interested partners

1. Encourage efforts that address how cave tour operations impact spatial and temporal patterns of bat species use at the site, and robustly evaluate how these patterns may change in response to any changes in cave management and tours implemented in the interest of bat stewardship.

Mine Site #1

Mine Site #1 consists of four nearby adits. Given the number of adit openings, and the resources required to monitor all of them, we recommend treating this complex as two different sites. Pending location of additional known or suspected bat roosts located elsewhere, the County may consider substituting newly discovered sites for the sites at this complex, with input from USFWS staff.

Monitoring method

1. Concurrent video emergence counts of the four features if feasible, otherwise consecutive emergence counts of the features (i.e., video two openings on one night, and two on another night). If resources are limiting, the shallow approximately 23 m long adit identified by prior surveys (XX4 in the attached report) as a likely bat night roost can be internally surveyed relatively easy and not included in a videoed emergence count.

Additional opportunities given available resources or interested partners

1. Pima County should remain vigilant for opportunities to secure the subsurface mineral rights here as a means to permanently protect the site.

Mine Site #2

Mine Site #2 is a single feature accessed by a single, gated entrance. This site is most important as a major winter roost for MACA, and winter occupancy is best assessed through exit counts as determined from evening bat emergence captured on infra-red video.

Monitoring method

1. Winter (November – January) emergence counts through infra-red video
2. Assess status of bat gate and work with Pima County Natural Resources, Parks and Recreation for any site-specific management needs.

Additional opportunities given available resources or interested partners

1. Video September emergence to assess potential LEYE use
2. Consider working with others to outfit female MACA at this site with transmitters to locate additional maternity roosts that may be in the area.

Mine Site #3 – discretionary site

This deep, vertical mine shaft is not being included in future committed bat monitoring efforts. However, pending any available discretionary resources or interested partners it would be useful to assess its use as a seasonal LEYE roost through a video exit count in late September as the species is known to be in the area at this time. During September most or all summer cave myotis roosts would likely be vacated, making it easier to assess LEYE presence on an exit video.

Monitoring method

1. This site will not be included in regularly scheduled monitoring commitments during upcoming bat monitoring.

Additional opportunities given available resources or interested partners

1. Mid to late September emergence counts through infra-red video to assess potential LEYE use of the site.
2. Pima County should remain vigilant for opportunities to secure the subsurface mineral rights here as a means to permanently protect the site.
3. Consider options for bat-friendly securing of the site to prevent human injury as well as other wildlife from falling into the feature.

Mine Site #5 – discretionary site

Mine Site #5 includes two nearby mine adits. Small numbers of MACA were noted using both sites in September. This site is not being included in the list of committed monitoring sites chosen for future efforts, though if there are any available discretionary resources it may be useful to more thoroughly describe their use by MACA (particularly the deeper of the two adits) during summer and winter.

Monitoring method

1. This site will not be included in regularly scheduled monitoring commitments during upcoming bat monitoring.

Additional opportunities given available resources or interested partners

1. Assess whether this is a MACA maternity roost by surveying earlier in the summer (before pups are born) by netting and examining female reproductive status (i.e., avoiding unnecessary entry into the feature).

2. Complete internal survey during winter of SHH2 or if unable to safely enter the feature use videoed exit counts.
3. Monitor both adits for site conditions (vandalism, portal collapse).

New monitoring sites for the 2021 effort

Pima County proposes to substitute approximately 23 additional potential bat roosting sites in its second round of monitoring to be completed in 2021, in lieu of some of the sites monitored during 2018, in coordination with USFWS input. These proposed substitutions are discussed below.

Status and bat occupancy of soil piping caves in Cienega Creek Natural Park

Between 1994-1996 William Peachey and Don Carter (Pima County Natural Resources, Parks and Recreation surveyed all of the known eroded soil piping features in Cienega Creek Natural Park that were occupied by Mexican long-tongued by bats and documented 13 distinct features (12 of which are on County owned land) that were being used by CHME. Occupancy of Cienega Creek Natural Preserve by CHME is largely during the summer and early fall, and is mainly made up of females with young. The highest number of CHME counted on any particular day surveying all of these features was 57 (38 adults and 19 young)(Carter and Peachey 1996).

During the next round of Pima County's bat monitoring efforts we propose a one time (potentially recurring on undetermined cycle as subsequent findings or management needs may dictate) re-evaluation of all of the potential soil piping features within Cienega Creek Natural Park that may host CHME (or other bat species such as COTO). This also includes two additional soil piping features that had been reconstructed in a USFWS Partners Grant to Pima County Natural Resources, Parks and Recreation. Relative to when these features were first surveyed in 1994-1996, there may be some that have eroded to the extent that they are no longer bat habitat, and there may also be additional features that are now suitable for bats. Given the potentially unstable nature of some of these features, surveyors will use utmost care when assessing a particular site and gauging its suitability for an internal survey. Sites will not be surveyed internally when there has been recent rainfall in the vicinity, or if there are signs of recent collapse of substrate near or at the entrance of a feature. All sites will also be surveyed by at least two people and will not be entered if the surveyors deem them to be possibly unstable.

An approach such as is described above is well suited to monitoring CHME within a region given the fact that this species tends to roost in very small groups, in sometimes rather small or shallow sites (see Appendix A). A onetime re-evaluation of all of the known roosts for this species within Cienega Creek Natural Preserve, as well as documentation of any additional roosts is an important part of the management plan for this area, as well as providing a more relevant snapshot of the status of CHME in this area.

Monitoring method

1. Internal surveys and site assessment during summer (approximately two days of field work).

Additional opportunities given available resources or interested partners

1. Work with Natural Resources, Parks and Recreation to evaluate condition and any management needs associated with already completed projects targeting soil piping stabilization efforts targeting bat habitat.
2. Work with the property owners whose land the Amber Adit (an adit within a limestone cave adjacent to Cienega Creek Natural Preserve) is situated on to evaluate its use by LEYE and CHME (it is a known CHME site).

Status and bat occupancy of caves and crevices in Colossal Cave Mountain Park

Carter and Peachey (1996) also monitored all of the known CHME roosts in Colossal Cave Mountain Park, and documented nine features (8 caves or rock crevices and 1 mine) that were being used by CHME (maternity roosts). The highest number of CHME counted on a particular day was 26. Two of these features are those that were included in the County's 2018 bat monitoring effort (Colossal Cave and Arkenstone Cave), but the others were not included in the 2018 monitoring effort. This proposed effort would provide an update to the census produced in 1996. Whether it is prudent to repeat this snapshot view of the status of CHME at these sites every three years (the Pima County bat monitoring interval) would need to be determined. Updating the population status and roost conditions of these sites in Colossal Cave Mountain Park and Cienega Creek Natural Preserve represents a substantial investment of resources, and we proposed that together with the already described recommended sites for the 2021 bat monitoring effort, that this more than satisfies the County's monitoring obligation for these bat species.

Monitoring method

1. Internal surveys, targeted video exit counts (Arkenstone Cave), and site assessment during summer (approximately two days of field work).
2. As resources allow, consider netting Bat Tunnel Exit in late July to assess whether the site is a COTO maternity colony (to be done in 2021 to prevent overly frequent netting at a single site given that it was netted in 2018).

Additional opportunities given available resources or interested partners

1. As opportunity allows, work with the Cave operators to ensure operations minimize disturbance to bat use of the cave. Continue to work towards a bat-compatible gate of the Bandit's Escape Exit to allow bats unimpeded use of the Bat Tunnel Entrance (i.e., Cave tours would then be routed through the Bandit's Escape, rather than the Bat Tunnel).

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Appendix A: 2018 Final Monitoring Report

BAT MONITORING IN PIMA COUNTY

Final Report

April 26, 2019

Sandy Wolf

Bat Research and Consulting

sandyawolf42@gmail.com

Table of Contents

Final Report on Monitoring Bats in Caves and Mines in the Pima County Preserve System, 2018

INTRODUCTION 3

OBJECTIVES 3

METHODS 3

RESULTS 4

DISCUSSION/RECOMMENDATIONS 4

- TARGETED SPECIES 6
  - Mexican long-tongued bat (CHME) 6
  - Townsend's big-eared bat (COTO) 7
  - California leaf-nosed bat (MACA) 8
  - Lesser long-nosed bats (LEYE) 8
- SITE PROTECTION 9

CONCLUSIONS 10

LITERATURE CITED 10

SITE REPORTS 11

FINAL REPORT ON MONITORING BATS IN CAVES AND MINES IN THE PIMA COUNTY PRESERVE SYSTEM, 2018

Sandy Wolf, Bat Research and Consulting

INTRODUCTION

As part of Pima County's commitments outlined in the County's Multi-species Conservation Plan, the county is responsible for implementing the Pima County Ecological Monitoring Program. One element of the program is to monitor site occupancy for 4 bat species at 10 sites (site list and access were provided by Pima County based on data collected by previous field efforts) within the Pima County Preserve System, and to monitor site condition. The 4 species are Townsend's big-eared bat (*Corynorhinus townsendii*, COTO), California leaf-nosed bat (*Macrotus californicus*, MACA), Mexican long-tongued bat (*Choeronycteris mexicana*, CHME), and lesser long-nosed bat (*Leptonycteris yerbabuenae*, LEYE). All 4 species are subterranean-obligates, which means they require caves or mines to survive. Pima County has not yet finalized selection of the 10 caves/mines; staff is inventorying and evaluating potential sites for use by the 4 targeted species as well as other bat species. We (Bat Research and Consulting) were contracted to assist with the monitoring effort.

OBJECTIVES

1. Survey 10 sites 2 times (approximately) between 15 April and 31 December, 2018 for bat use and site condition. Identify species of bat, number of individuals, and observed threats to the site (e.g., vandalism, evidence of human visitation, collapse of portal/internal workings).
2. Develop protocols for field methods for inventorying and monitoring bats, including internal survey of mine features, infrared video-recordings of bat evening emergences, bat captures, and DNA analysis and species determination of bat guano.

METHODS

We used different methods for monitoring sites based on each site's attributes, and what was previously known about the site and bat use of the site (data from Pima County and BLM). Methods included internal surveys, video-recording evening bat emergences, capturing bats, and deploying guano sheets to collect guano for analysis to determine species. Monitoring protocols for each survey method are discussed in detail in the Protocols for Field Methods for Inventorying and Monitoring Bats in Caves and Mines included in this report. Specific methods used for each site are given in Appendix A: Site Reports. Methods were selected and implemented so that disturbance to bats, when present, was minimized while obtaining necessary data. All methods are standard practices and discussed in the American Society of Mammalogists guidelines (Sikes et al. 2011) and the "bible" of bat research methods (Kunz and Parsons, 2009).

We held a scientific collecting license from Arizona Game and Fish Department (SP619854). We notified the appropriate Pima County, Colossal Cave Mountain Park, Border Patrol, and Bureau of Land Management personnel as necessary before going out to each site.

We cleaned gear after each field trip, following the most recent decontamination protocols for white-nose syndrome from the U.S. Fish and Wildlife Service.

Collected guano was sent to the Bat Ecology and Genetics Lab at Northern Arizona University for analysis.

RESULTS

Between May and December, 2018, we visited 10 sites at least one time: Arkenstone Cave, Colossal Cave, Karen's Cave, Mine Site #3, Mine Site #2, Mine Site #4, Mine Site #1, Mine Site #5, Mine Site #7, and Mine Site #6. Sites #1 and 5 had multiple features. We observed 3 of the 4 targeted species: Townsend's big-eared bats at 2 sites (Colossal Cave and Mine Site #1), California leaf-nosed bats at 2 sites (Mine Sites #2 and 5), and Mexican long-tongued bats at 2 sites (Arkenstone and Karen's Caves). We did not observe or find evidence of use by lesser long-nosed bats at any of the sites. We observed cave myotis (*Myotis velifer*) at Colossal Cave, and what were most likely cave myotis at 4 additional sites (Mine Sites # 3, 4, 5, 6). We collected guano for DNA analysis at Karen's Cave, Mine Site #1, and Mine Site #6. Analysis of this guano confirmed use by cave myotis at Mine Site #1 and 6, and use by big brown bats at Mine Site #1. Guano results from Karen's Cave indicated Mexican long-tongued bat use.

We were unable to survey 3 features: Mine Site #7 and two additional adits near Mine Site #5. There were bee hives at the portals of Mine Site #7 and one of the adits near Mine Site #5, and one adit near Mine Site #5 was closed with cable netting.

We observed and recorded the condition of each site's physical structure and evidence of human visitation or vandalism. All abandoned mines are subject to natural deterioration and some showed more signs of instability than others. Except for Colossal Cave, sites had unrestricted/unenforced access. We saw very little evidence of recent human visitation in the form of trash or tire tracks. Fortunately, most sites are in locations that do not receive much visitation. One of the most vulnerable is Mine Site #2, which is near to houses; the gate installed in 2007 was breached and needs repair. Another is Mine Site #6 in the Rancho Seco area, where there was evidence of rock-hounding. Prior field efforts by Pima County staff have also encountered people exploring the interior of this feature (Ian Murray, personal communication). We observed some trash from migrant use near the trail to Mine Site #4.

Results for each site are given in Appendix A: Site Reports.

DISCUSSION/RECOMMENDATIONS

Discussion and recommendations for each site are in the Appendix A: Site Reports. See below for a brief summary of site-specific monitoring strategies.

MONITORING SITES AND SELECTION STRATEGIES

Of the 10 sites we monitored, we think Mine Site #2, Colossal Cave, and Mine Site #1 are the best choices for permanent, long-term monitoring. Mine Site #2 and Colossal Cave have been occupied by California leaf-nosed bats and Townsend's big-eared bats, respectively, for many years, and therefore are stable colonies. Although the COTO colony in Mine Site #1 was first observed rather recently (in 2014), maternity colonies tend to be site faithful, and our observation of the colony again in 2018 provides evidence that the colony is stable.

Karen's Cave has been monitored periodically by Pima County staff since 1997. Mexican long-tongued bats have been observed from 1997 to the present, including 2018. Townsend's bats have not been observed since 2013 (Pima County data). We collected a bat guano sample (from an insectivorous bat species) that had been deposited at this site after the start of this project, but eDNA analysis of this sample and associated species determination will not be available until after this report is submitted. This site seems to be a good site to monitor CHME (but see discussion in section on Targeted Species under CHME).

Arkenstone Cave, Mine Site #5, Mine Site #6, and Mine Site #7 need additional inventory work on bats using these sites to determine whether they are suitable as long-term monitoring sites. For example, although guano

analysis from Mine Site #6 confirmed use of a non-target species (cave myotis), it is possible that other species use this site; the 2014 survey found evidence of COTO use. These sites are discussed in the next section on Targeted Species.

Mine Site #4, though a biologically valuable site given its status as a large cave myotis maternity site, is not a good choice for monitoring any of the 4 targeted species because of its physical characteristics and its use by large numbers of cave myotis spring through fall. It is not possible to conduct internal surveys or place guano sheets. Video-recording emergences requires 2 cameras and many lights, and does not yield high-quality video or permit species identification. Townsend's bats have been captured on the bench below the shafts (Pima County data), but although the bats were likely roosting in the mine, mist-netting is not a valid method to determine abundance; it can only confirm presence of a species. Mist-netting should be avoided when bats are pregnant or newly lactating (mid-May through early August) and is not likely to be successful in cool months when bats are torpid, so the time period for using mist-netting to document presence is narrow. In addition, failure to capture a targeted species cannot be interpreted as absence of that species in the mine or area. Acoustic data can document presence, but may miss Townsend's bats because of their call characteristics or placement of equipment.

Mine Site #3 is also not a good choice for monitoring targeted species, for similar reasons as those given for Mine Site #4. It seems to be primarily, or solely, a cave myotis summer roost (use by a maternity or bachelor colony is unknown), and is a deep shaft not safe to rappel (collar has loose material). Although it may be difficult to distinguish lesser long-nosed bats from cave myotis either acoustically or by video-recording depending on the proportion of lesser long-nosed bats to cave myotis and total abundance, the site should be surveyed in mid-late September for lesser long-nosed bats (see section on Targeted Species).

More inventory work on caves and mines in the Pima County Preserve system will identify additional sites that are occupied by bats and potential sites that are suitable as permanent, long-term monitoring sites. Multiple potential sites in each geographic area of interest should be surveyed in all seasons and over multiple years to determine how each site is used, when, and in what numbers, by a particular species. A particular site may be important one year and not the next because of changes in temperature and rainfall patterns that affect roost microclimates and prey availability, human disturbance, and the availability of alternate nearby sites. Although such intensive work can be costly and labor intensive, it is crucial that how, when, and in what numbers bats use a particular site is understood in relation to other nearby roosts before a site is selected for long-term monitoring. It is of no value to monitor a site that is used as an ephemeral, transitory, or auxiliary roost by a small number of bats when large numbers of the same species are roosting nearby, or in the same site at a different time of year.

It is much easier to determine abundance accurately, and therefore determine long-term trends, at a single-species roost, so such a roost is preferable to a multi-species roost for permanent long-term monitoring sites. Monitoring multi-species roosts adds to the difficulty of determining abundance, or even presence, of each species. For example, several hundred MACA (large for most MACA colonies) are more difficult to detect and accurately count if they are among thousands of LEYE, even with a high-speed camera and optimal lighting (pers. obs.). It may be impossible if the camera's field of view must be wide to cover a large entrance and species cannot be distinguished.

For a single-species roost, such as Arkenstone Cave, an Anabat Roost Logger acoustic detector can be used to find the time of year when activity is greatest (e.g., when adult numbers in a maternity colony peak before parturition). Abundance can then be obtained through evening bat emergences during that period of time in the future. Deploying a roost logger inside the entrance of a mine or cave for a couple years minimizes personnel time otherwise spent on many survey trips that may not be at optimal times. Batteries can last at

least 2 months; several quick trips to change the batteries will result in long-term data on relative activity throughout the year, and in the long run, will be more cost-effective in determining the best time for a survey.

TARGETED SPECIES

Mexican long-tongued bat (CHME)

The life-history characteristics of Mexican long-tongued bats make it difficult to determine trends in populations over time. Females migrate from Mexico in the spring to form small maternity colonies, often only a few bats (<20), and up to ~50 bats (O'Shea et al., 2018). Colonies are scattered over the landscape in low abundance overall (O'Shea et al., 2018). They frequently roost in the twilight/entrance area, and often fly out of the roost if it is approached by humans, rather than fly farther back into the roost. They are able to use a wide variety of shelters spring through fall, from shallow to deep caves or mines. Some may also overwinter in the Tucson area if they can find sufficiently warm roosts and food resources. Observing sign in unoccupied sites is not always possible because of a colony's small numbers: a few fecal "splats" can easily be lost in the substrate of a mine or cave, or lost in large amounts of similar guano if LEYE also inhabit the site. To determine population size over an area and through time, many roosts would need to be identified and monitored as simultaneously as possible (bats may move from one site to another nearby from day to day or within short periods of time). Even if all 10 of Pima County's chosen monitoring sites were occupied by this species, they would need to be in the same area, such as Cienega Creek Natural Preserve, for interpretation of trends to be valid.

We monitored 2 sites occupied by CHME: Arkenstone and Karen's Caves. In late May, we observed 3 bats in Arkenstone, and 1 in Karen's Cave. In late September, there were 5 bats in Arkenstone and 2 in Karen's Cave, possibly the same adults plus young-of-the-year. Although both sites would work as permanent monitoring sites, interpreting data from surveys at isolated sites every 2-3 years would be impossible. For example, we have monitored a cave in the Rincon Mountains annually since 2005 that always has 2-4 CHME in the same place on our August and September surveys. This past August we saw no CHME. Should we conclude the population of CHME in the area had decreased? As it happened, we observed 1 CHME when we returned in September (and the creek below the site had resumed flowing). In 1999, researchers visited 24 of 39 historical CHME roosts in Arizona and New Mexico and observed bats at 75% of them; the largest number at a site was 17. They were unable to conclude whether the population throughout this area had decreased or increased (O'Shea et al., 2018). These examples illustrate the difficulty of determining real trends in population with both frequent, repeated visits to one site and over time at many sites over a large geographic area.

We suggest that multiple sites in a relatively small area be considered a single long-term monitoring site. For example, in past years, Pima County personnel monitored several soil-piping features along Cienega Creek, including Karen's Cave, that were occupied by CHME. Surveyors would need to survey all sites on the same day but ensure they were not double-counting individuals if bats were flushed out of one site and possibly flew to another. Buehman Canyon is another potential area; multiple mines could be surveyed in one day. The survey would have to be repeated at the same time of year, either before young are born (to count the adult population), or in late summer (which would include young-of-the-year).

Another method would be to monitor bats that feed at hummingbird feeders. Bats are active at feeders in the Tucson area, particularly late in the summer and fall when agaves are finished blooming. They usually start appearing before lesser long-nosed bats arrive, although they may not be noticed by homeowners. A small number of CHME may not drink enough overnight to make the level of sugar water used readily detectable. A few years ago, we set up a feeder by the bat tunnel at Colossal Cave to attract LEYE, and video-recorded both CHME and LEYE at the feeder. Cave personnel maintained the feeder. If a feeder at the cave were maintained again, video-recordings from a night or 2 in September would verify presence in the area, although not provide abundance, and would be at least as valuable as surveying an isolated roost. A strategically placed trail camera

that has high resolution and uses infrared light may be sufficient to record videos throughout the night and identify species if a high-resolution infrared-sensitive camcorder or surveillance camera is not available.

Arizona Game and Fish Department is now maintaining the Bat and Hummingbird Feeder Study website and program (https://www.azgfd.com/wildlife/backyard-bats/), formerly run by the Town of Marana. Most homeowners cannot distinguish CHME from LEYE, but enough pictures are posted that presence of CHME could be verified. Some website contributors have trail-cams; a group of monitors could be solicited from this group and record CHME use after being trained to distinguish between species. Although the monitoring area for CHME would be, for the most part, the north and east sides of Tucson rather than the Pima County Preserve System, it is a large geographic area, and would provide sufficient data to determine presence/absence over time, dates of arrival and departure, and possibly relative amounts of activity from area to area. Although activity at feeders is greatest about the same time as LEYE are present, CHME remain active in some areas well into December and some appear to overwinter. Data from volunteer monitors specifically studying CHME would provide valuable information on how and when this species uses an artificial food source throughout the year.

Townsend's big-eared bat (COTO)

Locating and monitoring maternity colonies is the most effective method of monitoring long-term trends for this species in the Pima County Preserve System. In fall through winter, bats are usually found roosting singly and scattered throughout mines and caves, which makes observations of bats during internal surveys opportunistic. However, maternity colonies abandon their roosts easily if disturbed (Sherwin et al., 2009, O'Shea et al., 2018), so monitoring methods must avoid disturbance. Ideally, maternity colonies should be monitored before adults give birth so that the adult abundance is compared from year to year, rather than later in the season when adults plus volant young-of-the-year are counted. Reproduction and neonate survival is likely more variable from year to year than adult abundance.

The 4 adits of Mine Site #1 in Rancho Seco seem to be a good long-term monitoring site because a maternity colony was observed in 2014 (BLM data from Bat Conservation International surveys) and again in 2018; however, monitoring many entrances simultaneously would be labor intensive. Additional inventory work would determine which adits are used and when throughout the maternity season; the colony seems to switch from one adit to another (XX1 adit in July, 2014 and XX2 in June, 2018). Maternity colonies may use different roosts during pregnancy, birth, and post-birth depending on their physiological needs and the microclimatic conditions of the roosts (O'Shea et al., 2018). Long-term monitoring should consist of video-recording all 4 adits on the same night to ensure that the evening emergence at the occupied adit is captured. Camera images should be of high enough resolution to distinguish COTO from other species that may be present. Surveyors should be aware that XX2 adit probably connects to the vertical pit next to the portal, and XX3 may also connect (BLM data); bats may also exit from these features. Internal surveys in the future are not recommended because of this species' sensitivity to human disturbance. If all entrances cannot be monitored on the same night, we recommend conducting emergence counts at XX1 and XX4 on one night, and XX2, XX3, and the vertical shaft on the next. Guano analysis using eDNA methods from pellets deposited after the start of this study confirmed that samples taken from XX1 were from cave myotis, and samples from XX3 were from big brown bats. Therefore, it is important to distinguish species. On high-quality video, COTO are relatively easy to distinguish from cave myotis and big brown bats because of their large ears.

Colossal Cave is also a good site for long-term monitoring. We confirmed presence of Townsend's bats in early September by capturing 18 bats; 39% (7) were female. Use of the cave by a maternity colony of this species should be confirmed by captures in mid-late July when females are in late-lactation or finished and young are volant. Even though the cave supports other species, it is an easy site logistically to harp trap and capture bats. Bats should not be captured more frequently than once every 2-3 years. Data on abundance requires high-speed, high-resolution cameras at 2 entrances and a camera with good resolution at the Bandit's Escape

entrance. If video-recording all 3 entrances simultaneously is not feasible, the Bandit's Escape entrance could be covered with a tarp or shade cloth so bats use one of the other entrances.

Both Mine Site #1 and Colossal Cave are challenging sites to monitor, in one way or another. Pima County personnel should continue to survey mines and caves to locate additional maternity colonies. If another one or two maternity colonies were located, the easier sites logistically to monitor might be monitored more frequently than the others, depending on scarcity of resources.

California leaf-nosed bat (MACA)

California leaf-nosed bats do not hibernate or migrate; they remain active in an area year-round, moving from roost to roost as their physiological requirements and roost microclimates change through the seasons. The longest distance recorded from a winter to summer roost was 93 km; the longest movement documented was 137 km (O'Shea et al., 2018).

This species can reduce its body temperature somewhat, but cannot hibernate or go into full daily torpor (O'Shea et al., 2018). Winter roosts must be warm enough, and insects must be plentiful enough within a night's flying distance from the roost (<20 km) so that bats can keep a positive energy balance. Colonies can range from just a few individuals to up to hundreds of bats. Summer roosts are often shallow or with multiple entrances and airflow; bats often roost near the entrance (pers. obs., O'Shea et al., 2018). Abandoned mines or caves with a variety of structural characteristics in a relatively small geographic area are therefore crucial to this species for survival.

Mine Site #2 is a historical roost that has been used for decades, and only MACA have been observed there (with the exception of 3 male cave myotis captured in July, 2006; Wolf and Dalton data). Colony size peaks sometime from November through January (not necessarily the exact same time every year) and varies from ~700-900 (Wolf and Dalton data). Bats are present year-round with the fewest numbers (~50) during maternity season (both males and reproductive females present July, 2006; Wolf and Dalton data). The ratio of males to females of 103 bats captured in January 2015 was 52% male and 48% female (Wolf and Dalton data). Therefore, there must be other maternity sites in the Tucson Mountains or nearby areas that are occupied by the hundreds of females that do not remain at the mine. The mine should be surveyed in both winter and summer to monitor trends in both types of use.

Mine Site #5 complex in Rancho Seco may be a good long-term monitoring site, but more inventory work is necessary at all the features in all seasons, and in nearby areas. We observed only small numbers of bats (~25) and in September, when bats were probably between summer and winter roosts. There could well be a roost nearby with larger numbers in either summer or winter.

We recommend that county personnel continue to survey sites for MACA on Pima County lands. Mine Site #7 is not far from Mine Site #5. We were not able to enter because of an active bee hive at the portal, but an internal survey in cold weather would provide recent information, and guano (some was found in the 2014 survey) could be collected and analyzed for species.

Lesser long-nosed bats (LEYE)

We did not observe lesser long-nosed bats, although the species is known to be present in eastern Pima County late-August through mid-October. Colossal Cave was historically used as a maternity roost until the late 1960s when it was extirpated by the installation of an exhaust system. Although the exhaust system was removed in 1988, and other species currently occupy the cave, LEYE have not returned, although they forage in the area in late summer/early fall. Efforts are being made to make the cave more attractive to LEYE; we would consider use of the cave as a late-summer roost as a successful conservation effort.

Arkenstone Cave is probably not inhabitable by large numbers of LEYE because of the small, gated entrance. To our knowledge there are no records of use by large numbers of this species before it was gated, so it is possible it does not provide suitable temperature profiles or structural requirements.

If there are mines or caves within the Pima County Preserve System on lands with patches of agaves, or near agaves, they should be surveyed late July through August, when this species is present in southeastern Arizona to feed on agaves.

Lesser long-nosed bats begin appearing at hummingbird feeders in the Tucson metropolitan area, and at nearby roosts, in mid-late August after agaves have finished flowering in the nearby mountain ranges. They have been found sharing sites and roosting among cave myotis in southeastern Arizona (pers. obs.). For this reason, we recommend video-recording evening emergences of large cave myotis summer roost-sites within about 30 miles of the Tucson metropolitan area in mid-late September, when the LEYE population in this area is greatest. Most or all cave myotis have left maternity/summer bachelor sites by this time, so if LEYE are present, they will be readily distinguishable and identifiable. We recommend that Mine Site #3 be surveyed at this time for LEYE. Mines in Tucson Mountain Park, particularly deep shafts, should also be surveyed mid-late September; in the last several years, LEYE have begun to use hummingbird feeders on the west side of Tucson in the Tucson Mountain foothills and to the west of the Tucson Mountains in the Picture Rocks area. They appear at feeders too early to have flown from known roosts in the Rincon or Santa Rita Mountains; we believe there may be a roost in the Tucson Mountains. Although it is unlikely that LEYE would use a gated site such as Mine Site #2, the mine should also be surveyed with an emergence count in late September. The entrance is small enough (and therefore, the camera can be close or zoomed in) that LEYE and MACA are distinguishable with a camcorder and good infrared lighting.

SITE PROTECTION

The long-term monitoring sites chosen by the county should be protected from human visitation, as should other known roosts of bats that inhabit County lands; otherwise, managers will not know whether a change in abundance or use at a site is due to human visitation at that site, natural changes in the bat population (due to a variety of unknown causes), or changes due to humans at another site used by those bats. Effective measures to protect bat roosts include restricting vehicle access near the site, fencing the site area with high chain-link fencing, and installing a steel “bat-friendly” gate.

Many of the mine features had barb-wire fences across the portal (particularly the adits at Mine Site #1) that had been partially or fully bent back sometime in the past to allow easy access for humans. We do not recommend replacing the fencing. The location of the fences was directly in the flight path of bats entering or leaving the mines and thus a potential danger to the bats. In addition, barb-wire fences are not an effective deterrent to keep people out of adits.

Except for lesser long-nosed bats, the other 3 targeted species adapt well to steel “bat-friendly” gates. To provide the best protection for the roost from human access and allow the most unobstructed flight access for bats, the gate must be well designed for the site’s physical characteristics and anticipated level and type of vandalism, located in the best place, installed at an appropriate time of year to prevent disturbance to bats, and constructed by competent people. The colony should be monitored before and after the gate is installed (at the appropriate season) in addition to the regular monitoring schedule. If the roost is used by other species, these species should be considered in a gating project (e.g., large colonies of cave myotis do not adapt well to gates). We would not recommend adversely affecting a colony of cave myotis in order to gate a roost of another species. Another potential disadvantage of a gate is that it also keeps out other animals that may use the site, such as desert tortoises, javelina, and vultures.

The gate on Mine Site #2 needs an additional top bar to close the space where someone dug out the rock above the gate. At Colossal Cave, the county and cave personnel are interested in re-routing the wild cave tour route to avoid the bat room so that bats can roost undisturbed throughout the year. This would be possible if the gate at the Bandit's Escape entrance was replaced with one that could be easily opened and closed and used as the route's exit point, rather than the bat-room tunnel.

CONCLUSIONS

We observed Townsend's big-eared bats at 2 sites (Colossal Cave and Mine Site #1), California leaf-nosed bats at 2 sites (Mine Site #2, 5), and Mexican long-tongued bats at 2 sites (Arkenstone and Karen's caves). We did not observe or find evidence of use by lesser long-nosed bats at any of the sites. We also confirmed cave myotis at Colossal Cave through capture, and assume that large summer bat colonies at Mine Site #3 and 4 were largely cave myotis. Additionally, through eDNA analysis of bat guano, we confirmed that big brown bats were using at least one of the adits at Mine Site #1 and cave myotis were using a different adit at Mine Site #1 as well as Mine Site #6.

Bats are a difficult group of animals to monitor. Interpretation of data for long-term trends in site occupancy or abundance at a specific site or over a geographic area of interest is difficult. Each species has different natural history characteristics including roost and foraging requirements that change throughout the year and differ between species, age groups, and reproductive condition of individuals. Selecting only 10 sites to monitor 4 very different species is not sufficient to provide good information on each species. In addition, given the geographic extent of the Pima County Preserve System, its diverse ecosystems, and the very small number of monitored sites for each species, the scope of inference for the results of monitoring would not be valid beyond each individual site.

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SITE REPORTS

ARKENSTONE CAVE, Colossal Cave Mountain Park

Bat Surveys, 2018, by Bat Research and Consulting

Location/access: Sensitive information

Participants: Sandy Wolf, Dave Dalton

Objective: Video-record bat evening emergence from cave to determine bat species and abundance

Results summary:

May 30, 2018: 3 Mexican long-tongued bats (*Choeronycteris mexicana*)
September 26, 2018: 5 Mexican long-tongued bats

Methods: The only good place to video-record the emergence at this cave is right next to the entrance pit, looking down at the gate. The ideal position is to view an entrance from the side so emerging bats fly across the field of view, but that is not possible at this site if the goal is to have an image in which species can be identified (providing visual characteristics are unique). Equipment: Axis 1354 camera, 2 near-infrared lights (Wildlife Engineering Model IR6, peak emission 830 nm, spectral bandwidth 15 nm fwhm, 20° dispersion angle, total radiant flux 1.6 W each), Toshiba laptop computer, 2 5-Ah, gel-cell batteries.

Weather data:

May 30: Sunset: 1924 h. Weather at 1924 h: 84°, wind B1, clouds 10%- high, thin. Moon 99% illuminated, rose at 2029 h. Weather at 2100: 77°, wind B1, clouds 0%

September 26: Sunset: 1817 h. Weather at 1830 h: 81°, wind B1, clouds 0%. Moon 98% illuminated, rose at 1936 h. Weather at 1950: 79°, wind B2, clouds 0%

Result details:

May 30: At 1919 h, we saw 2 bats foraging high above the road (likely canyon bats, *Parastrellus hesperus*, which often emerge before sunset). One of us watched the cave entrance continually after the camera was set up, about 1900 h. At 1942 h, we started to record, and recorded until 2055 h. Three bats emerged between 2025 and 2033. Many crickets emerged during our visit, as well as the biggest conenose I have ever seen. We heard one poor-will.

Later, we watched the video and positively identified (100% confidence) 3 Mexican long-tongued bats emerge at 20:25:06 h, 20:26:30 h, and 20:33:30 h. We identified them by the muzzle shape or tail; both characteristics are unique to this species. We were surprised at how late the bats emerged.

September 26: At 1829 h, we started to record, and recorded until 1947 h (all bats were likely to have emerged by this time). Five bats emerged between 1841 and 1931 h. Many crickets emerged during our visit, and at 1937 h, a ringtail went into the cave carrying a rat. The ringtail poked his head out at 1941 h (without the rat) and went back in the cave, then came out at 1944 h without the rat.

Upon watching the video in slow motion, we positively identified 5 Mexican long-tongued bats emerging. We had 100% confidence for identification for 4 of the bats, and about 80% confidence for the fifth. We identified them by the muzzle shape or tail membrane; both characteristics are unique to this species.

Condition of site: There was no sign, including tracks, of recent human activity in the area on either visit. The cave is well protected; the entrance is small, hidden, and gated. The Arizona Trail and road are visible from the site, but the cave entrance is not visible from the trail, road, or hillsides.

Recommendations: Because there are invertebrates in the cave whose status is considered vulnerable, emergence counts are the preferred method of monitoring this site. To fully understand the seasonal use and peak colony size of this cave by Choeronycteris, surveys should occur about every 2 weeks from May through October or November. To determine if lesser long-nosed bats (*Leptonycteris yerbabuenae*) are present, surveys should occur August through mid-October.

We surveyed this cave at the peak of lesser long-nosed b at abundance in the area (26 September) and observed none. We do not expect this species to use the cave except perhaps by an individual or 2; the entrance is small and the gate further impedes access. Lesser long-nosed bats are less maneuverable than Mexican long-tongued bats and roost in much larger numbers; it is unlikely that they would find this site attractive because of the restricted access.

COLOSSAL CAVE, Colossal Cave Mountain Park

Bat Survey, 2018, by Bat Research and Consulting

Location/access: Colossal Cave

Participants: Sandy Wolf, Dave Dalton, Melanie Bucci. Several park personnel and their relatives were present and helped with data recording or observed the capture/processing activities.

Objective: Video-record bat evening emergence from all 3 entrances of the cave to determine number of bats roosting in the cave, and capture bats emerging from the bat-room tunnel entrance to determine species, sex, age, and reproductive condition.

Results summary: September 8, 2018: Captured: 99 Cave myotis (*Myotis velifer*), 18 Townsend's big-eared bats (*Corynorhinus towsendii*). Abundance estimate from video-recordings: including all bats captured, a total of 537 bats (net number out) emerged.

Methods: We video-recorded (from outside the entrances) the main cave entrance, the bat-room tunnel entrance, and the Bandit's Escape entrance. Equipment: Axis 1354 camera, records to Toshiba laptop computer via wired Ethernet, Sony SR11 camcorder, Axis 214 PTZ camera, records to Dell computer via wired Ethernet, 5 near-infrared lights (custom lights using Osram Ostar LED chip arrays SFH 4730, peak emission 850 nm, spectral bandwidth 30 nm fwhm, 32° dispersion angle, total output radiant flux 3 W each), solid state laser with dispersing optics, peak emission 808 nm, spectral bandwidth 2 nm fwhm, ~30° dispersion angle, total radiant flux 5 W, 3 20-Ah lithium-ion batteries.

We captured bats in a harp trap placed a few feet inside the entrance to the bat-room tunnel. Bats were removed from the trap bag and placed in individual cloth bags and taken several yards away to the processing table. Data recorded on each bat included species, sex, age, reproductive condition, and general body condition. Bats were marked with a temporary mark on their head, and released.

Afterwards, equipment was decontaminated according to the most current white-nose syndrome decontamination protocols.

Weather data: Sunset: 1841 h. Weather at 1841 h: 85°, wind calm, clouds 3%. Moon 1% illuminated, set at 1819 h.

Result details: Bats began flying at 1853 h. Recording ended at 2046 h at the main entrance, 2102 h at the bat-room entrance, and 2048 h at the Bandit's Escape entrance.

The first bat captured in the harp trap was at 1848 h. All bats captured until 1935 h were cave myotis, which was when the first Townsend's big-eared bat was captured. The trap was closed at 2015. Although some bats flew around or through the trap without being captured, we captured 99 cave myotis, and 18 big-eared bats. To reduce the time bats were held, we identified most of the cave myotis and some of the big-eared bats at the trap, sexed them, and released them without holding them for further processing. For all cave myotis captured, 40% (40) were female, 60% (59) were male; 37% (7) of the 19 males we examined had descended testes. For big-eared bats, 39% (7) big-eared bats were female, 61% (11) were males; 20% (1) of the 5 males we examined had descended testes.

We counted 502 bats (net number out) emerging from the cave's 3 entrances, plus 35 held for processing, for a total of 537 bats: 86 from the main entrance, 383 from the bat-room tunnel (plus 35 captured and held), and 33 from Bandit's Escape.

Condition of site: The county and cave personnel are interested in re-routing the wild cave tour route to avoid the bat room. This would be possible if the gate at the Bandit's Escape entrance was replaced with one that could be easily opened and closed and used as the route's exit point, rather than the bat-room tunnel. This is not under the scope of the monitoring contract; the county, with our assistance, is currently planning on replacing the existing gate in the near future.

Discussion/Recommendations: We surveyed in September because it was likely to be a time of peak abundance. We assume that most of the 537 bats occupying the cave were cave myotis; 85% of captured bats were cave myotis. According to park staff (Bill Savary, pers. comm.), pallid bats also occupy the cave at times. A harp-trapping session when this species is present would provide information on how pallid bats use the cave.

Previous work in 2011 (Wolf and Dalton) confirmed use by a maternity colony of Townsend's big-eared bats. Our survey this year was too late in the season for us to see epiphyseal gaps in metacarpal-phalangeal joints, which is indicative of juveniles. Resurveying in late July next year would confirm whether this species continues to use the cave as a maternity site.

Data for cave myotis from 2011 showed use primarily by males. The cave myotis occupying the cave in summer therefore may be a bachelor colony. If it were a maternity colony, assuming that each bat gave birth and the single youngster survived, it would be expected that about 75% of the bats would be female; data from this year showed 60% male. Cave myotis migrate in September, so it is possible that the cave is used as by migrating bats as well as the summer colony. Resurveying in late July (before migration) next year would provide more information on the use of the cave by cave myotis.

The harp trap impeded natural exit behavior of bats from the bat-room tunnel so conclusions cannot be drawn from the relative proportions of bats emerging from each entrance (16% from main, 78% from bat-room tunnel, and 6% from Bandit's Escape). In addition, there was a tour along the main tour route that might have affected behavior at the main entrance. Monitoring the entrances on evenings when bats are not captured and there are no tours would provide information on the relative use of each entrance and how it changes throughout the year. Cave Management could then decide whether or not to alter tour times during certain times of the year.

Harp trap, Colossal Cave

Cave myotis, Colossal Cave

Townsend's big-eared bat, Colossal Cave

KAREN'S CAVE, Cienega Creek Natural Preserve

Bat Surveys, 2018, by Bat Research and Consulting

Location/access: Sensitive information

Participants: Sandy Wolf, Dave Dalton

Objective: internal survey of cave, deploy and collect guano sheets

Results summary:

May 29, 2018: 1 Mexican long-tongued bat (*Choeronycteris mexicana*), set 3 guano sheets
September 29, 2018: 2 Mexican long-tongued bats, collected guano sheets, collected insectivorous bat guano from sheet in large room for DNA analysis.

Methods: We used white light for safety because we were unfamiliar with the site (29 May), because snakes or other animals could be present, and to identify bats as quickly as possible (red light is less bothersome to bats but insufficient for safety or quick identification). We used fine mesh bridal veil, laid down and held in place with rocks, to collect guano. We prefer bridal veil to sheets of plastic because it does not affect the airflow and humidity underneath it and therefore minimizes effects on invertebrates and microbiota.

Result details: May 29: We entered the west entrance to the cave using white light. In the first room, we saw a bat flying around. It hung up above us in a high chimney-like area in the ceiling where there was a small grate-like structure reinforcing the ceiling. We identified species with 100% confidence by its long cylindrical muzzle; it took only a few seconds, and then we moved out from under it and did not shine a light near the bat again. We put a large piece of bridal veil underneath the chimney area and across a large part of the room. We surveyed the rest of the cave. Although we saw no guano pellets from insectivorous bats or fecal splats from nectar-feeding bats, we put 2 small pieces of veil down under likely roosting spots near the east entrance where the ceiling was highest. Throughout the cave there was a large amount of scat from pack-rats and mice, as well as javelina and possibly ringtail. There was an old javelina skull outside the west entrance, mostly buried in debris.

September 29: We entered the west entrance to the cave using white light. In the first room, we saw a bat flying around, and then fly into the back part of the cave. We surveyed the rest of the cave. We collected one of the guano sheets near the east entrance (no guano on it); it had been dislodged, and the other one we put down was missing. We heard no bats in the inaccessible room near the east entrance. We returned through the cave and saw 2 *Choeronycteris* (100% confidence for species based on muzzle) hanging up in the big room at the top of the chimney. The large guano sheet in the big room had been mostly moved by water flow and was partly under dried mud. There were a few pellets of guano on it, which we collected for species identification through DNA. Throughout the cave there was a large amount of old scat from pack-rats and mice, as well as javelina. Except for the few guano pellets we collected that looked somewhat fresh, we saw only a couple others that were old. There was no evidence that a colony of an insectivorous bat used the cave, although it is possible that guano was washed away. The old javelina skull outside the west entrance that had been mostly buried in debris in May was washed down a few feet and more visible.

DNA analysis of guano: Pending results from laboratory.

Cave description: This is a soil piping cave that has been stabilized with man-made structures. The west entrance is a metal, corrugated culvert 5 feet in diameter and set in the cliff face. It opens into an oval room about 20 feet long, 15 feet wide, and about 10 feet high at its highest point. At the far end of the room, a metal culvert (30-inch diameter), set at floor level, leads to a narrow, breakdown passage about 40 feet long that

varies in height from about 3- 12 feet high. The passage ends with a metal corrugated culvert, 48 inches in diameter and 5-6 feet long, that stabilizes the east entrance to the cave. Just before the east entrance is a room off to the north. There are 3 holes that lead into this area but are too small for human access. We could see a large open space with a straight vertical wall, but could not see the ceiling, floor, or how big the room was. We listened for bats but did not hear anything. The east entrance is in a large eroded, sinkhole area, and is slightly higher in elevation than the west entrance. Another large sinkhole area to the north has a small hole, about 2 feet wide by 4 inches high, that likely leads to the room we could not get into. We also climbed up the cliff from near the west entrance, went under the fence (the Cienega Preserve boundary), and down to the sinkhole with the east entrance. It is easy to get from one entrance to the other outside the cave, with only a little scrambling.

Condition of site: May 29: The only sign of human activity in the vicinity of the cave was one old plastic water bottle between the cave and the creek.

September 29: There was no sign of human activity in the vicinity of the cave but a large amount of water had come through the area rather recently. The structure of the cave appeared the same as in our previous visit. Soil-piping caves are inherently unstable, but we saw no signs of imminent collapse.

Recommendations: Because there is an area of the cave that humans cannot access, an internal survey cannot provide information on bats' use of this area. Although the entrance to the room is very small, it is possible for a maneuverable bat (e.g., Townsend's big-eared bat) to use it. Emergence counts at times when bats are known to be there are needed to supplement data from internal surveys. Both entrances must be recorded.

It is unlikely that lesser long-nosed bats use this site as a day-roost; we surveyed at the peak time of abundance for this species in the area. We saw no feces from nectarivorous bats. Given the dirt floor, large amount of other scat (rodent, javelina) and evidence of water running through parts of the cave, it is not surprising that we saw nothing if only a small number of Mexican long-tongued bats use the cave. If large numbers of nectarivorous bats (i.e. lesser long-nosed bats) roosted here, I think we would have seen sign in areas where water didn't flow.

Mine Site #3, Buehman Canyon Preserve

Bat Surveys, 2018, by Bat Research and Consulting

Location/access: Sensitive information

Participants: Sandy Wolf, Dave Dalton

Objective: Video-record bat evening emergence from mine to determine abundance

Results summary: June 2, 2018. Over 1,000 bats emerged from the shaft.

Methods: We set up the camera outside the fence on the west side of the shaft, several yards to the west of the fence. The lights were on a U shaped bracket set up at the fence. It's preferable to have the lights with the camera, but the light would have reflected off the fence wires and closed the iris. The size of the shaft meant that the camera's field of view was too wide, and therefore the images of the bats too small, for species identification. Equipment: Axis 1354 camera, records to Toshiba laptop computer via wired Ethernet; 4 near-infrared lights (custom lights using 4 Osram Ostar LED chip arrays SFH 4730, peak emission 850 nm, spectral bandwidth 30 nm fwhm, 32° dispersion angle, total output radiant flux 14 W), 2 20-Ah lithium-ion batteries. Sony SR-11 camcorder.

Weather data: Sunset: 1927 h. Weather at 1927 h: 87°, wind B1-2, clouds 0%. Moon 87% illuminated, rose at 2253 h.

Result details: We saw 2 deer on the hike in. We checked the boiler house and saw no bats, only pack-rat nests and debris. At 1935 h, we started recording when a bat came out and went in (light-sampling). The exit started at 1936 h. Most bats flew over the top of the fence on the north, west, and south sides. Only a few flew over the east chain link fence, and only a few flew through the strands of barbed wire. I hand-held the camcorder and video-recorded the emergence for several minutes until 1958 h. During the emergence, we had to move the Axis camera's view higher; the view through the fence showed bats circling rather than flying out. We stopped recording at 2014 h, after the emergence was over.

We watched the video-recording and estimated at least 1,000 bats exiting (net out). We assume the bats were cave myotis (*Myotis velifer*) based on their exit behavior and acoustic data (Iris Rodden, Pima Co.) from 2016 and 2017.

Mine Description: Shaft is about 8 feet x 12 feet with 2 concrete hoist pillars at the west end. A fence surrounds the ant-lion collar. The east side of the fence is 6 ft high chain link with barbed wire above it. The other 3 sides consist of 6-strand barbed wire. Juniper, hackberry, and mesquite are growing around the shaft. Vegetation partially obstructs access for bats at the east end, inside the fence.

We did not have time to check the double adit above the shaft. Although we allowed plenty of extra time, we used it getting to the site. Neither of the recommended parking areas worked out.

Condition of site: We saw no signs of recent human visitation.

Recommendations: On the north and south sides of the shaft, there are strands of barbed wire about 8 feet high that are an obstruction and potential hazard to bats, particularly juveniles. We were able to remove the strand on the south side because it was unattached and dangling midway along its length. The strand on the north side should be removed the next time county personnel are in the area.

The video did not lend itself to a good count. With more experimentation with camera and light location, we may have found a better view, but would have lost recording part of the emergence. As it was, we moved the

camera angle slightly in the middle of the emergence, which briefly disrupted the recording and changed the view. Placing the lights at the fence prevented reflection off the wires which meant the scene was relatively well lit, but also caused bats to have shadows, which can result in double-counting when it's not clear what is a bat and what is a shadow. To get the best video of an emergence, a site visit and test video (when bats are exiting) are necessary to determine the optimal camera placement, field of view, and light set up. The test video isn't designed to get an accurate count, but is necessary to determine optimal camera placement and view to obtain the most accurate counts in the future.

This site does not seem to be a good one to include as one of the County's 10 monitoring sites. It seems to be primarily, or solely, a cave myotis site, which is not one of the 4 cave/mine-dwelling species targeted for monitoring. Any other species occupying the shaft during the same season as the cave myotis would not be able to be distinguished. However, this site should be assessed during mid-late September (i.e., exit count video) for potential use by lesser long-nosed bats (LEYE). Most cave myotis are gone by then, and with a camera with good resolution and sufficient infrared lighting, LEYE should be distinguishable (lack of tail membrane is diagnostic) if present. It would be difficult to identify species occupying the shaft at other times of the year by video-recording, although if it has never been surveyed during the cooler seasons, it should be. The effort required to get to the site, however, is significant, and might be a factor as to the site's suitability.

Mine Site #2 Tucson Mountain Park

Bat Surveys, 2018, by Bat Research and Consulting

Location/access: Sensitive information

Participants: Sandy Wolf, Dave Dalton

Objective: May 13, 2018: Site visit to design repairs to gate. Opportunistically, determine if bats are present, number and species.

December 23, 2018: Video-record bat evening emergence from cave to determine abundance

Results summary: May 13, 2018: Observed 7 California leaf-nosed bats (*Macrotus californicus*) from outside the gate. December 23, 2018: 738 California leaf-nosed bats
Methods:

May 13, 2018: We arrived at the mine at 0955 h. We approached the gate quietly and sat next to it for several minutes. From past experience, California leaf-nosed bats (*Macrotus californicus*) commonly roost just past the gate in the warmer months. There was sufficient ambient light to see several yards on the other side of the gate.

December 23, 2018: We arrived on site at 1650 h. We set up the camera to the west of the mine entrance, up against the hill. We were ready at 1710 h. Equipment: Sony SR-11 camcorder, 2 near-infrared lights (Wildlife Engineering Model IR6, peak emission 830 nm, spectral bandwidth 15 nm fwhm, 20° dispersion angle, total radiant flux 1.6 W each).

Weather data: December 23, 2018: Sunset: 1925 h. Weather at 1925 h: 65°, wind B0, clouds 3%. Moon 99% illuminated, rose at 1846 h.

Results: May 13, 2018: After a couple minutes, bats flew from farther down the adit and from the alcoves off the adit just past the gate. They flew around, coming right up to the gate, and hung up a few feet from the gate. We saw 7 at the same time. They hung up as individuals, sometimes flapping their wings. This is typical behavior. We are 100% certain these were California leaf-nosed bats; they were only a few feet away, and their ears, face, pelage, body shape, and flight pattern were readily identifiable.

December 23, 2018: Bats began exiting at 1739 h. We recorded from 1739-1840 h. As usual, bats circled around the entrance area, including over our heads and very close to us, as if they were checking us out. We counted a total of 738 bats (net out).

Condition of site: We tested 2 original gate keys and 2 copies; the originals worked, but only one of the copies did.

The gate was installed in spring 2007. Sometime between our visits in December, 2009 and May, 2011, someone dug out the back (roof) of the mine above the gate enough so that a human can climb over the gate and enter the mine. At the widest point, there is a space about 15 inches high above the top bar. We took measurements and designed an addition to the top of the gate to prevent human access, using 3-inch square tube, hardfaced. The addition could be prefabbed to minimize the amount of cutting and welding required on site.

The piece of steel that the County bolted to the gate as a repair was lying in the alcove next to the gate. Some of the gate bars were tagged. Someone had tried to drill a hole in the second bar from the top; this can be

repaired when the additional bar is installed. There was a little trash just inside the gate but did not appear to be recent.

There were water bottles and trash by the entrance to the ungated mine next to this one. Based on surveys in the past, this mine was used infrequently by bats. If future surveys confirm it is still used infrequently, it could be filled in (any bats present would need to be excluded first) or gated to keep people out.

The fence at the parking area was partially knocked down; people can just step over it to go across the wash and up the old road. The road showed no signs of ATV traffic and is getting overgrown. There was no sign of recent ATV traffic at the mine site on either visit, although there is access from the west.

When we got back to the vehicle after each trip, we talked to XXXXXXXX, the homeowner by the parking area. He acts as an unofficial steward and discourages people (successfully) from riding their ATVs through the fence and up the road to the mine. He also has a brush pile near the fence to keep people from breaking down the fence in one area. He said he was interested in talking to county personnel about being an official site steward and being kept informed about activity/work at the mine. I didn't ask for contact information but in 2013 it was XXX-XXXX. We told him we hoped that the gate would be repaired in the spring.

Discussion/Recommendations: Based on previous data from 2005-2009 (Wolf and Dalton, unpublished), abundance seems to be a bit lower than in the past, but we don't have recent data to compare (Table 1). We know that winter (November-January) is the season of peak abundance, but it would take multiple emergence counts over multiple years to narrow down the peak, and determine how variable it is from year to year. We (Wolf and Dalton) have video from emergences in December 2012, December 2013, and January 2014 that are uncounted (we video-recorded for fun and never got a chance to count). If time ever permits, we will count the recordings. At this point we do not know if visitors have disturbed the bats since the gate was breached, although it is likely. Once the gate is repaired, numbers may increase.

Table 1. Abundance estimates of *Macrotus californicus* at Mine Site #2. Data from emergence counts by Wolf and Dalton.

DATE	NUMBER OF BATS
June 28, 2004	14
September 3, 2004	81
September 9, 2004	43
May 25, 2005	49
June 15, 2005	35
November 2, 2005	568
December 24, 2005	624
February 13, 2006	504
November 15, 2006	258
December 15, 2006	883
December 30, 2006	814
January 16, 2007	762
February 15, 2007	661
February 20, 2007	442
March 18, 2007	392
April 25, 2007	346
November 22, 2009	919
December 12, 2009	851
December 23, 2018	738

Mine Site #4, Rancho Seco

Bat Surveys, 2018, by Bat Research and Consulting

Location/access: Sensitive information

Participants: Sandy Wolf, Dave Dalton

Objective: Video-record bat evening emergence from mine

Results summary: June 9, 2018: Total net bats out: ~7,500 Assume bats were all or almost all *Myotis velifer*.

Methods: The mine consists of a large pit on a hillside with a shaft at the west end and an adit above it, and a shaft at the east end. We set up the Axis camera on the north side of the pit looking down and across the west shaft and adit. The lights were on a U shaped bracket on the tripod with the camera. Equipment: Axis 1354 camera, records to Toshiba laptop computer via wired Ethernet; 4 near-infrared lights (custom lights using 4 Osram Ostar LED chip arrays SFH 4730, peak emission 850 nm, spectral bandwidth 30 nm fwhm, 32° dispersion angle, total output radiant flux 14 W), 2 20-Ah lithium-ion batteries. We set up a second camera on the south side of the pit looking north down and across the east shaft. Equipment: Sony SR-11 camcorder, 2 near-infrared lights (Osram, same as above, total output radiant flux 7 W), 1 lithium-ion battery. We started recording a few minutes before bats began to emerge and stopped about 65 minutes later after the exit had ended.

Weather data: Sunset: 1930 h. Weather at 1930 h: 82°, wind B1-2, clouds 20%. No moon. Rain over the Santa Rita Mountains to the east in the afternoon, but none in the Rancho Seco area.

Results: Total net bats out: 7,449 Net bats out from west shaft/adit: 5,231 Net bats out from east shaft: 2,218. Total should be reported as ~7,500 bats.

Two owls exited the west shaft at 2021 h.

Condition of site: We saw one water bottle on the trail not too far from the parking area. There was a rather large pile of water jugs and bottles a few feet off the trail by the wash, where the trail runs along the west side of the wash southeast of the mine. There were 2 metal camp chairs folded up on the bench by the pit that looked weathered and unused in a while. The east side of the pit is fenced on the bench but that is the only barrier. The sides of the pit do not look stable; we took care to stay back from the edge. The shafts and adit are inaccessible without ropes, and even then look pretty sketchy. Don Carter drew a partial map of the features based on his survey, which gives sufficient information to inform future surveyors that this site is best monitored externally through emergence counts, acoustic data, and possibly mist-netting.

Discussion/Recommendations: This is not an easy site to monitor, even with high-speed, high-sensitivity cameras and strong near- infrared lights. The size of the pit meant that it required 2 cameras, and that each camera's field of view was too wide, and therefore the images of the bats too small, for species identification. Some bats flew close to the west shaft camera, and we were able to tell they were not Townsend's big-eared or California leaf-nosed bats, but for most bats, identification isn't possible. In addition, although we placed the cameras at the locations best suited for an accurate count at each shaft, there was vegetation that partly obstructed the fields of view and reflected some of the infrared light. Reflected light caused each camera's iris to close somewhat. The result was that the scene was not as well lit as it could have been. Although we were aware of the problem when we set up, the vegetation was at the very edge of the pit and not where we could safely remove it. In spite of the problems, upon review, it looks like the west shaft recording yielded fairly accurate results. The east shaft was not as easy to count accurately because bats from the west shaft flew into the view, lighting was not as good, and there was more vegetation obstructing the view of bats flying.

We assume all, or most, bats emerging were cave myotis (based on past County data, and our observations). If any of the 4 cave/mine-dwelling species targeted for monitoring by the County were present (or other non-target species), it would be difficult to obtain good data on abundance by video-recording; the site does not lend itself to being able to identify species visually with video.

To positively identify species of bats inhabiting the mine, bats could be captured in mist-nets set on the bench below the pit. On 12 September 2013, 10 Townsend's big-eared bats were captured in addition to cave myotis (Pima County data). Many, possibly most, bats we observed flew high above the bench. To sample them, nets could be placed where we put the cameras; as we sat next to the cameras, we both had bats flying low and directly over our heads. Footing would be unsteady, especially on the rock pile on the south side of the pit, but with care could be done.

This site may not be a good one to include as one of the County's 10 bat monitoring sites. It is not possible to conduct internal surveys or place guano sheets. Video-recording emergences requires 2 cameras and many lights, and does not yield high-quality video or permit species identification. Acoustic data only give presence/absence, and may miss Townsend's bats because of their call characteristics. Capturing bats by mist-netting is feasible but invasive and is safest only on the bench, which is not close to where bats exit. It is likely to be successful only in the warm season. In addition, failure to capture a targeted species cannot be interpreted as absence of that species in the mine.

Mine Site #1, Rancho Seco

Bat Surveys, 2018, by Bat Research and Consulting

Location/access: Sensitive information

Participants: Sandy Wolf, Dave Dalton

Objective: Internal surveys, deploy and collect guano sheets, collect guano for DNA analysis for species identification.

Results summary: June 9, 2018: A small maternity colony with half-grown young (~50 adults and young) of Townsend's big-eared bats (*Corynorhinus townsendii*) in XX. Deployed 2 guano sheets in XX, 3 in XX, 1 in XX.

December 1, 2018: One torpid Townsend's big-eared bat (*Corynorhinus townsendii*) in XX. Collected guano from a guano sheet in XX and from scattered guano in XX.

Guano analysis from XX: cave myotis, from XX: big brown bat

Methods: We conducted internal surveys of the adits. We had descriptions of the features from a previous survey by BCI so we had a general idea of what to expect, although conditions can change greatly in 4 years. Safety gear included hard hats, white light, and a gas detector. We used bridal veil for guano sheets. As we surveyed the adits, we checked for snakes and other animals, guano and insect parts, and hazards. In June, when we encountered bats, we immediately retreated. One of us returned with a camcorder and infrared lights, approached slowly and low to the ground and recorded the colony; no one approached closely enough to cause the bats to fly. Once a group of bats was encountered, we ended the survey in that adit; we did not continue past the bats to prevent them from flying out of the mine or otherwise disturbing them.

Results:

XX1 June: No bats. A trench about 20 ft. long leads to the portal, which is half obstructed by slope creep. The short adit ends in a collapse. One guano pile part way in was old and moldy; we laid a guano sheet over it. December: No bats. The guano sheet we put over the guano pile part way in was gone although the rocks that held it in place were there. There were 2 small areas with scattered guano several feet in from the older pile. We took samples of guano for DNA analysis from the scattered guano and from the edge of the older pile. Later eDNA analysis of this pooled guano sample indicated that it was from cave myotis. Temperature inside the mine was 64° F.

XX2 June: Adit is adjacent to vertical pit. Just a few yards into adit (~25 ft.) was a colony of Townsend's big-eared bats with young. We retreated, came back with video-camera and infrared lights and recorded for a couple of minutes. Ended survey there. Air was blowing into the portal (~97° F outside). The barbed wire fence across the trench just outside the portal was bent back, but there was a wire hanging out into bat flight area. We bent it back and secured it to increase the unobstructed flight area and prevent injuries from bats flying into it. December: No bats seen. Many piles of guano along passage. Some guano was mothy (feels silky smooth from moth wing scales), brown and black, probably Townsend's. Some was more gritty (beetle exoskeleton parts) and similar to that in XX1. We surveyed the adit as far as the ore pile with the winze on the other side and the stope above. It was too dangerous to try to navigate around the winze and continue the survey in the passage beyond the winze (the 2014 BCI survey stopped here also). It would be possible to continue only with additional people and surveyors roped in. Air was blowing, probably from the winze, which probably connects to the vertical shaft nearby. The top of the stope was not visible. There was a wooden ladder leading up to the other stope, which is mid-way between the portal and the winze; the 2014 BCI didn't mention the ladder so

whether it is recent or not is unknown. There was an amazingly large amount of ringtail scat throughout the adit and in the stope.

XX3 June: No bats. Cold air (66° F) blowing strongly out of portal. Going in, first drift to right had small area of large, coarse (beetlely) guano (probably big brown bat guano); we laid down a guano sheet over guano. First drift to left, 18 inch diameter scattering of guano, medium-sized, beetlely (probably cave myotis) under a large flat rock back, put down guano sheet. Largest area of guano (probably Townsend's big-eared bat) between first and second ore chutes, vari-colored, medium-sized, mothy, rather fresh. Put down guano sheet over this area. Air blowing down from stope above fourth (and last) ore chute. Several vulture feathers (white-tipped feathers, immature bird) just past fourth ore chute. Scattering of guano along middle of passage began between second and third ore chute to portal, becoming more continuous and denser towards portal. December: One torpid (ears curled up) Townsend's big-eared bat hanging on the timber on the second ore chute; air was flowing down the stope/chute. Two guano sheets were missing; the one in the first right-hand drift (going in) was present and had a small amount of guano on it. Collected some for DNA analysis. Analysis of this guano sample later confirmed its identity as being from big brown bats.

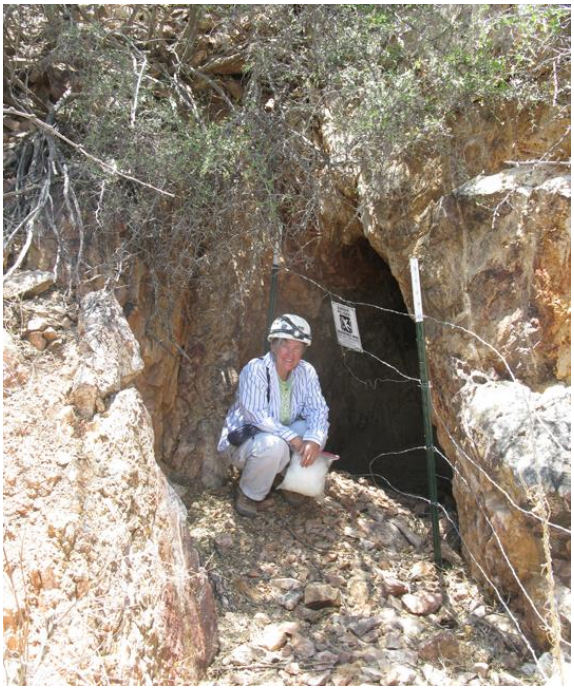
XX4 June: No bats. Half-dozen guano piles plus small scatterings along sill edges near ribs. Laid 2 guano sheets over largest densest piles; one was older and moldy, with small, stained pocket in back above guano; the other guano area was fresher and smaller. Two small black-tailed rattlesnakes. Turkey vulture feather part-way back. December: No bats. One of the 2 guano sheets was missing. The one present had some guano on it with a couple small triangular insect wings, but the guano looked the same as that in XX3 so we didn't collect it. There were also green insect wings inside the portal, indicating use of the mine by night-roosting bats.

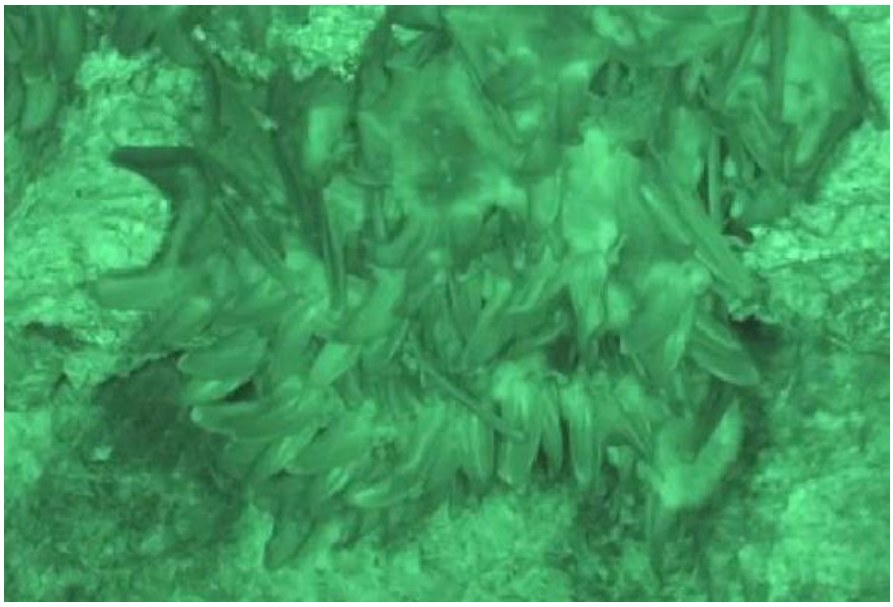
Condition of site: We saw no signs of recent human visitation. Tracks up the hillside between adits and between the highest mine and the wash are well-developed in places but are not continuous, so it is unclear whether they were made by people or javelina. There is debris/equipment left from mining days, but nothing recent. The fences across adit entrances are all partially bent back, which allows easy human access but also allows better bat access. In December, the dirt road leading to the mine from the main east-west road through Rancho Seco was much more overgrown than in June and showed no signs of being traveled on recently.

Discussion/Recommendations: Townsend's big-eared bats likely use all 4 adits at various times as microclimates in the adits and physiological needs of the bats change throughout the year. In 2014, BCI surveyors observed a colony of 150-200 Townsend's bats in XX1 on 24 July 2014. We likely only saw part of the colony and the rest was farther back in the mine. From the guano, cave myotis and big brown bats also inhabit the mine.

Fences were placed very near the portals. To allow for better bat access, they should be moved farther away from the portal, or kept bent back. Townsend's big-eared bats are maneuverable, but youngsters are less so. This species seems to adapt well to steel gates

Some critter (pack rats?) made off with 4 of the 6 guano sheets we put down in June. We searched the mines unsuccessfully for them. Perhaps the bridal veil provided coveted and superior nesting material. It would be worth trying plastic sheeting rather than bridal veil in the future. Although plastic affects the microclimate underneath it, it is not likely to have adverse effects to the mine when used as temporary guano sheeting.


XX2 portal


Townsend’s big-eared bats in XX2

Mine Site #5, Rancho Seco

Bat Surveys, 2018, by Bat Research and Consulting

Location/access: Sensitive information

Participants: Sandy Wolf, Dave Dalton

Objective: Internal survey to determine abundance and species of bats present

Results summary:

September 15, 2018: ~18 California leaf-nosed bats (*Macrotus californicus*) in XX1, 6 *Macrotus californicus* and 2 bats, probably cave myotis (*Myotis velifer*), in XX2.

December 1, 2018: No bats seen. Only partial survey of XX2.

Methods: We conducted internal surveys of the adits. We had descriptions of the features from a previous survey by BCI so we had a general idea of what to expect, although conditions can change greatly in 4 years. Safety gear included hard hats, white light, and a gas detector. Once a group of bats was encountered, we did not continue past the bats to prevent them from flying out of the mine, or otherwise disturbing them.

Result details:

XX1 September: We never entered the adit. Six California leaf-nosed bats (*Macrotus californicus*) hung just inside the portal; they were visible from outside the fence. 100% confidence for species identification (we used binoculars). About 6 m past portal is a collapse with a skylight above it. About 12 California leaf-nosed bats were visible from the surface just below the skylight. A few flew farther back but then immediately returned; the adit may only continue for a meter or so, but an internal survey when bats are not present is necessary to confirm this. December: No bats. Complete internal survey. Adit pinches out about 2 m past the vertical skylight; there is guano and a pack rat nest under the low ledge (~0.6-0.76 m high).

XX2 September: As we approached the portal, we saw 6 California leaf-nosed bats (MACA) just inside; they flew back into the mine so we continued the survey. Two bats, probably cave myotis (*Myotis velifer*, MYVE) based on visual characteristics, hung in a small dome (stained, so likely was repeatedly used by bats) about 15 m in. We did not continue farther. The MACA were flying around this area of the mine; we did not want to push them further or cause them to fly past us towards the portal. From the BCI survey notes, we went about 1/3 of the way. There was a large pile of guano several meters inside the adit and guano scattered throughout, along with lots of varied insect parts, including beetles. There was also an old javelina skeleton and a pack rat with a bowl nest and large pile of greens next to it. There was a muddy area near the portal, and the mine was very humid; slope creep obstructing part of the portal probably contributed to the humidity. December: A medium-large diamondback rattler stopped the survey about 10 m in from the portal. It was crawling towards the portal at a constricted point in the passage. Although we had a snake hook, there was no way to get the snake safely out of the mine because of the partially-collapsed portal that necessitates crawling in and out of. We shone our headlamps down into the adit past the snake and although we did not hear or see any bats, we cannot rule out the presence of bats towards the face of the mine. There were insect parts and guano near the portal.

XX3 September: The entrance is very small. Bees were flying all around the portal so we did not enter. December: No survey.

XX4 September: No survey- the adit and the one adjacent were both covered with steel cable net (~25 cm square, not diagonal). Net looks somewhat recent. December: No survey.

Condition of site: XX1: Fenced and signed. Large pit inside fence in front of adit; old water bottles in pit. XX2: Fenced and signed. First 15 m of adit shows evidence of instability- several large rocks had fallen along the adit. One piece of trash (food wrapper) outside the adit on the bench. XX3: Not fenced or signed. Dirty water bottles outside entrance. XX4: No signs of human activity other than installation of cable net that looks somewhat recent.

Discussion/Recommendations: BCI's description of XX1 is confusing and doesn't quite match our observations. It is unclear from BCI survey notes whether they completed an internal survey of XX3. If the adit opens up past the portal, it may be bat habitat. These sites, especially XX1 and XX2, warrant further monitoring for MACA in different seasons to determine how this species uses the site and when peak numbers occur, and if other species use the features. It seems to be a good site to include on Pima County's monitoring list for MACA.

The portal of XX2 should be monitored for continued collapse. It would be interesting to monitor temperature and humidity, and compare it to other sites occupied by MACA, as the partially-obstructed portal from slope creep likely contributes to microclimatic characteristics favored by this species at certain times of year. Maneuverable species such as MACA do not appear to have problems with small entrances; we have seen them use an entrance a smallish human male had to belly-crawl and squeeze through.

The cable net over XX4 and XX5 appears recent. The size of the net may not prevent use of the adits by maneuverable species that roost in relatively small numbers such as MACA, Townsend's big-eared bats, and Mexican long-tongued bats (*Choeronycteris mexicana*). Unfortunately, the only way to determine use of the site now is by monitoring evening emergences for day-roosting bats and all night long for night-roosting bats, which is inefficient and inconclusive if data are negative. However, monitoring is important to determine if this type of closure is a successful, bat-friendly method for keeping humans out. It may or may not be as "bat-friendly" as a steel gate with horizontal bars.

Mine Site #5 portal. Dave is by skylight.

Mine Site #5 California leaf-nosed bats.

Mine Site #7, Rancho Seco

Bat Surveys, 2018, by Bat Research and Consulting

Location/access: Sensitive information

Participants: Sandy Wolf, Dave Dalton

Objective: Internal survey to determine abundance and species of bats present

Results summary: September 15, 2018: no survey, bee hive at portal

Result details: We were unable to enter the mine because there was a substantial beehive just inside the portal with bees flying all around. They did not seem particularly aggressive, but we did not test this by trying to go past them and into the mine.

Condition of site: Barbed wire across entrance outside drip-line of portal. Grass along 2-track was mashed down due to recent vehicle traffic, but no other signs of recent human activity.

Discussion/Recommendations: This site is worth resurveying, perhaps in winter when bees are gone. The BCI survey (24 July 2014) reported the presence of guano from California leaf-nosed bats (MACA), Townsend's big-eared bats (COTO), and cave myotis, but no bats, and concluded it was used as a night roost. Based on only one survey, that conclusion seems premature. Based on its described length (29.6 m), and our observations that MACA and COTO roost near portals and in small groups at certain times of the year, this site could very well be a day roost.

Mine Site #6, Rancho Seco

Bat Surveys, 2018, by Bat Research and Consulting

Location/access: sensitive information

Participants: Sandy Wolf, Dave Dalton

Objective: Internal survey to determine abundance and species of bats present

Results summary: September 15, 2018: 6 bats, probably cave myotis. Took guano samples for DNA species identification; results confirmed guano was from cave myotis.

Methods: We conducted internal surveys of the adit. We had a description of the feature from a previous survey by BCI so we had a general idea of what to expect, although conditions can change greatly in 4 years. Safety gear included hard hats, white light, and a gas detector.

Result details: Heavy vegetation in deep trench. Some slope creep but portal is a walk-in. Small snake (black-necked gartersnake?) curled up under a piece of metal outside portal. Conducted complete internal survey (BCI survey data: 54 m long). Several small (~ 0.5 m diameter) discreet areas of guano along passage from near portal to face; all areas of guano looked like they were from same species. All but one looked old and were moldy. Took samples for DNA analysis from the one that looked the freshest. Above it were 5 bats with visual characteristics consistent with MYVE. Another bat was flying near the face. We left immediately. Later eDNA analysis of this guano sample confirmed that the sample was exclusively from cave myotis. We saw one sphinx moth wing, which is typical evidence of MACA. There was a 6 ft. step ladder at the stope and a grill from an air conditioner; apparently rock-hounds were collecting and sieving rocks from the stope. The rock at the top of the stope had green and yellow bands of minerals.

Condition of site: Portal used to have chain-link fencing across it, but it has been mostly torn down and pushed aside. Lots of trash near stope: plastic plates and lids, plastic fork, McDonald's Styrofoam cup; looks like people spent some time there. Looks like ATV tracks south of parking spot in road, looks challenging in a few places even for an ATV. Tank was dry and grassy with several cows.

Discussion/Recommendations: This site is worth rechecking in different seasons for California leaf-nosed bats and Townsend's big-eared bats. The site apparently receives rather frequent visitation from recreationalists; the PICO surveyors in December, 2017 met people that had been in the mine. Preventing vehicle/ATV access would be helpful in protecting bats that use this and possibly other mines in the area. Barriers across the portal other than a steel bat-friendly gate are either unlikely to be effective in keeping people out or in allowing bats necessary room for access.

Appendix B: Field Survey Protocols

PROTOCOLS FOR FIELD METHODS FOR INVENTORYING AND MONITORING BATS IN CAVES AND MINES

Sandy Wolf, Bat Research and Consulting

These protocols were written for use by surveyors periodically monitoring bats at cave or mine sites that have already been inventoried and where bats are known to be present or to have been present, and where species and type of use is known. They are not adequate for sites that have not been thoroughly inventoried for bats and other wildlife throughout the year, or where the complete internal structure (including connections to other features), hazards, and resources have not yet been described and mapped.

Regardless of the survey method, there should be sufficient personnel on each visit for safety. Monitoring surveyors should be provided with accurate location data; the datum for GPS coordinates is essential and EPE (estimated precision error) is helpful, particularly for features where entrances are close to each other or cannot be easily seen from a distance. Surveyors should also be given all information from inventory and previous monitoring work; this information will allow them to prepare for the survey and known hazards and conditions, and be better able to interpret changes they observe.

EXTERNAL SURVEY: VIDEO-RECORDED EMERGENCE COUNTS

External surveys, where observers video-record an evening bat emergence, should be used as the monitoring method of choice when bats are known to be present and the objective is to accurately determine abundance. It is the only method that should be used for maternity colonies, and at any time of year for vertical shafts and at sites with large colonies. Emergence counts are more accurate than estimates made during internal surveys where it may not be possible to observe all bats or make an accurate count without disturbing them. External surveys need expertise, particularly at large or complicated sites, but less expertise than internal surveys, or capture and handling of bats. They are also safer for surveyors and bats.

An emergence count produces an estimate of the number of bats in the roost. A census is possible if surveyors conduct an internal survey after the emergence is over to see if any bats remain in the roost. Video-recorded emergences yield important parameters, such as emergence length and peak number of bats out per minute, in addition to total number of net bats out. These can be compared over time and under various circumstances (e.g., installation of a gate). An emergence count is also the only method that can be used to determine the effects of gating on bat behavior.

If multiple species are present, a camera with high resolution and fast shutter speed can be used to identify individuals of easily distinguishable species (e.g., cave myotis and lesser long-nosed bats). Not every individual needs to be identified to estimate the relative proportion of each species. Emergence curves can sometimes be used to determine when the emergence of one species ends and another begins if species typically fly at different times, but this is not reliable without visually identifying as many bats as possible. Acoustic detection of other species that may be present can be conducted at the same time as the emergence count, although the relative proportion of each species cannot be determined.

Camera placement is critically important for good results. Bats on videos can be difficult or impossible to count if bats are flying towards or away from the viewer, or if there is a great amount of swirling

around. Sufficient infrared lighting that is well placed is also essential; bats will be missed in dark areas, and poorly placed lights can create shadows that result in double-counting.

The range of dates for an emergence count should be determined from inventory work. Roosts used during winter, migration, mating, or by transitory or summer bachelor colonies should be monitored at the time of peak abundance. Maternity colonies should be monitored ideally before young are born, but definitely before young are volant, so that the size of the adult population can be compared year to year. Timing is more difficult if births are asynchronous, as is the case with lesser long-nosed bats.

The end-point of an emergence should be standardized for all sites. Bats typically dribble out at the end of the emergence, and particularly at large maternity colonies (e.g., lesser long-nosed bats), they may continue going in and out of the entrance for hours. If the recording is stopped too soon, the abundance estimate will be low. Assuming there are no extenuating circumstances that could temporarily affect an emergence, a possible end-point criterion is: no net bats out for 10 minutes. For large (thousands) maternity colonies of lesser long-nosed bats or cave myotis, we may use an alternate method: when the number of net bats out is <10 for at least 10 minutes.

If capturing bats to determine sex, age, and reproductive condition is necessary, the emergence count should be conducted on a night prior to capture. Capturing bats at a roost will affect their exit behavior on that night, and possibly temporarily after that.

SURVEYOR QUALIFICATIONS

At least one of the surveyors should be familiar with the operation of the equipment and understand photographic principles (e.g., shutter speed, field of view, contrast ratio, depth of field), understand infrared lighting and the camera's sensitivity to same, know what a well-lit scene looks like and how to achieve it (including recognizing and minimizing reflective objects such as vegetation and rocks between the camera and bat flight path), and have experience with where the optimal camera placement is in relation to the physical structure of entrance and bat flight. All of these are necessary to obtain high quality video that yields the most accurate estimate.

All surveyors should be experienced in hiking and navigating in the dark in backcountry. They should be able to recognize and evaluate dangerous conditions on the surface near and above abandoned mines (Pierson et al. 1999).

Familiarity with the behavior of monitored species is helpful. For example, knowing which species are early or late flyers can help one recognize and thus record atypical behavior, and conversely, knowing what is typical behavior can prevent misinterpretation of observations.

EQUIPMENT/MATERIALS

1. Standard field and safety gear for the weather and environment
2. Copies of necessary permits
3. An equipment list: includes monitoring and personal gear to be taken in the field, decontamination materials and possible back-up equipment to be left in the vehicle
4. Standardized data form (and pens, pencils) that includes: date, site name, location, species, observers, sunset time, weather data, equipment used and set up, time recording begins, time first bat out, time recording ends, reason recording ends, if internal survey was conducted after emergence and results, comments (e.g., weather, bat behavior, site condition), room for a sketch of set-up

5. Infrared-sensitive camera for each entrance. Camcorder with internal hard drive and sufficient battery power, or surveillance camera (e.g., Axis P1354) with laptop in waterproof case and Ethernet cable. Although surveillance cameras require the use of a laptop to record the emergence, a long Ethernet cable (50-100 feet) allows the observer to sit away from the entrance and monitor the emergence on the laptop; this avoids the potential disturbance of an observer sitting with a camcorder close to the entrance. Minimum distance depends on the geography of the site, the flight path of the bats, and observer behavior (ability to sit still and be quiet).
6. Enough infrared lights for the size of each entrance, distance from the camera to the entrance, and sensitivity of the camera to infrared light. For example, for a 6 ft. by 6 ft. entrance, and a camera with the sensitivity of an Axis 1534 placed about 15-20 ft. away from the flight path of the bats, 2 WE IR6 lights (Bat Conservation and Management, www.batmanagment.com) are sufficient. Additional lights on a separate tripod may be necessary depending on the entrance; this requires additional skill for placement to avoid shadows that can be double-counted as separate bats.
7. Batteries and power cords for powering cameras and lights, and hardware for attaching lights to tripod. Lithium-ion batteries are lighter (more amp hours per pound) than gel-cell lead acid batteries, but care must be taken not to drop them. Batteries should have enough power to last longer than the expected length of the emergence to avoid having to change a battery and lose recording time
8. Rain protection for cameras (depending on camera model, this may be commercially available, or can be home-made). Bats will fly in the rain and if equipment is protected, monitoring can continue. Especially important if site is logistically difficult to monitor because of distance, terrain, amount of equipment needed to haul, and returning to site another night is costly. If exit behavior is altered by weather, however, another survey should be scheduled.
9. Pan/tilt unit (with a power source) mounted under the camera can be useful if the camera is mounted higher than the surveyor can reach in order to achieve the optimal angle and field of view, or must be remotely operated from recording/viewing laptop.
10. Method for stabilizing tripods for unexpected strong wind (e.g., hang battery or rock from tripod in a cloth bag)
11. As much as possible, a back-up of everything in case of failure
12. Thermometer
13. Watch
14. Headlamps with white light for hiking, red light for use during emergence
15. Camera to photograph equipment set up, site condition, etc.
16. If an entrance must be live-counted, night vision goggles and clicker counters
17. Small pruning shears for clipping grass or trimming vegetation, p-cord for tying back branches
18. Decontamination supplies for use at site after survey and at vehicle- soap, Clorox wipes, alcohol, garbage bags/containers, spare clothes and shoes

FIELD MONITORING PROTOCOL

Before monitoring visit

1. Develop safety plan and contacts for each field visit.
2. Conduct a test emergence count at the site. Determine how long it takes to get to the site to ensure there is adequate time for set-up, determine the best route to take when returning to vehicle in the dark, and check area around site for safety hazards.

A test count is necessary to figure out camera placement and angle (previous video recordings can be used for reference), especially if there are multiple entrances. For sites with multiple entrances, there should be a camera at each entrance.

During the test, move the camera if necessary throughout the emergence to compare views. For abundance estimates, the best camera view is to have bats fly across the screen. All bats exiting must be in the field of view, and large enough to distinguish individuals. Camera placement is best off to the side of the entrance so that it does not interfere with bat flight. For data on behavior at a gate (or at the site of the gate before construction) placement and angle of the camera should ensure that the total gate is in view, at as much of an angle to the gate as possible so that the individual space between bars (or sections of gate) that bats fly through can be distinguished. If this view makes obtaining accurate estimates of abundance more difficult, a different camera can be used outside the entrance with a better view for counting.

During the test video, experiment with the number, placement, and aiming of infrared lights until the optimally lit scene is obtained.

Trim, remove, or tie back vegetation if it obstructs view of the flight path. Vegetation may also reflect infrared light and cause the camera iris to close, darkening areas of the bat flight path and affecting the accuracy of the count.

3. If it is not possible to have a camera at every entrance to a site with multiple entrances, an observer with night vision equipment and supplemental infrared light should monitor the entrance where the fewest number of bats exit. Clicker counters, one for bats exiting and one for bats entering, are used to count bats. Live counts have many disadvantages compared to video-recorded counts, and should be avoided if at all possible.
4. View the test video and count a sample of bats to evaluate the field of view, angle, and lighting. Make notes for any adjustments needed to improve the video.

Before leaving for site

1. Do a full equipment set-up and operation test to ensure everything is working (e.g., batteries are charged), and all hardware and parts are present. Synchronize times on cameras. For surveillance cameras, check that exposure and frame rate are appropriate for the objectives of the visit (i.e. abundance count vs. species identification), and information that will appear on the video (e.g., site name, date, time) is correct.
2. Check items on equipment list as gear is packed and loaded into vehicle to ensure nothing is forgotten.
3. Avoid nights with rain or strong wind; bat exit behavior can be affected.
4. Plan to arrive at the site early enough to be set up and ready before sunset.

On site

1. Be quiet as possible, but do not whisper (higher proportion of ultrasonic sounds). Bats, especially Townsend's big-eared and California leaf-nosed maternity colonies, often roost close to the entrance in the twilight zone, and may be disturbed by noise outside the roost.
2. Set up equipment and check that everything is working, but then turn power off until ready to start recording.

3. Trim, remove, or tie back grass or vegetation in field of view between camera and entrance if necessary.
4. Be ready to begin recording no later than sunset and possibly before in cooler weather. Depending on species, season, and weather, bats may begin flying at or very shortly after sunset. Surveyors should keep an eye on the entrance as they are setting up to ensure atypical behavior is noted, even if it cannot be recorded. We observed several hundred lesser long-nosed bats flying before sunset in the rain in September.
5. Start recording at sunset, shortly before bats are expected to fly (based on past experience), or wait until you see first bat emerge. It is better to start recording early than to miss bats. If recording starts after bats start emerging, write down the number of bats missed. Camcorders do not always record time, so when you start to record, say the time into the camera's microphone as soon as you see the red REC light in the camera's eyepiece or LCD screen. Also say the date and site name. Shut the LCD screen (it uses more battery than eyepiece and produces more light).
6. Record weather data at sunset: temperature, % cloud cover, wind, precipitation. Record significant weather events during emergence that may affect bat behavior (e.g., strong wind, rain). Record weather conditions at end of emergence if significantly different from that at sunset. Note moon phase (% illumination) and moonrise/set time and if visible during emergence.
7. Infrared lights cannot be checked in daylight. Turn lights on when recording starts. Check when dark enough to ensure lights are aimed correctly to produce an evenly lit scene over the entire entrance and flight path. Watch and adjust camera angle, zoom, and lights are capturing the entire flight path of bats once the emergence starts. Depending on where the camera is placed in relation to the entrance, one person may choose to stay at the camera once the emergence starts to check that the set-up is correct and the equipment is operating correctly. If the person is very quiet, still, and does not use any visible light (red or white), this may be less disturbing to bats than to have someone approach the entrance to check equipment periodically.
8. Fill out data form.
9. During the emergence, observers should wait well away the entrance, not in front of entrance or in/near the flight path. They should be as quiet as possible, use red light and only when necessary.
10. Record until the predetermined criteria for the end of recording are met. If no bats exit and there does not seem to be a reason (bad weather, predator at entrance), wait at least 90 minutes after sunset before assuming no bats are present.
11. If possible, do an internal survey after the emergence to determine if, and how many, bats remain.
12. Clean and store gear, according to WNS protocol, and change clothes before entering vehicle.

After field visit

1. Check over equipment and gear and decontaminate according to WNS protocols.
2. Store batteries charged.
3. Watch video-recordings (VLC is a good, free viewer) as slow as is necessary to count accurately. Using a clicker counter, count bats exiting and entering each minute, record on spreadsheet (Figure 1); net number of bats out is estimate. The same person should count all videos for the same evening to standardize bias.
4. If a camcorder is used, download files to a computer and concatenate with a video editor (e.g., AVS Video Editor) to produce a single video for counting.

5. Store video files on a computer (or external hard drive) and back up on an external hard drive.

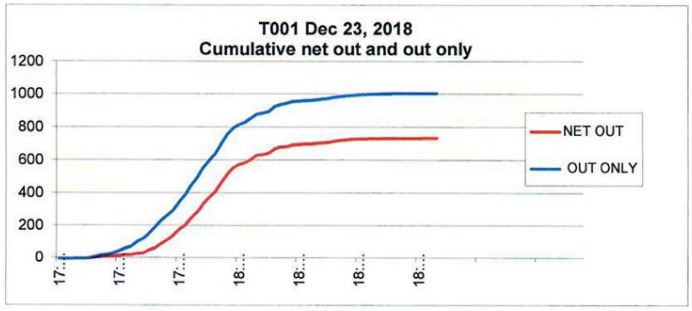
BAT SURVEY DATA SHEET

DATE:	23-Dec-18
NAME:	Dalton and Wolf
SITE:	T001 Tucson Bat Mine
SPECIES:	Macrotus californicus

738 net out total

TIME	#OUT	#IN	OUT-IN	NET OUT	CUMULATIVE OUT ONLY
17:39	1	0	0	1	1
17:40	1	0	1	2	2
17:41	0	0	0	2	2
17:42	1	1	0	2	3
17:43	1	1	0	2	4
17:44	2	0	2	4	6
17:45	7	6	1	5	13
17:46	11	6	5	10	24
17:47	3	1	2	12	27
17:48	9	8	1	13	36
17:49	13	10	3	16	49
17:50	17	13	4	20	66
17:51	13	13	0	20	79
17:52	28	19	9	29	107
17:53	20	18	2	31	127
17:54	30	9	21	52	157
17:55	40	28	12	64	197
17:56	41	13	28	92	238
17:57	29	8	21	113	267
17:58	35	6	29	142	302
17:59	50	11	39	181	352
18:00	44	21	23	204	396
18:01	57	11	46	250	453
18:02	49	12	37	287	502
18:03	58	9	49	336	560
18:04	46	6	40	376	606
18:05	40	4	36	412	646
18:06	58	5	53	465	704
18:07	58	6	52	517	762
18:08	41	1	40	557	803
18:09	21	2	19	576	824
18:10	15	3	12	588	839
18:11	23	3	20	608	862
18:12	27	1	26	634	889
18:13	6	3	3	637	895
18:14	11	1	10	647	906
18:15	28	2	26	673	934
18:16	13	1	12	685	947
18:17	7	5	2	687	954
18:18	11	0	11	698	965
18:19	2	1	1	699	967
18:20	3	0	3	702	970
18:21	2	1	1	703	972
18:22	4	0	4	707	976
18:23	4	1	3	710	980
18:24	3	0	3	713	983
18:25	8	1	7	720	991
18:26	4	0	4	724	995
18:27	4	0	4	728	999
18:28	4	0	4	732	1003
18:29	3	2	1	733	1006
18:30	2	2	0	733	1008
18:31	1	0	1	734	1009
18:32	1	0	1	735	1010
18:33	1	0	1	736	1011
18:34	0	0	0	736	1011
18:35	1	0	1	737	1012
18:36	0	0	0	737	1012
18:37	0	0	0	737	1012
18:38	0	0	0	737	1012
18:39	0	0	0	737	1012
18:40	1	0	1	738	1013 end record
18:41			0	738	1013
18:42			0	738	1013

Notes



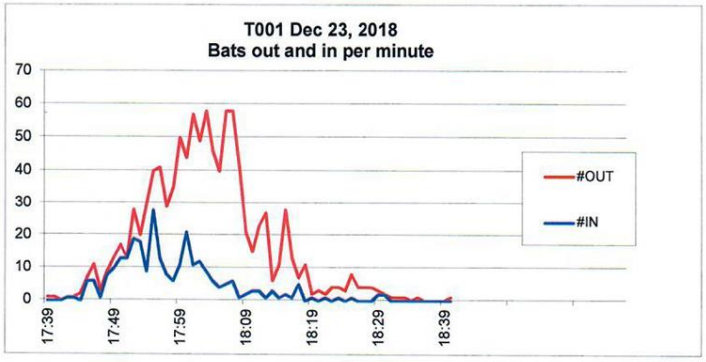


Figure 1. Sample spreadsheet for obtaining an emergence count estimate.

INTERNAL SURVEYS FOR ABANDONED MINES AND CAVES

As an inventory and monitoring method, an internal survey is the best way to determine past, present, and potential future use of a site, and the only way to determine cold season use (hibernating bats) (Sherwin et al. 2009). It is more cost-effective than an external survey because surveys at multiple sites can be conducted in one day, and if a complete survey is possible, results are more conclusive as to whether a site is used or has been used by bats. An internal survey is the best way to collect guano for DNA analysis; bats more often deposit guano under their roosting spots and along passages than at or outside the entrance to a roost.

During an internal survey, surveyors can collect data on environmental conditions (e.g., temperature, humidity, noticeable air flow, standing or dripping water) and will add to our knowledge of conditions associated with use by bats. For mines, data can be collected on the structural condition and stability of the portal and internal workings, noting changes such as slope creep or recent collapses that have occurred since the last survey. Mines will eventually reclaim themselves; data on changes in structural and environmental conditions over time may provide insight on changes in bat use.

At a site where an emergence count is conducted, an internal survey, if possible, provides data on bats remaining after the emergence is over. Without it, the emergence count only provides an estimate of abundance, not a census.

The Pima County Preserve System is comprised of low-mid elevation habitats. It is unlikely there are roost sites suitable for true hibernation for long periods (Townsend's big-eared bats prefer sites <10°C [Pierson et al. 1999]); therefore, an internal survey for the 4 target species at any time of year is likely to find active or temporarily torpid bats. Internal surveys as a monitoring method would be most suitable for fall, winter, or spring surveys where bat use is known to be ephemeral, and based on past inventory work, bat abundance is likely to be low (although migratory cave myotis colonies can number in the hundreds if present with the target species).

If a maternity colony is known to be present in a feature, surveyors should not conduct an internal survey; an emergence count should be used instead. Maternity colonies are easily disturbed; mothers may drop young, move to another area in the roost with less preferable microclimatic conditions, or abandon the roost entirely. If it is necessary to determine if young are present, surveyors can conduct an internal survey after an emergence count; however, great care must be taken not to disturb the young or the adults remaining to babysit them. Young that fall may not be able to be retrieved when the mother returns.

Internal surveys of mine shafts (vertical features) should not be conducted where bat use has been established. Rappelling down a shaft requires multiple people for safety and additional expertise; again, caving experience is not sufficient. An emergence count is safer, and potentially more accurate.

Monitoring caves with internal surveys should probably be avoided unless the objective is to put down guano sheets; emergence counts are more accurate for abundance estimates and safer for surveyors and the cave. Most caves are too complex to survey entirely; even large bat colonies can be missed if not in easily accessible and visible locations. In addition, caves are sensitive environments, often with delicate formations or surfaces easily damaged. If the situation warrants an internal survey, someone familiar with the cave should lead surveyors to ensure a complete survey, and in a manner that

minimizes damage to the cave and prevents anyone becoming lost. Surveyors should wear gloves and avoid touching speleothems; skin oils can damage growing formations.

Those involved in a project that includes internal surveys should ensure that if a rescue is necessary, search and rescue personnel that will be called first to the scene are trained and willing to go underground into caves or abandoned mines.

SURVEYOR QUALIFICATIONS

Surveyors who are not already knowledgeable and experienced in internal surveys as described generally below, and in more detail in Sherwin, et al. (2009), should not survey features unless they are being trained and accompanied by a professional surveyor. A trained professional has the expertise to be able to identify many species visually from a distance, and knows, for each species and type of colony, how best to approach bats, how to evaluate the behavior of the colony, how to balance affecting bats while obtaining necessary data, and when to end the survey if necessary to prevent disturbance.

Surveyors should be experienced and familiar with bats and bat sign for all bats that may be encountered on surveys in that geographic area (i.e. Pima County Preserve System). For example, some species are crevice dwellers and difficult to find even if roosting in groups; they may be in cracks, small domes, and shot holes. Some species roost openly on walls, ceiling, but may still be difficult to find if roosting individually, especially in hard to see areas such as high stopes and ore chutes. Often bats are visible only when approached from one direction and not another. Different species leave different evidence of use behind, for example, California leaf-nosed bats can leave fecal matter on the walls of a mine that do not look typical of bat guano. Nectar-feeding bats leave different guano and odor than insectivorous species. Bats often leave stains indicative of roosting areas; historical roosts of lesser long-nosed bats have a distinctive staining pattern. Surveyors should be able to find small amounts of guano, even a few scattered pellets if present, distinguish bat guano from rodent scat, and should be able to identify the species, or probable species, of the more distinguishable guano.

Surveyors should be trained in looking for, recognizing, and evaluating the numerous hazards that can be present in abandoned mines. Caving experience, no matter how extensive, does not prepare one for the potential dangers of abandoned mines. Surveyors decide when conditions warrant cessation of a survey or abort it before entering the feature. Abandoned mines are not safe, although some hazards can be minimized with training and experience. Proper attitude is important; surveyors must understand and respect the dangers, and recognize when conditions exceed their training or experience (Pierson et al., 1999, Sherwin et al., 2009).

EQUIPMENT/MATERIALS

1. Standard field and safety gear for the weather and environment
2. Copies of necessary permits
3. An equipment list: includes monitoring and personal gear to be taken in the field, decontamination materials and possible back-up equipment to be left in the vehicle
4. Standardized data form (and pens, pencils) that includes at minimum: date and time, site name, location, surveyors, weather, notes on condition of feature, particularly the portal, hazards noticed, water present, air flow, location and type of sign, species observed, how identified, and number, other wildlife present, whether survey was complete, notes on access, evidence of human visitation, photos if taken, room to sketch location of bats/guano
5. Safety gear: MSHA-approved hard hat, multi-gas detector (O2, CO, hydrocarbons, methane sensors), O2-generating self-rescuer, respirator with ammonia and particulate (e.g., dust with potential hantavirus, arenavirus, arsenic or lead-contaminated dust) filters used as necessary

6. Headlamp and 2 backups with powerful white light. Red light is less disturbing to bats, but is insufficient for moving safely through a mine or for seeing bats clearly from a distance. White light is overall less disturbing as it allows identification and estimate of abundance to be done faster and from a greater distance.
7. Probing pole if water is present, waders if necessary
8. An infrared-sensitive camcorder, and supplemental infrared lights and battery. If bats cannot be identified immediately, it is less disturbing to film them from a distance, using the zoom lens and only infrared light. Bats can be identified and counted on the video later.
9. Psychrometer
10. Guano sheet material if deploying, trash bag for collecting used ones
11. Collection vials and materials for collecting guano for DNA analysis, film canisters, ziplocs for collecting skeletal material
12. Still camera
13. Snake hook if experienced in use
14. Small pack to carry gear during survey that can easily be decontaminated
15. Disposable Tyvex suits are an option. However, suits are noisy and hot, and the footies do not have good traction. They should not be worn hiking in between mines.
16. For caves: gloves, and if needed, knee and elbow pads, other standard caving safety gear
17. Plastic bags for hiking backpack to store gear used inside mine and keep separate from other gear in pack, large plastic bags/containers to store potentially-contaminated gear from survey in vehicle
19. Cleaning supplies for on-site decontamination procedures after survey is complete: soap, Clorox wipes, alcohol, garbage bags/containers, spare clothes and shoes

FIELD MONITORING PROTOCOL

This is a general outline only and is not intended to describe everything that must be done to ensure a safe and effective survey.

Before monitoring visit

1. Surveyors should have access to, and be familiar with, all known data about the site, including internal structure, maps, hazards, and previous survey results
2. Develop a safety and communication plan. There should be at least 3 surveyors, 2 who go underground and one who stays on the surface. Sherwin et al. (2009) suggests one person focus only on human safety, the other focus on the biological survey. If a surface monitor is not available and the feature to be surveyed is simple, small, and stable, 2 surveyors can survey the mine with a safety check by phone. The safety check/surface monitor should have the exact location of the feature. Surveyors contact the safety check before entering, giving approximate time of return, and contact again after exiting. In an emergency, the protocol is to call 911 and ask specifically for a Pima County Sheriff Search and Rescue Deputy. That person will determine if a call out to Southern Arizona Rescue Association (SARA) is warranted.
3. Surveys should not be conducted during or after storms. Rain soaking into the ground makes portals and underground workings less stable and prone to collapse.
4. Ensure all safety and survey equipment is functioning correctly.

On site

1. Follow established safety protocol.

2. Assess structural conditions on the surface and at the portal, decide if an internal survey is safe. In Arizona, a common potential hazard is Africanized beehives near the entrances to some features, and this threat should also be assessed.
3. During survey, continuously assess safety conditions, and abort survey if deemed dangerous.
4. Proceed slowly enough to assess safety conditions and search for bats and sign: guano, staining, insect parts, bat carcasses/skeletons.
5. Prevent disturbance to bats by being quiet except for necessary communication.
6. If bats are encountered, identify quickly and estimate numbers. Leave immediately; do not take time staying in the area and shining lights on bats to count individuals. Do not stay in the area to take environmental data. The distance from surveyors to roosting bats before disturbance occurs varies by species and type of colony, physical characteristics of the roost, and surveyors' expertise.
7. Look for other wildlife species and their sign.
8. Collect data on environmental conditions.
9. Look for and make note of structural conditions.
10. Lay down or check guano sheets (probably not useful if surveys are over a year apart). Note amount and type of newly deposited guano. Collect guano for DNA analysis as per protocol.
11. Follow white-nose syndrome protocols after survey upon returning to the surface.

After field visit

1. Store guano samples for DNA analysis as per protocol.
2. Clean and decontaminate all clothing and gear according to the most current white-nose syndrome protocols (see section on white-nose syndrome decontamination).
3. Inspect all personal and safety gear for proper operation and condition.

ROOST LOGGERS

The Anabat Roost Logger (Titley Scientific, www.titley-scientific.com) acoustic detector is a passive detection method for long-term monitoring of bat activity in a roost. It cannot identify species or abundance, but it does document dates, times, and levels of echolocation activity as bats enter or leave the roost. It can be deployed just inside a cave or mine entrance, or at the entrance, which avoids the safety problems of internal surveys. The system is waterproof (but not submersible) and batteries last for weeks or months depending on how it is programmed. Proper placement for optimal results is important, and it is most effective at smaller entrances/passages where distance from the unit to flight path of bats is <10 m (closer is better).

This system is a valuable tool for inventory work or long-term monitoring (depending on the objective of the monitoring). Deploying one for a year at a site where bats or bat sign was observed will give information on which seasons bats use the site, when peak activity (and most likely abundance) occurs, and whether the site is used as a day or night (or both) roost. Although visits to the site are necessary periodically to change batteries, trained bat surveyors may not be necessary depending on the location of the logger, and visits can be done during the day to multiple sites.

Once data are collected, trained surveyors can time visits to determine species and type of use when data show surveys are most likely to produce the best information, thus reducing the number of trips for inventory work. If the roost is occupied by a single species, long-term deployment of a logger can provide valuable data on dates of arrival and departure, and relative abundance, whereas an emergence count once every 2-3 years provides an accurate estimate of abundance for that night only.

BAT CAPTURES

Capturing bats is not appropriate as a periodic, long-term monitoring method; before a site is selected for long-term monitoring, the species, type of use (e.g., maternity, transitory) and approximate size of the colony should already be known. For the 4 target species, it should be necessary only occasionally (e.g., at multi-species roost) to identify species during inventory work; species can usually be determined visually during an internal survey or from a video-recording. Captures are necessary if bats' age, sex, and reproductive condition are needed to determine the type of use in a roost (e.g., maternity vs. bachelor). Bats should only be handled by experienced people with pre-exposure rabies vaccine and up-to-date titer checks, and who hold a current permit from Arizona Game and Fish Department.

Bats should not be captured during late pregnancy or early lactation; capture sessions should be timed to avoid this period (in eastern Pima County, mid-May through early-August for MACA and CHME, May through mid-July for COTO, mid-April through early July for LEYE). The stress of being captured and held on pregnant bats and those with newborns can negatively affect survival of young. The ideal time to confirm maternity use at a site is after young have begun to fly and before bats have started to leave, or others move in from another roost. If a maternity colony roosts in the twilight zone, human activity at the entrance is too disturbing. In this case, bats should be captured away from the entrance.

EQUIPMENT

1. Gloves- thick enough to avoid bites, flexible enough to feel and hold bat safely. May need different gloves depending on species caught.
2. Cotton cloth bags for individual bats, preferably numbered, with string cord-locked closure
3. Mist net and poles, guy lines and tent stakes
4. Harp trap, shade cloth or tarps
5. Scissors for cutting net if necessary to free tangled bat (rare event if nets are adequately tended and bats removed immediately by skilled handlers)
6. Head lamps with red and white light
7. Tub with sticks to hang bat bags on, protected from cold and wind and predators, to hold temporarily until processing
8. Processing equipment in large clipboard- calipers, Pesola scale, data sheets, pen, pencil, bat species key, straw for blowing hair to see nipples on females
9. Plastic trash bags to store used bat bags, used nets, to transport for cleaning and decontamination.
10. Rollup or portable table for processing is helpful if a large sample of bats is anticipated.
11. Supplies for initial cleaning and WNS decontamination in field

PROTOCOL

A harp trap is better than a mist-net for mine or small cave entrances, especially when a large colony is present (Sikes et al. 2011). Spaces around the harp trap can be closed with tarps or shade cloth to keep bats from flying around the trap. A few will probably still fly through the trap, but it is an effective method of capture, and much less stress on the bats. If too many bats are caught, the trap should be moved away from the entrance. Bats can hurt each other if they are too crowded in the harp trap's bag.

If the entrance is too large for a harp trap, bats can be captured in mist-nets set near, but not directly in front of, a roost. Bats must be removed by skilled handlers; it is easy to break a bat's delicate wing

bones. If more bats are caught simultaneously than can be removed immediately, bats get tangled, are more difficult to remove, suffer more stress, and are more likely to be injured. Therefore, the mist-net must be placed in a position far enough away to catch only one or 2 bats at a time, but near enough and in a flight path to catch a sufficient sample for the desired objective. This requires skill and experience. Multiple handlers should be present to remove bats from a net, and the net should be closed when a sufficient number of bats are caught.

1. Do not open nets until just before bats fly to avoid capturing birds.
2. Monitor the mist-net or harp trap continuously, remove bats immediately.
3. Remove bats from the trap's bag or mist net and place in individual cloth bags until they are processed. Hang bags somewhere (e.g., large tub with wooden sticks across top) out of wind, cold, and safe from predators. Ensure bats are hung in the order they are caught.
4. If there are too many bats in the harp trap bag to quickly remove and bag individually, they can be transferred to a holding cage (separate species in different cages) to avoid suffocation/fighting, and another handler can transfer them to individual bags.
5. Close nets or move harp trap as soon as a sufficient sample of bats is obtained.
6. Process bats as soon as possible; other handlers should continue to monitor net/trap. Process bats in the order they were caught. Process bats near the site of capture but not in view of the entrance to avoid lights shining into the entrance or bats being affected by noise. Minimize talking.
7. Release each bat as soon as data is collected. Bats should be held for as short a time as possible, a maximum of 30 minutes (less in cool weather); pregnant or lactating bats should be held only a few minutes.
8. Release by holding bat in an open palm with arm upraised away from processing area and in the dark. Handler should wait until bat flies off on its own; under no circumstances should a bat be tossed into the air to release it.
9. Limit photographs to one or 2 animals and minimize the time a bat is photographed; flash in a bat's eyes temporarily impairs its vision.
10. Clean and decontaminate equipment according to WNS protocol, and store separately from gear not potentially contaminated in vehicle.
11. Do off-site cleaning and decontamination according to WNS protocol.

DNA ANALYSIS OF GUANO

Identifying bat species from their guano is a relatively recent method of determining bat use in a cave or mine. It is an ideal method because bats do not need to be present for surveyors to collect data and there is no disturbance. To collect guano for analysis, material is laid down either over old guano, underneath a known bat roosting area, or near the entrance to the cave or mine. The guano "sheet" can be bridal veil or plastic sheeting. Corners should be weighted down with rocks. Bridal veil is preferable in caves because plastic sheeting can allow moisture to collect under the sheet and affect microclimatic conditions, and in turn, microbiota and invertebrates. In a mine, for which there is generally less interest in the preservation of delicate ecosystems, plastic sheeting can be used, and is less expensive. In addition, plastic may be less attractive to animals. It is also possible to use material already in the mine, such as a board or rock, if old guano has been brushed off. After a specified length of time, surveyors return and collect samples. In addition to identification of the bat species, the time period when bats were present will be known.

Guano samples should be collected and stored according to the protocols provided by the laboratory conducting the analysis. Fresh samples are best. Care must be taken to avoid contamination with other DNA, including the collector's, during collection. Nitrile gloves, sterile tweezers, Ziploc bags, a marker, and alcohol are needed in addition to the collection vials provided by the lab. After collection, samples should be stored on ice in a cooler while in the field, then stored as directed until sent to the lab.

WHITE-NOSE SYNDROME DECONTAMINATION PROCEDURES

White-nose syndrome is a disease affecting hibernating bats that was first observed in a cave in New York in 2006. It is caused by the fungus *Pseudogymnoascus destructans* that attacks the skin, causing physiological changes that make bats come out of hibernation too often, use up energy reserves, and die. It has affected 11 species, resulting in 90-100% mortality of some colonies, and killing millions of bats as it continues to spread throughout the United States and Canada. The fungus spreads primarily through bat to bat contact, but bats can also be infected by contacting fungus on the floors and walls of caves and mines. It is possible for humans to transfer spores on clothing, shoes, or gear from one site to another. (www.whitenosesyndrome.org)

To prevent the inadvertent spread of the fungus by humans, guidelines and decontamination protocols (https://www.whitenosesyndrome.org) have been developed for people who come into contact with bats or their underground environments. The protocol is updated periodically; surveyors should use the most recent version. Surveyors should clean and decontaminate anything that has entered a roost, has been used outside a roost (i.e. emergence count equipment), or has been used to capture or process bats. This includes surveyor clothing and shoes.

Decontamination may not be necessary between each of multiple sites surveyed during the same day if sites are within a small geographic area where bats are likely to move among those roosts. The managing agency has the discretion to determine this amendment to the protocol and the size of the geographic area.

GENERAL GUIDELINES (not a substitute for following the most recent official protocol as posted on www.whitenosesyndrome.org)

1. Prepare for field work by choosing equipment, gear, and clothing that is easiest to decontaminate if choices are possible. Ideally, have certain gear dedicated for use only at a specific site.
2. When a survey is completed, clean as much as possible before packing for transport to vehicle, separate and package gear to keep uncontaminated gear clean during transport.
3. Before entering the vehicle, wash exposed skin, change and bag clothes and shoes. Package potentially-contaminated items to prevent spread to the vehicle.
4. Off-site, clean and thorough decontaminate all clothing and equipment according to protocol.

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Appendix C: Field Data Forms

Sample Emergence Count Spreadsheet

BAT SURVEY DATA SHEET

DATE:	6/9/18	**5,231 total**
NAME:	Dalton and Wolf	**peak 265**
SITE:	XX Mine west shaft	
SPECIES:	Myotis velifer	

TIME	#OUT	#IN	OUT-IN	NET OUT	CUMULATIVE OUT ONLY	Notes
19:35	0	0	0	0	0	
19:36	0	0	0	0	0	
19:37	0	0	0	0	0	
19:38	1	1	0	0	1	
19:39	0	0	0	0	1	
19:40	0	0	0	0	1	
19:41	1	1	0	0	2	
19:42	1	1	0	0	3	
19:43	2	0	2	2	5	
19:44	2	0	2	4	7	
19:45	1	0	1	5	8	
19:46	3	0	3	8	11	
19:47	13	0	13	21	24	
19:48	17	2	15	36	41	
19:49	22	0	22	58	63	
19:50	35	2	33	91	98	
19:51	32	0	32	123	130	
19:52	61	0	61	184	191	
19:53	63	0	63	247	254	
19:54	79	0	79	326	333	
19:55	97	0	97	423	430	
19:56	111	0	111	534	541	
19:57	169	0	169	703	710	
19:58	134	0	134	837	844	
19:59	107	0	107	944	951	
20:00	163	0	163	1107	1114	
20:01	198	0	198	1305	1312	
20:02	232	0	232	1537	1544	
20:03	**265**	0	265	1802	1809	
20:04	259	0	259	2061	2068	
20:05	247	0	247	2308	2315	
20:06	255	0	255	2563	2570	
20:07	247	0	247	2810	2817	

20:08	247	0	247	3057	3064	
20:09	264	0	264	3321	3328	
20:10	234	0	234	3555	3562	
20:11	230	0	230	3785	3792	
20:12	213	0	213	3998	4005	
20:13	162	0	162	4160	4167	
20:14	110	14	96	4256	4277	
20:15	124	5	119	4375	4401	
20:16	139	0	139	4514	4540	
20:17	100	0	100	4614	4640	
20:18	81	0	81	4695	4721	
20:19	80	0	80	4775	4801	
20:20	51	0	51	4826	4852	
20:21	59	0	59	4885	4911	owl out
20:22	60	0	60	4945	4971	
20:23	52	0	52	4997	5023	
20:24	56	0	56	5053	5079	
20:25	41	0	41	5094	5120	
20:26	24	0	24	5118	5144	
20:27	26	0	26	5144	5170	
20:28	17	0	17	5161	5187	
20:29	17	0	17	5178	5204	
20:30	20	0	20	5198	5224	
20:31	9	0	9	5207	5233	
20:32	9	0	9	5216	5242	
20:33	3	0	3	5219	5245	
20:34	8	0	8	5227	5253	
20:35	1	0	1	5228	5254	
20:36	1	0	1	5229	5255	
20:37	1	0	1	5230	5256	
20:38	1	0	1	5231	5257	
20:39	0	0	0	5231	5257	
20:40	0	0	0	**5231**	5257	stop record

Appendix D: Bat species observations on Pima County properties

Table 1. Bat species observation type by County property using available data from AZGFD Scientific Collecting License reporting, Pima County staff monitoring and netting data (i.e., at Agua Caliente Park), BLM-funded Bat Conservation International surveys of abandoned mines, and other available historical data. **Roost** = bat species has a roost site on the property; **Visual** = non-capture observation of a bat species on the property that does not indicate a roost; **Capture** = bat species captured while active, such as by mist net; **Acoustic** = bat species detected acoustically, usually while active and often away from a roost; **Salvage** = specimen salvaged dead and reported to AZGFD; **Historic** = prior records of a species using a site as a roost.

Pima County Property	Bat Species	Observation Type
MSCP covered bat species		
California leaf-nosed bat *Macrotus californicus*	Tucson Mountain Park Rancho Seco Mission Garden Agua Caliente Park Colossal Cave Mountain Park	Roost, Capture Roost Visual, Acoustic Capture Historic
Lesser long-nosed bat *Leptonycteris yerbabuenae*	Agua Caliente Park Cienega Creek Natural Preserve Colossal Cave Mountain Park	Capture Capture Capture, Historic
Mexican long-tongued bat *Choernonycteris mexicana*	A7 Ranch Agua Caliente Park Buehman Canyon Cienega Creek Natural Preserve Colossal Cave Mountain park	Roost Capture Roost Roost Roost
Pale Townsend's big-eared bat *Corynorhinus townsendii pallescens*	Agua Caliente Park Buehman Canyon Cienega Creek Natural Preserve Colossal Cave Mountain Park Diamond Bell Ranch Marley Ranch Old Hayhook Ranch Oracle Ridge Rancho Seco Tucson Mountain Park	Capture Roost Capture, Roost, Acoustic Capture, Roost, Acoustic Roost Roost Roost Roost Roost, Acoustic, Capture Roost, Acoustic
Western red bat *Lasiurus blossevillii*	Cienega Creek Natural Preserve	Capture
Western yellow bat *Lasiurus xanthinus*	Agua Caliente Park Cienega Creek Natural Preserve	Roost, Capture, Acoustic Capture
Non-covered bat species		
Big brown bat *Eptesicus fuscus*	Agua caliente park Arizona Sonora Desert Museum Cienega Creek Natural Preserve Colossal Cave Mountain Park	Capture, Acoustic Capture Capture, Acoustic Capture, Acoustic

Pima County Property	Bat Species	Observation Type
	Diamond Bell Ranch	Roost
	M Diamond Ranch	Acoustic
	Marley Ranch	Roost
	Oracle Ridge	Roost, Acoustic
	Rancho Seco	Roost
	Mission Garden	Acoustic
	Robles Barn	Roost
Big free-tailed bat *Nyctinomops macrotus*	Agua Caliente Park	Acoustic
California myotis *Myotis californicus*	Agua Caliente Park	Capture, Acoustic
	Cienega Creek Natural Preserve	Capture, Acoustic
	Oracle Ridge	Acoustic
Canyon bat *Parastrellus hesperus*	Agua Caliente Park	Acoustic
	A7 Ranch	Acoustic
	Arizona Sonora Desert Museum	Salvage
	Cienega Creek Natural Preserve	Capture
	Colossal Cave Mountain Park	Acoustic
	Oracle Ridge	Acoustic
	Rancho Seco	Acoustic
	Tucson Mountain Park	Roost, Acoustic
	Mission Garden	Acoustic
Cave myotis *Myotis velifer*	A7 Ranch	Acoustic
	Agua Caliente Park	Capture
	Arizona Sonora Desert Museum	Visual
	Bar V Ranch	Acoustic
	Buehman Canyon	Roost, Acoustic
	Cienega Creek Natural Preserve	Capture, Roost
	Colossal Cave Mountain Park	Acoustic, Capture, Roost
	Diamond Bell Ranch	Roost
	Marley Ranch	Roost, Acoustic
	Oracle Ridge	Acoustic
	Rancho Seco	Roost, Capture, Acoustic
	Tucson Mountain Park	Roost, Capture
	Robles Ranch	Roost, Capture
	Mission Garden	Acoustic
Fringed Myotis *Myotis thysanodes*	Oracle Ridge	Acoustic
	Rancho Seco	Acoustic, Capture, Roost
Hoary bat *Lasiurus cinereus*	Cienega Creek Natural Preserve	Capture
	Marley Ranch	Acoustic
	Agua Caliente Park	acoustic
Mexican free-tailed bat *Tadarida brasiliensis*	Agua Caliente Park	Capture
	Cienega Creek Natural Preserve	Capture
	Colossal Cave Mountain Park	Acoustic
	Marley Ranch	Acoustic
	Oracle Ridge	Acoustic
	Rancho Seco	Acoustic

Pima County Property	Bat Species	Observation Type
	Robles Ranch Mission Garden	Roost Acoustic
Pallid Bat *Antrozous pallidus*	Agua Caliente Park Canoa Ranch Cienega Creek Natural Preserve Colossal Cave Mountain Park Oracle Ridge Rancho Seco Robles Ranch Tucson Mountain Park	Capture Roost Capture, Roost Roost Roost, Acoustic Acoustic Roost Roost, Acoustic
Pocketed free-tailed bat *Nyctinomops femorosaccus*	Agua Caliente Park Cienega Creek Natural Preserve Oracle Ridge	Capture Capture Acoustic
Southwestern myotis *Myotis auriculus*	Agua Caliente Park	Capture
Western small-footed myotis *Myotis ciliolabrum*	Agua Caliente Park	Capture, Acoustic
Yuma myotis *Myotis yumanensis*	Agua Caliente Park	Capture, Acoustic
Western mastiff-bat *Eumops perotis*	Agua Caliente Park	Acoustic
Silver-haired bat *Lasionycteris noctivagans*	Agua Caliente Park	Acoustic
Arizona myotis *Myotis occultus*	Agua Caliente Park	Acoustic