LONG-TERM MONITORING PLAN FOR THE SAN DIEGO COUNTY MOUNTAIN LION POPULATION

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Table of Contents

Executive summary 3

1. Introduction 5

1.1. Description and ecological role. 5

1.2. Taxonomy and phylogeny. 5

1.3. Conservation status 6

1.4. Conservation threats 7

1.5. California mountain lion population monitoring. 8

2. Monitoring techniques for mountain lion populations with a robust scientific foundation. 10

2.1. Camera monitoring. 10

2.2. Hair sampling. 11

2.3. Scat sampling. 12

2.4. Capture and radio-collaring mountain lions. 12

2.5. Opportunistic data collection. 14

2.6. Environmental DNA (eDNA). 14

2.7. Aerial surveys using thermal imagery. 14

3. Techniques recommended for the long-term monitoring of the southern California mountain lion population. 15

3.1. Long-term fixed-location camera trap database. 16

3.2. Capture and radio-collaring mountain lions and spatial-mark -resight models. 19

3.3. Time-to-event and space-to-event models. 19

3.4. DNA analysis: hair sampling 20

3.6. Scat DNA 21

3.7. Facial recognition in camera trap surveys 21

4. Time-frame 22

5. Budget 23

6. Literature cited 24

Executive summary

This document seeks to provide the San Diego Association of Governments (SANDAG) with the UC Davis Wildlife Health Center recommendations on scientific techniques for long-term monitoring of mountain lions in southern California that can be applied under the scope of work of the San Diego Management and Monitoring Program (https://sdmmp.com/metrics/).

For each technique, we have considered the following factors:

1. Applicability to solitary carnivores and specifically to mountain lions, based on relevant past efforts using the technique,

2. Feasibility of employing the technique to obtain meaningful and repeatable results,

3. Quality of data and robustness of results associated with the technique, and

4. Costs and intervals associated with employing the method through time.

Key Findings:

1. From all the techniques discussed in the Literature Review (Task 6, University of California – Davis Agreement A37682 Amendment #2 SANDAG Contract #5005298 Amendment #2 (S890571) as well as in the Southern California Mountain Lion Workshop (October 11th, 2023) up to seven techniques, employed independently or in combination, may serve to generate the required data to inform long term monitoring of the mountain lion population.
2. The two main recommended techniques that can provide the most data (e.g., habitat use, reproduction, recruitment, survival, genetics, health/disease exposure) are: a) the establishment and maintenance of a long-term camera trap database at fixed locations and b) capture, radio-collaring, and telemetry.
3. The capture and radio-collaring of mountain lions provide the highest quality of certain data (e.g., genetics, health/disease exposure, survival-mortality) and it can also be included within the camera trap analysis to estimate population densities through spatial-mark-resight models.
4. Other non-invasive techniques recommended for long-term monitoring include: non-invasive DNA sampling (e.g., hair and scat) and camera-trapping density models. The use of scat DNA via scat detection dogs is recommended at 5-8-year intervals to serve in the estimation of population density. Randomly placed camera-traps can potentially also be used for population density estimation utilizing models that are currently under investigation such as time-to-event and/or space-to-event models.
5. There is potential to include newer techniques in capture-recapture models for population estimation as they emerge, such as using other sources of DNA from hair shafts, since they are viable in the environment for a longer period of time than DNA in hair follicles, or other

capture-recapture techniques based on facial recognition if that becomes more accurate with software advances.

6. As the genetic health of the southern California mountain lion population is severely compromised, we also recommend DNA collection effort via tissue and blood, since isolation of DNA from these types of samples is most reliable. This DNA collection will provide genetic diversity metrics (e.g., allelic richness, heterozygosity, internal relatedness) and will contribute to estimates of population density. Lower quality DNA from hair and scat may also yield this type of information with advances in genetic analysis.

1. Introduction

1.1. Description and ecological role.

Mountain lions (*Puma concolor*) are the largest non-pantherine felid. They are uniformly colored without body markings, typically light to dark tawny-brown with creamy-white underparts. The tail tip and the ear backs are dark brown to black, and the white muzzle is bordered by black (Hunter, 2015).

The mountain lion is the top carnivore in southern California and is important in maintaining the biodiversity and integrity of natural communities. Mountain lions are a key indicator of preserve system connectivity. They have very large territories and juveniles disperse long distances (Beier 1995; Zeller et al. 2017; Dellinger et al. 2020). They may act as an umbrella species since protecting land and improving connectivity for mountain lions could also benefit other species, especially those that are wide-roaming (Zeller et al. 2017). Mountain lions also influence food webs and the flow of energy through natural ecosystems. This species can change community composition and structure by affecting prey population dynamics, which impacts herbivory on plants and competition between herbivores (Ripple and Beschta 2006). In other cases, there may be short-term impacts on prey populations but no change in long-term dynamics (Hurley et al. 2011). The mountain lion is also a charismatic species that sparks public interest and fascination. The facts explained above plus their potential to act as an umbrella species, rank mountain lions among felids of the highest conservation priority (Dickman et al. 2015).

1.2. Taxonomy and phylogeny.

The mountain lion is the only species classified in the genus *Puma*. The jaguarondi (*Herpailurus yaguarondi*) is its closest relative. Both species are also related, although more distantly, to the cheetah (*Acinonyx jubatus*) and all these three species together comprise the *Puma* lineage.

Up to six subspecies in three geographic groups (North, Central, and South America) were formerly suggested based on genetic analysis:

- *Puma concolor cougar* (North America)
- *Puma concolor costaricensis* (Central America)
- *Puma concolor capricornensis* (East South America)
- *Puma concolor concolor* (N South America)
- *Puma concolor cabrerae* (central South America)
- *Puma concolor puma* (S South America).

Currently, only two subspecies are recognized: *Puma concolor concolor* (South America) *and Puma concolor cougar* (North and Central America) (Castello, 2020).

The remnant population in Florida, although usually classified as its own subspecies (*Puma concolor coryi),* is an ecologically and geographically distinct population of the North American subspecies *Puma concolor cougar,* since its isolation occurred only in the last 100 years and genetic data indicates its relatedness to other North American populations (Hunter, 2015).

1.3. Conservation status

The mountain lion is a specially protected species in California. The California Wildlife Protection Act of 1990 (Proposition 117) declared the mountain lion a “specially protected mammal under the laws of this state” (Cal. Fish & Game Code § 4800(a)). As a result, hunting of mountain lions is generally prohibited, and there are restrictions on taking, injuring, possessing, transporting, importing, or selling mountain lions (Cal. Fish & Game Code § 4800(b)). However, there are exceptions that allow for the removal or killing of mountain lions if they are perceived to be an imminent threat to public health or safety or pose a threat to the survival of threatened, endangered, candidate, or fully protected sheep species (Cal. Fish & Game Code § 4801).

Furthermore, if a mountain lion damages or destroys livestock or other property, a person may request a permit to “take” the mountain lion (Cal. Fish & Game Code § 4802). The California Department of Fish and Wildlife (CDFW) is responsible for issuing depredation permits, which authorize the removal of mountain lions in such cases. Currently, mountain lions are being evaluated to be listed as threatened under the California Endangered Species Act (CESA). While Proposition 117 already provides protections for mountain lions, listing them under CESA would offer supplementary safeguards and align with the goals of protecting and preserving their habitat and genetic diversity.

Southern California and Central Coast mountain lions would comprise an Evolutionarily Significant Unit (ESA), as is supported by genetic studies (Gustafson et al 2018). An ESA allows for the designation and listing of these populations as endangered under the CESA.

The six recognized subpopulations within the Southern California/Central Coast ESU of mountain lions are as follows:

1. Central Coast North – includes mountain lions in the Santa Cruz Mountains and East Bay.
2. Central Coast Central – represents a genetically distinct mountain lion population in the central coastal area.
3. Central Coast South – includes mountain lions in the Santa Monica Mountains.
4. San Gabriel/San Bernardino - consists of mountain lions in the San Gabriel and San Bernardino Mountains.
5. Santa Ana Mountains – represents a genetically distinct mountain lion population in the Santa Ana Mountains. San Diego County contains a portion of this mountain range, that is west of Interstate 15 and south of the Riverside County line. 6. Eastern Peninsular Range - includes mountain lions in the eastern portion of the Peninsular Range, the majority of which is in San Diego County .

Thus, San Diego County mountain lions exist as part of two of the six subpopulations that have been petitioned for listing under the California Endangered Species Act, and both populations are primarily dependent on gene flow from a third petitioned population, the San Gabriel/San Bernardino population to the north.

1.4. Conservation threats

There are a variety of threats facing mountain lions in southern California. Southern California's human population grew rapidly over the last half century. This rapid human population growth is leading to extensive habitat loss and fragmentation from urban and agricultural development (Vickers et al. 2015). Despite conservation of large blocks of habitat, many mountain lion populations are small and isolated by freeways and surrounded by development (Vickers et al. 2015, 2017; Dellinger et al. 2020). Mountain lions have unusually high mortality rates in southern California, primarily from vehicle strikes and human conflicts (for example, depredation permits) (Vickers et al. 2015). In particular, up to 60% of mortalities in the Santa Ana Mountains were due to vehicle collisions and in the eastern Peninsular Range depredation permits and vehicle collisions were the first and second most common causes of mortality(Vickers et al. 2015). The eastern Peninsular Range population was in the top tier of mortality rates in a statewide mortality analysis assessing all the genetic subpopulations (Benson et al. 2013) and mortalities related to humans were judged to be additive versus compensatory – raising the issue of sustainability of these two populations.

• Climate Vulnerability: Changing climate can increase the frequency, intensity, and duration of droughts and negatively affect lion populations by reducing prey availability (Stoner et al. 2018). Plant productivity in semi-arid regions is correlated with rainfall, and drought limits food availability for prey and causes prey populations to shrink. A reduction in prey availability can lower lion productivity and survival and adversely impact populations (Stoner et al. 2018).

• Human Use: A growing human population results in less habitat for mountain lions free from human disturbance. There are increasing interactions between lions and humans that can result in safety and livestock protection concerns and in the death of the lions from depredation permits (Vickers et al. 2015).

• Connectivity: Habitat loss and fragmentation are causing increasing risk to mountain lion populations in southern California. Mountain lions are constrained or blocked in moving between small, isolated populations, leading to a loss of genetic diversity, which has been reported from a molecular to a phenotypical level in Southern California (Huffmeyer et al. 2022). Loss of connectivity is leading to a potential extinction vortex in the Santa Ana Mountains population and is likely to similarly affect the Eastern Peninsular Ranges population over time (Ernest et al. 2014; Gustafson et al. 2018; Benson et al. 2019; Dellinger et al. 2020).

• Fire: Increasing frequency of large-scale wildfires in shrublands is leading to conversion of shrublands to invasive, nonnative annual grassland, a habitat infrequently used by mountain lions. Fire in linkages and corridors can be impactful as loss of shrub and tree cover can lead to decreased connectivity between habitat patches (Jennings et al. 2016). In urbanized regions affected by wildfires, mountain lions avoid burned areas and increase activities that elevate their risk of negative interactions with humans and conspecifics (Blakey et al. 2022).

• Urbanization: Loss and fragmentation of habitat are negatively impacting lion populations which require very large, unfragmented natural habitats to persist. Lions bordering urbanized and rural residential areas are at risk of death from vehicle collisions and conflicts with humans (Vickers et al. 2015; Dellinger et al. 2020).

1.5. California mountain lion population monitoring.

Population monitoring (e.g., population density, population trend) is a key element of a carnivore management strategy. Because most carnivores are highly mobile, usually secretive, and occur in relatively low densities, they are difficult to study and require greater and longer-term funding for monitoring compared to other taxa. Nonetheless, population monitoring schemes if planned properly, can provide reliable estimates of changes in population size over time and assess the impact of management actions on populations (Clevenger 1998). Therefore, reliable long-term research into trends is crucial in ecology and conservation studies (Sereno-Cadierno et al. 2023).

Several monitoring methods have been developed for large carnivores including telemetry, capture-mark-recapture, harvest data, sign survey among others (Wilson and Delahay 2001, Barea-Azcón et al. 2007). Noninvasive genetic sampling techniques, which use DNA extracted from animal sign such as hair, scat, saliva, urine, or regurgitates (Waits and Paetkau 2005), have become an effective method for studying wildlife populations and are the preferred monitoring methods for some species and populations (e.g., Rudnick et al. 2005, De Barba et al. 2010, Borthakur et al. 2011).

Monitoring mountain lion density and abundance is typically estimated using two approaches: a minimum count of known animals with associated monitoring data (e.g., radio-collar data and age class data) divided by the amount of habitat in a demarcated study area (Lambert et al. 2006, Cooley et al. 2009), or a spatial capture-recapture (including spatial mark-resight) framework using a variety of methods including, but not limited to, DNA from scat (Davidson et al. 2014), skin biopsies (Proffitt et al. 2015), or hair (Alldredge et al. 2019), or collars and/or cameras (Alexander and Gese 2018, Murphy et al. 2019, Loonam et al. 2021).

The minimum count approach has been used for many decades and has the benefit of generating a lot of detailed site-specific data on mountain lion populations (Beausoleil et al. 2013). However, such an approach is very resource-intensive and difficult to replicate. Murphy et al. (2022) conducted a review of techniques and determined approaches employing a spatial capture-recapture framework could estimate mountain lion populations but precision of estimates, primarily due to size of the study areas, was a concern that should be addressed prior to initiating any data collection efforts. In general, there is uncertainty related to how different data collection and analytical methods relate to one another (Beausoleil et al. 2021, Murphy et al. 2022). Additionally, some of the difficulty in comparing results is simply related to differences in how studies define spatial and temporal scales (Cooley et al. 2009, Dellinger et al. 2018) and what segment of the population (e.g., adults and/or subadults) density is being estimated for (Rinehart et al. 2014, Beausoleil et al. 2016). To this point, most efforts to estimate mountain lion density have been conducted once over a short time period (e.g., < 5 years) equal to or less than 1-2 generations as it relates to mountain lions, and/or most efforts occur at a spatial scale that does not allow extrapolation to large habitat areas or subpopulations; Beausoleil et al. 2013, Murphy et 6 al. 2022). Given that processes influencing mountain lion population viability occur over much longer temporal (van de Kerk et al. 2019) and larger spatial scales (Dellinger et al. 2020), there is a need to standardize efforts across a given jurisdiction (e.g., regional, state, or provincial level; Beausoleil et al. 2013) and replicate efforts regularly (e.g., every 5-10 years given mountain lion generation times; Jenks 2011) to understand population trends.

Mountain lions exist throughout San Diego County and have relatively low overall survival rates (Vickers et al. 2015, Benson et al. 2023). As a result, concerns have been raised about the possibility of long-term overall population decline and the need to monitor the population trajectory over time.

Long-term monitoring of this species is imperative to detect in a timely way any changes that this subpopulation may suffer which might put it on the verge of extinction. Different methodologies have been discussed under Task 6 - Literature Review (University of California – Davis Agreement A37682 Amendment #2 SANDAG Contract #5005298 Amendment #2 (S890571). Moreover, monitoring techniques for mountain lions have also been the focus of the Southern California Mountain Lion Workshop organized by the UC Davis Wildlife Health Center on October 11th, 2023.

2. Monitoring techniques for mountain lion populations with a robust scientific foundation from our literature review.

Our literature review explored and discussed three non-invasive techniques for monitoring mountain lion populations:

2.1) camera monitoring;

2.2) hair sampling;

2.3) and scat sampling.

We also discussed the modifications of more invasive approaches (i.e., radio-collaring) to inform long-term population monitoring of mountain lions in southern California. Finally, we discussed the utility of opportunistic data collection (e.g., sampling carcasses of recently deceased animals) and other alternatives such as the use of environmental DNA and aerial surveys using thermal imagery.

2.1. Camera monitoring.

The use of cameras is a widely utilized method for monitoring mountain lion populations. This method allows to gather valuable data on various aspects of mountain lion populations such as population density, distribution, behavior, activity patterns, and interactions with other species. It helps detect changes in habitat use and movement patterns, as well as interactions with prey or other individuals. This information contributes to a comprehensive understanding of mountain lion ecology and can inform conservation and management efforts.

Validation and improvement of camera monitoring methods are necessary to ensure reliable density estimates. Techniques like density modeling and occupancy analysis can be employed to enhance the accuracy of density estimates derived from camera monitoring. Camera monitoring can be paired with other monitoring techniques, such as radio-collar data, to improve inference and provide a more comprehensive understanding of wildlife populations. More recently, space-to-event and time-to-event (STE/TTE) models have attempted to derive local mountain lion densities using large-scale game camera monitoring efforts (Loonam et al.

2021). This method does not rely on individual identification of animals but utilizes information on the space between cameras with detections and time between single cameras with multiple detections.

In general, camera monitoring is a valuable tool in monitoring mountain lion populations, offering insights into their behavior, distribution, and interactions with the environment. When combined with other monitoring approaches, such as radio-collaring or genetic analysis, it provides a more comprehensive understanding of mountain lion populations.

2.2. Hair sampling.

Hair sampling is a population monitoring approach that involves collecting hair left behind by mountain lions. This method typically uses sticky rollers or wire brushes placed in strategic locations such as trails or marked scent stations. As mountain lions pass by these stations, their hair gets caught on the adhesive material.

The collected hair samples are then carefully removed from the sticky rollers or wire brushes and stored for further analysis. The hair samples are typically sent to a genetics lab capable of performing DNA analysis.

Hair sampling allows for genetic analysis of the collected hair samples, which provides valuable information about the population size, genetic diversity, and even individual identification of mountain lions. This method has been shown to be effective in estimating mountain lion densities, as well as assessing population structure and genetics.

However, hair sampling has some limitations and challenges. It is sensitive to site disturbance, meaning that other animal species visiting the sampling site can interfere with the collected hair samples. Regular site maintenance is required to ensure the effectiveness of the sampling equipment.

Additionally, the technique requires access to a genetics lab for DNA analysis, which can add to the overall costs of implementing this method. Coordinating and planning hair sampling efforts can be complex due to the opportunistic nature of collecting samples and the small sample sizes often obtained.

Broadly speaking, hair sampling is a non-invasive and informative method for monitoring mountain lion populations, but further refinement and validation of the technique are necessary for more accurate and reliable results.

2.3. Scat sampling.

Scat sampling is a non-invasive method that involves collecting feces left behind by mountain lions in their natural habitat. Scat samples are collected from various locations, such as trails, marking sites, or areas where mountain lions are known to frequent.

Once collected, the scat samples are preserved for further analysis. DNA analysis allows researchers to extract genetic information, such as identifying individual mountain lions, estimating population size, and assessing genetic diversity.

Scat sampling has been successfully used in Southern California and other regions to estimate mountain lion densities. It provides valuable insights into population dynamics, including changes in population size over time and the identification of individuals within the population. Moreover, scat sampling allows for assessing genetic diversity and understanding the genetic health of the population.

One of the advantages of scat sampling is that it is relatively non-invasive, minimizing disturbance to the animals and their habitat. It is also a cost-effective method compared to other techniques like camera monitoring or long-term collaring. However, scat sampling does require access to a genetics lab capable of performing DNA analysis, which adds to the associated costs.

Overall, scat sampling is a valuable tool for monitoring mountain lion populations, providing information about population size, genetic diversity, and individual identification. It complements other monitoring methods and contributes to a comprehensive understanding of mountain lion populations.

2.4. Capture and radio-collaring mountain lions.

Some research efforts using game cameras have combined them with parallel efforts to capture and mark a subset of the mountain lions in an area to develop density estimates (Rich et al. 2014, Murphy et al. 2019). These efforts can produce statistically robust results but any efforts to capture, mark, and radio-collar mountain lions is inherently costly. Capturing and radio-collaring mountain lions provides a wealth of information about that individual animal and, if enough animals in a given area are captured and radio-collared, then one can gain in-depth insight on multiple aspects of mountain lion ecology (e.g., demographics, genetics, diet, health, etc.). However, capturing and radio-collaring the requisite number of animals in a population to gain such levels of insight then quickly render game camera data less useful given how coarse it is compared to radio-collar and other data sets collected during and after captures. If survival rates are high enough, one approach could be long-term monitoring collars, which are cheaper than standard GPS radio-collars, on a few animals (that allow for individual identification) paired with game camera arrays and regular intervals (e.g., every 1-5 years). Such an approach could provide some more in-depth insight on aspects of demographics at a reduced cost.

There are already radio-collared mountain lions present in some areas within southern California. It is much cheaper to maintain radio-collars in an area compared to deploying radio-collars on new individuals. The telemetry data provided by these collared mountain lions adds precision to density estimates performed by camera-trap data analysis (e.g., spatially mark-resight models -SMR-; Murphy et al. 2019). While using generalized SMR, telemetry data from GPS collars are critical for accurately estimating density and improving parameter estimate precision (Murphy et al. 2019). Further, the isolated nature of the mountain lion populations in southern California, while not ideal from a conservation standpoint, do help ensure assumptions of closed populations during survey periods would not be violated, since population closure is a general assumption for most models used with unmarked or partially marked wildlife populations.

A less intensive variation of this technique is the use of long-term collars. Long-term monitoring collars could be used to maintain a level of understanding about local mountain lion survival and recruitment rates. Long-term collars take 1-2 locations/day and thus can last for five or more years, assuming that the animal lives that long, and are less than half the cost of regular collars when accounting for location fees. This means that recollaring does not have to occur regularly (currently recollaring is something that has to occur every 1-2 years to continue monitoring a given animal) which reduces costs. Further, long-term collars would allow low-intensity monitoring of adult females and attending young. This could take the form of placing a camera at a kill site once/month to check on the number of kittens present which would inform understanding of kitten survival. Having long-term collars on adult animals, including adult females, would also readily allow opportunities to place long-term monitoring collars on subadults just prior to dispersal. In short, if you have the mother radio-collared then you know where the subadults are and one can readily capture them on a kill. This would provide information on recruitment and habitat connectivity to name a few things. Lastly, it is well established that spatial data from radio-collars greatly improves population estimation methods involving non-invasive methods like those mentioned above (Murphy et al. 2019, 2022). Thus, this approach would bolster any of the methods previously mentioned.

2.5. Opportunistic data collection.

Carcasses from roadkill animals would provide an opportunity to opportunistically monitor various aspects of a mountain lion population including population genetics via a tissue sample, diet via stomach contents and stable isotope analyses of whisker samples, and health/disease status via analyzing blood and organ tissue.

2.6. Environmental DNA.

Environmental DNA (eDNA)—DNA shed from an organism in its environment—coupled with quantitative PCR (qPCR) analyses, has become a reliable and extremely sensitive mean for identifying rare species in aquatic systems. Franklin et al. (2019) use this methodology in surveys for Canada lynx, wolverine, and fisher. Samples were collected by means of snow-track testing (snow samples were collected from tracks of Canada lynx, fisher, and wolverine) and from snow collected at camera stations (snow samples were collected from areas where a rare carnivore was photographed). The authors stated that qPCR-based DNA analyses provide more reliable species identifications, reducing misidentification and missed detection errors.

Although this methodology may be applied to mountain lions' habitat presenting snow cover during the winter months, the usefulness in San Diego County would be limited due to the rarity of snow except in the highest elevations. This technique is also labor intensive in that it requires extensive time spent searching for tracks of this animal that is relatively rare on the landscape.

2.7. Aerial surveys using thermal imagery.

Havens and Sharp (1998) discussed the use of thermal imaging technology to provide a method for obtaining complete counts of animals with little risk of bias. The authors conducted a study in southwest Florida using thermal imagery to survey animals. They recorded thermal signatures of deer on video tape during flights along transects. The thermal imagery survey counted 42% more deer compared to standard visual aerial survey methods. The authors successfully located radio-collared Florida panthers using thermal imagery. The detectability of thermal contrast between biological objects and their background was sufficient for species identification. Due to the environmental temperature and habitat characteristics in southern California generally and in San Diego County specifically, obtaining enough thermal contrast to distinguish fauna may be constricted to a very few months out of the year, making this technique more challenging to apply successfully than in other areas.

3. Techniques recommended for the long-term monitoring of the San Diego County mountain lion population from literature review and workshop.

Our literature review and the discussions/round table at the Southern California Mountain Lion Workshop revealed what techniques are the most appropriate to perform long-term monitoring for the population of mountain lions in San Diego County. Since this is an evolving field, this document is focused on monitoring techniques as of November 2023. This is a living document, and our UC Davis mountain lion team will keep collecting new techniques applicable to mountain lions as they emerge from the scientific literature and could be included in the long-term monitoring strategy for San Diego County. Applicability of techniques and strategies will also depend on the factors mentioned above in the Executive Summary (e.g., applicability to southern California mountain lions, feasibility, replicability, data quality, data robustness, cost).

Herein we propose different techniques in order of relevance to our study site and characteristics of the mountain lion population.

3.1. Long-term fixed-location camera trap database.

Camera traps serve as a research tool for the study of the occurrence of elusive carnivores such as mountain lions. Researchers from UC Davis Wildlife Health Center Mountain Lion Project have been using this non-invasive methodology to detect mountain lions in the landscape for +20 years. Our team has secured collaboration with numerous other entities in the region (e.g., Irvine Ranch Conservancy, Orange County Parks, The Nature Conservancy, California State Parks, and potentially USGS and others) that include camera arrays within their wildlife monitoring techniques. These collaborations, when paired with new cameras that the UC Davis team anticipates placing in unmonitored areas can ensure the monitoring of a vast area of suitable habitat for mountain lions. These long-term placed cameras are very valuable in providing data regarding:

- Occurrence data
- Demographic metrics (e.g., kitten production over time)
- Genetic traits (e.g., kinked tails)

The analysis that can be included using this type of long-term placed cameras include but are not limited to:

- Photographic rates

  Photographic rates measured as an index of abundance (i.e., relative abundance index -RAI-) present some challenges to provide accuracy since photographic rates are influenced by the species abundance, movement patterns, camera trap setup, or habitat to name a few (Sollman 2018). Nevertheless, photographic rates should be interpreted as an index of activity, where activity of a species at a site can increase because a higher number of individuals use that site and/or because individuals use that site more frequently (Sollman 2018).

- Presence-only data

  Presence-only data, where there is no information on locations where the species is absent, are common in wildlife studies. Effective models are needed to explore species distribution or species use of habitat using presence-only data and the study design is critical in such models where only presence can be identified (Pearce and Boyce 2006). For modelling presence-only data, four approaches can be used:

  A) Describing the distribution of the presence-only records: This approach involves characterizing the spatial distribution of the species based on the recorded presence locations. It provides a descriptive analysis of the data without explicitly modeling the underlying environmental factors.
  B) Contrasting the distribution of presence records with pseudo-absences: In this approach, random background or pseudo-absence locations are generated to contrast with the presence locations. The environmental attributes of the presence and pseudo-

absence locations are compared to identify the factors associated with the species' presence.

C) Contrasting the distributions of presence records and available sites: Similar to the pseudo-absences approach, this method focuses on contrasting the environmental attributes of the presence locations with the available sites in the study area. The available sites can be selected randomly or based on certain criteria, such as a defined buffer around the presence locations.

D) Modeling abundance when abundance given presence is known: This approach is used when information on species abundance is available, in addition to presence records. Statistical models are developed to estimate the relative abundance or density of the species based on the presence data and other relevant covariates.

The choice of approach depends on the quality of the presence-only data and the research objectives since different approaches may provide different insights into the species distribution and habitat use.

- Occupancy

Occupancy models describe spatial patterns of animal occurrence (Sollmann 2018). Occupancy has been proposed as a proxy for abundance (Noon et al. 2012). In theory, occupancy and abundance share a predictable relationship. As population size increases, the number of sites occupied by members of that population should also increase (until all sites are occupied); likewise, a decrease in population size should lead to a decrease in the number of sites used (Gaston et al. 2000, Royle and Dorazio 2008). This is called an occupancy-abundance relationship, and therefore occupancy can be used as an index of abundance (Clarke et al. 2023).

- Habitat selection

Habitat is a key determinant of the distribution and abundance of wildlife. Based on habitat use and occupancy data, resource selection functions (RSFs) and related models can be used to estimate abundance (Boyce et al. 2016). RSFs and occupancy models can be used to estimate abundance by analyzing habitat selection and use patterns of animals. RSFs estimate the probability of habitat use based on the distribution of attributes associated with used resource units compared to available resource units. This information can then be used to calculate the relative probability of selection.

Occupancy models, on the other hand, estimate the presence or absence of animals in a given area. By combining occupancy data with habitat use information, abundance can be predicted. These models consider factors such as detection probability and can be adjusted to account for false absences or difficulties in detecting animals.

- Density

  Estimates of population density are essential for mountain lion management and conservation in southern California. Although estimation of population density in wildlife populations without individual recognition is more challenging than estimates in naturally or artificially marked wildlife populations, recently, certain camera trapping techniques make feasible to attain precise and accurate estimates of population density. Two of these techniques are highly recommended for mountain lions (see recommendation #4) but we may use our long-term place camera trap database to explore density with other techniques that have also used for other carnivores. Within the techniques employed to estimate density in unmarked wildlife populations we may consider the following ones to apply to San Diego County and other southern California mountain lions:

  A) Distance Sampling with Camera Traps (or Camera Trap Distance Sampling; CT-DS).
     Distance Sampling model was developed to estimate density from line- or point-transect surveys, and it has been adapted for use with camera trap data (Howe et al 2017). This model can be applied to low-density populations (Palencia et al. 2021).
  B) Random Encounter Model (REM).
     This model treats animals like ideal gas particles (i.e., randomly moving entities which are neither attracted to nor repelled by one another or landscape features (Rowcliffe et al. 2008; Gilbert et al. 2021). If animals behave like ideal gas particles, the rate at which they are detected by camera traps is a function of population density, animal movement, and the area within which cameras detect animals (Nakashima et al. 2018).
  C) The random encounter and staying time (REST).
     The random encounter and staying time model is an extension of the random encounter model. The REST treats animals like ideal gas particles but without requiring measures of animal movement speed. Instead, the model uses the time animals spend in the camera viewshed (i.e.,"staying time") as a proxy for animal movement speed, since the two measures are inversely proportional (Nakashima et al. 2018).

     These three models assume that camera traps are placed randomly with respect to animal movement, the study population is closed, animal movement and behavior are not affected by the camera trap, and the observations are considered independent events (Palencia et al. 2021). The general recommendations for the use of these three models to estimate density of unmarked populations are: REST could be recommended in scenarios of high abundance, CT-DS in those of low abundance while REM can be recommended when camera trap performance is not optimal, as it can be applied with less risk of bias (Palencia et al. 2021).

Due to the compromised genetic health of the southern California mountain lion subpopulation, long-term placed cameras may reveal further deterioration on the genetic status of the population by recording a decrease on kitten productivity over time or presence of morphological traits like kinked tails that have been associated with inbreeding in mountain lions.

3.2. Capture and radio-collaring mountain lions and spatial-mark-resight models.

Capture and radio-collaring can significantly enhance population estimation and monitoring efforts for mountain lions through their continuous tracking capabilities. Radio-collaring mountain lions provide a wealth of knowledge in regards to 1) demographics (e.g., survival, mortality, productivity, recruitment); 2) population genetics; 3) habitat connectivity and habitat use; 4) health/disease exposure; 5) foraging ecology; 6) behavior and population interactions; 7) human-wildlife conflict; and 8) response to environmental change (e.g., as habitat loss, climate change, or human disturbances).

Radio-collaring provides a vital source of detailed data on mountain lion populations, helping make informed decisions about population monitoring, management, and conservation efforts. By collaring a proportion of the mountain lion population other population density analyses can also be incorporated. When only a fraction of a population carries marks (i.e., radio-collars, ear tags), the spatial mark-resight (SMR) models can be implemented to estimate population density.

Mark-resight models need only a subset of the population to be marked (either naturally or from a single trapping-and-tagging event; Sollmann et al. 2013). The entire population is then resighted using a non-invasive survey technique (i.e., a camera trap survey; Sollmann et al. 2013). Incorporating this analysis for future density estimates adds value to our capture efforts and also supports the financial investment of the capture operations.

3.3. Time-to-event and space-to-event models.

The time-to-event (TTE) and space-to-event (STE) models represent more recent camera-based analysis that attempt to derive local mountain lion densities using large-scale camera trap monitoring efforts (Loonam et al. 2021, Clarke et al, 2023).
Due to our vast experience working with cameras these techniques may well be viable tools for population estimation in San Diego County and elsewhere in southern California using our own cameras but also investigating the potential value of other cameras placed by collaborative entities.

Some of the advantages of the TTE model include (Clarke et al. 2023):
The model requires less image processing time/effort than other models. Researchers need only "take" images until an animal is first in frame; any further images do not need to be processed (Moeller et al. 2018). The TTE model relies on whether an individual was within frame during each sampling period (Moeller et al. 2018). The TTE can account for spatial variation in density (Moeller et al. 2018). A model extension is available which compares densities at different camera stations as a result of habitat covariates (Loonam et al. 2021b). TTE studies are simple to scale up or down, since the number of cameras determines the

precision of estimates, the precision is not determined by camera density or coverage (Loonam et al. 2021a). A hundred cameras can be used to estimate density in a large area just as effectively as a small area (Loonam et al. 2021a).

On the other hand, some of the advantages found while using STE models include (Clarke et al. 2023):
The STE model does not require measurements of animal movement speed (Moeller et al. 2018). Instead, sampling occasions are folded into instants in time using camera traps' time-lapse function. The STE is not biased by animal movement speed (i.e., animals travelling slow vs fast). This differs with the TTE model, which is sensitive to animal movement speed (i.e., more reliable when animals move fast; Moeller et al. 2018). The STE model, as the TTE, does not rely on counts of animals in images; researchers need only record whether any animals were in the viewshed at each camera during a sampling occasion (Moeller et al. 2018). Consequently, the model is unaffected by weather, travelling and explorative behaviour, obstructions, camera malfunctions and other factors that make accurately counting the number of individuals challenging (Moeller et al. 2018). The STE uses time-lapse images, therefore there is essentially no uncertainty in detection probability when using time-lapse images (Moeller et al. 2018). STE studies are simple to scale up or down. Just the number of cameras determines the precision of estimates (Loonam et al. 2021a).

Conversely, some restrictions apply while using these models. One of the limitations of the TTE model is its sensitivity to movement speed. This model requires accurate measures of animal movement speed to set the sampling period (Moeller et al. 2018, Loonam et al. 2021a, Morin et al. 2022). Incorrect values of movement speed bias density estimates (Loonam et al. 2021b). Randomly-placed cameras may not collect sufficient images of elusive/rare species, therefore, this model is best suited to relatively common, high-density species (Moeller et al. 2018, Morin et al. 2022). The STE model does not apply well to very rare species, as detections of animals from randomly-placed cameras in time-lapse images can be too few to draw significant conclusions (Moeller et al. 2018, Loonam et al. 2021b), which may obligate to deploy more cameras to increase the number of detections (Moeller et al. 2018, Loonam et al. 2021a). Because animal movement speed is not an input, the STE is less precise than the TTE (Moeller et al. 2018, Loonam et al. 2021a). Depending on the time-lapse interval chosen, the STE can produce a high number of images (Morin et al. 2022).

Both models will also allow the analysis of population density of other species ecologically related to mountain lions such as deer, which increases the value of these techniques.

3.4. DNA analysis: hair sampling

Hair snares are more time consuming and financially challenging than other methods such as scat dog surveys for acquiring DNA for population estimation in southern California. However, the ability to use cameras at hair snares, and the information that they provide, may make the extra investment worthwhile if protocols are refined and if more effective snaring methods can be developed.

3.5. New DNA technique: hair shafts as new source of DNA.

We are currently investigating the potential use of hair shafts as source of DNA. Hair sequencing has been used in humans and it has potential to work in mountain lions. The California Conservation Genomics Program already has about 500 lion genomes from across the state. If this technique is proven to work, it would easily increase this database size, and allow identifying almost any lion's pedigree through its genome.

3.6. Scat DNA

Collecting fecal excrement (scat) deposited by animals can be used to examine a variety of information on wildlife species. Using molecular techniques to extract and analyze DNA that may be present on the surface of the scat sample provides data from as basic as presence/absence of a species in a specific area to identifying specific individuals along with their sex. Specially trained “scat dogs” are used to locate the fecal samples and by associating the GPS location of the sample with an individual animal it can allow for analysis of distribution, relative abundance, density and sex ratios. This technique may also provide genetic health metrics such as heterozygosity. Since this metric does not change in a short period of time (< 5 years), conducting a grid-based scat collection survey across the county conserved lands would not be advised to occur in the next year(s). This future sampling effort will compare to density/abundance estimate to the previous estimate generated in 2023 from the 2020 sampling effort. Opportunistically collected scat that is found by field personnel can also add to the total DNA detections in an area and be incorporated in models that use multiple sources of data (Vickers et al. 2022).

3.7. Facial recognition in camera trap surveys

The UCD-WHC team is currently evaluating trail camera photographs taken at hair snare stations used for DNA collection as a method for population estimation in the Santa Ana Mountain Range. Photographs taken at those stations, along with large numbers of other photographs of known mountain lions from UCD-WHC’s previous and ongoing studies in San Diego County will be utilized for software development and testing. This software development is aimed at identifying animals to species level, and mountain lions to the individual level, from trail camera photographs.

This work will help the collaborating researchers to fully develop the potential of trail camera arrays to assist in the long-term monitoring of the San Diego County mountain lion populations.

4. Time-frame

Technique	2024	2025	2026	2027
Long-term placement of cameras and documentation of photos from all source cameras in a database	X	x	x	X*
Capture and radio-collaring	x	x	x	X*
DNA sampling from captures and mortalities, and health monitoring via necropsies of deceased animals	x	x	x	X*
TTE-STE model trial in Santa Ana Range and deployment in eastern Peninsular Range if viable		X**	X**	X**
Hair shaft DNA extraction investigation and further hair snare trials	x	?	?	
Scat dog survey				X*
Mountain lion facial recognition software development	x	x	x	

*If funding is secured/ongoing

**If trial in Santa Ana Range is successful and funding becomes available

5. Budget

This budget expresses approximate costs based on previous efforts.

Technique	2024 Cost	2025 Cost	2026 Cost	2027 Cost
Long-term placement of cameras and documentation of photos from all source cameras in a database	$47,095	$47,095	$47,095	$47,095*
Capture and radio-collaring	$35,730	$35,730	$35,730	$35,730*
DNA sampling from captures and mortalities, and health monitoring via necropsies of deceased animals	$48,425	$48,425	$48,425	$48,425*
TTE-STE model trial in Santa Ana Range and deployment in eastern Peninsular Range if viable	Other funding		$175,000*	
Hair shaft DNA extraction investigation and further hair snare trials	No cost	$175,000**		
Scat dog survey				$175,000**
Mountain lion facial recognition software development	Dependent on entities conducting analyses	Dependent on entities conducting analyses	Dependent on entities conducting analyses	

*****Funding not secured

******DNA extraction and processing costs may not be included

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