

**U.C. Davis Wildlife Health Center California Mountain Lion Projects
2022 Annual Report
January 31, 2023**

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Introduction / overview

History of the UC Davis Wildlife Health Center Mountain Lion Study and current focus

This mountain lion (*Puma concolor*; puma, cougar) study is headed by Dr. Winston Vickers of the Wildlife Health Center at UC Davis (WHC). David Garcelon of the Institute for Wildlife Studies (IWS) is the PI for the northeastern California part of the study, and Dr. Jessica Sanchez of the San Diego Zoo Wildlife Alliance was a key contributor to several study elements in Southern California. From mid-April till December 1, 2022, Dr. Justin Dellinger co-Directed activities in all of the study areas with Dr. Vickers. Dr. Fernando Najera is expected to join the study as co-Director in March 2023. Dr. Vickers prepared this report with contributions from the other project collaborators.

Overall activities and research into mountain lions by the UC Davis WHC have been ongoing since the first capture of a mountain lion in the study in March of 2001. Over the course of this long term study, cougars were captured, sampled, and GPS collared in Orange, Riverside, San Diego, Modoc, Lassen Counties, and Kern Counties, and future capture activities are expected in the Gabilan Range and Pacheco Pass either in or in close proximity to Santa Clara, San Benito, Santa Cruz, and Monterey Counties (Figure 2).

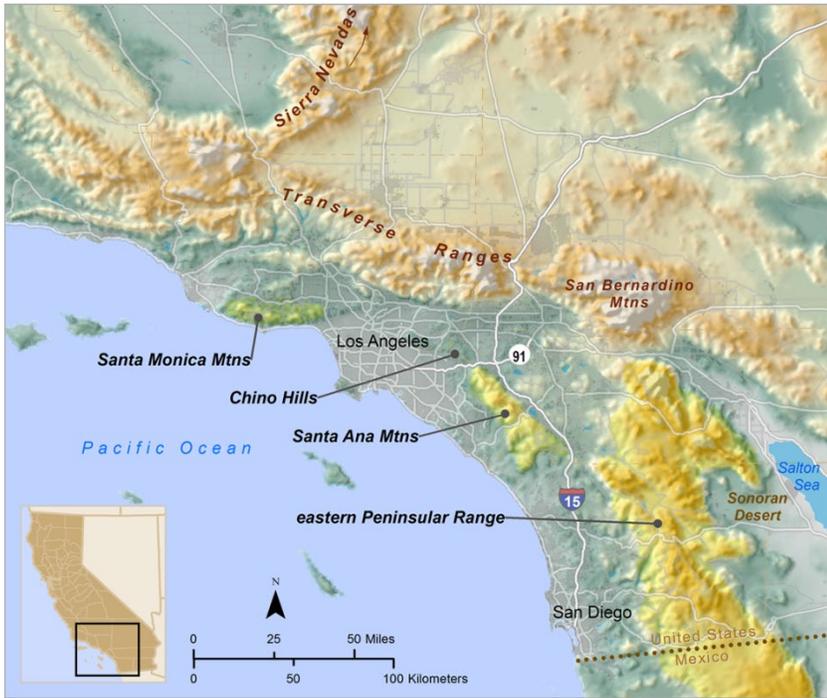
Figure 2.
Geographic focus areas in 2022:



Tissue samples have been obtained from deceased animals in San Bernardino County but no captures have occurred there. Activities of both types are anticipated in additional counties in the future.

The study in southern California (Figure 3) has incorporated mountainous areas including the Santa Ana Mountains (SAM) portion of the Peninsular Ranges (west of I-15) and the eastern Peninsular Mountain Ranges (ePR) east of I-15 (Figure 3), and has been conducted under authorization by the California Department of Fish and Wildlife (CDFW) via a number of Memoranda of Understanding and Scientific Collecting Permits.

Figure 3. Southern California mountain ranges where WHC study has been conducted since 2001.



In 2016, an amendment to our SCP was approved to allow expansion of the study scope and region to study of cougar ecology and interactions with pronghorn antelope (*Antilocapra americana*) in the northeastern corner of California (Modoc, Lassen, and Siskiyou Counties) (Figure 4). This second part of our study is being conducted in collaboration with, and with the financial support of, Dave Garcelon and the Institute for Wildlife Studies (IWS). The first capture in that study area was in February of 2016.

Figure 4. Northeastern California study area.



A new 3 year SCP was approved in early 2022 that allowed further expansion of the study areas to the Tehachapi Mountain Range north of Los Angeles (Figures 5 and 6) and the Gabilan Range (Figure 7) east of the Santa Cruz Mountains and the Pacheco Pass area (Figure 8). The focus in these study areas is on connectivity across barriers, specifically SR58 and I-5 in the Techachapi Range, and US 101 and SR152 as well as other highways in the Gabilan Range and Pacheco Pass areas. The first capture in those study areas was in late November of 2022.

Figure 5. Tehachapi Mountain Range between the Sequoia and Los Padres National Forests, with land status designated

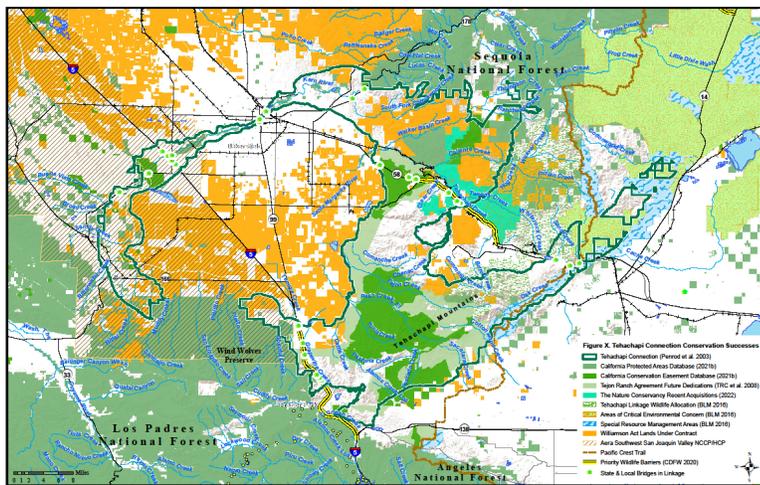


Figure 8. The Pacheco Pass area with recorded mountain lion vehicle kills and photos noted. Conserved lands in blue.



Broad study goals are to collect data and conduct analyses to inform the understanding of mountain lions in California in a number of areas:

- 1) Health, infectious disease and toxin exposure patterns and diet, along with relationships to anthropogenic and landscape factors, and sources of mortality;
- 2) Landscape connectivity as measured by movement data, especially across highways; genetic; camera monitoring of crossing structures; and studying dispersal patterns in more developed versus less developed landscapes.
- 3) Habitat use and movement patterns in response to anthropogenic influences, fire and fire management, and during various reproductive states and dispersal
- 4) Assessment of population genetic status and levels of inbreeding depression
- 5) Development and comparison of non-invasive sampling and population estimation methods, with the special goal of developing best practices and concrete plans for closely monitoring the most at-risk populations such as the SAM population. These methods include opportunistic and systematic scat sampling, hair sampling, camera-based methods, GPS collaring, and others.
- 6) Methods of reducing conflicts between mountain lions, domestic animals and people, especially in wild areas utilized by people, and rural areas where livestock are often kept in mountain lion habitat. This involves developing and testing deterrent devices and assessing mountain lion interaction patterns with humans and domestic animals in peri-urban or rural areas, and wildland parks.
- 7) Interactions with other wildlife species, especially species of special concern to CDFW such as pronghorn antelope in NE California, and possibly bighorn sheep in southern California.
- 8) The impacts of sound and light, or other anthropogenic influences, on mountain lion movement across highways and habitat use in and near other areas that contain levels of human influence.

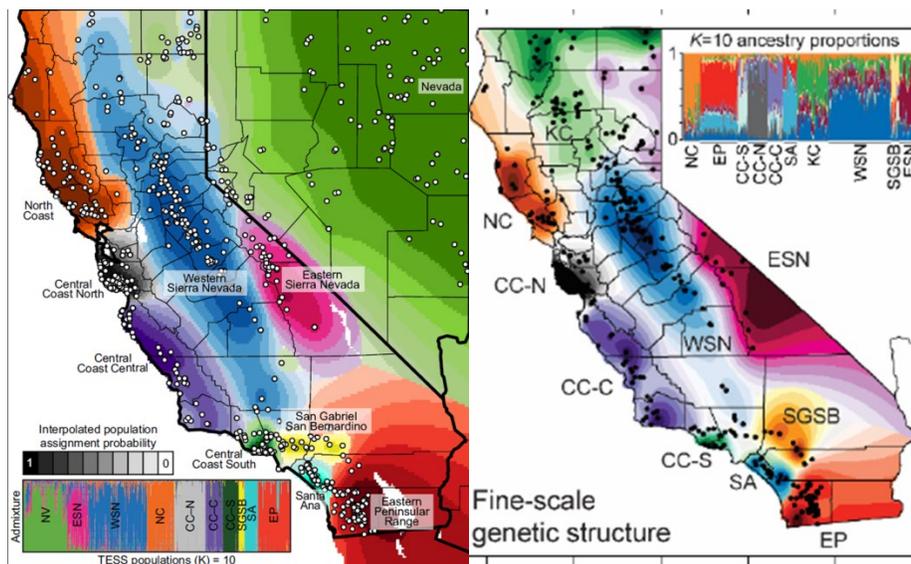
9) The differences between major habitat areas in all of the areas of focus listed above by collaborating on comparisons between our own study areas as well as the area in Sonoma-Napa Counties where study is overseen by collaborator Dr. Quinton Martins.

As we are now, we will continue to collaborate with other mountain lion researchers and CDFW throughout the state. The results will feed into policy and public outreach programs to promote habitat conservation, persistence of mountain lion populations long term, and improved wildlife and human well-being.

Status of mountain lions in California and relationship to our study areas:

Mountain lions in California have been characterized in multiple studies as being fragmented into multiple subpopulations, with barrier effects of highways and development likely to be the primary contributors (Gustafson et al. 2018, 2022; Figure 9).

Figure 9. Mountain lion subpopulations in California based on microsatellite (left figure) and Single Nucleotide Polymorphism (right figure) based analyses (Gustafson et al. 2018,2022).



From Gustafson et al. 2022:

“Fragmented populations along the Southern Coast and Central Coast had particularly low genetic diversity and strong linkage disequilibrium, indicating genetic drift and close inbreeding.” “Thus, extant variation at the broader scale has potential to restore diversity to local populations if management actions can enhance vital gene flow and recombine locally sequestered genetic diversity.”

The two subpopulations in the Santa Ana Mountains (SAM) and Santa Monica Mountains (SMM) are the most seriously genetically restricted subpopulations, with mountain lions in both mountain ranges being second only to the federally endangered Florida panther in their level of inbreeding, and are at risk of extirpation (Benson et al. 2019). A Population Viability Analysis for the SAM population found that there is a 11–21% risk of extirpation in the next 50 years due to low annual survival, and demographic, stochastic, and environmental factors, and a near certain likelihood of extirpation within a median time of 12 years if inbreeding depression should occur. (Benson et al. 2019; Figure 10).

Figure 10. From Benson et al. 2019. Risk of extirpation of the SMM and SAM populations of mountain lions if inbreeding depression occurs in the populations.

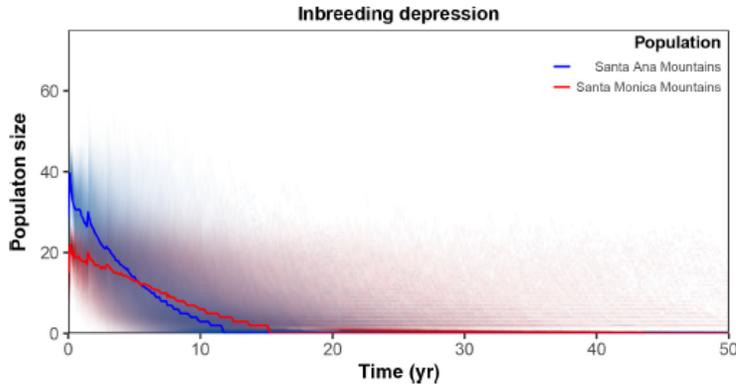


FIG. 4. Density-dependent demographic projections from individual-based population model showing predicted population sizes for mountain lions in the Santa Ana and Santa Monica Mountains over 50 yr based on 5,000 projections when we simulated inbreeding depression with the observed level of immigration.

The risk to the SAM and SMM subpopulations, as well as the ePR and San Bernardino and San Gabriel (SB/SG) populations over time, has led to a petition being filed to list all four of these subpopulations, plus the Central Coast and Santa Cruz Mountains populations, as threatened under the California Endangered Species Act (CESA).

Long-term viability of mountain lion populations in southern California and in the central coast of California is at-risk due to habitat fragmentation that limits connectivity and gene flow and contributes to deleterious effects of inbreeding (Benson et al. 2019). Though current work to restore connectivity in parts of this geographic area are underway (e.g., in the Santa Monica and Santa Ana Mountains), it is recognized that the success of these current efforts is still dependent on future efforts seeking to further restore connectivity throughout larger geographic area of southern California and the central coast of California (Gustafson et al. 2022).

Notably, the Santa Ana Mountains and the Peninsular Range in southern California must be connected to the Sierra-Nevada Mountains for these populations to remain viable (Dellinger et al. 2020a, Gustafson et al. 2022). This connectivity would be achieved through restoring connectivity that links the Peninsular Range to the Transverse Range and then the Tehachapi Mountains (Figure 11). In addition, the Santa Cruz Mountains, Diablo Range, Gabilan Range, and greater central coast area must be connected to one another, and to the Sierra-Nevada Mountains, for mountain lions to persist throughout the central coast (Dellinger et al. 2020a). There are several major transportation routes that would likely need improved wildlife crossing structures to realize such connectivity. Notably, I-10, I-15, US 101, SR-14, I-5, and SR-58 all need to be assessed with respect to current impacts on connectivity, and recommendations developed for improvement of passageways that allow safe wildlife movement.

Figure 11. Southern California Mountains with priority linkage areas. From South Coast Wildlands and The Nature Conservancy.



The Tehachapi Range is a key linkage between the Sierra Nevadas population of mountain lions and coastal and southern California populations. Highways SR58 and I-5 appear to be likely barriers to mountain lion and other wildlife movement through the Techachapis. Thus those highways and their potential barrier effects have become a new focus of our study.

As with the other mountain lion populations, those in the Gabilan Range, Santa Cruz Mountains, Diablo Range, and greater central coast area require connectivity in order for the populations in those ranges to be sustained over the long term. Multiple highways are barriers or potential barriers in that region (US 101, SR152, and others).

Planning for high speed rail construction suggests that barrier effects will be exerted by the rail infrastructure as well as noise associated with train operation. These concerns apply to both the Tehachapi and Gabilan/Pacheco Pass study areas, and helped prompt this current study in order to establish pre-existing connectivity patterns in those area.

Addressing such connectivity issues requires a step-wise approach in implementing and utilizing multi-faceted data collection efforts, and also requires several years of effort to produce meaningful sample sizes and results. Via our suite of projects, we seek to ensure that viable wildlife populations persist throughout southern California and the central coast of California by: 1) understanding current wildlife connectivity and gene flow throughout these areas, and 2) informing efforts to restore connectivity throughout these areas by informing efforts to improve existing crossing structures and/or construct new wildlife crossing structures, as well as conserving key habitat areas, especially in wildlife movement corridors and adjacent to highway crossing sites. These measures require investment of public and private funds, and these investments can only be targeted for the most effect by utilizing accurate data from appropriate wildlife studies. Providing this data is a role that our organization has played for 20+ years in many areas.

Methods of data acquisition and use

In 2022, cougar use and movement areas were detected via scouting / tracking and placement of trail cameras in Modoc and Lassen Counties (joint UCD-IWS project), in the Santa Ana Mountains especially around Whiting Ranch Wilderness Park in Orange County, in the Tehachapi Range in the vicinity of SR58 and I-5, and in the Gabilan Range and Pacheco Pass west and east of the town of Gilroy respectively. These camera data were used to define areas that were most promising for potential cougar capture activities, and to better understand puma patterns of use in the Santa Ana Mountains for use in population estimation efforts and human interactions studies as described below.

As per our SCP, road-killed deer were collected and stored frozen for later use at bait sites. When regular cougar use of an area was detected, road-killed deer were placed at bait sites and checked daily. If cougar feeding was detected at a bait site, cage traps were set and monitored continuously via radio-transmitters affixed to the trap doors. Cameras that could transmit images from the site via cell systems were also used at trap sites if possible. Cage traps were used for the majority of captures in 2022, however hounds were used for two. Traps were checked as quickly as possible (within half an hour in almost all cases) after detection of signals indicating a cage door had closed and / or receipt of cell camera pictures indicating a cougar had entered a trap. This assured that no animal was left within a trap unattended for lengthy periods of time, minimizing the potential for self-inflicted injury within the trap.

Anesthesia drugs and dosages listed in our SCP were administered by jabstick syringe, hand syringe, or dart rifle (in the case of hound captures). Capture methods were approved by the UC Davis Institutional Animal Use and Care Committee (IACUC Permit #22408).

Wildlife veterinarians Dr. Winston Vickers or Dr. Jessica Sanchez, or o-PI Dave Garcelon, administered or directly oversaw administration of all anesthesia, conducted and oversaw anesthetic monitoring, conducted all physical exams and sampling, and fitted all GPS-collars. Various students and wildlife biologists had opportunities to participate in captures as data recorders or other roles. We have been complimented on our attention to the well-being of the animals by the UC Davis Institutional Animal Care and Use Committee, and we feel that we have helped to set the standards of “best practices” for mountain lion captures and research.

Each animal captured had blood samples taken for assessment of exposure to infectious disease or toxins, and genetic analysis. Nasal, pharyngeal, and rectal swabs were taken in some cases for additional assessment of disease presence, especially viral entities such as SARS Cov2 (Covid 19 virus),

influenza, coronavirus, and microbiome analysis. Fecal samples were taken for assessment of intestinal parasites if possible, and a whisker was taken from most animals for other analyses.

Animals were examined, measured, weighed, ear tagged, and tattooed with their individual study ID number in one ear (unless anesthetic considerations dictated not completing one or more of these items). GPS-collars were applied if body weights exceeded 50 pounds. Cougars were released at the site of capture after recovery from anesthesia. All collars were equipped with either timed or on-demand drop off units, or cotton “spacers” that rot off after a period of time, or some combination of those, This assures that collars drop off the animals after a period of time and are not worn long term.

All data acquired to date from GPS-collared cougars in the southern California, Tehachapis, and Gabilans/Pacheco Pass study areas is or will be curated by Brian Cohen, GIS specialist at The Nature Conservancy office in San Diego. Data from cougars collared in northeastern California is being curated by IWS personnel. CDFW regional biologists and certain state staff were notified of all capture attempts prior to their initiation and invited to observe, depending on the area. CDFW biologists or other personnel have had access to our collar GPS data when requested for their conservation and management purposes. All data has also been provided to the San Diego County Management and Monitoring Program (SDMMP), and other entities whose use of the data could help inform conservation action.

Aliquots of sera samples from all live-captured cougars were stored frozen for later forwarding to CDFW veterinarian Dr. Deana Clifford and Jaime Rudd at the CDFW Wildlife Investigations Lab, and remaining blood, serum, etc is now stored at the University of California South Coast Research and Extension station in Irvine CA. and at the IWS facility in Blue Lake, CA. All deceased cougars recovered in the study areas by the study teams were necropsied at the California Animal Health and Food Safety Lab in San Bernardino if the carcass was not too deteriorated for necropsy. Data on causes of death and locations were added to our overall mortality databases and will be used in analyses that are ongoing. Sample disease and toxin exposure analysis plans are being developed with the collaboration of CDFW and Dr. Jessica Sanchez and others at the San Diego Zoo Wildlife Alliance.

Up until 2021, DNA samples from all live-captured cougars, or deceased cougars from the project area, were taken, processed, maintained, and analyzed or stored for later analysis at the genetics laboratory of Dr. Holly Ernest at the University of Wyoming. During 2021 and 2022 most samples stored at the Ernest Lab were forwarded to Dr. Mike Buchalski at the CDFW genetics lab, as well as new samples taken by our project teams in 2021 and 2022 (scat samples collected opportunistically or via scat sniffing dog team surveys). DNA samples isolated from swabs for saliva taken from puma-killed animals and hair from hair snare sites were also forwarded to the CDFW genetics lab for genotyping. These genotypes from all sources were then used in a population estimation analysis completed in 2022 (Vickers and Dellinger 2022; Appendix A), and are being currently utilized in pedigree updating in concert with Dr. Kyle Gustafson at Arkansas State University, Dr. Mike Buchalski at the CDFW Wildlife Genetics Laboratory, and Dr. Holly Ernest at the University of Wyoming.

In November of 2022, Rogue Detection Dogs was contracted to conduct a range-wide scat collection effort in the SAM. Dr. Dellinger developed a sampling plan based on a grid structure across the range and target areas within each grid based on previous mountain lion GPS data and habitat analyses. Samples collected were also forwarded to Dr. Buchalski at the CDFW genetics lab for DNA isolation. Results will be incorporated in the statewide mountain lion density analysis that is being completed by Dr. Dellinger, and resulting population estimates for the SAM will be compared to those obtained by

other methods.

Some swabs for viral analysis were analyzed at the San Diego Zoo virology lab, and additional analyses of serum for disease exposure analysis, and scat for metabarcoding to assess diet and parasite/other microbe presence is ongoing there under the oversight of Dr. Jessica Sanchez.

Deer used as bait were monitored for signs of disease in cooperation with CDFW veterinarian Dr. Brandon Munk. A summary table of deer collected and examined for disease by IWS and UCD biologists is submitted via the Mandatory Wildlife Reporting System at CDFW. Samples of lymph nodes were stored frozen for later analysis by Dr. Munk.

Capture reports were forwarded to CDFW supervisors Justin Dellinger, Dr. Deana Clifford, regional biologists, and other CDFW personnel specified in our SCP.

In order to study human interactions and potential interactions with pumas our team focused on Whiting Ranch Wilderness Park in Orange County (Figure 12). This park has had multiple incidences where mountain lions were sighted by park visitors or in a few cases had close encounters, either with some element of aggression including attack or apparent lack of fear on the part of the mountain lion involve. The goal of this study that was begun in 2021 and continued through most of 2022 was to better understand how mountain lions, humans, and prey species used Whiting Ranch Park temporally and spatially, and where activities of humans and mountain lions were most likely to overlap.

We placed over 30 cameras across the Whiting Ranch Wilderness Park property on trails and in likely mountain lion travelways, in addition to a group of cameras already present. In early 2022 we also captured and GPS collared 7 mountain lions that might potentially utilize the Park, and opportunistically collected scat and hair when they were encountered on trails or at mountain lion feeding sites. DNA from those samples was incorporated into our SAM population estimation study that was going on simultaneously (Appendix A) These samples, along with photos and GPS data from collared animals, were used to identify a number of individual animals that were moving through the Park periodically, as well as determine how often uncollared / unidentified animals also utilized the Park. Some monitoring is ongoing with both Park and WHC cameras. We intend to continue to analyze camera and GPS data to better understand the interplay of lions, prey species, and human visitors.

Figure 12. Whiting Ranch Wilderness Park (red outline, arrow) with conserved or partially conserved (ie Camp Pendleton Marine Base) lands (green) in the SAM

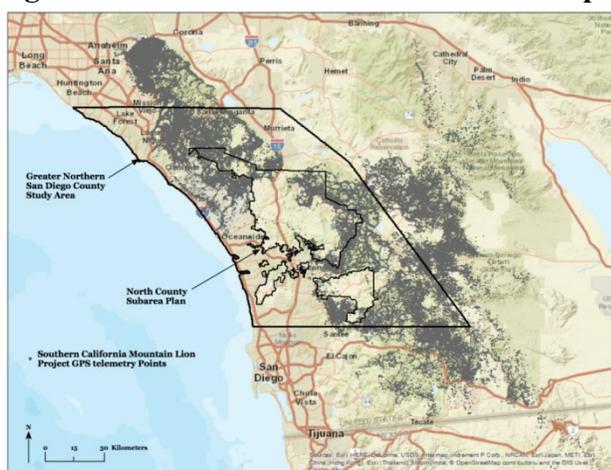


unless they were too young or below minimum weights for collar placement. Several hundred thousand data points have been collected (Figure 13) and utilized extensively in numerous analyses and studies, and by CDFW and various other agencies and entities for conservation purposes.

Some animals have been recaptured several times with no evidence of ill effects from previous capture or GPS-collar placement (significant weight or condition change) being noted.

Of all the handling events, no animals were recorded as having significant injuries that were capture related except for one mortality that occurred in the early years of the study. Capture-related tooth damage or abrasions were sometimes noted but were rare (Dellinger et al 2023 submitted), but we continue to monitor traps closely when set and sedate and remove animals from traps quickly. With hound captures we also take every precaution possible to prevent injury. We feel that the one mortality was preventable and have taken steps to assure that it does not ever happen again in the future. Despite the one mortality, we feel that our safety record during captures is consistent with or better than most other studies (Dellinger et al 2023 submitted).

Figure 13. All collared mountain lion data points through 2016. (Vickers et al. 2017)



In addition, we have handled or sampled opportunistically more than 100 animals after mortality for DNA and disease/toxin assessment, and have collected DNA non-invasively via hair or scat from a large number of individuals (exact numbers dependent on pending analysis at the CDFW wildlife genetics lab).

Additionally, some data has been acquired from 3 cougars captured in a USGS study conducted in the northern Santa Ana Mountains, and those data have been incorporated in our analyses.

Over the course of this long-term study (since early 2001), GPS data points collected from cougars in the southern CA study area have indicated that they have utilized lands as far northwest as the 605 Freeway in the Puente Hills region of Riverside County and as far south as the Parque Nacional Constitución de 1857 located approximately 50 miles south of the border in Mexico. They have ranged from the Pacific shore in the west to the San Jacinto and Peninsular mountain ranges in the east (Figure 13). In the Modoc/Lassen Co. study area GPS collared animals have ranged as far north as into Oregon, and east into Nevada. In the Techachapis study area, the collared mountain lions have circulated south of SR 58 and west of I-5 to date.

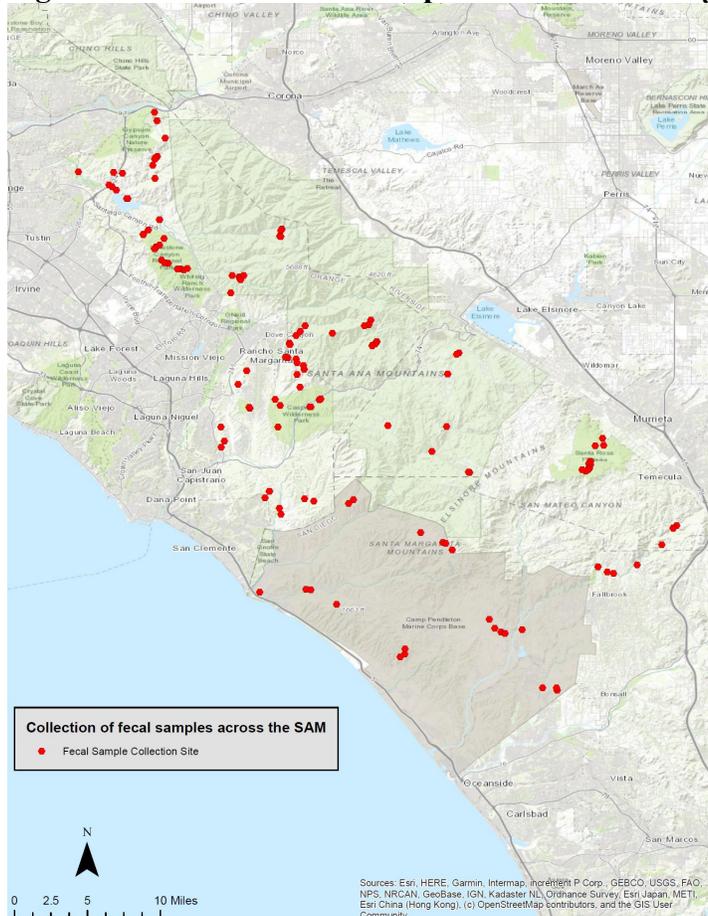
The status of collared cougars whose collars dropped off or stopped transmitting prior to recapture, and captured kittens that were never recaptured for GPS-collaring, is generally unknown. Some of those previously captured and / or collared cougars whose status is unknown are also likely deceased given the mortality rates for collared cougars in the study. Trail cameras have captured photos of some of these animals after their collars dropped off, and they were positively identified by their ear tags. Dates of these confirmations of live status are also recorded in our survival database. Mortalities of some of these individuals have been detected due to their ear tags or tattoos being noted in CDFW reports on depredations, vehicle strikes, or other mortality detections.

2022 and January 2023 Results

Summary of activities in the southern California study area (Orange, Riverside, San Diego Counties):

1. In 2022 we continued assessment of multiple different methods of population monitoring, specifically assessing the use of DNA and photos from hair snares deployed in a grid across the Santa Anas to estimate the population. The population estimation study also utilized identification of individual animals from photos and occurrence data, as well as DNA derived from blood and tissue samples collected opportunistically from captured and deceased animals for disease and genetics assessment and DNA from opportunistically collected scat. The report of that project is attached here as Appendix A.
2. Systematic collection of scat using scat-collection dogs and handlers from Rogue Detection Dogs. The collection effort was conducted across the entirety of the Santa Ana Range in 2022 (n=158 samples collected; Figure 14) to create density estimates to contrast with estimates derived from the hair snare-tissue-scat-photo based density and population estimates derived in 2020 in the eastern Peninsular Range (Appendix A). The quarterly report to CDFW regarding the progress of this effort is attached here as Appendix B.

Figure 14. Sites where scat samples were collected by scat detection dogs



3. We captured (n=9; Tables 1a,b) and GPS collared 8 mountain lions in 2022 in the SAM and ePR for studies of human interactions, survival, movement, connectivity, population estimation, and deterrent testing. One kitten was captured and released without handling during the attempt to capture its mother, F307. We assisted in capture of one abandoned kitten that later expired in captivity from disease. Samples collected at capture were also incorporated into disease and diet studies via serology, metabarcoding, and stable isotope analysis.

4. We continued to collect all deceased mountain lions (n=9 in 2022; n=2 in January 2023; Table 3) that were known in the study areas when possible to submit for necropsy at the California Animal Health and Food Safety Lab to assess overall health and disease and toxin exposure, survival and sources of mortality, collect DNA, and collect physical, demographic, and other information. Some individual animals found were too deteriorated from age or trauma to have necropsies performed but tissue samples were still collected.

5. In 2022 we continued to monitor crossing structures along I-15 (culverts and the Temecula Creek Bridge) with trail cameras for connectivity assessment, and to help guide planning by The Nature Conservancy and Caltrans for improvements at the Temecula Creek Bridge and possible construction of a new wildlife crossing structure (see report to The Nature Conservancy attached as Appendix C; Workshop report pending). We also led a multi-agency group planning for monitoring of crossing structures along I-10, and participated in planning with Caltrans and The Nature Conservancy for

improvements to wildlife crossings on SR79, SR 78, SR76, I-8, and other highways in San Diego County, as well as SR's 74 and 241 in Orange County.

6. We are using genetics with our partners at CDFW (M. Buchalski), U. of Wyoming (H. Ernest), and Arkansas State University (K. Gustafson) to update previous pedigrees and test the ability of different DNA analysis techniques to detect and characterize close relationships between individuals, as well as their subpopulations of origin.

7. We have tested a number of methods of depredation reduction and mortality prevention for both pumas and domestic animals. Some methods have been substantially effective in changing certain movement patterns of GPS-collared mountain lions and reducing returns of collared mountain lions to depredation or bait sites where testing was conducted. Testing has been conducted at multiple sites and on multiple collared animals throughout the study area, and with the collaboration of personnel at the San Diego Zoo Wildlife Alliance (SDZWA) - Dr. 's Jessica Sanchez, Charlie de la Rosa, Matthias Tobler, and others (See quarterly report (2nd quarter 2022) to SANDAG attached as Appendix D, and SDZWA report attached as Appendix E).

8 Also in collaboration with Dr. Sanchez with the SDZWA and the San Diego Conservation Genetics Lab, we initiated studies of DNA extraction techniques for mountain lion hair and swabs (mountain lion saliva from killed animals), metabarcoding of scat for diet and disease assessment, and disease exposure assessment from serum samples (SDZWA report attached as Appendix E). Metabarcoding of scat DNA will be conducted in collaboration with both Dr. Buchalski of the CDFW Wildlife Genetics Lab and the lab of Dr. Ben Sacks at UC Davis.

8. We contributed genetic samples to a statewide mountain lion genome study being led by the University of California, Santa Cruz. This study is expected to yield dramatically new insights into the population genetics dynamics in California pumas.

9. We contributed whisker samples to a stable isotope analysis to determine prey species differences across the state that is being conducted by Kyle Dougherty of Dr. John Benson's lab at the University of Nebraska, Lincoln with the assistance of the UC Davis lab that conducts stable isotope analyses.

10. We are collaborating with researchers from a number of other institutions (UCLA, U. of Texas, UC Davis, and the Universidad Complutense Madridd on a study of how mountain lions are affected by light and sound on approach to roadways. An abstract was presented at an international meeting relating to this effort and a peer reviewed paper is expected to be developed from these analyses. This information will inform mitigation strategies to reduce the barrier effect of roads on mountain lion movement, and roadkills.

11. We collaborated with Dr. Fraser Shilling of the UC Davis Road Ecology Center and Dr. Amy Collins on a study of wildlife response to traffic noise at crossing structures. A peer reviewed publication was published detailing the responses (Collins et al. 2022).

12. We have been tasked by the San Diego County Association of Governments with developing a long term monitoring plan for the ePR population. We have completed much of the experimental work in both the ePR and SAM, as well as a literature review on the subject (See 3rd Qtr report to SANDAG attached as Appendix F). We will be continuing to work on that plan and expect to have a completed product by the end of 2023.

Southern CA study 2022 capture detail:

In the southern CA study, 8 subadults and adults were captured, sampled, and all 8 were GPS-collared (Table 1a,b). One kitten (designated X308) was briefly cage captured then released, and the cage reset for the mother (F307) who was captured later the same night. All 8 GPS-collared animals were new individuals to the study. There were no recaptures. Seven individuals were captured via cage traps at bait / feeding sites, and one (designated M317) was captured by Irvine Animal Control via dart by assistant veterinarian on our SCP, Dr. Scott Weldy, in an Irvine office park building.

Table 1a. Statistics on mountain lions captured during reporting period in the southern CA study area

UCD ID	Capture Date	Method	Sex	Age(mo.)	Ear Tag	Tattoo	Status
X308	01/12/22	Cage	unkown	~3 (estimated visually based on size)	none	none	Lost
F307	01/13/22	Cage	F	37.0	7 orange (right)	307 (left)	Active
F312	01/20/22	Cage	F	27.4	148 orange/yellow (right)	312 (left)	Active
M313	02/05/22	Cage	M	19.7	13 yellow (left)	313 (right)	Active
F315	03/13/22	Cage	F	30.0	15 orange (right)	315 (left)	Active
M316	03/16/22	Cage	M	28.0	yellow 16 (left)	316 (right)	Lost
M317	03/22/22	Free range dart	M	21.0	155 yellow (left)	317 (right)	Active
F320	05/05/22	Cage	F	49.0	149 orange (right)	320 (left)	Active
M321	05/09/22	Cage	M	23.0	142 yellow (left)	321 (right)	Active

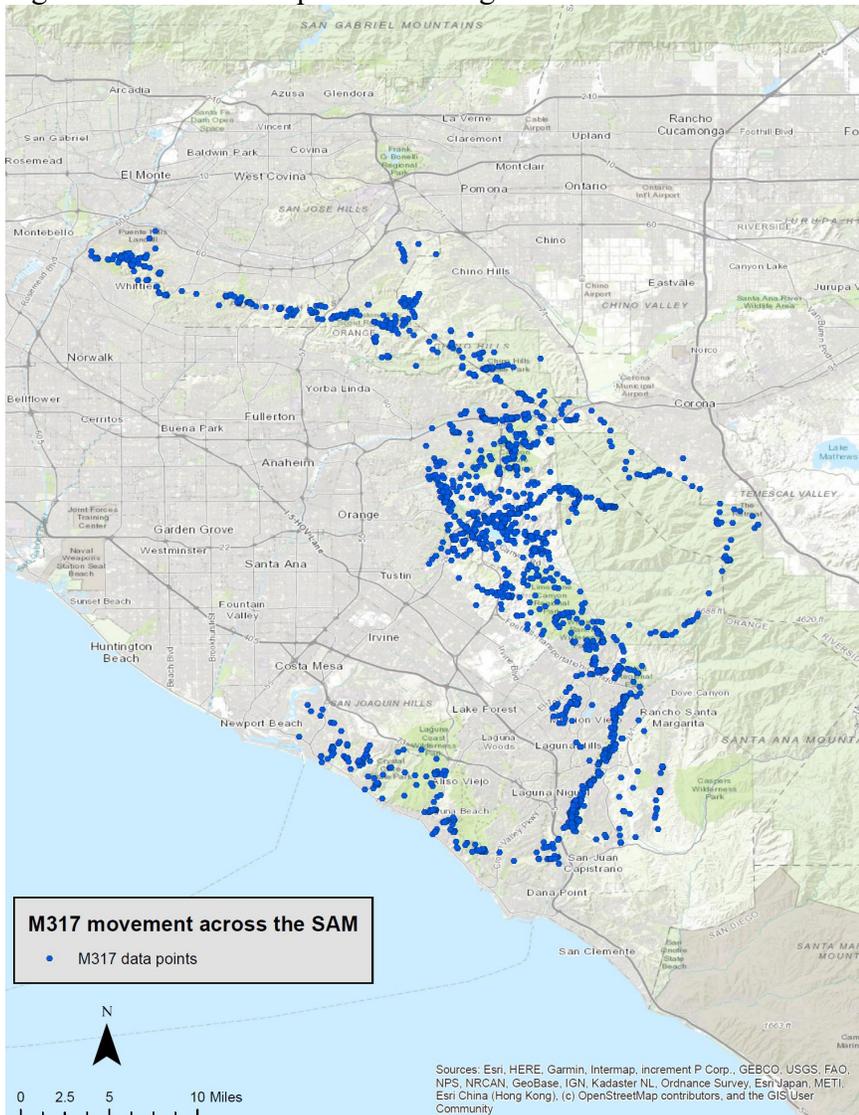
Table 1b. Locations of captures and animal disposition/Transmitter frequencies

UCD ID	General loations	County	UTM Zone	UTM Easting	UTM Northing	Injuries	Mort date	Mort cause	Transmitter frequency
X308	San Diego Zoo Safari Park	San Diego	11	500785.60	3663271.06	None	n/a	n/a	n/a
F307	San Diego Zoo Safari Park	San Diego	11	500785.60	3663271.06	None	n/a	n/a	160.020
F312	Cleveland National Forest	Orange	11	443701.08	3727669.68	Scarred Rt. Cornea	n/a	n/a	160.030
M313	Cleveland National Forest	Orange	11	443717.76	3727669.58	None	n/a	n/a	160.050
F315	Audubon Starr Ranch	Orange	11	449315.01	3720080.95	None	n/a	n/a	160.060
M316	Audubon Starr Ranch	Orange	11	448639.73	3722338.66	Numerous skin wounds all over the body apparently from intraspecies conflict. Skin lacerations face, body, legs in various stages of healing	n/a	n/a	160.010
M317	40 Waterworks Way, Irvine Ca	Orange	11	429063.45	3725314.01	None	n/a	n/a	160.463
F320	Audubon Starr Ranch	Orange	11	448645.99	3722295.38	Abrasion on top of nose	n/a	n/a	148.600
M321	Audubon Starr Ranch	Orange	11	448645.99	3722295.38	Right front paw had broken D1 and D5 claws, old injuries, D5 almost completely missing.	n/a	n/a	160.470
M325	Bear Mountain	Kern	11	351665.35	3898735.28	None	n/a	n/a	160.120
M326	Black Buck Canyon	Kern	11	315930.64	3863490.33	None	n/a	n/a	160.160
F327	Bear Mountain	Kern	11	349875.54	3898467.49	None	n/a	n/a	160.080

The mountain lion captured in the Irvine office park (after running into the open door of a building and being cornered in an office) in March of 2022, M317, has proved to be an especially interesting individual. After capture by dart by Dr. Weldy and Irvine Animal Control, M317 was then sampled and collared by our biologist Juan Gonzalez while under anesthesia and being monitored by Dr. Weldy at his regular veterinary practice. Once recovered from anesthesia, M317 was taken into the Santa Ana Mountains by Irvine Animal Control officers and released.

Since that time M317 has traversed wide areas of the Santa Anas and the Laguna Coast in Orange County, as well as the Chino Hills and Puente Hills in Riverside County (Figure 15). He was recaptured in a Newport Beach neighborhood and re-transported to the Santa Anas and re-released, and has continued to travel widely since then, becoming the first collared mountain lion in our study since 2005 to cross the 91 freeway (n=12 times to date; see roads discussion below).

Figure 15. M317 data points in Orange and Riverside Counties.



Roads as sources of mortality and barriers in the southern California study area:

Except for a portion of SR 241 in Orange County, Los Patrones Parkway in Orange County, portions of SR 76 west of I-15, and along I-15 south of Temecula, no major highways in the region have adequate fencing to prevent cougar intrusion onto roads and funnel them to safe crossing structures.

Our team has studied multiple regional highways at nearly 200 sites where known mountain lion crossings had occurred (based on high frequency GPS data, cameras, and roadkill), least cost path modeling indicating likely crossing points (Zeller et al 2017,2018), or expert opinion indicated likely crossings. All sites were examined in depth and all findings documented (Vickers et al. 2020).

Findings of that study included that most puma crossing points did not have adequate infrastructure present to allow safe passage, most crossing points had less than 50% of the land within a 500 meter radius conserved, and at most crossing points where adequate infrastructure was present, the land was not conserved at one or both ends of the structure (Vickers et al. 2020).

The lack of conservation at existing adequate structures was of special concern because of the potential for functional loss of some of the adequate structures due to development. Another major concern was that the majority of crossings were occurring at grade where no infrastructure existed, either crossing structures or exclusion fencing, and thus both mountain lions and drivers were being put at risk.

Given that our survival analysis indicated that females were killed as often as males on roads in this region and females are more crucial to the stability of the population, it increases the importance of decreasing this source of mortality as much as possible.

Our mortality data from 2022 and Jan 2023 brings home this point. Of the 10 known mortalities in the region, 60% (n=6) were from vehicle strikes with one additional mortality suspected to be related to a vehicle impact, though not on a highway. Other uncollared cougars may have been killed by vehicles or on depredation permits in the southern CA study area that we were not made aware of.

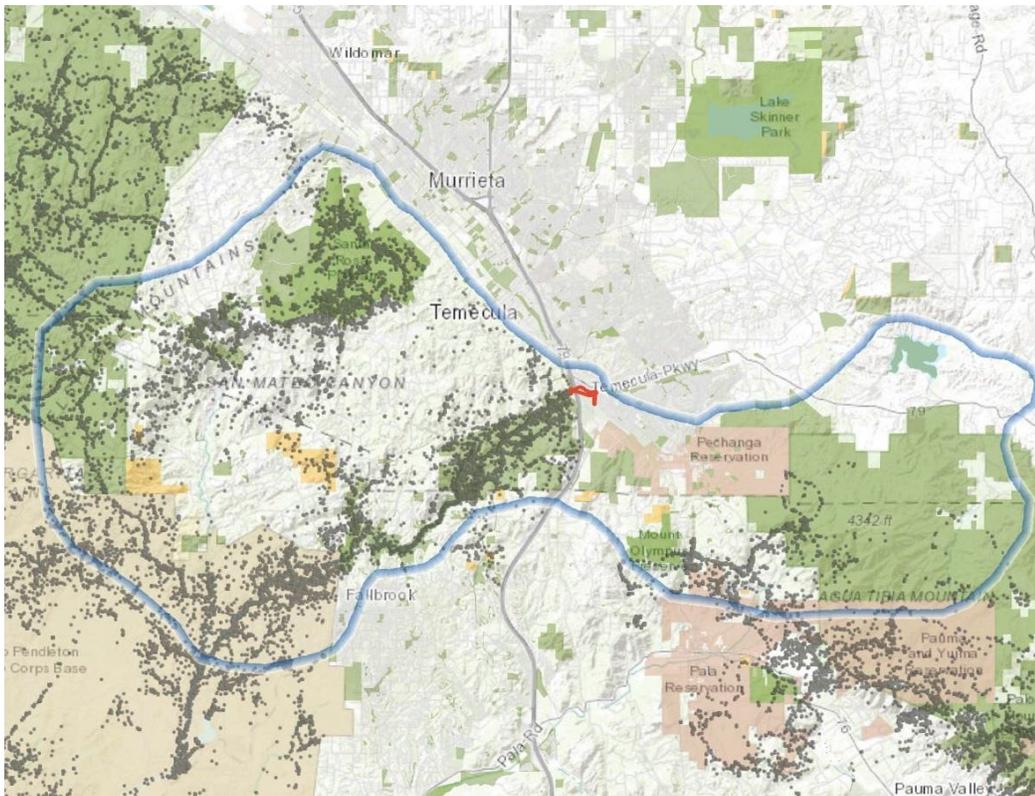
Table 3. Overall mortalities in the southern CA study area:

ID	Date of mortality	Population	Sex	Cause of death	Lat (WGS84)	Long (WGS 84)	Age	County
X314	1/1/2022*	ePR	?	Unknown - too deteriorated	33.06431	-116.95655	Est 1 yr	San Diego
M318	04/03/22	SAM	M	Vehicle strike	34.02773	-117.79962	Est 2 yr	Orange
F319	04/04/22	San Bernardinoos	F	Probable poaching	34.16983	-117.631700	10 yr	San Bernardino
X322	5/1/2022*	SAM	?	Unknown - possible vehicle strike	33.49522	-117.57771	Est 10 mo	Orange
F323	6/23/2022*	ePR	F	Vehicle strike	33.26242	-116.806752	10 mo	San Diego
F371	10/14/22	SAM	F	Disease/Abandoned by mother	33.71102	-117.61826	4 mo	Orange
X372	10/14/22	SAM	?	Vehicle strike	33.69628	-117.63288	4mo	Orange
X373	Before Nov 2022*	SAM	?	Unknown	33.80187	-117.68738	?	Orange
F374	12/01/22	SAM	F	Vehicle strike	33.51478	117.5731780	Est 24 mo	Orange
M375	01/06/23	ePR	M	Vehicle strike	32.815528	-116.526356	Juvenile per CDFW	San Diego
X376	01/20/23	SAM	?	Vehicle strike	33.67395	-117.64205	7 mo	Orange

In 2022 we continued to monitor crossing culverts on I-15, and Temecula Creek as it passes under the freeway south of Temecula, as well as Pechanga Creek east of its junction with Temecula Creek (Figure 16). During monitoring from 2018 - 2021, we observed only one mountain lion (uncollared) crossing under the Temecula Creek Bridge, and it crossed west to east. Two collared dispersal age mountain lions crossed I-15 west to east during that period. One crossed in Murrieta, and one near Escondido, but

both returned to the Santa Anas after relatively short periods of time.

Figure 16. SAM to ePR linkage area with mountain lion data through 2020 depicted. Red line is Temecula Creek at the I-15 bridge area.



In the period April 2021- late 2022, our cameras did not record any crossings of I-15 by mountain lions, but did record activity in and through the Temecula Creek crossing by other wildlife, though very little activity through large culverts further south. Our monitoring of Temecula Creek and Pechanga Creek SE of the junction of the two found that we photographed humans more often than all other terrestrial mammalian wildlife animals combined (see Vickers 2022 attached as **Appendix C**). During that period, no mountain lion activity was recorded near the freeway culverts, despite fairly regular presence in previous years (n=16 documentations during 2018-2021).

However, on the somewhat positive side, during 2022 and January 2023, our seven GPS-collared mountain lions in Orange and Riverside Counties, and one in San Diego County, have crossed busy freeways, tollroads, and state highways 260 times with no mortalities (Table 4). M317 has been particularly busy in that regard with 81 highway crossings due to his wide ranging travels as noted above (Table 4; Figures 17a,b,c). It is notable though that 3 other GPS-collared mountain lions had over 30 crossings each of these highways, with F315 having 69 crossings. This illustrates how mountain lions in southern California literally find it impossible to have territories in this landscape without crossing busy highways.

Table 4. Numbers of crossings of busy freeways, tollroads, and major highways in 2022 by 8 GPS-collared mountain lions.

Lion ID	Grand Total	Hwy Number										
		1	57	74	78	91	133	142	241	261	I-5	
F307	14				14							
F312	12								12			
F315	69			69								
F320	0											
M313	42								42			
M316	39			39								
M317	81	2	4			12	1	6	47	6	3	
M321	3			1					2			
Grand Total	260	2	4	109	14	12	1	6	103	6	3	

Figure 17a. M317 data with lines between data points (predominantly at 3 hour intervals) in the Chino Hills and Puente Hills, including multiple crossings of the 57 freeway.

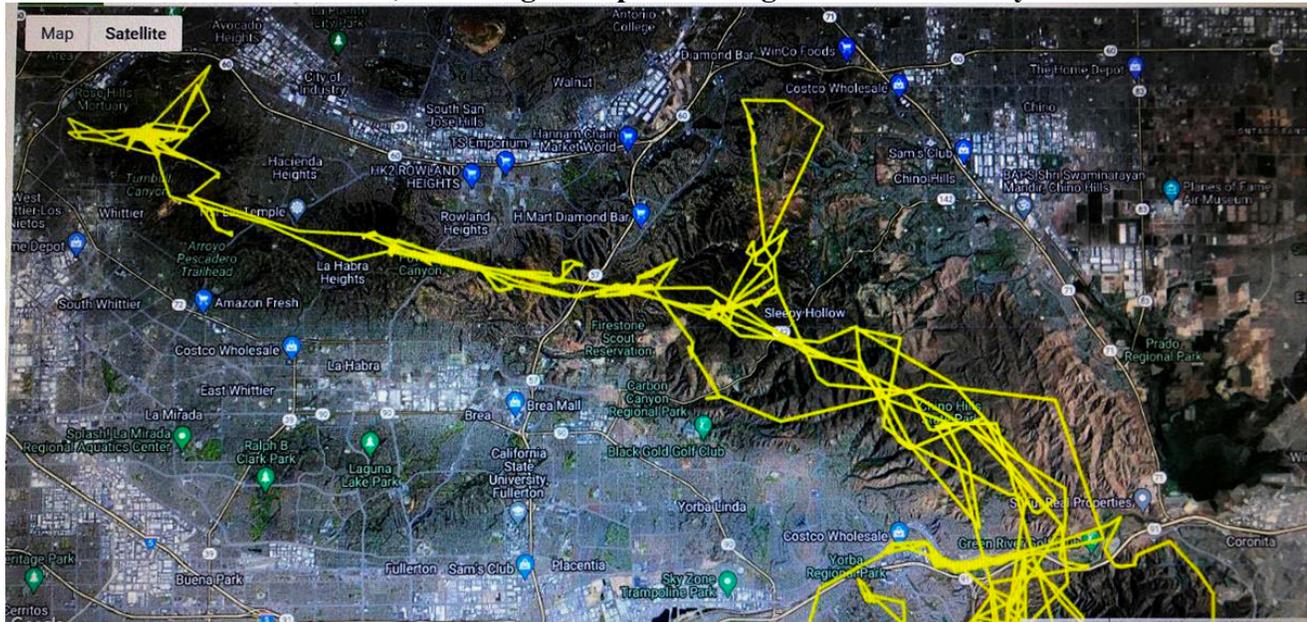


Figure 17b. M317 data with lines between data points (predominantly at 3 hour intervals) in the 91 Freeway vicinity.

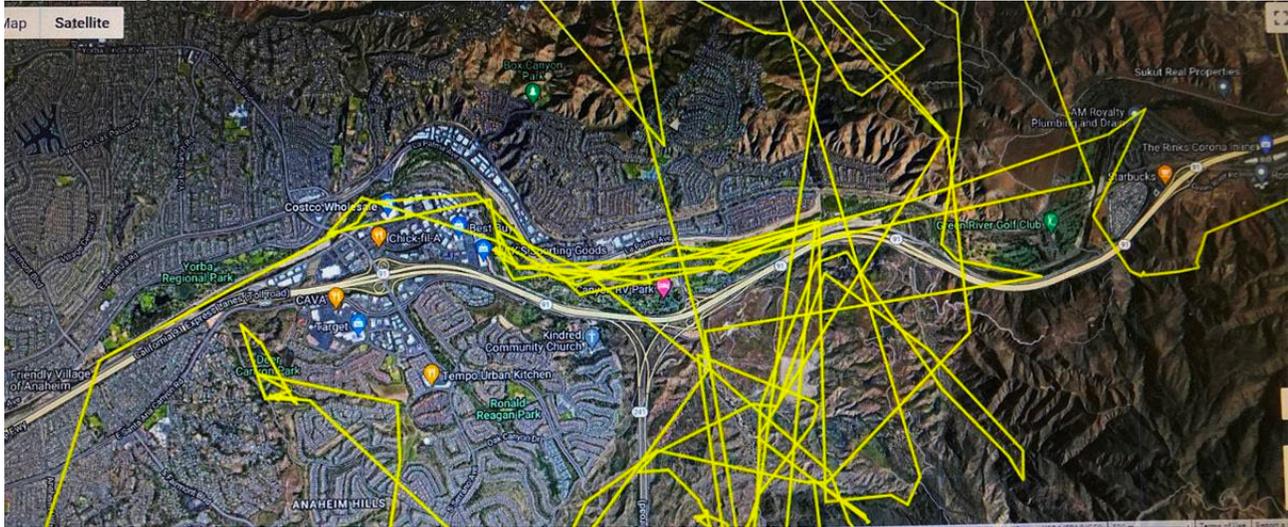
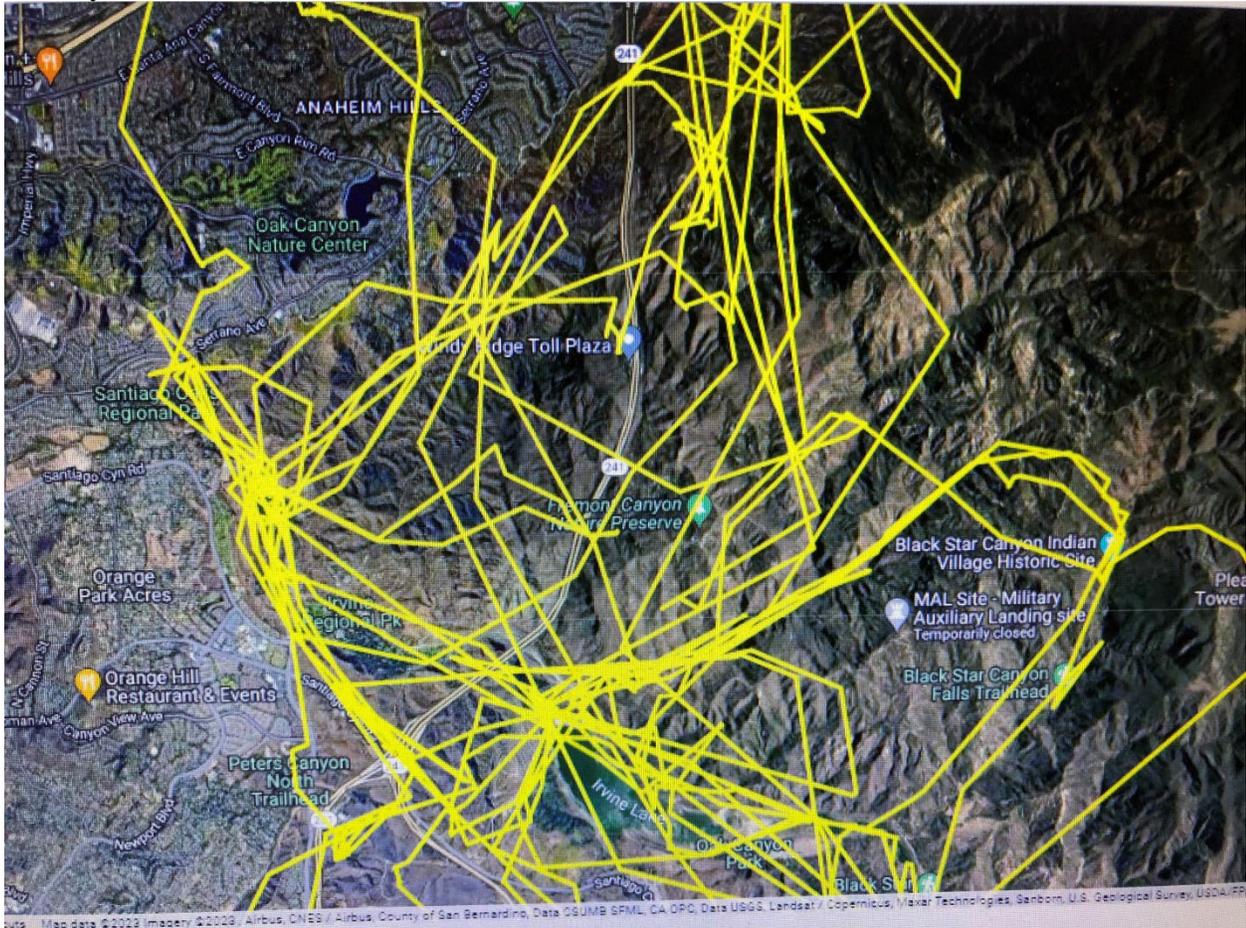


Figure 17c. M317 data with lines between data points (predominantly at 3 hour intervals) in the vicinity of the SR241 and SR261 toll roads.



Extensive tall and buried wildlife fencing with barbed wire outriggers (Figure 18) and safe crossing structures (bridges and large culverts; Figures 19,20), are present in the area where the majority of crossings of the SR241 and SR261 tollroads by collared mountain lions occurred. Since construction of this fencing project, no mountain lion, bobcat, or deer have been killed in the fenced section (approximately 6 miles), and only a small number of coyotes. This is a dramatic reduction from the period before the fencing was built. Four individual GPS-collared mountain lions crossed these two tollroads 109 times in the reporting period, tied with SR74 for the most crossings.

Figure18. SR241 Wildlife Protection Fence on SR241



Figure 19. SR241 Oak Canyon Wildlife Undercrossing.



Figure 20. SR 241 culvert dual purpose culvert (water conveyance and wildlife undercrossing)



SR74 (the Ortega Highway), also had 109 crossings, but by only 3 individuals. One of the 3 only crossed once, so 108 crossings were by 2 individuals (Figure 21).

Figure 21. Lines between data points from 3 GPS collared mountain lions in the vicinity of SR74 (arrows) that crosses the SAM east to west



Several safe crossing structures exist in the section of SR 74 where the majority of crossings occurred (Figure 22), but cameras at those structures have only captured these individuals using the structures a handful of times. It is most likely that the majority of these crossings have occurred at grade, endangering both the mountain lions and drivers.

Figure 22. A mountain lion and kittens use a safe crossing structure on SR74



Genetics, Disease, Mortality, and Toxins:

We continued to collect tissue samples with CDFW of deceased cougars in the study area, and record the locations and causes of death. We also have had every deceased mountain lion from the region necropsied at the California Health and Food Safety Lab if the carcasses were not too deteriorated. This will assist in assessment of health, disease, toxin exposure, and genetics of live and deceased cougars in the study area, as well as mortality patterns.

We had previously contributed serum and whole blood samples to a study of feline leukemia in pumas and domestic cats. That study was published (Petch et al. 2022) and determined that the source of the virus in pumas is usually domestic cats but puma to puma transmission can occur, and pro-viral loads were higher in populations that were more genetically restricted. Because epizootics of feline leukemia have affected puma populations in certain areas, and puma to puma transmission has been demonstrated, this raises the level of concern about this virus's ability to affect the small genetically restricted population in the SAM. The proximity of pumas to domestic cats in the SAM makes introduction of the virus to this population a possibility.

Our study's previous findings (Vickers et al. 2015) and those of a statewide survival study that will likely be published in 2023 (Benson et al. 2023). Both studies confirm that annual survival rates of collared pumas in our southern CA study area are low enough to be of significant concern for these populations.

Deaths due to vehicle collisions and secondary to depredation permits are the first and second most common causes of death in our southern California study area. In 2022, vehicle collisions continued to be the most common cause of death that we detected. All mortalities were uncollared animals so we would be unlikely to detect deaths other than vehicle strikes and depredation kills. However, we did detect carcasses for which a cause could not be determined and one abandoned kitten that died of illness at Ramona Wildlife Care after it was captured with Dr. Weldy's assistance (Table 3).

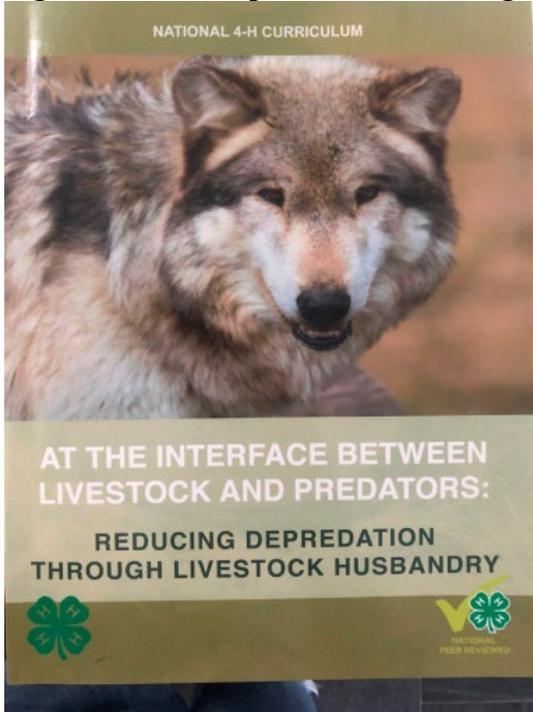
Both of the two referenced studies emphasize the need for long term monitoring of the at-risk Santa Anas population, a major goal of our current work in southern California.

Education and depredation prevention:

In 2022, we continued to educate and inform the general public, livestock and pet owners, agency personnel and managers, and others about mountain lions and how to most successfully coexist with them in the southern California landscape via prevention of depredation events.

In 2021, Dr. Vickers has worked to distribute a peer-reviewed curriculum that he co-authored for use by 4-H clubs to teach students how to prevent depredation (Cheung et al. 2019, Figure 23). That curriculum resulted from a collaboration with the Mountain Lion Foundation, UC Extension, and UC Davis Veterinary School

Figure 23. Cover photo from Cheung et al. 2019



In 2022, our team continued to assess the effectiveness of different hazing and deterrent devices on cougars and is collaborating with other mountain lion researchers and CDFW in that effort (Appendices D, E). Some deterrent effects were observed with motion triggered deterrent devices as well as with putting opaque material on livestock pen fences so that the pumas could not see where they were landing if they jumped the fence in some depredation sites. Repetitive exposure to deterrent devices, along with certain randomly played sounds, has appeared to be most effective at deterring cougars from frequenting an area. Further experimenting is ongoing in cooperation with the San Diego Zoo Wild Animal Park and other entities (Appendix E).

With producer Kate Remsen we created a series of short documentary films and one full length film about mountain lions in California that are intended as educational tools for use by various entities in the state such as CDFW, Parks Departments, and non-profits that interact with the public on the subject of mountain lions. As of this writing in early 2023 they have been viewed nearly 2 million times online at <https://camountainlions.com/> and elsewhere.

Developing and comparing population monitoring methods for the Santa Ana Mountains.

In 2021, we also began collaborating with USGS on testing the use of photos from the hair snare sites to potentially identify individual mountain lions for use in spatial capture-recapture modeling. Several thousand photos of known mountain lions from several study areas were incorporated in the library of images to be analyzed by Dr. Jeff Tracey of USGS. That effort has continued in 2022.

Another monitoring method, estimation of the population density based on DNA from scat collected systematically by scat detection dogs across the Santa Ana Range, was tested by conducting collections in this way in November 2022 (see 4th Quarter 2022 report to CDFW, Appendix B). Rogue Detection Dogs was contracted to do the scat collections as they had in multiple other parts of the state previously for CDFW. Isolation of the DNA from the scat samples and genotyping is being conducted by the CDFW Wildlife Genetics Lab. Results of those analyses are going to be included in a statewide

population estimate being prepared by Dr. Dellinger, and utilized to compare to density estimates derived from hair, scat, and other collections and reported in Vickers et al. (2022; Appendix A)

Analysis of population viability in the Santa Ana Mountains:

A population viability analysis published in 2019 (Benson et al.) suggests that increased connectivity and movement of mountain lions from the adjacent areas into the Santa Anas appears to be key to the viability of mountain lions in the Santa Anas in terms of reducing probability of extinction due to both demographic and genetic factors.

With immigration at a low level consistent with what has been documented during the last 15 years (3 males; Gustafson et al. 2017), the likelihood of extirpation of the population over the next 50 years is uncomfortably high (1 in 5 or 6 chance). At the current or lower levels of immigration, genetic diversity (heterozygosity) is predicted to decline substantially in the next 50 years raising concerns about inbreeding depression (Benson et al. 2019). If inbreeding depression develops then extirpation in a much shorter time frame (<15 yrs) becomes very likely without intervention (Benson et al. 2019).

Initial results of semen quality analysis in 2020 from two male lions in the study area that died (one from the Santa Anas and one from the eastern Peninsula Range population) teratospermia was present (high percentages of abnormal sperm; Huffmeyer et al. 2021). Poor semen quality was also documented in the SMM populations and new observations of kinked tails were noted there. These findings of signs of inbreeding depression amplify concerns relating to future decline of both of those relatively isolated populations.

Subsequently, an analysis of semen from an additional deceased male from the SAM yielded inadequate sperm numbers due to the young age of the male (17 months). Further analyses will be done as opportunities for sampling arise. Similar assessments are ongoing in the Santa Monica Mountains population and our collaborators at the SDZWA reproductive lab are assisting in these evaluations.

These findings highlight the importance of continuing to monitor the SMM and SAM populations closely, and slow the rate of loss of genetic diversity to avoid the potential for compromised demographic performance due to inbreeding depression. Dr. Benson's results suggest that movement of individuals into the Santa Ana's needs to occur at a higher rate than we have documented to date in order to avoid the projected further loss of genetic diversity. This again highlights the importance of maintaining and increasing linkages that allow mountain lions to move from adjacent areas into the Santa Anas.

Camera monitoring of pumas in the Whiting Ranch Wilderness Park in Orange County

Figure 24. Whiting Ranch Wilderness Park (red outline) and other conserved lands (green) and its relationship to residential development to the SW, S, SE, and some to the N..

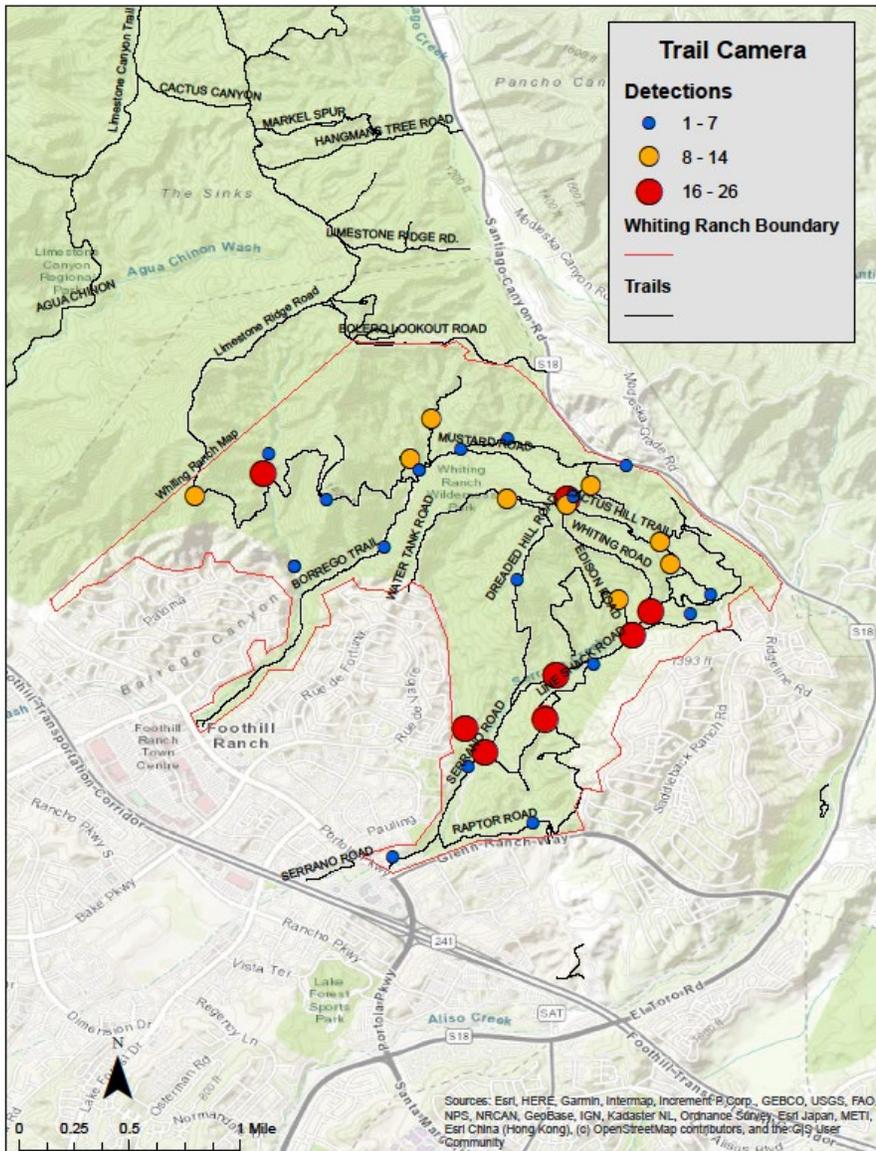


Beginning in August 2021, we placed more than 25 cameras in the Whiting Ranch Wilderness Park on Orange County, a small (less than 3 sq.mi) park that is bordered on one side by conserved lands, and on the other 3 sides by residential development and a highway, Santiago Canyon Rd that has moderately heavy traffic at times and has been the site of a number of collisions between mountain lions and vehicles. Approximately 10-12 cameras belonging to OC Parks were already active there and recording puma presence periodically. A total of over 40 cameras were active at some time or location in the Park during the study period from August 2021 to August 2022. The vast majority of the cameras were on trails also used by humans.

The Park is heavily visited by hikers, mountain bikers, and trail runners, with a level of visitation estimated to be over 97,000 visitors per year just prior to the SARs CoV2 pandemic (Monz et al. 2019). Visitation was thought to have gone up substantially during the period of our study. Our preliminary data from our cameras suggests that the Ranger impressions are correct.

One or more mountain lions were recorded on one or more cameras in the Park on over half of the calendar days during the study period (Figure 25). Four of the 7 mountain lions that were GPS collared in 2022 in the SAM used the park, with one of the females F312, and a male M313, apparently incorporating the Park in their regular territorial patterns. We found that one or more cameras detected every visit of collared mountain lions within the Park boundaries, so we felt that if any mountain lion entered the Park that it was being detected by at least one camera.

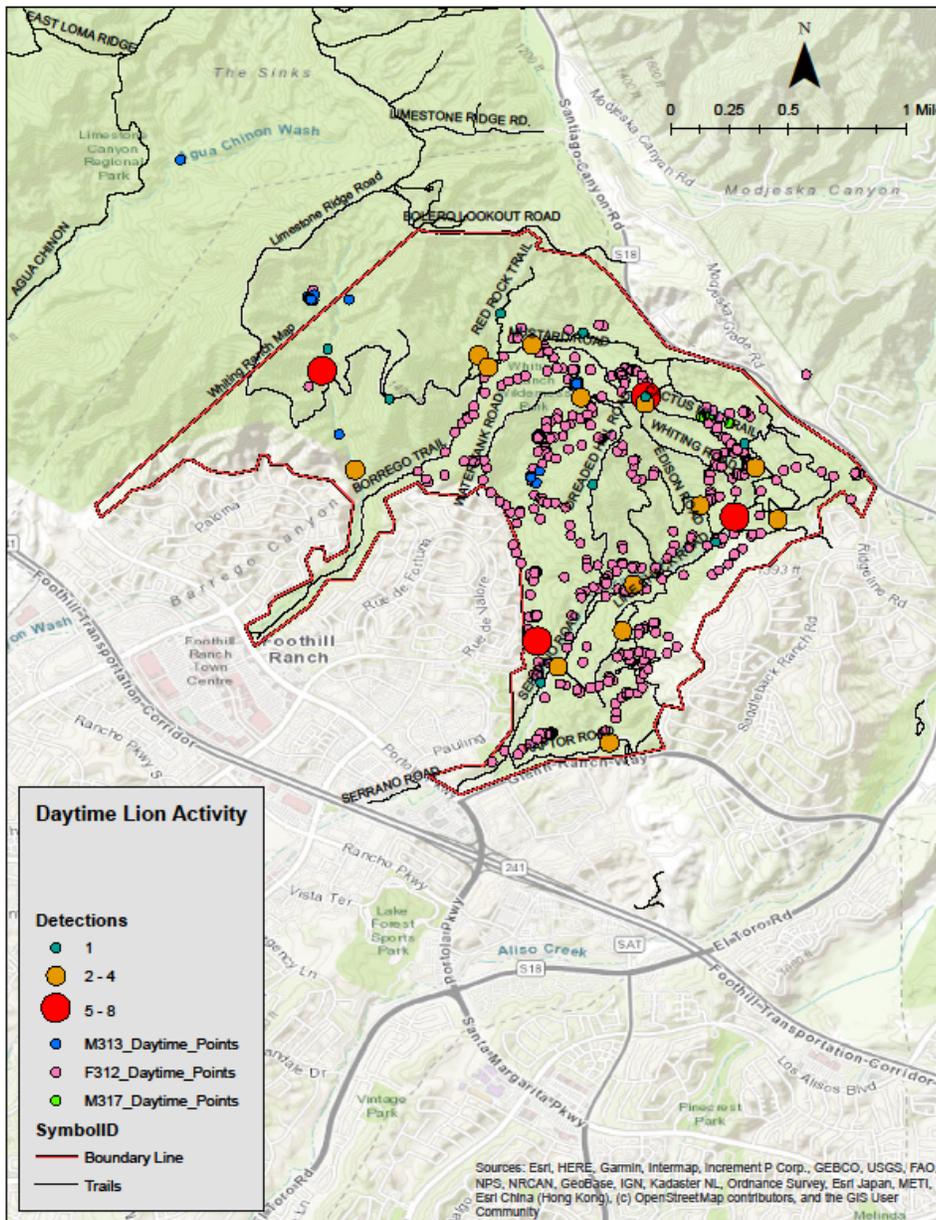
Figure 25. Locations where mountain lion detections occurred within the Park boundaries in the first 9 months of the study.



During the one year study period, mountain lion presence was recorded on camera 385 times, with 551 individual mountain lions being detected due to family groups being present in many instances. Collared individuals were the mountain lions photographed on 90 of the 385 occasions, but could have been photographed on other occasions before being collared.

The majority of the photos of mountain lions were at night, however there were a substantial number during the daytime hours when human visitors were on the trails. Figure 26 below depicts approximately the first 9 months of data and photos taken daylight hours, as well as daytime datapoints from 3 of the 4 collared mountain lions that used the Park.

Figure 26. Daylight photos taken of mountain lions in the Park and daylight data points of 3 collared mountain lions during the first 9 months of the study.



We are working with Orange County Parks staff to increase training of Rangers and education of Park visitors about mountain lions in order to minimize the likelihood of negative outcomes of encounters. We are also working to develop a graduate level analysis of the impacts of people on other wildlife in the Park and to better understand the interplay between people, mountain lions, and other wildlife.

In the northeastern California study area:

NE CA study capture detail:

From 1 January through 31 December 2022, 12 mountain lions were handled on the NE CA project (Table5). Five were adults (2 males, 3 females), and seven were juveniles (5 males, 2 females). The five of the adults were all GPS collared or re-collared, two of which were recaptures. Of the 7 juveniles captured, 4 were collared with as part of our dispersal study, and the remaining three were not collared as they were too young or did not meet our minimum body weight threshold. Of the two adult lions were recaptures: M216, a 55-month-old male whose previous collar had failed on 9/26/20 was recaptured in April; F204, a 63-month-old female whose collar had also failed was recaptured in December. All captures went without incident.

Table 5a. Statistics on mountain lions captured during reporting period in Modoc and Lassen counties, California.

ID	Date	Method	Sex	Age	Ear Tag	Tattoo
M282	1/30/2022	Cage Trap	M	19 mo.	282 -L-Org	N/A
M283	2/3/2022	Cage Trap	M	56 mo.	283-L-Org	283-R-Blk
M284	2/24/2022	Cage Trap	M	5 mo.	284-L-Org	N/A
F285	2/24/2022	Cage Trap	F	5 mo.	285-R-Org	N/A
F286	3/16/2022	Cage Trap	F	16 mo.	286-R-Org	N/A
M289	3/17/2022	Cage Trap	M	7 mo.	291-L-Org	N/A
M290	3/17/2022	Cage Trap	M	7 mo.	292-L-Org	N/A
F287	4/4/2022	Cage Trap	F	31 mo.	287-R-Org	N/A
M216*	4/14/2022	Cage Trap	M	55 mo.	216-L-Org	N/A
F340	4/19/2022	Cage Trap	F	120+ mo.	340-R-Org	N/A
M341	4/21/2022	Cage Trap	M	18 mo.	341-L-Org	341-R-Blk
F204*	12/14/2022	Cage Trap	F	63 mo.	204-R-Org	204-L-Blk

*Indicates a recaptured mountain lion.

Table 5b. Locations of captures and animal disposition/Transmitter frequencies

ID	Gen'l Locale	County	UTM Zone	UTM Easting	UTM Northing	Injuries	Mort. Date	Mort Cause	Trans. Freq.
M282	Yellowjacket Spring	Modoc	10	664317	4586620	None	N/A	N/A	160.165
M283	West Valley Reservoir	Modoc	10	716984	4566809	None	N/A	N/A	173.625
M284	West Valley Reservoir	Modoc	10	716984	4566809	None	N/A	N/A	N/A
F285	West Valley Reservoir	Modoc	10	716984	4566809	None	N/A	N/A	N/A
F286	Timbered Mountain	Modoc	10	687335	4621846	None	N/A	N/A	160.215
M289	Rail Mountain	Modoc	10	670146	4603307	None	3/19/2022	Testicular hernia	160.175
M290	Rail Mountain	Modoc	10	670146	4603307	None	N/A	N/A	N/A
F287	Whittemore Ridge	Modoc	10	691648	4611782	None	N/A	N/A	173.400
M216	West Valley Reservoir	Modoc	10	716984	4566809	None	N/A	N/A	173.675
F340	Whittemore Ridge	Modoc	10	691648	4611782	None	N/A	N/A	173.500
M341	Mud Lake2, Modoc Co.	Modoc	10	715447	4565048	None	N/A	N/A	160.170
F204	West Valley Reservoir	Modoc	10	716984	4566809	None	N/A	N/A	173.021

We only had one mortality of a collared lion during the reporting period. A juvenile male lion (M289) was captured on 3/17/22 along with its suspected sibling (M290). On 3/19/22 we received a mortality signal on lion M289 and recovered the body on 3/20/22. We transported the carcass to the WIL lab at CDFW and then to the UC Davis Veterinary Hospital for necropsy. Cause of death was determined to be

a testicular hernia and was not associated with the capture or handling. There was a mortality of a previously captured but uncollared dispersed male (M273) that was killed on 12/29/22 by a hunter near Keno, Oregon; 10T 588777, 4660858. It was originally captured on 3/4/21 near Cuppy Butte in Modoc County and aged at approximately 8 months old at the time of capture.

NE CA Project Research Detail:

Ten of the adult and juvenile lions that were captured in 2022 were tested for FIV and FeLV using the Witness Antibody Test Kits (Zoetis Inc.) and all were negative.

We collared juvenile mountain lions in 2021/2022 as part of a study examining dispersal characteristics in an un hunted population. It was hypothesized that in an un hunted population there may be fewer territories that become vacant compared to a hunted population, therefore creating different challenges and possibly a higher mortality rate from natural causes. The project is being conducted in collaboration with Utah State University and our biologist on the project will be using this aspect of our overall study as his graduate project.

In 2022, investigations of mountain lion diet in the NE California study area has generally followed what has been reported elsewhere for the species, with deer being the primary prey item. From January through August 2022, deer were the primary (44%; n=66) prey item found at clusters investigated with prey items present, with feral horses (36%), coyotes (6%), pronghorn (6%), and elk (3%) also recorded. Other prey items such as one bobcat and one beaver were also observed at kill sites. While one adult male lion accounted for 58% of the wild horses taken as prey, two female lions were responsible for the remaining 42% of the feral horses at kill sites investigated in 2022.

How Data from the NE CA study is being collected and used for mountain lion conservation throughout California:

- Contributed to paper on habitat selection of mountain lions in California (Dellinger et al. 2019)
- Contributed data to current work on a paper on mountain lion survival in California (J. Benson et al. in review, PNAS)
- Transferred whiskers collected from mountain lions captured in NE California to Dr. John Benson at University of Nebraska who is collaborating with CDFW on a project using stable isotopes to examine food habitats across California
- Collaborating on project to compare disease exposure of mountain lions across California in relation to human population density (with Dr. Jessica Sanchez, San Diego Wild Animal Park)
- Currently working on a paper for publication from a master's thesis at Humboldt State University (IWS supported this graduate student research) examining kill site habitat selection by mountain lions in Modoc and Lassen counties.
- Supporting a graduate school project (Utah State University) to examine dispersal behavior of mountain lions from NE California
- Currently working on a paper examining denning behavior of mountain lions in Modoc and Lassen counties (Brinkman et al. in prep)

-Currently working on paper examining pronghorn demographics in northeastern California which includes impacts of mountain lion predation (Garcelon et al. in prep)

-Currently working on a project to examine familial/pedigree analysis of mountain lions in northeastern California using genetic samples (Analyses pending)

Tehachapi Mountains Project

Current efforts are underway to improve habitat connectivity among isolated mountain ranges in southern California to support sustainable wildlife populations, including the Santa Monica, Santa Ana Mountains, Tehachapi Mountains and the Transverse Range. However, data show that the success of these current efforts is ultimately contingent upon restoring connectivity between these isolated mountain ranges and the Sierra Nevada Mountains (Figures 27,28). Thus, to successfully ensure viable wildlife populations in all portions of southern California, and along the central coast of California, there is a great need to better understand current wildlife connectivity and gene flow across existing major transportation routes.

To that end, in July 2022, we initiated our study in the Tehachapi Range with funding from the Wildlife Conservation Network (WCN) and The Nature Conservancy (in-kind). The Tehachapi Mountains have substantial prime mountain lion habitat, and we feel it is critical to understand as best we can the movement of mountain lions through the Range.

Figure 27. Connectivity areas of focus (red arrows) and major highways in the Tehachapis and southern California region.

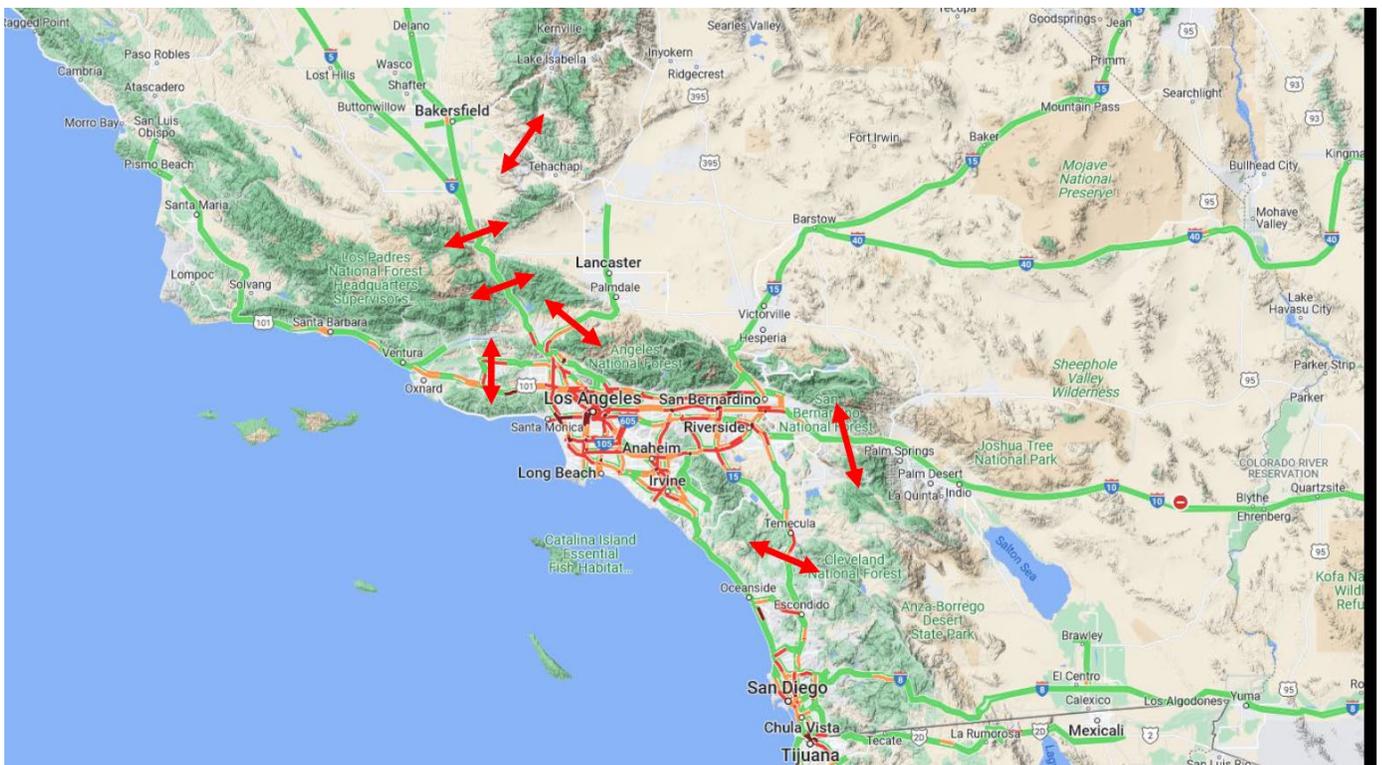
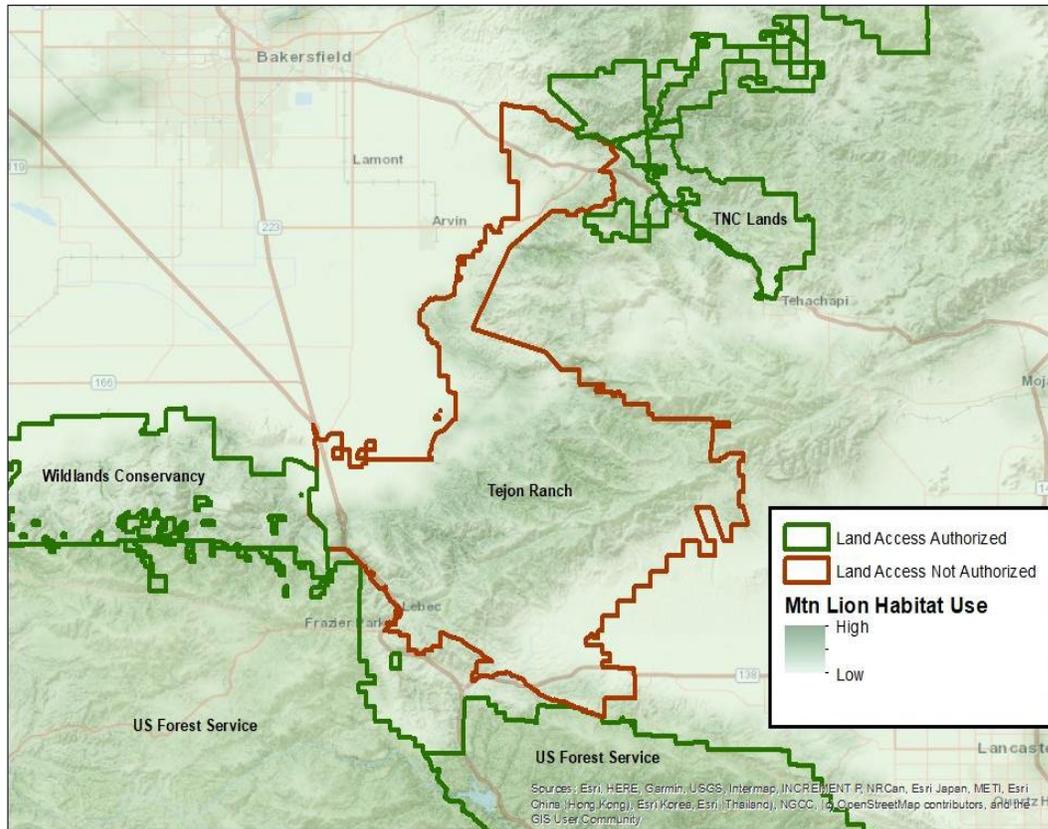


Figure 28. Mountain lion habitat in the Tehachapi Range with I-5 and SR 58 shown, and lands adjacent to those highways where proposed camera and mountain lion capture work would focus.



Tasks and Approach:

Task 1. Monitor use of existing crossing structures by multiple wildlife species using remote cameras (Ford et al. 2009): Caltrans and USFS, as well as CDFW, are willing collaborators with UCD in placing and monitoring cameras at crossings along SR58 and I-5 in the study area (Figures 29,30)

Figure 29. Focal area of planned camera monitoring of crossing structures along SR 58 in the Tehachapi Mountains with conserved lands shown.

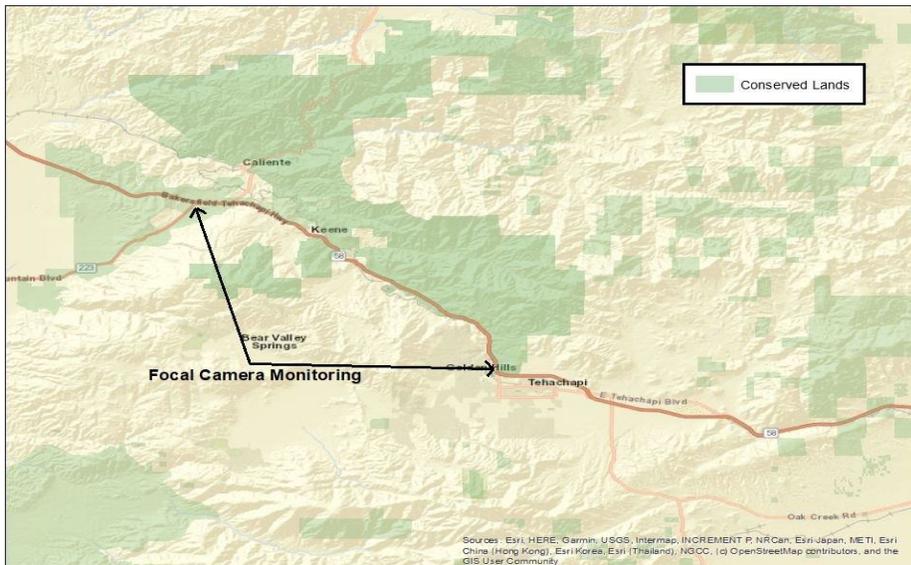
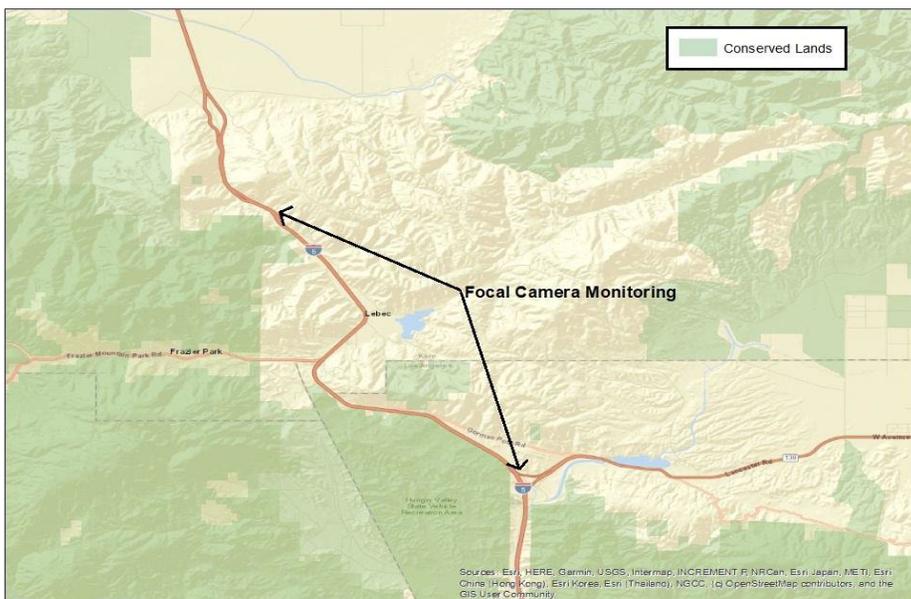


Figure 30. Focal area of planned camera monitoring of crossing structures along Interstate 15 in the Tehachapi Mountains with conserved lands shown.



Task 2. Determine if and where additional crossing structures might be needed by capturing and deploying GPS radio-collars on mountain lions (Zeller et al. 2017): Take samples for genetics studies and other purposes. Collars will be remotely programmed if possible to increase location fix rate to get finer scale road crossing data.

Task 3. Understand gene flow of mountain lions and other mammals by opportunistically gathering genetic samples e.g., from captures, roadkill, scat, and hair; (Gustafson et al. 2017): Genetic samples can help document the current level of gene flow in relation to major roadways with existing crossing structures. Though mountain lions are being targeted, this can be done opportunistically with multiple species. CDFW's Wildlife Genetics Lab has committed to storing and analyzing all mountain lion genetic samples and would take high quality genetic samples (e.g., tissue) from other species (Mike Buchalski – CDFW Wildlife Geneticist; michael.buchalski@wildlife.ca.gov). If connectivity is improved, additional genetic samples can be used to monitor changes in gene flow in response to mitigation measures.

Task 4. Compile results for informing efforts to improve/maintain connectivity given existing and future transportation infrastructure: After processing photos, occupancy and detection analyses will be used to analyze remote camera data (Dellinger et al. 2019). Resource and step selection function models will be used to analyze radio-collar location data (Dellinger et al. 2020b). Genetic samples will be analyzed using a single nucleotide polymorphism (SNP) assay to determine individual ID's and contribute to understanding of genetic connectivity (Buchalski et al. 2022).

Activities by Task

Task 1.

We have coordinated with CDFW and Caltrans biologists on combined monitoring of highway crossing structures along SR 58. Cameras were already in place at some crossings and being monitored by biologists from those agencies, but gaps remained. We have placed cameras at locations that were lacking monitoring and were agreed among the group to be high priority. Camera placement at crossings along I-5 is planned in 2023.

In August 2022 we deployed fifteen trail cameras among nine different culverts/crossings on highway 58 (Figure 31). These cameras were deployed with the objective of increasing the wildlife monitoring effort already in place by Caltrans and CDFW. Through this collaborative effort, a total of nineteen different crossings are currently being monitored within our study area along highway 58. Recent rains and snow have hampered camera servicing somewhat recently, but camera data up to mid December has shown that many species have used the crossings but no mountain lions have done so (Table 6). Humans and domestic animals including dogs, cats, and cattle have also been seen on many of the cameras.

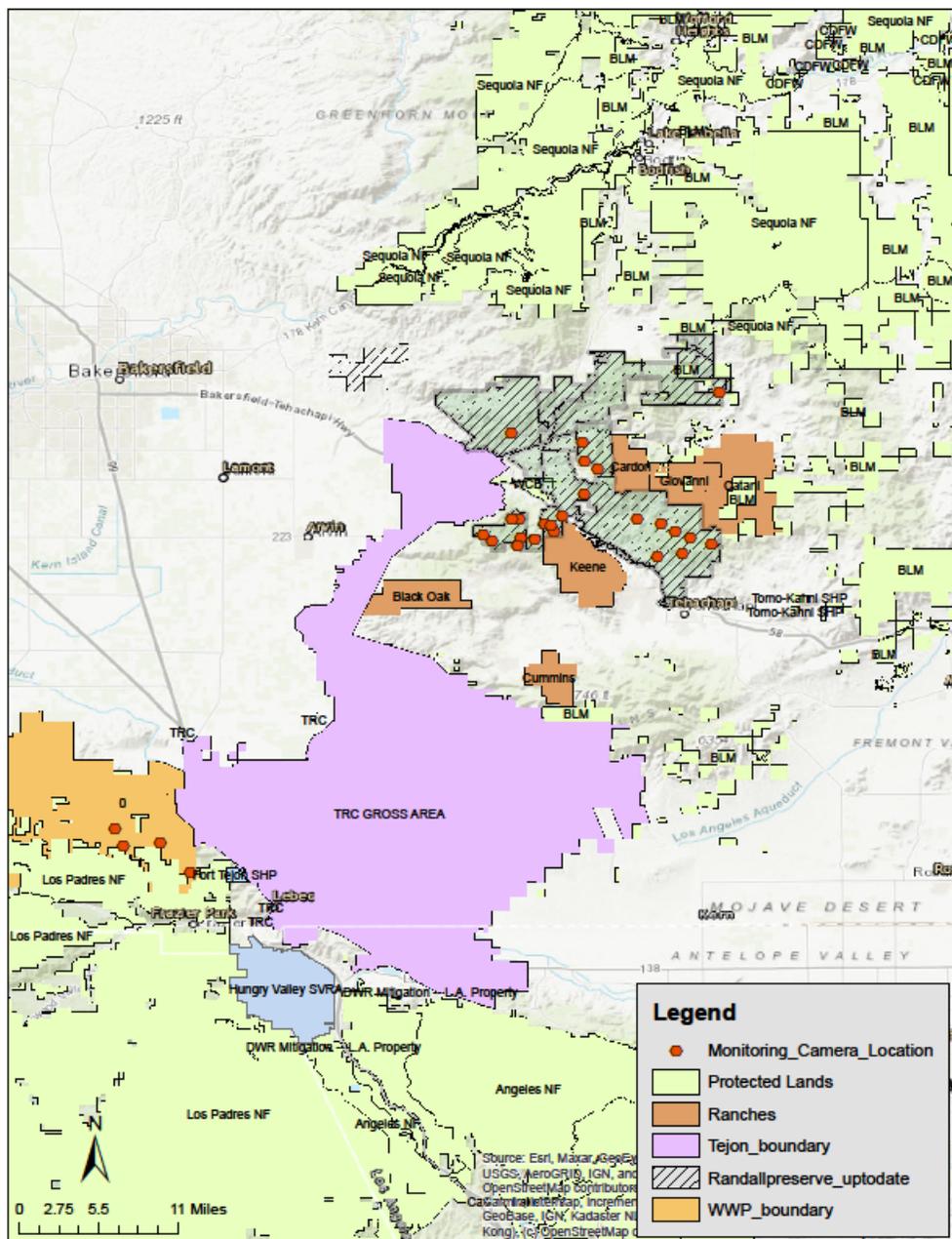
Table 6. SR58 Crossing camera data

Site	Total Animal Photos	Service Date	Lion	Bobcat	Coyote	Gray Fox	Kit Fox	Bear	Deer	Raccoon	Striped Skunk	Badger	Rabbit	Rodent
82.7 R1	0	9/7/2022	no	no	no	no	no	no	no	no	no	no	no	no
82.7 R2	306	9/7/2022	no	yes	yes	yes	no	yes	yes	yes	no	no	no	yes
82.7 R3	57	9/7/2022	no	no	yes	no	no	yes	yes	yes	no	no	no	no
84.75 R	3	9/7/2022	no	no	no	no	no	no	no	no	no	no	no	yes
84.75 L	190	9/7/2022	no	yes	yes	no	no	yes	yes	yes	no	no	no	no
86 R	0	9/7/2022	no	no	no	no	no	no	no	no	no	no	no	no
88.44 R	83	9/7/2022	no	yes	yes	no	no	yes	no	yes	no	no	no	no
89.02 L	45	9/7/2022	no	no	no	no	no	no	yes	yes	no	no	no	yes
99.81 L	28	9/7/2022	no	no	yes	no	no	no	no	no	no	yes	no	yes
99.81 R	18	9/7/2022	no	yes	no	no	no	no	no	no	no	yes	no	yes
100.5 L	0	9/7/2022	no	no	no	no	no	no	no	no	no	no	no	no
100.5 R	10	9/7/2022	no	yes	yes	no	no	no	no	no	no	no	yes	no
103.5 L	87	9/7/2022	no	yes	yes	no	no	no	yes	no	no	no	yes	no
105.75 R	180	9/7/2022	no	yes	yes	no	yes	no	no	no	no	no	yes	no
105.75 R	120	10/7/2022	no	yes	no	no	no	no	no	no	no	no	yes	no
103.5 L	47	10/7/2022	no	yes	no	no	no	no	yes	no	no	no	yes	no
100.5 R	29	10/7/2022	no	yes	yes	no	no	no	no	no	no	no	yes	no
99.81 R	87	10/7/2022	no	no	yes	no	no	yes	yes	no	no	no	no	no
99.81 L	70	10/7/2022	no	no	yes	no	no	yes	yes	no	no	no	no	no
86 R	0	9/7/2022	no	no	no	no	no	no	no	no	no	no	no	no
84.75 L	76	10/7/2022	no	yes	no	yes	no	no	yes	no	no	yes	no	yes
84.75 R	82	10/7/2022	no	yes	no	yes	no	no	no	no	no	no	no	yes
100.5 L	0	9/7/2022	no	no	no	no	no	no	no	no	no	no	no	no
89.02 L	33	10/7/2022	no	no	yes	no	no	no	yes	yes	no	no	no	no
88.44 R	213	10/7/2022	no	yes	yes	yes	no	yes	no	yes	no	no	no	no
82.7 R3	213	10/7/2022	no	no	yes	yes	no	yes	yes	yes	no	no	no	no
82.7 R1	183	10/7/2022	no	no	no	yes	no	no	yes	yes	no	no	no	yes
82.7 R2	86	10/7/2022	no	yes	no	yes	no	yes	no	yes	yes	no	no	yes
82.7 R1	475	11/13/2022	no	no	no	yes	no	no	yes	yes	no	no	no	yes
82.7 R2	167	11/13/2022	no	yes	no	yes	no	no	no	yes	no	no	yes	no
82.7 R3	59	11/13/2022	no	yes	yes	yes	no	no	yes	yes	no	no	no	no
84.75 L	25	11/13/2022	no	yes	yes	no	no	yes	no	yes	no	no	no	no
84.75 R	55	11/13/2022	no	yes	yes	yes	no	yes	no	yes	no	no	no	yes
88.44 R	17	11/13/2022	no	no	no	no	no	no	no	yes	no	no	no	no
89.02 L	79	11/13/2022	no	yes	yes	no	no	no	no	yes	no	no	yes	yes
99.81 L	96	11/13/2022	no	yes	yes	no	no	no	yes	no	no	no	no	yes
99.81 R	96	11/13/2022	no	yes	no	no	no	no	no	no	no	no	no	yes
100.5 R	110	11/13/2022	no	yes	yes	no	no	no	no	no	no	no	yes	yes
100.5 L	87	11/13/2022	no	yes	yes	yes	no	no	no	no	no	no	yes	no
103.45 L	39	11/13/2022	no	yes	yes	no	no	no	yes	no	no	no	yes	no
105.75 R	174	11/13/2022	no	yes	yes	no	no	no	no	yes	no	no	yes	no
100.5 L	115	12/14/2022	no	yes	yes	yes	no	no	no	no	no	no	yes	no
100.5 R	2	12/14/2022	no	yes	no	no	no	no	no	no	no	no	no	no
103.45 L	41	12/14/2022	no	yes	yes	yes	no	no	yes	no	no	no	yes	no
105.75 R	0	12/14/2022	no	no	no	no	no	no	no	no	no	no	no	no
82.7 R1	404	12/14/2022	no	yes	no	yes	no	yes	yes	yes	no	no	yes	yes
82.7 R2	24	12/14/2022	no	yes	no	no	no	no	no	no	no	no	yes	no
82.7 R3	79	12/14/2022	no	yes	yes	no	no	no	no	yes	no	no	no	yes
84.75 L	171	12/14/2022	no	yes	no	yes	no	no	yes	yes	yes	no	no	yes
84.75 R	54	12/14/2022	no	yes	yes	no	no	yes	yes	yes	no	no	no	no
86 L	219	12/14/2022	no	no	yes	no	no	no	no	no	no	no	no	no
88.44 R	99	12/14/2022	no	yes	yes	no	no	no	no	no	no	no	yes	no
89.2 L	36	12/14/2022	no	yes	no	yes	no	no	no	no	no	no	yes	no
99.81 L	25	12/14/2022	no	yes	no	no	no	no	no	yes	no	no	no	no
99.81 R	41	12/14/2022	no	yes	yes	no	no	yes	no	no	no	no	no	no

In addition to the wildlife monitoring effort along SR58, we have deployed up to twenty trail cameras throughout the habitat in our study area (The Nature Conservancy Randall Preserve, Wind Wolves

Preserve, National Forest Lands, and Hungry Valley State Park) to help us identify mountain lion activity in proximity to the highways (Figure 32). From August 2022 to December 2022, we have recorded fifty-five independent mountain lion detections and fifty-six total individuals (Figure 33, 34).

Figure 32. Monitoring camera locations in the Tehachapi Range habitat



Within TNC land, much of the camera effort was focused on the Loop Ranch (14 mountain lion detections) and Bear Mountain Ranch (11 mountain lion detections) on the North and South side of the SR58, respectively. Moreover, two mountain lions were successfully captured and fitted with GPS collars on Bear Mountain Ranch (see Task 2 below; Figures 33,34).

Along highway I-5, camera effort was focused within Hungry Valley State Park (9 mountain lion detections) and Wind Wolves Preserve (11 mountain lion detections). One mountain lion was successfully captured and fitted with GPS collar on Wind Wolves Preserve during this time.

There were other multiple detections throughout the study area including a family group of three mountain lions on 1/12/2023 near highway 1-5.

Figure 33. Mountain lion camera detections in the Tehachapi study area

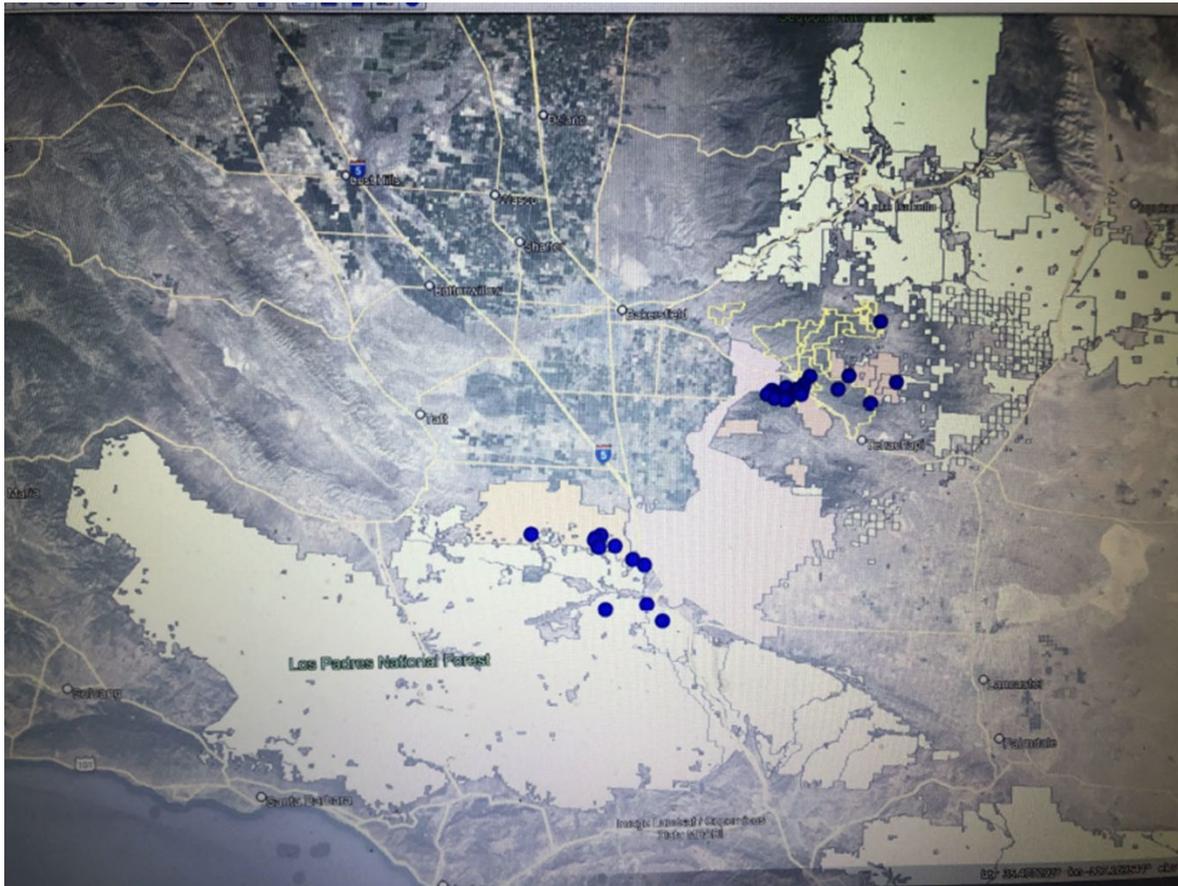
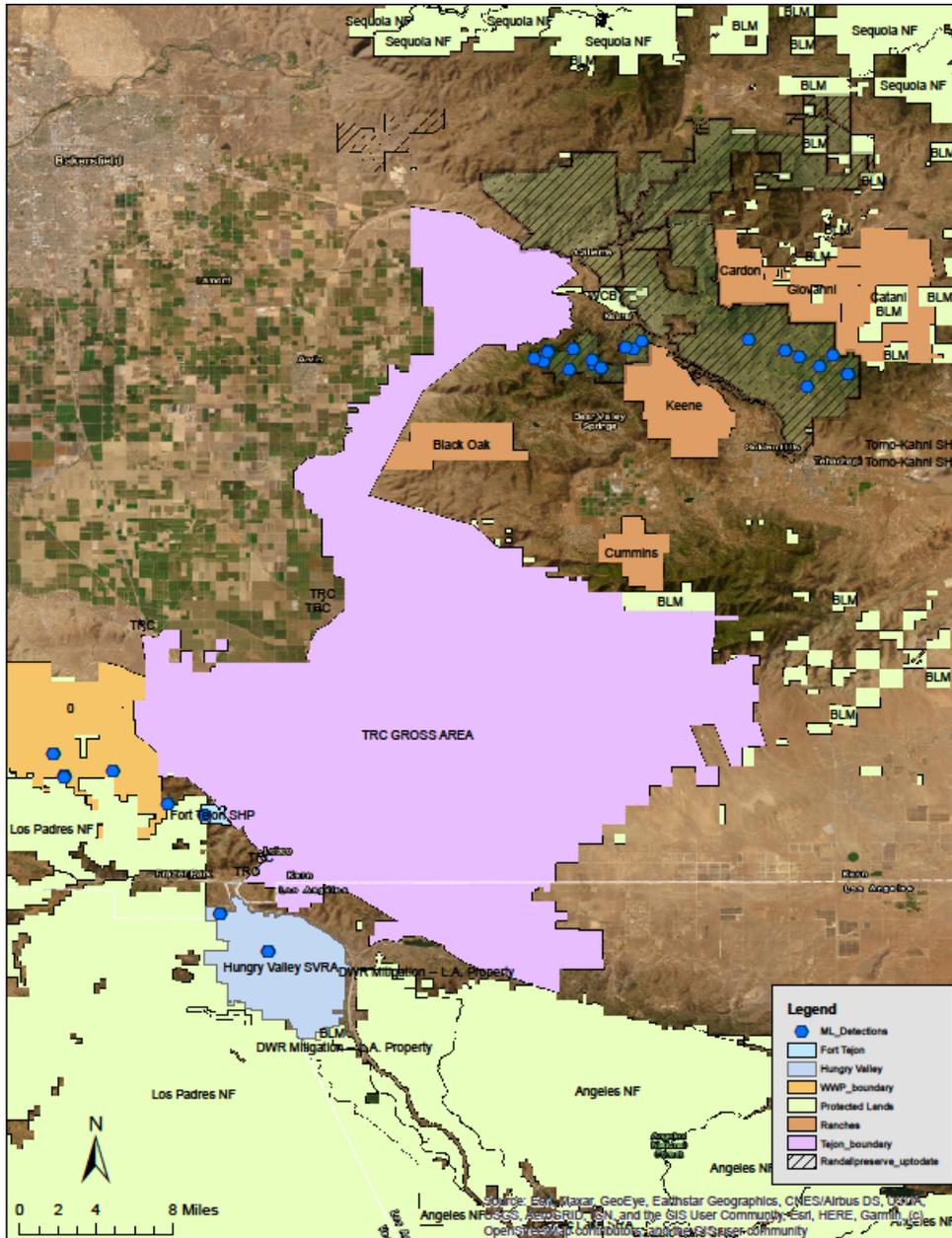


Figure 34. Camera mountain lion detections with conserved lands named



Task 2.

Baiting for capture of mountain lions had to be limited up until late November in much of the study area due to bear disturbance of bait sites. However, some baiting was initiated in November west of I-5 on the Wind Wolves Preserve where bears were not as numerous as in the areas around SR58. Beginning in late November we initiated efforts to capture mountain lions with hounds as well. During an approximately two week period in late November three mountain lions were captured (Tables 7a,b), one male (now designated M326) west of I-5 via cage trap, and one male (M325) and one female (F327) south of SR 58. Biological samples were taken for genetics, disease, and other studies at the time of

captures. Activities are ongoing to attempt additional captures, especially around I-5 and north of SR 58.

Table 7a. Statistics on mountain lions captured during reporting period in

UCD ID	Capture Date	Method	Sex	Age(mo.)	Ear Tag	Tattoo	Status
M325	11/22/22	Hounds	M	72.0	325 orange (left)	325 (right)	Active
M326	11/25/22	Cage	M	66.0	326 orange (left)	326 (right)	Active
F327	11/26/22	Hounds	F	45.0	327 orange (right)	327 (left)	Active

Table 7b. Locations of captures and animal disposition/Transmitter frequencies

UCD ID	General loations	County	UTM Zone	UTM Easting	UTM Northing	Injuries	Mort date	Mort cause	Transmitter frequency
M325	Bear Mountain	Kern	11	351665.35	3898735.28	None	n/a	n/a	160.120
M326	Black Buck Canyon	Kern	11	315930.64	3863490.33	None	n/a	n/a	160.160
F327	Bear Mountain	Kern	11	349875.54	3898467.49	None	n/a	n/a	160.080

All 3 mountain lions were mature adults and appear to be maintaining territories that are similar in size to those that we have documented elsewhere in our studies. None of the 3 have crossed either of the highways that we are most focused on (I-5 and SR58) to date. They, especially M325 and F327, have however crossed secondary roads periodically (Figures 35, 36, 37)

Figure 35. Mountain lion GPS collar data points in the Tehachapi Range. M326 (red), M325 (blue) and F327 (pink).

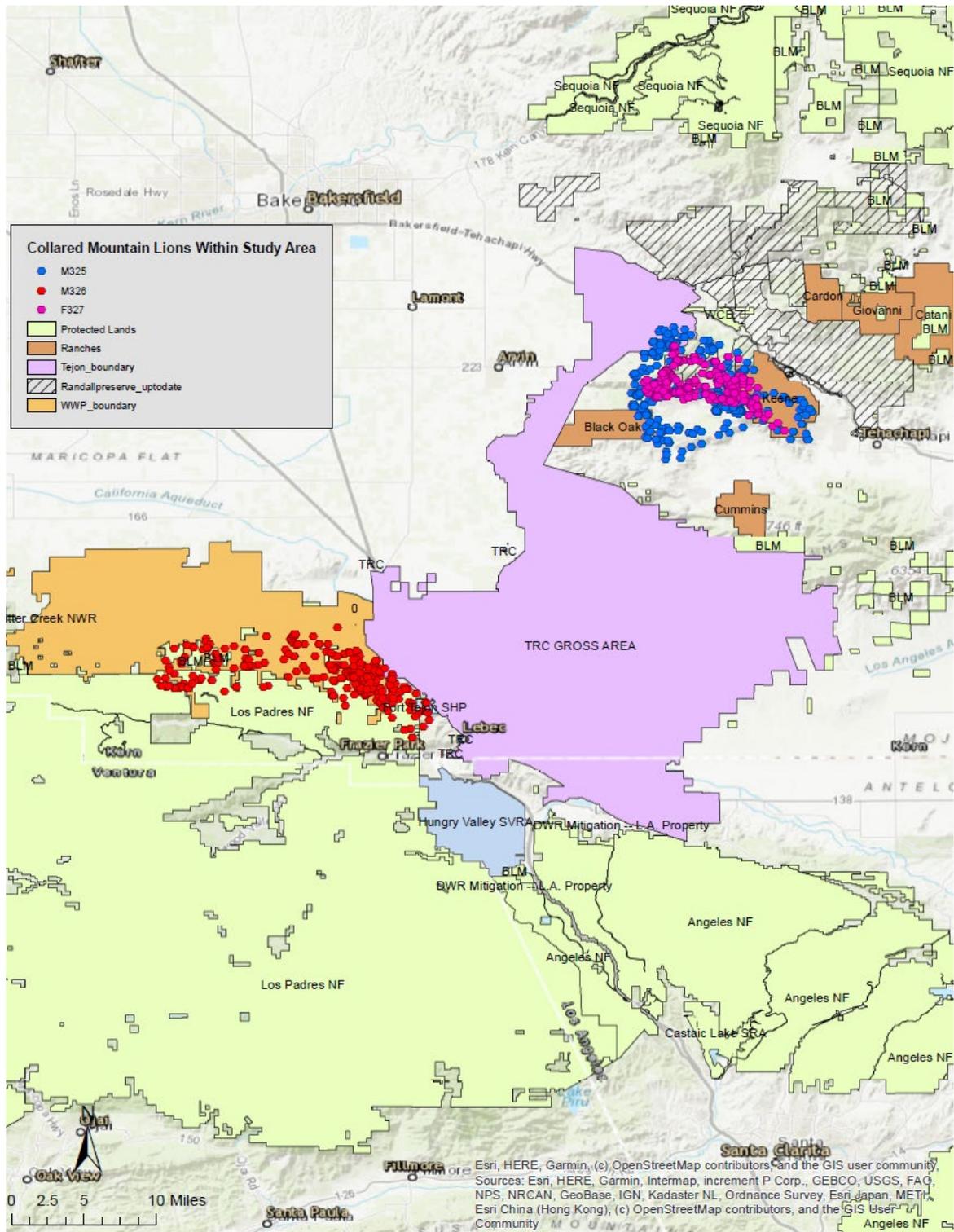


Figure 36. GPS data points of M325 (blue) and F327 (pink). Both animals commonly cross local paved roads in the Bear Valley Springs area, but have not crossed Hwys 223 or 58. Datapoints are 3 hours apart, lines between points do not represent actual paths of travel.

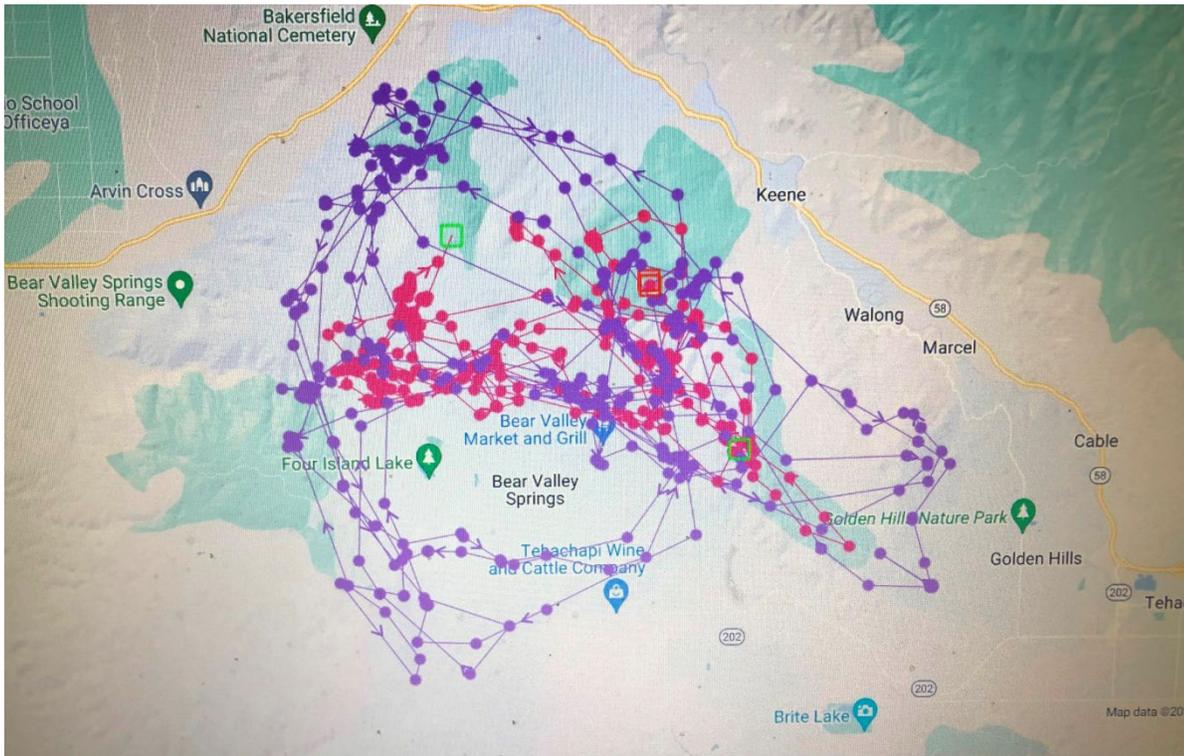
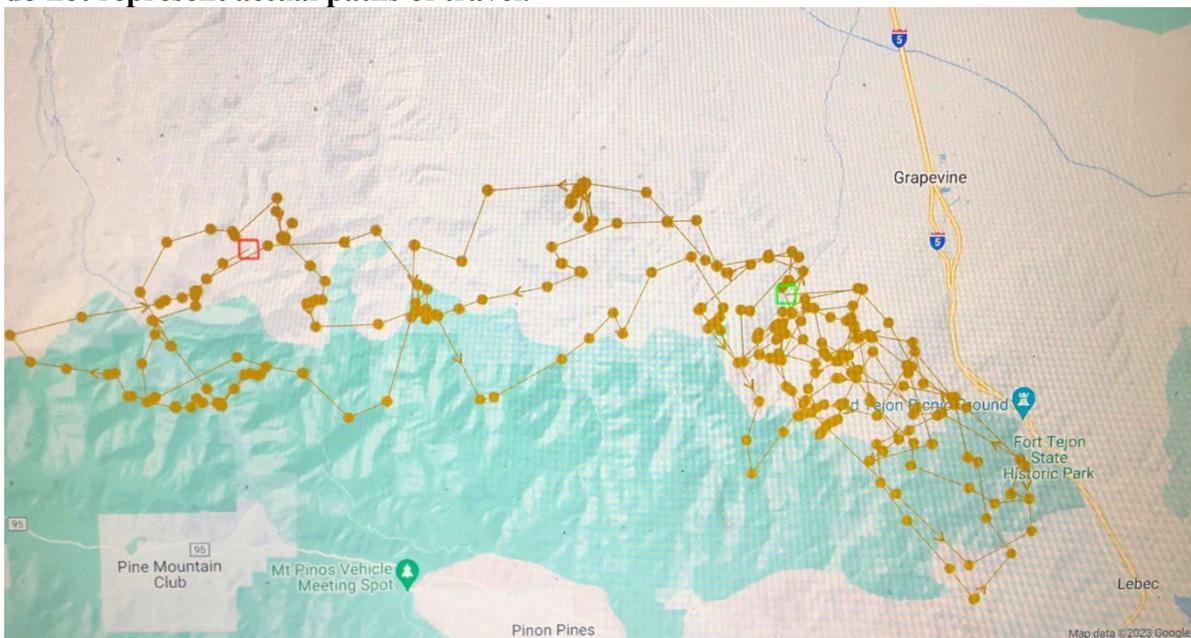


Figure 37. M326 GPS data points (Brown) in the region west of I-5. He has approached the Freeway somewhat closely but has not crossed. Datapoints are 3 hours apart, lines between points do not represent actual paths of travel.

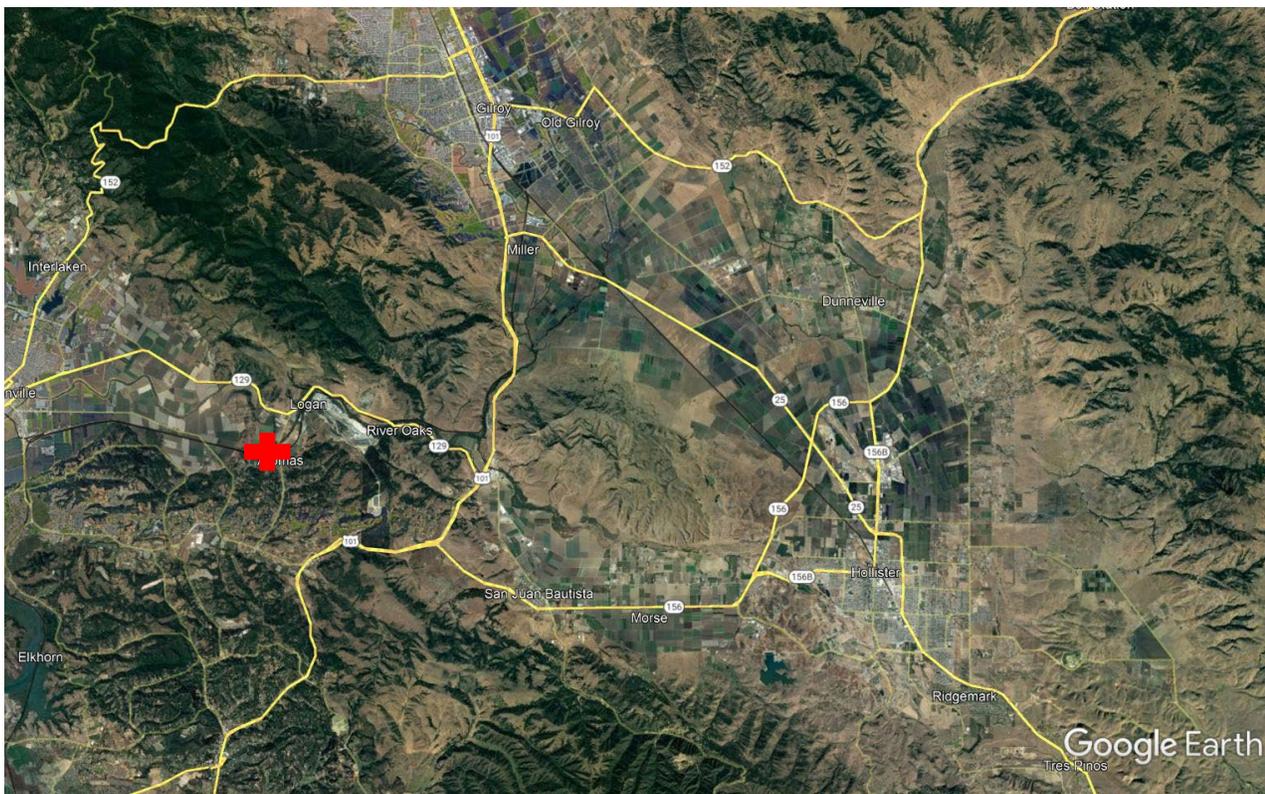


Northern Gabilans and Pacheco Pass Study

In August 2022, we initiated our studies in the Gabilan Range and Pacheco Pass areas with funding from WCN and the Santa Clara Valley Habitat Agency. As in the Tehachapi study area, the goals are highly focused on connectivity across major highways in the region, especially US101 and SR152 through the Pacheco Pass area (Figures 38,39,40). As noted previously in the report, this nexus of the Santa Cruz, northern Gabilans, the Diablo Range, and the coastal ranges to the south is a key area to understand in relation to connectivity for mountain lions and other wildlife, especially in advance of any high speed rail construction that will likely further limit connectivity and gene flow for mountain lions and other wildlife in this area.

Mountain lion GPS collar and genetic samples can help document the current level of gene flow in relation to major roadways with existing crossing structures. Though mountain lions would be targeted, this can be done opportunistically with multiple species. CDFW's Wildlife Genetics Lab has committed to storing and analyzing all mountain lion genetic samples and would take high quality genetic samples (e.g., tissue) from other species (Mike Buchalski – CDFW Wildlife Geneticist; michael.buchalski@wildlife.ca.gov). If connectivity was then improved, additional genetic samples could be used to monitor changes in gene flow in response to mitigation measures.. Thus, understanding current connectivity and gene flow can help mitigate impacts of future transportation infrastructure on viability of local wildlife populations.

Figure 38. Highway 101 near Rocks Ranch (red cross), and SR152 through Pacheco Pass are potential barriers for connectivity and gene flow of mountain lions in this portion of California.



Tasks and Approach:

Task 1. Determine if and where crossing structures might be needed by capturing and deploying GPS radio-collars on mountain lions (Zeller et al. 2017):

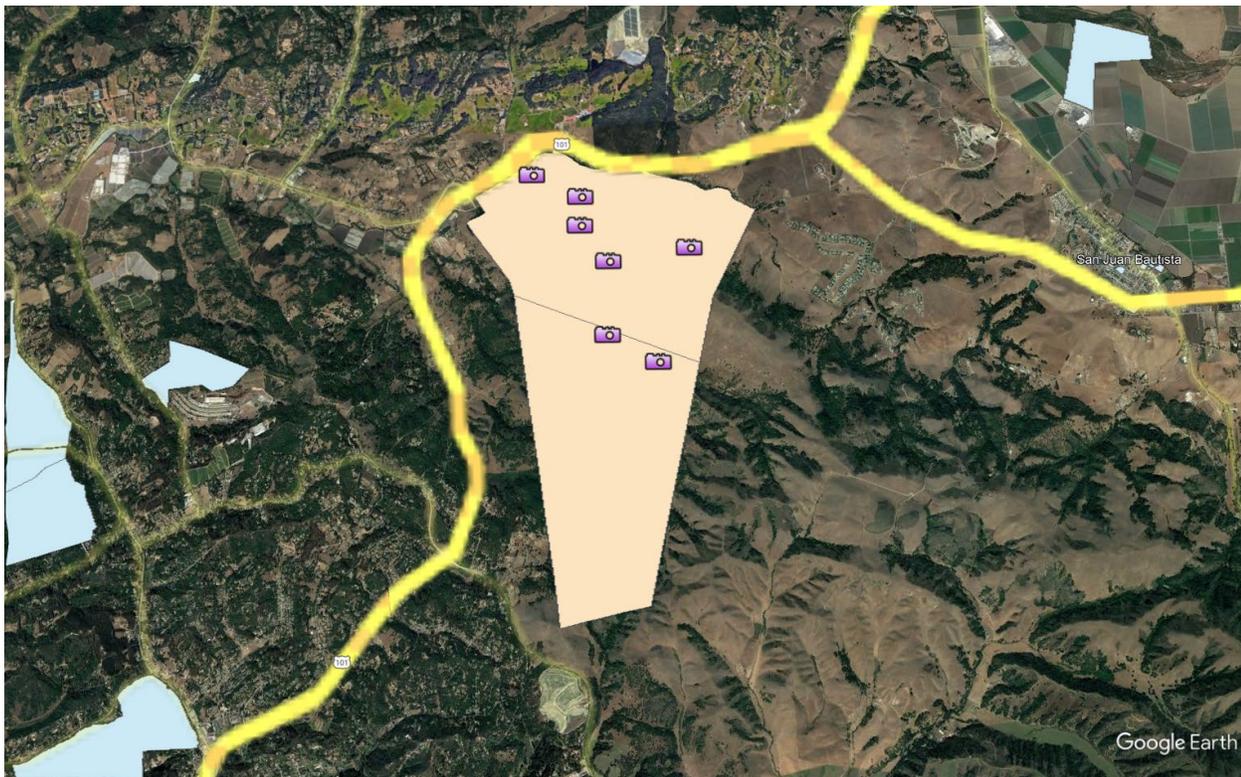
Progress made thus far on the Gabilan Mountains Lion Connectivity project has been primarily in planning and preparation, including coordinating access to additional properties, filling staff positions, procuring equipment and supplies (including securing locations for bait freezers and placing them), and securing permit approvals from regulatory agencies. Some road killed deer bait carcasses have been secured and contacts have been made with Caltrans and other various agencies in the area that are responsible for pickup and disposition of roadkill. It is our hope to build up our bait supply relatively quickly so as to be able to bait for capture during the colder weather.

In addition to the 2,600 acres of the Rocks Ranch and adjacent property owned/overseen by our partners at the Land Trust of Santa Cruz County (LTSCC), the WHC has successfully secured access to the southwest Nature Area of the Hollister Hills State Vehicular Recreation Area (SVCRA), and an 11,000-acre adjoining ranch owned by the Gabilan Cattle Company. This property is located in Monterey and San Benito Counties and is approximately 18-km southwest of Rocks Ranch and HWY 101. The property was initially purchased in 1929 by Rollin Reeves and was entered into a non-developmental conservation easement with The Nature Conservancy in 2006.

Rather than transport capture equipment and supplies between project sites, as initially planned, the WHC has coordinated to outfit each project area (Gabilans and Pacheco Pass, Tehachapi's, and Santa Ana's) with complete capture kits, including all necessary medical gear and controlled drugs. This has required adjustments to DEA licensing agreements, added hardware installation (drug safes, locking cabinets, and vehicle lock boxes), and additional staff training (Veterinary Assistant Controlled Substance Permits).

Heavy rains in mid-December and severe and persistent storms beginning on December 23, 2022, impaired field access and further delayed efforts to place field cameras and establish bait sites. However, since the rains ceased and landowners cleared us for access after roads dried, we have placed 7 field cameras on the Rocks Ranch property (Figure 41). Cameras will now be placed on the Gabilan Ranch and the southwest nature area of the Hollister Hills State Vehicular Recreation Area in order to determine the best areas for baiting for mountain lion capture.

Figure 41. Field camera locations on Rocks Ranch



Task 2. Understand gene flow of mountain lions and other mammals by opportunistically gathering genetic samples e.g., from captures, roadkill, scat, and hair; (Gustafson et al. 2017):

SCVHA/WHC biologists will respond to any reports of road killed mountain lions and collect pertinent genetic samples and work to coordinate getting opportunistic and purposeful collection (i.e., captures) of genetic samples to the CDFW Wildlife Genetics Lab. Primary genetic analysis of samples collected will be contributed by the California Department of Fish and Wildlife genetics lab. To date no captures or mountain lion mortalities have occurred in the area but non-invasive collection of scat for DNA extraction is likely as scouting of the landscape for mountain lion sign continues.

Task 3. Compile results for and consult on efforts to improve/maintain connectivity given existing and future transportation infrastructure.

As GPS collar data becomes available, WHC biologists will process it and develop resource and step selection function models to understand spatial connectivity (Dellinger et al. 2020b). Genetic samples will be analyzed using a single nucleotide polymorphism (SNP) assay to determine individual ID's and contribute to understanding of genetic connectivity (Buchalski et al. 2022). In addition, this Task includes attending/participating in meetings and consulting with pertinent groups to improve/maintain connectivity in the study area. Lastly, this Task also includes overall project funds management with assistance from administrative support personnel at the UC Davis WHC, and overall reporting.

In both the Gabilan Range and Pacheco Pass areas extensive pre-existing data exists from crossing monitoring cameras (Diamond et al. 2022, J. King/C. Duncan pers comm) but little data exists about mountain lion activities in the habitat on either side of the highways. We hope that our efforts will provide substantially more information to help secure or improve connectivity in those areas.

Discussion

Low annual survival rates of pumas in southern California, combined with restricted genetics and connectivity are a major concern especially for the population in the Santa Ana Mountains which is estimated to number between 16 and 21 adults (Beier et al. 1993, Benson et al. 2019), but evidence is mounting of concerns for the populations elsewhere in southern California as well.

These findings suggest that with projected increases in human population size in the region and the attendant loss of habitat and increased traffic and roads, that cougars in this mountain range are under increasing threat. The population east of I-15, though larger and potentially more robust, also faces the same array of increasing threats.

Even under current conditions of immigration into the Santa Ana's, the likelihood of extirpation of the population over the next 50 years is uncomfortably high (1 in 5 or 6 chance), and if inbreeding depression develops then extirpation in a much shorter time frame (<15 years) becomes much more likely without intervention (Benson et al. 2019). Thus the key to keeping robust populations of mountain lions in southern CA is to assure and improve connectivity, and reduce mortality rates.

In 2022, our team has been involved with extensive communication and collaboration with the three counties in the region, transportation agencies, non-profits such as The Nature Conservancy, and wildlife agencies, and others in an attempt to accomplish these goals. It is also critical that mortality levels be reduced across the region as much as possible. Our work on both depredation prevention and road crossings and fencing are aimed at this goal.

We feel that it is critical that education, investment in proper road crossings and fencing, habitat conservation, prevention of habitat fragmentation, and proper domestic animal husbandry all be employed to improve the potential for puma population persistence in the region, especially the Santa Ana Mountains population. These mitigation measures are under human control, but require increased financial and political support in order to accomplish real reductions in current rates of mortality. In light of the bias toward males in the known-mortality statistics for San Diego County, and the need for greater movement of males across I-15 into the Santa Anas, reducing mortalities of males (especially young males) east of the freeway should be a major priority. We are exploring other potential programs and pathways that might also be utilized to reduce threats to these populations.

In northeastern California collared mountain lion survival is substantially higher than survival of collared lions in southern California, despite some losses of collared lions over time to depredation permits. It is hoped that our work on depredation reduction that is focused in southern California may also benefit this population. Information from this study area on dispersal, female behavior, and prey choice may inform management decisions by CDFW in this area.

Though it has just begun, we feel that our work on connectivity assessment in the Tehachapis and northern Gabilan Range and Pacheco Pass will likely be informative to many levels of conservation action in those areas. Connectivity in both areas is critical to mountain lion persistence not only in those

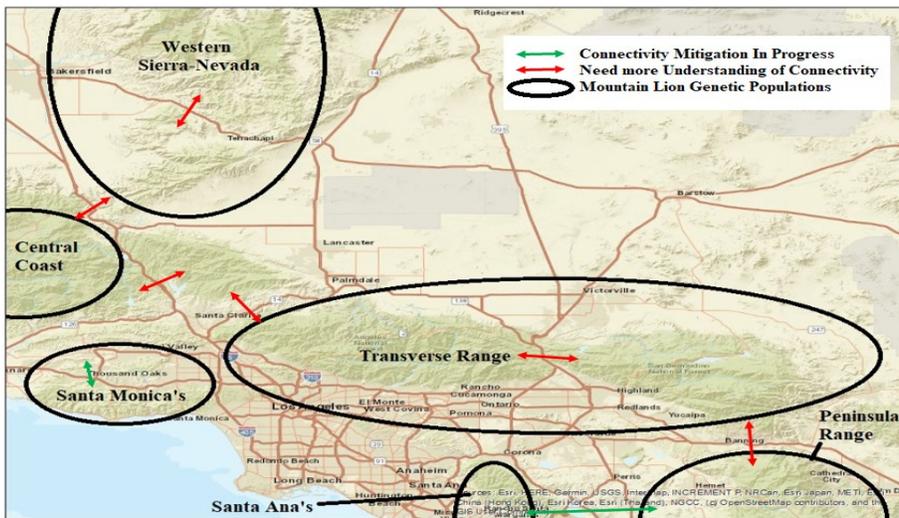
specific areas but in areas distant to those ranges. It is more and more evident from genetic studies and roadway studies that solutions need to be found via the movement of dispersing animals through the likely bottlenecks in exist It is our hope that over the full 3 year period of those studies that the data generated will also spur increased interest in conservation in the general public in that area.

Plans for 2023:

We will continue collaborations with most of the mountain lion researchers in CA and CDFW to conduct further genetic and disease research, connectivity and roads research, and research into deterrence devices to help protect livestock from predation. We are also actively collaborating with TNC, SANDAG, Caltrans, and others on roads improvements.

Mountain lion populations in both the central coast and southern California depend on connectivity through the Tehachapi Mountains to remain genetically viable. For southern California, additional work will still be required in the Transverse Range to ensure viability of mountain lions in the Santa Ana's and Peninsular Range (Figure 45).

Figure 45. Areas of connectivity focus in southern California that are necessary for long term persistence of mountain lion subpopulations there.



We will be continuing all the efforts detailed in this report through 2023.

The UCD team will continue to monitor the 7 mountain lions with active collars in the eastern Peninsular Range and the Santa Anas. We will be continuing collaring efforts in the Santa Anas to test lighter weight collars that could be worn longer term to assist in ongoing monitoring of the population, and at the San Diego Zoo Wild Animal Park. We will test deterrent devices and strategies on those animals as opportunities arise, or will create opportunities by continuing to test at bait sites or along common pathways.

We will complete the study of non-invasive methods of population estimation for the Santa Anas, with potential application to the eastern Peninsular and other subpopulations, and develop a long term

monitoring plan for the ePR and SAM populations.

We will continue our collaboration between IWS and the NE California study and the Audubon Canyon Ranch Living with Lions Project in Sonoma-Napa-Mendocino Counties, as well as the San Diego Zoo to conduct comparisons between the three areas relating to disease and toxin exposure, habitat utilization at fine scales, prey studies, and others.

We will continue and complete the study of mountain lion, prey, and human interactions in Whiting Ranch Wilderness Park.

In the NE CA study area specifically, IWS will also focus on:

- Determining mountain lion kitten survival through non-invasive photo monitoring.
- Continuing to investigate the diet of lions in northeastern California through visiting GPS clusters to identify kill sites.
- Determining mountain lion habitat use, density and range overlap to better understand changes in landscape use patterns if/when wolves establish in the study area.
- Developing further peer reviewed publications via another MS graduate student that IWS has taken on to work on mountain lion biology, specifically dispersal patterns and distances in the study area.

Samples and data from all study areas have, and will continue to be, contributed to statewide analyses of survival, genetics, habitat selection, and connectivity, and be available to CDFW.

Acknowledgments

We thank the following for their financial and in-kind support of project activities in 2020:

- The Wildlife Conservation Network
- San Diego County Association of Governments
- Santa Clara Valley Habitat Agency
- The Nature Conservancy
- Orange County Natural Communities Coalition
- California Department of Fish and Wildlife Local Assistance Grant Program
- The Foothill East Transportation Corridor Agency
- Land Trust of Santa Cruz County
- South Coast UC Research and Extension Station
- Caltrans
- San Diego State University Santa Margarita River Ecological Reserve
- Individual donors, who provide a key element of our project funding

We also thank the following entities for their cooperation and permissions for access to managed conserved lands at various times during the study period, without which the study would not have been possible:

- California Dept. of Fish and Wildlife
- California State Parks
- US Forest Service
- US Bureau of Land Management
- US Fish and Wildlife Service

- US Navy/Marines Camp Pendleton
- Orange, San Diego, and Riverside Counties and County Parks Departments
- The Irvine Ranch Conservancy
- City of San Diego Water District, Parks, and Environmental Services Departments
- Caltrans
- Foothill East Transportation Corridor Agency
- The Nature Conservancy
- San Diego State University
- Western Riverside County Conservation Authority
- Fallbrook Water District and others
- The Wildlands Conservancy
- The Conservation Fund
- Rivers and Lands Conservancy
- Rancho Mission Viejo
- Wind Wolves Preserve
- Gabilan Ranch
- Land Trust of Santa Cruz County
- Santa Clara Valley Habitat Agency
- Private landowners

We appreciate all those persons who have participated in project field or lab activities, data analyses and interpretation, scientific collaboration, or consultation with the project in 2022. We are especially appreciative of our field personnel - Jamie Bourdon, Armand Avery, Lina Vu, Juan Gonzolez, John Randolph, Juan Gonzalez, Alex Dwornik, Calvin Duncan, and other biologists for their dedication and willingness to put in long hours and much hard work to assure the success of the project and enhance puma conservation. We also acknowledge all the lab and other personnel at the SDZWA, and the UC Southcoast Research and Extension Station who have been great help and collaborators, and all of our key scientific collaborators from CDFW and various organizations and institutions that are named in this report (and others) who are too numerous to name but without whom none of the data and samples from the mountain lions would mean much. Finally, thanks to those others at the WHC and IWS who support the work administratively – Matt Blake, Lori Faherty, Leah Brewster, Jenn Larr, and others. And special appreciation to Julie King and Neal Sharma for their support in getting the new projects off the ground.

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2022 Reports and Publications

Vickers, W., J. Dellinger, J. Manning. Estimation of the Population of Mountain Lions in the Santa Ana Mountains and Comparison of Techniques for Population Estimation and DNA Collection, Wildlife Photo Technology Development, and Development of a Long-term Monitoring Plan and Collaborations for Mountain Lion Populations in Regional NCCP. Final Report. September 2022.

Vickers, W – Interim Quarterly reports to the San Diego County Association of Governments – Research into mountain lion depredation prevention, genetics, disease, and photo recognition software development. January, April, July, Oct. 2022

Vickers, W. and Dellinger, J.A. – Interim quarterly reports to the CDFW Local Assistance Grant Program. Project Titled: Use of fecal DNA for genetic evaluation and population size estimation of the mountain lion population in the Santa Ana Mountains, and comparison of results from those of other non-invasive sampling techniques. October, 2022; January, 2023

Gustafson, K.D, R.B. Gagne, M.R. Buchalski, T.W. Vickers, S.P.D. Riley, J.A. Sikich, J.L. Rudd, J.A. Dellinger, M.E.F. LaCava, H.B. Ernest. Puma connectivity could restore genomic diversity to at-risk coastal populations in California. 2022. *Evolutionary Applications*. DOI: 10.1111/eva.13341

Petch, R.; R. Gagne; E. Chiu; C. Mankowski; J. Rudd; M. Roelke-Parker; T. W. Vickers; K. A. Logan; M. Alldredge; D. Clifford; M. Cunningham; D. Onorato; S. Vandewoude. Feline leukemia virus frequently spills over from domestic cats to North American pumas. Accepted October 2022. *Journal of Virology*.

Shilling, F., D. Waetjen, T. Longcore, W. Vickers, S. McDowell, A. Oke, A. Bass, C. Stevens. March, 2022. Improving Light and Soundscapes for Wildlife Use of Highway Crossing Structures. A report to the University of California Institute for Transportation Studies. DOI:10.7922/G2V1233R

Collins, A., T. Winston Vickers, Fraser M. Shilling. Behavioral responses to anthropogenic noise at highways vary across temporal scales. *Frontiers in Ecology and Evolution*. 30 November 2022
Sec. Conservation and Restoration Ecology. Volume 10 – 2022.
<https://doi.org/10.3389/fevo.2022.891595>. This article is part of the Research Topic Ecological Impacts of Transportation Networks at Large Extents

Benson, J.F., K. D. Dougherty, P. Beier, W. M. Boyce, B. Cristescu, D. J. Gammons, D. K. Garcelon, J. M. Higley, Q. E. Martins, A. C. Nisi, S. P.D. Riley, J. A. Sikich, T. R. Stephenson, T. W. Vickers, G. M. Wengert, C. C. Wilmers, H. U. Wittmer, J. A. Dellinger. The Ecology of Human-Caused Mortality for a Protected Large Carnivore. Accepted with revisions. December 2022. *Proceedings of the National Academy of Sciences*.

Dellinger, J. A., Ana. F. Basto, T. Winston Vickers, Christopher C. Wilmers, Jeffrey A. Sikich, Seth P.D. Riley, Daniel Gammons, Quinton E. Martins, Heiko U. Wittmer, David K. Garcelon, Maximilian L. Allen, Bogden Cristescu, Deana L. Clifford. Evaluation of the effects of multiple capture methods and immobilization drugs on mountain lion welfare. Submitted January 2023 to *The Wildlife Society Bulletin*

Sanchez, J. and C. de la Rosa. Passing Shadows. The Quest to Save California's Mountain Lions. The San Diego Zoo Wildlife Alliance Journal. Jan-Feb 2023

2022 Abstracts / Posters

Rafa Barrientos, et al. Presentation to the 2022 Conference of the Infrastructure and Ecology Network Europe. Human footprint and mountain lion territory use in human-dominated landscapes

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Gustafson KD, Vickers TW, Boyce WM, Ernest HB. 2017 A single migrant enhances the genetic diversity of an inbred puma population. *R. Soc. open sci.* 4: 170115. <http://dx.doi.org/10.1098/rsos.170115>

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Dellinger, J.A., B. Cristescu, J. Ewanyk, D.J. Gammons, D. Garcelon, P. Johnston, Q. Martins, C. Thompson, T.W. Vickers, C.C. Wilmers, H.U. Wittmer, and S.G. Torres. 2020b. Using mountain lion habitat selection to inform management. *Journal of Wildlife Management*.

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Diamond, TD, A Sandoval, NP Sharma, ME Vernon, PD Cowan, AP Clevenger, and SC Lockwood. 2022. Enhancing ecological connectivity and safe passage for wildlife on highways between the southern Santa Cruz Mountains, Gabilan Range, and Diablo Range in California. *Pathways for Wildlife and Peninsula Open Space Trust*, Palo Alto, CA.