MAMMALS - JULY 2007

WILDLIFE RESEARCH REPORTS

JULY 2006 – JUNE 2007

MAMMALS PROGRAM

COLORADO DIVISION OF WILDLIFE
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Task No.	1	: Post-Release Monitoring of Lynx
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ABSTRACT

In an effort to establish a viable population of lynx (*Lynx canadensis*) in Colorado, the Colorado Division of Wildlife (CDOW) initiated a reintroduction effort in 1997 with the first lynx released in February 1999. From 1999-2007, 218 lynx were released in Colorado. We documented survival, movement patterns, reproduction, and landscape habitat-use through aerial (n = 9496) and satellite (n = 23,791) tracking. Most lynx remained near the core release area in southwestern Colorado. From 1999-June 2007, there were 98 mortalities of released adult lynx. Approximately 30.6% were human-induced which were attributed to collisions with vehicles or gunshot. Starvation and disease/illness accounted for 19.4% of the deaths while 35.7% of the deaths were from unknown causes. Reproductive females had the smallest 90% utilization distribution home ranges ($\bar{x}$ = 75.2 km^2, SE = 15.9 km^2), followed by attending males ($\bar{x}$ = 102.5 km^2, SE = 39.7 km^2) and non-reproductive animals ($\bar{x}$ = 653.8 km^2, SE = 145.4 km^2). Reproduction was first documented in 2003 with subsequent successful reproduction in 2004, 2005 and 2006. No dens were documented in 2007. From snow-tracking, the primary winter prey species (n = 506 kills) were snowshoe hare (*Lepus americanus*, annual $\bar{x}$ = 74.9%, SE = 4.6, n = 9) and red squirrel (*Tamiasciurus hudsonicus*, annual $\bar{x}$ = 16.5%, SE = 4.1, n = 9); other mammals and birds formed a minor part of the winter diet. Lynx use-density surfaces were generated to illustrate relative use of areas throughout Colorado and areas of use in New Mexico, Utah and Wyoming. Within the areas of high use in southwestern Colorado, site-scale habitat use, documented through snow-tracking, supports mature Engelmann spruce (*Picea engelmannii*)-subalpine fir (*Abies lasiocarpa*) forest stands with 42-65% canopy cover and 15-20% conifer understory cover as the most commonly used areas in southwestern Colorado. Little difference in aspect (slight preference for north-facing slopes), slope ($\bar{x}$ = 15.7°) or elevation ($\bar{x}$ = 3173 m) were detected for long beds, travel and kill sites (n = 1841). Den sites (n = 37)

however, were located at higher elevations ($\bar{x}$ = 3354 m, SE = 31 m) on steeper ($\bar{x}$ = 30°, SE = 2°) and more commonly north-facing slopes with a dense understory of coarse woody debris. The first year of a study to evaluate snowshoe hare densities, demography and seasonal movement patterns among small and medium tree-sized lodgepole pine stands and mature spruce/fir stands was completed in 2006-2007 and will continue through 2009 (see Appendix I of this report). Results to date have demonstrated that CDOW has developed lynx release protocols that ensure high initial post-release survival followed by high long-term survival, site fidelity, reproduction and recruitment of Colorado-born lynx into the Colorado breeding population. What is yet to be demonstrated is whether Colorado can support sufficient recruitment to offset annual mortality for a viable lynx population over time. Monitoring continues in an effort to document such viability.

WILDLIFE RESEARCH REPORT

POST RELEASE MONITORING OF LYNX (*LYNX CANADENSIS*) REINTRODUCED TO COLORADO

TANYA M. SHENK

P. N. OBJECTIVE

The initial post-release monitoring of Canada lynx (*Lynx canadensis*) reintroduced into Colorado will emphasize 5 primary objectives:

1. Assess and modify release protocols to ensure the highest probability of survival for each lynx released.
2. Obtain regular locations of released lynx to describe general movement patterns and habitats used by lynx.
3. Determine causes of mortality in reintroduced lynx.
4. Estimate survival of lynx reintroduced to Colorado.
5. Estimate reproduction of lynx reintroduced to Colorado.

Three additional objectives will be emphasized after lynx display site fidelity to an area:

6. Refine descriptions of habitats used by reintroduced lynx.
7. Refine descriptions of daily and overall movement patterns of reintroduced lynx.
8. Describe hunting habits and prey of reintroduced lynx.

Information gained to achieve these objectives will form a basis for the development of lynx conservation strategies in the southern Rocky Mountains.

SEGMENT OBJECTIVES

1. Complete winter 2006-07 field data collection on lynx habitat use at the landscape scale, hunting behavior, diet, mortalities, and movement patterns.
2. Complete winter 2006-07 lynx trapping field season to collar Colorado born lynx and re-collar adult lynx.
3. Complete spring 2007 field data on lynx reproduction.
4. Summarize and analyze data and publish information as Progress Reports, peer-reviewed manuscripts for appropriate scientific journals, or CDOW technical publications.
5. Complete the first year of field work to evaluate snowshoe hare (*Lepus americanus*) densities, demography and seasonal movement patterns among small and medium tree-sized lodgepole pine stands and mature spruce/fir stands (see Appendix I).

INTRODUCTION

The Canada lynx occurs throughout the boreal forests of northern North America. Colorado represents the southern-most historical distribution of lynx, where the species occupied the higher elevation, montane forests in the state. Little was known about the population dynamics or habitat use of this species in their southern distribution. Lynx were extirpated or reduced to a few animals in the state by the late 1970’s due, most likely, to predator control efforts such as poisoning and trapping. Given the isolation of Colorado to the nearest northern populations, the CDOW considered reintroduction as the only option to attempt to reestablish the species in the state.

A reintroduction effort was begun in 1997, with the first lynx released in Colorado in 1999. To date, 218 wild-caught lynx from Alaska and Canada have been released in southwestern Colorado. The goal of the Colorado lynx reintroduction program is to establish a self-sustaining, viable population of lynx in this state. Evaluation of incremental achievements necessary for establishing viable populations is an interim method of assessing if the reintroduction effort is progressing towards success. There are 7 critical criteria for achieving a viable population: 1) development of release protocols that lead to a high initial post-release survival of reintroduced animals, 2) long-term survival of lynx in Colorado, 3) development of site fidelity by the lynx to areas supporting good habitat in densities sufficient to breed, 4) reintroduced lynx must breed, 5) breeding must lead to reproduction of surviving kittens 6) lynx born in Colorado must reach breeding age and reproduce successfully, and 7) recruitment must equal or be greater than mortality over an extended period of time.

The post-release monitoring program for the reintroduced lynx has 2 primary goals. The first goal is to determine how many lynx remain in Colorado and their locations relative to each other. Given this information and knowing the sex of each individual, we can assess whether these lynx can form a breeding core from which a viable population might be established. From these data we can also describe general movement patterns and habitat use. The second primary goal of the monitoring program is to estimate survival of the reintroduced lynx and, where possible, determine causes of mortality for reintroduced lynx. Such information will help in assessing and modifying release protocols and management of lynx once they have been released to ensure their highest probability of survival.

Additional goals of the post-release monitoring program for lynx reintroduced to the southern Rocky Mountains included refining descriptions of habitat use and movement patterns and describing successful hunting habitat once lynx established home ranges that encompassed their preferred habitat. Specific objectives for the site-scale habitat data collection include: 1) describe and quantify site-scale habitat use by lynx reintroduced to Colorado, 2) compare site-scale habitat use among types of sites (e.g., kills vs. long-duration beds), and 3) compare habitat features at successful and unsuccessful snowshoe hare chases.

Documenting reproduction is critical to the success of the program and lynx are monitored intensively to document breeding, births, survival and recruitment of lynx born in Colorado. Site-scale habitat descriptions of den sites are also collected and compared to other sites used by lynx.

The program will also investigate the ecology of snowshoe hare in Colorado. A study comparing snowshoe hare densities among mature stands of Engelmann spruce (*Picea engelmannii*)/subalpine fir (*Abies lasiocarpa*), lodgepole pine (*Pinus contorta*) and Ponderosa pine (*Pinus ponderosa*) was completed in 2004 with highest hare densities found in Engelmann spruce/subalpine fir stands and no hares found in Ponderosa pine stands. A study to evaluate the importance of young, regenerating lodgepole pine and mature Engelmann spruce/subalpine fir stands in Colorado by examining density and demography of snowshoe hares that reside in each was initiated in 2005 and will continue through 2009 (see Appendix I).

Lynx is listed as threatened under the Endangered Species Act (ESA) of 1973, as amended (16 U. S. C. 1531 et. seq.)(U. S. Fish and Wildlife Service 2000). Colorado is included in the federal listing as lynx habitat. Thus, an additional objective of the post-release monitoring program is to develop conservation strategies relevant to lynx in Colorado. To develop these conservation strategies, information specific to the ecology of the lynx in its southern Rocky Mountain range, such as habitat use, movement patterns, mortality factors, survival, and reproduction in Colorado is needed.

STUDY AREA

Southwestern Colorado is characterized by wide plateaus, river valleys, and rugged mountains that reach elevations over 4200 m. Engelmann spruce-subalpine fir is the most widely distributed coniferous forest type at elevations most typically used by lynx. The Core Release Area is defined as areas bounded by the New Mexico state line to the south, Taylor Mesa to the west and Monarch Pass on the north and east and > 2900 m in elevation (Figure 1). The lynx-established core area is roughly bounded by areas used by lynx in the Taylor Park/Collegiate Peak areas in central Colorado and includes areas of continuous use by lynx, including areas used during breeding and denning (Figure 1).

METHODS

REINTRODUCTION

Effort

All lynx releases were conducted under the protocols found to maximize survival (see Shenk 2001). Estimated age, sex and body condition were ascertained and recorded for each lynx prior to release (see Wild 1999). Specific release sites were those used in earlier years of the project and were selected based on land ownership and accessibility during times of release (Byrne 1998). Lynx were transported from the Frisco Creek Wildlife Rehabilitation Center, where they were held from their time of arrival in Colorado, to their release site in individual cages. Release site location was recorded in Universal Transverse Mercator (UTM) coordinates and identification of all lynx released at the same location, on the same day, was recorded. Behavior of the lynx on release and movement away from the release site were documented.

Movement, Distribution and Relative Use of Areas by Lynx

To monitor lynx movements and thus determine distribution and relative use of areas all released lynx were fitted with radio collars. All lynx released in 1999 were fitted with Telonics™ radio-collars. All lynx released since 1999, with the exception of 5 males released in spring 2000, were fitted with Sirtrack™ dual satellite/VHF radio-collars. These collars have a mortality indicator switch that operated on both the satellite and VHF mode. The satellite component of each collar was programmed to be active for 12 hours per week. The 12-hour active periods for individual collars were staggered throughout the week. Signals from the collars allowed for locations of the animals to be made via Argos, NASA, and NOAA satellites. The location information was processed by ServiceArgos and distributed to the CDOW through e-mail messages.

Datasets.-- To determine recent (post-reintroduction) movement and distribution of lynx reintroduced, born or initially trapped in Colorado and relative use of areas by these lynx, regular locations of lynx were collected through a combination of aerial and satellite tracking. Locations were recorded and general habitat descriptions for each aerial location was recorded. The first dataset of lynx locations included all locations obtained from daytime flights conducted with a Cessna 185 or similar aircraft to locate lynx by their VHF collar transmitters (hereafter aerial locations). VHF transmitters have been used on lynx since the first lynx were released in February 1999. The second type of lynx location data was collected via satellite from the satellite collar transmitters placed on the lynx (hereafter satellite locations). Satellite transmitter collars were first used for lynx in April 2000. These satellite collars also contained a VHF transmitter which also allowed locating lynx from the air or ground. All locations were recorded in Universal Transverse Mercator (UTM) coordinates using the CONUS NAD27 datum.

Flights to obtain lynx aerial locations were typically conducted on a weekly basis throughout most summer and winter months and twice a week during the den search field season (May 15 – June 30), depending on weather and availability of planes and pilots. Flights were typically concentrated in the high elevation (> 2700 m) southwest quadrant of Colorado which encompasses the core lynx release and research area (Figure 1). Flights during the den seasons were conducted to obtain locations on all female lynx within the state wearing an active VHF transmitter. VHF transmitters were outfitted with sufficient batteries to last 60 months. The satellite transmitters were designed to provide locations on a weekly basis with sufficient batteries to last for 18 months.

Lynx may not be exhibiting typical behavior or habitat use within the first few months after their release in Colorado. Therefore, a subset of each of the aerial and satellite datasets was created that eliminated the first 180 days (approximately 6 months) of locations obtained for each lynx immediately after their initial release. As a result, the truncated aerial location dataset contained lynx locations from September 1999 through March 2007 while the truncated satellite location dataset began October 2000 and extended through March 2007.

Accuracy of both aerial and satellite locations varied with the environmental conditions at the time the location was obtained. Accuracy of aerial locations was influenced by weather with accuracy ranging from 50 - 500 meters. Satellite location accuracy was also influenced by atmospheric conditions and position of the satellites. Satellite location accuracy ranged from 150 meters -10 km.

Movement and Distribution.-- To document all known lynx locations maps were generated with all aerial and satellite locations displayed. Due to lynx movements outside of Colorado, particularly into the states of New Mexico, Utah and Wyoming we further evaluated lynx use throughout those three states, as well as the data would allow. All individual lynx located at least once in these 3 states (non-truncated datasets) were identified and tallied for each year. To document consistency and known use of these states after the initial effect of being reintroduced was minimized (i.e., 180 days post-release), each individual lynx located at least once in these states from the truncated datasets were identified and tallied.

Relative Use.-- To document relative use of areas by lynx, 90% kernel use-density surfaces were calculated for truncated satellite and aerial lynx locations using the ArcGIS Spatial Analyst Kernel Density Tool. Due to differences in data collection frequency and accuracy between datasets, the truncated satellite and truncated aerial data were analyzed separately for generating the lynx use-density surfaces.

These use-density surfaces fit a smoothly curved surface over each lynx location. The surface value was highest at the location of the point and diminished with increasing distance from the point. A fixed kernel was used with a smoothing parameter of 5 km, reaching 0 at the search radius distance from the point. Only a circular neighborhood was possible. The volume under the surface equaled the total value for the point. The use-density at each output GIS raster cell was calculated by adding the values of all the kernel surfaces from all the lynx point locations that overlaid each raster cell center. The kernel function was based on the quadratic kernel function described in Silverman (1986, p. 76, equation 4.5). The use-density surfaces were calculated at 100 m resolution. To enhance graphic displays of higher use-density areas, density values representing single locations were not displayed.

Home Range

Annual home ranges were calculated as a 95% utilization distribution using a kernel home-range estimator for each lynx we had at least 30 locations for within a year. A year was defined as March 15 – March 14 of the following year. Locations used in the analyses were collected from September 1999 – January 2006 and all locations obtained for an individual during the first six months after its release were eliminated from any home range analyses as it was assumed movements of lynx initially post-release may not be representative of normal habitat use. Locations were obtained either through aerial VHF surveys or locations or the midpoint (ArcView Movement Extension) of all high quality (accuracy rating of 0-1km) satellite locations obtained within a single 24-hour period. All locations used within a single home range analysis were taken a minimum of 24 hours apart.

Home range estimates were classified as being for a reproductive or non-reproductive animal. A reproductive female was defined as one that had kittens with her; a reproductive male was defined as a male whose movement patterns overlapped that of a reproductive female. If a litter was lost within the defined year a home range described for a reproductive animal were estimated using only locations obtained while the kittens were still with the female.

Survival

Survival was estimated as ragged telemetry data using the nest survival models in Program MARK (White and Burnham 1999).

Mortality Factors

When a mortality signal (75 beats per minute [bpm] vs. 50 bpm for the Telonics™ VHF transmitters, 20 bpm vs. 40 bpm for the Sirtrack™ VHF transmitters, 0 activity for Sirtrack™ PTT) was heard during either satellite, aerial or ground surveys, the location (UTM coordinates) was recorded. Ground crews then located and retrieved the carcass as soon as possible. The immediate area was searched for evidence of other predators and the carcass photographed in place before removal. Additionally, the mortality site was described and habitat associations and exact location were recorded. Any scat found near the dead lynx that appeared to be from the lynx was collected.

All carcasses were transported to the Colorado State University Veterinary Teaching Hospital (CSUVTH) for a post mortem exam to 1) determine the cause of death and document with evidence, 2) collect samples for a variety of research projects, and 3) archive samples for future reference (research or forensic). The gross necropsy and histology were performed by, or under the lead and direct supervision of a board certified veterinary pathologist. At least one research personnel from the CDOW involved with the lynx program was also present. The protocol followed standard procedures used for thorough post-mortem examination and sample collection for histopathology and diagnostic testing (see Shenk 1999 for details). Some additional data/samples were routinely collected for research, forensics, and archiving. Other data/samples were collected based on the circumstances of the death (e.g., photographs, video, radiographs, bullet recovery, samples for toxicology or other diagnostic tests, etc.).

From 1999–2004 the CDOW retained all samples and carcass remains with the exception of tissues in formalin for histopathology, brain for rabies exam, feces for parasitology, external parasites for ID, and other diagnostic samples. Since 2005 carcasses are disposed of at the CSUVTH with the exception of the lower canine, fecal samples, stomach content samples and tissue or bone marrow samples to be delivered by CDOW to the Center for Disease control for plague testing. The lower canine, from all carcasses, is sent to Matson Labs (Missoula, Montana) for aging and the fecal and stomach content samples are evaluated for diet.

Reproduction

Females were monitored for proximity to males during each breeding season. We defined a possible mating pair as any male and female documented within at least 1 km of each other in breeding season through either flight data or snow-tracking data. Females were then monitored for site fidelity to a given area during each denning period of May and June. Each female that exhibited stationary movement patterns in May or June were closely monitored to locate possible dens. Dens were found when field crews walked in on females that exhibited virtually no movement for at least 10 days from both aerial and ground telemetry.

Kittens found at den sites were weighed, sexed and photographed. Each kitten was uniquely marked by inserting a sterile passive integrated transponder (PIT, Biomark, Inc., Boise, Idaho, USA) tag subcutaneously between the shoulder blades. Time spent at the den was minimized to ensure the least amount of disturbance to the female and the kittens. Weight, PIT-tag number, sex and any distinguishing characteristics of each kitten was also recorded. Beginning in 2005, blood and saliva samples were collected and archived for genetic identification.

During the den site visits, den site location was recorded as UTM coordinates. General vegetation characteristics, elevation, weather, field personnel, time at the den, and behavioral responses of the kittens and female were also recorded. Once the females moved the kittens from the natal den area, den sites were visited again and site-specific habitat data were collected (see Habitat Use section below).

Captures

Captures were attempted for either lynx that were in poor body condition or lynx that needed to have their radio-collars replaced due to failed or failing batteries or to radio-collar kittens born in Colorado once they reached at least 10-months of age when they were nearly adult size. Methods of recapture included 1) trapping using a Tomahawk™ live trap baited with a rabbit and visual and scent lures, 2) calling in and darting lynx using a Dan-Inject CO_2 rifle, 3) custom box-traps modified from those designed by other lynx researchers (Kolbe et al. 2003) and 4) hounds trained to pursue felids were also used to tree lynx and then the lynx was darted while treed. Lynx were immobilized either with Telazol (3 mg/kg; modified from Poole et al. 1993 as recommended by M. Wild, DVM) or medetomidine (0.09mg/kg) and ketamine (3 mg/kg; as recommended by L. Wolfe, DVM)) administered intramuscularly (IM) with either an extendible pole-syringe or a pressurized syringe-dart fired from a Dan-Inject air rifle.

Immobilized lynx were monitored continuously for decreased respiration or hypothermia. If a lynx exhibited decreased respiration 2mg/kg of Dopram was administered under the tongue; if respiration was severely decreased, the animal was ventilated with a resuscitation bag. If medetomidine/ketamine were the immobilization drugs, the antagonist Atipamezole hydrochloride (Antisedan) was administered. Hypothermic (body temperature < 95^{o} F) animals were warmed with hand warmers and blankets.

While immobilized, lynx were fitted with replacement Sirtrack™ VHF/satellite collar and blood and hair samples were collected. Once an animal was processed, recovery was expedited by injecting the equivalent amount of the antagonist Antisedan IM as the amount of medetomidine given, if medetomodine/ketemine was used for immobilization. Lynx were then monitored while confined in the box-trap until they were sufficiently recovered to move safely on their own. No antagonist is available for Telezol so lynx anesthetized with this drug were monitored until the animal recovered on its own in the box-trap and then released. If captured and in poor body condition, lynx were anesthetized with either Telezol (2 mg/kg) or medetomodine/ketemine and returned to the Frisco Creek Wildlife Rehabilitation Center for treatment.

HABITAT USE

Gross habitat use was documented by recording canopy vegetation at aerial locations. More refined descriptions of habitat use by reintroduced lynx were obtained through following lynx tracks in the snow (i.e., snow-tracking) and site-scale habitat data collection conducted at sites found through this method to be used by lynx. See Shenk (2006) for detailed methodologies.

DIET AND HUNTING BEHAVIOR

Winter diet of reintroduced lynx was estimated by documenting successful kills through snow-tracking. Prey species from failed and successful hunting attempts were identified by either tracks or remains. Scat analysis also provided information on foods consumed. Scat samples were collected wherever found and labeled with location and individual lynx identification. Only part of the scat was collected (approximately 75%); the remainder was left in place in the event that the scat was being used by the animal as a territory mark. Site-scale habitat data collected for successful and unsuccessful snowshoe hare kills were compared.

SNOWSHOE HARE ECOLOGY

A study designed to evaluate the importance of young, regenerating lodgepole pine and mature Engelmann spruce / subalpine fir stands in Colorado by examining density and demography of snowshoe hares that reside in each was initiated in 2005.

Specifically, the study was designed to evaluate small and medium lodgepole pine stands and large spruce/fir stands where the classes “small”, “medium”, and “large” refer to the diameter at breast height (dbh) of overstory trees as defined in the United States Forest Service R2VEG Database (small = 2.54–12.69 cm dbh, medium = 12.70–22.85 cm, and large = 22.86–40.64 cm dbh; J. Varner, United States Forest Service, personal communication). The study design was also developed to identify which of the numerous density-estimation procedures available perform accurately and consistently using an innovative, telemetry augmentation approach as a baseline. Movement patterns and seasonal use of deciduous cover types such as riparian willow were assessed. Finally, the study was designed to further expound on the relationship between density, demography, and stand-type by examining how snowshoe hare density and demographic rates vary with specific vegetation, physical, and landscape characteristics of a stand.

RESULTS

REINTRODUCTION

Effort

From 1999 through 2006, 218 lynx were reintroduced into southwestern Colorado (Table 1). No lynx were released in 2007. All lynx were released with either VHF or dual VHF/satellite radio collars so they could be monitored for movement, reproduction and survival. The CDOW does not plan to release any additional lynx in 2008.

Movement Patterns and Distribution

Numerous travel corridors were used repeatedly by more than one lynx. These travel corridors include the Cochetopa Hills area for northerly movements, the Rio Grande Reservoir-Silverton-Lizardhead Pass for movements to the west, and southerly movements down the east side of Wolf Creek Pass to the southeast through the Conejos River Valley. Lynx appear to remain faithful to an area during winter months, and exhibit more extensive movements away from these areas in the summer.

A total of 9496 aerial and 23791 satellite locations were obtained from the 218 reintroduced lynx, radio-collared Colorado kittens (n = 14) and unmarked lynx captured in Colorado (n = 2) as of June 30, 2007. The majority of these locations were in Colorado (Figure 2). Some reintroduced lynx dispersed outside of Colorado into Arizona, Idaho, Iowa, Kansas, Montana, Nebraska, Nevada, New Mexico, South Dakota, Utah and Wyoming (Figure 2). The majority of surviving lynx from the reintroduction effort currently continue to use high elevation (> 2900 m), forested terrain in an area bounded on the south by New Mexico north to Independence Pass, west as far as Taylor Mesa and east to Monarch Pass. Most movements away from the Core Release Area were to the north.

Relative Use

The lynx use-density surfaces resulting from the fixed kernel analyses provided relative probabilities of finding lynx in areas throughout their distribution. A single use-density surface was calculated separately for both the aerial (n = 8058) and satellite truncated datasets (n = 16240).

Relative Use in Colorado.-- All 218 lynx released in Colorado, all radio-collared kittens and 2 captured unmarked adults were located at least once in Colorado. The majority of these lynx remained in Colorado. The use-density surfaces within Colorado were displayed separately for both the aerial (Figure 3) and satellite truncated datasets (Figure 4). Of the total locations available in the truncated datasets used to generate the use-density surfaces, 7953 of the aerial locations and 13,241 of the satellite locations were in Colorado. Aerial and satellite use-density surfaces indicated similar high use-density areas. Satellite locations indicated broader spatial use by lynx because satellite collars provided more locations than flights.

The use-density surface for lynx use in Colorado indicates two primary areas of use. The first is the Core Research Area (see Figure 1) and a secondary core centered in the Collegiate Peaks Wilderness (Figures 3 and 4). High use is also documented for 1) the area east of Dillon, on both the north and south sides of I70 and 2) the area north of Hwy 50 centered around Gunnison and then north to Crested Butte. These last 2 high use areas are smaller in extent than the 2 core areas.

Relative Use in New Mexico.-- Combining the non-truncated aerial (*n* = 81) and satellite lynx location (*n* = 928) datasets, lynx used New Mexico consistently and with an increasing number of individuals from 1999 through 2006 (Table 2). Data for 2007 represents only a partial year and thus trend in numbers of individuals using New Mexico for 2007 cannot be made, however continued use of New Mexico into 2007 was documented Sixty lynx (37 females: 23 males) were found within New Mexico from February 1999 through March 2007 (Table 2). Excluding all aerial and satellite lynx locations collected in the first 180 days after release (truncated datasets; *n* = 61 aerial locations, *n* = 569 satellite locations), a total of 35 individual lynx (22 females: 13 males) were found within New Mexico from September 1999 through March 2007 (Table 3).

The decrease in number of lynx frequenting New Mexico in 2001 through 2003 (Tables 2 and 3) was more likely due to fewer satellite collars functioning in those years rather than indicating less use of the area by lynx. The satellite transmitters placed on lynx in 2000 were failing and no new lynx were released or re-collared in 2001 and 2002. This decrease in satellite locations is present throughout the lynx distribution and is also reflected in the numbers presented below for Utah and Wyoming

The use-density surface for lynx use in New Mexico indicates the primary area of use being located either immediately south of the Colorado border and south of the Conejos River Valley (an area of high use in Colorado) or east of Taos (Figure 5). The use-density surfaces throughout both Colorado and New Mexico are displayed so that lynx use within New Mexico can be directly compared to lynx use throughout Colorado (Figure 6).

Relative Use in Utah.-- Combining the non-truncated aerial (*n* = 10) and satellite lynx location (*n* = 574) datasets, lynx used the analysis area consistently and with an increasing number of individuals from 1999 through 2006 (Table 4). Data for 2007 represents only a partial year and thus trend in numbers of individuals using the state for 2007 cannot be made, however continued use of Utah into 2007 was documented. Twenty-two lynx (7 females: 15 males) were found within Utah from February 1999 through March 2007 (Table 4). Excluding all aerial and satellite lynx locations collected in the first 180 days after release (truncated datasets; *n* = 7 aerial locations, *n* = 399 satellite locations), 17 individual lynx (6 females: 11 males) were found within Utah from September 1999 through March 2007 (Table 5).

The use-density surface for lynx use in Utah indicates the primary area of use being located in the Uinta Mountains (Figure 7). The use-density surfaces throughout both Colorado and Utah are displayed so that lynx use within Utah can be directly compared to lynx use throughout Colorado (Figure 8).

Relative Use in Wyoming.-- Combining the non-truncated aerial (*n* = 34) and satellite lynx location (*n* = 1780) datasets, lynx used the analysis area consistently and with an increasing number of individuals from 1999 through 2006 (Table 6). Data for 2007 represents only a partial year and thus trend in numbers of individuals using the state for 2007 cannot be made, however continued use of the Wyoming into 2007 was documented. Thirty-three lynx (14 females: 19 males) were found within

Wyoming from February 1999 through March 2007 (Table 6). Excluding all aerial and satellite lynx locations collected in the first 180 days after release (truncated datasets; n = 28 aerial locations, n = 1533 satellite locations), 27 individual lynx (13 females: 14 males) were found within Wyoming from September 1999 through March 2007 (Table 7).

The use-density surface for lynx use in Wyoming indicates the primary area of use being located either immediately north of the Colorado border in the Medicine Bow National Forest or in the northwest quadrant of the state including areas in Yellowstone and Teton National Parks and the Laramie Range (Figure 9). The use-density surfaces throughout both Colorado and Wyoming are displayed so that lynx use within Wyoming can be directly compared to lynx use throughout Colorado (Figure 10).

Home Range

Reproductive females had the smallest 90% utilization distribution annual home ranges ($\bar{x}$ = 75.2 km^2, SE = 15.9 km^2, n = 19), followed by attending males ($\bar{x}$ = 102.5 km^2, SE = 39.7 km^2, n = 4). Non-reproductive females had the largest annual home ranges ($\bar{x}$ = 703.9 km^2, SE = 29.8 km^2, n = 32) followed by non-reproductive males ($\bar{x}$ = 387.0 km^2, SE = 73.5 km^2, n = 6). Combining all non-reproductive animals yielded a mean annual home range of 653.8 km^2 (SE = 145.4 km^2, n = 38).

Survival

Initial survival rate estimates for reintroduced lynx were completed, however, further analyses need to be conducted before estimates will be presented. As of June 30, 2007, CDOW was actively monitoring/tracking 71 of the 120 lynx still possibly alive (Table 8). There are 50 lynx that we have not heard signals on since at least June 30, 2006 and these animals are classified as 'missing' (Table 8). One of these missing lynx is a mortality of unknown identity, thus only 49 are truly missing. Possible reasons for not locating these missing lynx include 1) long distance dispersal, beyond the areas currently being searched, 2) radio failure, or 3) destruction of the radio (e.g., run over by car). CDOW continues to search for all missing lynx during both aerial and ground searches. Two of the missing lynx released in 2000 are thought to have slipped their collars.

Mortality Factors

Of the total 218 adult lynx released, we have 98 known mortalities as of June 30, 2007 (Table 9). The primary known causes of death included 30.6% human-induced deaths which were confirmed or probably caused by collisions with vehicles or gunshot. Starvation and disease/illness accounted for 19.4% of the deaths; starvation was a significant cause of mortality in the first year of releases only. An additional 35.7% of known mortalities were from unknown causes.

Mortalities occurred throughout the areas where lynx moved, including 13 in New Mexico, 4 in Wyoming and Nebraska, 3 in Utah and 1 each in Arizona, Kansas and Montana (Figure 2, Table 10).

Reproduction

Field crews weighed, photographed, PIT-tagged the kittens and took hair, blood and saliva samples from the kittens for genetic work in an attempt to confirm paternity. Lynx kittens weigh approximately 200 grams at birth and do not open their eyes until they are 10-17 days old. Kittens were processed as quickly as possible (11-32 minutes) to minimize the time the kittens were without their mother. While working with the kittens the females remained nearby, often making themselves visible to the field crews. The females generally continued a low growling vocalization the entire time personnel were at the den. In all cases, the female returned to the den site once field crews left the area. At all dens the females appeared in excellent condition, as did the kittens.

2003.-- Nine pairs of lynx were documented during the 2003 breeding season (March and April) from the 17 females we were monitoring. In May and June, 6 dens and a total of 16 kittens were found in the lynx Core Release Area in southwestern Colorado (Table 11, Figure 1). The kittens weighed from 270-500 grams. The dens were scattered throughout the Core Release Area, with no dens found outside the core area. All the dens were in Engelmann spruce/subalpine fir forests in areas of extensive downfall. Elevations ranged from 3240-3557 m.

Four of the 6 females that we know had kittens in summer 2003 were still with kittens at the end of April 2004. Two of those females still had 2 kittens with them at that time. Visual observations in February 2004 of one female with 2 kittens indicated all 3 were in good body condition. The mortality of female YK00F16 and her 1 kitten in October 2003 from plague was not due to poor habitat or prey conditions, and thus we might assume she would have raised the 1 kitten to this stage as well. Three probable kitten deaths from female YK00F19 were from 1 litter that most likely failed very early. Through snow-tracking in winter 2003-04 an unknown female (no radio frequency heard in the area of the tracks) we also documented 1-2 additional kittens born spring 2003 and still alive in winter 2004.

Of the 16 kittens we found in summer 2003, we documented the following by April 2004: 6 confirmed alive, 7 confirmed dead, and 3 some evidence dead. Although we tried, we were not able to capture any of the 6 surviving kittens to fit them with radio-collars for subsequent monitoring.

2004.-- In Spring 2004, 26 females from the releases in 1999, 2000 and 2003 had active radio-collars. Of these, we documented 18 possible mating pairs of lynx during breeding season. All 4 of the females that had kittens with them through winter 2003-04 bred again spring 2004; 2 with the same male they successfully bred with spring 2003. During May-June 2004 we found 11 dens and a total of 30 kittens (Table 11). The kittens weighed from 250-770 grams. Three of the 11 females with kittens were from the 2003 releases. Three additional litters were documented after denning season through either observation of a female lynx with kittens or snow-tracking females with kittens that were not one of the 11 females found on dens. From the size of the kittens they would have been born during the normal denning season in May or June. Nine additional kittens were observed from these litters for a total of 39 known kittens born in 2004. Two of these additional litters were documented from direct follow-ups to sighting made by the public and reported to CDOW.

Two females that had kittens in 2003 and reared at least part of their litters through March 2004, bred and had kittens again in 2004. Two of the litters documented by direct observation or snow-tracking are from females whose collars were no longer functioning. Seven kittens born in 2004 were captured at approximately 10-months of age and fitted with dual satellite/VHF collars. Six of the 7 were still alive and being monitored as of June 30, 2006. The cut collar of one kitten CO04M15 was left at the Silverton Post Office on October 25, 2005. We assume this lynx is dead.

2005.-- In spring 2005 we had 40 females from the releases in 1999, 2000, 2003 and 2004 that had active radio-collars. We documented 23 possible mating pairs of lynx during breeding season. During May-June 2005 we visited 16 dens and found a total of 46 kittens (Table 11). An additional female (BC03F10) had a den we were not able to get to during May or June due to high water during spring run-off. Female BC03F03 was hit and killed on I-70 on 5/19/2005. She had 2 fetuses in her uterus, so would have contributed to reproduction this year had she lived.

All of the 2005 dens were scattered throughout the high elevation areas of Colorado, south of I-70. Most of the dens were in Engelmann spruce/subalpine fir forests in areas of extensive downfall. Elevations ranged from 3117-3586 m. Four of the females would not leave the den until we reached out to pick up a kitten.

One female, YK00F10 has had litters 3 years in a row. In 2003 she had 4 kittens and raised 2 through the winter. In 2004 she had 2 kittens and raised both through the winter, in 2005 she had 4 kittens again. She has had all 3 litters in the same general area and has had the same mate for 3 years. Eight additional females had their second litter in Colorado in 2005. Three females from the 2004 releases had litters in 2005. Year 2005 was the second consecutive year that we had females released the prior spring find a territory and a mate within a year and produced live young. In reproduction season 2004 we had 3 females released in spring 2003 that also produced live young the next year. Of those 3, 2 successfully raised at least part of their litters through winter 2005.

Seven kittens born in 2005 were captured at approximately 10-months of age and fitted with dual satellite/VHF collars. One of the 7 was still alive and being monitored as of June 30, 2007.

2006.-- In spring 2006, 42 females were being monitored. We found 4 dens in May and June 2006 with 11 kittens total (Table 11). Lynx CO04F07, a female lynx born in Colorado in 2004, was the mother of one of these litters which documented the first recruitment of Colorado-born lynx into the Colorado breeding population. There were at least 2 surviving kittens as of spring 2007. We were unsuccessful in capturing these kittens for collar placement.

The percent of tracked females found with litters in 2006 was lower (0.095) than in the 3 previous years (0.413, SE = 0.032, Table 11). However, all demographic and habitat characteristics measured at the 4 dens that were found in 2006 were comparable to all other dens found (Table 11). Mean number of kittens per litter from 2003-2006 was 2.78 (SE = 0.05) and sex ratio of females to males was equal ($\bar{x}$ = 1.14, SE = 0.14).

2007.-- During May and June 2007 we monitored 34 females for reproduction (Table 11). No dens were found.

Den Sites.-- A total of 37 dens have been found from 2003-2006. All of the dens except one have been scattered throughout the high elevation areas of Colorado, south of I-70. In 2004, 1 den was found in southeastern Wyoming, near the Colorado border. Dens were located on steep ($\bar{x}_{slope} = 30^o$, SE=2^o), north-facing, high elevation ($\bar{x}$ = 3354 m, SE = 31 m) slopes. The dens were typically in Engelmann spruce/subalpine fir forests in areas of extensive downfall of coarse woody debris (Shenk 2006). All dens were located within the winter use areas used by the females.

Captures

Two adult lynx were captured in 2001 for collar replacement. One lynx was captured in a tomahawk live-trap, the other was treed by hounds and then anesthetized using a jab pole. Five adult lynx were captured in 2002; 3 were treed by hounds and 2 were captured in padded leghold traps. In 2004, 1 lynx was captured with a Belisle snare and 6 adult lynx were captured in box-traps. Trapping effort was substantially increased in winter and spring 2005 and 12 adult lynx were captured and re-collared. Eight reintroduced lynx were captured in winter and spring 2006. In 2007, 11 reintroduced adult lynx were captured and re-collared. All lynx captured in Colorado from 2005-2007 were caught in box-traps.

In addition, as part of the collaring trapping effort, 14 Colorado-born kittens were captured and collared at approximately 10-months of age. Seven 2004-born kittens were collared in spring 2005, and 7, 2005-born kittens were collared in spring 2006. We were not successful at capturing and collaring any kittens born in 2006 in winter 2006-07. We did however, capture 2 adults (approximate age 2 years old) in winter 2006-07 that had no PIT-tags or radio collars. We assume these 2 lynx were from litters born in Colorado that were never found at dens (i.e., why there were no PIT-tags). All lynx captured for collaring or re-collaring were fitted with new Sirtrack ™ dual VHF/satellite collars and re-released at their capture locations.

Seven adult lynx were captured from March 1999-June 30, 2007 because they were in poor body condition (Table 12). Five of these lynx were successfully treated at the Frisco Creek Rehabilitation Center and re-released in the Core Release Area. One lynx, BC00F7, died from starvation and hypothermia within 1 day of capture at the rehabilitation center. Lynx QU04M07 died 3 days after capture at the rehabilitation center. Necropsy results documented starvation as the cause of death that was precipitated by hydrocephalus and bronchopneumonia (unpublished data T. Spraker, CSUVTH).

Seven lynx were captured (either by CDOW personnel or conservation personnel in other states) because they were in atypical habitat outside the state of Colorado (Table 12). They were held at Frisco Creek Rehabilitation Center for a minimum of 3 weeks, fitted with new Sirtrack ™ dual VHF/satellite collars and re-released in the Core Release Area in Colorado. Five of these 7 lynx were still alive 6 months post-re-release but 3 had already dispersed out of Colorado and 2 stayed in Colorado through June 30, 2007. Two lynx died within 6 months of re-release: 1 died of starvation in Colorado and the other died of unknown causes in Nebraska. Two lynx captured out of state and re-released currently remain in Colorado.

HABITAT USE

Landscape-scale daytime habitat use was documented from 9496 aerial locations of lynx collected from February 1999-June 30, 2007. Throughout the year Engelmann spruce - subalpine fir was the dominant cover used by lynx. A mix of Engelmann spruce, subalpine fir and aspen (*Populus tremuloides*) was the second most common cover type used throughout the year. Various riparian and riparian-mix areas were the third most common cover type where lynx were found during the daytime flights. Use of Engelmann spruce-subalpine fir forests and Engelmann spruce-subalpine fir-aspen forests was similar throughout the year. There was a trend in increased use of riparian areas beginning in July, peaking in November, and dropping off December through June.

Site-scale habitat data collected from snow-tracking efforts indicate Engelmann spruce and subalpine fir were also the most common forest stands used by lynx for all activities during winter in southwestern Colorado. Comparisons were made among sites used for long beds, dens, travel and where they made kills. Little difference in aspect, mean slope and mean elevation were detected for 3 of the 4 site types including long beds, travel and kills where lynx typically use gentler slopes ($\bar{x}$ = 15.7°) at an mean elevation of 3173 m, and varying aspects with a slight preference for north-facing slopes. See Shenk (2006) for more detailed analyses of habitat use.

DIET AND HUNTING BEHAVIOR

Winter diet of lynx was documented through detection of kills found through snow-tracking. Prey species from failed and successful hunting attempts were identified by either tracks or remains. Scat analysis also provided information on foods consumed. A total of 506 kills were located from February 1999-April 2007. We collected over 900 scat samples from February 1999-April 2007 that will be analyzed for content. In each winter, the most common prey item was snowshoe hare, followed by red squirrel (*Tamiusciurus hudsonicus*; Table 13). The percent of snowshoe hare kills found however, varied annually from a low of 55.56% in 1999 to a high of 90.77% in winter 2002-2003.

SNOWSHOE HARE ECOLOGY

The first year of a study to evaluate snowshoe hare densities, demography and seasonal movement patterns among small and medium tree-sized lodgepole pine stands and mature spruce/fir stands was completed and preliminary results presented (Appendix I).

DISCUSSION

In an effort to establish a viable population of lynx in Colorado, CDOW initiated a reintroduction effort in 1997 with the first lynx released in winter 1999. From 1999 through spring 2007, 218 lynx were released in the Core Release Area.

Locations of each lynx were collected through aerial- or satellite-tracking to document movement patterns and to detect mortalities. Most lynx remain in the high elevation, forested areas in southwestern Colorado. The use-density surfaces for lynx use in Colorado indicate two primary areas of use. The first is the Core Research Area (see Figure 1) and a secondary core centered in the Collegiate Peaks Wilderness (Figures 3, 4). High use is also documented for 1) the area east of Dillon, on both the north and south sides of I70 and 2) the area north of Hwy 50 centered around Gunnison and then north to Crested Butte. These last 2 high use areas are smaller in extent than the 2 core areas.

Dispersal movement patterns for lynx released in 2000 and subsequent years were similar to those of lynx released in 1999 (Shenk 2000). However, more animals released in 2000 and subsequent years remained within the Core Release Area than those released in 1999. This increased site fidelity may have been due to the presence of con-specifics in the area on release. Numerous travel corridors within Colorado have been used repeatedly by more than 1 lynx. These travel corridors include the Cochetopa Hills area for northerly movements, the Rio Grande Reservoir-Silverton-Lizardhead Pass for movements to the west, and southerly movements down the east side of Wolf Creek Pass to the southeast to the Conejos River Valley.

Lynx appear to remain faithful to an area during winter months, and exhibit more extensive movements away from these areas in the summer. Reproductive females had the smallest 90% utilization distribution home ranges ($\bar{x}$ = 75.2 km^2, SE = 15.9 km^2), followed by attending males ($\bar{x}$ = 102.5 km^2, SE = 39.7 km^2) and non-reproductive animals ($\bar{x}$ = 653.8 km^2, SE = 145.4 km^2). Most lynx currently being tracked are within the Core Release Area. During the summer months, lynx were documented to make extensive movements away from their winter use areas. Extensive summer movements away from areas used throughout the rest of the year have been documented in native lynx in Wyoming and Montana (Squires and Laurion 1999).

Current data collection methods used for the Colorado lynx reintroduction program were not specifically designed to address the reintroduced lynx movements or use of areas in other states. In particular, the core research and release area were in Colorado. Therefore, the number of aerial locations obtained would be far fewer in other states than in Colorado which would bias low the number of lynx and intensity of lynx use documented outside the state. In contrast, obtaining satellite locations is not biased by the location of the lynx. Satellite locations are, however, biased by the shorter time the satellite transmitters function, approximately 18 months versus 60 months for the VHF transmitters used to obtain the aerial locations. However, data collected to meet objectives of the lynx reintroduction program were used to provide information to help address the question of lynx use outside of Colorado. Due to the rarity of flights conducted outside Colorado, only use-density surfaces generated from satellite locations were used to document relative lynx use of areas in New Mexico, Utah and Wyoming.

New Mexico and Wyoming have been used continuously by lynx since the first year lynx were released in Colorado (1999) to the present (Tables 2, 6). Lynx reintroduced in Colorado were first documented in Utah in 2000 (Table 4) and are still being documented there to date. In addition, all levels of lynx use-density documented throughout Colorado are also represented in New Mexico, Utah and Wyoming from none to the highest level of use (Figures 5, 7, 9). One den was found in Wyoming. Although no reproduction has been documented in New Mexico or Utah to date, documenting areas of the highest intensity of use and the continuous presence of lynx within these states for over six years does suggest the potential for year-round residency of lynx and reproduction in those states.

The use-density surface for lynx use in New Mexico indicates the primary areas of use being located immediately south of the Colorado border and south of the Conejos River Valley (an area of high use in Colorado) or east of Taos (Figure 5). In Utah, the primary area of use is located in the Uinta Mountains (Figure 7). Lynx use in Wyoming is focused in 2 primary areas, the Medicine Bow National Forest in south-central Wyoming and in the northwest quadrant of the state including areas in Yellowstone and Teton National Parks and the Laramie Range (Figure 9).

From 1999-June 2007, there were 98 mortalities of released adult lynx. Human-caused mortality factors are currently the highest causes of death with approximately 30.6% attributed to collisions with vehicles or gunshot. Starvation and disease/illness accounted for 19.4% of the deaths while 35.7% of the deaths were from unknown causes. Lynx mortalities were documented throughout all areas lynx used, including 28 (28.6%) occurring in other states (Figure 2, Table10). Half of the out-of-state mortalities were documented in New Mexico.

Reproduction is critical to achieving a self-sustaining viable population of lynx in Colorado. Reproduction was first documented from the 2003 reproduction season and again in 2004, 2005 and 2006. Lower reproduction occurred in 2006 (Table 11) but did include a Colorado-born female giving birth to 2 kittens, documenting the first recruitment of Colorado-born lynx into the Colorado breeding population. No reproduction was documented in 2007. The cause of the decreased reproduction in 2006 and 2007 is unknown. One possible explanation would be a decrease in prey abundance.

Additional reproduction is likely to have occurred in all years from females we were no longer tracking, and from Colorado-born lynx that have not been collared. The dens we find are more representative of the minimum number of litters and kittens in a reproduction season. To achieve a viable population of lynx, enough kittens need to be recruited into the population to offset the mortality that occurs in that year and hopefully even exceed the mortality rate to achieve an increasing population.

The use-density surfaces depict intensity of use by location. Why certain areas would be used more intensively than others should be explained by the quality of the habitat in those areas. Characteristics of areas used by lynx, as documented through aerial locations and snow-tracking of lynx in the Colorado core research area, include mature Engelmann spruce-subalpine fir forest stands with 42-65% canopy cover and 15-20% conifer understory cover (Shenk 2006). Within these forest stand types, lynx appear to have a slight preference for north-facing, moderate slopes ($\bar{x}$ = 15.7°) at high elevations ($\bar{x}$ = 3173 m; Shenk 2006).

Snow-tracking of released lynx also provided information on hunting behavior and diet through documentation of kills, food caches, chases, and diet composition estimated through prey remains. The primary winter prey species (n = 506) were snowshoe hare (Table 12) with an annual $\bar{x}$ = 74.9% (SE = 4.6, n = 9) and red squirrel (annual $\bar{x}$ = 16.5%, SE = 4.1, n = 9). Thus, areas of good habitat must also support populations of snowshoe hare and red squirrel. In winter, lynx reintroduced to Colorado appear to be feeding on their preferred prey species, snowshoe hare and red squirrel in similar proportions as those reported for northern lynx during lows in the snowshoe hare cycle (Aubry et al. 1999). Environmental conditions in the springs and summers of 2003 and 2006 resulted in high cone crops during their following winters based on field observations, resulting in increased red squirrel abundance. This may partially explain the higher percent of red squirrel kills, and thus a lower percent of snowshoe hare kills, found in winters 2003-04 and 2006-07 (Table 12).

Caution must be used in interpreting the proportion of identified kills. Such a proportion ignores other food items that are consumed in their entirety and thus are biased towards larger prey and may not accurately represent the proportion of smaller prey items, such as microtines, in lynx winter diet. Through snow-tracking we have evidence that lynx are mousing and several of the fresh carcasses have yielded small mammals in the gut on necropsy. The summer diet of lynx has been documented to include less snowshoe hare and more alternative prey than in winter (Mowat et al., 1999). All evidence suggests reintroduced lynx are finding adequate food resources to survive.

Mowat et al. (1999) suggest lynx and snowshoe hare select similar habitats except that hares select more dense stands than lynx. Very dense understory limits hunting success of the lynx and provides refugia for hares. Given the high proportion of snowshoe hare in the lynx diet in Colorado, we might then assume the habitats used by reintroduced lynx also depict areas where snowshoes hare are abundant and available for capture by lynx in Colorado. From both aerial locations taken throughout the year and from the site-scale habitat data collected in winter, the most common areas used by lynx are in stands of Engelmann spruce and subalpine fir. This is in contrast to adjacent areas of Ponderosa pine, pinyon juniper, aspen and oakbrush. The lack of lodgepole pine in the areas used by the lynx may be more reflective of the limited amount of lodgepole pine in southwestern Colorado, the Core Release Area, rather than avoidance of this tree species.

Hodges (1999) summarized habitats used by snowshoe hare from 15 studies as areas of dense understory cover from shrubs, stands that are densely stocked, and stands at ages where branches have more lateral cover. Species composition and stand age appears to be less correlated with hare habitat use than is understory structure (Hodges 1999). The stands need to be old enough to provide dense cover and browse for the hares and cover for the lynx. In winter, the cover/browse needs to be tall enough to still provide browse and cover in average snow depths. Hares also use riparian areas and mature forests with understory. Site-scale habitat use documented for lynx in Colorado indicate lynx are most commonly using areas with Engelmann spruce understory present from the snow line to at least 1.5 m above the snow. The mean percent understory cover within the habitat plots is typically less than 15% regardless of understory species. However, if the understory species is willow, percent understory cover is typically double that, with mean number of shrubs per plot approximately 80, far greater than for any other understory species.

In winter, hares browse on small diameter woody stems (<0.25"), bark and needles. In summer, hares shift their diet to include forbs, grasses, and other succulents as well as continuing to browse on woody stems. This shift in diet may express itself in seasonal shifts in habitat use, using more or denser coniferous cover in winter than in summer. The increased use of riparian areas by lynx in Colorado from July to November may reflect a seasonal shift in hare habitat use in Colorado. Major (1989) suggested lynx hunted the edge of dense riparian willow stands. The use of these edge habitats may allow lynx to hunt hares that live in habitats normally too dense to hunt effectively. The use of riparian areas and riparian-Engelmann spruce-subalpine fir and riparian-aspen mixes documented in Colorado may stem from a similar hunting strategy. However, too little is known about habitat use by hares in Colorado to test this hypothesis at this time.

Lynx also require sufficient denning habitat. Denning habitat has been described by Koehler (1990) and Mowat et al. (1999) as areas having dense downed trees, roots, or dense live vegetation. We found this to be in true in Colorado as well (Shenk 2006). In addition, the dens used by reintroduced lynx were at high elevations and on steep north-facing slopes. All females that were documented with kittens denned in areas within their winter-use area.

SUMMARY

From results to date it can be concluded that CDOW developed release protocols that ensure high initial post-release survival of lynx, and on an individual level, lynx demonstrated they can survive long-term in areas of Colorado. We also documented that reintroduced lynx exhibited site fidelity, engaged in breeding behavior and produced kittens that were recruited into the Colorado breeding population. What is yet to be demonstrated is whether current conditions in Colorado can support the recruitment necessary to offset annual mortality in order to sustain the population. Monitoring of reintroduced lynx will continue in an effort to document such viability.

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Prepared by ____________________________________
Tanya M. Shenk, Wildlife Researcher

Table 1. Lynx released in Colorado from February 1999 through June 30, 2007. No lynx were released in 2001, 2002 or 2007.

Year	Females	Males	TOTAL
1999	22	19	41
2000	35	20	55
2003	17	16	33
2004	17	20	37
2005	18	20	38
2006	6	8	14
TOTAL	115	103	218

Table 2. All individual lynx (n = 60) documented through either aerial or satellite locations (non-truncated datasets) by year in New Mexico from February 1999 – March 2007.

	Year								
Lynx ID	1999	2000	2001	2002	2003	2004	2005	2006	2007
AK99F10	X								
AK99F13		X							
AK99F17	X								
AK99F3		X							
AK99F5					X		X		
AK99F8	X								
AK99M11		X							
AK99M26	X								
AK99M9		X							
BC99M4	X			X					
YK99F3		X							
YK99M3	X								
YK99M6	X	X							
YK99M7	X								
AK00F2								X	X
AK00F5							X		
BC00F10		X							
BC00F14				X					
BC00F6		X							
BC00F8		X	X			X	X		
BC00M04								X	
BC00M11		X						X	
BC00M4		X						X	
YK00F11							X		
YK00F2				X	X				
YK00F4			X						
YK00F7								X	
BC03F03					X	X			
BC03F04					X				
BC03F06						X			
BC03F08					X				
BC03M02					X	X			
BC03M05					X				
BC03M08						X	X	X	
QU03F01					X				
QU03F04						X	X		
QU03F07					X	X	X		
BC04F02						X	X		
BC04F03						X			
BC04F05						X			
BC04M02						X			
BC04M13						X	X		
QU04F05						X	X		
QU04F08						X			
QU04F09						X			
QU04M02						X			
QU04M04						X			
BC05F04							X		
BC05M02							X		
QU05F03							X		
QU05M01							X		
QU05M05							X	X	
YK05F01							X		
YK05M01							X	X	
BC06F05								X	
BC06F07								X	X
BC06F09								X	
BC06M12								X	
YK06F01								X	
YK06M01								X	
Total Lynx	8	11	2	3	9	17	17	14	2

Table 3. All individual lynx (*n* = 35) documented at least 180 days after their initial release (truncated datasets) through either aerial or satellite locations, by year in New Mexico from September 1999 – March 2007.

	Year							
Lynx ID	2000	2001	2002	2003	2004	2005	2006	2007
AK99F13	X							
AK99F3	X							
AK99F5				X		X		
AK99M11	X							
AK99M9	X							
BC99M4			X					
YK99F3	X							
YK99M6	X							
AK00F2							X	X
AK00F5						X		
BC00F14			X					
BC00F8		X			X	X		
BC00M04							X	
BC00M11							X	
BC00M4							X	
YK00F11						X		
YK00F2			X	X				
YK00F4		X						
YK00F7							X	
BC03F03					X			
BC03F06					X			
BC03M02					X			
BC03M08					X	X	X	
QU03F04					X	X		
QU03F07					X	X		
BC04F02					X	X		
BC04M13					X	X		
QU04F05					X	X		
QU04F08					X			
QU04F09					X			
QU05M05							X	
YK05M01							X	
BC06F07							X	X
BC06M12							X	
YK06F01							X	
Total Lynx	6	2	3	2	12	10	11	2

Table 4. All individual lynx ($n = 22$) documented through either aerial or satellite locations (non-truncated datasets) by year in Utah from February 1999 – March 2007.

Lynx ID	Year							
	2000	2001	2002	2003	2004	2005	2006	2007
AK99F5						X		
AK00F5								X
AK00M3		X						
BC00M09					X			
BC00M13	X							
YK00F7							X	X
BC03F03				X				
BC03M06								X
BC03M08				X				
BC03M10								X
QU03F03					X	X		
BC04M01					X	X		
QU04M04							X	
QU04M05					X			
BC05M01								X
BC05M03						X	X	
CO05F20							X	
QU05F05						X	X	
QU05M03						X		
QU05M08						X		
YK05M01							X	
YK06M01							X	
Total Lynx	1	1	0	2	4	7	7	5

Table 5. All individual lynx ($n = 17$) documented at least 180 days after their initial release (truncated datasets) through either aerial or satellite locations, by year in Utah from September 1999 – March 2007.

Lynx ID	Year							
	2000	2001	2002	2003	2004	2005	2006	2007
AK99F5						X		
AK00F5								X
AK00M3		X						
BC00M09					X			
YK00F7							X	X
BC03M06								X
BC03M10								X
QU03F03					X	X		
BC04M01					X	X		
QU04M04							X	
BC05M01								X
BC05M03						X	X	
CO05F20							X	
QU05F05						X	X	
QU05M03						X		
YK05M01							X	
YK06M01							X	
Total Lynx	0	1	0	0	3	6	7	5

Table 6. All individual lynx (n = 33) documented through either aerial or satellite locations (non-truncated datasets) by year in Wyoming from February 1999 – March 2007.

Lynx ID	Year						
	1999	2001	2003	2004	2005	2006	2007
AK99M6	X						
BC00F14			X	X			
BC00M13		X		X			
YK00F11						X	
BC03F03				X			
BC03M02				X			
BC03M06				X			
BC03M09			X	X			
QU03M01				X			
BC04F02					X		
BC04M01				X			
BC04M08				X	X	X	
BC04M13					X		
CO04F10				X			
CO04M05				X			
CO04M06				X			
QU04F01				X	X	X	
QU04F02				X	X	X	X
QU04F07				X			
QU04M04					X	X	
QU04M05				X	X	X	
BC05M03					X	X	
BC05M08					X		
MB05F01						X	
MB05F02						X	X
MB05F03						X	
QU05F04					X	X	X
QU05F05					X	X	
QU05F08					X	X	X
QU05M08					X	X	
YK05M03					X		
BC06M10						X	
BC06M13						X	X
Total Lynx	1	1	2	16	14	16	5

Table 7. All individual lynx (n = 27) documented at least 180 days after their initial release (truncated datasets) through aerial or satellite locations, by year in Wyoming from September 1999 – March 2007.

	Year					
Lynx ID	2001	2003	2004	2005	2006	2007
BC00F14		X	X			
BC00M13	X		X			
YK00F11					X	
BC03F03			X			
BC03M02			X			
BC03M06			X			
BC03M09		X	X			
QU03M01			X			
BC04F02				X		
BC04M08			X	X	X	
BC04M13				X		
CO04F10			X			
CO04M05			X			
CO04M06			X			
QU04F01			X	X	X	
QU04F02			X	X	X	X
QU04M04				X	X	
QU04M05			X	X	X	
BC05M03					X	
MB05F01					X	
MB05F02					X	X
MB05F03					X	
QU05F04				X	X	X
QU05F05				X	X	
QU05F08				X	X	X
QU05M08				X	X	
BC06M13					X	X
Total Lynx	1	2	14	11	15	5

Table 8. Status of adult lynx reintroduced to Colorado as of June 30, 2007.

Lynx	Females	Males	Unknown	TOTALS
Released	115	103		218
Known Dead	54	43	1	98
Possible Alive	61	60		120
Missing	23	27		49[a]
Monitoring/tracking	38	33		71

[a] 1 is unknown mortality

Table 9. Causes of death for all lynx released into southwestern Colorado 1999-2006 as of June30, 2007.

Cause of Death	Mortalities		
	Total (%)	In Colorado (%)	Outside Colorado (%)
Unknown	35 (35.7)	20 (57.1)	15 (42.9)
Gunshot	13 (13.3)	7 (53.8)	6 (46.2)
Hit by Vehicle	12 (12.2)	8 (66.7)	4 (33.3)
Starvation	10 (10.2)	9 (90.0)	1 (10.0)
Other Trauma	8 (8.1)	7 (87.5)	1 (12.5)
Plague	7 (7.1)	7 (100)	0 (0)
Probable Gunshot	5 (5.1)	4 (80)	1 (20)
Predation	3 (3.1)	3 (100)	0 (0)
Probable Predation	3 (3.1)	3 (100)	0 (0)
Illness	2 (2.0)	2 (100)	0 (0)
Total Mortalities	98	70 (71.4)	28 (28.6)

Table 10. Known lynx mortalities (n = 28) and causes of death documented by state outside of Colorado from February 1999 – June 30, 2007.

Lynx ID	State	Date Mortality Recorded	Cause of Death
AK99F8	New Mexico	7/30/1999	Starvation
Unknown	New Mexico	2000	Hit by Vehicle
AK99M11	New Mexico	1/27/2000	Unknown
YK99M06	New Mexico	6/19/2000	Probable Gunshot
AK99F13	New Mexico	6/22/2000	Unknown
YK00F04	New Mexico	4/20/2001	Gunshot
BC99M04	New Mexico	6/7/2002	Gunshot
QU05M01	New Mexico	8/22/2005	Unknown
QU04F05	New Mexico	8/26/2005	Hit by Vehicle
QU03F07	New Mexico	9/15/2005	Unknown
BC00M04	New Mexico	7/19/2006	Unknown
YK06F01	New Mexico	10/19/2006	Unknown
BC03M08	New Mexico	10/19/2006	Unknown
BC06F07	New Mexico	1/8/2007	Gunshot
AK99M06	Nebraska	11/16/1999	Gunshot
AK99M01	Nebraska	1/11/2005	Snared (Other Trauma)
QU05M08	Nebraska	10/1/2006	Unknown
MB05F02	Nebraska	2/13/2007	Gunshot
BC00F14	Wyoming	7/28/2004	Unknown
QU04F07	Wyoming	9/21/2004	Unknown
BC06M10	Wyoming	8/15/2006	Vehicle Collision
QU04F02	Wyoming	3/14/2007	Unknown
AK00M03	Utah	7/2/2001	Unknown
QU05M03	Utah	10/26/2005	Unknown
YK06M01	Utah	12/4/2006	Unknown
YK99F01	Arizona	9/15/2005	Gunshot
YK00M03	Kansas	9/30/2005	Vehicle Collision
YK05M03	Montana	11/8/2005	Unknown

Table 11. Lynx reproduction summary statistics for 2003-2007. No reproduction was documented from 1999-2002 or in 2007.

Year	Females Tracked	Dens Found in May/June	Percent Tracked Females with Kittens	Additional Litters Found in Winter	Mean Kittens Per Litter (SE)	Total Kittens Found	Sex Ratio M/F (SE)
2003	17	6	0.353		2.67 (0.33)	16	1.0
2004	26	11	0.462	2	2.83 (0.24)	39	1.5
2005	40	17	0.425	1	2.88 (0.18)	50	0.8
2006	42	4	0.095		2.75 (0.47)	11	1.2
2007	34	0	0.0			0	
Total						116	1.14 (0.14)

Table 12. Lynx captured because they were in poor body condition or were in atypical habitat and their fates 6 months post re-release and as of June 30, 2007.

Lynx ID	Date of Capture	State Where Captured	Reason For Capture	Date of Re-release	Status 6 Months Post Re-release	Current Status
BC99F6	3/25/1999	Colorado	Poor body condition	5/28/1999	Dead	Died 7/19/1999 in Colorado from vehicle collision
AK99M9	3/24/2000	Colorado	Poor body condition	5/3/2000	Missing	Last located 5/3/2000, collar failure
AK99F2	4/18/2000	Colorado	Poor body condition	5/22/2000	Alive in Colorado	Last located 7/30/2003 in Colorado
BC00F7	2/11/2001	Colorado	Poor body condition	N/A	Dead	Died at Rehab Center on 2/12/2001
BC00M13	3/21/2001	Wyoming	Poor body condition	4/24/2001	Alive in Colorado	Last located 10/26/2004 in Colorado
BC03M08	9/5/2003	Colorado	Poor body condition	1/1/2004	Alive in Colorado	Died in New Mexico of unknown causes 10/19/06
QU04M07	2/2/2006	Colorado	Poor body condition	N/A	Dead	Died at Rehab Center on 2/5/2006 from hydrocephalous and pneumonia
BC04M01	11/5/2004	Utah	Atypical habitat	12/5/2004	Alive in Colorado	In Colorado as of 6/30/2007
QU04F02	4/10/2005	Nebraska	Atypical habitat	5/7/2005	Alive in Wyoming	Died 3/14/2007 in Wyoming (good habitat) of unknown causes
QU05M08	11/25/2005	Wyoming	Atypical habitat	4/18/2006	Dead	Died of unknown causes in Nebraska 10/1/2006
QU04M04	12/5/2006	Utah	Atypical habitat	1/20/2007	Dead in Colorado	Died of starvation in Colorado, found 3/19/07
YK00F7	12/12/2006	Utah	Atypical habitat	1/20/2007	Alive in Utah	In Utah as of 6/30/2007
YK05M02	1/1/2007	Kansas	Atypical habitat	2/2/2007	Alive in Iowa	In Iowa as of 6/30/2007
BC04M08	1/22/2007	Wyoming	Atypical habitat	2/15/2007	Alive in Colorado	In Colorado as of 6/30/2007

Table 13. Number of kills found each winter field season through snow-tracking of lynx and percent composition of kills of the three primary prey species.

		Prey (%)			
Field Season	n	Snowshoe Hare	Red Squirrel	Cottontail	Other
1999	9	55.56	22.22	0	22.22
1999-2000	83	67.47	19.28	1.20	12.05
2000-2001	89	67.42	19.10	8.99	4.49
2001-2002	54	90.74	5.56	0	3.70
2002-2003	65	90.77	6.15	0	3.08
2003-2004	37	67.57	27.03	2.70	2.70
2004-2005	78	83.33	10.26	0	6.41
2005-2006	50	90.00	0.08	0	0.02
2006-2007	41	61.00	39.0	0	0

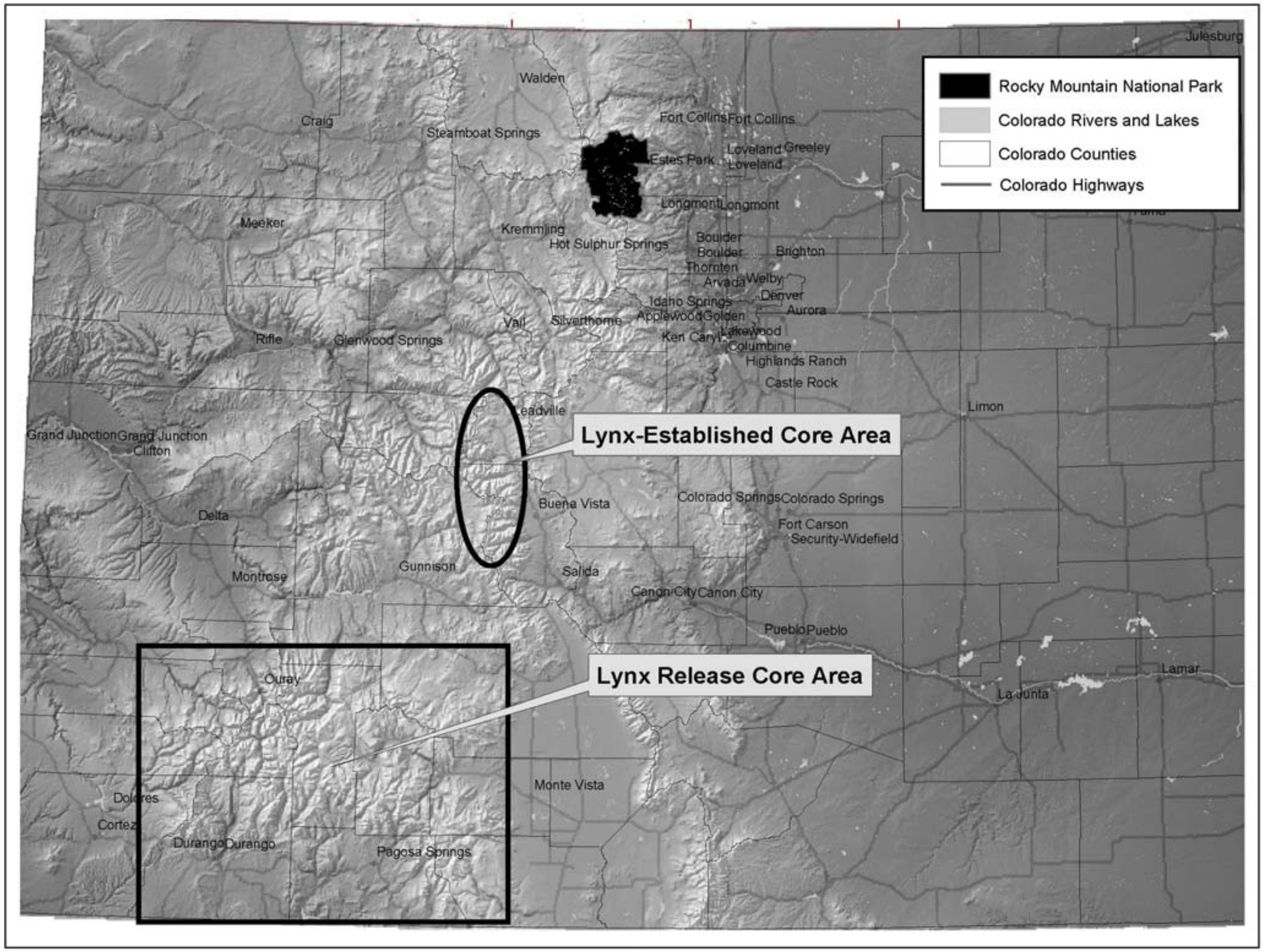

Figure 1. Lynx are monitored throughout Colorado and by satellite throughout the western United States. The lynx core release area, where all lynx were released, is located in southwestern Colorado. A lynx-established core use area has developed in the Taylor Park and Collegiate Peak area in central Colorado.

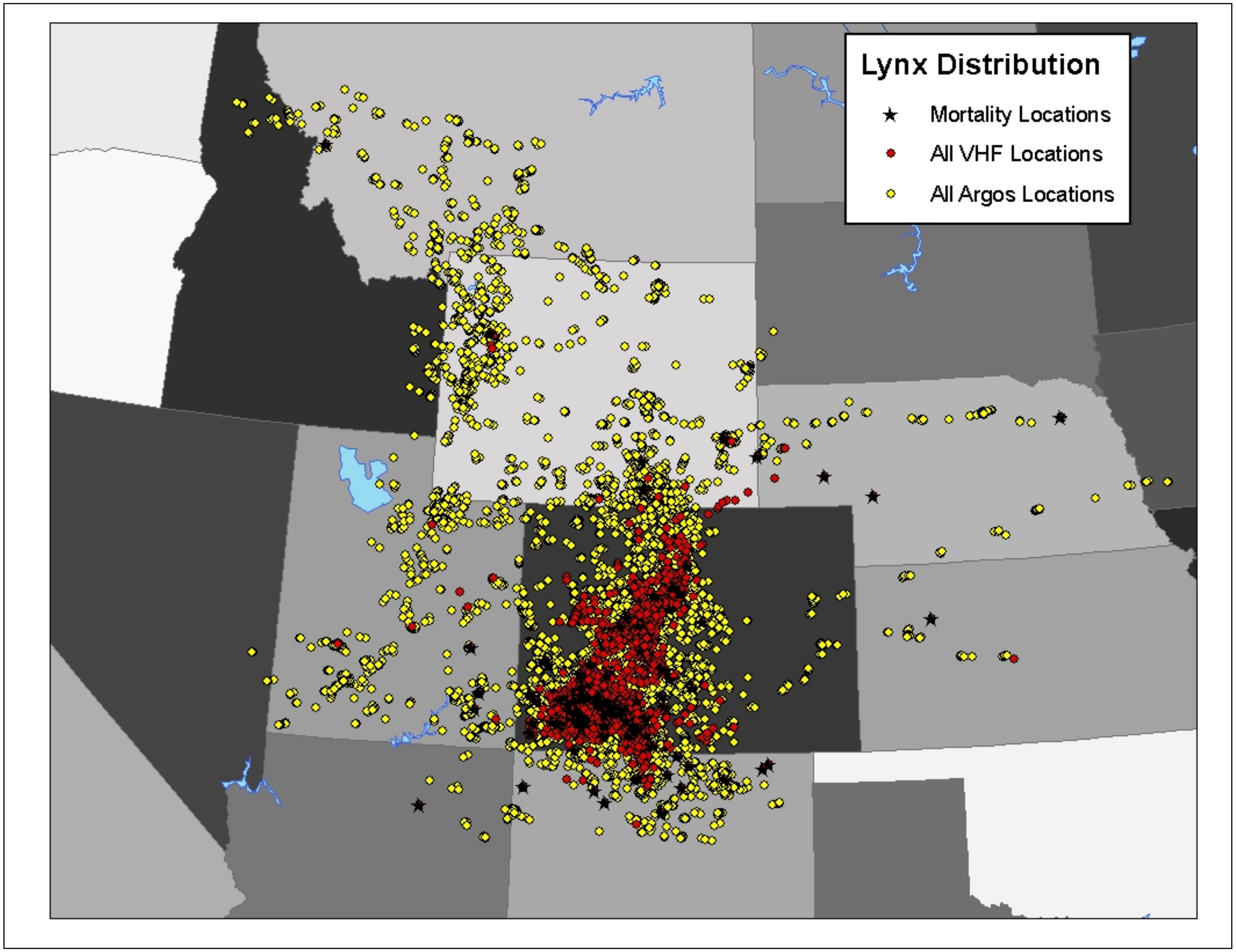

Figure 2. All documented lynx locations (non-truncated datasets) obtained from either aerial (yellow circles) or satellite (red circles) tracking from February 1999 through June 30, 2007. All known lynx mortality locations (*n* = 97) are displayed as stars.

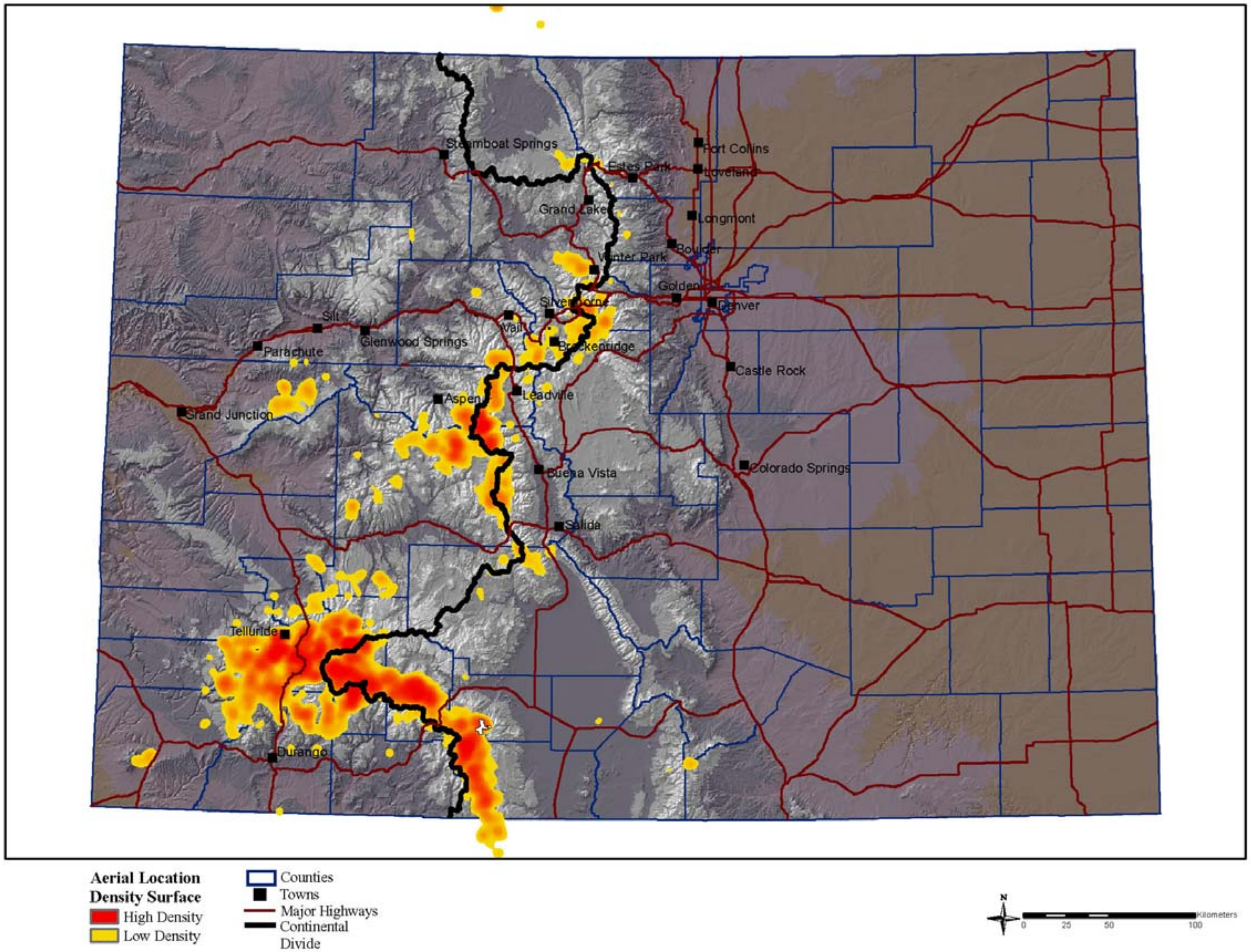

Figure 3. Use-density surface for lynx aerial locations (truncated dataset) in Colorado from September 1999-March 2007

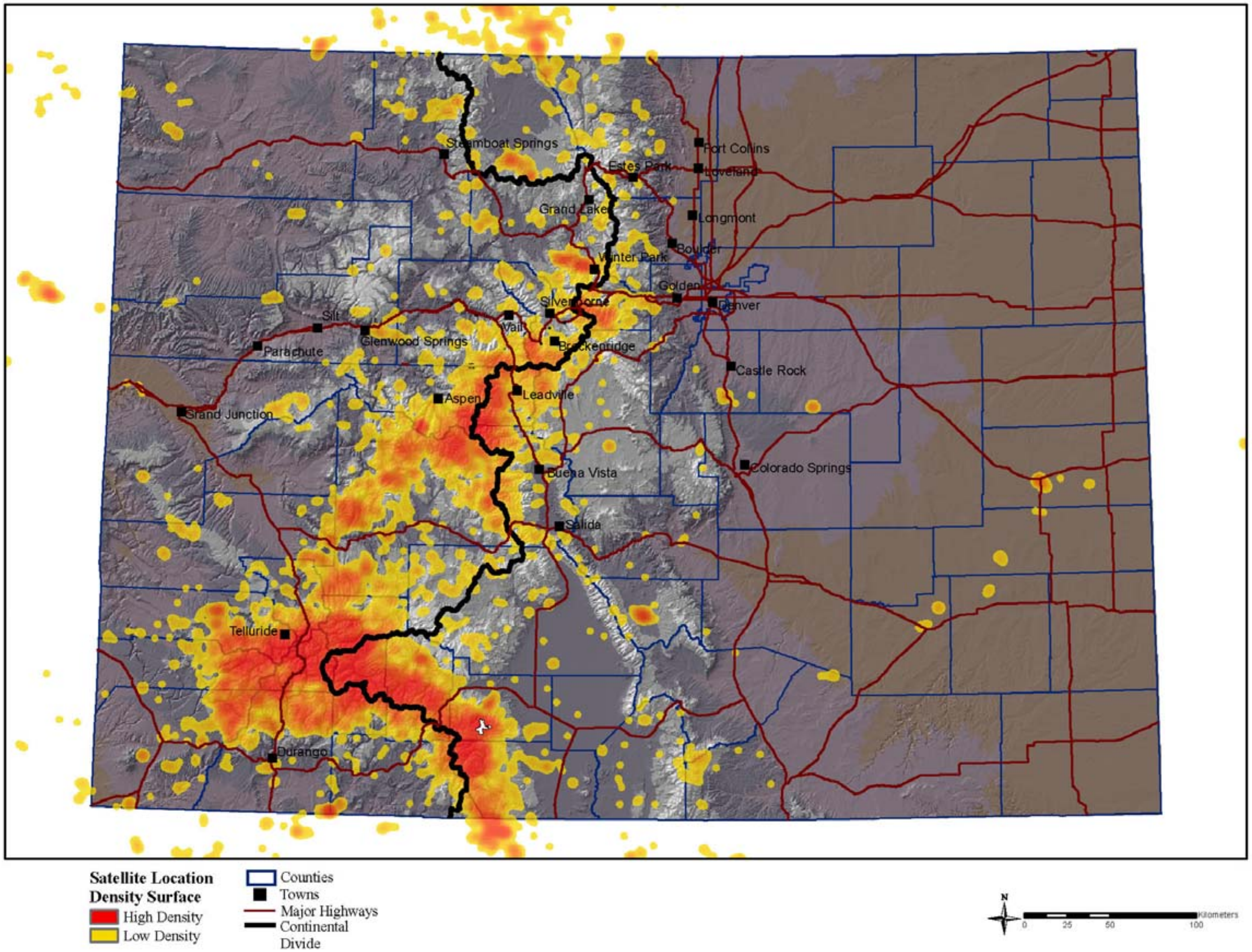

Figure 4. Use-density surface for lynx satellite locations (truncated dataset) in Colorado from September 1999-March 2007.

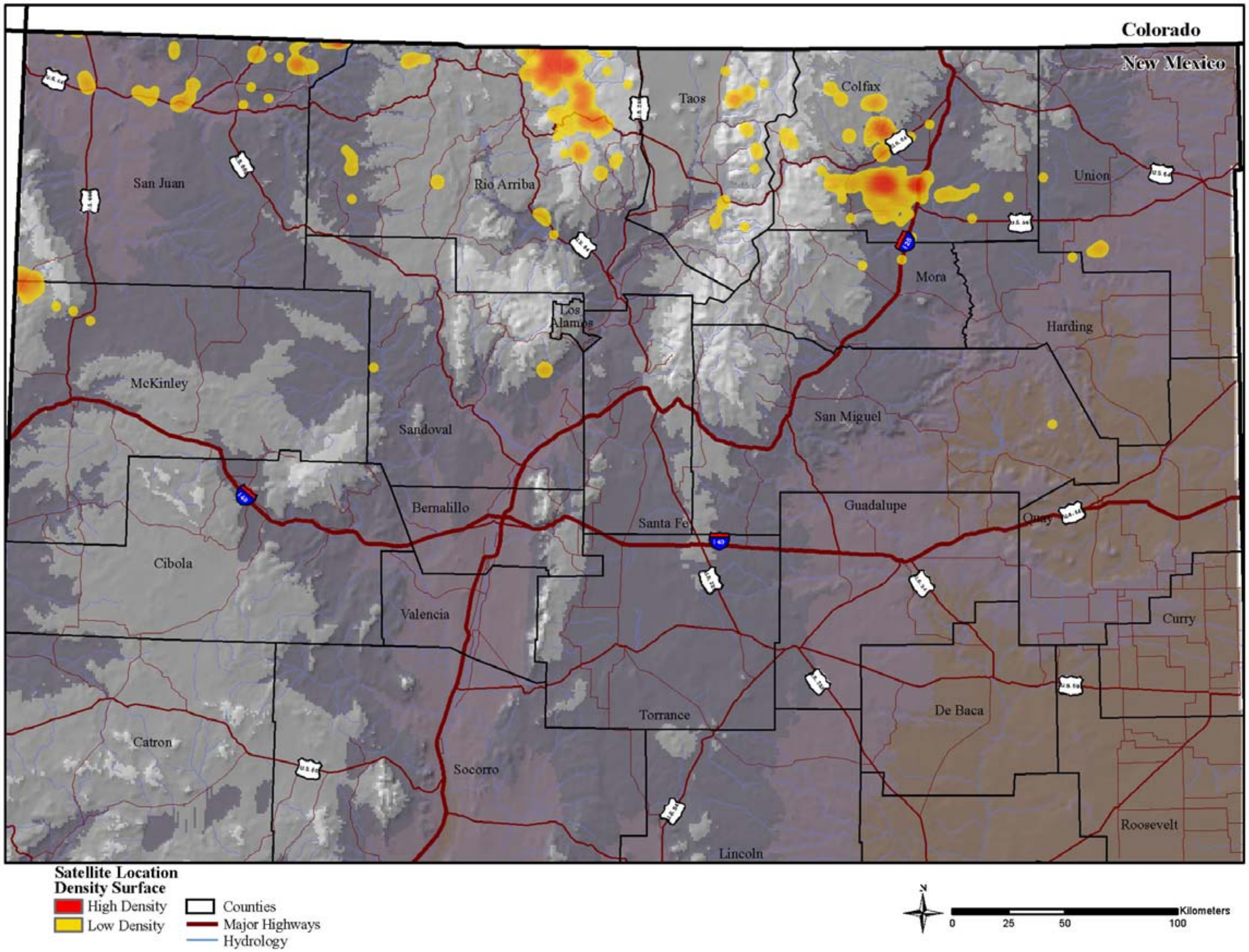

Figure 5. Use-density surface for lynx satellite locations (truncated dataset) in New Mexico from September 1999-March 2007

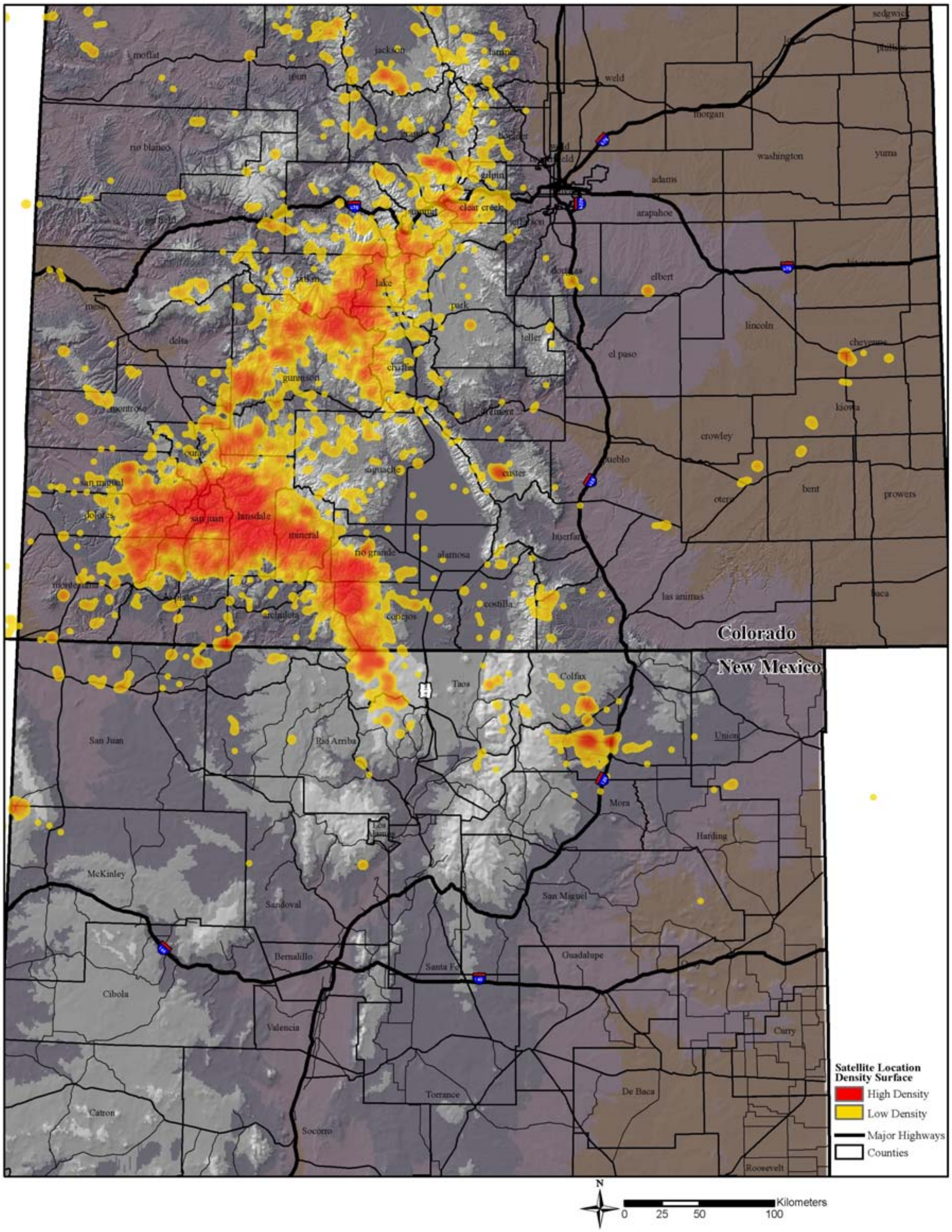

Figure 6. Use-density surface for lynx satellite locations (truncated dataset) in Colorado and New Mexico from September 1999-March 2007.

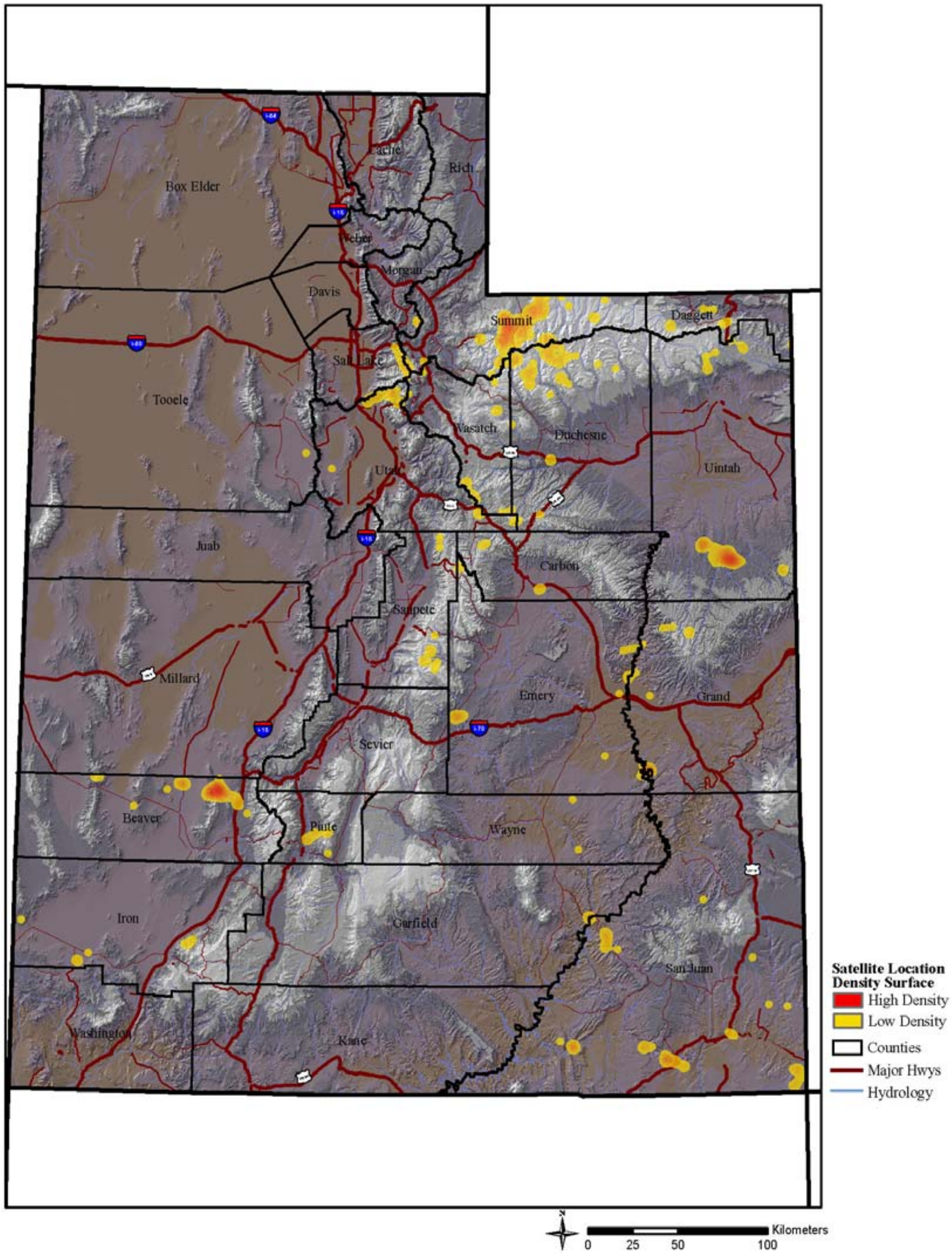

Figure 7. Use-density surface for lynx satellite locations (truncated dataset) for Utah from September 1999-March 2007.

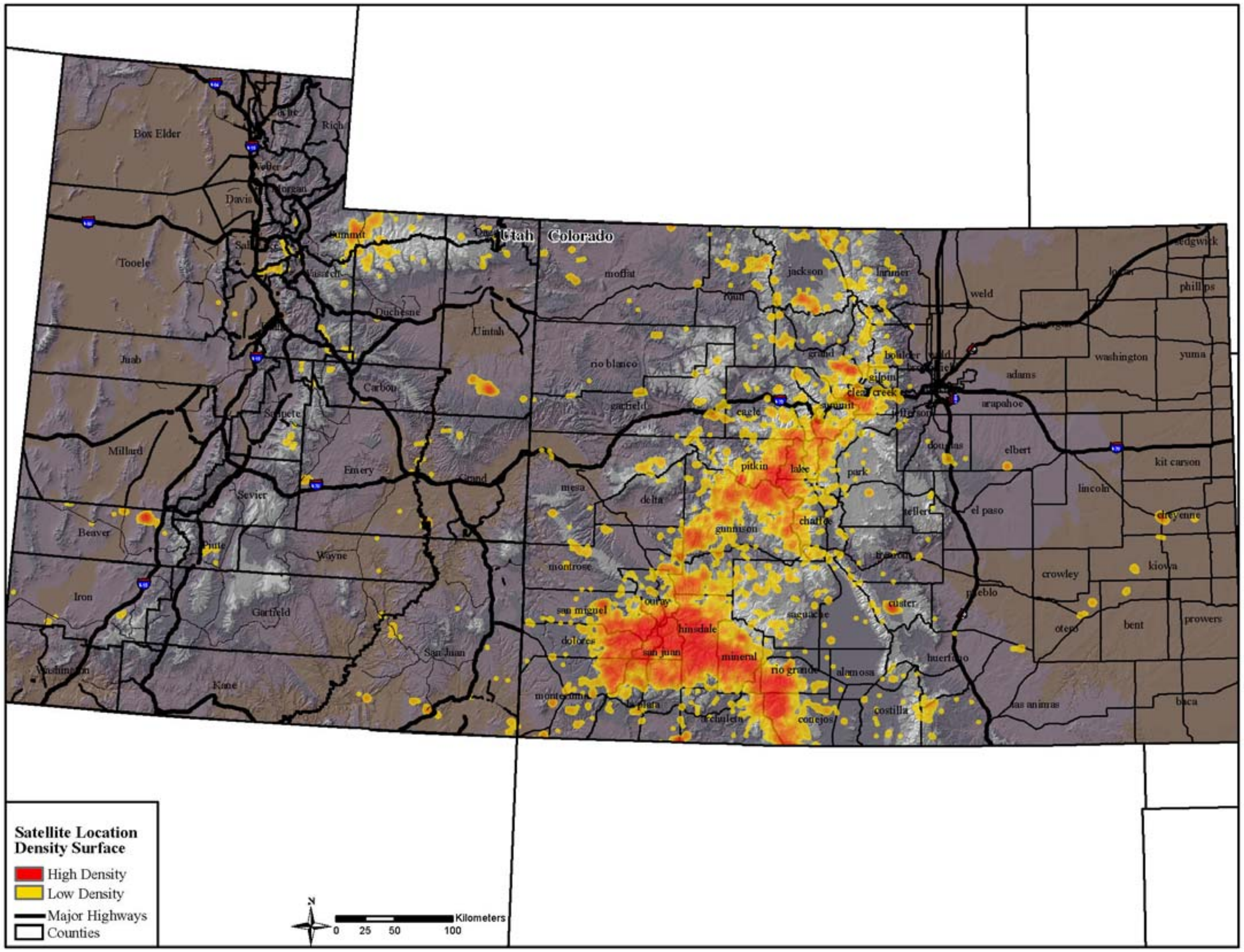

Figure 8. Use-density surface for lynx satellite locations (truncated dataset) in Colorado and Utah from September 1999-March 2007.

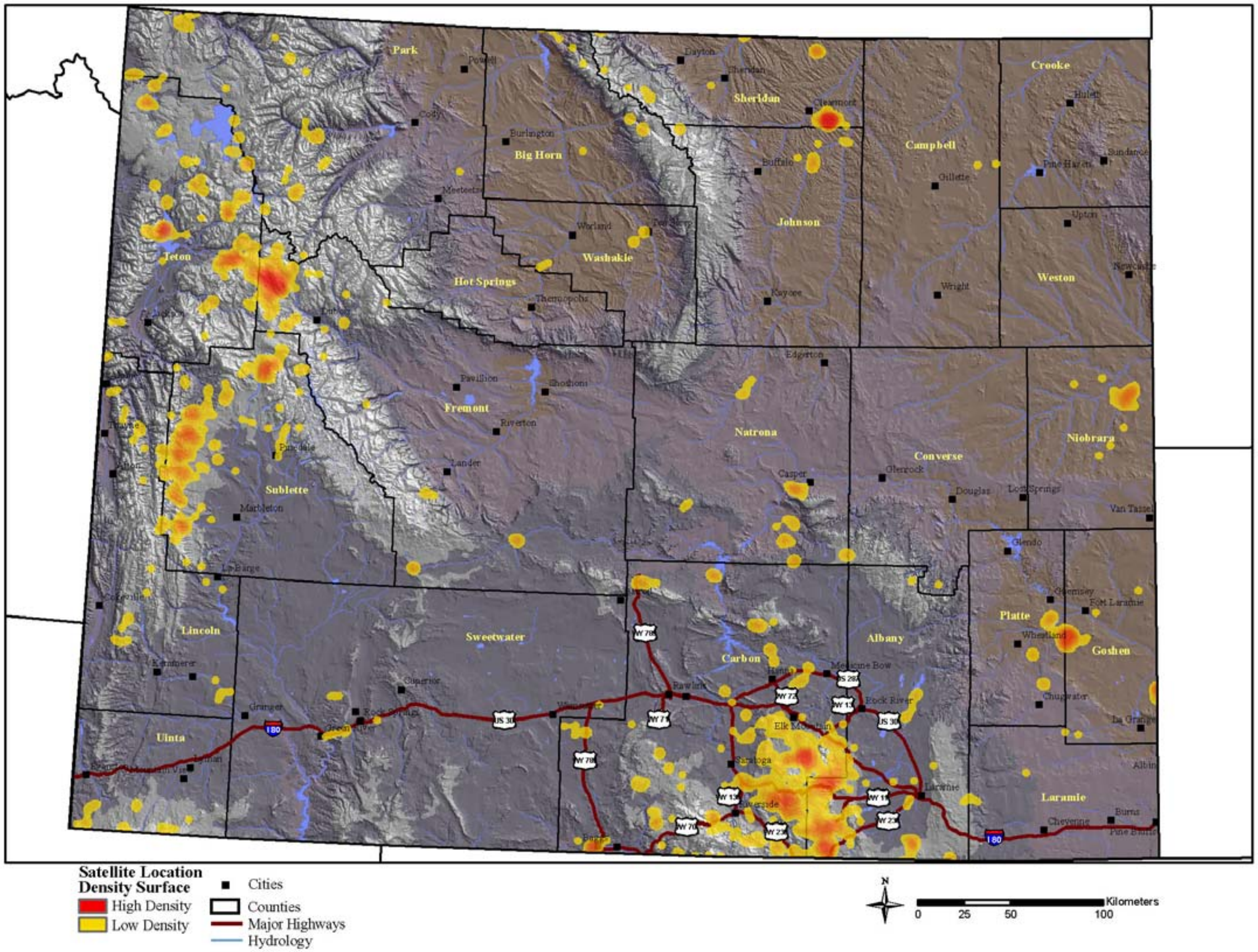

Figure 9. Use-density surface for lynx satellite locations (truncated dataset) in Wyoming from September 1999-March 2007.

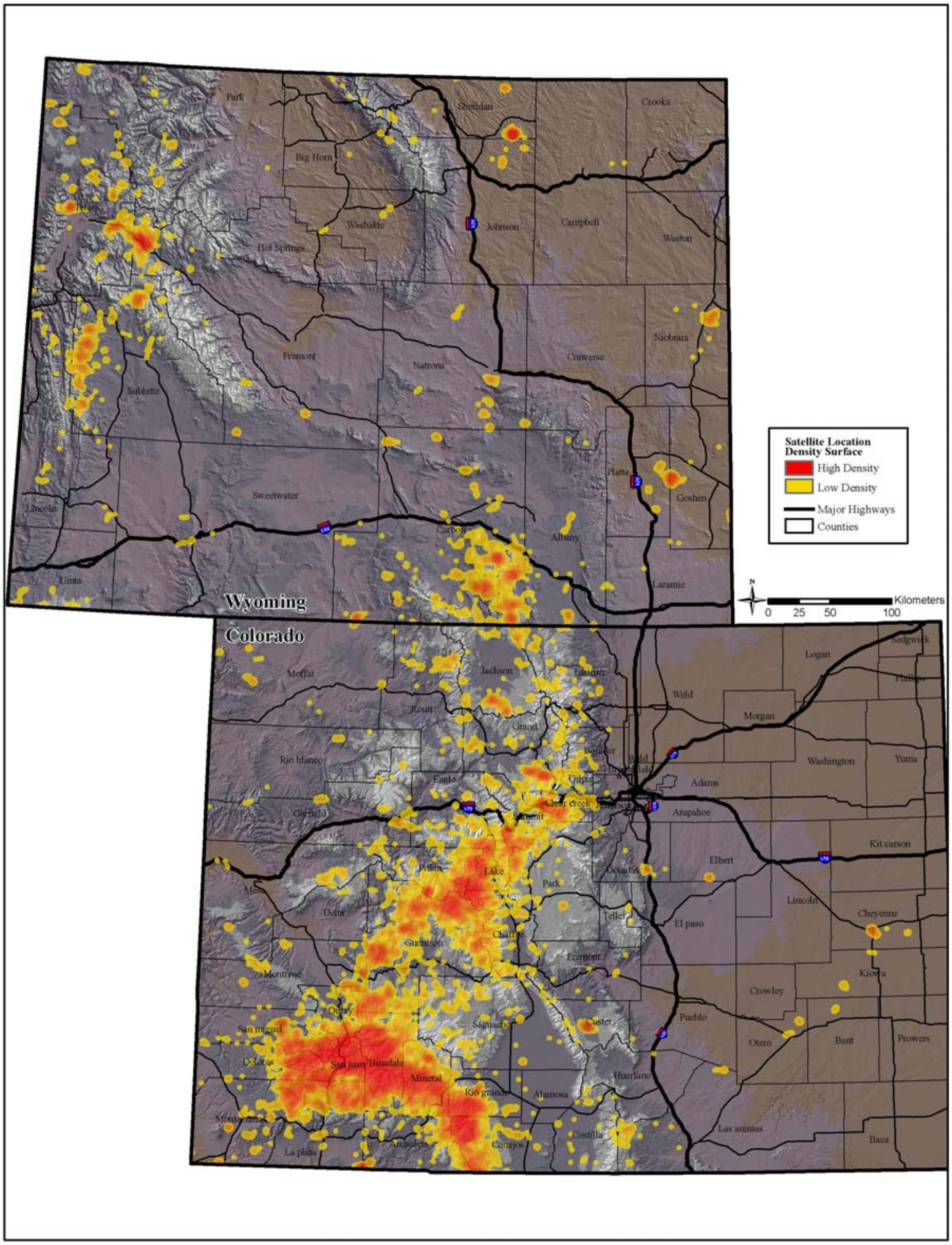

Figure 10. Use-density surface for lynx satellite locations (truncated dataset) in Colorado and Wyoming from September 1999-March 2007.

APPENDIX I

Colorado Division of Wildlife
July 2006 - June 2007

WILDLIFE RESEARCH REPORT

State of	Colorado	: Division of Wildlife
Cost Center	3430	: Mammals Research
Work Package	0670	: Lynx Conservation
Task No.	2	: Density, Demography, and Seasonal Movements of Snowshoe Hare in Colorado
Federal Aid Project:	N/A	:

Period Covered: July 1, 2006- June 30, 2007

Author: J. S. Ivan, Ph. D. Candidate, Colorado State University

Personnel: T. M. Shenk, CDOW and G. C. White of Colorado State University.

All information in this report is preliminary and subject to further evaluation. Information MAY NOT BE PUBLISHED OR QUOTED without permission of the author. Manipulation of these data beyond that contained in this report is discouraged.

ABSTRACT

A program to reintroduce the threatened Canada lynx (*Lynx canadensis*) into Colorado was initiated in 1997 with the first lynx release in 1999. Analysis of scat collected from winter snow tracking indicated that snowshoe hares (*Lepus americanus*) comprised 65–90% of the winter diet of reintroduced lynx. Thus, existence of lynx in Colorado and success of the reintroduction effort hinge at least in part on maintaining adequate and widespread populations of hares. Beginning in July 2006, I initiated a study to assess the relative value of 3 forest stand types (mature [“large”] spruce/fir, sapling [“small”] lodgepole pine, pole-sized [“medium”] lodgepole pine) that purportedly provide high quality hare habitat in Colorado. Estimates and comparisons of survival, recruitment, finite population growth rate, and maximum (late summer) and minimum (late winter) snowshoe hare densities for each stand will provide the metrics for assessing value. Number of individuals captured, number of captures, and number of locations obtained per hare during the first year of the project appear adequate for attaining the objectives of this study. Some hare deaths due to capture myopathy (most likely cause) occurred during initial trapping periods in both the summer and winter sampling seasons. However, changes to the trapping protocol, trapping schedule, and bait provided seem to have alleviated the problem. Densities during summer were highest in small lodgepole stands (0.47 hares/ha, 95% CI: 0.41-0.54), followed by large spruce/fir (0.18 hares/ha, 95% CI: 0.12-0.25) and medium lodgepole (0.02 hares/ha, 95% CI: 0.01-0.03). During winter, densities in small lodgepole stands dropped and became more variable across replicates (0.18 hares/ha, 95% CI: 0.01-0.35). Medium lodgepole stands gained hares (0.07 hares/ha, 95% CI: 0.05-0.10). Spruce/fir stands remained at the same density as during summer (0.17 hares/ha, 95% CI: 0.11-0.23).

WILDIFE RESEARCH REPORT

DENSITY, DEMOGRAPHY, AND SEASONAL MOVEMENTS OF SNOWSHOE HARE IN COLORADO

JACOB S. IVAN

P. N. OBJECTIVE

Assess the relative value of 3 forest stand types (old spruce/fir, sapling lodgepole, pole-sized lodgepole) that purportedly provide high quality snowshoe hare (*Lepus americanus*) habitat by estimating survival, recruitment, finite population growth rate, and maximum (late summer) and minimum (late winter) snowshoe hare densities for each type.

SEGMENT OBJECTIVES

1. Complete mark-recapture work across all replicate stands during late summer (mid-July through mid-September) and winter (mid-January through March).
2. Obtain daily telemetry locations on radio-tagged hares for 10 days immediately after capture periods, as well as monthly between primary trapping sessions.
3. Locate, retrieve, and refurbish radio tags as mortalities occur.
4. Summarize initial sampling efforts and provide initial density estimates for Wildlife Research Reports for Colorado Division of Wildlife (CDOW).

INTRODUCTION

A program to reintroduce the threatened Canada lynx (*Lynx canadensis*) into Colorado was initiated in 1997 with the first lynx released in 1999. Since that time, 204 lynx have been released in the state, and an extensive effort to determine their movements, habitat use, reproductive success, and food habits has ensued (Shenk 2006). Analysis of scat collected from winter snow tracking indicates that snowshoe hares (*Lepus americanus*) comprise 65–90% of the winter diet of reintroduced lynx (T. Shenk, Colorado Division of Wildlife, unpublished data). Thus, as in the far north where the intimate relationship between lynx and snowshoe hares has captured the attention of ecologists for decades, the existence of lynx in Colorado and success of the reintroduction effort may also hinge on maintaining adequate and widespread populations of hares.

Colorado represents the extreme southern range limit for both lynx and snowshoe hares (Hodges 2000). At this latitude, habitat for each species is less widespread and more fragmented compared to the continuous expanse of boreal forest at the heart of lynx and hare ranges. Neither species exhibits dramatic cycles as occur farther north, and typical lynx (≤2–3 lynx/100km^2; Aubry et al. 2000) and hare (≤1–2 hares/ha; Hodges 2000) densities in the southern part of their range correspond to cyclic lows form northern populations (2-30 lynx/100 km^2, 1–16 hares/ha; Aubry et al. 2000, Hodges 2000, Hodges et al. 2001).

Whereas extensive research on lynx-hare ecology has occurred in the boreal forests of Canada, literature regarding the ecology of these species in the southern portion of their range is relatively sparse. This scientific uncertainty is acknowledged in the “Canada Lynx Conservation Assessment and Strategy,” a formal agreement between federal agencies intended to provide a consistent approach to lynx conservation on public lands in the lower 48 states (Ruediger et al. 2000). In fact, one of the explicit guiding principles of this document is to “retain future options…until more conclusive information concerning lynx management is developed.” Thus, management recommendations in this agreement are decidedly conservative, especially with respect to timber management, and are applied broadly to cover all habitats thought to be of possible value to lynx and hare. This has caused controversy where recommendations conflict with competing resource management goals. Accurate identification and detailed description of lynx-hare habitat in the southern Rocky Mountains would permit more informed and refined management recommendations.

A commonality throughout the snowshoe hare literature, regardless of geographic location, is that hares are associated with dense understory vegetation that provides both browse and protection from elements and predators (Wolfe et al. 1982, Litvaitis et al. 1985, Hodges 2000, Homyack et al. 2003, Miller 2005). In western mountains, this understory can be provided by relatively young conifer stands regenerating after stand-replacing fires or timber harvest (Sullivan and Sullivan 1988, Koehler 1990, Koehler 1990, Bull et al. 2005) as well as mature, uneven-aged stands (Beauvais 1997, Griffin 2004). Hares may also take advantage of seasonally abundant browse and cover provided by deciduous, open habitats (e.g., riparian willow [*Salix* spp.], aspen [*Populus tremuloides*]; Wolff 1980, Miller 2005). In drier portions of hare range, such as Colorado, regenerating stands can be relatively sparse, and hares may be more associated with mesic, late-seral forest and/or riparian areas than with young stands (Ruggiero et al. 2000).

Numerous investigators have sought to determine the relative importance of these distinctly different habitat types with regards to snowshoe hare ecology. Most previous evaluations were based on hare density or abundance (Bull et al. 2005), indices to hare density and abundance (Wolfe et al. 1982, Koehler 1990, Beauvais 1997, Miller 2005), survival (Bull et al. 2005), and/or habitat use (Dolbeer and Clark 1975). Each of these approaches provides insight into hare ecology, but taken singly, none provide a complete picture and may even be misleading. For example, extensive use of a particular habitat type may not accurately reflect the fitness it imparts on individuals, and density can be high even in "sink" habitats (Van Horne 1983). A more informative approach would be to measure density, survival, and habitat use simultaneously in addition to recruitment and population growth rate through time. Griffin (2004) employed such an approach and found that summer hare densities were consistently highest in young, dense stands. However, he also noted that only dense mature stands held as many hares in winter as in summer. Furthermore hare survival seemed to be higher in dense mature stands, and only dense mature stands were predicted (by matrix projection) to impart a mean positive population growth rate on hares. Griffin's (2004) study occurred in the relatively moist forests of Montana, which share many similarities but also many notable differences with Colorado forests including levels of fragmentation, species composition, elevation, and annual precipitation.

Density estimation is a key component in assessing the value of a particular stand type and is the common currency by which hare populations are compared across time and space. However, density can be a difficult metric to estimate accurately. Density estimation based on capture-recapture methods is a well-developed field (Otis et al. 1978, White et al. 1982), but is often too costly and labor intensive to be implemented on scales necessary to effectively monitor density over a biologically meaningful area. Also, density can be difficult to assess from grid-trapping efforts because it is often unclear how much area was effectively sampled by the grid (Williams et al. 2002:314). Different approaches can produce density estimates that differ by an order of magnitude even when calculated from the same data (Zahratka 2004). Indices such as pellet plot counts and distance sampling of pellet groups can be used to estimate density, but each of these has limitations as well (Krebs et al. 1987, Eriksson 2006).

Pellet plot counts are typically conducted by laying out numerous rectangular or circular plots along transect lines randomly placed within a study site. All pellets occurring within the plot are counted and removed on an annual basis. The mean number of pellets per plot is then inserted into a regression equation that gives an estimate of hare density (Krebs et al. 1987). Estimates from this technique correlate well with density estimates derived from simultaneous mark-recapture studies occurring in the same area (Krebs et al. 2001, Murray et al. 2002, Mills et al. 2005, Homyack et al. 2006). However, because fecal deposition rates can vary by season and diet, and because pellet decomposition rates can vary with altitude, climate, aspect, precipitation, and cover type, region-specific, stand-specific, and/or season-specific equations should be developed before this technique is employed for a given area and season (Krebs et al. 2001, Prugh and Krebs 2004, Murray et al. 2005). Density estimates vary with plot size and shape, requiring equations specific to these geometric considerations as well (McKelvey et al. 2002). Pellet counts tend to yield more precise and unbiased density estimates when plots are visited and cleared more than once per year (e.g., plots cleared in the fall and then counted in the spring to estimate winter density) because variability in deposition and decomposition rates is reduced (Homyack et al. 2006). However, this requires considerably more work and expense than an annual survey. Some studies have conducted pellet plot counts without first clearing plots (e.g., Bartmann and Byrne 2001). This saves time and money, but requires the ability to discern fresh (this year) pellets from old pellets, which can be difficult and is generally not a recommended approach (Prugh and Krebs 2004, Murray et al. 2005).

Distance sampling is a well-developed method for estimating the density of objects in a given area (Buckland et al. 2001). In general, observers walk a pre-defined sampling transect and record each object of interest along with the perpendicular distance of that object from the transect line. This information is then used to develop a detection function which is in turn used to estimate density (Buckland et al. 2001). The method assumes all objects on the line are seen with certainty, objects are not double-counted, distance measures are accurate, and transect lines are located randomly within a study area (Buckland et al. 2001). Recently, distance sampling has been used to indirectly estimate hare density by first estimating the pellet group density of hares, then using fecal deposition and decomposition rates as a link back to hare density (Eriksson 2006). In general, distance sampling is more efficient than pellet plot counts as it does not require the tedious layout of hundreds of plots or counting individual pellets. This advantage is most recognizable in situations where pellet groups occur at low densities. Conversely, at extremely high densities, it may become difficult to distinguish pellet groups, and plots may be preferable (Marques et al. 2001). Regardless, distance sampling of pellet groups to estimate animal density also requires habitat and season specific decomposition and defecation rates, which can be difficult to obtain (Marques et al. 2001).

For this project, I have chosen to provide land managers with information relating demographic rates, as well as density, to forest stand characteristics. Thus, I will use mark-recapture techniques as data from such an approach can provide information on both density and demography. I will address the "effective trapping area" issue using a new approach that augments mark-recapture data with telemetry locations of animals using the grid.

The study outlined below is designed principally to evaluate the importance of young, regenerating lodgepole pine (*Pinus contorta*) and mature Engelmann spruce (*Picea engelmannii*)/ subalpine fir (*Abies lasiocarpa*) stands in Colorado by examining density and demography of snowshoe hares that reside in each (Figure 1). My hope is that information gathered from this research will be drawn upon as managers make routine decisions, leading to landscapes that include stands capable of supporting abundant populations of hares. I assume that if management agencies focus on providing habitat, hares will persist.

Specifically, I will evaluate small and medium lodgepole pine stands and large spruce/fir stands where the classes "small", "medium", and "large" refer to the diameter at breast height (dbh) of overstory trees as defined in the United States Forest Service R2VEG Database (small = 2.54–12.69 cm dbh, medium = 12.70–22.85 cm, and large = 22.86–40.64 cm dbh; J. Varner, United States Forest Service, personal communication). I also intend to identify which of the numerous density-estimation procedures available perform accurately and consistently using an innovative, telemetry augmentation approach as a baseline. I will assess movement patterns and seasonal use of deciduous cover types such as riparian willow. Finally, I will further expound on the relationship between density, demography, and stand type by examining how snowshoe hare density and demographic rates vary with specific vegetation, physical, and landscape characteristics of a stand.

Hypotheses

1) In general, snowshoe hare density in Colorado will be relatively low (≤0.5 hares/ha) compared to densities reported in northern boreal forests, even immediately post-breeding when an influx of juveniles will bolster hare numbers.
2) Snowshoe hare density will be consistently highest in small lodgepole pine stands, followed by large spruce/fir and medium lodgepole pine, respectively.
3) Survival will generally be highest in mature (large) spruce/fir stands followed by small and medium lodgepole pine, respectively.
4) Finite population growth rate will be consistently at or above 1.0 in mature spruce/fir stands with survival contributing most significantly to the growth rate. Finite growth rates for the lodgepole pine stands will be more variable.
5) Snowshoe hares will significantly shift their home ranges to make use of abundant food and cover provided by riparian willow (and/or aspen) habitats in summer.
6) Snowshoe hare density, survival, and recruitment will be highly correlated with understory cover and stem density.

STUDY AREA

The study area stretches from Taylor Park to Pitkin in central Colorado (Figure 2). Elevation ranges from 2700 m to 4000 m. Sagebrush (*Artemisia spp.*) dominates broad, low-lying valleys. Most montane areas are covered by even-aged, large-diameter lodgepole pine forests with sparse understory. Moist, north-facing slopes and areas near tree line are dominated by large-diameter Engelmann spruce/subalpine fir. Interspersed along streams and rivers are corridors of willow. Patches of aspen occur sporadically on southern exposures. This area was chosen over other potential study areas in the state because 1) it contained numerous examples of the 3 stand types of interest (more southern regions lack naturally occurring stands of lodgepole pine), 2) it was not subject to confounding effects of large-scale mountain pine beetle outbreak as were more northern stands, and 3) an adequate number of radio frequencies were available to support a large study with hundreds of radio-tagged individuals.

Within the study area I selected sample stands based on the following: Potential replicate stands were required to be 1) close enough geographically to minimize differences due to climate, weather, and topography, but are far enough apart to be considered independent, 2) adjacent to one or more riparian willow corridors, 3) within 1 km of an access road for logistical purposes, 4) of suitable size and shape to admit a 16.5-ha trapping grid, and 5) consistent in their management history (i.e., replicate lodgepole pine stands were clear-cut and/or thinned within 1-2 years of each other).

I queried the U.S. Forest Service R2VEG GIS database using the criteria listed above to initially develop a suite of potential sample stands. I further narrowed this suite after obtaining updated stand-level information from local USFS personnel (Art Haines, Silviculturalist, USFS Gunnison Ranger District, personal communication). Finally, I ground-truthed potential stands and qualitatively assessed their representativeness and similarity to other potential replicates. Given the numerous constraints imposed, very few stands met all criteria. Thus, I was unable to randomly select sample stands from a population of suitable stands. Rather, I subjectively chose the “best” stands from among the handful that met my criteria. Small lodgepole stands rarely occur on the landscape in patches large enough to fit a full

7 x 12 trapping grid. To accommodate this, I sampled 6 replicate small lodgepole stands (rather than 3) using 6 x 7 trapping grids (1/2 size).

METHODS

Experimental Design/Procedures

Variables.--The response variables of interest for this project include stand-specific snowshoe hare density (*D*), apparent survival (ϕ), recruitment (*f*), finite population growth rate (λ), and a metric of seasonal movement. Density is the number of hares per unit area and will be estimated using a variety of conventional techniques as well as a rigorous method that incorporates radio telemetry. The stand-specific demographic parameters will be estimated primarily from capture-mark-recapture methods. As such, apparent survival is defined as the probability that a marked animal alive and in the population at time *i* survives and is in the population at time $i + 1$. Apparent survival encompasses losses due to both death and emigration. Recruitment is the number of new animals in the population at time $i + 1$ per animal in the population at time *i*. New recruits can arise from on-site reproduction as well as immigration. The finite population growth rate is the number of animals in a given age class at time $i + 1$ divided by the number present at time *i*. Shifts in home range will be assessed by comparing the seasonal proportion of telemetry locations in deciduous habitats using multi-response permutation procedures (MRPP; Zimmerman et al. 1985, White and Garrott 1990).

Potential explanatory variables for snowshoe hare density, demographics, and movement include general species composition and structural stage of each stand in which response variables are measured. Additionally, stem density, horizontal cover, and canopy cover (to a lesser extent) are highly correlated with snowshoe hare abundance and habitat use (Wolfe et al. 1982, Litvaitis et al. 1985, Hodges 2000, Zahratka 2004, Miller 2005). Thus, I will further characterize vegetation in each stand by measuring stem density by size class (1-7 cm, 7.1-10 cm, and >10 cm), percent canopy cover, percent horizontal cover of understory and basal area. Basal area is an easily obtainable metric that may be correlated with the other variables and is recorded routinely during timber cruises, whereas the others are not. Thus, it might prove a useful link for biologists designing management strategies for snowshoe hare. Additionally, I will record physical covariates such as ambient temperature, precipitation, and snow depth at each stand during sampling periods as well as precipitation 1-3 years prior to sampling. Finally, I will calculate potentially important landscape metrics such as patch size and level of fragmentation.

Sampling.--All trapping and handling procedures have been approved by the Colorado State University Animal Care and Use Committee and filed with the Colorado Division of Wildlife. Snowshoe hares breed synchronously and generally exhibit 2 birth pulses in Colorado (although in some years, some individuals may have 3 litters), with the first pulse terminating approximately June 5–20 and the second approximately July 15–25 (Dolbeer 1972). To obtain a maximum density estimate, I began data collection on the first suite of sites immediately following the second birth pulse in late July. Along with a crew of 5 technicians, I deployed one 7 × 12 trapping grid (50-m spacing between traps; grid covers 16.5 ha) in the large spruce/fir and medium lodgepole stands within the first suite, along with 2 6 × 7 grids in 2 small lodgepole stands. Grid set up and trap deployment followed Griffin (2004) and Zahratka (2004). Grid locations and orientation within each stand were chosen subjectively to accommodate logistical constraints and to ensure that hares using the grid had ample opportunity to use adjacent riparian willow zones. Traps were deployed in all 4 stands in a single day. As traps are deployed, they were locked open and "pre-baited" with apple slices and commercial rabbit chow. During winter, hay cubes were added to traps as well (see Discussion). On days 2-4, the crew continued pre-baiting, replacing apples and rabbit chow as necessary. The purpose of this extended pre-baiting was to maximize capture rates when trapping began. This minimized the number of trap-nights needed to capture the desired number of animals which in turn minimized trapping-related stress as well as the likelihood that

American marten (*Martes americana*) keyed into trap lines and preyed on entrapped hares, as has occurred in previous studies (J. Zahratka, personal communication). During pilot work in winter 2005, I observed low but increasing capture rates (<0.20) during the first 3 nights of trapping, with higher, more stable capture probabilities after 3 days (approximately 0.35–0.45). Thus 3 days of pre-baiting seems reasonable.

Traps were set on the afternoon of the 4th day and checked early each morning and again in the evening on days 5–9. By checking traps in both morning and evening I prevent hares from being entrapped >13 hours, which should minimize capture stress. A crew of 2 people worked together on each grid to check traps and process captures as quickly as possible. All captured hares were coaxed out of the trap and into a dark handling bag by blowing quick shots of air on them from behind. Hares remained in the handling bag, physically restrained with their eyes covered, for the entire handling process. Each individual was aged, sexed, marked with a passive integrated transponder (PIT) tag and temporary ear mark (to track PIT tag retention), then released. Aging consisted of assigning each individual as either juvenile (<1 year old, <1000 g) or adult (≥1 year old, ≥1000 g) based on weight. This criterion is accurate through the end of September at which point juveniles are difficult to distinguish from adults (K. Hodges, University of British Columbia; P. Griffin, University of Montana, personal communication). After the first day of trapping, all captured hares were scanned for a PIT tag prior to any handling and those already marked were recorded and immediately released. Traps and bait were completely removed from the grid on day 10.

In addition to PIT tags and ear marks, I radio collared up to 10 hares captured on each grid with a 28-g mortality-sensing transmitter (BioTrack, LTD) to facilitate unbiased density estimation as well as assessment of seasonal movements. I expected heterogeneity in snowshoe hare movements and use of the grid area, with potential bias surfacing due to location at which a hare is captured (e.g., hares captured on the edge of a grid may use the grid area differently than those captured at the center), and differential behavioral responses to trapping (e.g., young individuals may have lower capture probabilities and thus may be more likely to be captured on later occasions). To guard against the first potential bias, I randomly selected a starting trap location each morning and ran the grid systematically from that point. Thus, the first several hares encountered (and collared) were as likely to be from the inner part of the grid as from the edge. To protect against the second potential source of bias, I refrained from deploying the final 3 collars until days 4 and 5 of the trapping session.

Immediately following the removal of traps, the field crew began work locating each radio-collared hare 1–2 times per day for 10 days. Most locations were obtained by triangulation from relatively close proximity, but some were obtained by "homing" on a signal (Samuel and Fuller 1996, Griffin 2004) taking care not to push hares while approaching them. Because hares are largely nocturnal (Keith 1964, Mech et al. 1966, Foresman and Pearson 1999), I made an effort to conduct telemetry work at various times of the night (safety and logistics permitting) and day to gather a representative sample of locations for each hare.

The crew gathered telemetry locations for radio-collared hares on the initial sites for 8 to 10 days. Then the 10−day trapping procedure and 8 to 10−day telemetry work were repeated on the 3 grids comprising suite 2 (Figure 3). The cycle was repeated once more for grids in suite 3 (Figure 3). The entire process was repeated during the winter when densities should have been at a minimum.

In summary, for any given 9-week sampling period, I collected data from 12 total grids, 1 spruce/fir, 1 medium lodgepole, and 2 small lodgepole across 3 replicates. Sampling will occur during 2 such 9-week periods each year – once in late summer and once in late winter – and will continue for 3 years. During the interim between intensive trapping and telemetry work, monthly telemetry checks were conducted from the air to track mortalities and facilitate retrieval of collars from dead hares. Telemetry work was also occur during "pre-baiting" days after the initial summer sampling session to determine which hares were still alive and immediately available to be sampled by the grid during the ensuing trapping period.

Vegetation sampling at each stand will follow protocols established through previous snowshoe hare and lynx work in Colorado (Zahratka 2004, T. Shenk, Colorado Division of Wildlife, personal communication). Specifically, on each of the 12 live-trapping grids, I will lay out 5 × 5 grids (3-m spacing) of vegetation sampling points centered on 15 of the 84 trap locations (Figure 4; 9 points will be sampled on each of the ½-sized small lodgepole stands). At each of the 25 vegetation sampling points, I will record: 1) distance to the nearest woody stem 1.0–7.0 cm, 7.1–10.0 cm, and >10.0 cm in diameter at heights of 0.1 m and 1.0 m above the ground (to capture both summer [0.1 m] and winter [1.0 m] stem density; Barbour et al. 1999), 2) horizontal cover in 0.5-m increments above the ground up to 2 m (Nudds 1977), and 3) canopy cover [present or absent] using a densitometer. Additionally, at the center of all 15 vegetation sampling grid points (i.e., at the trap location), I will measure basal area using an angle gauge. These measurements will be gathered once at the start of the project, unless conditions change due to disturbance such as fire. Temperature will be monitored hourly at each grid during the 6-week intensive sampling periods using data loggers. During winter sampling periods, snow depth measurements will be recorded daily at the same 15 trap locations used to quantify the vegetative attributes of that stand.

Data Analysis

Density.--I assumed that hare populations were demographically and geographically closed during the short 5-day mark-recapture sampling periods. To obtain a density estimate for each grid, I used the Huggins closed capture model (Huggins 1989, 1991) in Program MARK (White and Burnham 1999) with some modifications. The basic Huggins estimator (no individual covariates) is based on the fact that if p_j is the probability that a hare in the population is captured (and marked) for the first time on trapping occasion *j*, then $p^* = 1-(1-p_1)...(1-p_5)$ is the probability that an individual is captured at least once during a 5-day trapping period (i.e., *j* = 1,…,5). Accordingly, the basic Huggins estimator for population size, $\hat{N}$, is $\hat{N} = M_{t+1} / p^*$ where M_{t+1} is the total number of hares captured. The estimator can be re-written to allow each of the M_{t+1} individuals captured to have their own p^*. In that case, $\hat{N} = \sum_{i=1}^{M_{t+1}} 1/p_i^*$. Presumably hares that reside near the edge of a grid encounter fewer traps and are less likely to be captured than hares residing near the center of a grid. To account for this, I took advantage of the Huggins model with individual covariates to model p^* by using the logit link function of program MARK to model p_i^* as a function of d_i, where d_i is distance from the edge of the grid for hare *i* based on mean capture coordinates. A naïve density estimate for each grid would then be $\hat{D} = \hat{N} / A$ where *A* is the area of the grid. However, this gives full credit to all hares, even those whose home range only partially overlaps the grid, which results in a density estimate that is biased high. To correct for this bias, I determined the proportion, $(\tilde{p}_k)$, of telemetry locations for each of the *k* = 1,…,10 radio-collared hares that fell within the "naïve grid area." By incorporating data from multiple grids, a logistic regression model was developed to estimate $\tilde{p}_i$ for all M_{t+1} animals captured on a grid based on distance from the edge of the grid for hare *i* (d_i). Replacing the numerator (i.e., 1) in the Huggins estimator with $(\tilde{p}_i)$, gives a density estimate, $\hat{D} = \left(\sum_{i=1}^{M_{t+1}} \tilde{p}_i / p_i^* \right) \Big/ A$.

The above-stated approach assumes that radio-collared hares neither gravitate toward nor avoid the former grid area after the 5 days of trapping, 10–20 locations per hare is enough to provide a reasonable representation of the proportion of time they spend on the grid, and their use of the grid area is representative of other hares that were captured but not collared (i.e., that the logistic regression model of $\tilde{p}_i$ is a useful model). I contend that this type of estimate from grid-based trapping can be construed as a relatively unbiased estimate of density. Using these point estimates and their associated confidence intervals, I compared hare density among seasons and stand types. I will also compare these "true" density estimates to those that would have been obtained using other available methods such as ½ mean maximum distance moved (Wilson and Anderson 1985, Williams et al. 2002:314-315), full mean maximum distance moved (Parmenter et al. 2003), ½ trap interval (Parmenter et al. 2003), "nested grids" (White et al. 1982:120-131), and Program DENSITY (Efford et al. 2004).

Demography.--I will analyze mark-recapture data using Pradel temporal symmetry models (Pradel 1996, Nichols and Hines 2002) in a robust design framework (Williams et al. 2002:523-554), which will be available in Program MARK by summer 2006. Thus, I will treat summer and winter sampling occasions as primary periods, and the 5-day trapping sessions within each as secondary periods. The Pradel temporal symmetry models employ both forward and reverse-time evaluation of capture histories to provide estimates of apparent survival ($\hat{\phi}$) and seniority ($\hat{\gamma}$). Apparent survival, ϕ_i, is the probability that a marked animal alive and in the population at time *i* survives and is in the population at time *i* + 1. The seniority parameter, γ_i, is the reverse-time analogue of survival. Reading backward through a capture history, it is the probability that a marked animal alive and in the population at time *i* was alive and in the sampled population at time *i* − 1. If *N* is the number of animals present in the population, $N_i\phi_i \approx N_{i+1}\gamma_{i+1}$ and $N_{i+1}/N_i = \phi_i/\gamma_{i+1} = \lambda_i$. Also, if f_i is recruitment rate, or the number of recruits at time *i* + 1 per animal present at time *i*, then $N_{i+1} = N_i\phi_i + N_i f_i$. Rearranging and substituting into the previous equation gives $f_i = \phi_i\left(1/\gamma_i - 1\right)$. Thus, using Pradel models, one can estimate recruitment and finite population growth rate in addition to survival (Pradel 1996, Nichols and Hines 2002).

I will use Akaike's Information Criterion corrected for small sample size (AICc; Burnham and Anderson 1998) to determine whether models with time-dependent parameters or constant parameters are best supported by the data. I will derive estimates of the above-mentioned parameters from the best model or from model averaging. I anticipate pooling capture data across sites to obtain $\hat{\phi}_i$, $\hat{\lambda}_i$, and $\hat{f}_i$ for each stand type for each interval between primary sampling periods (5 estimates of each). I also anticipate simply estimating these parameters for "generic hares", treating both juveniles and adults as a single group or age class. Given that juveniles are morphometrically indistinguishable from adults by their first fall of life (K. Hodges, University of British Columbia; P. Griffin, University of Montana, personal communication), adult and juvenile survival rates are similar (Griffin 2004), and there is little evidence for age-specific differences in pregnancy rates or litter size (Dolbeer 1972), this approach seems justified. However, if I happen to capture sufficient numbers of juveniles and adults, the design I have laid out here allows for treating the age classes separately. This, in turn, may permit me to decompose the contribution that f_i makes to λ_i into the portion of that contribution due to on-site reproduction and that due to immigration (Nichols et al. 2000). Similarly, it may also be possible using my telemetry data to decompose apparent survival, ϕ_i, into emigration and mortality. Such fortuitous situations would facilitate the identification of source and sink habitats if they exist.

Seasonal Movements.--I will assess whether snowshoe hares seasonally shift their home ranges using the multi-response permutation procedure (MRPP; Zimmerman et al. 1985, White and Garrott 1990:134-135). Under this approach, telemetry locations are grouped by season (summer and winter), and an MRPP statistic is calculated as the weighted average of the distance between all possible pairs of locations within groups compared to the average distance between all possible pairs ignoring groups. The null hypothesis is that the distribution of locations is the same for both groups (seasons). Sufficiently small values of the test statistic suggest that within group distances are smaller than distances measured ignoring groups, which is evidence against the null in favor of a group (seasonal) effect. P-values are obtained by calculating the percentile of the observed test statistic relative to all possible test statistics that could be computed by re-arranging the data into all possible groups of 2. The MRPP procedure is sensitive and can detect even small changes in use of an area (White and Garrott 1990:136). I propose a priori that changes in proportional use of deciduous habitats <0.10 in magnitude are unlikely to be biologically significant.

Vegetation.--I will calculate mean stem density, horizontal cover, canopy cover, and basal area for each season–stand type as well as temperature, precipitation, snow depth information, and landscape metrics. These will be entered into the MARK design matrix as covariates to population size (~density) and survival in a random effects analysis. As such, I will be able to quantify the amount of variation in population size or survival that is due to differences in vegetation, landscape, or weather relative to the amount left to other causes.

Sample size.--I conducted power analyses to determine the probability of discerning meaningful differences in density and survival for hares occupying different stand types. For density, I postulated that foraging lynx likely do not discriminate among stands that differ by only a few hares. However, it seems probable that if hare density in one stand is twice that of another, a lynx would choose the former given the opportunity. Thus, I conducted power calculations to determine the probability of distinguishing differences in densities between 2 stand types in which one had twice the density of hares as the second. Specifically, using the Huggins closed capture model (Huggins 1989, Huggins 1991) in Program MARK, I specified the number of hares (N) present in each of 2 groups (i.e., 2 stand types), allowed capture (p) and recapture (c) probabilities to vary with time but constrained them to be equal and the same for each group, then simulated this scenario 1000 times for a range of realistic capture probabilities. For each simulation I calculated a 95% confidence interval for the mean difference in $\hat{N}$ between the 2 groups and determined the proportion of all simulations in which this confidence interval did not include zero. This proportion is the power, or probability of discerning a difference between the 2 groups when one actually exists. I compared 2-fold differences in density at the low (5 vs. 10 hares/grid) and high (15 vs. 30 hares/grid) end of the range of hare numbers and I expect to observe (Zahratka 2004). I also simulated the power to detect differences between 17 and 39 hares/grid, corresponding to recently published cut-points for low and high hare densities in the context of lynx conservation (Mills et al. 2005). Given capture/recapture probabilities I observed during winter 2005 (approximately 0.35–0.45), I expect to have reasonable power to detect 2-fold differences in density even if I encounter relatively few hares per grid (Figure 5).

I conducted power analyses for survival in a similar manner using the Huggins estimator (Huggins 1989, Huggins 1991) in a robust design framework (Williams et al. 2002:524-556). For this analysis, I specified 3 primary periods (e.g., 3 years) with 5 secondary occasions for each. I established either 30 or 45 hares in each of 2 groups (i.e., pooled an expected 10-15 hares/grid across the 3 grids in a given habitat type), specified a different survival rate for each, and allowed p and c to vary with time but constrained them to be equal and the same for each group as before. I then specified a general model that assumed survival rates varied among groups and a second, reduced model that assumed survival rates were the same for each group. After 1000 simulations under a given scenario of hare numbers, capture probabilities, and survival rates, I conducted a likelihood ratio test between each pair of general and reduced models. As before, I used the proportion of significant tests as an estimate of power to detect differences in survival.

I compared survival rates of 0.4 vs. 0.5, 0.3 vs. 0.5, and 0.2 vs. 0.5. These rates span the range of annual hare survival rates reported in the literature (Dolbeer 1972, Dolbeer and Clark 1975, Griffin 2004). Also, because each comparison is anchored at 0.5, these calculations provide a conservative estimate of power due to the nature of binomial probabilities. That is, I would be more likely to distinguish the difference between 0.1 and 0.2 than between 0.4 and 0.5 even though the difference in both cases is 0.1 because the sampling variance of the estimate for the same sample size is maximal at 0.5 and declines to 0 for survival rates of 0 or 1. Results indicate that I have ≥80% chance of discerning real differences in survival of ≥0.3 (Figure 6), but only 40-65% chance (depending on number of hares captured) of detecting a difference of 0.2, and very little chance of detecting differences smaller than 0.2. However, I plan to combine my telemetry data with my trapping data in the MARK Robust design model using separate groups for each data type. This should enhance my precision and power, thus making the prospect of detecting differences as small as 0.2 a possibility.

To complete a power analysis for $\hat{\lambda}$ requires running simulations of Pradel models in a robust design framework. This capability is not yet available in Program MARK, so such an analysis has not been completed. Sampling 15 vegetation plots per trapping grid provided reasonably precise characterizations of similar stands in similar locations during a previous study (Zahratka 2004). I trust this level of sampling will be adequate for the present study as well. If not, more plots can be established at a later date given that vegetative characteristics are unlikely to change appreciably over a few years.

RESULTS AND DISCUSSION

Much of the analysis presented above is not possible or meaningful without several seasons of data, especially the survival, recruitment, and growth rate models. Below, I present a basic summary, relevant observations, and initial density estimates from the inaugural year of this project.

I captured 75 hares 166 times during July-September 2006. I captured 99 hares 243 times during January-March 2007 (Table 1). Fourteen of these individuals were captured during both the summer and winter sampling sessions. During summer, I captured over twice as many individuals in small lodgepole stands as in spruce/fir. I captured only a few individuals in medium lodgepole stands. During winter, captures were more evenly distributed among the stands (Table 1).

During the initial trapping session of the summer trapping period, 6 hares were captured, handled, and released (seemingly without harm) but were found dead in traps 1-3 days later. I collected the carcasses and submitted them for necropsy. Cause of death was attributed to capture myopathy, which is relatively common in lagomorphs (Laurie Baeten DVM, and Lisa Wolfe DVM, Colorado Division of Wildlife, personal communication). I subsequently altered my trapping protocol to further minimize both the amount of time a hare could be entrapped as well as the handling time at each capture. No trap deaths occurred during the remainder of the sampling season aside from 4 hares that succumbed to predation while inside traps.

During the initial 2 trapping sessions of the winter trapping period, 6 more hares were captured, handled, and released multiple times, again with seemingly little adverse reaction, only to be found dead on a subsequent trapping occasion. Several more hares died during the 10-day telemetry session immediately following trapping. These "telemetry deaths" could have been due to natural causes, effects of capture, or a combination of both. Again, carcasses were submitted for necropsy, and again capture myopathy was cited as a potential cause of death. Further examination of the data indicated that hares trapped ≥3 days in a row were much more likely to die in a trap or during telemetry than other hares. Thus, I further modified the trapping protocol by locking traps open on day 3 of the 5-day trapping period so that hares could not be trapped more than 2 days in a row. Additionally, I began providing hay cubes in the traps as roughage to complement the high quality alfalfa pellets and apples. After implementing these changes, I did not observe any further trap-deaths or telemetry-deaths for the rest of the season.

I averaged 9.9 and 6.3 locations per radio-tagged hare during the summer and winter sampling sessions, respectively (Table 2). Thus, "proportion of time on grid," which is critical to my density estimation procedure, was based on relatively few points per individual for the first 2 sampling periods, and I was unable to attain my goal of 10-20 locations per individual. Following the winter field season, I conducted a series of simulations to examine the effects of sample size on precision of density estimates. I found that 1) the variability between hares ("proportion on grid" ranges from 0.00-1.00) overwhelms the variability within hares (i.e., the binomial variance for proportion of time on grid for any single individual, which decreases as number of locations increases), and 2) given a fixed effort, the variance of the density estimate is minimized by increasing the number of individuals collared as opposed to increasing the number of locations per individual. Thus, it is better to radio-tag more hares and get fewer locations than to tag fewer hares and get more locations. I will continue to deploy as many collars as possible, and will strive for 10-20 locations per individual, but the level of sampling achieved during the first 2 field seasons appears sufficient to detect the large differences in density that occur on the landscape.

During summer, density estimates followed hypotheses 1) and 2) above. Specifically, hare density in small lodgepole stands was twice that observed in spruce/fir, which was more than twice that observed in medium lodgepole stands (Figure 7). However, even the relatively high density found in the small lodgepole stands was relatively low compared to densities that have been reported in other parts of hare range (Griffin 2004, Hodges 2000). However, different methods for computing density make this type of comparison difficult.

During winter, hare densities remained the same in spruce/fir stands. Hare density in medium lodgpole stands more than doubled, although still remained relatively low compared to other stand types. Density in the small lodgepole stands dropped significantly compared to summer levels and was more variable among replicates. Hare density is likely driven by availability of food and cover. I submit that the interplay between food, cover, and snow depth provides a plausible explanation for the density patterns observed during the first year of this study. Spruce/fir stands probably provide adequate access to both necessities during both summer and winter due to their uneven-aged, multi-layered structure. Medium lodgepole stands, on the other hand, apparently provide very little forage/cover for hares during summer as the canopy in these stands is generally ≥1 meter off the ground. However, in winter, accumulated snow may bring that canopy back into reach for hares. Conversely, small lodgepole stands provided abundant food and cover during summer, but accumulated snow during winter brings hares closer to the crowns of the young trees, which then provide less cover.

SUMMARY

- The number of snowshoe hares captured, the number of captures, and the number of locations obtained per hare during the first year appeared adequate for attaining the objectives of this study.

- Some deaths due to capture myopathy (most likely cause) occurred during initial trapping periods in each sampling season. Changes to the trapping protocol, trapping schedule, and bait provided seem to have alleviated the problem.

- Snowshoe hare densities during summer were highest in small lodgepole stands, followed by large spruce/fir and medium lodgepole. During winter, densities in small lodgepole stands dropped and became more variable across replicates. Medium lodgepole stands gained hares. Spruce/fir stands remained at the same density as during summer.

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Table 1. Number of snowshoe hares (*Lepus americanus*) captured during 5-day trapping sessions conducted during July-September 2006 and January-March 2007 on 3 medium lodgepole, 3 spruce/fir, and 6 small lodgepole stands on the Gunnison National Forest, Taylor Park and Pitkin, Colorado.

	Number of Hares Captured (Total Captures)		
	Summer 2006	Winter 2007	Both Summer and Winter
Medium Lodgepole	3	24	2
Small Lodgepole	50	40	10
Spruce/Fir	22	35	2

Table 2. Number of snowshoe hares (*Lepus americanus*) radio-collared and tracked during 10-day sessions immediately following 5-day trapping periods July-September 2006 and January-March 2007 on the Gunnison National Forest, Taylor Park and Pitkin, Colorado.

	Summer 2006	Winter 2007
Number of Hares Collared	41	79
Number of Locations	407	510
Number of Locations/Hare	9.9	6.5

Figure 1. Purported high quality snowshoe hare habitat in Colorado. From left to right: small lodgepole pine, medium lodgepole pine, and large Engelmann spruce/subalpine fir.

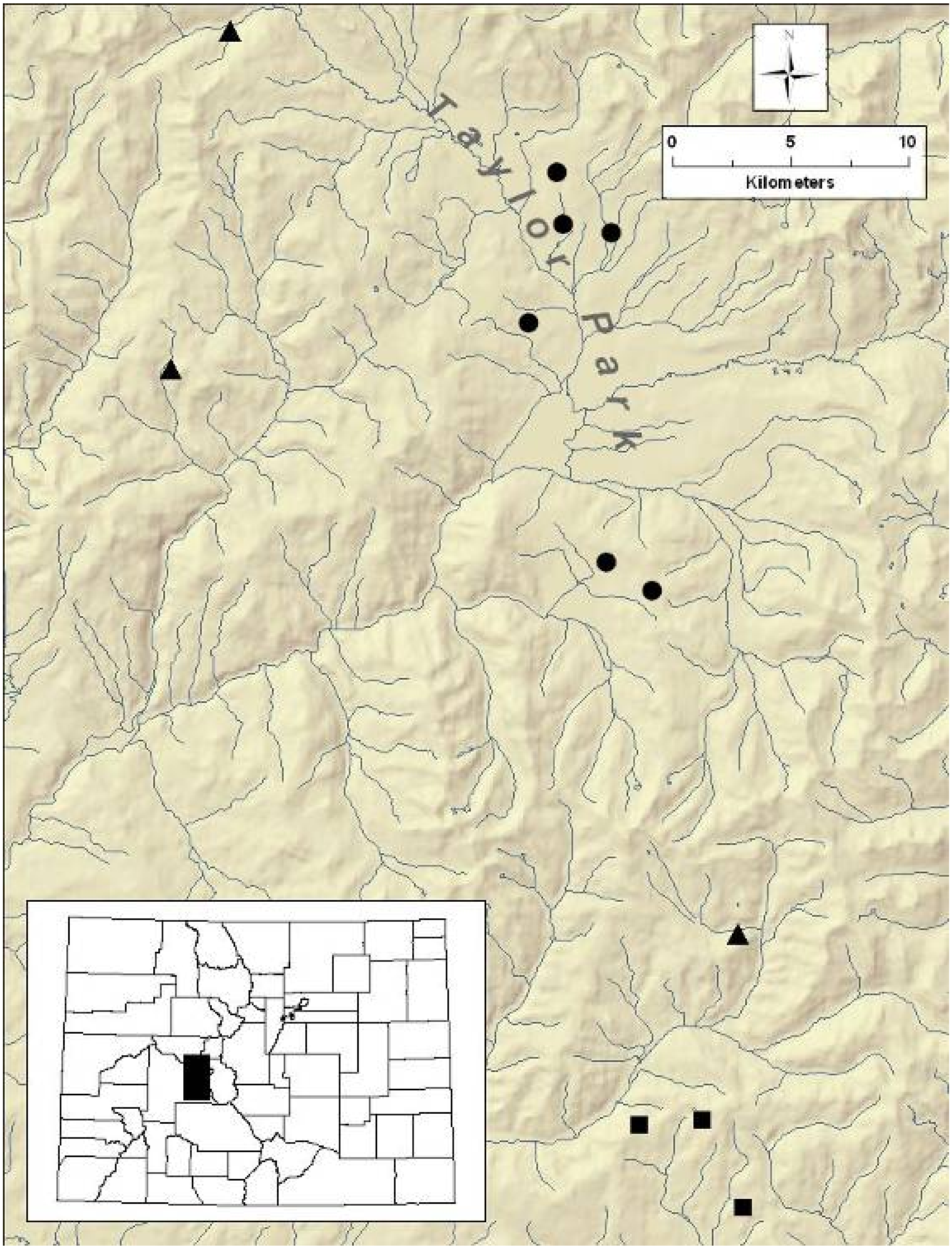

Figure 2. Study area near Taylor Park and Pitkin, Colorado including medium lodgepole (squares), small lodgepole (circles), and spruce/fir (triangles) stands selected for mark-recapture sampling.

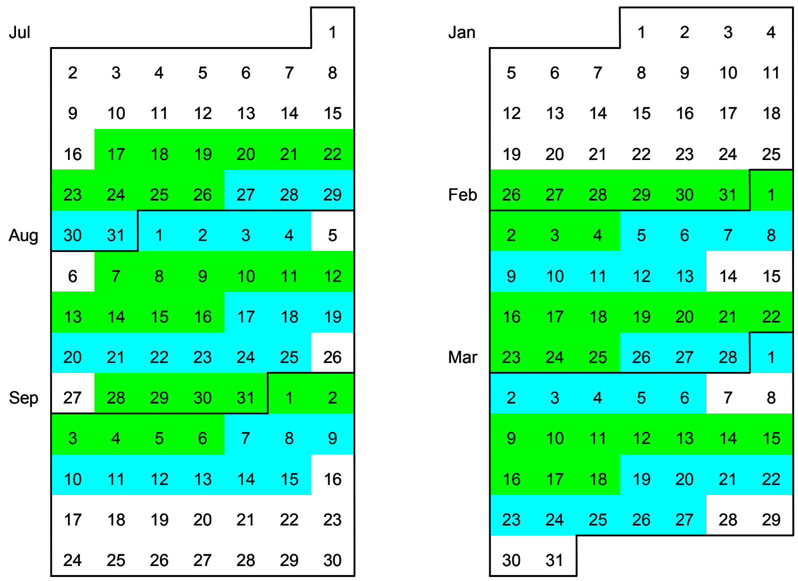

Figure 3. Approximate annual data collection schedule for trapping (■) and telemetry (■). Dates and weeks will change depending on calendar year and pay schedule. During telemetry work, the 6-person crew will be divided into 2 teams, only one of which will be working at any given time. Monthly locations on radio-collared hares will also be collected in the interim between the intensive sampling periods indicated here.

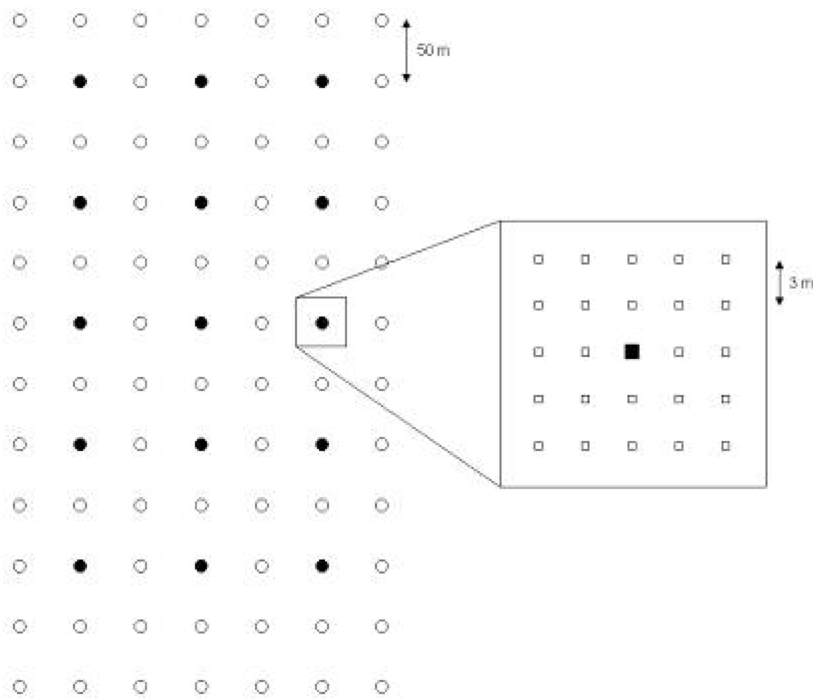

Figure 4. 15 trap locations (•) on 7 × 12 trapping grid where vegetation will be sampled by measuring stem density horizontal cover, and canopy cover at the 25 points on each 5 × 5 subgrid (inset). In addition, basal area will be measured at the trap location (■) on which each of the 15 subgrids are centered.

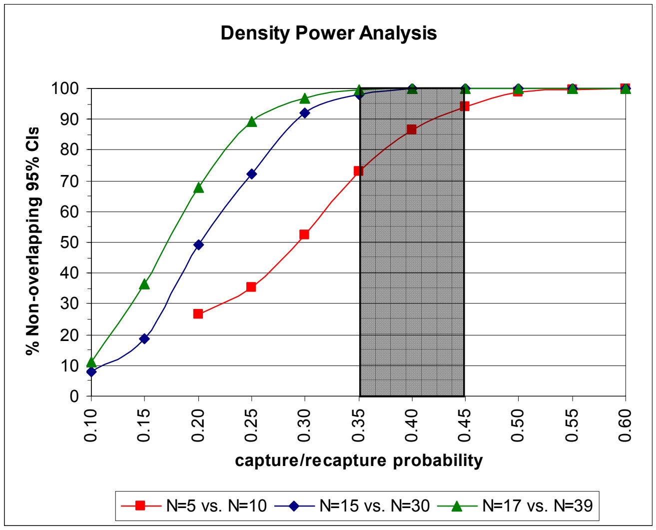

Figure 5. Power for distinguishing differences in snowshoe hare density between 2 habitat types when a difference actually exists. Gray area indicates the capture probability realized by the 3rd day of trapping during a pilot study in winter 2005. *N* indicates number of hares per grid, a range of roughly 0.1 (*N* = 5) to 0.7 hares/ha (*N* = 39).

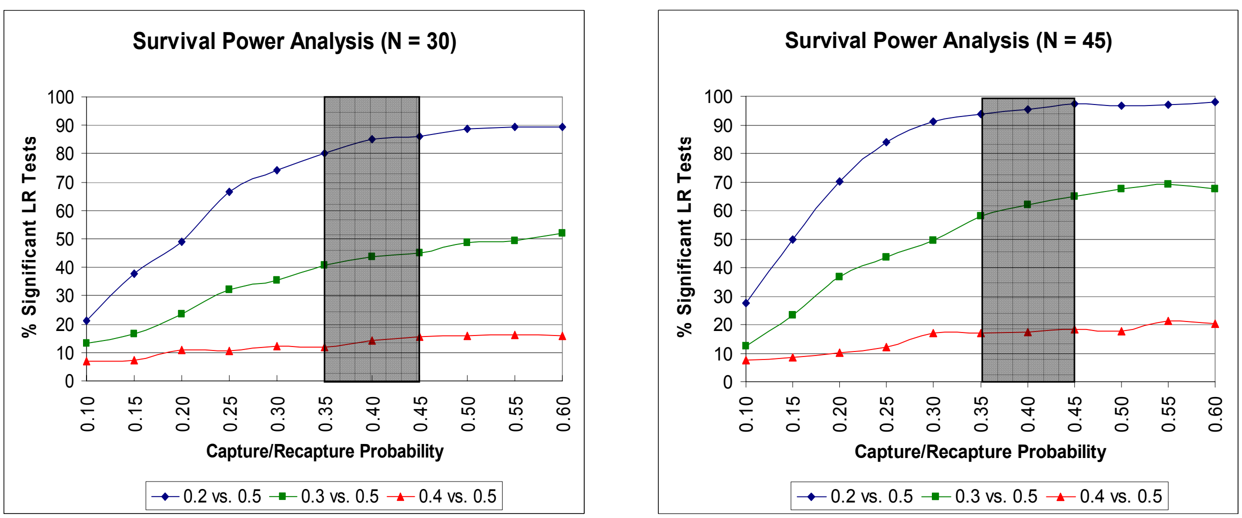

Figure 6. Power, or probability of distinguishing differences in snowshoe hare survival between 2 habitat types when differences actually exist. *N* = 30 (left) and *N* = 45 (right) correspond to reasonable estimates of the number of hares I expect to capture in each habitat type. Gray area indicates the capture probability realized by the 3rd day of trapping during a pilot study in winter 2005.

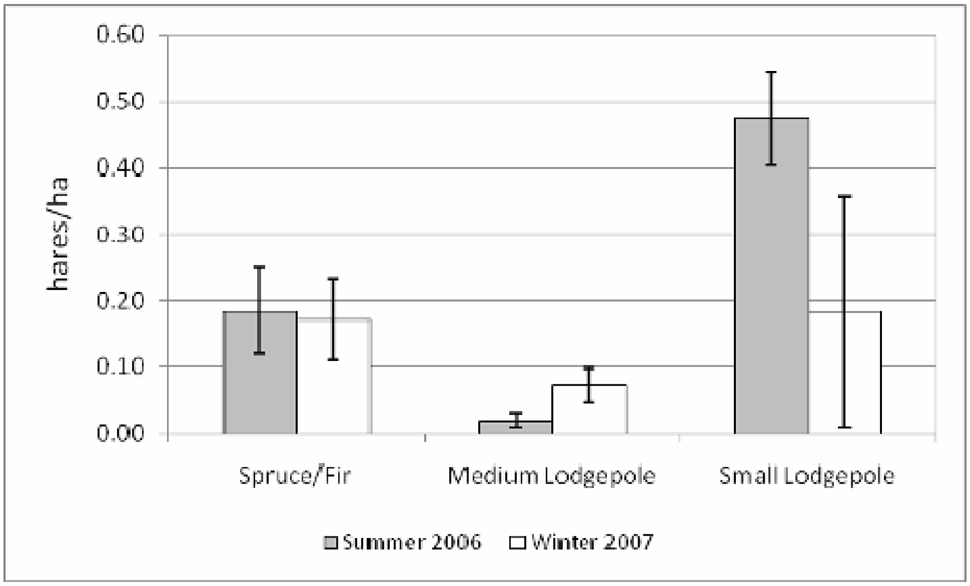

Figure 7. Snowshoe hare density and 95% confidence intervals in 3 types of stands in central Colorado as determined by mark-recapture with telemetry augmentation, July-September 2006 and January-March 2007.

Colorado Division of Wildlife
July 2006 – June 2007

WILDLIFE RESEARCH REPORT

State of	Colorado	:	Division of Wildlife
Cost Center	3430	:	Mammals Research
Work Package	3001	:	Deer Conservation
Task No.	4	:	Effect of Nutrition and Habitat Enhancements on Mule Deer Recruitment and Survival Rates
Federal Aid Project:	W-185-R	:	

Period Covered: July 1, 2006 – June 30, 2007

Authors: C. J. Bishop, G. C. White, D. J. Freddy, and B. E. Watkins

All information in this report is preliminary and subject to further evaluation. Information MAY NOT BE PUBLISHED OR QUOTED without permission of the author. Manipulation of these data beyond that contained in this report is discouraged.

ABSTRACT

We measured mule deer (*Odocoileus hemionus*) population parameters in response to a nutrition enhancement treatment to evaluate the relative importance of habitat quality as a limiting factor of mule deer in western Colorado during November 2000 – January 2005. The nutrition enhancement treatment increased survival of fetuses to the yearling age class by 0.14–0.20 depending on year and fawn sex, although 95% confidence intervals slightly overlapped 0. The nutrition treatment also had a positive effect on annual adult doe survival. Survival of does receiving the treatment ($\hat{S}$ = 0.879, SE = 0.0206) was higher than survival of control does ($\hat{S}$ = 0.833, SE = 0.0253). Our estimate of the population rate of change, $\hat{\lambda}$, was 1.15–1.17 for treatment deer and 1.02–1.06 for control deer, with some overlap in 95% confidence intervals. The treatment caused $\hat{\lambda}$ to increase by 0.139 (95% CI: –0.0152, 0.2941) during 2001–02, 0.113 (95% CI: –0.0009, 0.2279) during 2002–03, and 0.145 (95% CI: 0.0176, 0.2723) during 2003–04.Our results provide a foundation for focusing deer management efforts on improving habitat quality in western Colorado pinyon-juniper (*Pinus edulis*-*Juniperus osteosperma*) ecosystems with corresponding research efforts to quantify the effects of habitat manipulations on deer performance. During the past year, we had 2 papers published in peer-reviewed journals, we completed final data analyses, and we prepared 3 other manuscripts for publication. The published manuscripts included: 1) a manuscript on the effectiveness of vaginal implant transmitters (Journal of Wildlife Management 71(3):945–954), and 2) a manuscript documenting malignant catarrhal fever in the Uncompahgre deer population (Journal of Wildlife Diseases 43(3):533–537). We also completed a publication on mule deer habitat guidelines for the Colorado Plateau ecoregion, which will be published by the Western Association of Fish and Wildlife Agencies. The estimated publication date is January 2008.

WILDLIFE RESEARCH REPORT

EFFECT OF NUTRITION AND HABITAT ENHANCEMENTS ON MULE DEER RECRUITMENT AND SURVIVAL RATES

CHAD J. BISHOP, GARY C. WHITE, DAVID J. FREDDY, AND BRUCE E. WATKINS

P. N. OBJECTIVE

To determine experimentally whether enhancing mule deer nutrition during winter and early spring via supplementation increases fetal survival, neonatal survival, overwinter fawn survival, or ultimately, population productivity.

SEGMENT OBJECTIVES

1. Complete final data analyses to support preparation of manuscripts.
2. Prepare manuscripts for submission to scientific journals for publication.
3. Complete dissertation as part of PhD requirements at Colorado State University

INTRODUCTION

Mule deer (*Odocoileus hemionus*) numbers apparently declined during the 1990s throughout much of the West, and have clearly decreased since the peak population levels documented during the 1940s–1960s (Unsworth et al. 1999, Gill et al. 2001). Biologists and sportsmen alike have concerns as to what factors may be responsible for declining population trends. Although previous and current research indicates multiple interacting factors are responsible, habitat and predation have typically received the focus of attention. A number of studies have evaluated whether predator control increases deer survival, yet results are highly variable (Connolly 1981, Ballard et al. 2001). Together, predator control studies with adequate rigor and statistical power indicate predation effects on mule deer are variable as a result of time-specific and site-specific factors. Studies which have demonstrated deer population responses to predator control treatments have failed to determine whether predation is ultimately more limiting than habitat when considering long term population changes. Numerous research studies have evaluated mule deer habitat quality, but virtually no studies have documented population responses to habitat improvements. In many areas where declining deer numbers are of concern, predation is common yet habitat quality appears to have declined. The question remains as to whether predation, habitat, or some other factor is more limiting to mule deer in these situations, and whether habitat quality can be improved for the benefit of deer. It may also be that no single factor is responsible for observed deer declines, and a more comprehensive understanding of multi-factor interactions is needed.

We designed and implemented a field experiment where we measured deer population responses to a nutrition enhancement treatment to further understand the causative factors underlying observed deer population dynamics. We conducted the study on the Uncompahgre Plateau in southwest Colorado, where several predator species were present in abundant numbers: coyotes (*Canis latrans*), mountain lions (*Felis concolor*), and bears (*Ursus americanus*). In addition to predation, myriad diseases in combination have proximately affected survival of the Uncompahgre deer population (Pojar and Bowden 2004, B. E. Watkins, Colorado Division of Wildlife, unpublished data). Predator numbers were not manipulated in any manner during the course of the study. All factors were left constant with the exception of deer nutrition. Deer nutrition was enhanced by providing supplemental feed to deer occupying a treatment area during winter. We measured December fawn recruitment and overwinter fawn survival in response to the treatment to determine whether deer nutrition was ultimately more limiting than predation or disease. A second phase of research was initiated in 2005 to quantify deer population parameters in response to manipulations of pinyon-juniper (*Pinus edulis*-*Juniperus osteosperma*) habitat (Bergman et al. 2006). The objective of this research is to determine whether habitat can be effectively improved for mule deer by introducing disturbance into late-seral pinyon-juniper stands.

STUDY AREA

We non-randomly selected two experimental units (A–B) within mule deer winter range on the Uncompahgre Plateau (Figure 1) to facilitate a cross-over experimental design for evaluating the effects of enhanced deer nutrition during winter on annual population performance. Unit A received a nutrition enhancement treatment during the first 2 winters of research (2000 – 2002) while Unit B served as a control unit. During winters 2002–03 and 2003–04, Unit B received the treatment while Unit A served as the control. In late April and May, prior to fawning, deer from the winter range experimental units migrated to summer range. We defined the summer range study area by movements of the radio-collared deer captured on winter range; summer range encompassed >1000 mi^2 covering the southern portion of the Uncompahgre Plateau and adjacent San Juan Mountains (Figure 2). Winter range elevations ranged from 1830 m (6000 ft) in Shavano Valley to 2318 m (7600 ft) adjacent to the Dry Creek Rim above Shavano Valley. Winter range habitat was dominated by pinyon-juniper with interspersed sagebrush adjacent to agricultural fields in the Shavano and Uncompahgre Valleys. Summer range elevations occupied by deer ranged from 1891 m (6200 ft) in the Uncompahgre Valley to 3538 m (11,600 ft) in Imogene Basin southwest of Ouray, CO. Summer range habitats were dominated by spruce-subalpine fir (*Picea* spp.-*Abies lasiocarpa*), aspen (*Populus tremuloides*), sagebrush, ponderosa pine (*Pinus ponderosa*), Gambel oak (*Quercus gambelii*), and to a lesser extent, pinyon-juniper at lower elevations. Bishop et al. (2005) provide a detailed study area description.

METHODS

Refer to Bishop et al. (2005) for field methodology employed during 2000–2005. During fiscal year 2006-07, we had 2 papers published in peer reviewed wildlife journals, which were the result of work completed in the previous year. Our primary research efforts were focused on data analysis and the preparation of manuscripts for publication in scientific journals. We spent much of the year evaluating dependence among deer siblings with respect to fetal and neonatal survival analyses. Essentially all statistical analyses are based on an assumption that sample units are independent. In survival analyses, the assumption pertains to independence of fates. That is, we must assume that the death or survival of one sample unit is not related to the fate of another. Predation and maternal condition are both good examples of mechanisms that could cause sibling neonates to lack independent fates. If a coyote or bear kills twin fawns because both were together, clearly those mortality events were not independent. Similarly, a lack of independence would occur if twin fawns each die of starvation because their dam is in poor condition. Data are considered overdispersed when the independence assumption is violated. Overdispersion does not generally affect point estimates, but rather causes variances to be underestimated. We estimated overdispersion in fetal and neonatal survival datasets and incorporated a data bootstrap procedure into Program MARK (White and Burnham 1999), making it easier for others to conduct similar analyses. The procedure in MARK can be generalized to any situation where multiple individuals are marked from the same litter, clutch, pair, trap site, etc. Once we completed the overdispersion analysis, we spent the remainder of the year conducting final data analyses that quantified the effect of enhanced nutrition on population performance. We then prepared several manuscripts that incorporated the various analyses we conducted. The principal investigator also completed a draft of his PhD dissertation.

RESULTS AND DISCUSSION

A comprehensive presentation and discussion of preliminary results was provided by Bishop et al. (2005). These results have not changed and therefore we do not repeat them here. The final results are contained in peer-reviewed manuscripts that have either already been published or will be submitted for publication in 2007 or early 2008. The following manuscripts were published in 2007 (abstracts are provided in Appendix I):

Bishop, C. J., D. J. Freddy, G. C. White, B. E. Watkins, T. R. Stephenson, and L. L. Wolfe. 2007. Using vaginal implant transmitters to aid in capture of mule deer neonates. Journal of Wildlife Management 71:945–954. .

Schultheiss, P. C., H. Van Campen, T. R. Spraker, C. J. Bishop, L. L. Wolfe, and B. Podell. 2007. Malignant catarrhal fever associated with ovine herpesvirus-2 in free-ranging mule deer in Colorado. Journal of Wildlife Diseases 43:533–537.

The following book chapter was completed should be published by January 2008:

Watkins, B. E., C. J. Bishop, E. J. Bergman, A. Bronson, B. Hale, B. F. Wakeling, L. H. Carpenter., and D. W. Lutz. 2007. Habitat guidelines for mule deer: Colorado Plateau shrubland and forest ecoregion. Mule Deer Working Group, Western Association of Fish and Wildlife Agencies.

The following draft manuscripts were prepared in 2007 and will be submitted for publication in 2007 or early 2008 (abstracts are provided in Appendix II):

Bishop, C. J., G. C. White, and P. M. Lukacs. In review. Evaluating dependence among mule deer siblings in fetal and neonatal survival analyses. Journal of Wildlife Management.

Bishop, C. J., G. C. White, D. J. Freddy, B. E. Watkins, and T. R. Stephenson. In review. Effect of enhanced nutrition on mule deer population performance. Journal of Wildlife Management OR Wildlife Monographs.

Bishop, C. J., B. E. Watkins, L. L. Wolfe, D. J. Freddy, and G. C. White. In review. Evaluating mule deer body condition during late winter using serum thyroid hormone concentrations. Journal of Wildlife Management.

The following draft dissertation was prepared in 2007 and submitted for review at Colorado State University (abstract is provided in Appendix III):

Bishop, C. J. In review. Effect of enhanced nutrition during winter on the Uncompahgre Plateau mule deer population. Dissertation, Colorado State University, Fort Collins, USA.

We intend to pursue several additional manuscripts as time allows, listed below in order of priority.

1. Evaluating dependence of fates among mule deer siblings in Colorado, Idaho, and Montana. Journal of Wildlife Management.
2. Bovine viral diarrhea isolation and seroprevalence in a free-ranging mule deer (*Odocoileus hemionus*) population in southwest Colorado. Journal of Wildlife Diseases.
3. Spatial patterns in mortality causes of neonatal mule deer across a land use gradient in southwest Colorado. Journal of Wildlife Management.

4. Evaluation of mule deer age and sex ratios as a response variable in field research. Journal of Wildlife Management.

SUMMARY

Enhanced winter nutrition of free-ranging deer caused an increase in both fetus-neonate survival and overwinter fawn survival, resulting in higher yearling recruitment. Overwinter adult doe survival increased as a result of the treatment, and therefore annual survival was higher among treatment than control adult does. Combining all parameter estimates into a deterministic population model, the treatment population indicated an exceptionally high rate of increase while the control population was stable and indicative of the overall Uncompahgre deer population during 2000–2004. The nutrition enhancement treatment was artificial in the sense that we applied it only to test whether habitat quality was ultimately more limiting than predation or other factors. Our results to do not provide support for managing deer populations with nutrition supplements because our treatment delivery approach could not be applied to a large number of animals over a large area. Rather, our results provide a foundation for focusing deer management efforts on improving habitat quality in western Colorado pinyon-juniper ecosystems with corresponding research efforts to quantify the effects of habitat manipulations on deer. We are presently in the process of conducting final data analyses and preparing and submitting manuscripts for publication in scientific journals.

LITERATURE CITED

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BISHOP, C. J., G. C. WHITE, D. J. FREDDY, AND B. E. WATKINS. 2005. Effect of nutrition and habitat enhancements on mule deer recruitment and survival rates. Federal Aid in Wildlife Restoration Project W–185–R, Job Progress Report. Wildlife Research Report, Colorado Division of Wildlife, Fort Collins, USA.

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Prepared by ________________________
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Year	Unit A	Unit B
2000-01	Treatment	Control
2001-02	Treatment	Control
2002-03	Control	Treatment
2003-04	Control	Treatment

Figure 1. Schematic representation of experimental units and nutrition enhancement treatment allocation. Units A and B were located in winter range habitat on the Uncompahgre Plateau in southwest Colorado. The nutrition enhancement cross-over design encompassed 4 years.

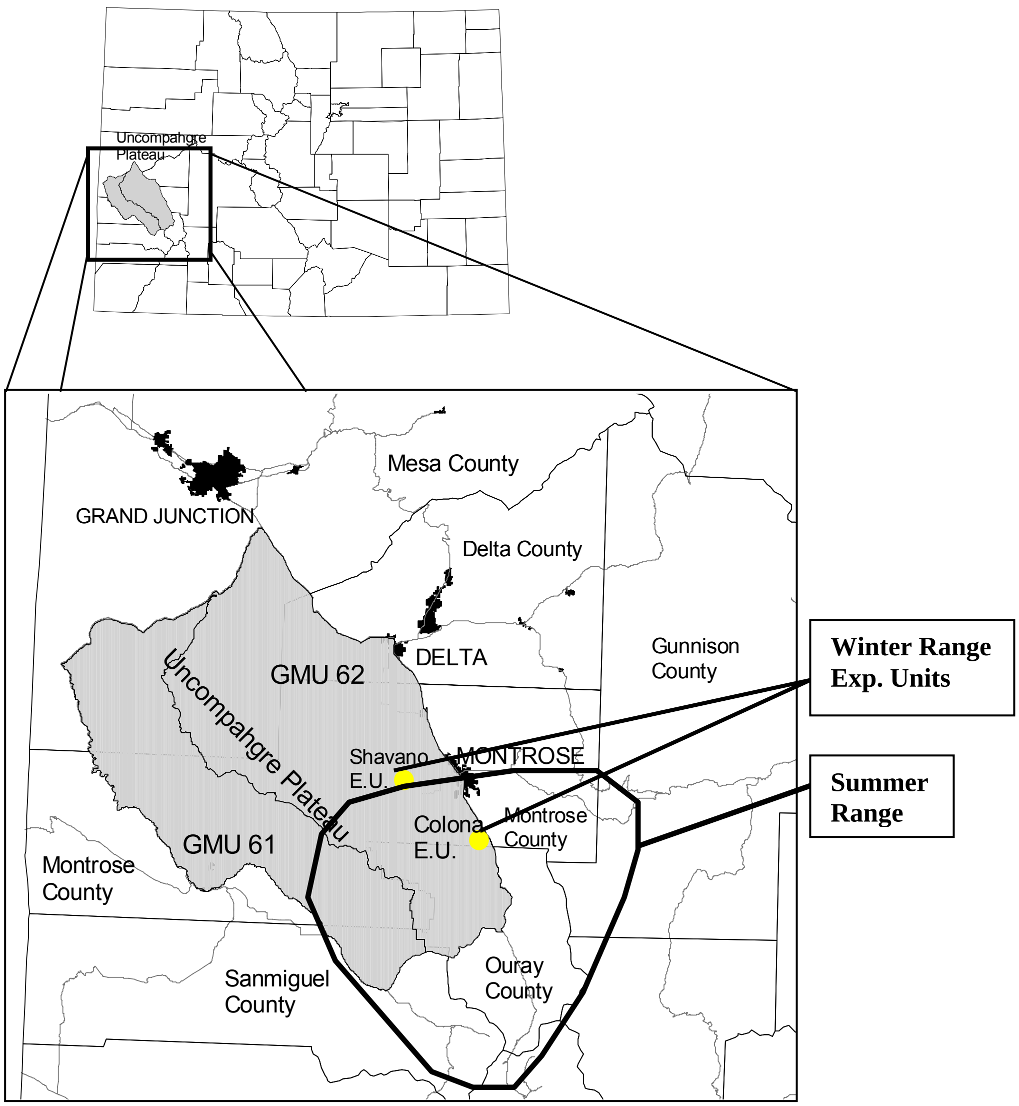

Figure 2. Location of Colona and Shavano (Units A and B) experimental units on the Uncompahgre Plateau, southwest Colorado; and location of the summer range study area encompassing the southern Uncompahgre Plateau and adjacent San Juan Mountains.

APPENDIX I

The following manuscript (referenced here by Abstract) was published in the Journal of Wildlife Management in 2007.

USING VAGINAL IMPLANT TRANSMITTERS TO AID IN CAPTURE OF MULE DEER NEONATES

CHAD J. BISHOP, DAVID J. FREDDY, GARY C. WHITE, BRUCE E. WATKINS, THOMAS R. STEPHENSON, AND LISA L. WOLFE

ABSTRACT

Estimating survival of the offspring of marked female ungulates has proven difficult in free-ranging populations yet could improve our understanding of factors that limit populations. We evaluated the feasibility and efficiency of capturing large samples (i.e., >80/year) of neonate mule deer (*Odocoileus hemionus*) exclusively from free-ranging, marked adult does using vaginal implant transmitters (VITs, n = 154) and repeated locations of radio-collared does without VITs. We also evaluated the effectiveness of VITs, when used in conjunction with in utero fetal counts, for obtaining direct estimates of fetal survival. During 2003 and 2004, after we placed VIT batteries on a 12-hour duty cycle to lower electronic failure rates, the proportion that shed ≤3 days prepartum or during parturition was 0.623 (SE = 0.0456), and the proportion of VITs shed only during parturition was 0.447 (SE = 0.0468). Our neonate capture success rate was 0.880 (SE = 0.0359) from does with VITs shed ≤3 days prepartum or during parturition and 0.307 (SE = 0.0235) from radio-collared does without VITs or whose implants failed to function properly. Using a combination of techniques, we captured 275 neonates and found 21 stillborns during 2002–2004. We accounted for all fetuses at birth (i.e., live or stillborn) from 78 of the 147 does (0.531, SE = 0.0413) having winter fetal counts, and this rate was heavily dependent on VIT retention success. Deer that shed VITs prepartum were larger than deer that retained VITs to parturition, indicating a need to develop variable-sized VITs that may be fitted individually to deer in the field. We demonstrated that direct estimates of fetal and neonatal survival may be obtained from previously marked female mule deer in free-ranging populations, thus expanding opportunities for conducting field experiments. Survival estimates using VITs lacked bias that is typically associated with other neonate capture techniques. However, current vaginal implant failure rates, and overall expense, limit broad applicability of the technique.

Citation: Bishop, C. J., D. J. Freddy, G. C. White, B. E. Watkins, T. R. Stephenson, and L. L. Wolfe. 2007. Using vaginal implant transmitters to aid in capture of mule deer neonates. Journal of Wildlife Management 71:945–954.

The following manuscript (referenced here by Abstract) was published in the Journal of Wildlife Diseases in 2007:

MALIGNANT CATARRHAL FEVER ASSOCIATED WITH OVINE HERPESVIRUS-2 IN FREE-RANGING MULE DEER IN COLORADO

PATRICIA C. SCHULTHEISS, HANA VAN CAMPEN, TERRY R. SPRAKER, CHAD J. BISHOP, LISA L. WOLFE, AND BRENDAN PODELL

ABSTRACT

Malignant catarrhal fever (MCF) was diagnosed in 4 free-ranging mule deer (*Odocoileus hemionus*) in January and February of 2003. Diagnosis was based on typical histologic lesions of lymphocytic vasculitis and PCR identification of ovine herpesvirus-2 (OHV-2) viral genetic sequences in formalin fixed tissues. The animals were from the Uncompahgre Plateau of southwestern Colorado. Deer from these herds occasionally resided in close proximity to domestic sheep *(Ovis aries)*, the reservoir host of OHV-2, in agricultural valleys adjacent to their winter range. These cases indicate that fatal OHV-2 associated MCF can occur in free-ranging mule deer exposed to domestic sheep that overlap their range.

Citation: Schultheiss, P. C., H. Van Campen, T. R. Spraker, C. J. Bishop, L. L. Wolfe, and B. Podell. 2007. Malignant catarrhal fever associated with ovine herpesvirus-2 in free-ranging mule deer in Colorado. Journal of Wildlife Diseases 43:533–537.

APPENDIX II

The following draft manuscripts (referenced here by Abstract) were prepared in 2007 and will be submitted to the Journal of Wildlife Management.

EVALUATING DEPENDENCE AMONG MULE DEER SIBLINGS IN FETAL AND NEONATAL SURVIVAL ANALYSES

CHAD J. BISHOP, GARY C. WHITE, AND PAUL M. LUKACS

ABSTRACT

The assumption of independent sample units is potentially violated in deer (*Odocoileus* spp.) fetal and neonatal survival analyses where twin and triplet siblings comprise a high proportion of the sample. Violation of the independence assumption causes sample data to be overdispersed relative to a binomial model, and therefore requires a variance inflation factor, *c*, to obtain appropriate estimates of sampling variances. We evaluated overdispersion in fetal and neonatal mule deer (*O. hemionus*) datasets where more than half of the sample units were comprised of siblings. We developed a likelihood function for estimating fetal survival when the fates of some fetuses are unknown, and we used several variations of the binomial model to estimate neonatal survival. We compared theoretical variance estimates obtained from these analyses with empirical variance estimates obtained from data bootstrap analyses to estimate the overdisperion parameter, *c*. Our estimates of *c* for fetal survival ranged from 0.678 to 1.118, which provided virtually no evidence of overdispersion. For neonatal survival, 3 different models indicated that $\hat{c}$ ranged from 1.1 to 1.4 and averaged 1.24–1.26, providing evidence of limited overdispersion (i.e., limited sibling dependence). Our results indicate that fates of sibling mule deer fetuses and neonates may often be independent even though they have the same dam. Predation tends to act independently on sibling neonates because of dam-neonate behavioral adaptations, although we observed several cases of a predator(s) killing siblings. The effect of maternal characteristics on sibling fate dependence is less straightforward and may vary by circumstance. We recommend that future neonatal survival studies incorporate additional sampling intensity to accommodate modest overdispersion (i.e., $\hat{c}$ = 1.25), which would facilitate a corresponding $\hat{c}$ adjustment in a model selection analysis using quasi-likelihood without a reduction in power.

EFFECT OF ENHANCED NUTRITION ON MULE DEER POPULATION PERFORMANCE

CHAD J. BISHOP, GARY C. WHITE, DAVID J. FREDDY, BRUCE E. WATKINS, AND THOMAS R. STEPHENSON

ABSTRACT

Concerns over declining mule deer (*Odocoileus hemionus*) populations during the 1990s prompted research efforts to identify and understand key limiting factors of deer. Similar to past deer declines, a top priority of state wildlife agencies was to evaluate the relative importance of habitat and predation. We therefore evaluated the effect of enhanced nutrition of deer during winter and spring on fecundity and survival rates using a field experiment involving free-ranging mule deer on the Uncompahgre Plateau in southwest Colorado. The nutrition enhancement treatment represented an instantaneous increase in nutritional carrying capacity of a pinyon (*Pinus edulis*) and Utah juniper (*Juniperus osteosperma*) winter range, and was intended to simulate optimum habitat quality. Prior studies on the Uncompahgre Plateau indicated predation and disease were the most common proximate causes of deer mortality. By manipulating nutrition and leaving predation as it was, we determined whether habitat quality was ultimately a critical limiting factor of the deer population. We measured fetal, neonatal, and overwinter fawn survival, and annual adult doe survival, which we then used to estimate population rate of change as a function of enhanced nutrition. Pregnancy and fetal rates were high for all deer, regardless of the nutrition treatment. Fetal and neonatal survival rates were higher among deer that received the nutrition enhancement treatment than deer that served as experimental controls. Overwinter fawn survival was considerably higher among treatment deer than control deer. Overwinter survival increased by 0.16–0.31 depending on year and fawn sex, and none of the 95% confidence intervals associated with the effect overlapped 0. The nutrition enhancement treatment increased survival of fetuses to the yearling age class by 0.14–0.20 depending on year and fawn sex, although 95% confidence intervals slightly overlapped 0. The nutrition treatment also had a positive effect on annual adult doe survival. Survival of does receiving the treatment ($\hat{S}$ = 0.879, SE = 0.0206) was higher than survival of control does ($\hat{S}$ = 0.833, SE = 0.0253). Our estimate of the population rate of change, $\hat{\lambda}$, was 1.15–1.17 for treatment deer and 1.02–1.06 for control deer, with some overlap in 95% confidence intervals. The treatment caused $\hat{\lambda}$ to increase by 0.139 (95% CI: –0.0152, 0.2941) during 2001–02, 0.113 (95% CI: –0.0009, 0.2279) during 2002–03, and 0.145 (95% CI: 0.0176, 0.2723) during 2003–04. We documented density dependence in the Uncompahgre deer population because survival of fawns and does increased considerably in response to enhanced nutrition. We found strong evidence that coyote (*Canis latrans*) predation of ≥6 month old fawns and adult does was compensatory. Our results demonstrate that observed coyote predation is not useful for evaluating whether coyotes are negatively impacting a deer population. We also found evidence that mountain lion (*Puma concolor*) predation was compensatory. Disease was not compensatory among adult does. We found that winter range habitat quality was a limiting factor of the Uncompahgre Plateau deer population. We recommend the implementation and evaluation of habitat treatments designed to set back succession and increase productivity of late-seral pinyon-juniper habitats that presently dominate the landscape because of the absence of fire.

EVALUATING MULE DEER BODY CONDITION DURING LATE WINTER USING SERUM THYROID HORMONE CONCENTRATIONS

CHAD J. BISHOP, BRUCE E. WATKINS, LISA L. WOLFE, DAVID J. FREDDY, AND GARY C. WHITE

ABSTRACT

Body condition of ungulates is ultimately a determinant of fecundity and survival rates. Researchers have recently developed ultrasonography and body condition scoring techniques that allow reliable estimation of body fat in several ungulate species, but the approach is not feasible to employ in all circumstances, particularly in routine population monitoring programs. There remains a need for a reliable blood chemistry index that could be used to assess relative condition of different deer populations or groups. We evaluated the relationship between estimated body fat of free-ranging mule deer and serum concentrations of total thyroxine (T4), total triiodothyronine (T3), free T4 (FT4), and free T3 (FT3), during late February–early March in southwest Colorado. Deer body fat varied widely because we imposed a nutrition treatment on one-half of our sample. Concentrations of T4 and FT4 were 48.23 nmol/l (SE = 3.88) and 12.69 pmol/l (SE = 1.12) higher, respectively, in deer that received the nutrition treatment than deer that did not receive the treatment. Our optimal model of estimated body fat included T4, $T4^2$, FT4 and deer chest girth ($\%\hat{Fat} = -4.8015 - 0.0946 \times T4 + 0.000603 \times T4^2 + 0.1474 \times FT4 + 0.1426 \times$chest girth, $r^2 = 0.609$). Ultrasound and body condition scoring should be used to estimate body fat whenever possible. However, in cases where only a blood sample can be obtained, we documented the potential utility of T4 and FT4 during late winter for evaluating relative body condition of mule deer.

APPENDIX III

The following draft dissertation (referenced here by Abstract) was prepared in 2007 and will be submitted to Colorado State University.

EFFECT OF ENHANCED NUTRITION DURING WINTER ON THE UNCOMPAHGRE PLATEAU MULE DEER POPULATION

CHAD J. BISHOP

ABSTRACT

Mule deer (*Odocoileus hemionus*) populations declined across much of the West during the 1990s, prompting state wildlife agencies to pursue explorations of mule deer limiting factors. The greatest concern of agencies and sportsmen was whether declining habitat quality or predation was ultimately responsible for the observed declines. In Colorado, the Uncompahgre Plateau mule deer population received the most attention, having substantially declined from the 1980s through the late 1990s. Biologists hypothesized that poor winter range habitat quality was the primary cause of the observed decline. In contrast, many of the Colorado Division of Wildlife's (CDOW) external constituents hypothesized that high rates of predation were keeping the mule deer herd below nutritional carrying capacity. The habitat quality hypothesis indicated CDOW should pursue habitat improvements in the pinyon (*Pinus edulis*) and juniper (*Juniperus osteosperma*) winter range, whereas the predator hypothesis suggested CDOW should pursue efforts to reduce predator populations, particularly coyote (*Canis latrans*) populations. The competing hypotheses represented very different paradigms of population limitation. I therefore evaluated the effect of enhanced nutrition during winter on the Uncompahgre deer population as a way to evaluate the importance of habitat quality versus that of predation.

I conducted a field study incorporating a crossover experimental design to quantify the effect of enhanced nutrition on fetal, neonatal, overwinter fawn, and annual adult doe survival rates. I captured and radio-collared samples of deer in 2 experimental units (EUs) on winter range. I delivered the nutrition treatment to deer occupying one EU (treatment) and did not administer the treatment to deer in the other EU (control). Established field techniques were not sufficient to allow me to quantify the effect of the treatment on fetal and neonatal survival. I therefore pursued an exploration of vaginal implant transmitters as a mechanism to capture necessary samples of newborn fawns on summer range exclusively from radio-collared does that occupied the winter range EUs (Chapter 1). This effort allowed me to estimate fetal and neonatal survival as a function of the treatment. In broad terms, I demonstrated that direct estimates of fetal and neonatal survival may be obtained from previously marked female mule deer in free-ranging populations, thus expanding opportunities for conducting field experiments.

I encountered additional challenges with estimation of fetal and neonatal survival. First, I was unable to determine the fate of all fetuses that I documented in utero. I therefore developed a likelihood function for estimating fetal survival when the fates of some fetuses are unknown (Chapter 2). Second, a majority of my fetal and neonatal samples were comprised of siblings, indicating my data were potentially overdispersed. Overdispersion causes sample variances to be underestimated and requires a variance inflation factor, c. To estimate c, I compared theoretical variance estimates with empirical variance estimates obtained from data bootstrap analyses (Chapter 2). I found little evidence of overdispersion in my fetal survival data, and I found modest overdispersion in my neonatal sample data ($\hat{c}$ = 1.25). Although some overdispersion was detected, my result indicated that fates of sibling mule deer neonates may often be independent even though they have the same dam and use the environment similarly. I discuss reasons for this in Chapter 2.

After resolving issues with fetal and neonatal survival estimation, I quantified the effect of the nutrition enhancement treatment on fetal, neonatal, overwinter fawn, and annual adult doe survival (Chapter 3). I then used these parameter estimates, along with estimated fecundity rates, in an age-structured, deterministic population model to estimate the effect of the treatment on the population rate of change, λ. The treatment caused $\hat{\lambda}$ to increase by an average of 0.133 (SD = 0.0168) during the 3 years of my study. I documented density dependence in the Uncompahgre deer population because survival of fawns and does increased considerably in response to enhanced nutrition. I found strong evidence that coyote predation of ≥6-month-old fawns and adult does was compensatory. Finally, I found that winter range habitat quality was a limiting factor of the Uncompahgre Plateau deer population.

I completed my principal study objectives in the first 3 chapters of the dissertation. However, my research afforded the opportunity to evaluate the utility of serum thyroid hormones in mule deer as an index to body condition (Chapter 4). Concentrations of total thyroxine (T4) and free T4 (FT4) were substantially higher in treatment deer than control deer. I also found that serum thyroid hormones were highly correlated with estimated body fat in mule deer during late winter. Concentrations of T4 and FT4 could be useful for evaluating relative condition of different deer groups or populations, and for roughly estimating body fat of individual animals during late winter.

n summary, I demonstrated that winter range habitat quality was ultimately limiting the Uncompahgre mule deer population. Observed predation was primarily compensatory, particularly of ≥6-month-old fawns and adult does. My findings indicate that CDOW should implement and evaluate habitat treatments in late-seral pinyon-juniper habitat as a means to increase habitat productivity for mule deer. My findings provide no support for predator reduction programs.

Colorado Division of Wildlife
July 2006 – June 2007

WILDLIFE RESEARCH REPORT

State of	Colorado	: Division of Wildlife
Cost Center	3430	: Mammals Research
Work Package	3001	: Deer Conservation
Task No.	2	: Evaluation of Winter Range Habitat Treatments on Over-Winter Survival and Body Condition of Mule Deer.
Federal Aid Project	W-185-R	:

Period Covered: July 1, 2006 - June 30, 2007

Author: E.J. Bergman; project cooperators, C.J. Bishop, D.J. Freddy and G.C. White

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All information in this report is preliminary and subject to further evaluation. Information MAY NOT BE PUBLISHED OR QUOTED without permission of the author. Manipulation of these data beyond that contained in this report is discouraged.

ABSTRACT

We completed the second year of a multi-year, multi-area study to assess the impacts of landscape level winter range habitat improvement efforts on mule deer population performance. This study is occurring on the Uncompahgre Plateau and in adjacent valleys in southwestern Colorado. Data collection and analysis for this second year were consistent with that of the pilot study and first year of the study. We measured over-winter fawn survival and total deer density on 4 annual study areas, as well as on a fifth variable area that had previously not been involved in the study. Additionally, on 2 of the study areas we estimated body condition of does. Compared to results from other research throughout the west, as well as on the Uncompahgre Plateau, survival estimates for 6-month old mule deer fawns were highly variable between areas, but tended to be above average (mean survival rate of 0.76 (0.04 SE)). Preliminary evidence suggests that areas that have received habitat treatments have higher survival. However, based on estimates of total body fat for adult female deer, there was no apparent distinction between treatment and reference study areas. Point estimates of deer density on the 5 study areas during the winter of 2006-2007 were consistent with those colleted during the winter of 2005-2006. Attempts to refine and improve the precision of density estimates between the two winters were marginally successful and will be further refined during the next year of data collection. During the 2006-2007 winter, we also initiated a pilot study concerning elk spatial use as it relates to competition with deer as well as predator-prey dynamics. Due to the timing of the study, these data have will not be retrieved from elk GPS collars until fall 2007.

WILDLIFE RESEARCH REPORT

EVALUATION OF WINTER RANGE HABITAT TREATMENTS ON OVER-WINTER SURVIVAL AND BODY CONDITION OF MULE DEER

ERIC J. BERGMAN

P.N. OBJECTIVE

To experimentally assess whether mechanical/chemical treatments of native habitat vegetation will increase over-winter mule deer fawn survival, adult doe body condition, and localized deer densities on Uncompahgre Plateau in southwest Colorado.

SEGMENT OBJECTIVES

1. Capture and radio-collar the minimum necessary sample (n=25) of 6 month-old fawns during November through early January in each of 5 study areas.
2. Measure over-winter fawn survival from mid-December through mid-June.
3. Estimate late-winter deer densities in each study area via helicopter resighting of marked deer.
4. Capture and sample a minimum number of adult female deer (n=30) to estimate late-winter body condition in 2 study areas.
5. Capture and radio-collar 10 adult female elk with GPS or VHF collars.

INTRODUCTION

A common trend among many terrestrial, mammalian systems is a tendency to cycle between population highs and lows (Jedrzejewska and Jedrzejewski 1998, Krebs et al. 2001, Clutton-Brock and Pemberton 2004). While the true cause of these cycles is likely a merger of habitat quality, weather, disease, predation, sport hunting, competition and community population dynamics, it is often necessary or intriguing for wildlife managers and ecologists to identify the primary limiting factor to population growth. Without exception, mule deer populations have also demonstrated a tendency to show large fluctuations. Several dramatic declines have been observed since the turn of the 19th century (Connolly 1981, Gill 2001, Hurley and Zager 2004). However, only one period of increase, a general trend during the 1940's and 1950's, has been noted. The most recent and pressing decline took place during the 1990's (Unsworth et al. 1999). Colorado has not escaped these tendencies, with certain parts of the state experiencing population declines by as much as 50% between the 1960's and present time (Gill 2001, B. Watkins personal communication). Primarily due to the value of mule deer as a big game hunting species, wildlife managers' challenges are two-fold: understanding the underlying causes of mule deer population change and managing populations to dampen the effects of these fluctuations.

In Colorado, the role of habitat as the limiting factor for mule deer populations was recently tested. Specifically, the role of forage quality and quantity on over-winter fawn survival was tested using a treatment/reference cross-over design with *ad libitum* pelleted food supplements as a substitute for instantaneous high quality habitat improvements (Bishop et al. 2004). The primary hypothesis behind this research concerned the interaction between predation and nutrition. If supplemental forage treatments improved over-winter fawn survival (i.e. if predation did not prevent an increase), then it could be concluded that over-winter nutrition was the primary limiting factor on populations. As such, preliminary evidence suggests that nutrition enhancement treatments increased fawn survival by as much as 20% (C.J. Bishop, personal communication). This research effectively identified some of the underlying processes in mule deer population regulation, but did not test the effectiveness of acceptable habitat management techniques. Due to the undesirable effects of feeding wildlife (e.g. artificially elevating density, increased potential for disease transmission and cost), a more appropriate technique for achieving a high quality nutrition enhancement needs to be assessed.

Based on this past research and the above mentioned objectives, we designed and initiated a multi-year, multi-area study to assess the impacts of landscape level winter range treatments on mule deer population performance. This study is being conducted on the Uncompahgre Plateau and adjacent valleys in southwestern Colorado. Due to the active habitat treatment history in this area, the Uncompahgre Plateau stood out as the most opportune place for addressing these issues. Additionally, there are several tracts from 2 state wildlife areas that are located in key locations, thereby allowing additional habitat treatments to occur on the level and schedule necessary of this project. To assess the impacts of habitat treatments on mule deer in these areas, we are measuring over-winter fawn survival, mule deer density and late winter body condition.

In addition to the above mentioned objectives, the opportunity to explore deer/elk interactions, as well as predator-prey dynamics is available in our study areas. As part of a pilot study to assess these interactions, we distributed elk GPS collars across the south end of the Uncompahgre Plateau where the density of radio-marked deer and mountain lions is highest (for further details on this pilot work, see Appendix I). Preliminary data will give basic information regarding elk distribution throughout the year, which can then be compared to similar data for deer and the spatial distribution of mountain lion kill sites.

STUDY AREA

At the onset of this study (Bergman et al. 2005), we identified 2 pairs of treatment/reference study areas, stratified into historically known high and low deer density areas. The selection process for these pairs of experimental units followed several strict guidelines:

1) Treatment/reference units could not be further than 10km apart, but needed to have adequate buffer to minimize the movement of animals between the treatment and reference areas.
2) Reference study areas could not have received any mechanical treatment during the past 30 years.
3) Strata were defined by winter range type (all experimental units had to be in pinyon/juniper winter range) and deer density.
4) Treatment units needed to have received mechanical treatment in the past, but also had to be capable of receiving further treatments during the study period.

Each winter a 5th study area is added to increase the level of inference that can be drawn from this study. For each of the 4 winters that will cover the study period, this 5th study area shifts between 4 randomly selected areas. The treatment history on each of these additional study areas varies, but is representative of what can be expected of typical winter-range treatments. During the second winter of this study, this 5th study area fell on the Colona Tract (~5km^2) of Billy Creek State Wildlife Area (approximately 15km south of Montrose, CO). The treatment history of Colona Tract is primarily composed of brush mowing and chemical control of weeds and dry land fertilization of preferred species.

The high density treatment area is located on the Billy Creek tract of Billy Creek State Wildlife Area (approximately 20km south of Montrose, CO). The high density reference area is located around Beaton Creek (approximately 15km south of Montrose, CO and approximately 5km north of Billy Creek State Wildlife Area). Both of the high density study areas are located in GMU 65 (DAU D-40). The low density treatment area is located on Peach Orchard Point, on/near Escalante State Wildlife Area (approximately 25km southwest of Delta, CO). The low density reference area is located on Sowbelly and Tatum draws (approximately 25km west of Delta, CO and approximately 8km from Peach Orchard Point). Both of the low density study areas are located in GMU 62 (DAU D-19). Shavano Valley was also located in GMU 62 (DAU D-19) to the west of Montrose, CO.

METHODS

Twenty-five mule deer fawns were captured and radio-collared in each of the 5 study areas. Fawns were captured via baited drop-nets (Ramsey 1968, Schmidt et al. 1978, Bartmann et al. 1992) and helicopter net-gunning (Barrett et al. 1982, van Reenen 1982) between mid-November and late-December. Ten adult female elk were captured via helicopter net-gunning during this same period. Fawns were fitted with radio collars made of vinyl belting and equipped with mortality sensors, which after remaining motionless for 4 hours, increase the pulse rate of received signals. To make fawn collars temporary, one end of the collar was cut in half and reattached using rubber surgical tubing; fawns shed the collars after approximately 6 months. Elk were collared with either permanent VHF collars or temporary GPS collars that were fitted with timed blow-off devices.

On a daily basis, from December through May, we monitored the radioed fawns in order to document live/death status. This allowed us to determine accurately the date of death and estimate the proximate cause of death. Daily monitoring was done from the ground to maximize efficient collection of mortalities and assessment of cause specific mortality. Weekly aerial telemetry flights were conducted to insure that all deer were heard at least once a week, allowing weekly survival estimates for each study area.

Additionally, throughout the winter field season and as part of a related pilot field study, investigations of mountain lion GPS clusters were conducted (see Appendix I for details).

To estimate body composition, an additional 30 adult female deer were captured via helicopter net-gunning and fitted with temporary radio-collars, also having mortality sensors, in late-February within each of the 2 high density study areas. For body condition work, we relied on methods that employed the use of ultrasonography to estimate total body fat (Stephenson et al. 1998, Cook 2000, Stephenson et al. 2002). Blood samples were also collected for endocrinology and pregnancy tests.

During late winter (early-March) we estimated deer density on each of our study areas. Helicopter based mark-resight techniques were used for density estimation (Gill 1969, Bartmann et al. 1986, Kufeld et al. 1980, Freddy et al. 2004).

Preliminary survival analyses were conducted on the first two years of data. In addition to including individual covariates (fawn sex and mass), we explored the role of habitat treatment history on survival. Due to the preliminary nature of these analyses and the ongoing status of the habitat treatment work, we did not attempt to rank individual study areas. Rather, our analyses were conducted such that areas were included and compared using three different approaches. With the first approach, areas were included individually and a unique survival rate was calculated for each area. The second approach allowed for 3 levels of habitat treatment intensity (untreated, single treatment or ongoing treatments). The final approach did not attempt to segregate treated areas by treatment history. Rather, any area with any treatment history was treated similarly, resulting in a unique survival rates being calculated for untreated (reference) areas and a different, unique survival rate being calculated for all other areas.

All survival models were conducted in program MARK (White and Burnham 1999). Known-fate models were tested using the logit link function. All models are compared using Akaike's Information Criterion corrected for small sample size (Burnham and Anderson 2003).

RESULTS AND DISCUSSION

Minimum necessary sample sizes were met in all study areas for all components of this research (n = 25 fawns per area for survival work, n = 30 adult females in two areas for body condition assessment, n=10 adult female elk for pilot data collection). Capture related mortalities occurred on 1 of 186 occasions (0.5%, 1 fawn). Two fawns died of unknown causes within 1week of capture and were censored from the survival analysis. Mean mass of all fawns was 36.5 kg (Table. 1).

Estimates of fawn survival collected during this study have been above average compared to results from other research throughout the west, as well as on the Uncompahgre Plateau. Across our 5 study areas, estimated survival rates ranged between 0.63 (0.10 SE) and 0.92 (0.05 SE), with mean survival rate of 0.76 (0.04 SE) (Table 2). While these rates are lower than those measured during the 2005-2006 winter, they remain higher than long term averages reported in the literature (Unsworth et al. 1999). Of particular note, survival rates in our low-density study areas have been exceptionally high during each winter of this study. Based on historical climate trends across the Uncompahgre Plateau, it is apparent that the stratification of treatment/reference study areas by density also captured a regional effect of climatic variation on over-winter fawn survival.

Preliminary survival models indicate that the individual parameter most influencing over-winter fawn survival continues to be fawn mass (Table 3). Of particular interest to this study, however, is that incorporation of treatment history into model structure improved performance beyond that of models that only incorporate individual covariates. While ΔAIC_c scores reflect minimal differentiation between models that incorporate treatment history under different strategies, ΔAIC_c scores for models that do not include treatment history and estimate survival rates for each individual area are noticeably higher. When categorized by treatment intensity, 6 study areas are split into 3 categories. When categorized as treatment/reference, 6 study areas are split into 2 categories. Yearly effects on survival rates appear to have no meaningful contribution to model performance. While model results indicate that habitat treatments have a positive influence on survival, standard errors for parameter estimates remain high, effectively dampening any conclusions that can be drawn This is an artifact of the preliminary nature of these analyses and is ultimately linked to having collected only half of the necessary data. As the study progresses and more study areas are included, a treatment intensity effect is likely to be detected if it exists.

Late winter body condition estimates for adult females during the winter of 2006-2007 were again higher than those collected during previous winters on the Uncompahgre Plateau (Bishop et al. 2004 and C.J. Bishop, personal communication). However, based on estimates of total body fat, there was no apparent distinction between our study areas. This apparent lack of distinction was also present in the levels of the T3 and T4 hormone (nmol/l) observed between study areas (Table 4). Differences in pregnancy rates, based on PSPB, between study areas were not observed. Overall, pregnancy rates were high in both study areas (Billy Creek = 27/30, Buckhorn = 25/28). Titers for BT and EHD were observed at low/moderate rates in Billy Creek (BT = 4/21, EHD = 2/21). Samples were not collected at Buckhorn.

During the 2006-2007 winter, efforts were made to improve precision within our density estimation techniques. Specifically, the number of helicopter resight surveys was increased and attempts were made to increase the total number of marks in the population via temporary marking with paint pellets. It was found that marking deer with pellets during the time period of our flights (late-March) was not a viable mechanism for increasing the number of marks in the population. On 5 markings attempts, which accounted for 90 hours of effort, no deer were successfully marked. The lack of success in these attempts was primarily attributed to our inability to get deer to respond to bait sites due to increasing availability of new natural forage. To be successfully marked, deer needed to be hit with 5-10 pellets on the dorsal region of their body. In order to successfully mark deer with this many pellets, deer needed to be within a 50-80 foot range. During early-winter months (late-November through early-January), it is not difficult to bring large groups of deer within this range as natural food resources are typically restricted by snow cover. However, during late-winter months natural food resources such as grass and browse species were becoming increasingly available. In contrast to this, the number of helicopter resight flights was increased by 2 on all study areas. Comparison of precision for density estimates between 2005-2006 and 2006-2007 showed an overall improvement during the second winter (i.e. range of 95% confidence intervals were reduced) by 31%-35% via increasing the number of resight occasions (Figure 1). However, while not quantifiable, it is believed that further increasing the number of resight occasions would likely not result in substantial further improvements. The use of helicopter resights as a density estimation technique is dependent on the behavior that deer exhibit as a helicopter flies overhead. Sightability of deer is greatly improved by deer movement. With the addition of more flights during this past winter, deer appeared to adapt a behavioral strategy of standing under trees rather than running. While this behavior cannot be quantified, if actually occurring, it introduces bias into the data collection technique. As such, we conclude that increasing the number of resight occasions can improve precision, but there is a point of diminishing returns that can quickly be surpassed as deer grow accustomed to helicopter disturbance.

Due to the timing of the pilot work on elk distribution, and to the timing of the elk GPS collar release mechanisms, elk location data will not be collected and analyzed until fall of 2007. Also due to timing of pilot work on predator/prey interactions, analysis of mountain lion GPS cluster data will not occur until fall 2007.

SUMMARY

Survival rates for mule deer fawns across our study areas averaged 76% with a measured high of 92% and measured low of 62%. Overall body condition parameter estimates for late-winter adult female deer were moderate to high, highlighting the general mild winter conditions that were observed throughout deer winter range in southwestern Colorado. Preliminary evidence of higher deer survival in treatment areas was observed, but we do not have enough data to draw strong conclusions at this preliminary stage. Estimates of total deer density across our study areas are in line with historical estimates. Precision of density estimates have improved with modification to techniques and additional years of data collection will be needed to determine if habitat treatment effects can potentially be detected.

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Table 1. Mean mass (n) and sex of mule deer fawns captured on the Uncompahgre Plateau from late-November through early-January of each year, 2005-2006 and 2006-2007. All fawns were captured by baited drop-nets or helicopter net-gunning. Mass is reported in kg.

Area	Year	Male	Females	Total
Billy Creek	2005	37.1 (14)	32.0 (11)	34.9 (25)
Buckhorn	2005	37.4 (11)	35.0 (15)	36.0 (26)
Colona	2005	39.4 (11)	37.2 (14)	38.2 (25)
Peach Orchard	2005	37.0 (11)	35.3 (14)	36.1 (25)
Sowbelly	2005	37.1 (16)	34.2 (9)	36.1 (25)
Billy Creek	2006	38.3 (12)	34.4 (12)	36.5 (25)
Buckhorn	2006	36.7 (10)	34.7 (15)	35.5 (25)
Colona	2006	38.1 (12)	32.5 (12)	35.4 (24)
Peach Orchard	2006	37.0 (13)	35.5 (12)	36.3 (25)
Sowbelly	2006	44.3 (8)	35.5 (15)	38.7 (25)

Table 2. Over-winter mule deer fawn survival rates for study areas across the Uncompahgre Plateau, for both winters of the study. Billy Creek, Peach Orchard, Colona and Shavano represent treatment areas. Buckhorn and Sowbelly are reference areas. Peach Orchard and Sowbelly are considered low-density study areas. Sample size equals 25 fawns in each area.

	2005-2006	2006-2007
Area	$\hat{S}$ (S.E.)	$\hat{S}$ (S.E.)
Billy Creek	0.83 (0.76)	0.72 (0.09)
Buckhorn	0.76 (0.88)	0.63 (0.10)
Colona	N.A.	0.68 (0.09)
Shavano	0.76 (0.85)	N.A.
Peach Orchard	0.88 (0.65)	0.92 (0.05)
Sowbelly	1.00 (0.00)	0.88 (0.07)
Other	0.83 (.108)	N.A.

Table 3. Preliminary survival model results for radio collared fawns on the Uncompahgre Plateau from the winters of 2005-2006 and 2006-2007.

Model	AICc	ΔAICc	ω_i
Treatment Reference + Mass	544.784	0.00	0.294
Treatment Reference + Mass + Sex	544.954	0.17	0.270
Treatment Type + Mass	545.592	0.81	0.196
Treatment Type + Mass + Sex	545.860	1.08	0.172
Area + Mass	548.908	4.12	0.037
Area + Mass + Sex	549.877	5.09	0.023
Area	555.220	10.44	0.002
Treatment Reference	555.279	10.50	0.002
Treatment Type	555.615	10.83	0.001
Area + Year	555.655	10.87	0.001
Treatment Reference + Year	556.371	11.59	0.000
Treatment Type + Year	556.426	11.64	0.000

Table 4. Late-winter body condition estimates for female adult mule deer on the Uncompahgre Plateau in 2 study areas each year, 2005-2006 and 2006-2007. Sample sizes were 30 does in each area. Mean T3 and T4 samples are reported in nmol/l. Parameters marked with an asterisk designate a significant difference at the 0.05 level.

	Parameter	Billy Creek	Buckhorn	Sowbelly
	% Body Fat	8.80% (2.02 S.E.)	N.A.	9.81% (2.88 S.E.)
2005-2006	T3*	1.12 (0.28)	N.A.	1.41 (0.51 S.E.)
	T4	70.69 (20.94)	N.A.	79.97 (15.80 S.E.)
	% Body Fat	7.61% (1.94 S.E.)	7.03% (1.80 S.E.)	N.A.
2006-2007	T3	1.55 (0.53)	1.42 (0.31)	N.A.
	T4	88.23 (19.53)	78.07 (22.34)	N.A.

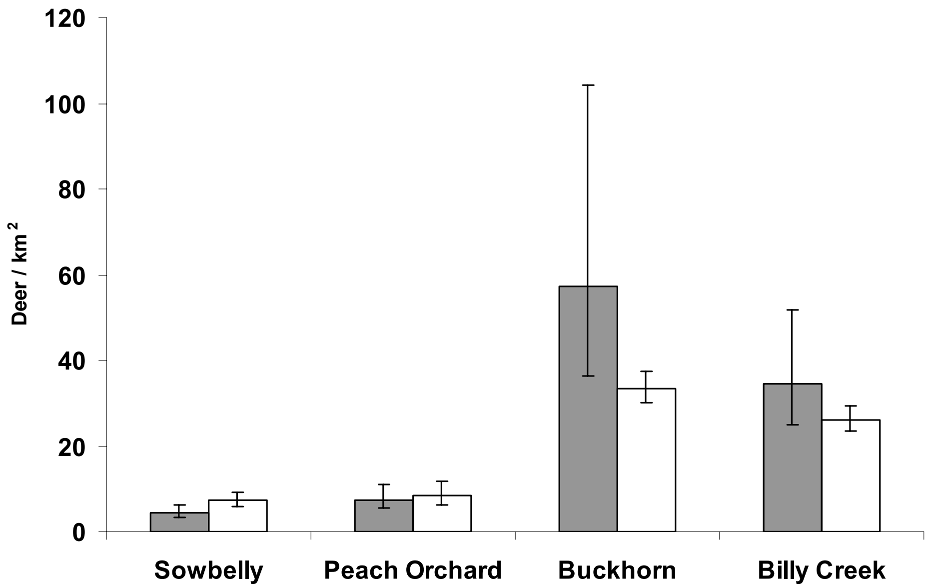

Figure 1. Deer density estimates for the 4 permanent study areas. Dark boxes reflect data from the 2005-2006 winter, white boxes reflect data from the 2006-2007 winter. Error bars represent the 95% confidence intervals for density estimates.

APPENDIX I

PROGRAM NARRATIVE PILOT STUDY PLAN
FOR MAMMALS RESEARCH
FY 2006-07

State of:	Colorado	: Division of Wildlife
Cost Center:	3430	: Mammals Research
Work Package:	3001	: Deer Conservation
Task No.:	3	: Pilot Evaluation of Predator-Prey Dynamics on the Uncompahgre Plateau
Federal Aid Project No.:	W-185-R	:

Pilot Evaluation of Predator-Prey Dynamics on the Uncompahgre Plateau.

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Pilot Study Plan Approval

Prepared by: Eric J. Bergman	Date: November 2006
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Reviewed by: ______________	Date: ______________
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Reviewed by: ______________ Biometrician	Date: ______________
Approved by: David J. Freddy, Mammals Research Leader	Date: November 2006

PROGRAM NARRATIVE PILOT STUDY PLAN

PILOT EVALUATION OF PREDATOR-PREY DYNAMICS ON THE UNCOMPAHGRE PLATEAU.

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NEED

Predator prey interactions have always been a topic of keen interest for wildlife managers and ecologists. However, due to the complexities of studying natural systems, behavioral theories pertaining to the subject are often developed in invertebrate, aquatic or small mammal systems, often under controlled laboratory conditions (Mathews et al. 2006, Schmitz 2006, Werner and Peacor 2006). Similarly, many models are developed within theoretical frameworks (Keeling et al. 2000, Mitchell and Lima 2002). While developing theories under these conditions is almost inherently necessary, their subsequent transition to free ranging systems is not frequent (Ryall and Fahrig 2006). Of the free ranging systems where theories are developed and tested, most deal with avian species (Lima and Bednekoff 1999, Roth et al. 2006), where as application to large mammalian systems is less frequent. Of the mammalian predator prey systems that have been studied, most have been conducted in preservation/park settings that largely exclude human influence (Kunkel and Pletscher 1999, Kunkel et al. 1999, Krebs et al. 2001, Creel and Creel 2002, Mao et al. 2005, Wilmers et al. 2006,). Additionally, due to the small number of large scale studies that have been conducted, the ability of managers to draw inference to separate systems (i.e. different species or different ecosystems) is limited. While this existing body of work is invaluable, extrapolation of theories to large mammalian systems could be limited and basing wildlife management decisions on this information may be tenuous.

Due to the value of mule deer, elk and mountain lions as recreationally hunted species in Colorado, there is much interest in understanding the nature and relationship between the population dynamics of these species. However, resulting from the dearth of information pertaining to the interactions of these 3 species, a vast array of opinions and theories pertaining to their impacts on each other have been propagated. As a management agency, the Colorado Division of Wildlife is responsible for supporting or refuting claims with biological data that were collected in a scientifically unbiased manner. To date, these data are largely unavailable.

Currently, the opportunity to develop a predator prey study exists on the Uncompahgre Plateau in southwestern Colorado. Two large scale research programs, independently studying mountain lion and mule deer, are underway in the same geographic area. Thus, the initial framework to study a top carnivore, and what are thought to be its primary prey species, is in place. However, to date there is little or no information pertaining to elk distribution or population dynamics in this area. The addition of elk spatial data will allow us to assess the feasibility of developing a full study addressing the influence and interactions of mountain lions, mule deer, and elk.

OBJECTIVES

The specific objectives for our pilot study are: 1) design and implement a pilot study that assesses mountain lion space use in relation to mule deer and elk distribution, 2) design and implement a sampling protocol that allows estimation of mountain lion use rates of deer and elk, 3) quantify prey selection patterns (number of elk versus number of deer), and 4) evaluate the feasibility of expanding this into a full study.

EXPECTED RESULTS

As part of the ongoing mountain lion research program, adult mountain lions have been captured and collared with GPS radio collars (Logan 2005). These collars are programmed to attain GPS locations 4 times per day. By design, GPS locations can be remotely downloaded from these radio collars via UHF signal by researchers at any chosen time. Through the investigation of GPS clusters of lion locations, a preliminary assessment of prey selection and ungulate use rates can be made (Anderson 2003, Logan 2005).

Additionally, as part of the ongoing mule deer research program, quantification of mule deer survival rates is also being done. With nominal, additional flight time, distributions of radio collared deer can be estimated at bi-weekly intervals. Finally, the addition of 6-12 radio (VHF and GPS) collars on elk will allow us to collect elk group locations and distribution data at the same interval as those collected for mule deer.

As a result of simultaneously collecting these data for all three species, we expect to make a preliminary assessment of the basic interaction behaviors of mountain lions and their 2 primary prey species on the Uncompahgre Plateau. Primarily, we expect to estimate kill rates for mountain lions, quantify prey selection behavior in light of prey distribution, and gather information on prey switching behavior by mountain lions.

APPROACH

Capture and Handling Methods: To date and as part of a completed mule deer study, approximately 150 adult female mule deer are marked with VHF radio collars in the area of interest (Bishop et al. 2005). Additionally, 25 mule deer fawns will be captured and radio-collared within the eastern portion of the study area between late-November and late-December 2006 as part of the ongoing mule deer research (Bergman et al. 2005; capture protocols previously approved by CDOW ACUC). All mule deer were, or will be, captured with baited drop-nets (Ramsey 1968, Schmidt et al. 1978, Bartmann et al. 1992) or via helicopter net-gunning (Barrett et al. 1982, van Reenen 1982). As part of the ongoing mountain lion research project, 7 mountain lions currently wear GPS collars that allow on-demand interaction with researchers and additional mountain lions will continue to be captured during the identified pilot study period. Mountain lions have and will continue to be captured primarily via pursuit by dogs as well as in live traps (Logan 2005, capture protocols previously approved by CDOW ACUC). As part of this pilot study, adult female elk (6-10) will be captured via helicopter net-gunning (Appendix A) during late-December/January 2006-07 with 6 adult females fitted with drop-off GPS/VHF collars and up to 6 adult females fitted with VHF permanent collars.. Elk will be captured on the eastern portion of the study area, directly overlapping areas including radio collared mule deer and mountain lion. Sample sizes for elk reflect an estimate of what we believe will be an adequate number of elk to provide an initial estimation of elk spatial use in the study area. As preliminary data are collected, future sample sizes will be modified in a full study plan.

Ungulate Survival and Location Monitoring: On a daily basis, from December through May, we will monitor the radioed fawns and adult female deer and elk in order to document live/death status. This will allow us to determine accurately the date of death and estimate the proximate cause of death. For animals not heard from the ground, we will conduct weekly flights to assess live/death status. Additionally, every other week we will collect aerial locations on a random sample of the collared ungulates, allowing us to estimate temporal variation in distribution. Detailed locations of GPS collared elk will not be available until self-actuating mechanisms cause GPS collars to drop-off elk in September 2007. However, during the same bi-weekly monitoring flights, we will attempt to obtain a visual location and approximate group size count for elk groups containing marked elk. This will allow us to collect broad scale group dynamic data for comparison to fine scale spatial data.

Identification of Mountain Lion GPS location clusters: Clusters of GPS locations thought to represent lion-killed ungulate sites can be determined subjectively by inspection, or objectively using a standard algorithm to group points together (Anderson and Lindzey 2003). Either approach may be effective at finding puma kill sites, but an objective approach provides a sound sampling frame from which statistical inference can be made about clusters that are not physically investigated. We have chosen to develop an objective clustering routine that will group GPS locations together that are spatially and temporally within a sampling window.

The clustering routine is designed to identify clusters in five unique selection sets in order to identify clusters containing two or more points, those that contain missing locations, and those that are represented by single points. The clustering algorithm is written in Visual Basic and is designed to run within ARCGIS (Alldredge and Schuette, CDOW unpubl. data 2006). The widths of the spatial and temporal sampling windows are user specified, in order to meet multiple applications and research needs. This will enable adjustment of the sampling frames to improve cluster specifications as needed.

The initial step is to prepare data files for ARCGIS. The main step priority is to number all downloaded GPS lat-long location records consecutively to provide a time stamp that can be used in the program. Failed locations must be numbered within the data files to maintain the proper time step (i.e. two locations that are separated by a missing location must be time stamped in such a way that the clustering algorithm can recognize that a missing location existed between the records). At this point data files can be imported to ARCGIS and coordinates converted to UTMs if necessary.

The initial selection set of clusters (S_1) is based on clusters consisting of two or more points within a specified distance and time interval. Working with temporal and spatial variables simultaneously is difficult, so we chose to create an association matrix of the combined variables. The units for time are based on GPS locations so that the time between consecutive downloads is one. Lion locations are attempted 4 times a day, so that one day consists of 4 time-steps. The association matrix is then constructed as

$$A_{ij} = \frac{1}{e^{d_{max}/d_{ij}}}\left(1 - \left|\frac{t_i - t_j}{t_{max}}\right|\right)$$

where A_{ij} is the association in time and space between points i and j, d_{max} is the maximum distance between two points to be considered a cluster, d_{ij} is the distance between points i and j, t_{max} is the maximum number of time steps between points to be considered in a cluster, and t_i and t_j are the times for locations i and j. This formula weights the distance between two locations heavier than the time between two locations. It also causes the association A_{ij} to be negative for any locations that are outside the temporal window (separated by more time-steps than t_{max}). The association between two locations within the specified time interval will be greatest for those locations that are spatially closer together. So, the largest value in the association matrix will correspond to the 2 points that are spatially the closest and within the time interval. Initially, d_{max} is set at 200 m and t_{max} is set at 16 time steps [4 DAYS] .

The initial cluster is selected by choosing the 2 points with the largest association value from the association matrix. The distance is checked to verify that the points are within the specified maximum distance, d_{max}, and if it is, the centroid of the two points is calculated. An association vector $\overrightarrow{A_c}$ is made by calculating the association among the centroid and all other points using the above formula. If all values in $\overrightarrow{A_c}$ are negative, then no points are within the specified time interval, so no additional points can be added to the cluster. Then the greatest association value A_{cmax} is selected from $\overrightarrow{A_c}$ and the distance from the centroid to the point corresponding to A_{cmax} is compared to d_{max}. If the distance is less than d_{max} then the point is added to the cluster and a new centroid is calculated using all cluster points and a new vector $\overrightarrow{A_c}$ is constructed using the new centroid. This procedure is repeated until no additional points are added to the cluster because either no points are within the specified time interval or the distance from the centroid to all points is greater than d_{max}.

After each cluster is constructed these points are omitted from the association matrix and a new cluster is started by again selecting the greatest value from the matrix and verifying that the distance between points are less than d_{max}. Points are again added to this cluster as previously described. This entire procedure is repeated until no 2 locations meet the temporal or spatial criteria. All clusters are given a unique identifier, which is based on the animal identification and the Julian date. This completes the selection set for clusters with two or more locations, which likely have a high probability of being a kill site.

Additional selection sets can be constructed from the remaining points as single location clusters. However, not all locations are equal, so the remaining selection sets are created based on whether points are associated with missing locations and based on distance between consecutive locations. The second selection set (S_2) of clusters is created from any 2 points that are within a distance d_{miss}, and are separated by 1 or more missing locations. The cluster is considered to be the area within the distance d_{max} of each of the known locations (2 areas make up the cluster, and d_{miss} is initially set at 500 m).

The final 2 cluster selection sets consist of consecutive points that are within the ranges d_{max} to d_2 (S_3) and d_2 to d_3 (S_4). To construct these selection sets, the distance between consecutive points is examined and if the distance is within the range d_{max} to d_2 (500 m) then the initial point is added as a cluster to the set S_3, or if the distance is within the range d_2 to d_3 (1000 m) then the initial point is added as a cluster to the set S_4. These single-point clusters are assumed to have radius d_{max}.

Points not used in selection sets S_1 through S_4 can then be used in a final selection set S_5. These points represent larger movements between consecutive locations and thus are thought to have low probabilities of being associated with a kill site, although these points could be associated with use of small prey items, or kill sites where a lion was physically disturbed away from a kill site. These single-point clusters are also assumed to have radius d_{max}.

Sampling of Mountain Lion GPS location clusters: A primary objective of the pilot study is to determine the probability that a given cluster represents a lion feeding site. Specifically, we will evaluate lion feeding sites as a function of the cluster association matrix. Using the clustering algorithm described above, we will attempt to classify each sampled cluster as a lion feeding site (1) or not a feeding site (0). We expect a high proportion of S_1 clusters to represent lion feeding sites. Conversely, we expect a moderate proportion of S2 and S3 clusters, and a low proportion of S4 and S5 clusters, to represent lion feeding sites. A secondary objective of the pilot study is to gather preliminary biological data regarding lion prey utilization, primarily with respect to deer and elk. The secondary objective is most efficiently accomplished by sampling S_1 clusters with greater intensity than other clusters. We therefore structured our sampling approach to allow adequate estimation of the proportion of clusters that are lion feeding sites for each cluster set, while more intensively sampling S_1 clusters than all others.

With no previous evidence to indicate similarities among individuals based on sex, age, or parental status, sampling will be stratified by each individual puma. GPS collars will be downloaded once a month for each lion and data will be analyzed through the clustering algorithm. Clusters within 2 weeks of the download date will be selected for the sampling frame, which will make the maximum time between the predation event and sampling about 1 month by the time field technicians can get to and assess evidence at each cluster site. Clusters will be randomly chosen from each selection set for each individual every month in the following manner: S_1 = 2 clusters, S_2 = 1 cluster, S_3 = 1 cluster, and S_4 and S_5 = 1 cluster on alternating months. Five clusters will be sampled each month for each lion, for a total of 30 clusters per lion from 1 November 2006, to 30 April 2007. As time allows, additional clusters can be sampled from the selection sets, which will be used as a validation data set.

Our approach forces constant sampling of each cluster set over time regardless of the frequency of clusters within a given set. This will prevent a scenario where nearly all sampled clusters in a given month are from sets, S_3, S_4 and/or S_5 (i.e., low probability of finding feeding sites). Our assessment of prey utilization depends on relatively constant detection of lion feeding sites over time to avoid bias. However, for each cluster set, the true proportion of clusters representing feeding sites may possibly change over time corresponding to changes in lion use of feeding sites. If the GPS download data indicate major changes in set-specific cluster frequencies over the sampling period, we may choose to use a proportional-allocation sampling approach in future years.

Assuming a binomial distribution and 0.90 of S_1 clusters represent lion feeding sites, we will be able to estimate the true proportion with a 95% confidence interval of +/– 0.07. Assuming 0.5 of S_2 clusters represent lion feeding sites, we will be able to estimate the true proportion with a 95% confidence interval of +/– 0.17. Assuming 0.3 of S_3 clusters represent feeding sites, we will be able to estimate the true proportion with a 95% confidence interval of +/– 0.15. Finally, assuming 0.1 of S_4 and S_5 clusters represent feeding sites, we will be able to estimate the true proportion with a 95% confidence interval of +/– 0.10. These precision levels are acceptable for the pilot study, which will facilitate development of an optimal sampling scheme in future years for evaluating lion prey utilization from GPS cluster-location data. Finally, regarding our secondary objective of collecting preliminary prey use data, we should be able to estimate the overall proportion of feeding sites represented by deer (or the proportion of feeding sites represented by elk) with a 95% confidence interval of +/– 0.05 (Anderson and Lindzey 2003, Logan 2005).

We anticipate using the following strategy to initially detect clusters in the field. For S_1 clusters, we will go to each lion GPS location in the cluster and visually inspect the immediate area for prey remains. We anticipate discovering prey remains at one or more of the cluster points but if remains are not found, we will go to the central GPS location of the cluster and spiral out overlapping view fields to a distance of 50 m beyond the outermost GPS location associated with the cluster. This should produce a thorough search and provide adequate information to judge whether or not a kill site was likely located with the cluster. For S_2 through S_5 clusters, we will go to each lion GPS location and spiral out 150 m around each point, and depending on vegetation type and density, spend a minimum of 1 hour and a maximum of 2 hours per location point to judge whether the cluster is a kill site. Data will be recorded on a standard form (Appendix B). We will also initiate attempts to quantify omission errors (missing kill remains at a cluster site) as time allows.

Estimating deer, elk, and lion distributions: We will examine locations, movements, and kernel home ranges of mule deer, elk, and lions for spatial overlap and time synchrony using ArcGIS. Our initial analyses will be descriptive and should provide insight into patterns of lion movements and feeding sites in relation to major ungulate species. Based on past observations, we do not expect deer distributions to fluctuate greatly during the winter. However, we do expect elk distributions to fluctuate depending on weather and time. As distribution varies, we also expect to observe variability in group size depending on elevation and vegetation type. We would anticipate being able to generate correlations between species of prey killed by lions and the relative presence of prey within lion home ranges.

Hypothesis Testing: This preliminary sampling effort of lion clusters and ungulate distributions will not test any hypotheses but will provide estimates with measures of precision for lion kill rates and proportions of deer and elk killed. However, provided the cluster sampling protocols prove to be functional, future work may be able to examine the likelihood that:

Lion prey mass is positively related to lion mass (male lions kill larger prey than female lions),
Lions prey on deer and elk in proportion to availability (no selection by lions)
Lions prey on sex or ages of deer or elk populations in proportion to availability (no selection by lions.
Lions alter their use of prey among seasons of the year (lions prey-switch between deer and elk)
Maternal lion home ranges include the highest available densities of ungulate prey and, many other investigative hypotheses

LOCATION OF WORK

This pilot study will be conducted on the southern half of the Uncompahgre Plateau in southwestern Colorado, near Montrose, Colorado. The study area is defined by the existing boundary for the ongoing mountain lion research project with prey populations being monitored only in the eastern half of the lion study area (Figure 1).

SCHEDULE OF WORK

Time	Activity
Fall 2006, ongoing	Capture of adult mountain lions
November 2006	Initiate sampling of GPS location clusters for lions
November-December 2006	Capture and deploy 25 VHF collars on mule deer fawns
	Capture and deploy 6 GPS/VHF collars on adult female elk
	Capture and deploy up to 6 VHF collars on adult female elk
December 2006-May 2007	Monitor mule deer and elk distribution and survival
	Monitor mountain lion distribution and survival
	Continue sampling of GPS lion clusters
June-August 2007	Analyze cluster data and deer, elk, and lion spatial distributions and provide preliminary report on the effectiveness of cluster sampling techniques and feasibility of full study

ESTIMATED COSTS

Salaries of permanent employees, as well as many other logistical costs (vehicles and flights) will be covered by existing project funds in the CDOW mule deer research, carnivore research and terrestrial management programs. Additional expenditures specific to this pilot study will include:

ITEM	COST
Wildlife Technician I for 6-months @ $12.49/hour	$13,975
GPS/VHF drop-off Collars for 6 elk @ $3,055 each	18,330
Helicopter Capture costs 12 elk @$600 each	7,200
Vehicles mileage (15K), lease for State Temporary Vehicle (6 months/$25/mo)	4,775
TOTAL COST	$44,280

RELATED FEDERAL PROJECTS

This pilot study is collaborative with 3 studies (2 ongoing, 1 completed) through the mammals research program of the Colorado Division of Wildlife. As mentioned above, the mule deer to be utilized in this pilot study were captured and radio collared as part of a previous mule deer research study (Bishop et al. 2004) as well through an ongoing mule deer study (Bergman et al. 2005). Each of these studies has received Federal-Aid research funds. Additionally, this pilot study is collaborative with an ongoing mountain lion research study through the Colorado Division of Wildlife that does not currently receive Federal-Aid research funds.

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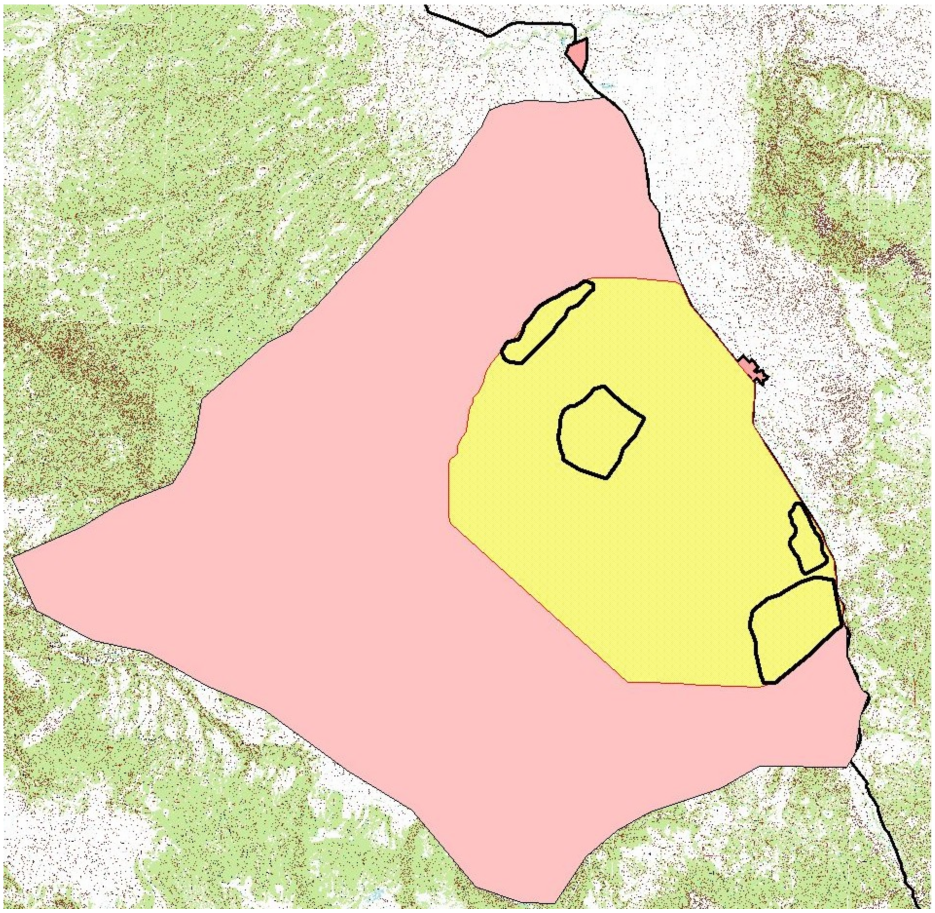
Figure 1. Ongoing mountain lion study area (solid gray), extensive mule deer study area (stippled gray), and intensive mule deer study areas (heavy black line polygons) located on the southeastern portion of the Uncompahgre Plateau immediately west of Montrose, Colorado (right center small shaded polygon).



APPENDIX A

HELICOPTER NET-GUNNING CAPTURE PROTOCOL FOR ELK

Helicopter net-gunning (Barrett et al. 1982, van Reenen 1982) is an established procedure for effectively and safely capturing elk and other cervids with minimal capture-related mortality (Bartmann et al. 1992, White and Bartmann 1994, Freddy 2001, Bishop 1998). Net-gunning will be done by Quicksilver Air, Inc., or other qualified vendor, through the current Division of Wildlife capture contract.

Capture and Transport Methods: Wild elk will be pursued and netted by the helicopter net-gunning crew. The crew will consist of one pilot and one net-gunner and up to two muggers. Netted animals will immediately be blind-folded and hobbled. Netted animals will be immediately processed and released by the capture crew at the site of capture site. The capture crew will be responsible for deploying radio collars on captured animals. Elk will be captured with net-guns in mid to late-December and in late-February or early-March in the southeast portion of GMU 62 and in the western portion of GMU 65. December snow depths rarely exceed 50 cm where elk will be captured, and mean daily temperatures during December have averaged –4.4 °C (24 °F) during the 1990's. Under these conditions, elk can be captured safely without undue risk of hyperthermia (defined as anything over 103°). Maximum allowable pursuit time, or time necessary to chase and net a target animal, will not exceed 8 minutes and will be shortened to less than 5 minutes depending on weather conditions and animal behavior. For example, in warmer conditions (e.g. >4°C), pursuit times will be minimized, particularly if unfavorable snow conditions are present. The areas where capture will occur have variable, but high, elk densities. Throughout the study area, encounter rates with different groups of elk will be high, minimizing the need to pursue any given animal for a lengthy period. However, a spotter plane will be used to facilitate capture. The spotter plane will identify unique elk groups from higher elevations and communicate coordinates of the groups to the helicopter via radio. This approach will allow the helicopter to efficiently navigate to the group and quickly identify an individual for capture. Additionally, if any potential barriers that could potentially jeopardize any animal during a chase exist (i.e. fences, roads, etc.), the capture crew will be informed of their location in order minimize/remove the potential for injury around these obstacles. In the case of roads, DOW personnel will monitor/control traffic to reduce the potential of having vehicles stop in the capture area.

The helicopter pilot and handling crew will be in radio contact with one another and with a ground crew at the helicopter refueling site. In the event of an accident, the Montrose Division of Wildlife office will be contacted by radio, and necessary emergency services will be sent to the site. The ground crew will have direct radio access to the Montrose County Sheriffs Office, Colorado State Police, Search and Rescue, and NLEEC (National Law Enforcement Emergency Channel).

Training and Personnel: The helicopter net-gunning crew will be instructed as to procedures for minimizing stress and injury to the animals. Specifically, they will be instructed on pursuit times, transport distances, and safe handling procedures. The handling crew, comprised of DOW personnel, will be instructed on proper care and handling procedures to minimize stress and risk of injury to the captured elk. Eric Bergman will be ultimately responsible for all animal care and handling.

PROCEDURES AND MANIPULATIONS OF ANIMALS

Capture: As stated above, netted animals will immediately be blind-folded and hobbled. Elk will immediately be radio-collared. Elk will be removed from the net, and the blind-fold and hobbles will be checked. Elk will be radio-collared and approximately aged by qualitatively evaluating height and wear of incisors and premolars (yearling, 2-5 years, 6-9 years, ≥10 years). All radio collars will be of fixed-size and individually fitted to each animal.

If a captured elk suffers a broken leg, back, neck, pelvis, or other similar wound, it will be euthanized at the capture site with a gunshot to the head following euthanasia protocols of the Colorado Division of Wildlife Animal Care and Use Committee.

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APPENDIX B

PUMA CLUSTER INVESTIGATION FORM

Puma ID ____________. Puma Class (check): ____ AM; ____ AFS; ____ AFC-_____ *mo.;* ____ AM & AF.
Cluster No. _________; Random Sample (y/n) ____; Cluster Dates ____________________________________;
No. of GPS Locations at Cluster _____; Individual Location IDs: ____.
Investigator(s) ____; Date investigated ______________________.
Cluster Location (e.g., drainage, mt. name) ____.

Findings: (y/n)
Prey cache ___: Species ___________; Sex (M/F) _______; Age _______ (known or est); Incisor collected? ___.
Puma sign ___: Tracks? ____; Bed with hair? ____; Scrape(s)? _____; Scat(s) _____, collected? ____________.
Other ___: Describe (e.g., bear sign, deer beds) ____.

Dist. from closest GPS cluster location (pt # ___________) to Hand-held GPS fix where puma sign found = ____ m.

Site Details: [*If matches individual GPS location(s), list number(s):* ______________________________]

Map Projection: NAD 27; Zone ______; UTM (E x N) _________________; Elev (m) _______; Aspect ____.
Terrain features (e.g., boulders, cliffs) ____.

% Ground Cover (≥70 cm tall over 5 m area) ____; Vegetation association ______________________________

Dominant plant species at site (in order): ____

Predation Evidence:
Carcass: Cached? ____; Number of cache sites ___. Drag marks: Present? __; Length (m) _____; Blood/hair? ___.
Rumen: Present?____; Removed from carcass? _____ Covered? _____________________________________.
Tooth marks: Present? ____; Location______________________; Distance between canines ___________mm
Subcutaneous or internal hemorrhage ____.
Hair plucked? ____. Carcass fed upon? ___; Approx. % consumed:____. Point of first feeding:________________; Parts eaten:____. Large leg bones broken? ________.
Signs of predation sequence (e.g., tracks, chase)?____ .
If found, UTM (E x N) of prey KILL site:____

Other predators / scavengers present (incl. other pumas)?____
Carnivore tracks: Present? ___ ; Species? _______________; If puma, heel pad width (mm) _________________.
Scrapes? ___ ; No. & dist. (m) from carcass ______________. Puma scat?____; on scrape or in mound? ________.

Prey Condition:
Antlers/horns present?(size) ________. External parasites?______________. Approx. date of death: __________.
Fat on internal organs?____.
Pregnant? ________; No. fetuses ________. Lactating? ____. Dependent young? ________________________.
Femur marrow: Too old (x)_____ Consistency-_________________________; Color-____________________.
Injuries or disease?(e.g., jaw necrosis):____.

Prey Cause of Death:
Certain puma ___; Probable puma ___; Non-puma ____; Unknown ____. Bait station? ____; Scavenged? ____.
If puma not cause, give probable cause & support notes (e.g., other predator) ______________________________
____.

Samples Collected:
Skull ____; Jaw ___; Incisors ____; Parasites ___; Puma feces __; Collar ____; Eartag ___; Photos ____;
Other_____________

Notes? (on back) ___

Colorado Division of Wildlife
July 2006 – June 2007

WILDLIFE RESEARCH REPORT

State of	Colorado	: Division of Wildlife
Cost Center	3430	: Mammals Research
Work Package	3001	: Deer Conservation
Task No.	5	: Multispecies Investigations Consulting Services for Mark-Recapture Analysis
Federal Aid Project:	W-185-R	:

Period Covered: July 1, 2006 - June 30, 2007

Author: G. C. White

Personnel: C. Bishop, D. J. Freddy, T. M. Shenk, P. Lukacs, R. Kahn, F. Pusateri, E. O'Dell, D. Martin, P. Schnurr, K. Navo, B. Andelt, A. Linstrom, P. Conn, B. McClintock, G. Davidson, and J. Ivan.

All information in this report is preliminary and subject to further evaluation. Information MAY NOT BE PUBLISHED OR QUOTED without permission of the author. Manipulation of these data beyond that contained in this report is discouraged.

ABSTRACT

Progress towards meeting the objectives of this job include:

1. Consulting assistance to Colorado Division of Wildlife (CDOW) on harvest surveys, terrestrial inventory systems, and population modeling procedures was provided. Computer code written in SAS to compute these estimates and display results graphically was provided. Specific input involved estimation of variances for design of the E-2 elk aerial survey.

2. Support for the CDOW DEAMAN software package for the storage, summary, and analysis of big game population and harvest data was provided. I met with the CDOW software group to discuss conversion of DEAMAN to a central server application.

3. Consultation with CDOW Terrestrial Biologists in the use of DEAMAN and population modeling procedures continued. Numerous questions were answered via meetings with biologists, and via email. A workshop on modeling Colorado's deer, elk, and antelope populations was conducted for biologist in Glennwood Springs, August, 2006.

4. A paper comparing the population levels of swift foxes in eastern Colorado to a previous study in cooperation with CDOW was accepted for publication in Southwestern Naturalist: Martin, D. J., G. C. White, and F. M. Pusateri. 2007. Monitoring swift fox populations in eastern Colorado. Southwestern Naturalist. In Press.

5. A paper on estimation of abundance and demography using age-at-harvest and mark-recovery data was submitted to Environmental and Ecological Statistics: Conn, P. B., G. C. White, and J. L. Laake. 2007. Estimating abundance and demography using age-at-harvest and mark-recovery data: a Bayesian approach. Environmental and Ecological Statistics. Submitted.

6. A paper on Bayesian methods to analyze age-at-harvest data was submitted to Biometrics: Conn, P. B., J. L. Laake, D. R. Diefenbach, G. C. White, and M. A. Ternent. 2007. Bayesian analysis of wildlife age-at-harvest data. Biometrics. Submitted.

7. A paper on the use of vaginal implant transmitters in cooperation with CDOW was submitted and accepted for publication in the Journal of Wildlife Management: Bishop, C. J., D. J. Freddy, G. C. White, B. E. Watkins, T. R. Stephenson, and L. L. Wolfe. 2007. Using vaginal implant transmitters to aid in capture of neonates from marked mule deer. Journal of Wildlife Management 71:945–954.

8. A paper resulting from collaboration with Montana colleagues resulted in a publication in the Journal of Wildlife Management: Pac, D. F., and G. C. White. 2007. Survival and cause-specific mortality of male mule deer under different hunting regulations in the Bridger Mountains, Montana. Journal of Wildlife Management 71:816–827.

9. A paper resulting from Sherri Huwer's M.S. project in cooperation with CDOW was accepted for publication in the Journal of Wildlife Management: Huwer, S. L., D. R. Anderson, T. E. Remington, and G. C. White. 2007. Evaluating the importance of forbs to sage-grouse using human-imprinted chicks. Journal of Wildlife Management. In Press.

10. A paper on mountain sheep populations in Rocky Mountain National Park was published in the Journal of Wildlife Management: McClintock, B. T., and G. C. White. 2007. Bighorn sheep abundance following a suspected pneumonia epidemic in Rocky Mountain National Park. Journal of Wildlife Management 71:183–189.

11. A paper on extending the mark-resight estimator using a beta-binomial distribution was published in the Journal of Agricultural, Biological, and Ecological Statistics: McClintock, B. T., G. C. White, and K. P. Burnham. 2006. A robust design mark-resight abundance estimator allowing heterogeneity in resighting probabilities. Journal of Agricultural, Biological, and Ecological Statistics 11:231–248.

12. A paper resulting from the May, 2005 Elk and Deer Workshop was published in the Wildlife Society Bulletin: Mason, J. R., L. H. Carpenter, M. Cox, J. C. Devos, J. Fairchild, D.J. Freddy, J. R. Heffelfinger, R. H. Kahn, S. M. McCorquodale, D. F. Pac, D. Summers, G. C. White, and B. K. Williams. 2006. A case for standardized ungulate surveys and data management in the western United States. Wildlife Society Bulletin 34:1238–1242.

13. A paper describing the use of closed captures models to estimate population size with Program MARK remains in press in Environmental and Ecological Statistics: White, G. C. 2007. Closed population estimation models and their extensions in program MARK. Environmental and Ecological Statistics. In Press.

14. A paper on the application of multistate models in Program MARK was published in the Journal of Wildlife Management: White, G. C., W. L. Kendall, and R. J. Barker. 2006. Multistate survival models and their extensions in program MARK. Journal of Wildlife Management 70:1521–1529.

15. A paper on the estimation of female grizzly bears was published in the Journal of Agricultural, Biological, and Ecological Statistics: Cherry, S., G. C. White, K. A. Keating, M. A. Haroldson, C. C. Schwartz. 2007. Evaluating estimators of the numbers of females with cubs-of-the-year in the Yellowstone grizzly bear population. Journal of Agricultural, Biological, and Ecological Statistics 12:195–215.

16. A paper on estimation of nest survival was published in Studies in Avian Biology: Heisey, D. M., T. L. Shaffer, and G. C. White. 2007. The ABCs of nest survival: theory and application from a biostatistical perspective. Studies in Avian Biology 34:13–33.

17. A paper on extending the mark-resight population estimation method was submitted to Environmental and Ecological Statistics: McClintock, B.T., G. C. White, K. P. Burnham, and M. A. Pryde. 2007. A robust design mixed effects mark-resight model for estimating abundance when sampling is without replacement. Environmental and Ecological Statistics. Submitted.

18. A paper on estimation of random effects with Bayesian methods was submitted to Environmental and Ecological Statistics: White, G. C., K. P. Burnham, and R. J. Barker. 2007. Evaluation of some Bayesian MCMC random effects inference methodology applicable to bird ringing data. Environmental and Ecological Statistics. Submitted.

19. A paper on analysis of small count data was submitted to Condor: McDonald, T. L., and G. C. White. 2007. A comparison of regression models for small counts. Condor. Submitted.

20. A paper on the impact of previous capture on sampling probabilities with DNA hair-snag grids for grizzly bear populations was submitted to the Journal of Wildlife Management: Boulanger, J., and G. C. White. 2007. Influence of past live captures on detection probabilities of grizzly bears using DNA hair snagging methods. Journal of Wildlife Management. Submitted.

21. A paper on detecting trends in the Yellowstone grizzly bear population was submitted to Ursus: Harris, R. L., G. C. White, C. C. Schwartz, and M. A. Haroldson. 2007. Population growth of Yellowstone grizzly bears: uncertainty and future monitoring. Ursus. Submitted.

22. A graduate research project (Ph. D.) in cooperation with CDOW to develop statistical models to monitor puma and black bear populations in Colorado based on checks of harvested animals and DNA and/or radio-tracking data was completed. The graduate student is Paul B. Conn. The dissertation is: Conn, P. B. 2007. Bayesian Analysis of Age-at-Harvest Data with Focus on Wildlife Monitoring Programs. Ph. D. Dissertation, Colorado State University, Fort Collins. 184 pp.

23. A research study to examine the impact of nutrition on the decline of mule deer fecundity during the last 20 years was continued in cooperation with Chad Bishop and CDOW. Portions of this work will serve as his doctoral dissertation in addition to his full-time duties as a researcher with CDOW.

24. A graduate research project (M. S.) was continued in cooperation with CDOW to evaluate line transect methodology for estimating pronghorn populations in eastern Colorado. The graduate student is Aaron Linstrom, and the project is in addition to his full-time duties as a biologist with CDOW.

25. A graduate research project (M. S.) in cooperation with CDOW to evaluate methods of redistributing elk in and around Great Sand Dunes National Park was started and then discontinued. The student, Greg Davidson, switched his work to evaluate habitat use by elk on the Grand Mesa. A report on the San Luis Valley elk work is nearly completed.

26. A graduate research project (Ph.D.) in cooperation with CDOW to evaluate snowshoe hare densities relative to lodge pole pine and mixed conifer habitats was continued. The graduate student is Jake Ivan.

27. Development of the design of a monitoring system for white-tailed prairie dogs in western Colorado and eastern Utah was continued in cooperation with CDOW with P. Schnurr, K. Navo and B. Andelt.

A final draft of a manuscript on the use of occupancy monitoring for white-tailed and Gunnison prairie dogs was given to B. Andelt for submission to the Journal of Wildlife Management on 20 February, 2007.

28. Preliminary analysis of monitoring data on black-tailed prairie dogs in eastern Colorado in cooperation with CDOW was continued. This effort is in cooperation with Francie Pusateri and Eric O'Dell of CDOW.

WILDLIFE RESEARCH REPORT

CONSULTING SERVICES FOR MARK-RECAPTURE ANALYSES

GARY C. WHITE

P. N. OBJECTIVE

Provide expert biostatistical and experimental design services to the Colorado Division of Wildlife, Wildlife Programs Branch.

SEGMENT OBJECTIVES

1. Provide biostatistical support to implement and analyze CDOW hunter harvest surveys.

2. Provide professional oversight, critiques, and analytical support to CDOW terrestrial management and avian and mammals research sections.

3. Convey to CDOW research and management sections new and pertinent information obtained in various collaborative projects conducted with other agencies and entities.

RESULTS, DISCUSSION, SUMMARY

See ABSTRACT for summary of key activities and publications.

Prepared by: ____________________________________
Dr. Gary C. White, Department of Fish,
Wildlife, and Conservation Biology
Colorado State University

Colorado Division of Wildlife
July 2006 – June 2007

WILDLIFE RESEARCH REPORT

State of	Colorado	: Division of Wildlife
Cost Center	3430	: Mammals Research
Work Package	3001	: Deer Conservation
Task No.	6	: Population Performance of Piceance Basin Mule
		: Deer in Response to Natural Gas Resource
		: Extraction and Mitigation Efforts to Address
		: Human Activity and Habitat Degradation
Federal Aid Project:	W-185-R	:

Period Covered: July 1, 2006 – June 30, 2007

Authors: C. R. Anderson, and D. J. Freddy

Personnel: M. Alldredge, E. Bergman, C. Bishop, R. Kahn, P. Lukacs, T. Remington, M. Schuette; G. White, Colorado State University; H. Sawyer, Western Ecosystems Technology, Inc.

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ABSTRACT

A researcher FTE vacancy was filled with a newly hired person in December 2006 who became the project leader for this project. No preliminary field work could be completed in winter-spring 2007 as originally planned for FY2006-07 but assessments of potential study areas, resource inventory maps, and tentative study plan outlines were completed by June 2007. As such, field work for this project will begin winter 2007 and be centered in the Piceance Basin area of northwestern Colorado which is currently undergoing intensive natural gas development in one of the most extensive and important mule deer winter and transition range areas within the state. Our approach will be to experimentally evaluate habitat treatments that may rehabilitate the landscape to benefit mule deer and to evaluate human-activity management alternatives to reduce the disturbance impacts on mule deer. This project will require a long-term commitment of at least 10-years from private industry, the BLM, and the CDOW to assess if sustainable mule deer populations can persist within a highly disturbed landscape following implementation of beneficial habitat treatments and development practices.

WILDLIFE RESEARCH REPORT

POPULATION PERFORMANCE OF PICEANCE BASIN MULE DEER IN RESPONSE TO NATURAL GAS RESOURCE EXTRACTION AND MITIGATION EFFORTS TO ADDRESS HUMAN ACTIVITY AND HABITAT DEGREDATION

CHARLES R. ANDERSON AND DAVID J. FREDDY

P. N. OBJECTIVE

To develop approaches to provide for energy extraction in a manner that maintains viable mule deer populations for future recreational and ecological purposes.

SEGMENT OBJECTIVES

1. Consult with regional personnel to select potential study sites for addressing habitat mitigation and energy development practices that benefit mule deer.
2. Plot historic and current energy development activities to assess potential treatment and control sites for experimental evaluation.
3. Develop draft study proposals for peer review and funding solicitation.

INTRODUCTION

Extraction of natural gas from areas throughout western Colorado has raised concerns among many public stakeholders and the Colorado Division of Wildlife that the cumulative impacts associated with this intense industrialization will dramatically and negatively affect the wildlife resources of the region. Concern is especially high for mule deer due to their recreational and economic importance as a principal game species and their ecological importance as one of the primary herbivores of the Colorado Plateau Ecoregion. Research evaluating the most effective strategies for minimizing and mitigating these activities will greatly enhance future management efforts to sustain mule deer populations for future recreational and ecological values. Our primary goal of this study is to develop approaches to provide for energy extraction in a manner that maintains viable mule deer populations for future recreational and ecological purposes. This may be accomplished by restoring or enhancing habitat conditions on or adjacent to disturbed sites and by modifying development practices.

Due to the extensive energy development that is projected to occur over the next 20 years throughout much of the mule deer winter range in the northern Rocky Mountains of the western US, innovative approaches to energy development and mitigation methods are essential to sustain viable mule deer populations in the region. Impacts from development and conversely success of mitigation efforts are often assumed but rarely demonstrated, and these assumptions can only be confirmed by application of well designed research efforts conducted over sufficiently long time periods to measure responses. This project proposes to identify habitat mitigation and energy development approaches that sustain mule deer survival and recruitment during and after habitat disturbance from development activities. This effort will require coordination and cooperation between Colorado Division of Wildlife and the major energy companies. We anticipate this partnership will benefit mule deer populations and foster the evolution of wildlife management and energy development practices that are compatible with other wildlife and human values associated with maintaining functional ecosystems over the long term.

STUDY AREA

The proposed study sites represent 6 segments of mule deer winter range in the Piceance Basin, southwest of Meeker, Colorado (Figure 1), and the primary energy companies developing these areas include Encana and Exxon-Mobile (Figure 2). Because of the varying levels of development and deer densities relative to differing winter population segments in the Piceance Basin, different experimental units (i.e., mule deer winter ranges) are uniquely suited for addressing different questions. Experimental designs monitoring mule deer responses to treatment (e.g., habitat mitigation) and control areas are necessary to differentiate cause-effect relationships from development versus environmental factors. Suitable control areas require that little or no previous development has occurred and that no development occurs during the experimental time frame. Ideally, both temporal and spatial control areas would be monitored to make valid comparisons to developed and subsequently mitigated sites; temporal controls provide measures of natural variability in mule deer population parameters over time and spatial controls provide measures of variability due to differences in geography. Once spatial and temporal variation is accounted for, inferences can be made relative to development disturbance or mitigation effects on mule deer.

The North Ridge, Story/Willow Creek, and Yellow Creek deer population segment areas (Figure 1) currently exhibit little to no development, but it is currently unknown whether or not these areas will be developed in the future; there is potential for future oil shale development in the Story/Willow Creek and Yellow Creek deer areas. North Ridge appears least likely to be developed because it is outside of the current oil shale lease area and only a few wells have historically been drilled on or adjacent to the area, whereas the same cannot be said of the Story/Sprague or Yellow Creek areas. Thus, North Ridge would appear best suited as a temporal control site for comparison to other developed winter ranges within the Piceance Basin and may also serve as a geographic control for the Crooked Wash deer population segment located immediately north and adjacent to the Piceance Basin. The Story/Willow Creek and Yellow Creek deer may provide spatial controls for the Magnolia and Ryan Gulch deer population segments, respectively, but future development potential in these areas is unknown. If these areas become developed in the future (either for oil shale or natural gas), they would provide BACI (Before-After-Control-Impact) type comparisons strengthening our inference of development impacts on mule deer population performance.

Magnolia, Crooked Wash, and Ryan Gulch deer areas have historically received relatively high development activity and currently exhibit moderate-high development, and appear likely to be developed extensively in the future based on the gas development layers currently available (Colorado Oil and Gas Conservation Commission). Pretreatment data in these areas will be represented by parameters associated with developed sites and the measured response will be in the form of habitat treatments and/or differing development practices, which will be measured in comparison to the control sites.

We propose including 3 control sites (1 temporal/spatial control and 2 spatial controls) and 3 treatment sites to investigate mule deer response to habitat and/or development treatments (e.g., directional versus non-directional drilling, piping versus trucking condensate, etc.) across a range of deer densities (Table 1). We would strive to split high intensity extraction study sites into 2 halves with one half serving as the 'control' [standard development] and one half serving as the 'treatment' [improved development approach or improved habitat]. The above scenario addresses the potential for establishing control and treatment sites for evaluating mule deer population response to habitat treatments and/or development treatments, and may allow larger scale mule deer responses from energy development to be addressed by comparing control site parameters to developed site parameters; smaller scale inference would require collection of pretreatment data at developed sites (e.g., similar to mitigation treatments in the proposed design) and may not be possible unless the Yellow Creek or Story/Willow Creek areas are developed in the future. Modified versions of the proposed design could be implemented depending on the level of funding available and the degree to which industry willing to collaborate with this effort. We consider 3 study sites, likely North Ridge, Magnolia, and Crooked Wash, as the minimum number of study sites necessary to adequately address the objectives of this project; the additional proposed study areas will allow increased flexibility in the questions that are addressed and increase our inference relative to mule deer responses to habitat treatments and modifications of development practices. Furthermore, if we are not able to evaluate potential mitigating industrial operation and/or habitat improvements, this study would likely only have the potential to document negative impacts of intense energy extraction practices on mule deer.

RESPONSE VARIABLES

To allow for competing hypotheses in regards to potential development and mitigation effects, 4 primary response variables will be measured including (1) overwinter fawn survival, (2) deer density, (3) habitat use patterns, and (4) adult female body condition.

(1) To determine if mitigation and/or development treatments elicit a chronic survival response with a long-term population level effect, we will measure over-winter fawn survival in all experimental units. Based on past research (White and Bartmann 1998), treatment effects of 15% change in survival appear biologically significant.

(2) To determine if habitat treatments or development practices elicit a brief survival response with a long-term population level effect, we will estimate deer density to determine if there is a difference in carrying capacity between treatment and control experimental units. Because mule deer may respond to development or mitigation at variable rates, we may not be able to detect differences in fawn survival, but estimating deer density will still allow us to determine if development or mitigation efforts have a population level effect.

(3) To determine if habitat treatments or development practices elicit a shift in habitat use patterns, we will examine changes in Resource Selection Probability Functions (RSPF; Sawyer et al. 2006) pre- and post-habitat treatments, between areas exhibiting development practices, and compare RSPFs between developed and non-developed sites. We will infer population level impacts if fawn survival and/or deer densities differ relative to changes or differences in habitat use patterns.

(4) To determine if adult female mule deer respond positively to habitat treatments and/or changes in development practices, percent body fat and loin depth will be measured annually during late winter (Bishop et al. 2005, Bergman et al. 2005). We would expect a relatively rapid response in body condition following habitat or development treatments, indicating that treatments are having the intended positive effect.

PROJECT EXPENSES

Estimating fawn survival, deer density, deer behavioral responses, female body condition, and implementing small scale habitat improvements are costly endeavors involving the purchase of numerous standard VHF radio-collars, specialized GPS radio-collars, helicopter flight hours for deer capture/collaring and aerial surveys, machinery to physically alter the habitat, and personnel to adequately perform day-to-day data collection. If large scale habitat treatments are needed or desired, funding in addition to the estimates below will be required as habitat treatments cost \$300 to \$1,000/acre depending on the most appropriate treatment for a locale. Minimum cost estimates to design, implement, and evaluate responses of mule deer to habitat mitigation options range form \$580,500 to \$1,161,00 (most preferred design) per year depending on project design (Table 2).

LITERATURE CITED

BISHOP, C. J., G. C. WHITE, D. J. FREDDY, and B. E. WATKINS. 2005. Effect of nutrition on mule deer recruitment and survival rates. Wildlife Research Report, Colorado Division of Wildlife, Fort Collins. USA.

BERGMAN, E. J., C. J. BISHOP, D. J. FREDDY, and G. C. WHITE. 2005. Evaluation of winter range habitat treatments on over-winter survival and body condition of mule deer. Study Plan, Colorado Division of Wildlife, Fort Collins, USA.

SAWYER, H., R. M. NIELSON, F. LINDZEY, and L. L. MCDONALD. 2006. Winter habitat selection of mule deer before and during development of a natural gas field. Journal of Wildlife Management 70:396-403.

WHITE, G. C., and R. M. BARTMANN. 1998. Effect of density reduction on overwinter survival of free-ranging mule deer fawns. Journal of Wildlife Management 62:214-225.

Prepared by ___________________________________
Charles R. Anderson, Wildlife Researcher

Table 1. Relative density of natural gas wells and mule deer and experimental designation for potential study sites in the Piceance Basin, Colorado, for addressing mule deer response to natural gas development practices and habitat mitigation.

	Relative density			Experimental
Study area	Inactive wells	Active wells	Mule deer	designation
North Ridge	Very low	None	High	Temporal/spatial control
Crooked Wash	High	High	High	Treatment
Story/Willow Creek	Low	Low	Moderate	Spatial control
Magnolia	High	High	Moderate	Treatment
Yellow Creek	Moderate	Low	Low	Spatial control
Ryan Gulch	High	Moderate	Low	Treatment

Table 2. Estimated costs for CDOW to conduct desired mule deer research in the Piceance Basin to assess impacts of natural gas extraction on mule deer and evaluate approaches to mitigate habitat impacts. Project should be conducted for 10 years to allow for adequate time to measure biological responses, 2008-2018.

Cost Estimates Per Year Per Study Site (2008 dollars) Piceance Basin Mule Deer Research		Minimum Study One Control Site & Two Treatment Sites Cost Per Year (2008 dollars)	Acceptable Two Control Sites & Two Treatment Sites Cost Per Year (2008 dollars)	Best Three Control Sites & Three Treatment Sites Cost Per Year (2008 dollars)
Telemetry collars & equipment	$70,000	$580,500	$774,00	$1,161,000
Helicopter Deer Capture & Surveys	$68,500			
Other Field Operations & Equipment	$15,000			
12 months TFTE Personnel (Tech I)	$30,000			
Vehicle Yearly Lease Plus Mileage (4x4 PU, & 45,000miles)	$20,000			
Total cost Per Study Site/Yr	$193,500			

Figure 1. Proposed mule deer study sites relative to natural gas development in the Piceance Basin, Colorado, July 2007.

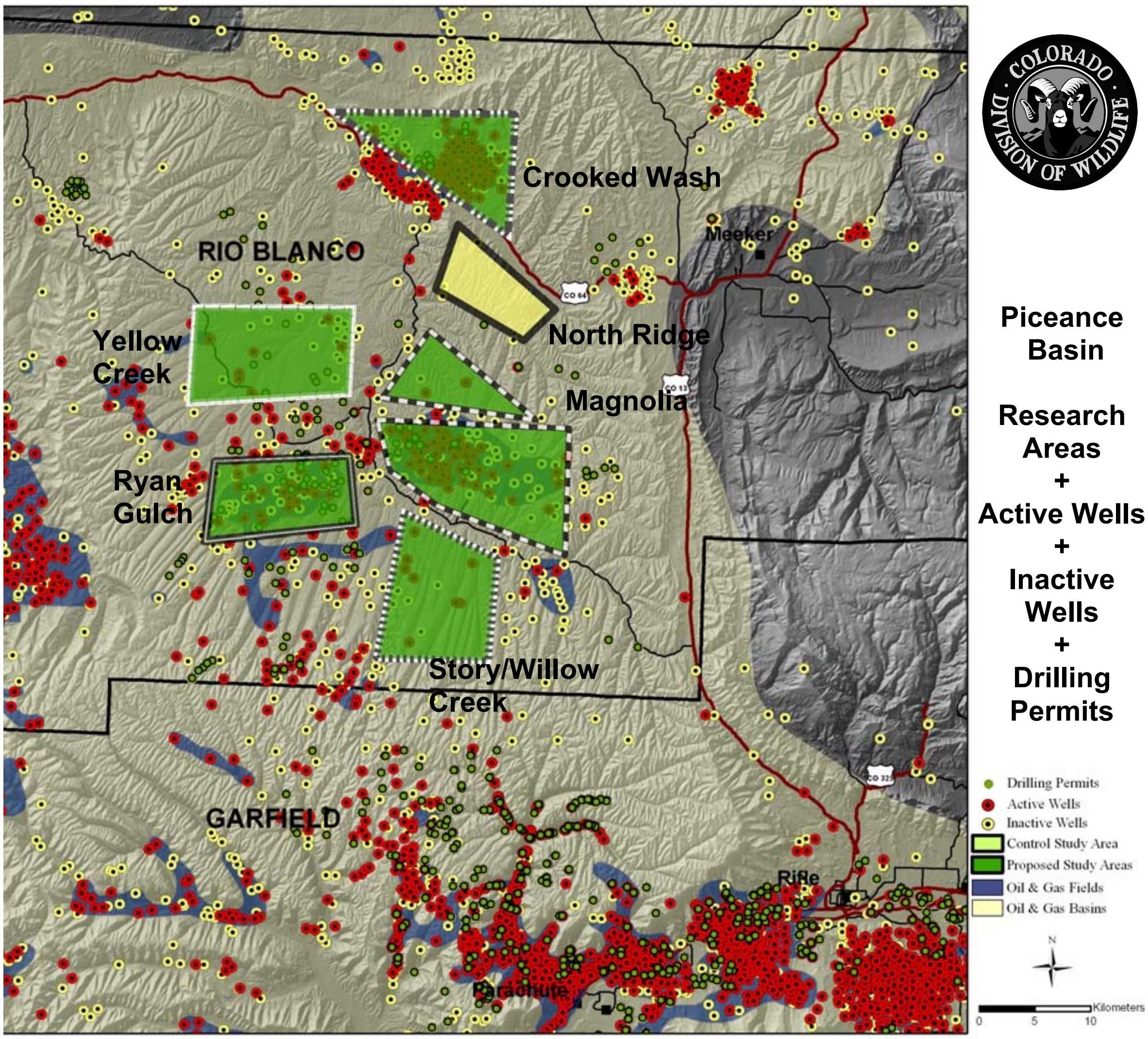
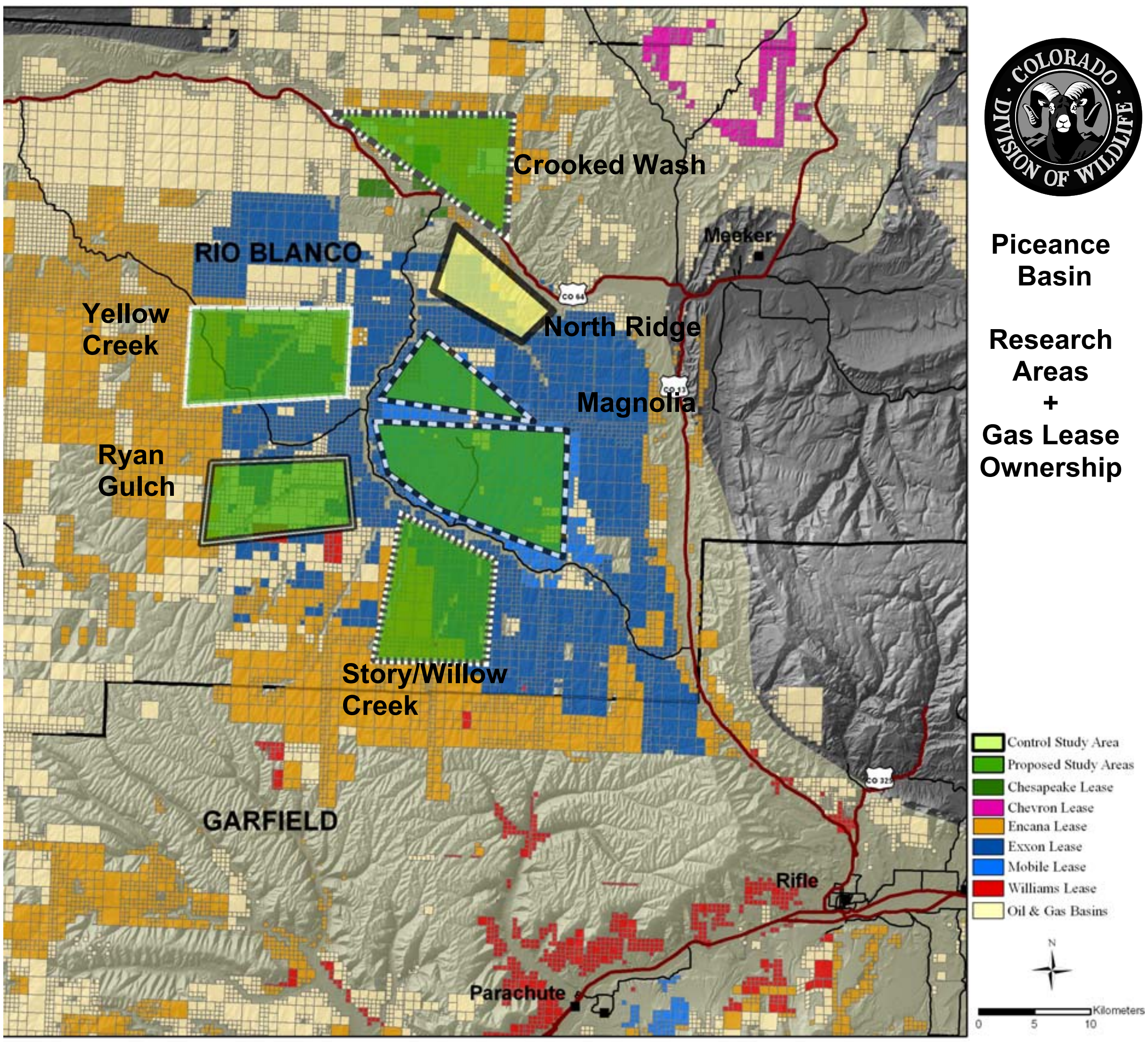


Figure 2. Proposed mule deer study sites relative to the primary energy companies controlling natural gas leases in the Piceance Basin, Colorado, July 2007.



Colorado Division of Wildlife
July 2006 – June 2007

WILDLIFE RESEARCH REPORT

State of	Colorado	: Division of Wildlife
Cost Center	3430	: Mammals Research
Work Package	3003	: Predatory Mammals Conservation
Task No.	1	: Puma Population Structure and Vital Rates on the Uncompahgre Plateau
Federal Aid Project:	N/A	:

Period covered: August 1, 2006—July 30, 2007

Author: K. A. Logan.

Personnel: K. Logan, B. Bavin, B. Dunne, J. Mannas, S. Waters, K. Crane, T. Mathieson, M. Caddy, and T. Bonacquista of CDOW; S. Young, and J. McNamara of U.S.D.A. Wildlife Services; volunteers and cooperators including: private landowners, U.S. Forest Service, Bureau of Land Management, and Colorado State Parks with financial support received from The Howard G. Buffett Foundation and Safari Club International Foundation.

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ABSTRACT

Research continued on puma population characteristics and dynamics on the Uncompahgre Plateau. Puma capture efforts resulted in a total of 54 puma captures (9-10 adult females [1 female captured twice], 7 adult males [1 male probably captured 3 times, and another captured twice], 1 subadult female, 0-1 other subadults, and 30 cubs [4 captured twice each]). Efforts to capture, sample, and mark pumas with the use of trained dogs extended from November 13, 2006 to May 11, 2007. This resulted in 22 puma captures, including 1 adult female, 1 adult male, 2 adult males, and 2 male cubs captured and processed for the first time. One female (adult or subadult) and 2 cubs were not handled for safety reasons. Capture efforts with ungulate carcasses and cage traps resulted in 8 puma captures, including 4 adult females, 2 adult males, 1 subadult female, and 1 female cub. Of those animals, 1 adult female, 1 adult male, the subadult female, and the female cub were captured for the first time. Capture efforts during November 2006 through May 2007 enabled us to estimate a minimum count of 24 independent pumas detected on the Uncompahgre Plateau study area during that time. The count included 16 females and 8 males. We captured, sampled, and marked 26 puma cubs produced by 10 females. Twenty-three of the cubs were examined at 8 nurseries when the cubs were 29 to 41 days old. Since the start of this study, 38 cubs from 13 litters aged 29 to 42 days old had a sex ratio of 21 males:17 females. The mean (±*SD*) and extremes of litter sizes were 2.84 (±0.99), 1 to 4. Eight birth intervals for 7 different females averaged 14.99 months (*SD* = 3.40), and ranged from 11.7 to 20.5 months. Four gestation periods averaged 92.0 days (*SD* = 1.68). Of 9 adult males and 12 adult females radio-monitored to quantify survival and agent-specific mortality rates, 1 male and 1 female are known to have died from natural causes. Of 6 subadult pumas monitored via radio-telemetry, none died. Thirty-nine puma cubs (20 males, 19 females) have been monitored by radiotelemetry for varying durations. Among those, 12 deaths were documented, including 7 from intra-species strife, 1 killed by coyotes, 1 killed by a vehicle, and 3 died due to research activities.

Twenty adult pumas (7 males, 13 females) fit with GPS collars since field research began in December 2004 have yielded 113 to 2,759 locations per individual puma. Winter activity areas were estimated for 12 (9 female, 3 male) GPS-collared adult pumas. As an index to the vulnerability of puma mothers to sport-harvest we monitored mother-cub distances from an airplane during November to March. Puma mothers were ≥520 m from their cubs during 16.3% of the observations (mean distance = 1,120 m, *SD* = 1,214.40, range = 616 to 4,101). These results were similar to our results the previous winter (15.2%). A collaborative effort to investigate puma use of ungulates on the Uncompahgre Plateau resumed. GPS clusters were investigated for 13 GPS-collared adult pumas (8 female, 5 male). A total of 257 clusters were investigated. Mule deer and elk were about equally important to pumas as food. Preliminary comparisons between our current puma research on the Uncompahgre Plateau (31 months duration) and results of the Anderson et al. (1992) puma research on the plateau (7 years duration 1981-1988) were made where appropriate. Proposed work includes: continuing investigations of puma use of ungulates, developing and testing methods and models to estimate puma abundance, and collaborating with colleagues to assess puma health. In addition, we will consider how research of pumas on developed areas on the Uncompahgre Plateau can contribute to the CDOW’s efforts to study puma-human interactions on the Colorado Front Range.

WILDLIFE RESEARCH REPORT

PUMA POPULATION STRUCTURE AND VITAL RATES ON THE UNCOMPAHGRE PLATEAU, COLORADO

KENNETH A. LOGAN

P. N. OBJECTIVE

Quantify puma population sex and age structure; estimate puma population vital rates, including: reproduction rates of females, age-stage survival rates, and immigration and emigration rates; quantify agent-specific mortality rates— all to improve the Colorado Division of Wildlife's (CDOW) model-based approach to managing pumas in Colorado.

SEGMENT OBJECTIVES

1. Continue gathering data on puma population sex and age structure.
2. Continue gathering data for estimates of puma reproduction rates.
3. Continue gathering data to estimate puma sex and age-stage survival rates.
4. Continue gathering data to estimate agent-specific mortality rates.
5. Continue gathering data on puma movements for the development of sampling methods for mark-resight or recapture population estimates that might involve sampling puma DNA-genotypes, trail cameras, or direct observations.
6. Begin gathering data on spatial relationships of puma mothers to their cubs during the Colorado puma hunting season as a preliminary assessment of the vulnerability of puma mothers to sport-hunting harvest.
7. Evaluate other data sources that could come from this research that can be developed into other puma research relevant to CDOW biologists and managers.

INTRODUCTION

Colorado Division of Wildlife managers need reliable information on puma biology and ecology in Colorado to develop sound management strategies that address diverse public values and the CDOW objective of actively managing puma while "achieving healthy, self-sustaining populations"(CDOW 2002-2007 Strategic Plan:9). Although 4 puma research efforts have been made in Colorado since the early 1970s and puma harvest data is compiled annually, reliable information on certain aspects of puma biology and ecology, and management tools that may guide managers toward effective puma management is lacking.

Mammals Research staff held scoping sessions with a number of the CDOW's wildlife managers and biologists. In addition, we consulted with other agencies, organizations, and interested publics either directly or through other CDOW employees. In general, CDOW staff in western Colorado highlighted concern about puma population dynamics, especially as they relate to their abilities to manage puma populations through regulated sport-hunting. Secondarily, they expressed interest in puma―prey interactions. Staff on the Front Range placed greater emphasis on puma―human interactions. Staff in both eastern and western Colorado cited information needs regarding effects of puma harvest, puma population monitoring methods, and identifying puma habitat and landscape linkages. Management needs identified by CDOW staff and public stakeholders form the basis of Colorado's puma research program, with multiple lines of inquiry (i.e., projects):

Improve our ability to manage puma hunting with enhanced scientific bases, strategies, and tools

- Puma population characteristics (i.e., density, sex and age structure).
- Puma population dynamics and vital rates (i.e., birth rates, survival rates, emigration rates, immigration rates, population growth rates).
- Field methods and models for assessing and tracking changes in puma populations.
- Relative vulnerability of puma sex and age classes to hunter harvest.

Improve our understanding of puma habitat needs and interrelationships of puma management units

- Puma habitat use, movements, and use of landscape linkages.
- Puma recruitment patterns (i.e., progeny, immigration, emigration).
- Models for identifying puma habitat and landscape linkages.

Improve our understanding of the puma's role in the ecology of other species

- Relationships of puma to mule deer, elk, and other natural prey.
- Relationships of puma to species of special concern, e.g., desert bighorn sheep.

Improve our understanding of puma-human interactions and abilities to manage them

- Behavior of puma in relation to people and human facilities.
- Puma predation on domestic animals.
- Effects of translocating nuisance pumas.
- Effects of aversive conditioning on pumas.

While all projects cannot be addressed concurrently, understanding their relationships to one another is expected to help individual projects maximize their benefits to other projects that will assist the CDOW to achieve its strategic goal in puma management (Fig.1).

Management issues identified by managers translate into researchable objectives, requiring descriptive studies and field experiments. Our goal is to provide managers with reliable information on puma population biology and to develop useful tools for their efforts to *adaptively manage* puma in Colorado to maintain healthy, self-sustaining populations.

The highest-priority management needs are being addressed with this intensive population study that focuses on puma population dynamics using sampled, tagged, and GPS/radio-collared puma. Those objectives include:

1. Describe and quantify puma population sex and age structure.
2. Estimate puma population vital rates, including: birth rates, age-stage-specific survival rates, emigration rates, immigration rates.
3. Estimate agent-specific mortality rates.
4. Improve the CDOW's model-based management approaches with Colorado-specific data from objectives 1—3. Consider other useful models.

Concurrently with the tasks associated with the objectives above, significant progress will be made toward a 5th objective, which will initially be subject to *pilot study*— develop methods that yield reliable estimates of population abundance (i.e., numbers and density) and attendant annual population growth rates, such as, direct capture-resight, and DNA genotype capture-recapture.

TESTING ASSUMPTIONS AND HYPOTHESES

Hypotheses associated with main objectives 1—5 of this puma population research are structured to test assumptions guiding puma management in Colorado.

1. Recreational puma hunting management in Colorado Game Management Units (GMUs) is guided by a model to estimate allowable harvest quotas to achieve one of two puma population objectives: 1) maintain puma population stability, or 2) cause puma population decline (CDOW, Draft L-DAU Plans, 2004). Basic model parameters are: puma population density, sex and age structure, and annual population growth rate. Parameter estimates are currently chosen from literature on studies in western states that are deemed to provide reliable information. Background material used in the model assumes a moderate annual rate of growth of 15% (*i.e.,* $\lambda = 1.15$) for the adult and subadult puma population (J. Apker, Carnivore Management Specialist, CDOW, Monte Vista). This assumption is based upon information with variable levels of uncertainty (e.g., small sample sizes, data from habitats dissimilar to Colorado). The key assumption is that the CDOW can manage puma population growth through recreational hunting: for a stable puma population hunting removes the annual increment of population growth (i.e., as estimated from estimates of population density, structure, and λ); for a declining population, hunting removes more than the annual increment of population growth. Parameters influencing λ include population density, sex and age structure, female age-at-first-breeding, age-specific natality, sex- and age-specific survival, immigration and emigration. A descriptive study will ascertain these population parameters in an area that appears typical of puma habitat in western Colorado and will yield defensible population parameters based upon contemporary Colorado data. This study will be conducted in a 5-year *reference period* (i.e., absence of recreational hunting) to allow puma life history traits to interact with the main habitat factors that appear to influence puma population growth (e.g., prey availability and vulnerability, Pierce et al. 2000, Logan and Sweanor 2001). Contingent upon results in the *reference period*, a subsequent 5-year *treatment period* is planned. The *treatment period* will involve the use of controlled recreational hunting to manage the puma population into a decline phase.

   H_{1a}: Population parameters measured during a 5-year *reference period* (in absence of recreational puma hunting) in conifer and oak communities with deer, elk and other prey populations typical of those communities in Colorado will yield an estimated annual adult plus subadult population growth rate that will match or exceed $\lambda = 1.15$, which is currently assumed in the CDOW's model-based management.

   H_{1aA}: Population parameters measured during a 5-year *reference period* (absence of recreational puma hunting) in conifer and oak communities with deer, elk and other prey populations typical of those communities in Colorado will yield an estimated annual adult plus subadult population growth rate that will be substantially lower (i.e., ≥50% lower, $\lambda \leq 1.075$) than the assumed $\lambda = 1.15$.

   H_{1b}: Population parameters during a 5-year *treatment period* (controlled puma hunting) will differ substantially from those measured during the preceding 5-year *reference* period (hunting closure) and will yield an estimated annual adult plus subadult population growth rate that will be approximately $\lambda = 0.8$ for at least the first 2 years of the *treatment period*. Hunting-caused mortality will be strongly additive, and will require removal of the annual growth increment (of adults plus subadults) plus 20% (e.g., assume $\lambda = 1.15$, so, $0.15 \times 0.2 + 0.15 = 0.18$; $0.18 \times 100 = 18\%$ annual harvest of adults plus subadults).

   H_{1bA}: Population parameters during a 5-year *treatment period* (controlled puma hunting) will not differ substantially from those measured during the preceding 5-year *reference* period (hunting closure), and the adult plus subadult population will not decline on average as a result of hunting mortality. Hunting-caused mortality, reproduction, immigration, and emigration might be compensatory.

2. Considering limitations (i.e., methods, number of years, assumption violations) to the Colorado-specific studies on puma densities cited above (Currier et al. 1977, Anderson et al. 1992, Koloski 2002), managers assume that puma population densities in Colorado are within the range of those

quantified in more intensively studied populations in Wyoming (Logan et al. 1986), Idaho (Seidensticker et al. 1973, Alberta (Ross and Jalkotzy 1992, and New Mexico (Logan and Sweanor 2001). The CDOW assumes density ranges of 2.0—4.6 puma/100 km^2 to extrapolate to Data Analysis Units to guide the model-based quota-setting process. Likewise, managers assume that the population sex and age structure is similar to puma populations described in the intensive studies. Using capture, mark, re-capture techniques developed and refined during the study to estimate the puma population, the following will be tested:

H_{2a}: Puma densities during the 5-year *reference period* (absence of recreational puma hunting) in conifer and oak communities with deer, elk and other prey populations typical of those communities in Colorado will vary within the range of 2.0—4.6 puma/100 km^2 and will exhibit a similar sex and age structure to puma populations in Wyoming, Idaho, Alberta, and New Mexico.

3. The increase and decline phases of the puma population make it possible to test hypotheses related to shifts in the age structure of the population which have been linked to harvest intensity in Wyoming and Utah.

H_{2b}: The puma population on the Uncompahgre Plateau study area will exhibit a young age structure after hunting prohibition at the beginning of the *reference period*. During the 5 years of hunting prohibition, greater survival of independent puma will cause an older age structure in harvest-age puma (i.e., adults and subadults) as suggested by the work of Anderson and Lindzey (2005) in Wyoming and Stoner (2004) in Utah.

H_{2c}: As hunting is re-instated in the *treatment period*, the age structure of harvested puma and the harvest-age puma in the population will vary as observed by Anderson and Lindzey (2005) in Wyoming and Stoner (2004) in Utah.

Desired outcomes and management applications of this research include:

1. Quantification of variations in puma population density, sex and age structure, growth rates, vital rates, and an understanding of factors affecting them will aid adaptive puma management by yielding population parameters useful for estimating puma population abundance, evaluation of management alternatives, and effects of management prescriptions.
2. Testing assumptions about puma populations, currently used by CDOW managers, will help those managers to biologically support and adapt puma management based on Colorado-specific estimated puma population characteristics, parameters, and dynamics.
3. Methods for estimating puma abundance (capture-mark-recapture) of known reliability will allow managers to “ground truth” modeled populations and estimate effects of management prescriptions designed to achieve specified puma population objectives in targeted areas of Colorado. Ascertaining puma numbers and densities during the project will require development of reliable monitoring techniques based on capture-mark-recapture methods and models. Potential methods include direct and DNA genotype capture-recapture. Study plans to develop and test feasible field and analytical methods will be developed in the future after we have learned the logistics of performing those methods, after we have preliminary data on puma demographics and movements which will inform suitable sampling designs, and when we have adequate funding.
4. This information will be disseminated to citizen stakeholders interested in pumas in Colorado, and thus contribute to informed public participation in puma management.

STUDY AREA

The study area for the puma population research is on the Uncompahgre Plateau (in Mesa, Montrose, Ouray, and San Miguel Counties, Fig. 2). The study area includes about 2,253 km^2 (870 mi.^{2}) of the southern halves of Game Management Units (GMUs) 61 and 62, and about 155 km^2 (60 mi.^{2}) of the northern edge of GMU 70 (between state highway 145 and San Miguel River). The area is bounded by state highway 348 at Delta, 25 Mesa road and Forest Service road FS503 to Nucla, state highway 97 to state highway 141 to state highway 145 to Placerville, state highway 62 to Ridgeway, U.S. highway 550 to Montrose, and U.S. highway 50 to Delta.

The study area seems typical of puma habitat in Colorado that has vegetation cover that varies from the pinon-juniper covered foothills starting from about 1,700 m elevation to the spruce-fir and aspen forests growing to the highest elevations of about 3,000 m. Mule deer (*Odocoileus hemionus*) and elk (*Cervus elaphus*) are the most abundant wild ungulates available for puma prey. There are cattle and domestic sheep raised on summer ranges on the study area. Year-round human residents live along the eastern and western fringe of the area, and there is a growing summer residential presence especially on the southern end of the plateau. A highly developed road system makes the study area well accessible for puma research efforts. A detailed description of the Uncompahgre Plateau is in Pojar and Bowden (2004).

METHODS

Reference and Experimental Treatment Periods

This research is structured in two 5-year periods: a *reference period* (years 1―5) and a *treatment period* (years 6―10). The *reference period* is expected to cause a population increase phase. The *treatment period* will be managed to cause a population decline phase. In both phases, puma population structure, and vital rates will be quantified, and some management assumptions and hypotheses regarding population dynamics will be tested. Contingent upon results of pilot studies, we will also estimate puma numbers, population growth rates, evaluate enumeration methods, and test other hypotheses (Logan 2004).

The *reference period*, without recreational puma hunting as a major limiting factor, is consistent with the natural history of the current puma species in North America which evolved life history traits during the past 10,000―12,000 years (Culver et al. 2000) that enable puma to survive and reproduce (Logan and Sweanor 2001). In contrast, puma hunting, with its modern intensity and ingenuity, might have influenced puma evolution in western North America for the past 100 years. Hence, the *reference period*, years 1―5, will provide conditions where individual puma in this population (of estimated sex and age structure) express life history traits interacting with the environment without recreational hunting as a limiting factor. Theoretically, the main limiting factors will be catchable prey abundance (Pierce et al. 2000, Logan and Sweanor 2001). This should allow researchers to understand basic system dynamics *before* the treatment (i.e., controlled recreational hunting). In the *reference period*, all puma in the study area will be protected, except for individual puma that might be involved in depredation on livestock or human safety incidents. In addition, all radio-collared and ear-tagged puma that range in a buffer zone, that includes the northern halves of GMUs 61 and 62, will be protected from recreational hunting.

The *reference period* will allow researchers to quantify baseline demographic data on the puma population to estimate parameters for the CDOW's model-based approach to puma management. Moreover, it will allow researchers to develop and test puma enumeration methods when population growth is known to be in one direction― increasing. Without the hunting closure, pilot data for enumeration methods could be confounded by not knowing if the population was increasing, declining, or stable. The *reference period* will also facilitate other operational needs (because hunters will not be killing the animals) including the marking of a large proportion of the puma population for capture-mark-recapture estimates, and the gathering of movement data from GPS-collared puma to help formalize exact sampling designs for enumeration methods.

During the *treatment period*, years 6―10, experimentally structured recreational puma hunting will occur on the same study area with the intent of causing a decline phase in the puma population by using management prescriptions structured from information learned during previous years. Using recreational hunting for the treatment is consistent with the CDOW's objectives of manipulating natural tendencies of puma populations, particularly survival, to maintain either population stability or population suppression (CDOW, Draft L-DAU Plans, 2004). Theoretically, puma survival will be influenced mainly by recreational hunting, which will be quantified by agent-specific mortality rates of radio-collared puma. The portion of adults and subadults in the population will be reduced by approximately 20% in year 6 and 20% more in year 7. The 20% change was identified by Division managers that requested enumeration tools that might detect 20% changes in puma populations. For managers, detecting the magnitude of puma population decline phases is probably more important that detecting the magnitude of population increase phases. This will also allow quantification of puma population characteristics and vital rates and initial tests of enumeration methods during a decline phase.

Additional reductions may be made to test enumeration methods and other hypotheses that may be related to effects of hunting (i.e.,: relative vulnerability of puma sex and age classes to hunting, variations in puma population structure due to hunting) and puma—prey interactions (i.e., lines of research identified in the Colorado Research Program, Fig. 1). Those decisions can be made later in project development and as late as years 8—10. The killing of tagged and collared puma during the *treatment period* will not hamper operational needs (as it would during the start-up years), because by the beginning of this period, a large majority of independent puma in the population will be marked, and sampling schemes will be formalized.

Puma on the study area that may be involved in depredation of livestock or human safety incidences may be lethally controlled. Researchers that find that GPS-collared puma have killed domestic livestock will record such incidents to facilitate reimbursement to the property owner for loss of the animal(s). In addition, researchers will notify the Area Manager of the CDOW of Wildlife if they perceive that an individual puma may be a threat to public safety.

Field Methods

Puma Capture: Realizing that puma live at low densities and capturing puma is difficult, as a starting point, our logistical aim will be to have a *minimum* of 6 puma in each of 6 categories (36 total) radio-tagged in any year of the study if those or greater numbers are present. The 6 categories are: adult female, adult male, subadult female, subadult male, female cub, male cub. Our aim is to provide more quantitative and precise estimates of puma demographics than were achieved in earlier Colorado puma studies. This relatively large number of puma might represent the large majority of the puma population on the study area, and will provide the basic data for age- and sex-specific reproductive rates, survival rates, agent-specific mortality rates, emigration rates, and movement data pertinent to sampling designs for various projects.

Assuming that the puma population density on the study area is relatively low at the beginning of this study— about 1 adult/100 km^2 and the sex ratio is equal (Anderson et al. 1992, Logan and Sweanor 2001:167), then there might be 22 adults, 11 males and 11 females. Also assuming that the total population contains 10% subadults and 34% cubs (Logan and Sweanor 2001), then there might be 4 subadults and 13 cubs with equal sex ratios in a total population of 39 puma. If we achieve our logistical aim in the first 1—2 years (recognizing that the population might grow), then we should be able to quantify population characteristics and vital rates for a majority of the puma population in those years and build upon the tagged number in each subsequent year. Thus, our inferences will pertain to the large majority of the puma population, if not the population on the study area, instead of a relatively small sample of it. We anticipate it may take 2 years to mark the large majority of puma in the population. In addition, the study area is large and will require some time to learn to access it efficiently.

Puma capture and handling procedures have been approved by the CDOW Animal Care and Use Committee (file #08-2004). All captured puma will be examined thoroughly to ascertain sex and describe physical condition and diagnostic markings. Age of adult puma will be estimated initially by the gum-line recession method (Laundre et al. 2000) and dental characteristics of known-age puma (Logan and Sweanor, unpubl. data). Ages of subadult and cub puma will be estimated initially based on dental and physical characteristics of known-age puma (Logan and Sweanor unpubl. data). Body measurements recorded for each puma will include at a minimum: mass, pinna length, hind foot length, plantar pad dimensions. Tissue collections will include: skin biopsy (from the pinna receiving the 6 mm biopsy punch for the ear-tags) and blood (30 ml from the saphenous or cephalic veins) for genotyping individuals, parentage and relatedness analyses; disease screening; hair (from various body regions) and fecal DNA for genotyping tests of field gathered samples. Universal Transverse Mercator Grid Coordinates on each captured puma will be fixed via Global Positioning System (GPS, North American Datum 27).

Puma will be captured year-round using 4 methods: trained dogs, cage traps, foot-hold snares, and by hand (for small cubs). Capture efforts with dogs will be conducted mainly during the winter when snow facilitates thorough searches for puma tracks and the ability of dogs to follow puma scent. The study area will be searched systematically multiple times per year by four-wheel-drive trucks, all-terrain vehicles, snow-mobiles, walking, and possibly horse- or mule-back. When puma tracks ≤1 day old are detected, trained dogs will be released to pursue puma to capture.

Puma usually climb trees to take refuge from the dogs. Adult and subadult puma captured for the first time or requiring a change in telemetry collar will be immobilized with Telazol (tiletamine hydrochloride/zolazepam hydrochloride) dosed at 5 mg/kg estimated body mass (Lisa Wolfe, DVM, CDOW, attending veterinarian, pers. comm.). Immobilizing agent will be delivered into the caudal thigh muscles via a Pneu-Dart® shot from a CO_2-powered pistol. Immediately, a 3m-by-3m square nylon net will be deployed beneath the puma to catch it in case it falls from the tree. A researcher will climb the tree, fix a Y-rope to two legs of the puma and lower the cat to the ground with an attached climbing rope. Once the puma is on the ground, its head will be covered, its legs tethered, and vital signs monitored (Logan et al. 1986). (Normal signs: pulse ~70—80 bpm, respiration ~20 bpm, capillary refill time ≤2 sec., rectal temperature ~101°F average, range = 95—104°F) (Kreeger 1996).

A cage trap will be used to capture adults, subadults, and large cubs when puma can be lured into the trap using road-killed or puma-killed ungulates (Sweanor et al. 2005). Efficiency of the trap might be enhanced by using an automated digital call box that emits puma vocalizations (Wildlife Technologies, Manchester, NH). A cage trap will be set only if a target puma scavenges on the lure (i.e., an unmarked puma, or a puma requiring a collar change). Researchers will continuously monitor the set cage trap from about 1 km distance by using VHF beacons on the cage and door. This allows researchers to be at the cage to handle captured puma within 30 minutes. Puma will be immobilized with Telazol injected into the caudal thigh muscles with a pole syringe. Immobilized puma will be restrained and monitored as described above. If non-target animals are caught in the cage trap, we will open the door and allow the animal to leave the trap.

Foot-hold snares will be used to capture adults, subadults, and large cubs only when safe snare sites at puma kills can be located as described by Logan et al. (1999). Snares set at puma kills will be monitored continuously with VHF beacons on the snares from about 1 km distance. We will not set snares at sites where tracks indicate that other mammals (e.g., deer, elk, bear, bighorn sheep, livestock) are also active. Puma will be immobilized with Telazol injected into the caudal thigh with a pole syringe. Vital signs will be monitored during the handling procedures. Efficiency of snares might also be enhanced with the use of an automated call box with puma or prey vocalizations.

Small cubs (≤10 weeks old) will be captured using our hands (covered with clean leather gloves) or with a capture pole. Cubs will be restrained inside new burlap bags during the handling process and will not be administered immobilizing drugs. Cubs at nurseries will be approached when mothers are away from nurseries (as determined by radio-telemetry). Cubs captured at nurseries will be removed from the nursery a distance of ~100 m to minimize disturbance and human scent at nurseries. Immediately after handling processes are complete, cubs will be returned to the exact nurseries where they were found (Logan and Sweanor 2001).

Marking, Global Positioning System- and Radio-telemetry: Puma do not possess easily identifiable natural marking, such as tigers (see Karanth and Nichols 1998, 2002), therefore, the capture, marking, and GPS- or VHF- collaring of individual puma is essential to a number of project objectives, including estimating vital rates and gathering movement data on puma to formalize designs for developing and testing enumeration methods. Adult, subadult, and cub puma will be marked 3 ways: GPS/VHF- or VHF-collar, ear-tag, and tattoo. The identification number tattooed in the pinna is permanent and cannot be lost unless the pinna is severed. A colored (bright yellow or orange), numbered rectangular (5 cm x 1.5 cm) ear-tag (Allflex USA, Inc., DFW Airport, TX) will be inserted into each pinna to facilitate individual identification during direct recaptures. Cubs ≤10 weeks old will be ear-tagged in only one pinna.

Locations of GPS- and VHF-collared puma will be fixed about once per week from light fixed-wing aircraft (e.g., Cessna 182) fitted with radio signal receiving equipment (Logan and Sweanor 2001). This monitoring will enable researchers to find GPS-collared puma to acquire remote GPS location reports from the ground, monitor the status (i.e., live or dead) of individual puma, and to recover carcasses for necropsy. It will also provide simultaneous location data on mothers and cubs. GPS- and VHF-collared puma will be located from the ground opportunistically using hand-held yagi antenna. At least 3 bearings on peak aural signals will be mapped to fix locations and estimate location error around locations (Logan and Sweanor 2001). Aerial and ground locations will be plotted on 7.5 minute USGS maps (NAD 27) and UTMs along with location attributes will be recorded on standard forms. GPS locations will be mapped using ArcGIS software.

Adult and subadult female pumas will be fitted with GPS collars (approximately 400 g each, Lotek Wireless, Canada). Initially, GPS-collars will be programmed to fix and store puma locations at 4 times per day to sample daytime, nighttime, and crepuscular locations (i.e., 0:00, 06:00, 12:00, 19:00). GPS locations for puma will provide precise, quantitative data on puma movements mainly to provide data to formalize study designs, to test assumptions for capture-mark-recapture methods for this project, and to assess the relevance of puma DAU boundaries. The GPS-collars also will provide basic information on puma movements and locations to design other pilot studies in this program on vulnerability of puma to sport-harvest, habitat use, and predation frequency on mule deer and elk.

Subadult male pumas will be fitted initially with conventional VHF collars (Lotek, LMRT-3, ~400 g each) with expansion joints fastened to the collars, which allows the collar to expand to the average adult male neck circumference (~46 cm). If subadult male puma reach adulthood on the study area, we will recapture them and fit them with GPS collars.

VHF radio transmitters on GPS collars will enable researchers to find those pumas on the ground in real time to acquire remote GPS data reports, facilitate recaptures for re-collaring, and to check on their reproductive and physical status. VHF transmitters on GPS- and VHF-collars will have a mortality mode set to alert researchers when puma have been immobile for at least 3 hours so that dead puma can be found to quantify survival rates and agent-specific mortality rates by gender and age.

We will attempt to collar all cubs in observed litters with small VHF transmitter mounted on an expandable collar (~100g, MOD 210, Telonics, Inc., Mesa, Arizona) when cubs weigh 2.3—11 kg (5—25 lb). Cubs with mass ≥11 kg can still wear these small expandable collars until they are about 12 months old. Cubs approaching the age of independence (~11—14 mo. old) may be fit with Lotek LMRT-3 VHF collars (~400 g) with expansion links. Cubs will be recaptured to replace collars as necessary. Monitoring radioed cubs allow quantification of survival rates and agent-specific mortality rates (Logan and Sweanor 2001).

Capture-Mark-Recapture: Capture-mark-recapture methods will be evaluated initially as a pilot study. Capturing and marking puma is time consuming, and would lengthen the time to thoroughly search the study area for capturing and marking puma during capture-recapture occasions needed for population estimation. Therefore, we will capture and mark pumas prior to performing capture-recapture or re-sight occasions using using methods such as houndsmen teams or trail cameras. In addition, by marking puma before capture-recapture occasions begin, we will have opportunities to capture female puma at different stages of their reproductive status, and thus reduce the chance that mothers in a stage with suckling cubs and small activity areas are not detected and marked on the study area. After cubs are weaned, the mothers' activity area expands (Logan and Sweanor 2001). The probability of females having suckling cubs in winter is naturally small; that season exhibits the lowest rate of births (Logan and Sweanor 2001). Capture-recapture occasions to estimate the population of independent puma may not begin until the end of the second winter or the third winter when we have a large majority of the puma population sampled and marked. Occasions performed at that time will be viewed as a pilot study allowing us to examine the logistics of the field methods, the extent to which model assumptions are met, performance of field methods (e.g., detection differences by sex or life stage as revealed by GPS data on collared puma), and precision of capture-recapture models used to estimate the puma population.

Analytical Methods

Population Characteristics: Population characteristics each year will be tabulated with the number of individuals in each sex and age category. Age categories, as mentioned, include: adult (puma ≥24 months old, or younger breeders), subadults (young puma independent of mothers, <24 months old that do not breed), cubs (young dependent on mothers, also known as kittens) (Logan and Sweanor 2001). When data allow, age categories may be further partitioned into months (for cubs and subadults) or years (for adults).

Reproductive Rates: Reproductive rates will be estimated for GPS- and VHF-collared female puma directly (Logan and Sweanor 2001). Genetic paternity analysis will be used to ascertain paternity for adult male puma (Murphy et al. 1998). Methods will be tested in Dr. M. Douglas's Laboratory (Colorado State University, Department of Fishery and Wildlife Biology).

Survival and Agent-specific Mortality Rates: Radio-collared puma will provide known fate data which can be used to estimate survival rates for each age stage using the binomial survival model (Williams et al. 2001:343-344) or analyzed in program MARK (White and Burnham 1999, Cooch and White 2004). Agent-specific mortality rates can be analyzed using proportions and Trent and Rongstad procedures (Micromort software, Heisey and Fuller 1985). Cub survival curves for each gender will be plotted with survival rate on age in months (Logan and Sweanor 2001:119).

Population Estimates: Capture-recapture models will be evaluated initially as a pilot study to estimate the parameters of primary interest— absolute numbers of independent puma (i.e., number of adult and subadult puma present in the survey area) and puma density (i.e., number of independent puma/100 km^2) each winter— December through March— when snow facilitates detection and capture of puma, provided that we meet model assumptions. The December—March period also corresponds with Colorado's puma hunting season. The population of interest is independent puma (i.e., adults and subadults) because those are the puma that can be legally killed by recreational hunters. Furthermore, adults comprise the breeding segment of the population and subadults are non-breeders that are potential recruits into the adult population in ≤1 year. Thus, the sampling unit is the individual independent puma (~≥1 yr. old).

General assumptions for closed capture-recapture models are: (1) the population is closed; (2) animals do not lose their marks during the interval; (3) all marks are correctly noted and recorded at each trapping occasion; (4) each animal has a constant and equal probability of capture on each capture occasion. Open population models allow the assumption of closure to be relaxed (Otis et al. 1978, White et al. 1982, Pollock et al. 1990). The robust design is a combination of closed and open models; thus, assumptions are a combination of the assumptions for closed and open population methods (Kendall 2001).

To analyze capture-recapture data, closed, open, and the robust design models are available in program MARK. Akaike's Information Criterion will be used to select the most parsimonious models based on AICc score ranks and the difference in AIC (ΔAIC) between models (Burnham and Anderson 1998). MARK results also include estimates of abundance.

Because the precision of estimates for small populations is sensitive to the probability of capture (White et al. 1982, Pollock et al. 1990), our operational goal will be to achieve capture probabilities of at least 0.5 for each animal per capture occasion. Capture simulations using MARK software (Cooch and White 2004) indicate that greater capture probabilities and more capture occasions yield more precise estimates. The capture probability for the simplest closed model [M(o)], which assumes that every member of the population has the same probability of capture *(p)* for each sampling period, suggest that for a population of 30 animals (i.e., adults plus subadult puma, which might be present by the end of year 2, see Puma Capture above) *p* must equal 0.5 for 3 capture occasions to attain a coefficient of variation (*V)* of 0.1. If 6 capture occasions are used, then a *p* of 0.3 might yield a *V* of 0.09.

In addition, behavior, movements, survival and mortality of GPS- and VHF-collared puma will allow direct biological examinations of assumptions of geographic and demographic closure (White et al. 1982) and variation in capture probability of individual puma and puma classes (i.e., adult females, adult males, subadult females, subadult males). If capture probabilities vary by puma class, we will examine if data stratification is necessary or possible (depending upon sample size). For example, we might expect the larger home ranges of male puma to expose them to more search routes, thus, this may increase their probability of capture. If the assumption of demographic closure cannot be satisfied, then open population models and the robust design would be more appropriate (Pollock et al. 1990, Williams et al. 2001). Collared puma will allow us to determine the number of marked puma present in the search area each capture-recapture occasion. Furthermore, GPS locations (4 fixes/day) on individual puma will provide data on the probability that puma may temporarily move out of and back into the survey area between capture occasions. Unmarked puma that are subsequently GPS-collared should provide such information, too.

ArcView geographic information system software will be used to map and analyze puma locations, movements, and home ranges. It will also be used to map and quantify attributes of the study area and sampling frames.

Rate of Population Increase: Finite rates of increase ($\lambda = N_{t+1}/N_t$) between consecutive years and average annual rates of increase *(r)* for 3- to 5-year periods and levels of precision will be calculated (Caughley 1978, Van Ballenberghe 1983) and plotted.

Functional Relationships: Graphical methods will be used to examine functional relationships between puma density and vital rates, relationships between puma density estimated with direct capture-recapture methods (i.e., houndsmen teams) and possibly later (depending upon funding) by using estimates from DNA genotype or other mark- recapture methods. Linear regression procedures and coefficients of determination can be used to assess these functional relationships if data for the response variable are normally distributed and the variance is the same at each level. If the relationship is not linear, data is non-normal, and variances are unequal, we will consider appropriate transformations of the data for regression procedures (Ott 1993). Non-parametric correlation methods, such as Spearman's rank correlation coefficient, can also be used to test for monotonic relationships between puma abundance and other parameters of interest (Conover 1999).

Statistical analyses will be performed using SYSTAT and SAS software. The risk of committing a type I error (i.e., rejecting a null hypothesis that is actually true) will be controlled at alpha = 0.10 because we will normally have small population sizes (typical of studies of large obligate carnivores). The higher alpha level will increase the probability of detecting a change and reduce the risk of a type II error (i.e., failing to reject a null hypothesis that is false). For managers, the risk of a type II error is probably more important.

RESULTS AND DISCUSSION

Segment Objective 1

Field research to quantify puma population structure, vital rates, and causes of mortality for this report extended from August 2006 to July 2007. Our searches to detect puma presence covered the entire study area. We allocated most of our effort in areas where we consistently found tracks that we thought were of unmarked pumas, particularly in the northeast and southwest areas where we found little or no evidence of pumas during the previous 2 years. We made 54 puma captures during the period (9-10 adult females [1 adult female captured twice], 7 adult males [1 adult male probably captured 3 times, another captured twice], 1 subadult female, 0-1 other subadults, and 30 cubs [4 of them captured twice each]).

As our main method to capture, sample, and mark adult and subadult pumas, we used trained dogs from November 13, 2006 to May 11, 2007. Those efforts resulted in 78 search days, 177-178 puma tracks detected, 45-47 pursuits, and 22 puma captures (Table 1). Puma capture efforts (i.e., search days) with dogs in this period was similar to our efforts in the 2 previous efforts (Table 2). But, the frequency of pursuits and puma captures has increased over the 2 previous periods. In addition, the number of adult and subadult pumas captured for the first time declined from 11 (Oct. 2005 to Apr. 2006) to 6 (this period). This included 1 adult female or subadult puma that could not be handled for safety reasons (see Tables 3 and 4). Of the pumas we captured, but could not handle, it is probable that we captured and marked 1 adult male (M51) and 1 adult female (F50) in subsequent capture efforts.

Our puma capture efforts using ungulate carcasses and cage traps extended from August 2006 to July 2007. We used 64 road-killed mule deer, 7 road-killed elk, 3 puma-killed mule deer, and 1 puma killed elk at 26 sites to capture pumas 8 times (Tables 5). Pumas scavenged 16 of 71 (22.5%) of the road-killed ungulate carcasses used for bait. This was similar to the results last years (16 of 80, 20%).

Five pumas were captured, sampled, and marked for the first time by using dogs and cage traps, (Table 3). Fifteen recaptures of 13 marked pumas were made with the use of dogs and cage traps; GPS/VHF collars were replaced as needed (Table 6). We captured, sampled, and marked 26 cubs in 10 litters that were captured by hand at nurseries (Table 7).

Search efforts throughout the study area also revealed the presence of at least 4 other independent females and 1 independent male. The tracks we found of those animals were too old to pursue (i.e., probability of capture with the dogs was negligible). We could separate the activity of those pumas from the GPS- and VHF- collared pumas in time and space. In addition, 2 of the females were in association with cubs. One female was followed by 2 cubs about 5 to 6 months old in December and January, when we captured but could not handle 1 or 2 of the cubs (Table 4). Another female was followed by 1 large cub (probably a male) likely 10 or more months old. And another female on the southwest portion of the study area might have been an adult if it were associated with a female cub (~6 mo. old) that was hit and killed by a vehicle on highway 62 on January 28, 2007.

Our search and capture efforts during November 2006 through May 2007 enabled us to estimate a minimum count of 24 independent pumas detected on the Uncompahgre Plateau study area. The count included 16 females and 8 males. Of those, 12 adult females and 7 adult males were probably marked animals (79% of independent pumas detected). Of the remainder, 2 females were adults because they were followed by cubs, and 2 females and 1 male were of unknown independent status (i.e., either subadult or adult). Figure 2 indicates the estimated use areas of those independent pumas. Some of the animals range outside the borders of the study area, as indicated by movements of GPS- and VHF-collared pumas. There appears to be variation in puma numbers on the west and east slopes of the study area. The west slope count includes 8 independent pumas (5 females— 4 marked, 1 unmarked; 3 males— 2 marked, 1 unmarked). The east slope count includes 16 independent pumas (11 females— 8 marked, 3 unmarked; 5 males— all marked). Female home ranges overlap other female home ranges extensively, and are overlapped by male home ranges. Male home ranges overlap multiple female home ranges, and overlap other male home ranges somewhat.

Anderson et al. (1992) studied pumas on the east slope of the Uncompahgre Plateau (i.e., GMU 62) during 1981 to 1988. Sport-hunting was banned during that study as it is in this study Reference Period. As our current effort results in larger samples and progresses in time through the Reference and Treatment periods, similarities and differences in results of the 2 research efforts, now separated by more than 15 years, should illuminate reliable knowledge for puma management in Colorado. Our current puma research on the Uncompahgre Plateau has been underway for 2.7 years (compared to 7 years of Anderson et al. 1992). Our data analysis at this stage of the research is not by any means exhaustive or complete, yet, our data set enables some preliminary comparisons with Anderson's completed work (Anderson et al. 1992).

In the Anderson et al. (1992) study, the average capture effort with dogs was 91.1 days per winter (range = 32 to 136, $n = 7$) resulting in an average capture effort of 13.9 days per puma. Of 189 pursuits of pumas, 110 (58%) were successful (either of radio-collared or non-collared animals). They captured 47 pumas for an average capture rate of 13.9 days per puma. Eight other pumas, all female cubs ≤7 months old, were caught in steel leg-hold traps by trappers, and were added to the study animal population.

So far, in our 3 winters, the average effort is 79.3 days (range = 78 to 82). Of 123 pursuits, 50 (41%) were successful. We captured and GPS- or VHF-collared 25 pumas for the first time, yielding a capture rate of 10.0 days per capture. Other capture efforts and results between the 2 studies are not comparable, because Anderson et al. (1992) did not routinely attempt to capture pumas using cage traps or at nurseries like we are. In their effort, Anderson et al. (1992) captured 57 pumas, of which 49 were radio-collared. In our current effort, we captured, sampled, and marked 68 pumas, of which 61 were radio-collared.

Puma mass recorded by Anderson et al. (1992:86) for pumas having an estimated age ≥24 months, averaged 61.6 kg for 8 males, (*SD* = 5.7, range = 51.8 to 70.8) and 44.5 kg for 14 females (*SD* = 3.6, range = 38.5 to 49.9). So far in our current study, mass for pumas ≥24 months old averaged 59.8 kg for 9 males (*SD* = 8.1, range 40 to 68 kg) and 38.4 kg for 11 females (*SD* = 4.9, range = 31 to 46). Sexual dimorphism has been described for puma throughout the species range (Young and Goldman 1946) and has been explained as a potential result of sexual selection (Logan and Sweanor 2001:109).

Segment Objective 2

We captured, sampled, and marked 26 puma cubs produced by 10 females (Table 7). Twenty-three of the cubs were examined at 8 nurseries when the cubs were 29 to 41 days old. The sexes were 17 males and 6 females. Four other cubs, including 2 males and 2 females, were caught when they were about 158 to 215 day old. In addition to those offspring, 2 cubs, about 152 to 183 days old, were detected in association with an unmarked female that we pursued. One or 2 of those cubs were captured in different events; the female sex was determined for one of the cubs. But, neither cub could be handled safely for further sampling. The estimated birth month for the 10 litters were April (1), May (1), July (5), August (2), and September (1).

During the past 27 months of this work we compiled data on puma reproduction that was heretofore not available for Colorado. We examined 38 cubs from 13 litters aged 29 to 42 days old where we were reasonably sure that we examined all the cubs at the nurseries. The sex ratio of the observed cubs was 21 males:17 females. The mean (±*SD*) and extremes of litter sizes were 2.84 (±0.99), 1 to 4. The distribution of puma births by month indicate puma births extending from March into September, with 18 of 20 births occurring May to September (Fig. 3). In addition, 8 birth intervals for 7 different female pumas averaged 14.99 months (*SD* = 3.40), and ranged from 11.7 to 20.5 months (Table 8). Based on observations (from GPS data) of associations between 4 mothers and putative sires, 4 gestation periods averaged 92.0 days (*SD* = 1.68), which is consistent with average puma gestation reported in literature (i.e., mean ± *SD* = 91.9 ± 4.1, Anderson 1983:33, mean = 91.5 ± 4.0 Logan and Sweanor 2001:414).

Anderson et al. (1992:47) reported of “17 postnatal litters about 10-240 days in estimated age from 12 individual females, the mean (±SD) and extremes of litter sizes were 2.41 ± 0.8, 1-4”. “Because most postnatal young were not handled, their sex ratio is unknown” (Anderson et al (1992:48). In addition, because cubs were first observed at older ages, it is likely that some post-natal mortality had occurred. This is one explanation for smaller litters observed by Anderson et al. (1992).

Anderson et al. (1992:47-48) found that of 10 puma birth dates 7 were during July, August, and September, 2 in October, and 1 in December, with most breeding occurring April through June. Data on our 20 litters adds to Anderson’s data (Fig. 3), and indicates puma births in Colorado occurring in every month except January and November (so far). Our data suggests that the majority of puma breeding activity occurs February through June. Anderson’s observation of two 12-month birth intervals for one female (Anderson et al. 1992:48) is at the low range of our observations (above).

<u>Segment Objective 3 & 4</u>

From December 2, 2004 (start of our research) to July 31, 2007, we radio-monitored 9 adult male and 12 adult female pumas to quantify survival and agent-specific mortality rates (Table 9). One adult male is known to have died. M4 was about 37 to 45 months old when he was killed by an unidentified male puma along the southeast boundary of the study area. We lost contact with 3 adult males apparently due to GPS/VHF collar failure (M1, M6, M27). Evidence in the field suggests that all 3 males might still be alive. One adult female is known to have died. F50 was about 29 to 31 months old when she apparently died of natural causes (exact agent could not be identified).

We have radio-monitored 6 subadult pumas (Table 10). None of those died while we were monitoring them. F23 has become a breeding adult on the study area. M5 dispersed from his natal area and the study area at about 13 months old and went to the northwest slope of the Uncompahgre Plateau where he has apparently established an adult territory. M49 was orphaned at 9 months old when his mother F50 died. He has since dispersed from his natal area and the study area to the northeast slope of the Uncompahgre Plateau. We continue to monitor his status. On the other hand, we have lost contact with 2 subadult males and 1 subadult female. Puma M11 became a subadult at 13 months old and dispersed from his natal area at 14 months old. He was last located in the Dolores River valley between Stapletone and Stoner, Colorado, on December 14, 2006. F52 dispersed from the study area before we lost track of her in the area of the Black Canyon of the Gunnison in mid-May 2007. We lost track of M31 seven days after he was captured. He might have dispersed from the study area. Efforts to locate him by flying over and around the study area have not been successful. Dispersal rates and distances will be reported after we have compiled more complete data. In addition to the subadults discussed above, a non-marked female puma about 18 to 24 months old was killed by a vehicle November 4, 2006 on highway 550, which forms the southeast boundary of our study area. The female appeared to be in good health (41 kg), was not pregnant, and was not lactating.

Anderson et al. (1992) found that all 9 radio-collared male pumas dispersed from their natal areas, and 2 of 6 radio-collared females did not disperse from their natal areas (A. E. Anderson, Sep. 1993, errata for Anderson et al. 1992:61). Mean ± *SD* and range of dispersal distances (km) for 8 males, aged 10 to 13 months old at dispersal, were 86.2 ± 51.3, 23 to 151. For 4 females, aged 11 to 31 months old at dispersal, mean ± *SD* and range of dispersal distances (km) were 37.0 ± 15.3, 17 to 54 (Anderson et al. 1992:63).

The current closure on sport-hunting on the study area and protection of marked pumas from sport-harvest on the buffer area on the northern portion of the Uncompahgre Plateau for the Reference Period appears to be operating, so far. None of the adult or subadult pumas wearing functional GPS- or VHF- collars have died due to human causes. This reference condition enables us to quantify puma survival rates and agent-specific mortality rates of adult and subadult pumas (i.e., harvest-age pumas) in the absence of direct human-caused mortality factors related to sport-hunting. So far, survival of radio-monitored adult and subadult pumas in the study and buffer areas appears to be high. In addition, the population sex and age structure can be examined in this reference condition. As indicated in Figure 4, the adult age structure appears to be indicative of high survival rates during the past 3 winters without sport-hunting mortality. These data will be valuable in comparisons of sex and age structure during the Treatment Period and with the structure of harvested pumas in other regions of Colorado. But, we will wait for greater sample sizes (i.e., greater numbers of radio-monitored pumas and duration) before we develop more quantitative analyses of survival rates and agent-specific mortality rates and attendant inferences.

Thirty-nine puma cubs (20 males, 19 females) have been monitored by radiotelemetry for varying durations (Table 11). Three males (M5, M11, M49) are known to have survived to the subadult or adult stages (Table 10). Seven cubs (F13, F18, M22, F33, F34, F36, and M37) were killed and eaten by other pumas. At least 4 of those were subjects of male-induced infanticide. Sex of the puma involved in each of the other cases could not be determined. In addition, cub F45 was apparently killed by coyotes when she was 280 to 283 days old. F45 was separated from her adopted mother, F2, and also appeared to be emaciated at the time of her death. Cub F17 was killed by a vehicle on highway 550 when she was about 330 days old. She was not radio-collared at the time, but GPS data from her mother, F16, showed her in the vicinity of her offspring. Thus, F17 was probably still dependent on F16. Three cub deaths were due to our research activities, namely problems with the expandable radiocollars. F35 died at 37 days old probably as a result of starvation caused when the transmitter box got caught in her mouth. M42 died at 106 days old apparently from complications of septicemia caused by an infection at the axis of the right foreleg. The cub put his right foreleg through the expandable collar and the collar material lacerated the right underarm as the animal grew, enabling the infection. M60 died at 49 days old, apparently from starvation. He apparently could not keep up with the movements of his mother, because he had put his right foreleg through the expandable collar, restricting his mobility. In addition to these deaths, 1 unmarked female cub (~6 mo. old) was killed by a vehicle on highway 62 on the southwest boundary of the study area on January 28, 2007 (mentioned earlier). We lost contact with a number of cubs because they shed their expandable radiocollars (Table 11). As this study proceeds, some cubs with which we have lost contact will be re-captured, re-observed, or harvested, and thus, provide more complete survival information.

Clearly, data on cub survival and mortality are still preliminary. At this time, we can say that a minimum of 12 deaths occurred in 39 radio-collared cubs that we monitored for varying periods of time. This represents a minimum 0.31 mortality rate (12/39), including research-related causes. Subtracting the 3 research-related deaths, the minimum mortality rate is 0.25 (9/36). The main cause of death is being killed by another puma (0.78, 7/9). These rates should be interpreted as only rudimentary information. More complete data on cub survival and mortality will be forthcoming as our efforts continue.

Anderson et al. (1992:50) reported on the fates of 21 radio-collared pumas (11 <24 months old, 10 ≥24 months old) from a total of 49 in the previous study where pumas were not hunted. Yet, 19 of those pumas died due to human causes, attributed to: legal kill outside the study area (7), capture-related (6), predator management (3), illegal kill (2), and suspected predacide (1). Other causes of mortality included, intraspecies strife (1) and disease (1). Actual age-stage and annual survival rates and agent-specific survival rates from our current effort will be compared with the Anderson et al. (1992) data set at a later date when we have greater samples, duration in research time, and more complete fate data (i.e., pumas currently without functional collars) to make such comparisons meaningful. Differences might be illuminated. For example, research of a puma population in New Mexico that was not hunted for 10 years indicated that the major cause of death for both sexes and all age stages of pumas was intraspecifies strife, and male-induced infanticide (Logan and Sweanor 2001).

Although we have observed 3 male pumas disperse from natal areas, and no females disperse, our current research is too short in duration and samples too small yet to make meaningful comparisons with Anderson's earlier effort, particularly regarding offspring dispersal rates, distances moved, and philopatry. Dispersal and philopatry have been explained as life history strategies in pumas that assist gene flow, colonization, population maintenance, and individual survival and reproductive success (Logan and Sweanor 2001). Thus, such strategies would be expected to be conserved, and expressed in puma populations at different times and different locations. In addition, because puma emigration and immigration (i.e., via dispersal) have been shown to be important processes in puma population dynamics (Sweanor et al. 2000), we need larger samples and longer research duration in this study to estimate those parameters.

Segment Objective 5

Twenty adult pumas (7 males, 13 females) were fit with Lotek 4400S GPS collars since field research began in December 2004. The collars are programmed to fix 4 locations per day (00:00, 06:00, 12:00, and 19:00). The number of GPS locations per individual puma ranged from 113 to 2,759 (Table 12). Winter activity areas for GPS-collared pumas were estimated (Table 13) with fixed kernel and minimum convex polygon home range estimators (ArcView 3.2 Animal Movement Extension). These estimates are intended for use in developing the sampling frame for the puma population estimation pilot project (see Introduction). In addition, 5 adult and subadult pumas have been monitored with VHF radiocollars (Table 14).

Anderson et al. (1992) provided an exhaustive analysis of seasonal puma home ranges and movements using data collected from VHF-collared animals during 1982 to 1988. We have not yet conducted an exhaustive analysis of adult puma home ranges and movements with the GPS data from our current puma research efforts. Instead, we provide only limited descriptive information in Tables 13, 14 and Fig. 2. Given the different types of location data and analytical methods, only broad descriptive comparisons might be made between the 2 studies at this time. Elemental similarities in home range attributes of pumas in the Anderson et al. (1992) research and our current effort, include: current home ranges of some puma overlap extensively with home ranges of puma documented by Anderson et al (1992), home ranges of male and female pumas are large, male home ranges are larger than female home ranges, male home ranges overlap multiple female home ranges, female home ranges overlap other female home ranges sometimes extensively, male home ranges overlap other male home ranges to a lesser extent than female home ranges. These characteristics are generally similar for pumas in other populations that have been studied with adequate intensity and duration (Beier and Barrett 1993, Logan and Sweanor 2001), and reflect behavioral strategies of male and female pumas that seem to contribute to individual survival and reproductive success (Logan and Sweanor 2001).

Segment Objective 6

To investigate the potential that puma hunters might detect puma mothers away from their cubs, we continued gathering data on spatial associations of puma mothers and their cubs during the puma hunting season, which extends from November through March each winter in Colorado. Female pumas are fair game in Colorado, unless they are accompanied by 1 or more cubs. Mothers that are caught away from their cubs could be legally harvested. Such incidents would result in cubs being orphaned. Orphaned cubs that ≤6 months old could have a survival rate (to the subadult stage) of <0.05. Orphaned cubs 7 to 12 months old might have a survival rate (to the subadult stage) of about 0.7 (K. Logan, unpublished data).

From November 7, 2006 to March 22, 2007 we located 1 to 4 radio-collared families of puma mothers and cubs from an airplane 49 times (Table 15).To assess whether mothers were apart or in close association with cubs, we needed to consider error in aerial locations. We recovered 7 puma radiocollars that we located from the airplane and fixed with GPS and then fixed the actual locations of collars on the ground with GPS. Range of location error was 20 to 520 m (mean = 282.86, *SD* = 164.75). We decided to use distances greater than the extreme high range of location error (520 m) as the metric to decide if puma mothers might be detected away from their cubs by hunters. Forty-one (83.7%) of observations located mothers and cubs ≤500 m apart, within the extreme margin of location error. Mothers were ≥520 m from their cubs during 8 (16.3%) of the observations (mean distance = 1,120 m, *SD* = 1,214.40, range = 616 to 4,101). The results for last winter were similar to our results the previous winter (15.2% and 16.3%, Table 15).

Anderson et al. (1992:70-71) recorded 69 instances of simultaneous aerial locations of 7 pairs of puma mothers and dependent young. They reported that mothers and young were together in 21 (30.4%) of those instances, and they were 1 to 2.2 km apart in 48 (69.6%) of those instances.

Segment Objective 7

Intensive effort to quantify puma use rates on ungulates by investigating puma GPS clusters continued during this period as an expansion of our pilot effort in the first research year (Logan 2005). That work proved the reliability of the GPS technology to allow us to gather quantitative information on ungulate prey use rates by pumas. In summary, 7 GPS-collared adult pumas (3 males, 4 females) used 61 mule deer, 48 elk, 2 porcupines, and 1 beaver found at 139 puma GPS clusters we investigated.

The current work is a collaborative effort among CDOW Mammals Researchers (M. Alldredge, E. Bergman, C. Bishop, D. Freddy, and K. Logan). This was another pilot effort because it involved the development and testing of clustering parameters, clustering routine, associated computer programs, and field investigation protocols. Here we report only the general summary of the pilot field investigations of puma GPS clusters from October 2006 to April 2007. Five types of puma GPS clusters (Bergman et al. 2006) were investigated for 13 GPS-collared adult pumas (8 female, 5 male). The sample unit was the individual puma. The field effort focused on investigating a sample of randomly chosen clusters from each cluster type. In addition, when other non-random S1 clusters (i.e., clusters with the highest probability of ungulate use detection) were conveniently located to random clusters targeted for investigation, field personnel would attempt to investigate those clusters, too. A total of 257 clusters were investigated, including 63 non-random S1 clusters, and 173 random clusters (S1, S2, S3, S4, S5). Mule deer and elk were about equally important to pumas as food (Tables 16, 17, 18). Other mammals were rarely found. The next step in this investigation involves examining the performance of all aspects of the GPS cluster investigations and modifying cluster parameters and field protocols to maximize the efficiency and reliability of our continuing efforts to quantify ungulate use by pumas on the Uncompahgre Plateau.

We will make further progress to designing and implementing a pilot project to investigate puma population estimation methods on the Uncompahgre Plateau. CDOW personnel Mat Alldredge, Chad Bishop, Ken Logan (Mammals Research) and Paul Lukacs (Terrestrial) met with Dr. Gary White (Colorado State University) June 21, 2007 to discuss possible approaches to estimating puma numbers by using capture-recapture methods and models. Another method we will explore, with the collaboration Mammals Researcher Chuck Anderson, is helicopter-based puma track probability sampling.

We will evaluate the potential for collaborative research on puma-human relationships on the Uncompahgre Plateau with the developing CDOW puma-human research on the Colorado Front Range. To date, we have gathered location data on 10 (7 adult females, 3 adult males) GPS-collared pumas with activity areas on the developed southeast portion of our study area, which includes: Fairway Pines, Loghill Village, and Fisher Creek subdivisions, numerous other private homes, Fairway Pines golf course and driving range, all adjacent to Ridgeway State Park (Fig. 2). In addition, 2 new subdivisions and golf courses are underdevelopment on the southeast quarter of the Uncompahgre Plateau. This is the same area that Anderson et al. (1992:80) received 17 useable questionnaires on puma observations from residents, and also had some radio-collared pumas frequenting these same developments. Linking puma-human research on the Uncompahgre Plateau and Front Range provides opportunities for increasing sample size (i.e., puma numbers, study sites) and observing variation in puma-human relationships.

We collaborated with Dr. Sue VandeWoude (CSU) to develop a pilot study titled: *Puma concolor immune health— Relationship to management paradigms and disease.* Tissue samples (i.e., blood, saliva, feces) from pumas we capture are collected and shipped to her laboratory for analyses. That project will be expanded to *The effects of urban fragmentation and landscape connectivity on disease prevalence and transmission in North American felids*. A description of that project and preliminary results on infectious disease surveillance on 21 pumas (13 female, 8 male) sampled on the Uncompahgre Plateau are presented in Appendix I.

SUMMARY

Manipulative, long-term research on puma population dynamics, effects of sport-hunting, and development and testing of puma enumeration methods began in December 2004. After 31 months of effort, 68 pumas have been captured, sampled, marked, and released. Of those 61 were radio-collared. Age stages we have monitored have included 21 adults, 6 subadults, and 46 cubs. Data from the marked animals are used to quantify vital rates and puma population dynamics in a *reference* situation (i.e., without sport-hunting off-take). Data on research efforts and puma capture, fates, reproduction, and activity areas are presented. During November 2006 through May 2007 a minimum count of 24 independent pumas were detected on the Uncompahgre Plateau study area. The count included 16 females and 8 males. Of those, 12 adult females and 7 adult males were probably marked animals (79% of independent pumas detected). Our efforts to quantify reproduction are yielding reliable data for Colorado on puma litter sizes, offspring sex ratios, and birth intervals. In this reference period, survival of adult and subadult pumas appears to be high. So far, the main cause of death in puma cubs is infanticide by males. Twenty adult pumas (13 females, 7 males) have been fitted with GPS collars, yielding 113 to 2,759 locations per puma. Our evaluations on the frequency that puma mothers on the Uncompahgre Plateau are away from their cubs >520 meters during the Colorado hunting season is low (15.2 to 16.3%). Intensive efforts to quantify puma use of ungulates on the Uncompahgre Plateau continued. Mule deer and elk appeared to be about equally important as puma food. Preliminary comparisons of aspects of puma biology were made between our new research effort on the Uncompahgre Plateau and that of Anderson et al. (1992) in GMU 62 during 1981 to 1988. Research efforts for year 4 will focus on increasing numbers and distribution of sampled, marked, and GPS/radio-collared pumas on the study area for the principal objectives of this research. In addition, we will continue to investigate puma use of mule deer and elk, develop a pilot project to estimate pumas, and consider incorporating our data on pumas on the Uncompahgre Plateau to address questions pertaining to research on puma-human relationships in Colorado. All of these efforts should enhance the Colorado puma research and management programs.

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Table 1. Summary of puma capture efforts with dogs from November 13, 2006 to May 11, 2007, Uncompahgre Plateau, Colorado.

Month	No. Search Days	No. & type of puma tracks found[a]	No. & type of pumas pursued	No. & I.D. or type of pumas captured
November	8	12 tracks: 3 male, 7 female, 2 cub	7 pursuits: 1 male, 4 females, 2 cubs	3 pumas captured 4 times: 1 male (not handled[b]), F3 twice (not handled once), cub M42 (died)
December	16	49 tracks: 7-8 male, 19-20 female, 22-23 cub	13 pursuits: 3 males, 4-5 females, 5-6 cubs	5 pumas captured 6 times: 1 male (not handled), F50, 1 female or subadult puma (not handled), cub M49 captured twice (not handled once), 1 cub (not handled)
January	19	56-58 tracks: 19 male, 30 female, 7-9 cub	9-10 pursuits: 3 males, 3 females, 3-4 cubs	3 pumas captured: 1 male (not handled), M51, 1 female cub (not handled)
February	8	4 tracks: 1 male, 1 female, 2 cub	3 pursuits: 1 female, 2 cubs	2 pumas captured: cubs M44 & M56
March	14	31-33 tracks: 8 male, 13 female, 10-12 cub	12 pursuits: 4 male, 5 females, 3 cubs	7 pumas captured: M29, F7, F23 (not handled), F24 (not handled), cubs M43 (not handled), M56 (not handled), & 1 unmarked female cub (not handled)
April	11	23 tracks: 13-16 male, 5-8 female, 2 cub	1 pursuit: 1 male	0 puma captured
May	2	2 tracks: 2 female	1 pursuit: 1 female	0 pumas captured
TOTALS	78	177-178 tracks: 51-55 male, 77-81 female, 45-50 cub	45-47 pursuits: 12 males, 18-19 females, 15-17 cubs	22 captures of 16 individuals: 7 pumas captured for the 1st time-M49, F50, M51, M56, & 1 female or subadult (not handled) & 2 female cubs (not handled), 1 adult male caught twice (not handled), 12 marked pumas were recaptured 15 times (including 4 caught for the 1st time this year).

[a] Puma hind-foot tracks with plantar pad widths >50 mm wide are assumed to be male; ≤50 mm are assumed to be female.

[b] Pumas are not handled for a variety of safety reasons: tree to dangerous to climb for researchers, puma treed near river, creek or cliff, puma might fall from tree after drug induction.

Table 2. Summary of puma capture efforts with dogs, December 2004 to May 2007, Uncompahgre Plateau, Colorado.

Period	Track detection effort	Pursuit effort	Puma capture effort	Effort to capture a puma for the first time
Dec. 2, 2004 to May 12, 2005	109/78 = 1.40 tracks/day	35/78 = 0.45 pursuit/day 78/35 = 2.23 day/pursuit	14/78 = 0.18 capture/day 78/14 = 5.57 day/capture	11 pumas captured for first time (minus M1, F3, & large female) 11/78 = 0.14 capture/day 78/11 = 7.09 day/capture
Nov. 21, 2005 to May 26, 2006	149/82 = 1.82 tracks/day	43/82 = 0.52 pursuit/day 82/43 = 1.91 day/pursuit	14/82 = 0.17 capture/day 82/14 = 5.86 day/capture	7 pumas captured for first time 7/82 = 0.08 capture/day 82/7 = 11.71 day/capture
Nov. 13, 2006 to May 11, 2007	177/78 to 182/78 = 2.27-2.33 tracks/day	45/78 to 47/78 = 0.58-0.60 pursuit/day 78/47 to 78/45 = 1.66-1.73 day/pursuit	22/78 = 0.28 capture/day 78/22 = 3.54 day/capture	7 pumas captured for first time 7/78 = 0.09 capture/day 78/7 = 11.14 day/capture

Table 3. Adult and subadult pumas captured for the first time, sampled, tagged, and released from December 2006 to January 2007, Uncompahgre Plateau, Colorado.

Puma I.D.	Sex	Estimated Age (mo.)	Mass (kg)	Capture date	Capture method	Location
F50	F	25-27	31	12-14-06	Dogs	West Fork Dry Creek
M51	M	44-49	61	01-07-07	Dogs	Lindsay Canyon
F52	F	18-20	38	01-10-07	Cage trap	Paco-Chu-Puk Campground, Ridgway State Park
F54	F	30	36	01-12-07	Cage trap	Pleasant View, Pleasant Valley
M55	M	24-36	62	01-21-07	Cage trap	Dallas Creek, Pleasant Valley

Table 4. Pumas that were captured with aid of dogs, but were not handled and marked at that time for either safety reasons or they escaped, November 2006 to May 2007, Uncompahgre Plateau, Colorado.

Puma sex	Age stage	Capture date	Location	Comments
Male	adult	11-13-06	East Fork Dry Creek	Unmarked puma climbed difficult spruce tree beside creek. This puma is probably M51 (captured & marked 01-07-07, identified with distinguishing notch in margin of right pinna).
Female, F3	adult	11-21-07	Dry Creek Basin	Puma F3 climbed dangerous tree adjacent to creek.
Unknown sex	cub	12-02-06	Dry Creek Basin	Unmarked cub was bayed on edge of high cliff. This cub was member of family comprised of an adult female & 2 cubs, which was probably pursued again on 01-25-07.
Unknown sex	subadult male or female, or adult female	12-05-06	Dry Creek Basin	This unmarked puma was treed on the same day that we captured & handled cub M49. This puma could have been M49's mother or a subadult puma (sex uncertain).
Male	adult	12-18-06	Lower East Fork Dry Creek	Unmarked puma climbed difficult fir tree on steep slope. This puma was probably M51 (captured & marked 01-07-07, identified with distinguishing notch in margin of right pinna).
Female	cub	01-25-07	Piney Creek	Unmarked cub climbed tree. Anesthesia was attempted with pole syringe. Cub jumped from tree, apparently with subcutaneous injection. Cub was pursued unsuccessfully by researchers on foot. Dogs were not released on partially sedated cub for safety reasons. This cub was member of family comprised of an adult female & 2 cubs, which was initially pursued on 12-02-06.
Female	cub	03-01-07	Dolores Canyon	Unmarked cub associated with puma F2; was probably her unmarked cub, sibling of M38. Cub climbed difficult spruce tree adjacent to creek.
Female, F23	adult	03-07-07	San Miguel River at Pinyon	Puma F23 climbed a cottonwood tree close to the San Miguel River. We did not attempt to anesthetize F23 to replace her non-functional GPS collar for safety reasons.

Table 5. Summary of puma capture efforts with ungulate road-kill baits, puma kills, and cage traps from August 2, 2006 to July 26, 2007, Uncompahgre Plateau, Colorado.[a]

Month	No. of Sites	Puma activity & capture effort results[b]
August	5	Puma scavenged a mule deer carcass on 08-07-06. Cage trap set. Black bear caught & released. Puma F16 was in the area. Puma did not return.
September	4	No puma activity detected.
October	10	Male puma scavenged a mule deer carcass 10-27-06. Cage trap set & monitored 10-27 to 28-06. Puma did not return.
November	12	Male puma scavenged a mule deer carcass on 11-06-06. Set & monitored cage trap 11-06 to 10-06. Puma F16 walked around cage trap, but did not enter on 11-09-06.
January	3	Subadult female F52 captured at adult female mule deer she killed 01-10-07, Ridgway State Park. Adult female F54 and her cub F53 were captured at an adult female mule deer kill 01-12-07, Pleasant Valley. Adult male puma M55 was captured at a mule deer fawn kill 01-21-07, Dallas Creek.
March	7	Adult male puma M29 was temporarily caught in cage trap set on an adult elk cow he had killed 03-15-07. But, M29 escaped out of back of the trap as researchers arrived. An ear-tagged male cub of puma F3 was observed feeding on a mule deer carcass 03-26-07; F3's family was in the vicinity. Female puma scavenged on a mule deer carcass 03-29-07. Cage trap was set. Puma F30 was recaptured, and her VHF collar was changed to a GPS collar.
April	7	Male puma, probably M29, scavenged a mule deer carcass ~04-01-07. Female puma walked by same carcass (as above) on ~04-02-07, but did not feed. During ~04-05 to 08-07 a puma completely scavenged the same mule deer carcass. Puma F3 and her cubs consumed a mule deer carcass 04-06 to 10-07. Puma F30 consumed a mule deer fawn carcass 04-08-07. Puma F30 consumed a mule deer carcass 04-24-07. Male puma scavenged on mule deer carcass 04-10-07. Cage trap set. Male puma M55 walked up to cage trap (GPS data), but did not enter. Pumas F30 & M55 fed on a mule deer carcass 04-17 to 20-07. Female puma scavenged a mule deer carcass 04-23-07. Cage trap set. Puma F8 was recaptured; her non-functional GPS collar was replaced with a VHF collar. Male puma M55 scavenged a mule deer carcass 04-30-07. Female puma scavenged a mule deer carcass. 04-27-07. Cage trap set. Puma F16 recaptured, and her GPS collar was changed with a new GPS collar.
May	6	Male puma M55 scavenged on a mule deer carcass 05-08-07. Female puma killed a mule deer doe 05-10-07. Cage trap set. Puma did not return or did not enter the trap. Male puma M55 scavenged a mule deer carcass 05-22-07.
June	5	Male puma M55 scavenged on an elk carcass 06-06-07.
July	4	No puma activity detected.

[a] We used 64 road-killed mule deer, 7 road-killed elk, 3 puma-killed mule deer, and 1 puma-killed elk at 26 different sites. Of the road-killed ungulate baits, 16 of 71 (22.5%) were scavenged by pumas.

[b] Eight pumas were captured, including: 2 adult males (M29, M55), 4 adult females (F54, F30, F8, F16), 1 subadult female (F52), and 1 female cub (F53).

Table 6. Pumas recaptured with dogs and cage traps, November 2006 to April 2007, Uncompahgre Plateau, Colorado.

Puma I.D.	Recapture date	Mass kg	Estimated Age (mo.)	Capture Method	Process
F3	11-21-06	Observed	63	Dogs	None
F3	11-22-06	41	63	Dogs	Changed GPS collar
M42	11-27-06	4.8	3.5	Dogs	Cub died due to infection & stress
M49	12-12-06	Observed	5	Dogs	None
M44	02-14-07	Observed	6	Dogs	None
M43	03-01-07	Observed	6.5	Dogs	None
M56	03-01-07	Observed	6.5	Dogs	None
F7	03-03-07	33	88	Dogs	Changed GPS collar
F23	03-07-07	Observed	31	Dogs	None
M29	03-05-07	Observed	91	Cage trap	None
F24	03-22-07	Observed	71	Dogs	None
M29	03-27-07	60	91	Dogs	Changed GPS collar
F30	03-29-07	37	44	Cage trap	Changed VHF collar to GPS collar
F8	04-23-07	37	46	Cage trap	Changed GPS collar
F16	04-28-07	48	51	Cage trap	Changed GPS collar

Table 7. Puma cubs sampled July 2006 to August 2007 on the Uncompahgre Plateau Puma Study area, Colorado.

Cub I.D.	Sex	Estimated birth date[a]	Estimated age at capture (days)	Mass (kg)	Mother	Estimated age of mother at birth of this litter (mo)
M38	M	July 29, 2006	41	2.9	F2	67
Unm.[b]	F	"	215	Observed	"	"
M39	M	August 13, 2006	29	1.9	F8	37
F40	F	"	"	1.8	"	"
F41	F	"	"	1.3	"	"
M42	M	"	"	1.5	"	"
M43	M	August 13, 2006	33	2.4	F7	82
M44	M	"	"	2.5	"	"
F45	F	"	"	1.7	"	"
M56[c]	M	"	185	9.6	"	"
M46	M	September 17, 2006	31	2.2	F3	61
M47	M	"	"	2.2	"	"
M48	M	"	"	2.5	"	"
M49[c]	M	July 1, 2006	158	10.0	F50	21
F53[c]	F	July 1, 2006	196	15.0	F54	24
F57	F	April 16, 2007	35	2.3	F25	94
M58	M	May 24, 2007	34	2.3	F16	52
F59	F	"	"	2.2	"	"
M60	M	"	"	2.0	"	"
M61	M	"	"	1.7	"	"
M62	M	July 14, 2007	34	1.8	F24	75
M63	M	"	"	2.1	"	"
M64	M	"	"	1.7	"	"
M65	M	"	"	1.9	"	"
F66	F	July 17, 2007	37	2.1	F30	48
M67	M	"	"	3.0	"	"
M68	M	"	"	3.3	"	"

[a] Estimated age of cubs sampled at nurseries is based on the starting date for GPS location foci for mothers at nurseries.
[b] This unmarked female cub was captured on 03-01-07 in association with adult female puma F2. This cub could be the sibling of cub M38, offspring of F2, which we were not able to capture previously with M38 (its tracks were observed).
[c] Estimated ages of M49 and F53 were based on morphometric comparisons with known-age cubs (Logan and Sweanor 2001, and unpublished data).

Table 8. Puma reproduction, Uncompahgre Plateau, Colorado, 2004-2007.

Consort pairs and estimated ages[a]				Dates pairs consorted[b]	Estimated birth date[c]	Estimated birth interval (mo.)	Estimated gestation	Observed number of cubs[d]
Female	Age (mo.)	Male	Age (mo.)					
F2	53				05/28/05			3
F2	67				07/29/06	14.0		2
F3	36				08/01/04			1
F3	50	M6	37	06/22-24/05	09/26/05	13.8	93-95	2
F3	62				09/17/06	11.7		3
F7	67				05/19/05			2
F7	82				08/13/06	14.9		4
F8	24				06/26/05			2
F8	37				08/13/06	13.4		4
F16	32				09/22/05			4
F16	52				05/24/07	19.9		4
F23	21				05/30/06			3
F24	75	M29	92	04/12-15/07	07/14/07		90-93	4
F25	74				08/01/05			1
F25	94				04/16/07	20.5		1
F28	36				06/09/06			2
F28	48	M29	88	12/27-29/06	03/30/07	11.7	92-93	≥2 tracks
F30	48	M55	34	04/16-20/07	07/17/07		88-92	3
F50	21				07/01/06			1
F54	24				07/01/06			1

[a] Ages of females were estimated at litter birth dates. Ages of males were estimated around the dates the pairs consorted.
[b] Consort pairs indicate pumas that were observed together based on GPS data.
[c] Estimated birth dates were indicated by GPS data of mothers at nurseries.
[d] Observed number of cubs do not represent litter sizes as some cubs were observed when they were 5 to 6 months old after postnatal mortality could have occurred in siblings. Only cub tracks were observed with F28.

Table 9. Summary for individual adult puma survival and mortality, December 2004 to July 2007, Uncompahgre Plateau, Colorado.

Puma I.D.	Monitoring span	No. days	Status: Alive/Lost contact/Dead; Cause of death
M1	12-08-04 to 08-16-06	616	Lost contact— failed GPS/VHF collar. M1 ranged principally north of the study area.
M4	01-28-05 to 12-28-05	333	Dead; killed by a male puma. Estimated age at death 37—45 months.
M5	08-01-06 to 07-31-07	365	Alive. Born on study area; offspring of F3. He was independent of F3 by 13 months old, and dispersed from his natal area at about 14 months old. Established adult territory on northwest slope of Uncompahgre Plateau at the age of 24 months.
M6	02-18-05 to 02-22-06	369	Lost contact— failed GPS/VHF collar.
M27	03-10-06 to 07-05-06	117	Lost contact— failed GPS/VHF collar. M27 ranged principally north of the study area.
M29	04-14-06 to 07-31-07	473	Alive.
M32	04-26-06 to 07-31-07	461	Alive.
M51	01-07-07 to 07-31-07	205	Alive.
M55	01-21-07 to 07-31-07	191	Alive.
F2	01-07-05 to 07-31-07	935	Alive.
F3	01-21-05 to 07-31-07	921	Alive.
F7	02-24-05 to 07-31-07	887	Alive.
F8	03-21-05 to 07-31-07	862	Alive.
F16	10-11-05 to 07-31-07	658	Alive.
F23	02-05-06 to 03-07-07	396	Lost contact— failed GPS/VHF collar.
F24	01-17-06 to 07-31-07	560	Alive.
F25	02-08-06 to 07-31-07	538	Alive.
F28	03-23-06 to 07-31-07	495	Alive.
F30	04-15-06 to 07-31-07	472	Alive.
F50	12-14-06 to 03-26-07	102	Died of natural causes; exact agent unknown.
F54	01-12-07 to 07-31-07	200	Alive.

Table 10. Summary of subadult puma survival and mortality, December 2004 to June 2006, Uncompahgre Plateau, Colorado.

Puma I.D.	Monitoring span	No. days	Status: Alive/Survived to adult stage/ Lost contact/Dead; Cause of death
M5	09-16-05 to 06-30-06	308	Alive; independent and dispersed from natal area at 13 months old. Established adult territory on northwest slope of Uncompahgre Plateau.
M11	06-21-06 to 12-14-06	176	Lost contact. Independent at 13 months old. Dispersed from natal area at 14 months old. Last location in Dolores River valley Dec. 14, 2006.
F23	01-04-06 to 02-04-06	31	Alive; survived to adult stage; gave birth to first litter at ~21 months old.
M31	04-19-06 to 04-26-06	7	Lost contact. Probable disperser. M31's estimated age at capture was 25 months, at the lower margin of puberty for puma. He may have been a dispersing subadult, and could have moved away from the study area.
M49	03-26-07 to 07-31-07	127	M49 was orphaned at about 9 months old, when his mother F50 died of natural causes. He dispersed from his natal area at about 10 months old and has been ranging on the northeast slope of the Uncompahgre Plateau.
F52	01-10-07 to 05-15-07	125	Lost contact. Dispersed from study area as a subadult. F52's last location was Crystal Creek, a tributary of the Gunnison River east of the Black Canyon.

Table 11. Summary for individual puma cub survival and mortality, December 2004 to 2007, Uncompahgre Plateau, Colorado.

Puma I.D.	Estimated Age at capture (days)	Estimated survival span from 1st capture to fate or last monitor date	Age to last monitor date alive or at death (days)	Status: Alive/Survived to subadult stage/ Lost contact/Disappeared/ Dead; Cause of death	Mother I.D.
M5	183	02-04-05 to 07-31-07	907	Survived to subadult stage by 09-16-05; independent at ~13 mo. old. Dispersed from natal area by 09-29-05 at 14 mo. old .	F3
F9	31	06-27-05 to 4-19-06	329	Lost contact— shed radiocollar 04-19-06—04-26-06.	F2
F10	31	06-27-05 to 11-20-05—12-29-05	207-246	Lost contact— shed radiocollar 08-10-05; last tracks of F10 with mother F2 & siblings F9 & M11 observed 11-20-05. F10 disappeared by 12-30-05.	F2
M11	31	06-27-05 to 12-14-06	535	Survived to subadult stage by 06-21-06, independent at 13 mo. old. Dispersed from natal area by 07-11-06 at 14 mo. old.	F2
F12	42	07-01-05 to 12-08-05—01-26-06	245-294	Lost contact— shed radiocollar 07-28-05—08-01-05. Tracks of F12 found in association with mother F7 on 12-08-05. F12 disappeared by 01-27-06 when she was not visually observed with F7, and her tracks were not seen in association with F7's tracks.	F7
F13	42	07-01-05 to 08-28-05	100	Dead; killed and eaten by a puma (sex unspecified).	F7
F14	26	07-22-05 to 02-07-06—03-10-06	226-257	Lost contact— shed radiocollar 01-20-06—01-25-06. Tracks of F14 were observed with tracks of mother F8 & sibling M15 on 02-07-06. Disappeared by 03-11-06, only tracks of F8 & M15 were found.	F8
M15	26	07-22-05 to 06-06-06	345	Lost contact— shed radiocollar 06-06-06—06-14-06.	F8
F17	34	10-26-05 to 08-18-06	330	Dead. Lost contact— shed radiocollar 06-06-06—06-14-06. Killed by a car on highway 550 on 08-18-06. Probably dependent on F16.	F16
F18	34	10-26-05 to 07-20—27-06	301-308	Dead; probably killed by another puma. Multiple bite wounds to skull. 10 mo. old. Born 9/22/05	F16
M19	34	10-26-05 to 07-27-06	306	Lost contact— shed radiocollar 07-27-06—08-02-06.	F16
M20	34	10-26-05 to 05-24-06	244	Lost contact— shed radiocollar 05-24-06—05-25-06.	F16
F21	37	11-02-05 to 06-30-06	277	Alive.	F3
M22	37	11-02-05 to 12-21-05—12-22-05	86-87	Dead; killed and eaten by male puma 12-21-05—12-22-05.	F3
M26	183	02-08-06 to 03-21-06	224	Lost contact— shed radiocollar 03-21-06—03-24-06.	F25

Puma I.D.	Estimated Age at capture (days)	Estimated survival span from 1st capture to fate or last monitor date	Age to last monitor date alive or at death (days)	Status: Alive/Survived to subadult stage/ Lost contact/Disappeared/ Dead; Cause of death	Mother I.D.
F33	31	06-30-06 to 07-31-06	62	Dead. Probably killed and eaten by a male puma 08-01 to 03-06. GPS data on M29 indicate he was *not* involved.	F23
F34	31	06-30-06 to 07-31-06	62	Dead. Probably killed and eaten by a male puma 08-01 to 03-06. GPS data on M29 indicate he was *not* involved.	F23
F35	31	06-30-06 to 07-07-06	38	Dead; research-related fatality.[a]	F23
F36	29	07-08-06 to 07-28-06	74	Dead. Killed and eaten by a male puma 08-22-06. GPS data on M29 indicate he was *not* involved.	F28
M37	29	07-08-06 to 07-28-06	74	Dead. Killed and eaten by a male puma 08-22-06. GPS data on M29 indicate he was *not* involved.	F28
M38	41	09-08-06 to 02-20-07	165	Lost contact— shed radiocollar found 03-06-07.	F2
M39	29	09-11-06 to 09-20-06 to 04-25-07	9 226	Lost contact— shed radiocollar by 09-20-06, but seen alive on that date. Tracks of 2 cubs following F8 on 04-25-07.	F8
M40	29	09-11-06 to 09-20-06 to 04-25-07	9 226	Lost contact— shed radiocollar by 09-20-06, but seen alive on that date. Tracks of 2 cubs following F8 on 04-25-07.	F8
F41	29	09-11-06 to 10-05-06	24	Lost Contact— shed radiocollar or died (blood on collar) between 10-05-06 (last live signal) & 10-13-06 (collar found).	F8
M42	29	09-11-06 to 11-27-06	77	Dead; research-related fatality.[b]	F8
M43	33	09-15-06 to 03-01-07	167	Treed, visually observed 03-01-07.	F7
M44	33	09-15-06 to 02-14-07	152	Treed, visually observed 02-14-07; sibling (?) M56 also captured, sampled, & marked for 1st time.	F7
F45	33	09-15-06 to 5-20 to 23-07	280-283	Dead. Multiple puncture wounds on braincase— parietal & occipital regions; consistent with bites from coyote. F45 switched families, moving from F7 to F2 about 12-19 to 20-06. Last date F45 was with F2 was 04-17-07.	F7
M46	31	10-18-06 to 12-15-06	58	Lost contact— shed radiocollar. Tracks of all cubs observed following F3 12-15-06.	F3
M47	31	10-18-06 to 12-15-06	58	Lost contact— shed radiocollar. Tracks of all cubs observed following F3 12-15-06.	F3
M48	31	10-18-06 to 12-15-06	58	Lost contact— shed radiocollar. Tracks of all cubs observed following F3 12-15-06.	F3

Puma I.D.	Estimated Age at capture (days)	Estimated survival span from 1st capture to fate or last monitor date	Age to last monitor date alive or at death (days)	Status: Alive/Survived to subadult stage/ Lost contact/Disappeared/ Dead; Cause of death	Mother I.D.
M49	153	12-05-06 to 07-31-07	238	M49 was orphaned when his mother died on about 03-26-07.	F50
F53	183	01-12-07 to 02-23-07	42	Lost contact— shed radiocollar 2-23-07.	F54
M56[c]	183	02-14-07 to03-01-07	15	Lost contact— shed radiocollar 2-27-07. M56 observed 03-01-07.	F7 (?)
F57	35	05-21-07 to 06-06-07	16	Lost contact— shed radiocollar 06-07-07. Live mode 06-06-07.	F25
M58	34	06-27-07		Not radio-collared.	F16
F59	34	06-27-07 to 08-21-07	55	Alive.	F16
M60	34	06-27-07 to 07-11-07	14	Dead; research-related mortality.[d]	F16
F61	34	06-27-07 to 06-29-07	2	Radiocollar malfunction.	F16
M62	34	08-17-07		Not radio-collared.	F24
M63	34	08-17-07		Not radio-collared.	F24
M64	34	08-17-07		Not radio-collared.	F24
M65	34	08-17-07		Not radio-collared.	F24
F66	37	08-23-07		Radio-collared.	F30
M67	37	08-23-07		Not radio-collared.	F30
M68	37	08-23-07		Not radio-collared.	F30

[a] Cub F35 probably starved between 06-30-06 & 07-07-06 after the transmitter on the expandable collar got in its mouth.
[b] Cub M42 died after being captured by dogs, probably from stress of capture associated with severe infection of laceration under right foreleg caused by expandable radiocollar.
[c] Cub M56 was captured in association with F7 and her cubs M43 and M44. He may have been missed at the nursery when M43 and M44 were initially sampled and marked.
[d] Cub M60 died probably of starvation. The expandable radiocollar was around the neck and right shoulder, possibly restricting movement.

Table 12. Numbers of GPS locations for pumas captured on the Uncompahgre Plateau, Colorado, December 2004 to July 2007.

Puma I.D.	Sex	Age stage	Dates monitored [a]	No. locations	Acquisition rate average, range, *n*[b]
M1	M	adult	12-08-04 to 07-20-06	1,864	76, 69—84, 14
M4	M	adult	01-28-05 to 12-28-05	910	70, 57—84, 10
M6	M	adult	02-18-05 to 11-23-05	926	84, 73—93, 9
M27	M	adult	03-11-06 to 06-21-06	316	77, 67—84, 3
M29	M	adult	04-14-06 to 07-30-07	1,165	69, 56—81,13
M51	M	adult	01-07-07 to 07-30-07	630	76, 66—87, 6
M55	M	adult	01-21-07 to 07-22-07	558	79, 68—91, 6
F2	F	adult	01-07-05 to 07-11-07	2,759	75, 43—91, 30
F3	F	adult	01-21-05 to 07-30-07	2,474	78, 55—90, 24
F7	F	adult	02-24-05 to 07-30-07	2,401	68, 26—92, 27
F8	F	adult	03-21-05 to 10-04-06	1,516	67, 41—81, 17
F16	F	adult	10-12-05 to 06-12-07	1,797	73, 41—90, 23
F23	F	subadult,	01-04-06 to 02-04-06	113	79, 45—92, 6
		adult	02-05-06 to 07-17-06	511	
F24	F	adult	01-17-06 to 07-25-07	1,816	82, 65—93, 18
F25	F	adult	02-09-06 to 07-02-07	1,408	69, 55—87, 16
F28	F	adult	03-24-06 to 07-11-07	1,394	74, 53—89, 16
F30	F	adult	03-30-07 to 07-25-07	381	83, 58—94, 4
F50	F	adult	12-14-06 to 03-26-07	361	87, 76—94, 4
F52	F	subadult	01-10-07 to 05-08-07	383	83, 70—92, 3
F54	F	adult	01-12-07 to 07-20-07	615	82, 77—86, 6

[a] GPS collars on pumas are remotely downloaded at approximately 1-month intervals. The last date in *Dates monitored* includes last location from the last GPS data download for an individual puma in this report.

[b] *n* = number of remote downloads.

Table 13. Estimated use areas of GPS-collared pumas during November through March, Uncompahgre Plateau, Colorado.[a]

Puma I.D.	No. locations	Time span	No. months	95% Fixed kernel (km^2)	50% Fixed kernel (km^2)	100% Minimum convex polygon (km^2)
F2	151	11-01-06 to 03-31-07	5	78.6	13.3	102.3
F3	130	11-22-06 to 03-31-07	4.3	138.9	12.2	164.0
F7	114	11-01-06 to 03-31-07	3.9[b]	66.7	10.6	66.8
F8	147	11-01-05 to 03-31-06	5	33.7	5.4	43.3
F16	144	11-01-06 to 03-31-07	5	53.6	7.0	59.9
F24	150	11-01-06 to 03-31-07	5	117.7	18.9	148.9
F25	147	11-01-06 to 03-31-07	5	52.0	6.5	79.8
F28	146	11-01-06 to 03-31-07	5	61.8	6.3	105.4
F50	103	12-14-06 to 03-26-07	3.4	70.0	15.8	91.2
M1	149	11-01-05 to 03-31-06	5	1,132.7	302.3	779.8
M29	97	11-01-06 to 03-31-07	3.4[c]	349.1	25.0	379.0
M51	85	01-07-07 to 03-31-07	2.8	231.0	31.0	281.2

[a] Use areas were estimated by using the Animal Movement extension in ArcView 3.2. One location per day was randomly chosen from up to 4 locations fixed per day per puma to reduce autocorrelation.

[b] Due to GPS collar failure, GPS locations were not fixed for F7 from 01-30 to 03-02-07.

C Due to GPS collar failure, GPS locations were not fixed for M29 from 02-09 to 03-26-07.

Table 14. VHF-radio-collared independent pumas on the Uncompahgre Plateau, Colorado, 2007.

Puma I.D.	Sex	Age stage	Dates monitored	No. locations
M5	M	Subadult	09-16-05 to 07-31-06	36
		Adult	08-01-06 to 07-30-07	37
F8	F	Adult	04-23-07 to 07-30-07	14
F30	F	Adult	04-15-06 to 03-29-07	43
M31	M	Subadult	04-09-06 to 04-26-06	2
M32	M	Adult	04-26-06 to 07-30-07	50

Table 15. Summary of puma mother and cub associations by distance (m) during airplane flights, November through March each winter.

Monitoring period	Month	No. flights	No. puma families[a]	Ages of cubs (mo.)	No. observations with mothers & cubs ≤520 m apart	No. observations with mothers & cubs >520 m apart
Nov. 9, 2005 to March 29, 2006	Nov.	3	4	2—6	10	2
	Dec.	4	4	3—7	16	4
	Jan.	5	4	4—8	16	4
	Feb.	4	5	5—9	16	2
	Mar.	2	5	6—10	9	0
	Totals	18	4—5	2—10	67	12[b]
Nov. 7, 2006 to March 22, 2007	Nov.	4	4	2—3	10	1
	Dec.	4	4	2—5	11	1
	Jan.	5	3	4—6	9	3
	Feb.	4	4	5—7	9	2
	Mar.	3	1	8	2	1
	Totals	20	1—4	2—8	41	8[c]

[a] All puma mothers wore GPS-radiocollars. At least 1 cub in the litter wore a VHF radiocollar.
[b] Mean = 1,060 m, *SD* = 325.99, range = 650—1,600.
[C] Mean = 1,120 m, *SD* = 1,214.40, range = 616—4,101.

Table 16. General results of puma GPS cluster investigations pilot project, October 2006 to April 2007, Uncompahgre Plateau, Colorado.

Cluster Types Investigated	No.	Animals found at all clusters	No.	Animals found at random clusters	No.
S1 Non-random	63	Mule deer	63	Mule deer	33
S1 Random	84	Elk	58	Elk	31
S2 Random	11	Beaver	1	Coyote	2
S3 Random	29	Coyote	2	Total	66
S4 Random	30	Total	124		
S5 Random	40				
Totals	257				

Table 17. Sex and age classes of mule deer found at puma GPS cluster investigations, October 2006 to April 2007, Uncompahgre Plateau, Colorado.

Sex & age of mule deer	**All clusters**			**Random clusters**		
	Female	**Male**	**Unknown**	**Female**	**Male**	**Unknown**
Fawn	0	1	18	0	1	10
Yearling	1	6	3	1	3	2
2+ year	6	6	1	5	1	1
Unknown age 1+ yr.	1	1	6	1	1	4
Unknown age	0	0	13	0	0	3
Totals	8	14	41	7	6	20

Table 18. Sex and age classes of elk found at puma GPS cluster investigations, October 2006 to April 2007, Uncompahgre Plateau, Colorado.

Sex & age of elk	**All clusters**			**Random clusters**		
	Female	**Male**	**Unknown**	**Female**	**Male**	**Unknown**
Calf	3	1	18	1	0	10
Yearling	13	2	3	7	1	3
2+ year	4	3	4	1	2	3
Unknown age 1+ yr.	0	0	4	0	0	2
Unknown age	0	0	3	0	0	1
Totals	20	6	32	9	3	19

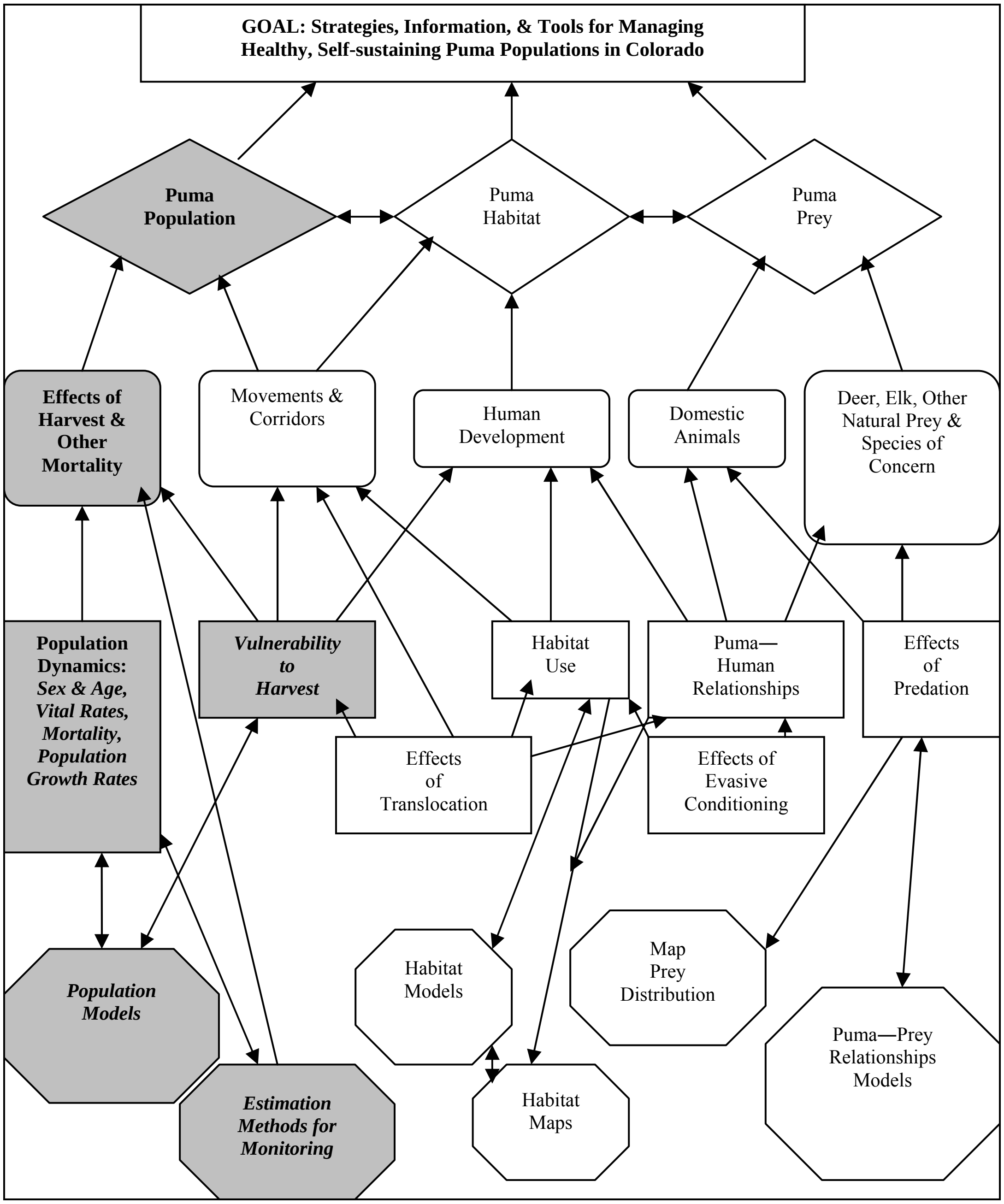

Figure. 1. An ecologically-based conceptual model of the Colorado Puma Research Program that provides the contextual framework for this and proposed puma research in Colorado. Gray-shaded shapes identify areas of research addressed by this report for the puma management goal (at top).

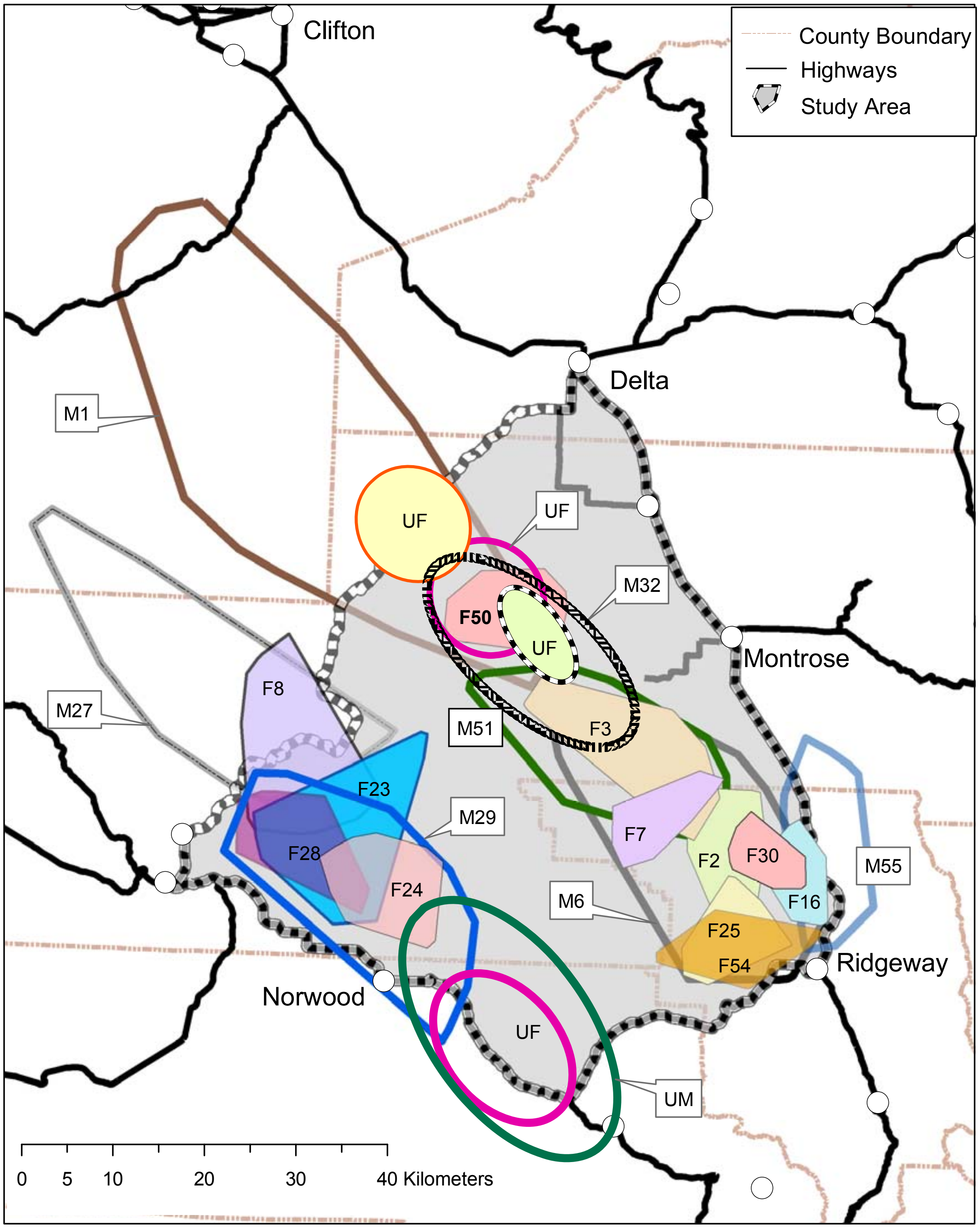

Figure 2. Schematic of home ranges of GPS-collared (polygons) and non-collared (ellipses) independent pumas (adults and subadults), intended to show the minimum count and location of independent pumas detected on the study area during November to May period, 2006-2007, Uncompahgre Plateau, Colorado. M & F signify male & female, followed by the identification number of the puma. UF and UM signify uncollared and unsampled female and male pumas, respectively.

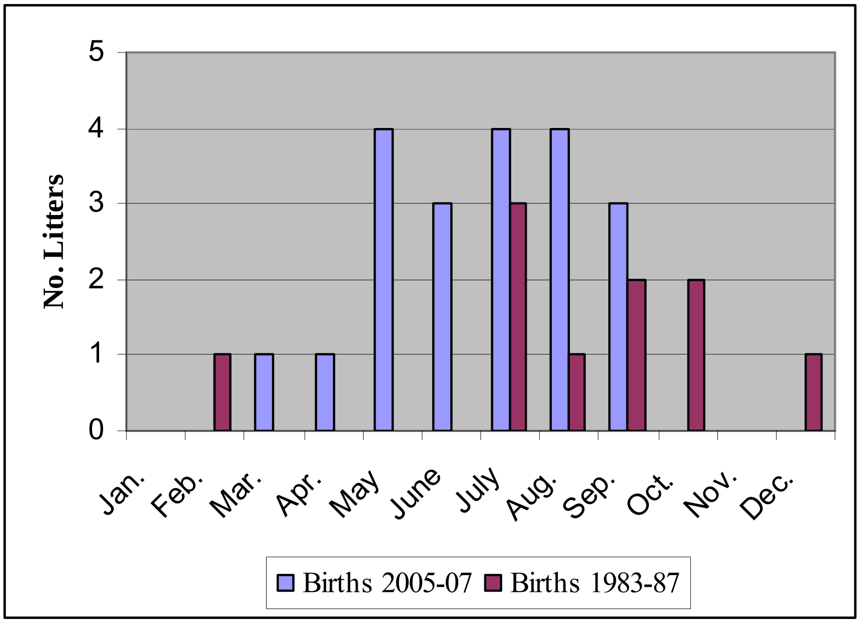

Figure 3. Puma births (n = 20 litters) detected by month during the current research effort, 2005 to 2007, and during the earlier effort by Anderson et al. (1992), 1983 to 1987 (n = 10 litters).

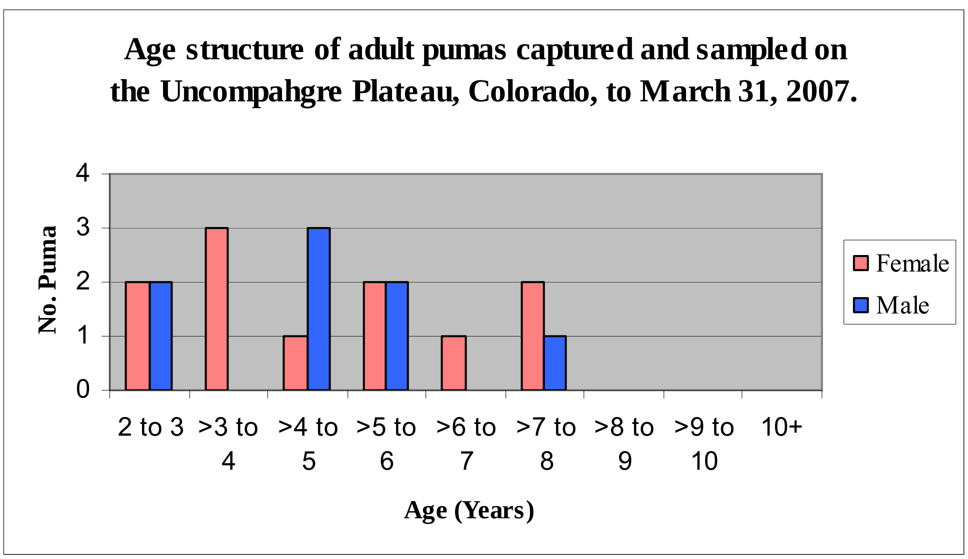

Figure 4. Age structure of adult pumas captured and sampled on the Uncompahgre Plateau, Colorado, on March 31, 2007, after 3 winters (Nov. through Mar., 2004-05 to 2006-07) of protection from sport-hunting mortality. In addition, no other human-caused mortalities have been documented in the GPS/radio-collared sample of adults. This age structure assumes that puma M1, M6, and M27 (which had non-functional GPS collars) were alive. Evidence was found on the ground that indicated that all 3 of those males were alive. Pumas M1, M5, and M27 range north of the study area and were protected from legal sport-harvest. Mean ± *SD* of adult female and adult male ages, respectively: 4.90 ± 1.80 yr. (58.82 ± 21.62 mo.), 4.76 ± 1.62 yr. (57.12 ± 19.39 mo.).

APPENDIX I
COLLABORATIVE PROJECT ON DISEASE SURVEILLANCE IN WILD FELIDS.

College of Veterinary Medicine and Biomedical Sciences
Department of Microbiology, Immunology & Pathology
1619 Campus Delivery
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970-491-6144 (voice)
970-491-0603 (fax)

TO: Ken Logan, Mammals Researcher, Colorado Division of Wildlife, Montrose, CO.
FROM: Sue VandeWoude, DVM, Associate Professor, DMIP
RE: Disease Seroprevalence in UP Pumas
DATE: August 26, 2007

Attached please find the consolidated report on infectious disease surveillance for the mountain lion samples you have provided to our laboratory as an adjunct to your CDOW ongoing studies. Our laboratory has performed puma-lentivirus (PLV) antibody screening using a sensitive western blot assay developed in our laboratory and found 13 of 18 samples conclusively positive (72%), with two additional samples inconclusive and one not tested. Dr. Michael Lappin, a veterinary internal medicine specialist with expertise in feline infectious disease has tested a subset of 6 samples for antibodies to Feline Calicivirus (FCV), Feline Herpes Virus (FHV), Feline parvovirus (FPV), *Toxoplasma gondii* (IgM, indicating recent infection, IgG indicating past exposure), and *Bartonella hensalae* (the agent associated with cat scratch disease). At least one of six animals tested has been positive for each of these agents. Further results are pending from the remaining samples you have provided for these 5 assays. In addition, Dr. Martin Scriefer at Fort Collins CDC has also tested 6 animals for evidence of antibodies to the agent responsible for plague (*Yersinia pestis*). Interestingly, 3 of 6 animals demonstrate significant exposure to this agent as well. These specific agents were selected for analysis in order to provide a variety of types of agents (viruses: PLV, FCV, FHV, FPV; bacteria: *Bartonella henselae* and *Yersinia pestis*; and coccidian: *T. gondii*), a variety of modes of transmission (direct intra-specific contact, PLV; direct contact with domestic cats, FCV, FHV, FPV; arthropod transmission, *B. henselae, Y. pestis*; prey ingestion, *T. gondii, Y. pestis*). Further, at least three of these agents (PLV, FCV, B. henselae) result in chronic infections, allowing the possibility of determining genetic relatedness among organisms isolated from different individuals, and three of these agents (*B. henselae, Y. pestis, T. gondii*) are also potential zoonotic agents. As you are aware, our laboratory has recently been awarded a 5 year NSF Ecology of Infectious Disease grant entitled, “The effects of urban fragmentation and landscape connectivity on disease prevalence and transmission in North American felids”, with co-PI Dr. Kevin Crooks, an associate professor in the Warner College of Natural Resources at CSU. The aims of this grant are to model the effects of urbanization and resultant habitat fragmentation on disease dynamics in large carnivore species as described on the following page. The letter of support provided by you and Dave Freddy were pivotal in demonstrating a large cohort of capable and active field collaborators willing to provide samples to support our studies. The mountain lion field work being led by your team, and the newly initiated studies by your colleague, Dr. Mat Alldredge, have provided us with renewed enthusiasm for developing our collaborations to support the goals of our study. We foresee the opportunity to interact in a mutually beneficial partnership to further the goals of all of our studies, and to maximize the information that can be gleaned about these important and ecologically significant species. We anticipate that the data we are generating will be useful for comparative seroprevalence of different geographic populations of bobcats and pumas, and for genetic phenotyping of pathogens to compare relationships among diseases spread by arthropod vectors, domestic cats, feral rodents, and inter-specific contacts. As we discussed during your recent visit to CSU, these samples are most valuable to us if we can receive them directly as quickly as possible after collection. I have provided an SOP providing information about the types of samples that will be most valuable, and a draft of a 'permissions' document that you can use with each sample submission to provide us with guidance for any testing that is permissible on the materials we receive. This latter document will be filed and recorded electronically. We will continue to provide annual updates and communications about any publications that utilize the data resulting from your samples. Again thank you for providing these extremely valuable samples, and we look forward to our continued collaborations.

Sincerely,
Sue VandeWoude

THE EFFECTS OF URBAN FRAGMENTATION AND LANDSCAPE CONNECTIVITY ON DISEASE PREVALENCE AND TRANSMISSION IN NORTH AMERICAN FELIDS

PROJECT SUMMARY

Sue VandeWoude (co-PI), Kevin Crooks (co-PI), Michael Lappin, Mo Salman, Walter Boyce, Ken Logan, Mat Alldredge, Carolyn Krumm, Don Hunter, Lisa Lyren, Seth Riley, Jennifer Troyer

The objective of this study is to model the effects of urbanization and resultant habitat fragmentation on disease dynamics in large carnivore species--ecologically pivotal organisms that are sensitive to human disturbances. Bobcats, puma, and domestic cats will be evaluated simultaneously in three divergent ecosystems: high mountain desert (Colorado), everglades (Florida), and Mediterranean scrub habitat (California). The research will: 1) assess the relationship between habitat fragmentation and prevalence of viral, bacterial, and parasitic pathogens across a gradient of urbanization, 2) use transmission dynamics of selected disease agents as markers of connectivity of fragmented populations, and 3) evaluate the effect of urbanization on the incidence of cross-species disease transmission. The results of this research will give wildlife managers a better understanding of how urbanization affects their local wildlife and assist them in future disease management planning. The combination of a uniquely qualified, broadly based research team with an extensive dataset on large carnivores from across the country presents an unprecedented opportunity to investigate the disease dynamics in these rare and difficult to study species. The research efforts of each regional team will support and provide new insights for all of the regions involved, not simply their own. Training of graduate students in ecology, infectious disease, and epidemiology will be emphasized, as will training for pre- and post-doctoral veterinarians. Results will be made widely available to other scientists, conservation practitioners, and the general public. This research has a tremendous capacity to broadly impact areas of public and post-graduate education, career development for new investigators and persons from underrepresented groups, and to enhance understanding of complex infectious disease ecological problems using extensive multi-disciplinary collaborations.

Table 1. Appendix I. Preliminary results of infectious disease surveillance for puma, Uncompahgre Plateau, Colorado.

Puma ID	Sex	Capture Date	GPS NAD27 U.T.M.: Zone, E, N	PLV[a]	FCV[b]	FHV[c]	FPV[d]	T.g. [e] IgM	T.g.[e] IgG	B.h. [f]	Y.p. [g]
UPCO 3	F	1/21/2005	13S, 241606, 4251510	-	+[h]	+	+	-	+	-	++
UPCO 7	F	2/24/2005	13S, 246328, 4244230	+	+	-	-	-	+	-	+++
UPCO 7	F	3/30/2006	13S, 245901, 4247627	+	P[h]	P	P	P	P	P	P
UPCO 7	F	3/3/2007	13S, 247645, 4246097	I[h]	P	P	P	P	P	P	P
UPCO 8	F	3/21/2005	12S, 727808, 4239029	I	-	-	-	-	+	-	++
UPCO 4	M	1/28/2005	13S, 257565, 4239606	+	-	-	-	-	+	+	I
UPCO 5	M	2/4/2005	13S, 240577, 4251037	-	-	+	+	-	+	-	I
UPCO 6	M	2/18/2005	13S, 247399, 4254006	+	-	-	-	-	+	-	I
UPCO 25	F	2/8/2006	13S, 258374, 4230480	+	P	P	P	P	P	P	P
UPCO 28	F	3/23/2006	12S, 722868, 4240115	+	P	P	P	P	P	P	P
UPCO 29	M	4/14/2006	12S, 723458, 4242340	+	P	P	P	P	P	P	P
UPCO 31	M	4/19/2006	12S, 746919, 4225441	+	P	P	P	P	P	P	P
UPCO 23	F	1/4/2006	12S, 730188, 4234861	-	P	P	P	P	P	P	P
UPCO 27	M	3/10/2006	12S, 722339, 4245212	-	P	P	P	P	P	P	P
UPCO 30	F	4/15/2006	13S, 248551, 4242095	-	P	P	P	P	P	P	P
UPCO 50	F	12/14/2006	12S, 753639, 4260149	+	P	P	P	P	P	P	P
UPCO 51	M	1/7/2007	13S, 238783, 4252390	+	P	P	P	P	P	P	P
UPCO 52	F	1/10/2007	13S, 258058, 4236260	I	P	P	P	P	P	P	P
UPCO 54	F	1/12/2007	13S, 252688, 4228050	+	P	P	P	P	P	P	P
UPCO 55	M	1/21/2007	13S, 258133, 4228691	+	P	P	P	P	P	P	P
UPCO 24	F	1/17/2006	12S, 737151, 4233273	+	P	P	P	P	P	P	P
			% Seroprevalance = No. animals positive/Total animals tested * 100	72	33	33	33	0	100	17	50

[a] PLV is Puma Lentivirus.
[b] FCV is Feline Calicivirus.
[c] FHV is Feline Herpesvirus.
[d] FPV is Feline Panleukopenia Virus
[e] T. g. is *Toxoplasma gondii*.
[f] B. h. is *Bartonella hensalae*.
[g] Y. p. is *Yersinia pestis*.
[h] Results: + (positive result), P (Pending result), I (Inconclusive result).

Colorado Division of Wildlife
July 2006 - June 2007

WILDLIFE RESEARCH REPORT

State of	Colorado	: Division of Wildlife
Cost Center	3430	: Mammals Research
Work Package	3003	: Predatory Mammals Conservation
Task No.	2	: Cougar Demographics and Human Interactions Along the Urban-Exurban Front Range of Colorado
Federal Aid Project:	N/A	:

Period Covered: July 1, 2006 - June 30, 2007

Author: M.W. Alldredge

Personnel: K. Griffin, D. Kilpatrick, M. Miller, F. Quartarone, M. Sirochman, L. Wolfe, D. Freddy CDOW; B. Posthumus, Jeffco Open Space; S. Oyler-McCance, USGS.

All information in this report is preliminary and subject to further evaluation. Information MAY NOT BE PUBLISHED OR QUOTED without permission of the author. Manipulation of these data beyond that contained in this report is discouraged.

ABSTRACT

Projects were undertaken to evaluate genetic techniques and examine statewide genetic population structure for cougars and bears. Genetic techniques were developed for both cougars and bears for both microsatellite and mitochondrial DNA. Using 50 samples each from cougars and bears, an initial attempt was made to determine genetic population structure for the state. Based on these data, the concept of a megapopulation is a more realistic representation of the cougar and bear populations, as opposed to ideas of subpopulations within the state. Further investigation is warranted to increase the power of these tests and to make management recommendations.

In addition to genetics, a pilot field study was initiated to determine the feasibility of conducting a long-term front-range cougar-human interaction study. With considerable time and effort, research agreements were achieved with Jefferson County, Boulder County, and Boulder City open-space agencies. Internal CDOW protocols were also developed that outlined the working relationship between research and management personnel in regards to dealing with various levels of cougar-human conflict. During this year, 2 cougars were caught and one adult male was fitted with a GPS transmitter. The GPS transmitter is working adequately, with an approximate 56% acquisition success rate. Given this success rate we will keep the daily number of acquisitions high, currently set at 8 per day. Based on the inter-agency agreements, trapping operations, and GPS data, we recommend proceeding with the full scale cougar-human interaction study on the front-range of Colorado.

WILDLIFE RESEARCH REPORT

COUGAR DEMOGRAPHICS AND HUMAN INTERACTIONS ALONG THE URBAN-EXURBAN FRONT RANGE OF COLORADO

MATHEW W. ALLDREDGE

P.N. OBJECTIVE

1. To assess cougar (*Puma concolor*) population demographic rates, movements, habitat use, prey selectivity and human interactions along the urban-exurban front-range of Colorado.
2. Develop methods for delineating population structure of cougars and black bears (*Ursus americanus*) and estimating population densities of cougars for the state of Colorado.

SEGMENT OBJECTIVES

1. Determine the efficacy of using microsatellites or mtDNA to delineate female cougar and black bear subpopulations across the state of Colorado.
2. Evaluate differences in DNA quantity from either a cougar scat surface collection or a cross-sectional collection.
3. Evaluate differences in DNA quantity from successive cougar feces depositions to determine the variation in quantities of genetic material in scats. Quantify differences in epithelial shedding rates.
4. Evaluate temporal, environmental, and seasonal effects on cougar DNA quantity and quality for both controlled and uncontrolled conditions.
5. Determine the effectiveness of cage traps and hounds for capturing cougars on the Front-Range of Colorado.
6. Determine functionality and suitability of GPS collars on cougars in Front-Range habitats.
7. Implement cougar-human risk protocols and communications within CDOW and among public entities and determine if modifications are necessary.
8. Determine the feasibility of aversive conditioning techniques on cougars within urban/exurban areas, including use of hounds and shotgun-fired bean bags or rubber bullets.
9. Evaluate political/social response to cougar research activities.

INTRODUCTION

Cougar management is a growing concern for the Colorado Division of Wildlife (CDOW). Cougar conservation and the maintenance of viable populations is a statewide issue as CDOW is charged with the management of cougars. However, the nexus between cougar conservation and human health and safety is becoming a high priority issue within the urban and exurban areas of the state. Cougar conflicts (livestock depredation, pet depredation, and direct human interaction) within urban and exurban areas appear to be increasing as humans continue to encroach on historical cougar habitats. Because of the diversity of cougar management issues across the state, cougar research has focused on statewide issues of cougar population structure and methods of estimating population demographics, and on urban/exurban issues of cougar/human interaction.

Genetic techniques for monitoring or research of rare, elusive, and wide ranging species are of particular interest as other techniques are either impractical or financially prohibitive. Genetic techniques for monitoring and research of cougars in Colorado may be invaluable as alternative techniques are expensive and in many situations may not be possible. Capture and handling of cougars is expensive, time consuming, and may not give representative samples of the population. Large dispersal distances of cougars, especially males, will require impractically large study areas in order to understand demographic patterns that are affected by immigration. Capture may not even be possible in suburban and exurban areas of Colorado as logistical constraints associated with private land owners will likely prohibit the use of many capture techniques.

Noninvasive genetic sampling (Hoss et al. 1992, Taberlet and Bouvet 1992) has the potential to provide a realistic method of sampling a population of interest. Noninvasive sampling techniques include the use of hair snares, and scat collections (Harrison et al. 2004, Smith et al. 2005). The use of scats for sampling cougar populations may be particularly useful and provide a representative sample of the population. Scat collections can either be done by searching transects with human observers (Harrison et al. 2004) or with trained dogs (Smith et al. 2005). Scats could also be collected from kill sites. Kill sites would probably need to be based on mortalities of radio-collared ungulate populations. Data from noninvasive sampling techniques are useful in describing dispersal patterns and estimating population size. Noninvasive genetic data are error prone, which in many cases is because of the quantity and quality of genetic material relative to the collection of noninvasive samples. Therefore, one objective over the last year has been to develop a study to evaluate degradation rates of DNA in fecal samples with respect to time and temperature.

Use of genetic data for other purposes, such as delineating subpopulations, is also very useful for managing cougar and bear populations in the state. In these cases the goal is to examine local characteristics of the genetic data and determine if it is different among areas (subpopulation structure) or is similar across all areas (panmictic population). Nuclear DNA is inherited from both the mother and the father and therefore is less likely to describe sex-linked population structure. Male cougars and bears generally disperse over greater distances than females and therefore a female population substructure may be easier to detect than male population substructure. Examination of cougar and bear population structure has been examined using nuclear DNA but few studies have examined cougar population structure using mitochondrial DNA (mtDNA). Mitochondrial DNA is only inherited from the female in mammals and therefore lends itself to delineating female cougar population substructures. A second objective has been to determine if any genetic population structure can be identified for cougars and bears across the state by examining nuclear and mtDNA from statewide female cougar and bear harvest.

At the local scale efforts have been made to initiate a cougar/human interaction study on the Front-Range of Colorado. Given that cougars currently coexist with humans within urban/exurban areas along Colorado's Front-Range, varying levels of cougar-human interaction are inevitable. The CDOW is charged with the management of cougar, with management options ranging from minimal cougar population management, to dealing only with direct cougar-human incidents, to attempted extermination of cougars along the human/cougar spatial interface. Inaction and extermination are management starategies that are not practical nor acceptable to the majority of the human population. In the 2005 survey of public opinions and perceptions of cougar issues, 96% of the respondents agreed that it was important to know cougars exist in Colorado, and 93% thought it was important that they exist for future generations (CDOW, unpublished data).

There is a growing voice from the public that CDOW do more to mitigate potential conflicts, and the Director of CDOW has requested that research efforts be conducted to help minimize future human/cougar conflicts. In order to meet these goals CDOW believes we need to directly test management prescriptions in terms of desired cougar population and individual levels of responses.

Long-term study objectives for the Front-Range Cougar Research project will involve directly testing management responses of cougars at various levels of human interaction, as well as collecting basic information about demographics, movement, habitat use, and prey selection. The CMGWG (2005) recommend that part of determining the level of interaction or risk between cougars and humans is to evaluate cougar behavior on a spectrum from natural, to habituated, to overly familiar, to nuisance, to dangerous. The CMGWG (2005) clearly state that there is no scientific evidence to indicate that cougar habituation to humans affects the risk of attack.

Studying individual and population level responses of cougars will require capturing and radio-collaring cougars, as well as standardizing responses of CDOW personnel to human/cougar interactions. Therefore, in this initial year, we need to test various cougar capture techniques in urban/exurban areas of interest for effectiveness and public acceptance and to assess the reliability of GPS collars as monitoring tools to assess cougar responses to management prescriptions. More importantly, clearly defined protocols have been developed within CDOW (APPENDIX II-B, sub-appendix I) to direct how researchers and field managers should deal with various levels of risk to human health and safety, and these protocols need to be tested and evaluated in the field.

A large portion of the Front-Range is a mosaic of private, city, county, State, and Federal public lands. An assessment of capture techniques will allow future assessments of research feasibility and limitations that might be imposed by various land ownerships. Testing capture techniques and potential management actions will also allow for an assessment of the receptiveness of future research within the various political/social environments.

STUDY AREA

GENETICS

Identifying population structure for cougars and bears is a statewide effort. The initial effort for cougars is based on the entire female segment of harvested cougars for the state. The female harvest for bears is much larger, so the sample involved a group of bears from each of the northwest, northeast, southwest, and southeast state regional portions of bear habitat, in an attempt to capture the greatest genetic diversity for the state through spatial separation of sample areas.

The genetic degradation study will be conducted at the Foothills Wildlife Research Facility, located in Fort Collins, Colorado. This is the facility where 3 sibling cougars have been raised in captivity and are part of other ongoing research efforts.

COUGAR/HUMAN INTERACTION

The pilot field study is being conducted in Boulder and Jefferson counties, in an area from near Interstate 70 north to approximately Lyons, Colorado, which is also a likely area for addressing long-term research objectives (see Study Plan APPENDIX II-B, Figure 1). This area is comprised of many land ownerships, including private, Boulder City, Boulder County, Jefferson County, and State and Federal owned lands. Therefore, we have been directly involved with Boulder City and Boulder and Jefferson county governments to obtain agreements from these entities on conduct of research and protocols for dealing with potential human/cougar interactions prior to conducting any research efforts.

METHODS

GENETICS

Genetic samples for the statewide population structure were obtained from statewide voluntary tooth collections from harvested cougars and bears. DNA was extracted from teeth using the DNeasy Blood and Tissue Kit (see Study Plan APPENDIX I-A, sub-appendix I). Following extraction, samples were sent to Sara Oyler-McCance at the Rocky Mountain Center for Conservation Genetics and Systematics, for PCR and sequencing (again, see sub-appendix I for specific methods). A major effort for this year was for the laboratory to develop and test primers for both cougar and bear genetic work. All future work with this laboratory will not incur these upfront costs of initial setup for these species. The cougar genetic degradation portion of the genetic study will be initiated in fall 2007.

COUGAR/HUMAN INTERACTION

A major effort involved coordinating access and research agreements with Boulder City, and Boulder and Jefferson counties. These agreements were essentially achieved after several meetings with these entities to discuss the research and also involved public meetings with Boulder city and county advisory boards. Final, signed working agreements among these entities and CDOW were not entirely finalized until late August 2007.

Through August 2007, cougar capture efforts only occurred on Jefferson County open space lands (White Ranch and Lacy properties) because a signed agreement had only been achieved with Jefferson County. Capture efforts with hounds occurred during April and May 2007. Baiting, using deer and elk carcasses, has been conducted regularly during April-August 2007. Bait sites are monitored using digital trail cameras to determine bait site activity. Cage traps were generally used for capture when cougars removed the bait and cached it. Captured cougars were anesthetized, monitored for vital signs, aged, measured, and ear-tagged. Adults were fitted with GPS collars. Subadults were released without collars because of dispersal potential and difficulty fitting a static collar to a young cougar that may grow substantially. For detailed capture and handling procedures see the study plan APPENDIX II-B.

RESULTS AND DISCUSSION

Two project study plans were completed in anticipation of long-term research efforts to improve the management of cougars in Colorado: 1) Evaluating genetic techniques for application to cougar population studies (see study plan APPENDIX I-A), and 2) Pilot Study--Front-Range cougar-human interactions: Feasibility assessment of techniques and protocols (see study plan APPENDIX II-B).

Additionally, considerable effort was focused on developing CDOW internal protocols for dealing with intra-agency coordination between research and management of conflict cougars involved in the research project. A working document was approved and signed by the CDOW Director defining internal CDOW protocols for dealing with research cougars in urban areas that outlined procedures, involvement, and communication between research and field management personnel based on perceived risk to public safety (see APPENDIX II-B, sub-appendix I). Standardized protocols are needed to maintain requisite levels of consistency within study populations. These protocols will also focus liability, political, and social pressures on CDOW as a whole, and not on individuals or sections within CDOW. One objective of this pilot year field study was to test the management and communication protocols and modify protocols as needed prior to conducting large scale research studies requiring larger commitments of time and resources.

GENETICS

Using 6 samples each for cougars and bears, the Rocky Mountain Center for Conservation Genetics and Systematics laboratory was successfully able to develop primers and successfully sequence nuclear DNA for both species. The laboratory was also successful in developing methods to analyze mtDNA for bears. Analysis of mtDNA for cougars has been difficult due to the transposition of mtDNA into the nuclear DNA for cougars.

DNA was extracted from all 308 bear teeth collected from harvested bears during 2006. Of those, 49 females were selected for genetic sequencing, representing 4 distinct spatial groups across the state (Figure 1). Using 8 microsatellite loci, all 49 female bears were genotyped. Using program STRUCTURE (2007), the data indicate that bears in Colorado function as one megapopulation, rather than as distinct subpopulations (Figure 2). Examination of control region mtDNA haplotypes revealed minimal spatial structure as well (Figure 3). Given this data further examination of both microsatellite and mtDNA is warranted for conclusive evidence of the true population structure of bears within the state.

DNA was extracted from all 128 cougar teeth collected from harvested cougars during 2006-2007. Additionally, DNA was extracted from samples taken from the Uncompahgre Plateau cougar study (Logan 2006), and from samples taken in the northern Front Range as part of C. Krumm's research project (Krumm, unpublished data). All female samples from the harvest plus additional samples from the other research projects were selected for genetic sequencing for a total of 54 samples across the state. Given the limited sample size for females, spatially distinct groups were not available. Using 15 microsatellite loci, all 54 samples were genotyped. The data revealed almost no population structure for cougars across the state (Figure 4). Again, we do feel that, for completeness, additional samples should be analyzed with regard to population structure in order to have conclusive evidence about the lack of population substructure for cougars within the state.

COUGAR/HUMAN INTERACTION

During 2006-07, several internal CDOW meetings were held to discuss the interaction between research and field management personnel operations when dealing with Front-Range cougars. These meetings resulted in the development of the "Colorado Division of Wildlife Protocols for Front-Range Cougar Pilot Research Project," which outlines how cougars at various levels of human interaction will be handled with regard to research and management issues of human health and safety. This document also outlines the personnel which will be involved in decisions about cougars, and internal and external communication about the project. In March, 2007, the director of CDOW signed the document, accepting the protocols as operationally feasible (APPENDIX II-B, sub-appendix I).

On January 25, 2007, CDOW held a meeting with Jefferson County, Boulder City, and Boulder County open space administrations to discuss the Front-Range cougar program. All entities supported the project. On February 16, 2007, CDOW received a signed research agreement from Jefferson County Open Space (JCOS) approving access and research activities on their properties. Meetings with Boulder county Parks and Open Space Advisory Committee (POSAC) were held in March and April, 2007. The POSAC board recommended support for the project, by unanimous vote, following the April 2007 meeting. On May 22, 2007, CDOW presented the project to the Boulder county commissioners, whom approved the project. Similar meetings were held with Boulder City Open Space and Mountain Parks board of trustees. At the final meeting on March 28, 2007, the board recommended approving the project and allowing access to Boulder city open space lands. Research agreement letters for both Boulder county and city had final signature authority by late August, 2007.

In May, 2007, cougar capture efforts commenced on the White Ranch and Lacy properties, which are owned and managed by JCOS. During May, 8 capture days were spent using hounds, resulting in 2 unsuccessful chases of cougars, 2 successful chases of bears, and 4 days with no chases. The 2 cougar chases were initiated from cougars identified at bait sites set-up on the White Ranch property.

From May 2 through August 31, 2007, 165 bait nights (one bait night equals one active bait site per night) were conducted, resulting in 10 visits by cougars. Only 2 of these 10 visits by cougars resulted in a cougar taking the bait and providing a trapping opportunity. On both of these occasions a cage trap was set. On June 14, 2007, a subadult male cougar was caught on the Lacy property, ear-tagged and released without a collar because of the likelihood of dispersal out of the area. On July 14, 2007, an adult male cougar (AM04) was caught on the White Ranch property, and was fitted with a GPS collar. Other than cougars, skunks, fox, and bears visited bait sites frequently. From July 18 to August 31, bears took the bait from bait sites on 14 occasions.

On August 17, 2007, the GPS collar on cougar AM04 was downloaded. Acquisition success rate was 56% of 288 total attempts. Most of the acquired locations were "good" locations with dop ratings < 10. Failed acquisitions were generally grouped together, indicating the cougar was in a location with poor GPS coverage for some period of time. The successful GPS locations indicate that cougar AM04 uses an area extending from the capture site on White Ranch, north to the city of Boulder (Figure 5). Some of the locations are in urban/exurban areas, but use of these areas is limited and appears to be random.

SUMMARY

The Rocky Mountain Center for Conservation Genetics and Systematics laboratory is now prepared to perform nuclear and mitochondrial genetic work on cougars and black bears. Based on sample sizes of approximately 50 individuals each, nuclear or mitochondrial DNA are suggesting that bears and cougars each represent a megapopulation within the state of Colorado. However, we feel further investigation is warranted to increase the strength of evidence.

Initiating the Front-Range cougar project was also successful, as working agreements have been implemented with all 3 municipal open space entities within our study area. We have also demonstrated the limited success of baiting and cage trapping to capture cougars, at least during summer months. We were successful in capturing 2 cougars and were able to GPS collar one adult male. Location acquisition success for the GPS collar is low (56%) but with 8 acquisitions per day, valuable information is still attainable from the GPS collars.

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Prepared by ____

Mathew W. Alldredge, Wildlife Researcher

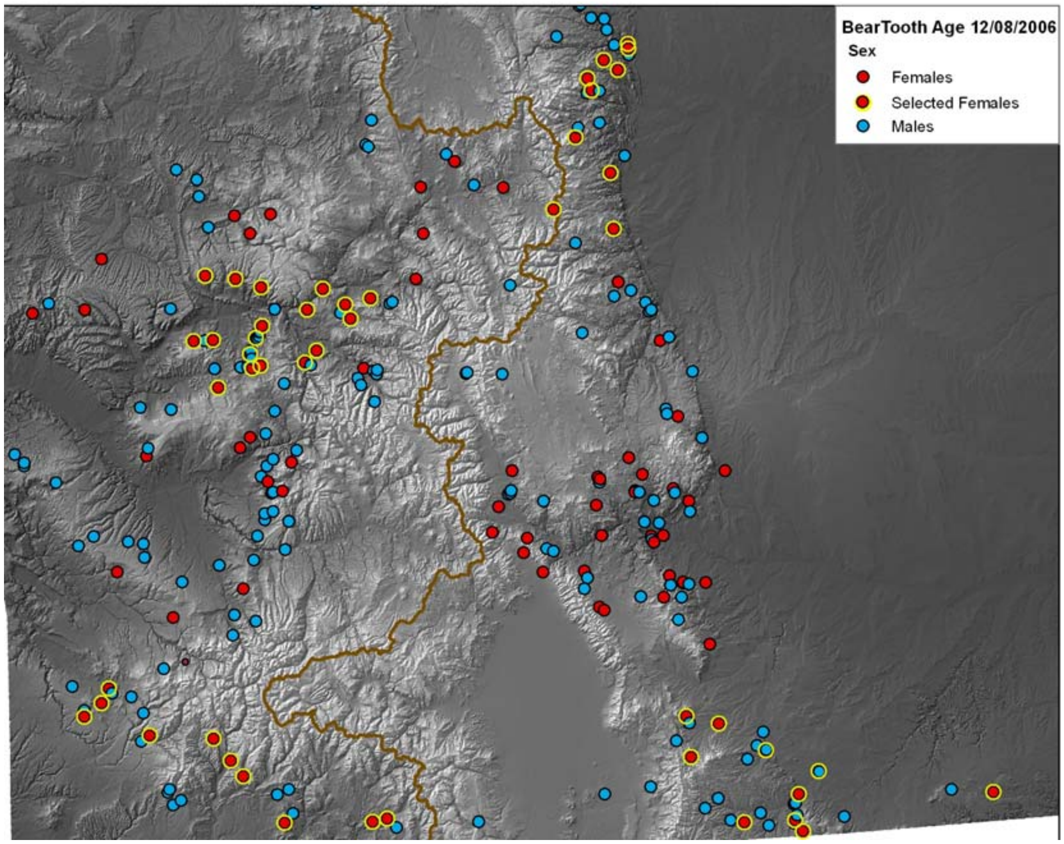

Figure 1. Locations of 308 bears harvested in Colorado during the 2006 hunting season. Locations for the 49 female bears from four distinct locations for genetic identification of population structure are highlighted.

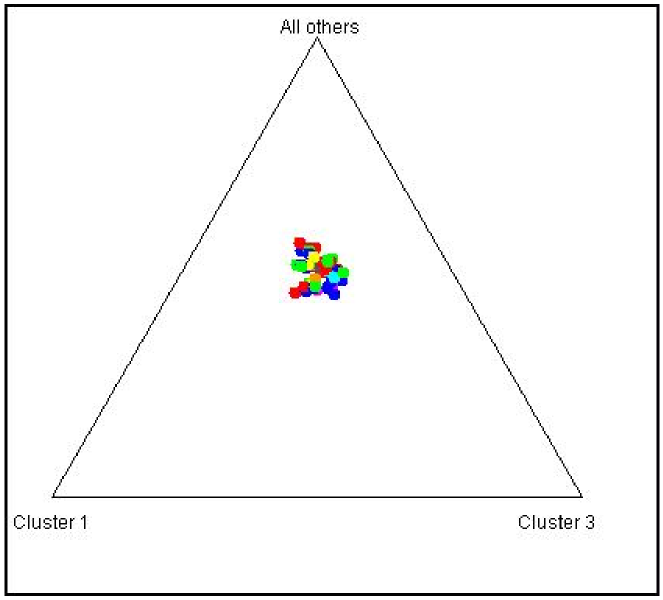

Figure 2. Cluster diagram of microsatellite data identifying little evidence of population substructure for bears across the state of Colorado based on 49 individual females harvested in 2006 from four distinct spatial locations representing the northeastern, northwestern, southwestern, and southeastern portions of Colorado.

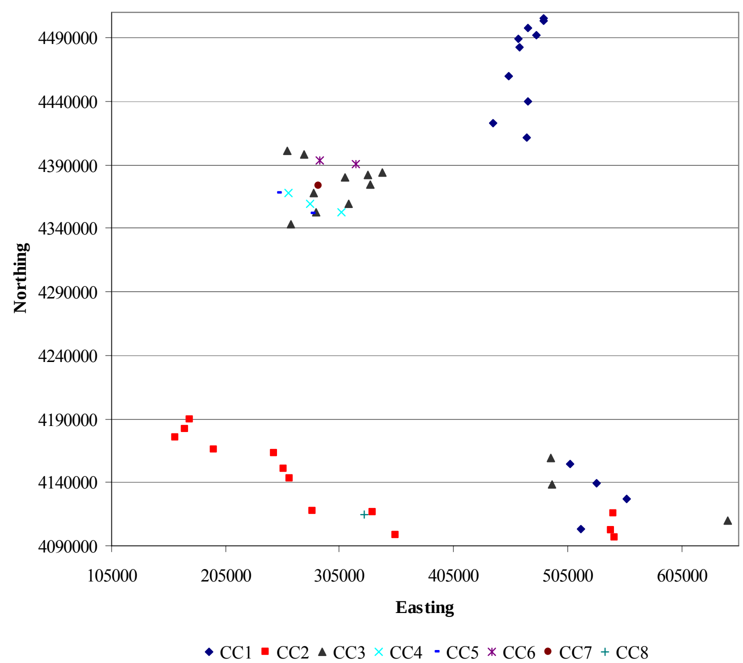

Figure 3. Mitochondrial DNA haplotypes for bears plotted by harvest location for 49 bears harvested in the 2006 hunting season. Although there are 8 haplotypes represented in the data, only 3 occur frequently.

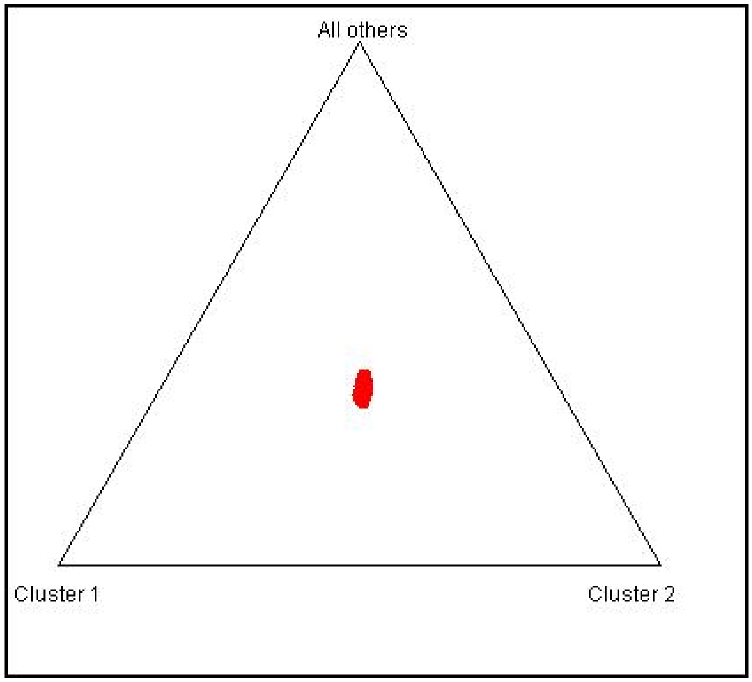

Figure 4. Cluster diagram of microsatellite data identifying little evidence of population substructure for cougars across the state of Colorado based on 54 individual females.

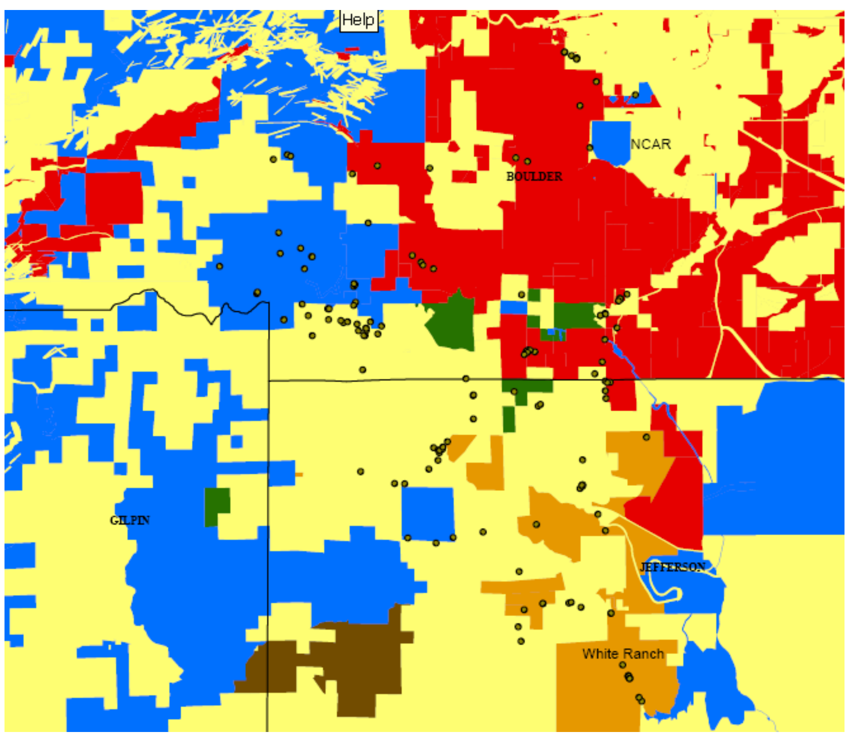

Figure 5. GPS locations for cougar AM04 from July 14, 2007 (date of capture) to August 17, 2007.

APPENDIX I-A

PROGRAM NARRATIVE STUDY PLAN
FOR MAMMALS RESEARCH
FY 2006-07

State of Colorado	: Division of Wildlife
Cost Center 3430	: Mammals Research
Work Package 3003	: Predatory Mammals Conservation
Task No.	: Evaluating Genetic Techniques for Application to Cougar Population Studies

EVALUATING GENETIC TECHNIQUES FOR APPLICATION TO COUGAR POPULATION STUDIES

Principal Investigators
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Paul Lukacs, Biometrician
Sara Oyler-McCance, Rocky Mountain Center for Conservation Genetics and Systematics, USGS
David J. Freddy, Wildlife Research Leader, Mammals Research
Cooperators
Michael Miller,
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Karen Griffin,
Jerry Apker,
Ken Logan
Others

STUDY PLAN APPROVAL

Prepared by:	Mathew Alldredge		Date:	
Submitted by:	Mathew Alldredge		Date:	4/18/2007
Reviewed by:	Mike Phillips		Date:	5/7/2007
	Eric Bergman		Date:	5/24/2007
			Date:	
Reviewed by:	Paul Lukacs		Date:	6/25/07
	Biometrician			
Approved by:	Dave Freddy		Date:	6/25/07
	Mammals Research Leader			

PROGRAM NARRATIVE STUDY PLAN
FY 2006-07

EVALUATING GENETIC TECHNIQUES FOR APPLICATION TO COUGAR POPULATION STUDIES

A study proposal submitted by:

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David J. Freddy, Wildlife Research Leader, Mammals Research

NEED

The use of genetic techniques for wildlife research is becoming increasingly common. Genetic information can be used for species identification, evaluation of diets, gender identification, maternal and paternal lineage, and individual identification. This information, in turn, can then be used to describe population demographics, such as sex ratios, population size, dispersal patterns, and local subpopulations.

Genetic techniques for monitoring or research of rare, elusive, and wide ranging species are of particular interest as other techniques are either impractical or financially prohibitive. Genetic techniques for monitoring and research of cougars in Colorado may be invaluable as alternative techniques are expensive and in many situations may not be possible. Capture and handling of cougars is expensive, time consuming, and may not give representative samples of the population. Large dispersal distances of cougars, especially males, will require impractically large study areas in order to understand demographic patterns that are affected by immigration. Capture may not even be possible in suburban and exurban areas of Colorado as logistical constraints associated with private land owners will likely prohibit the use of many capture techniques.

Genetic samples can be obtained from tissue samples from harvested lions or other removals associated with depredation or human conflict. Samples from these sources may not be representative of the cougar population, however, as harvest is not truly random but is affected by harvest pressure and vulnerability of various sex and age classes. Juvenile males may be most vulnerable to harvest as they are very mobile and may not have established home ranges. Adult females, especially those with cubs, are likely the least vulnerable to harvest, except at high harvest levels (Anderson and Lindzey 2005). Hunter selectivity and sex biased harvest quotas will also influence the composition of the harvested population. These data are still important but other data are necessary for specific demographic questions, such as population estimation.

Noninvasive genetic sampling (Hoss et al. 1992, Taberlet and Bouvet 1992) has the potential to provide a realistic method of sampling a population of interest. Noninvasive sampling techniques include the use of hair snares, and scat collections (Harrison et al. 2004, Smith et al. 2005). The use of scats for sampling cougar populations may be particularly useful and provide a representative sample of the population. Scat collections can either be done by searching transects with human observers (Harrison et al. 2004) or with trained dogs (Smith et al. 2005). Scats could also be collected from kill sites. Kill sites would probably need to be based on mortalities of radio-collared ungulate populations. Data from noninvasive sampling techniques are useful in describing dispersal patterns and estimating population size. Noninvasive genetic data are error prone, which in many cases is because of the quantity and quality of genetic material relative to the collection of noninvasive samples (Study plan A—Degradation of Genetic Markers from Fecal Samples).

Use of genetic data for other purposes, such as delineating subpopulations, is also very useful for managing cougar populations in the state but do not require the amount of randomization of sampled individuals that is required for population estimates. In these cases the goal is to examine local characteristics of the genetic data and determine if it is different among areas (subpopulation structure) or is similar across all areas (panmictic population). Nuclear DNA is inherited from both the mother and the father and therefore is less likely to describe sex-linked population structure. Male cougars generally disperse over greater distances than females and therefore a female population substructure may exist, which has not been described in previous studies. Examination of cougar population structure has been examined using nuclear DNA but few studies have examined cougar population structure using mitochondrial DNA (mtDNA). Mitochondrial DNA is only inherited from the female in mammals and therefore lends itself to delineating female cougar population substructures (Study plan B—An Assessment of female cougar population substructure using mitochondrial DNA).

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PROGRAM NARRATIVE STUDY PLAN PART A
FY 2006-07

DEGRADATION RATES OF GENETIC MARKERS FROM FECAL SAMPLES

A study proposal submitted by:

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David J. Freddy, Wildlife Research Leader, Mammals Research

NEED

Noninvasive genetic sampling of cougars may prove to be a useful method to estimate population size of cougars across the state, which is something that has not been successfully done using traditional methods. The use of hair is probably not an effective method for felids because of the amount of shedding, which limits the number of samples that actually produce DNA. Scats however, should provide a useful method of collecting genetic material as epithelial cells are shed from the intestinal track and appear in the feces. Using species specific primers, DNA from the epithelial cells are amplified during PCR. Important considerations that must be made are the timing of collections and the method of sampling in order to make valid inferences about population size.

Defining a sampling scheme for the collection of scats is critical to obtaining unbiased estimates of population size. Using radio-collared or captured cats and invasively collected genetic samples could be used as a means of identifying cats or producing the initial marking event but resampling must be done independently of knowledge about cat locations. Potentially transects could be searched for cougar scats but this is likely to be inefficient and yield low sample sizes. The use of scat-searching dogs may also be possible but efficiency may still be low, especially in large areas where access may be limited. Another option would be to use hunter harvest as the resampling event. Concerns with this would be sex and age bias in the harvest, but stratification may overcome these issues. A final potential approach would be to mark prey and locate scats from known kill sites. The only potential bias with this would be if marking prey affected how cats selected prey. This approach would benefit from the short time span between scat deposition and collection.

Other considerations when using scat are the quantity and quality of genetic material that can be obtained from the sample, which are factors of time since deposition and environmental conditions. Extrinsic factors, such as season, diet, and sample age, can cause variation in genotyping error rates (Lucchini et al. 2002, Murphy et al. 2002, Piggott 2004). Piggot (2004) found that red fox and rock-wallaby samples collected during the summer in Australia produced fewer genetic errors than samples collected during the winter. They attribute this to warm-dry summers compared to rainy-moist winters. Lucchini et al. (2002) found that wolf feces collected in the western Italian Alps during the winter produced higher quality genetic samples than samples collected during the spring and summer, and fresh samples were better than older samples. Winter samples collected in this study were found on snow, indicating that the samples were likely frozen for the majority of the time following deposition. Both of these studies indicate that fresher samples have lower genetic error rates than older samples.

Potential weaknesses with noninvasive genetic sampling are low success rates, contamination concerns, and high genotyping error rates (Taberlet et al. 1999, Waits and Leberg 2000). Even small genotyping errors (< 1%) cause reasonably large overestimation of population size (Waits and Leberg 2000). Some of these errors include allelic dropout (alleles fail to amplify) and false alleles (spurious amplification of an allele that does not exist for a given individual) (Flagstad et al. 2004, Piggott 2004). Lucchini et al. (2002) only used wolf scats less than 2 weeks old and found that, of the 130 samples analyzed, 64% successfully sexed and 53% successfully genotyped an individual. Inaccurate genotyping due to allelic dropout occurred in 18% of the PCR samples and amplification of false alleles was negligible. Piggot (2004), using captive red fox found that 6 month old feces did not amplify and amplification rates decreased significantly over time. This study also demonstrated that allelic dropout occurred more rapidly than amplification of false alleles. Similar problems have been documented for genetic work on cougar. Ernest et al. (2000) document 17 of 32 fecal samples collected in Yosemite Valley, California, did not amplify because samples were either of nonfelid origin, DNA was degraded, or fecal compounds inhibited detection. In a similar study of cougar in the Peninsular Ranges of California, it was found that only 77% of scats that provided DNA (63% of the samples) also provided the resolution to identify an individual (Ernest et al. 2002).

Paetkau (2003), in a review of 21 population inventory studies, states that it is unclear what the frequencies of errors are for different types of errors in relation to the type and quality of the samples being collected and suggests that use of noninvasive sampling will be limited unless evidence is provided to support the reliability of the data. It is also evident that siblings or closely related individuals provide the greatest challenge to genetically distinguishing individuals. A marker system with the power to distinguish a small number of closely related individuals will also have sufficient power to distinguish among a large number of unrelated individuals (Waits et al. 2001).

Noninvasive genetic sampling of cougar using feces is one of the few methods available that may provide realistic estimates of cougar populations in Colorado, which is important because these populations are being exploited. Before genetic sampling can be conducted an assessment of potential genetic error rates must be made in order to appropriately design the sampling protocols and determine if such a method can provide reliable estimates. Currently there are 3 sibling cougar in captivity at the CDOW Wildlife Health Lab, which can be used to produce known age feces. These scat samples can be subjected to both controlled and uncontrolled environmental conditions in order to accurately asses the magnitude of error rates that may be expected if scats are used for noninvasive population estimation of cougar.

In addition to these captive cougar, there are also numerous cougar that are currently being monitored using GPS collars. Invasive genetic samples have been collected from all of these monitored cougar. Current efforts to identify kill sites of these cougar based on GPS clusters could also be used to collect cougar scats with a known age of within a few days. These scats could be marked and genetic samples could be collected at various time intervals and assessed for error rates for these scats, which are subject to different environmental conditions.

OBJECTIVES

1. Evaluate differences in DNA quantity from either a scat surface collection or a cross-sectional collection.
2. Evaluate differences in DNA quantity from successive feces depositions to determine the variation in quantities of genetic material in scats. Quantify differences in epithelial shedding rates.
3. Evaluate temporal, environmental, and seasonal effects on DNA quantity and quality for both controlled and uncontrolled conditions.

EXPECTED RESULTS OR BENEFITS

Cougar in Colorado provide hunting opportunities across the majority of their current range, and the Colorado Division of Wildlife is responsible for setting reasonable harvest quotas in order to reach management objectives. This is an inherently difficult task, especially when realistic estimates of population size are not attainable. This study will provide basic information on degradation rates with respect to time, environmental condition, and season, which can then be used to design sampling protocols for noninvasive genetic sampling for the purpose of population estimation. This study will also provide estimates of genotyping error rates which can be incorporated in population estimation models to account for misidentification of individuals.

APPROACH

Working Hypothesis 1: In this investigation we will first test if the quantity of DNA sample is affected by whether the sample is taken along the surface of the scat or if it is taken as a cross-section of the scat. Quantity of DNA in feces can be low and therefore, it will be beneficial to sample scats in order to maximize DNA quantity in the sample. It may also be that genetic degradation due to some environmental factors is greater on the exposed surface of scats, which may indicate benefits of a cross-sectional sampling approach if genetic yield is sufficient.

In the case of how to sample scats to maximize genetic quantity, we are only interested in large differences between the proposed collection methods. Small differences will be of little biological significance. Therefore, we will collect paired samples from 20 scats, within 12 hours of deposition. A longitudinal sample along the surface of the scat, and a cross-sectional sample, including the surface, will be collected from each of the 20 scats. DNA will be extracted from the samples and DNA quantity will be measured using real time PCR.

Scat will be collected from three sibling cougars that are in captivity at the Foothills Wildlife Research Center in Fort Collins, Colorado. Scats will be collected from a known individual, and within six hours of deposition. Upon collection a 100 mg sample will be collected longitudinally along the surface and a 100 mg sample will be collected as a cross-section of the scat. Each scat will be assigned a sample number and each collection will be given the sample number and identified as to the collection method so that samples can remain paired. Samples will then be frozen at -20 C, until DNA extraction. DNA extraction will be done using QIAamp® DNA Stool Mini Kit (Appendix I), a commonly used method for extraction. Following extraction, DNA quantity will be measured using real time PCR(Appendix I).

DNA quantity could vary due to many factors, however, pairing samples should account for sources of variation other than collection method. A paired t-test will be used to test for significant differences in DNA quantity associated with collection method.

Working Hypothesis 2: We will also test the variability in DNA quantity over successive depositions of feces. It is likely that the amount of epithelial cells shed varies over time, which will affect the quantity of DNA in feces. Understanding this variability will be important in sorting out other sources of variation in this study and will also help to determine if multiple scats should be collected from a single individual, if possible. For example, if DNA quantities can be extremely low in some scats, then this will be a potential reason for amplification failures.

The variable of interest is, again, DNA quantity. In some respects the only real interest is the minimum amount of DNA that appears in feces. If minimum levels are low enough so that DNA amplification is a concern then the proportion of time that these low levels occur is also of interest.

Therefore, we will collect samples from all feces over a 20 day period from the 3 captive cougars located at the Foothills Wildlife Research Center. From this we will determine the general form of the distribution of DNA quantity within cougar scats and basic descriptive statistics, including the minimum observed quantities of DNA.

It will also be possible to determine if variation in DNA quantity varies with respect to a temporal pattern, since we are working with captive cougar. The captive cougars will be maintained so that feces can be associated with the individual (possibly using orally fed fluorescent markers) and the time series of collections will be tracked. From this we will be able to determine if there is any temporal pattern in DNA quantity. This information may assist in methods of collecting feces from kill sites located in the field where there may be multiple scats from an individual cougar.

Scat will be collected from three sibling cougar that are in captivity at the Foothills Wildlife Research Center in Fort Collins, Colorado. Scats will be collected from all individuals, and within 12 hours of deposition over a 20 day period. Two of the captive cougars will be fed a fluorescent marker, so that scats can be associated with an individual cougar. Upon collection, a 100 mg sample will be collected longitudinally along the surface of the scat. Each scat will be assigned a sample number and each collection will be given the sample number, which will identify the time of deposition as well. Samples will then be frozen at -20 C, until DNA extraction. DNA extraction will be done using the QIAamp® DNA Stool Mini Kit (Appendix I). Following extraction, DNA quantity will be measured using real time PCR (Appendix I).

Of primary interest will be basic statistics to determine the range (min, max), mean, and variability of DNA quantity in scat samples. We will also determine the general shape of the distribution and the minimum sufficient statistics necessary to describe the distribution. We will also examine the data for predictable patterns in changes in DNA quantity over time using a time series analysis or curve fitting routine.

Working Hypothesis 3, part A: Our final hypothesis that we are testing is the rate of genetic degradation that occurs temporally, under various environmental conditions. Some of these experiments will be done under controlled conditions, so that a relationship can be determined for the rate of degradation with respect to a specific factor (part A). Other analyses will involve scats that are subjected to more naturally occurring environmental conditions to help develop a better understanding of reasonable time frames following deposition that scats should be collected given particular environmental conditions (part B).

Scats will be collected from three sibling cougars that are in captivity at the Foothills Wildlife Research Center in Fort Collins, Colorado. Scats will be collected from all individuals, and within six hours of deposition. Two of the captive cougars will be fed a fluorescent marker, so that scats can be associated with an individual cougar. Each scat collected will be assigned a collection number that identifies the time of deposition. Scats from each individual will then be randomly assigned to one of the three treatment groups (-5 C, +5 C, and +15 C), such that 20 scats from each individual will be in each temperature regime. Scats will be sub-sampled by removing a 200 mg longitudinal sample at 2 weeks, and 1, 2, 3, 4, and 6 months post deposition. After collection, each sample will be frozen at -20 C, until DNA extraction. DNA will then be extracted using QIAamp® DNA Stool Mini Kit extraction kits.

Response variables that will be measured are number of incorrect identifications, allelic dropout rates (actual number of alleles that dropout in any given sample), and number of false alleles. The primary analysis will be a logistic regression on the dichotomous identification variable, treating the three temperature regimes as covariates. Additional analyses will summarize the rate at which alleles dropout and the occurrence of false alleles.

A total of 60 scats will be collected and sub-sampled at each time period. Based on simulating data under a logistic model and assumed error structure, the probability of detecting that the error rate is larger than 5% when the true error is 10%, is approximately 0.63. It is likely however, that genetic degradation will occur over a short time interval, which will render the logistic model less effective. In this case data will be pooled into pre- and post-degradation groups and misidentification rates compared between the groups. In this case the power of the comparison will be greater than that for the logistic analysis. Additionally samples will be analyzed by time period, and once misidentification rates are greater than 50% for a time period, no sequencing will be conducted on scats at larger time periods.

PCR and DNA sequencing will be done in the Rocky Mountain Center for Conservation Genetics and Systematics laboratory. Individual cougars will be screened and genotyped using 9 -12 nuclear microsatellite loci isolated from domestic cat (Menotti-Raymond and O'Brien 1995, Menotti-Raymond et al. 1999). Three recent studies have used sets of these primers successfully on mountain lion (Ernest et al. 2000, Sinclair et al. 2001, Anderson et al. 2004). We will choose a set of these primers for our work. PCRs will be performed using a M13-tailed forward primer as described by Boutin-Ganache et al. (2001). Each 12.5µl reaction will contain 125µM each dNTP, 1X *Taq* buffer (Kahn et al. 1998), 0.034µM M13-tailed forward primer, 0.5µM non-tailed reverse primer, 0.5µM M13 dye-labeled primer with Beckman Coulter dyes D2, D3 or D4 (Proligo), and 0.31U *Taq* polymerase (Promega). The thermal profile for both the forward dye-labeled and the M13 dye-labeled reactions will be as follows with the appropriate annealing temperature varying by locus: preheat at 94°C for 1 min, denature at 94 ºC for 1 min, anneal for 1 min, and extend at 72 ºC for 1 min for 35 cycles. The PCR products will be diluted and run on the CEQ8000 XL DNA Analysis System (Beckman Coulter). All loci will be run with the S400 size standard (Beckman Coulter) and analyzed using the Frag 3 default method.

Working Hypothesis 3, part B: The second experiment within Hypothesis 3 is to test the rate of genetic degradation that occurs with respect to time under more natural environmental conditions. This phase of the study will be conducted in the second year and data from the first year's degradation study will be used to refine the design of this second phase. Therefore, sample sizes and timing of collections are speculative. Part of this experiment will involve repeated freeze-thaw cycles to determine the number of freeze-thaw cycles that can occur before genetic material is unusable. The second part will be to expose scats from known individuals to natural environmental conditions in order to assess degradation rates that will be encountered in field situations.

Scat will be collected from three sibling cougar that are in captivity at the Foothills Wildlife Research Center in Fort Collins, Colorado for the freeze-thaw experiment. A total of 40 scats will be collected within six hours of deposition utilizing all three cougars. Two of the captive cougars will be fed a fluorescent marker, so that scats can be associated with an individual cougar. Each scat collected will be assigned a collection number that identifies the time of deposition. Scats will then be subjected to 3 freeze-thaw cycles per week (-5 C to +5 C). Scats will be sub-sampled by removing a 200 mg longitudinal sample following 5, 10, 15, 20, and 25 freeze-thaw cycles. DNA will then be extracted using the QIAamp® DNA Stool Mini Kit and then DNA amplification and sequencing will be conducted as described above. Treating each level of freeze-thaw cycle as a treatment group with a binomial response variable of correctly identifying an individual, the probability of detecting an error rate >0.05 when the true error rate is 0.1 is 0.047.

Response variables that will be measured are number of incorrect identifications, allelic dropout rates (actual number of alleles that dropout in any given sample), and number of false alleles. The primary analysis will be a binomial analysis of correct identification rates at each level of freeze-thaw cycles. Additional analyses will summarize the rate of allelic dropout and the occurrence of false alleles.

As part of ongoing research projects, cougars are being captured and monitored with GPS collars, as well as individually genetically identified from tissue samples. Kill or feeding sites are being identified and investigated based, on these GPS locations, which provides the opportunity to locate cougar scat exposed to natural environmental conditions with an approximate known date of deposition. Exposure times range from 2 weeks to 8 weeks post-deposition, based on current field protocols for kill site investigation. As scats are found during these field investigations, a 200 mg longitudinal sample will be collected. Following the above procedures for extraction, amplification and sequencing, the individual will be identified based on DNA and compared to the assumed known individual's genetic identification. The target sample size for this will be 200, which will give us an adequate sample to determine an approximate time frame post-deposition at which genetic identification error rates will increase to unacceptable levels for mark-recapture population analyses.

LOCATION OF WORK

The captive cougar are located at the Wildlife Health Laboratory in Fort Collins Colorado, which is also where DNA extraction will be done. Free-ranging cougars currently involved in research projects are located in the front-range of Colorado and on the Uncompahgre Plateau, in western Colorado. Genetic analyses will be conducted at the Rocky Mountain Center for Conservation Genetics and Systematics, located at Denver University, Denver, Colorado.

SCHEDULE OF WORK

Time	Activity
April 2007	Study Plan Approval
April-June 2007	Part A—scat collections from sibling cougar
April-Nov 2007	DNA extraction and sequencing
Nov-Dec 2007	Statistical Analysis
Jan 2008	Preliminary report—Decision on implementation of Part B
Jan-Feb 2008	Scat collections from sibling cougar (freeze-thaw)
Jan-April 2008	DNA extraction and sequencing
Mar 2007-May2008	Scat collection from free-ranging cougars
Mar 2007-May2008	DNA extraction
Jan-June 2008	DNA sequencing
June-September	Final analyses and report

ESTIMATED COSTS

Activity/category	2006-2007	2007-2008
Part A		
Supplies (extraction kits)	$500	$1,000
DNA sequencing (1200 samples at $50 per sample)		$60,000
Part B		
Supplies (extraction kits)		$500
DNA sequencing (400 samples at $50 per sample)		$20,000
Total	$500	$85,000

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PROGRAM NARRATIVE STUDY PLAN PART B
FY 2006-07

AN ASSESSMENT OF COUGAR AND BLACK BEAR POPULATION SUBSTRUCTURES USING MITOCHONDRIAL AND NUCLEAR DNA.

A study proposal submitted by:

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NEED

Understanding population structure provides important information for management strategies. A clear example is applying different management strategies to adjacent game management units (GMU's). If a single population is represented across both GMU's, then it is not possible to evaluate either management strategy because the effects are on the population and thus, are confounded across the unit boundaries.

Metapopulation structure of cougar populations in California were demonstrated using telemetry and the structure was attributed to small isolated habitat patches created through increased human development (Beier 1993). Logan and Sweanor (2001) suggest that a source-sink population dynamic existed for populations in southern New Mexico cougar populations. Cougar habitats in southern New Mexico are relatively small and are separated by areas of non-habitat, which is typical for population structuring of populations for a species that is capable of dispersing across the areas of non-habitat. However, Logan and Sweanor (2001) claim that the high dispersal rates documented in southern New Mexico make it unlikely that any genetic subdivision exists among cougar subpopulations.

The idea of managing cougar populations relative to source-sink areas is also promoted in the "Cougar Management Guidelines" (Cougar Management Guidelines Working Group 2005). However, this does not specify that source-sink populations exist naturally or if they are artificially created. In many situations it is likely that the source-sink dynamic is artificially created through hunting pressure in more accessible areas and along the human interface in rural areas to avoid conflict. Understanding whether the source-sink dynamic is part of a natural population structure or is artificially created within a spatially intact population has important management considerations.

Genetic analysis of cougar populations is one method that has been successfully used to identify population substructures for cougars in California (Ernest et al. 2003), Wolverine (Tomasik and Cook 2005), and feral pigs (Hampton et al. 2004). In the case of feral pigs, sink areas were artificially created through control efforts and source areas were identified using genetic techniques.

Genetic population structure for cougar populations throughout the west, has been evaluated extensively, but generally through the use of microsatellite markers. In California, genetic subdivision of cougar populations was defined and was associated with geographic boundaries and isolation by distance (Ernest et al. 2003). McRae et al. (2005) also found some genetic substructuring for cougar populations in southern New Mexico and Arizona and similarly attributed this to geographic barriers and distance. However, genetic substructuring has not been identified across several other western states based on microsatellite markers. Analysis of microsatellite markers have shown single large panmictic populations for Utah (Sinclair et al. 2001), Southwestern Colorado, Wyoming, and western South Dakota (Anderson et al. 2004), and northern New Mexico, northern Arizona, Utah, and Colorado (McRae et al. 2005). Culver et al. (2000) using an mtDNA analysis actually suggest that the entire North American population is a single homogeneous population coming from a late Pleistocene recolonization.

Little analysis of cougar population substructuring has been attempted using mtDNA, which is especially well suited to studying mammal populations with male-biased dispersal (DeYoung and Honeycutt 2005). Tomasik and Cook (2005) were able to describe three distinct populations of wolverine using mtDNA, which were not well defined in other studies that used nuclear markers. However, neither mtDNA nor microsatellite markers were able to differentiate population structure that was identified through radiotelemetry, indicating gene flow between the subpopulations is mediated by both sexes (Cronin et al. 2006).

Because mtDNA has the potential to provide an inexpensive method for assessing cougar population substructure across the state of Colorado, it should be evaluated for its efficacy. Given the evidence against such substructuring across the state using microsatellite markers this should be pursued as a pilot study to first determine if any differences can be detected in mtDNA across the state. If differences are detected, then a more intensive sampling scheme can be used to actually delineate potential subpopulations. Because many of the same large carnivore population management issues could be associated with black bears in Colorado, we propose to opportunistically include samples from harvested black bears to assess evidence for population structuring across the state during this initial effort (C. Anderson, personal comm.).

OBJECTIVES

Determine the efficacy of using mtDNA to delineate female cougar and black bear subpopulations across the state of Colorado.

EXPECTED RESULTS OR BENEFITS

In order to manage cougar and black bear populations it is important to understand the population dynamic affecting local and regional populations. Identification of female substructuring of the population across Colorado would influence how management strategies are implemented in the state. Evaluating the use of mtDNA and nuclear DNA as a potential method to delineate populations will help determine whether future efforts are warranted.

APPROACH

The general hypothesis for this pilot study is that mtDNA and/or nuclear DNA can be used to delineate subpopulations of cougars and black bears across the state of Colorado. Given recent genetic studies using microsatellite markers to investigate substructuring in cougar populations, which included Colorado populations, and documented dispersal distances, we expect limited results from mtDNA as well. Investigation of mtDNA as a method to investigate the existence of subpopulations within the state is warranted given speculation about source-sink dynamics of cougar populations within Colorado.

Tissue or blood samples will be obtained from cougars distributed across all available cougar habitat within the state of Colorado. Currently, approximately 70 tissue samples have been collected across the state for law enforcement purposes. About 40 additional tissue or blood samples have been collected along the northern front-range of Colorado and are stored at the Wildlife Health Lab. The majority of these samples will be used for this pilot study. Tissue samples have also been collected as part of the Uncompahgre Plateau, cougar research study, which should also be available for analysis.

Starting in 2006, voluntary tooth collection was initiated for all harvested bears and cougars in the state of Colorado to determine age structures in the harvest. DNA will be collected from all teeth collected from harvested bears and cougars. As of January, 2007, approximately 300 teeth have been collected from harvested bears and DNA has been extracted. This sampling effort provides genetic samples from bears and cougars across the state and will provide some areas with more intensive sampling efforts. This will allow for the use of genetic clustering algorithms to be used as well as direct comparisons among intensively sampled areas.

An initial analysis will be conducted by analyzing genetic samples from 10 to 15 female bears and cougars within each of 4 geographically distant areas across the state. Based on these genetic samples we will determine if nuclear or mtDNA can be used to identify differences among these 4 distinct groups. If no differences appear to exist, then analyses of remaining samples will not be done and we will conclude that genetic substructure in the bear or cougar population across Colorado does not exist. If differences are found among these four groups, then the remaining samples will be analyzed and substructures of the Colorado bear or cougar population will be identified.

DNA will be extracted using the DNeasy Blood and Tissue Kit, from collected tissues and teeth, following the procedures outlined in appendix I.

We will attempt to amplify and sequence approximately half of the black bear mitochondrial control region using primers developed previously (Shields and Kocher 1991, Ward et al. 1991). Amplification will be accomplished using our standard 25µl polymerase chain reaction (PCR) as in St. John et al. (2005) [125µM each dNTP, 1X *Taq* buffer (67mM Tris-HCl pH 8.0, 6.7 mM $MgSO_4$, 16.6mM NH_4SO_4 10mM β-mercaptoethanol; Kahn et al. 1998), 0.5µM each primer and 0.625U *Taq* polymerase (Promega)]. PCR products will be cleaned with shrimp alkaline phosphatase and exonuclease 1 (USB) followed by dye terminator cycle sequencing reactions performed with the Beckman-Coulter Quick Start Sequencing Kit according to the manufacturer's protocol. These will be precipitated according to the manufacturer's specifications, resuspended in 30 µL of formamide and run on a CEQ 8000 XL Data Analysis System using method LFR-b (Beckman-Coulter).

Individuals will be screened and genotyped using 9 -12 nuclear microsatellite loci isolated from various related mammals (Paetkau and Stobeck 1994, Paetkau et al. 1995, Taberlet et al. 1997, Paetkau et al. 1998). Two recent studies have used sets of these primers successfully in black bear molecular studies (Woods et al. 1999, Boersen et al. 2003). We will choose a set of these primers for our work. PCRs will be performed using a M13-tailed forward primer as described by Boutin-Ganache et al. (2001). Each 12.5µl reaction will contain 125µM each dNTP, 1X *Taq* buffer (Kahn et al. 1998), 0.034µM M13-tailed forward primer, 0.5µM non-tailed reverse primer, 0.5µM M13 dye-labeled primer with Beckman Coulter dyes D2, D3 or D4 (Proligo), and 0.31U *Taq* polymerase (Promega). The thermal profile for both the forward dye-labeled and the M13 dye-labeled reactions will be as follows with the appropriate annealing temperature varying by locus: preheat at 94°C for 1 min, denature at 94 ºC for 1 min, anneal for 1 min, and extend at 72 ºC for 1 min for 35 cycles. The PCR products will be diluted and run on the CEQ8000 XL DNA Analysis System (Beckman Coulter). All loci will be run with the S400 size standard (Beckman Coulter) and analyzed using the Frag 3 default method.

We will attempt to amplify and sequence approximately half of the mitochondrial control region using primers that we will design from published sequences in related species (Freeman et al. 2001). Amplification will be accomplished using our standard 25µl polymerase chain reaction (PCR) as in St. John et al. (2005) [125µM each dNTP, 1X *Taq* buffer (67mM Tris-HCl pH 8.0, 6.7 mM $MgSO_4$, 16.6mM

NH_4SO_4 10mM β-mercaptoethanol; Kahn et al. 1998), 0.5µM each primer and 0.625U *Taq* polymerase (Promega)]. PCR products will be cleaned with shrimp alkaline phosphatase and exonuclease 1 (USB) followed by dye terminator cycle sequencing reactions performed with the Beckman-Coulter Quick Start Sequencing Kit according to the manufacturer's protocol. These will be precipitated according to the manufacturer's specifications, resuspended in 30 µL of formamide and run on a CEQ 8000 XL Data Analysis System using method LFR-b (Beckman-Coulter).

Individuals will be screened and genotyped using 9 -12 nuclear microsatellite loci isolated from domestic cat (Menotti-Raymond and O'Brien 1995, Menotti-Raymond et al. 1999). Three recent studies have used sets of these primers successfully on mountain lion (Ernest et al. 2000, Sinclair et al. 2001, Anderson et al. 2004). We will choose a set of these primers for our work. PCRs will be performed using a M13-tailed forward primer as described by Boutin-Ganache et al. (2001). Each 12.5µl reaction will contain 125µM each dNTP, 1X *Taq* buffer (Kahn et al. 1998), 0.034µM M13-tailed forward primer, 0.5µM non-tailed reverse primer, 0.5µM M13 dye-labeled primer with Beckman Coulter dyes D2, D3 or D4 (Proligo), and 0.31U *Taq* polymerase (Promega). The thermal profile for both the forward dye-labeled and the M13 dye-labeled reactions will be as follows with the appropriate annealing temperature varying by locus: preheat at 94°C for 1 min, denature at 94 ºC for 1 min, anneal for 1 min, and extend at 72 ºC for 1 min for 35 cycles. The PCR products will be diluted and run on the CEQ8000 XL DNA Analysis System (Beckman Coulter). All loci will be run with the S400 size standard (Beckman-Coulter) and analyzed using the Frag 3 default method.

LOCATION

This study will be conducted statewide. Project headquarters will be in the Fort Collins office. DNA extraction will be done at the Wildlife Health Lab.

SCHEDULE OF WORK

Time	Activity
Jan 2007	Submit study plan for CDOW review
Sept 2006-Mar 2007	Collection of samples—primarily from hunter harvest
Jan 2006-May 2007	DNA extraction
Feb-July 2007	Genetic Sequencing (first 50 samples)
Aug-Sept 2007	Analysis of sequence data
Oct 2007	Prepare final report
Nov-Dec 2007	Genetic Sequencing of remaining samples (if necessary)
Jan-April 2008	Final report on genetic structure across the state

ESTIMATED COSTS:

Budget for each Species—Includes Genetics Laboratory Setup and Calibration		
Category	2006-2007	2007-2008
Personnel		
Co-investigators		
DNA extraction (Karen Griffin)	**$200.00**	
Sequencing (Rocky Mountain Center for Conservation Genetics and Systematics)		
Initial Startup (8 samples)		
Salary with fringe	$3850.00	
Primers	$300.00	
Microsatellite Sequencing	$307.20	
mtDNA sequencing	$142.08	
Other supplies, equipment	$689.89	
Indirect	$793.38	
Total	**$6082.55**	
Analysis of 50 teeth (Estimated cost of $58.44 per tooth)		
Sequencing	$444.00	
Microsatellite analysis	$640.00	
Salary with fringe	$1150.00	
Other supplies, equipment	$306.98	
Indirect	$381.15	
Total	**$2922.13**	
Analysis of remaining teeth (approximately 250 @ $58.44 /sample)		**$14,610.00**
Total	**$9404.68**	**$14,610.00**

Related Federal Projects: N/A

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APPENDIX I
EXTRACTION OF DNA

Extraction from animal tissue: Using the DNeasy Blood and Tissue Kit

Procedure is based on the Protocol: Purification of Total DNA from Animal Tissues (Spin-Column Protocol), from the DNeasy Blood and Tissue HandBook (Qiagen, 2006)

PROCEDURE

1. Cut up to 25 mg tissue into small pieces, and place in a 1.5 ml microcentrifuge tube. Add 180 µl Buffer ATL.
2. Add 20 µl proteinase K. Mix thoroughly by vortexing, and incubate at 56̊C until the tissue is completely lysed. Vortex occasionally during incubation to disperse the sample.
3. Vortex for 15 s. Add 200 µl Buller AL to the sample, and mix thoroughly by vortexing. Then add 200 µl ethanol (96-100%), and mix again thoroughly by vortexing.
4. Pipet the mixture from step 3 (including any precipitate) into the DNeasy Mini spin column placed in a 2 ml collection tube. Centrifuge at > 6000 x g (8000 rpm) for 1 min. Discard flow-through and collection tube.
5. Place the DNeasy Mini spin column in a new 2 ml collection tube, add 500 µl Buffer AW1, and centrifuge for 1 min. at > 6000 x g (8000 rpm). Discard flow-through and collection tube.
6. Place the DNeasy Mini spin column in a new 2 ml collection tube, add 500 µl Buffer AW2, and centrifuge for 3 min at 20,000 x g (14,000 rpm) to dry the DNeasy membrane. Discard flow-through and collection tube.
7. Place the DNeasy Mini spin column in a clean 1.5 ml or 2 ml microcentrifuge tube, and pipet 200 µl Buffer AE directly onto the DNeasy membrane. Incubate at room temperature for 1 min. and then centrifuge for 1 min. at > 6000 x g (8000 rpm) to elute.
8. Recommended: for maximum DNA yield, repeat elution once as described in step 7.

Following DNA extraction, sample should be divided into at least two aliquots for storage or further analysis.

Extraction from animal teeth: Using the DNeasy Blood and Tissue Kit

1. Place whole pre-molar tooth into a 1.5 ml microcentrifuge tube containing glass beads.
2. Add 180 ul Buffer ATL and 20ul proteinase K. Mix thoroughly by vortexing.
3. Incubate at 56C for approximately 3 hours. Vortex occasionally during incubation to disperse the sample.
4. Follow DNeasy Blood & Tissue kit protocol, starting with step 3 in protocol (see above).

Extraction from animal feces: Using the QIAamp® DNA Stool Mini Kit

Procedure is based on the protocol: Protocol for DNA Isolation from Larger Amounts of Stool and Protocol using Stool Tubes for Isolation of DNA from Stool for Human DNA Analysis, from the QIAamp® DNA Stool Mini Kit Handbook (Qiagen 2001).

1. Weigh the stool sample (we will use ~500 mg) and add 10 volumes of Buffer ASL (e.g. add 5 ml Buffer ASL to 500 mg stool). Vortex vigorously for 1 min or until the stool sample is thoroughly homogenized.
2. Pipet 2 ml of lysate into a labeled 2 ml microcentrifuge tube.
3. Go to step 4 of Protocol using Stool Tubes for Isolation of DNA from Stool for Human DNA Analysis.
4. Centrifuge sample at full speed for 1 min to pellet stool particles.

5. Pipet 1.4 ml of the supernatant into a new 2 ml microcentrifuge tube and discard the pellet.
6. Add 1 InhibitEX tablet to each sample and vortex immediately and continuously for 1 min or until the tablet is completely suspended. Incubate suspension for 1 min at room temperature to allow inhibitors to adsorb to the InhibitEX matrix.
7. Centrifuge sample at full speed for 3 min to pellet stool particles and inhibitors bound to InhibitEX.
8. Immediately after the centrifuge stops, pipet all of the supernatant completely into a new 1.5 microcentrifuge tube and discard pellet. Centrifuge the sample at full speed for 3 min.
9. Pipet 25 µl Proteinase K into a new 2 ml microcentrifuge tube.
10. Pipet 600 µl supernatant from step 8 into the 2 ml microcentrifuge tube containing Proteinase K.
11. Add 600 µl Buffer AL and vortex for 15 s.
12. Incubate at 70°C for 10 min.
13. Add 600 µl of ethanol (96-100%) to the lysate, and mix by vortexing.
14. Label the lid of the QIAamp spin columns provided in a 2 ml collection tube. Carefully apply 600 µl lysate from step 13 to the QIAamp spin column without moistening the rim. Close the cap and centrifuge at full speed for 1 min. Place the QIAamp spin column in a new 2 ml collection tube, and discard the tube containing the filtrate.
15. Carefully open the QIAamp spin column, apply a second aliquot of 600 µl lysate and centrifuge at full speed for 1 min. Place the QIAamp spin column in a new 2 ml collection tube, and discard the tube containing the filtrate.
16. Repeat step 15 to load the third aliquot of lysate onto the spin column.
17. Carefully open the QIAamp spin column and add 500 µl Buffer AW1. Centrifuge at full speed for 1 min. Place the QIAamp spin column in a new 2 ml collection tube, and discard the collection tube containing the filtrate.
18. Carefully open the QIAamp spin column and add 500 µl Buffer AW2. Centrifuge at full speed for 3 min. Discard the collection tube containing the filtrate.

REAL TIME PCR

Because we are interested in quantifying the amount of cougar DNA that is extracted from feces collected under differing conditions, merely using an estimate of DNA quantity from a spectrophotometer is problematic. This is because the DNA extracted from feces is likely comprised of DNA from prey and/or bacteria in addition to DNA from the mountain lion itself. To address this issue, we will attempt a new technique known as real time PCR to quantify the amount of target (i.e. mountain lion) DNA in the extraction. Real time or quantitative PCR detects products during the exponential phase of the reaction allowing for a more precise measurement of starting material since a direct relationship between quantity of target and product exists during this phase. An absolute quantitation method would be used so that different reactions can be compared. Three to four hydrolysis probes (such as TaqMan® probes) complementary to target species will be utilized in conjunction with dilutions of the DNA extractions.

APPENDIX II-B

PROGRAM NARRATIVE PILOT STUDY PLAN
FOR MAMMALS RESEARCH
FY 2006-07

State of	Colorado	: Division of Wildlife
Cost Center	3430	: Mammals Research
Work Package	3003	: Predatory Mammals Conservation
Task No.		: Front-Range Cougar-Human Interactions: Feasibility Assessment of Techniques and Protocols

FRONT RANGE COUGAR-HUMAN INTERACTIONS PILOT STUDY: FEASIBILITY ASSESSMENT OF FIELD TECHNIQUES AND PROTOCOLS

Principal Investigators
Mathew W. Alldredge, Wildlife Researcher, Mammals Research
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Cooperators
Scott Hoover, NE Regional Manager
Mark Leslie, Dave Clarkson, Liza Hunholz, Reid DeWalt,
NE Area Wildlife Managers
Janet George, NE Senior Terrestrial Biologist

Pilot Study Plan Approval

Prepared by:	Mathew W. Alldredge		Date:	12/2006
Submitted by:	Mathew W. Alldredge		Date:	1/25/2007
Reviewed by:	Brent Bibles		Date:	2/15/2007
			Date:	
			Date:	
Reviewed by:	Paul Lukacs		Date:	1/31/2007
	Biometrician			
Approved by:	Dave Freddy		Date:	3/2/2007
	Mammals Research Leader			

PROGRAM NARRATIVE PILOT STUDY PLAN
FY 2006-07

FRONT RANGE COUGAR-HUMAN INTERACTIONS PILOT STUDY; FEASIBILITY ASSESSMENT OF FIELD TECHNIQUES AND PROTOCOLS

A pilot study proposal submitted by:

Mathew W. Alldredge, Wildlife Researcher, Mammals Research
David J. Freddy, Mammals Research Leader

NEED

Animals have two choices as human populations encroach on wildlife habitats: they can leave the area or coexist with humans in altered habitats. When large animals choose to coexist, there are bound to be interactions between animals and humans. This is especially true for cougars in many areas across the United States, including the front-range of Colorado. Torres et al. (1996) found that cougar depredation on pets was strongly associated with new housing developments in California from 1979 to 1993. Cougar-human conflicts are inevitable in urban/exurban areas if viable cougar populations are to be maintained in these areas (Cougar Management Guidelines Working Group (CMGWG) 2005). Cougars are territorial, which implies they are space limited, and are also obligate carnivores on large ungulates, specifically deer and elk. Therefore, the options for cougars to relocate in response to human encroachment are limited. Potential prey for cougars also coexist with humans in relatively large numbers, which maintains these urban/exurban areas as effective cougar habitat.

Although cougar attacks on humans are rare (CMGWG 2005), they have increased in recent decades. From 1890 to 1990 there were a documented 9 fatal attacks and 54 non-fatal attacks on humans in the United States and Canada (Beier 1991, Fitzhugh et al. 2003). Seven fatal and 38 non-fatal attacks on humans occurred following Beier's 1991 publication and Fitzhugh et al.'s 2003 publication. Cougar attack rates on the front-range of Colorado have been estimated at one in every 2.2 million person-days. The increase in attacks also corresponds to a large increase in human-cougar incidents, which are likely due to habitat reduction, human encroachment, increased human recreational activities, and possible increases in cougar populations (CMGWG 2005). Torres et al. (1996) found no differences associated with gender in the likelihood of a cougar attack on humans. However, Ruth (1991) did find that sub-adults were the age group most likely to interact with humans. The CMGWG (2005) found that a combination of inexperience and unfamiliarity with their environment, as well as hunger, may cause young cougars to have more negative interactions with humans.

Given that cougars currently coexist with humans within urban/exurban areas along Colorado's front-range, varying levels of cougar-human interaction are inevitable. The Colorado Division of Wildlife (CDOW) is charged with the management of cougar, with management options ranging from minimal cougar population management, to dealing only with direct cougar-human incidents, to attempted extermination of cougars along the human/cougar spatial interface. Neither, inaction or extermination are practical options nor would they be agreeable to the majority of the human population. In the 2005 survey of public opinions and perceptions of cougar issues, 96% of the respondents agreed that it was important to know cougars exist in Colorado, and 93% thought it was important that they exist for future generations (CDOW, unpublished data).

CDOW wildlife managers are faced with decisions about how to manage cougar populations and individual cougars in order to maintain viable populations and maintain acceptable levels of human safety. Defining acceptable levels of human safety is difficult because people's perceptions are different when interactions do not directly affect them. In the 2005 public opinions survey, only 44% of respondents felt it acceptable to destroy a cougar that attacks and injures or kills a person that is recreating in cougar habitat, while 49% found eliminating the cougar unacceptable (CDOW, unpublished data). Other difficulties associated with managing cougar populations in areas with high levels of human interaction are caused by the limited amount of information that is currently known about cougars in these exurban situations and responses of cougars to management prescriptions (CMGWG 2005).

There is a growing voice from the public that CDOW do more to mitigate potential conflicts (CMGWG 2005), and the Director of CDOW has requested that research efforts be conducted to help minimize future human/cougar conflicts. In order to meet these goals CDOW believes we need to directly test management prescriptions in terms of desired cougar population and individual levels of responses.

Long-term study objectives for the Front-Range Cougar Research project will involve directly testing management responses of cougars at various levels of human interaction. The CMGWG (2005) recommend that part of determining the level of interaction or risk between cougars and humans is to evaluate cougar behavior on a spectrum from natural, to habituated, to overly familiar, to nuisance, to dangerous. These categories are defined as (CMGWG 2005):

> Habituated—frequent use of developed area and cougars appear comfortable in the presence of humans.
>
> Overly familiar—a cougar purposefully approaches a human, or allows a human to approach it after the cougar has seen the human.
>
> Nuisance—cougar exhibits overly familiar behaviors more than once.
>
> Dangerous—displayed non-defensive aggression towards humans (postures, vocalizations, and actions communicating an intent to harm).

Note that aggressive behaviors could also be defensive if the cougar perceives the human as a threat to itself, its young, or a food source, or if the cougar is surprised or harassed by humans. The CMGWG (2005) describes cougar behaviors and the level of risk to humans as perceived by the authors (Appendix I, Table 1). We have added an additional column that categorizes the level of risk, which will be used to determine management treatments that will be applied during research efforts. Although cougars may habituate to human developments and activities (Ruth 1991), both habituated and non-habituated cougars may experiment with humans as potential prey (Aune 1991). The CMGWG (2005) clearly state that there is no scientific evidence to indicate that cougar habituation to humans affects the risk of attack.

Clearly, cougars representing a danger to human health and safety should be removed, but the appropriate response to cougars that are overly familiar or habituated to humans is unclear. There have been no studies confirming the effectiveness of aversive conditioning (CMGWG 2005). There is also a paucity of information on relocation success. The information that does exist suggests that relocation distances should be large (Ruth et al. 1998) and that survival may be low (Ross and Jalkotzy 1995, Ruth et al. 1998)

Studying individual and population level responses of cougars will require capturing and radio-collaring cougars, as well as standardizing responses of CDOW personnel to human/cougar interactions. Therefore, in this initial year, we need to test various cougar capture techniques in urban/exurban areas of interest for effectiveness and public acceptance and to assess the reliability of GPS collars as monitoring tools to assess cougar responses to management prescriptions. More importantly, clearly defined protocols have been developed within CDOW (Appendix I) to direct how researchers and field managers should deal with various levels of risk to human health and safety, and these protocols need to be tested and evaluated in the field.

A large portion of the front-range is a mosaic of private, and city or county owned public lands. An assessment of capture techniques will allow future assessments of research feasibility and limitations that might be imposed by various land ownerships. Testing capture techniques and potential management actions will also allow for an assessment of the receptiveness of future research within the various political/social environments.

A working document has been developed, defining internal CDOW protocols for dealing with cougar in urban areas that outline procedures, involvement, and communication between research and field management personnel based on perceived risk to public safety (Appendix I). Standardized protocols are needed to maintain requisite levels of consistency within study populations. These protocols will also focus liability, political, and social pressures on CDOW as a whole, and not on individuals or sections within CDOW. One objective of this pilot year study is to test management and communication protocols and modify protocols as needed prior to conducting large scale research studies requiring larger commitments of time and resources.

OBJECTIVES

1. Determine the effectiveness of cage traps and hounds for capturing cougars on the Front-Range of Colorado.
2. Determine functionality and suitability of GPS collars in front-range habitats.
3. Implement cougar-human risk protocols and communications within CDOW and among public entities and determine if modifications are necessary.
4. Determine the feasibility of aversive conditioning techniques on cougars within urban/exurban areas, including use of hounds and rubber bullets.
5. Evaluate political/social response to cougar research activities.

EXPECTED BENEFITS

Information obtained from this pilot study will be valuable in designing future studies in terms of logistical and social limitations. Determining the effectiveness of capture methods as well as GPS collars will define the limitations of future studies and realistic objectives. Testing cougar-human risk protocols is also essential in order to construct research projects that will fit within CDOW operational constraints and avoid liability issues with the public. Finally, assessing public support and reactions to research efforts during this pilot study is important so that the possibilities of losing support during longer term research efforts is minimized or at least understood.

APPROACH

Long-term research efforts will test the working hypothesis that specific management actions can affect the distribution, behavior or population structure of cougars in order to minimize negative cougar/human interactions. In order to successfully test such hypotheses we will need to capture, GPS collar, VHF collar, and monitor cougars along the front-range and maintain control over other external anthropogenic factors. A pilot study will be initiated in January 2007 to test the logistical, social, and political constraints that will govern the conduct of cougar research along the front-range. As these constraints are understood, long-term research studies will be designed and implemented to test the efficacy of particular management prescriptions.

The pilot study will be conducted in Boulder and Jefferson counties, in an area from near Interstate 70 north to approximately Lyons, Colorado, which is also a likely area for addressing long-term research objectives. This area is comprised of many land ownerships, including private, Boulder city, Boulder county, Jefferson county, and state and federally owned lands. Therefore, we will be directly involved with Boulder city and Boulder and Jefferson county governments and will obtain agreements from these entities on conduct of research and protocols for dealing with potential human/cougar interactions prior to conducting any capture efforts. These entities will also be informed when research efforts are being conducted on their respective properties.

Prior to conducting this pilot study, management treatments addressing human/cougar interactions that may take place with individual collared cougars will be agreed upon. This agreement will be among the local DWM's, AWM's, biologists and research staff, and will be within the "Internal Colorado Division of Wildlife Protocols for the Front-Range Cougar Research Project," developed and approved by CDOW. Agreements will include information transfer among CDOW research, management, and external entities, as well as management actions to be tested on cougars at various levels of human interaction.

We will attempt to capture 2 cougars in 3 areas; Boulder city open space on the south side of Boulder, Boulder county open space to the north of Boulder, and Jefferson county open space to the south of Boulder. Two to four cougars will be captured using cage traps baited with deer carcasses. Deer carcasses will either be from known cougar cache sites or from road kill. An additional two to four cougars will be captured using hounds to tree individual cougars. Hounds used will be experienced in capturing cougar and may be provided by USDA APHIS-Wildlife Services under the auspices of a working agreement existing between Wildlife Services and CDOW. With complex land ownerships, hounds may be used in small numbers and on leashes as part of a cooperative effort with Wildlife Services to evaluate the effectiveness of leashed hounds. A leash will be necessary to prevent hounds from following cougars onto private lands where we lack permission for pursuit and capture.

All captured cougars will be fitted with Lotek 4400S GPS collars. Additionally, cougars will be ear-tagged in each ear with uniquely identifiable numbers, tattooed if required by directive W-20, and a genetic sample collected using an ear-punch. Sex, approximate age from tooth wear, weight and morphometric measurements will be recorded. Vital signs will also be monitored during handling of cougars. See Appendix II for a detailed description of capture and handling protocols.

In order to develop an understanding of how cougars are using habitats during normal activity periods, responding to human activity diurnally, and to optimize battery life for GPS collars, location acquisition will occur eight times per day and information downloads will occur once per month. Additional monitoring may be done using the VHF signal from the collars. If aversive conditioning is being considered for an individual the collar may be downloaded more frequently to determine the timing and effectiveness of aversive conditioning. Additionally, if an individual cougar is thought to be a risk to human safety more frequent downloads may be required.

Capture efforts will be assessed based on the number of successes versus the number of attempts and the amount of effort per success. Also a descriptive assessment of feasibility for the various methods will be made. Similarly, acquisition rates will be determined for the GPS collars, accounting for time of day and other temporal fluctuations to determine if these collars will provide the desired data for cougars along the front-range. Responses of collared cougars to aversive conditioning or other management actions will also be monitored and summarized descriptively, which will provide information for designing future studies. At this time, we consider aversive conditioning treatments on cougars to potentially be: multiple captures and handling of cougars, single or multiple treatments using rubber buckshot fired from a shotgun, and single or multiple chases using hounds, and potential combinations of capture, hound chases, and rubber buckshot. During the pilot project all cougars will be potential candidates for aversive conditioning in order to evaluate methods of implementing these techniques in a preventative manner.

Protocols for managing cougars at various levels of human interaction and for internal and external communication will also be evaluated and modified throughout this pilot study phase. Periodic assessments will be made by all individuals directly involved with the pilot work to determine if the protocols are sufficient to provide the structure to conduct more intensive research efforts. Public response to research efforts, including capture methods, aversive conditioning, and monitoring, in terms of public complaint, will also be evaluated and assessed in relation to future research efforts. Protocols will be assessed via internal CDOW discussions and in discussions with other cooperating agencies. Public attitudes will be assessed via media articles or coverage of the project and via interviews or limited surveys/interviews of individual citizens.

The above information will then be used to design future research efforts specifically developed to directly assess management prescriptions. The logistical, social, and political information obtained from this pilot effort will define the limits of what can be done, where the project can be conducted, and possibly add direction to initial efforts of studying the human-cougar interface. Long-term studies will begin by 2008, and previously collared cougars will likely be integrated into these studies.

LOCATION OF WORK

The pilot study will be conducted in Boulder and Jefferson counties, in an area from Interstate 70 north to approximately Lyons, Colorado, which is also a likely area for addressing long-term research objectives (Figure 1).

SCHEDULE OF WORK

Time	Activity
Jan-Feb 2007	Study Plan/ACUC Approval
Jan-Feb 2007	Open Space entities approval/access
Mar-April 2007	Capture effort
Mar-Dec 2007	Monitoring
April-Dec 2007	Aversive conditioning

ESTIMATED COSTS

Category	2006-2007
Personnel	
Field Technician(s) (6 months)	$14050
Operating Expenses	
Cage Traps (2)	$5000
Field/Capture Equipment	$4000
Lotek GPS collars (6) & download receiver (purchased in 2005 budget year)	($32,000)
Telemetry Receivers & Antennas	$1900
Aphis (houndsmen)	$7000
Travel	$1,000
Total	32,950

RELATED PROJECTS

Uncompahgre cougar research project near Montrose, CO conducted by CDOW and cougar project being conducted in and around Rocky Mountain National Park by NPS-USGS.

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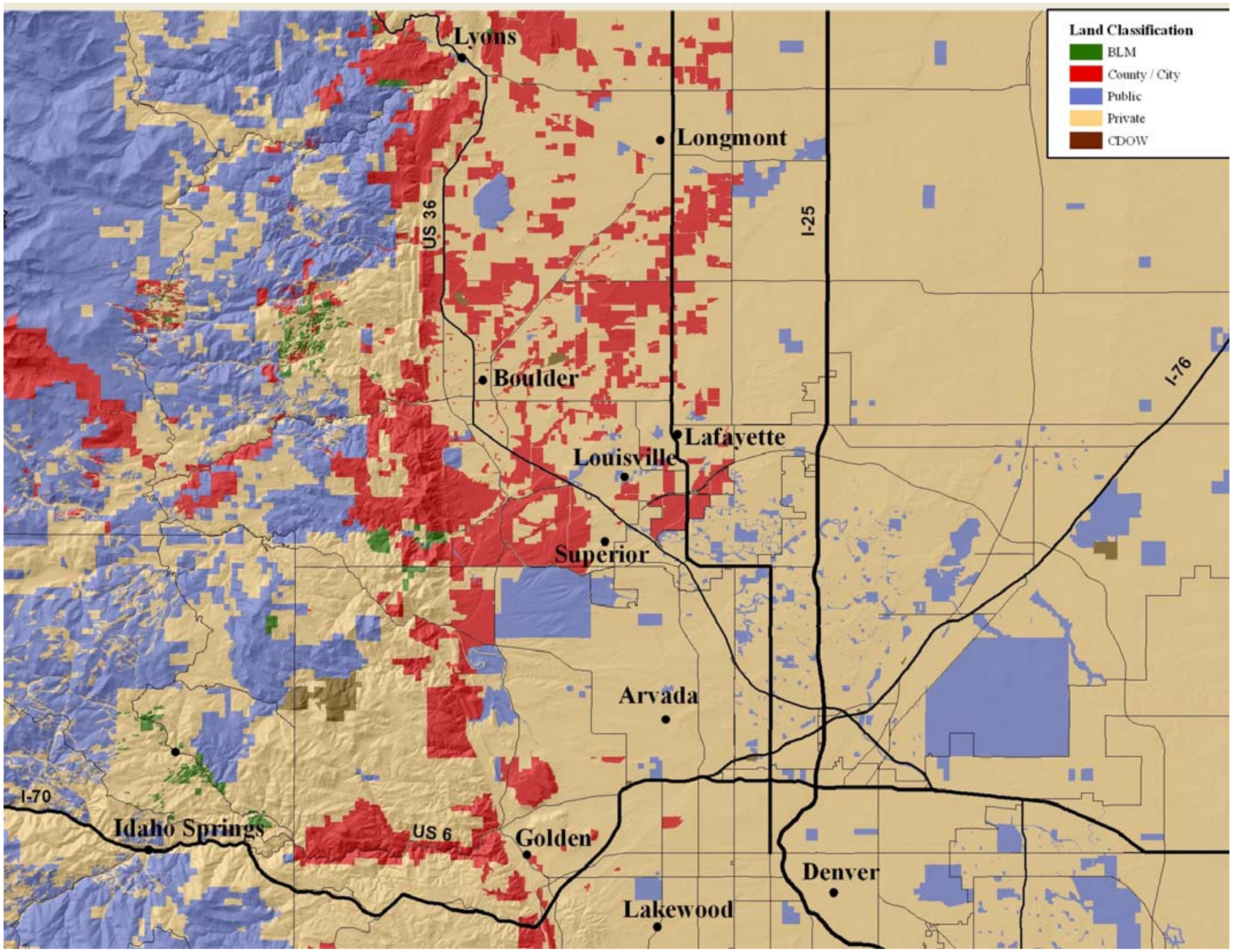
Figure 1. Initial study area 2007.



APPENDIX I

COLORADO DIVISION OF WILDLIFE PROTOCOLS FOR FRONT RANGE COUGAR PILOT RESEARCH PROJECT

Public safety will be the fundamental issue guiding decisions on how to respond to and manage human interactions involving cougars radio-collared for the Colorado Division of Wildlife (CDOW) Front-Range cougar research project. CDOW Administrative Directive W-20 will serve as a basic guideline for managing cougar incidents. These protocols amend Administrative Directive W-20 and provide guidance specific to the Front-Range cougar research project. Human safety will not be compromised for research purposes; original guidelines in Directive W-20 will be explicitly followed for cougar-human interactions defined as 'Level D-Attack' in W-20. These amendments allow additional flexibility and options for managing lower level cougar-human interactions as part of the research and management evaluation process.

Under the management guidelines of Directive W-20, section C, it is specified that any cougar that is tranquilized, handled and released by the Division under the authority of W-20 will be ear-tagged with the appropriate color tag for that region, and will be tattooed on the inside of the ear prior to release. All cougars captured for research purposes will also be ear-tagged with the appropriate color for the region using a tag code starting with an R followed by a three digit number. Cougars will only be tattooed on the inside of the ear if they would have been tranquilized, handled and released by the division under the authority of W-20 regardless of the associated research project. If tattooing does occur, the tattoo will match the code used on the ear-tag.

The purpose of the Front Range cougar project is to expand our understanding of how to better manage cougar-human interactions within the expanding suburban-rural human environment so that we can sustain both the existence of cougars and ensure public safety. For this study to succeed we must capture and radio-collar cougars that live in or near the suburban-rural environment to acquire basic information on cougar movements and prey selection and the potential for cougars to interact with humans. An inherent risk is that some radio-collared cougars will, at some point in time, likely interact to some degree with humans.

These management protocols will provide CDOW managers and researchers an initial menu of choices to consistently guide decisions involving interactions between radio-collared cougars and humans. Cougars will be radio-collared by capturing cougars during planned and systematic efforts or opportunistically during low-level human-interaction circumstances. Protocols address 5 major topics: A) radio-collared cougars, B) project communications, C) research data, D) external media, and E) cooperators awareness of ongoing proposed project protocols and study plan. These protocols will be a 'living document' that will evolve as the research project progresses with the input of field managers and researchers. Changes to the protocols will occur through informed discussions among CDOW managers and scheduled as needed as the research project unfolds or objectives are modified.

A. INTERNAL *CDOW* PROTOCOLS FOR MANAGING FRONT RANGE RESEARCH RADIO-COLLARED COUGARS

a. Cougar-Human Interaction Levels

Interactions involving radio-collared cougars and humans will span a potential range from benign to dangerous as depicted in the diagram below (**Levels I – V**).

I.. Cougar not seen, or detected, by public, nor near human dwellings or infrastructure.

II. Cougar sighted by public or passes near human infrastructure but no level of interaction between cougar and humans and not perceived as a safety concern.

III. Cougar seen by public or passes near human infrastructure with some low level of interaction between cougar and humans but no threatening behavior documented.

IV. Cougar seen by public or passes near human infrastructure with a level of interaction between cougar and humans reasonably considered to be threatening to humans.

V. Cougar seen by public or passes near human infrastructure with a level of interaction between cougar and humans considered to be dangerous to humans.

Defining the risk to humans that could be associated with observed cougar behaviors is difficult. We relied on the interpretations of cougar behavior as outlined in the Cougar Management Guidelines and adapted these interpreted levels of risk to our cougar-human interaction Levels 1 -5 (Table 1). Interpretations of cougar behavior would be highly dependent on the observer's skills and experience and the skills and experience of CDOW personnel who would interview the person who had the interaction with the cougar. In threatening or dangerous interactions (Levels 4 and 5), investigating personnel would attempt to determine whether the cougar was defending an animal carcass, kill site, den site, or young.

Table 1. Interpretations of cougar behaviors occurring during cougar-human interactions in order of increasing risk to humans. Columns 1-3, except for 'Attack" behavior, were copied from the 'Cougar Management Guidelines Working Group, 2005, Wild Futures Press' while column 4 represents Levels of Interaction as defined for these Front Range Cougar project protocols.

Human Observation of Cougar Behavior	**Interpretation of Cougar Behavior**	**Level of Likely Human Risk**	**Front-Range Cougar Risk Category**
Cougar opportunistically viewed at a distance	Secretive	Low	Non-threatening, Level 2
Cougar flight or hiding	Avoidance	Low	Non-threatening, Level 2 or 3
Cougar lack of attention, various movements not directed towards person.	Indifference or actively avoiding inducing aggression	Low	Non-threatening, Level 2 or 3
Cougar has various body positions, ears up, may be shifting positions, intent attention, following behavior	Curiosity	Low, provided human response is appropriate	Non-threatening to threatening, Level 3 or 4
Intense staring, following and hiding behavior	Assessing success of attack	Moderate	Threatening, Level 4
Hissing, snarling, vocalization	Defensive behaviors, attack may be imminent	Moderate depending on distance between human and cougar	Threatening, Level 4
Crouching, tail twitching, intense staring, ears flattened like wings, body low to ground, head may be up	Pre-attack	High	Dangerous, Level 5
Ears flat, fur out, tail twitching, body and head low to ground, rear legs "pumping"	Imminent attack	Very High and Immediate	Dangerous, Level 5
Cougar attempts to or actually strikes, claws, or physically comes into contact with human.	Attack	Extremely High	Dangerous, Level 5

An indirect interaction between humans and cougars involves cougars and domestic pets or livestock and such interactions do occur along the Front Range. There is the possibility that pet-cougar interactions may be a signal that a cougar may be inclined to eventually become involved in a cougar-human interaction. Similar to cougar-human interactions, we propose a gradient of cougar-pet/livestock interactions that would be assessed relative to the risk of these cougar behaviors to humans (Table 2). Key distinctions among cougar-pet/livestock interactions are whether the incident happened in an open space area and 'off-leash', within a confined area such as a fenced yard, within animal/livestock holding pen, or while the pet/livestock was on leash/halter and accompanied by a human. Definitions of domestic pet and domestic livestock will follow guidelines established for W-20.

Table 2. Interpretations of cougar behaviors occurring during cougar-pet/livestock interactions in order of increasing risk to humans.

Human Observation of Cougar Behavior Associated with Pet or Livestock	Interpretation of Cougar Behavior	Front-Range Cougar Risk Category when Occurs in Open Space or Similar Areas away from Dwellings	Front-Range Cougar Risk Category When Occurs in Confined Area or On Leash Accompanied by Human
Cougar seen in proximity to domestic pet/livestock	Secretive or possibly Curious	Non-threatening, Level 1	Non-threatening, Level 2
Cougar displays flight or hiding	Avoidance	Non-threatening, Level 1	Non-threatening, Level 2
Cougar approaches pet/livestock, displays various body positions, ears up, intent attention, following behavior	Curiosity or possibly assessing success of attack	Non-threatening, Level 2	Non-threatening, Level 3, providing human response is appropriate.
Hissing, snarling, vocalizations	Defensive behavior, or possible attack	Non-threatening, Level 2	Non-threatening, Level 3, or Threatening Level 4 if pet closely accompanied by a human
Crouching, tail twitching, intense staring, body near or low to ground, rear legs may be 'pumping'	Pre-attack or Imminent Attack	Non-threatening, Level 3	Non-threatening Level 3, or Threatening Level 4 if pet closely accompanied by a human
Cougar kills or injures pet	Attack Occurred	Level 3	Threatening Level 4, or Dangerous Level 5 if pet closely accompanied by a human
Cougar kills or injures livestock	Attack Occurred	Level 3	Threatening Level 4, or Dangerous Level 5 if livestock closely accompanied by a human

b. Decision Process for Evaluating Responses to Cougar-Human Interactions

Abbreviations in this section used in reference to CDOW personnel positions are: District Wildlife Manager (DWM), Area Wildlife Manager, (AWM), Regional Manager (RM), Wildlife Researcher (WR), Wildlife Research Leader (RL), Terrestrial Section Manager (TSM).

At any level of cougar-human interaction, the minimum Decision Response Team will consist of the primary WR, the area DWM, and the appropriate area AWM, unless immediate action is needed to benefit public safety whereby the AWM could act independently of the Decision Response Team. Input and options provided by all 3 of these persons will be assessed by the group which will attempt to reach a consensus decision. The Decision Response Team will objectively weigh the options available for each interaction/situation and make the most appropriate decision that considers the objectives of the research project while maintaining public safety. The decision will be a process of informed judgment. The AWM, or AWM designee, will be the official CDOW representative for the final decision. If the Decision Response Team cannot reach a decision of consensus, then the AWM will engage the RM, RL, and TSM in the decision process. At any level of response, any member of the response team may opt to consult with appropriate adjoining AWMs, RM, RL and TSM. The AWM will be responsible for forwarding situational and decision information to appropriate field personnel via internal email, phone, or via the Public Information Specialist. The CDOW Regional Public Information Specialist will be responsible for providing information to the CDOW Denver Public Information Specialist and the media.

As the level of cougar-human interaction increases from **Level 1 to Level 5**, the decision rationale shall shift and become more weighted towards public safety and preventing further cougar-human interactions as opposed to assessing or moderating cougar behavior. Decisions would therefore shift towards reducing imminent risks to humans.

Examples of Cougar-Human Interaction Decision Options:

Example situations representing radio-collared cougar interaction **Levels 1-5** and possible response decision options for responding to the interaction situation are presented below. The known history/behavior of a cougar in relation to levels of human interaction ***will weigh heavily*** on research/management decision options. We emphasize that the situations described below are not all encompassing. Furthermore, there may be rare situations where cougar-human interactions occur that prevent responsive management options because of extraneous factors such as access, snow conditions, or proper identification of the interacting cougar.

Level 1. A radio-collared cougar is known to remain in open space lands, utilize natural prey, and utilize areas near public trails based on radio-telemetry information but is not known to have been seen by the public or involved in any level of interaction.

Research/Management Options:

a. No management prescriptions are applied to the cougar.
b. 'Cougar In Area' signs may or may not posted on nearby public trails.
c. Aversive conditioning tactics are applied to the cougar consistent with the research study design.

Level 2. A radio-collared cougar is known to remain primarily in open space lands and utilize natural prey but is seen by the public near a public trail or is seen or is otherwise documented to occasionally be near human residences or businesses. Additionally, a cougar not previously radio-collared is seen by the public near a public trail or is seen or is otherwise documented to occasionally be near human residences or businesses

Research/Management Options:

a. No management prescriptions are applied to the radio-collared cougar.
b. The cougar is captured and radio-collared and subjected to management prescriptions consistent with the research study design.
c. "Cougar In Area" signs may or may not be posted on nearby public trails.
d. "Cougar In Area" signs are posted near human infrastructure. Persons living or working within affected human infrastructure are directly contacted by CDOW.
e. Aversive conditioning tactics are applied to the cougar consistent with the research study design. Aversive conditioning tactics may include; pursuing cougar with trained hounds, pepper spray application to cougar, or impacting cougar with rubber pellets fired from a shotgun.
f. Cougar is captured for the first time, or recaptured and relocated to an appropriate area of natural habitat consistent with the research study design. Relocation distances shall not be constrained by Directive W-20.

Level 3. A radio-collared cougar is known to use open space lands and areas having considerable human infrastructure. The cougar has been, or is likely to have been seen by the public on more than 1 occasion near human residences, businesses, or schools and there is reasonable concern for public safety but the cougar has not been perceived as exhibiting any current or past level of threatening behavior. Additionally, a cougar not previously radio-collared is known or likely seen by the public on more than 1 occasion near human residences, businesses, or schools and there is reasonable concern for public safety because of proximity, but the cougar has not been observed as exhibiting any current or past level of threatening behavior.

Research/Management Options:

a. No management prescriptions are applied to the radio-collared cougar but monitoring of cougar behavior is intensified by obtaining multiple telemetry locations per day and attempting multiple visual monitoring sessions per day.
b. The non-collared cougar is captured and radio-collared, subsequent behavior is closely monitored by obtaining multiple telemetry locations per day and attempting multiple visual monitoring sessions per day.
c. Warnings are posted or communicated to the appropriate public using signs or other media.
d. Newly collared or previously collared cougars could be subjected to management prescriptions consistent with the research study design.
e. Aversive conditioning tactics are applied to the cougar consistent with the research study design.
f. Cougar is recaptured and relocated to an appropriate area of natural habitat consistent with the research study design.
g. Cougar is recaptured, additional aversive conditioning tactics are applied to the cougar, and the cougar is relocated to an appropriate area of natural habitat consistent with the research study design.

Level 4. A radio-collared cougar is known to use open space lands and areas having considerable human infrastructure. The cougar has been or is likely to have been seen by the public on several occasions near human residences, businesses, or schools, or there is 1 documented interaction where the behavior of the cougar was reasonably considered to be somewhat threatening to humans but there was no evidence of attacking humans (such as cougar defending an animal carcass, kill site, den site, or young as demonstrated by snarling and vocalizing without stalking). Additionally, a cougar not previously radio-collared is known or likely seen by the public on several occasions near human residences, businesses, or schools, or there is 1 documented interaction where the behavior of the cougar was reasonably considered to be somewhat threatening to humans but there was no evidence of attacking humans (such as cougar defending an animal carcass, kill site, den site, or young as demonstrated by snarling and vocalizing without stalking).

Research/Management Options:

a. Warnings are posted or communicated to the appropriate public using signs or other media, and,
b. The non-collared cougar is captured and radio-collared, or if involving a previously radio-collared cougar, the subsequent behavior of either cougar is closely monitored by obtaining multiple telemetry locations per day and attempting multiple visual monitoring sessions per day.
c. Aversive conditioning tactics are applied to the cougar consistent with the research study design.
d. Cougar is initially captured and radio-collared, or recaptured, additional aversive conditioning tactics are applied to the cougar, and the cougar is relocated to an appropriate area of natural habitat consistent with the research study design.
e. If a cougar is involved in 1, Level 4 interaction and subsequently becomes involved in another Level 4 interaction, the cougar is euthanized.
f. Cougar is captured and euthanized.

Level 5. A cougar, whether previously radio-collared or non-collared, is involved in 1 interaction where the behavior of the cougar was highly threatening to humans or an attack of a human occurred.

Research/Management Options:

a. Actions follow protocols outlined in W-20, Level D-Attack. Attempts are made to capture the cougar and likely euthanize the cougar.

Cougars that must be euthanized will be necropsied by the Colorado State University pathology laboratory with reports provided to the Area Wildlife Manager, primary Wildlife Researcher, and the CDOW Wildlife Health Section. Remains of the cougar, such as head, hide, and tissue will be disposed of based on existing CDOW Regional guidelines with decisions the responsibility of the appropriate AWM.

B. INTERNAL *CDOW* PROTOCOLS MANAGING FRONT RANGE COUGAR RESEARCH COMMUNICATIONS

This research project will demand frequent and routine communication between Research, Terrestrial biologists, Area Field Operations personnel, and CDOW Regional and Denver Public Information Specialists. Timing of routine field activities such as baiting, trapping, capturing, and handling of cougars and monitoring of radio-collared cougars will be communicated frequently via email or phone in order to achieve coordinated success of such activities and to maintain informed local knowledge of radio-collared cougar behavior and whereabouts.

For cougar-human interaction concerns, the minimum communication tree will be the WR, DWM, AWM, RL, and Senior Terrestrial biologist responsible for the geographic area(s) containing the field research activities and/or inhabited by the radio-collared cougar. Communication should be by cell phone, communications radio, or email as needed for appropriate expediency. Frequency of communication will be decided mutually among these 5 persons. Behavior of individual cougars, and especially changes in behavior of cougars, may necessitate changes in frequency of communication.

As the potential for a cougar-human interaction increases from Level 1 to Level 5 as judged by the Decision Response Team based on acute or cumulated changes in cougar behavior or cougar location, communication frequency will increase, and ultimately communications will be a part of and dictated by the Decision Response Team. At any time the AWM or RL can expand the communications tree to include the TSM, RM, or other CDOW representatives but will also be responsible for sending the communications to these additional levels. The AWM will be responsible for disseminating appropriate information to other appropriate agencies or entities. The AWM will communicate with the Regional Public Information Specialist who will be responsible for coordinating activities with and providing information to the Denver Public Information specialist and the media.

C. INTERNAL *CDOW* PROTOCOLS FOR MANAGING FRONT RANGE COUGAR RESEARCH DATA

Because the cougar project will be high in profile and involves human safety issues, there will be a constant demand for information because of the perceived 'need to know' both by internal CDOW staff and the public. Finding the correct balance between the time spent obtaining information and the time spent distributing information, and to whom, will be a learning process and there will be real costs involved in personnel time. Furthermore, what information is distributed and to whom will be a learning process. The Decision Response Team shall clearly state that no 'real-time' data of cougar activity will be released, primarily because 'real-time' data capabilities will not be possible within the scope of the project.

Under current CORA guidelines, subject to legal interpretation, the raw, non-summarized data obtained during an on-going research project is protected from being obtained by the public via CORA. Examples of raw data would be the actual latitude-longitude or UTM coordinates of cougar locations or locations of critical den sites or kill-caches. Our intent is that this raw data would not be released to the public at-large, not only to protect the cougars as individuals, but also to protect our copyright on the data the agency has obtained. The current lynx reintroduction project sets a precedent for this approach with the caveat that lynx are a threatened species.

As part of the *internal-only* communication process and internal agency 'knowledge gathering' the WR will, once per month, provide the appropriate DWM, AWM, RL, and Senior Terrestrial biologist with e-maps (jpeg files) showing the distribution of radio-collared cougars in relation to important topographic and cultural features, so that these CDOW individuals are adequately aware of cougar locations and movement patterns. If cougar behavior changes such as to increase the likelihood of cougar-human interactions, monitoring of the cougar using VHF telemetry will be increased and frequency of internal communications will increase appropriately. The AWM and WR will work together to provide a reasonable frequency of *'internal-only'* information transfer with both individuals cognizant of the trade-offs between study objectives and needs and human safety issues. Cooperating public land-use agencies will be provided the same information on the same established schedule so as to keep these entities similarly informed.

The AWM, WR, RL, and the Regional Public Information Specialist will cooperatively discuss what type of information is released to the public and when such releases occur. However, as the public learns that CDOW has gained information about cougars in suburban-rural areas because CDOW radio-collared cougars and employed GPS collars and can map detailed cougar locations, post-event, CDOW can expect a variety of demands for information that will need to be addressed and a rising challenge as to how often and in what detail information is provided. The WR will provide a written report by 1 October summarizing the progress of the research project on an annual basis to Area and Regional personnel. This report will be available to the public through our standard Wildlife Research Report distribution process.

D. EXTERNAL COMMUNICATIONS

The Front Range cougar research project will attract the interest, curiosity, and involvement of the media such as newspapers, magazines, radio, and television. Appropriately interacting with the media will be important to maintaining credibility with the public and with providing educational opportunities to the public. Requests by the media for involvement with the research project should be assessed as consistently and appropriately as possible by the Decision Response Team. The Decision Response Team shall clearly state that no 'real-time' data of cougar activity will be released, primarily because 'real-time' data capabilities will not be possible within the scope of the project. We propose that requests be assessed as a dichotomy of cougar-human 'non-incident' and 'incident' requests (Table 3).

Table 3. Guidelines for responding to requests from the media.

Media Involvement Request	**Request Associated with Non-Interaction Cougar-Human Activity**	**Request Associated with Cougar-Human Interaction**
Field Trip on Project Activities	Media schedules time with CDOW Field Personnel; Activity will not jeopardize key field activities such as capturing & handling cougars or create unnecessary safety issues. Researcher identifies most appropriate time for activity to the Decision Response Team. Decision Response Team will notify Regional Public Information Specialist.	Likely Not Appropriate, Follow W-20 Guidelines-
Filming or Photographing Project Activities	Filming/photography to be done by CDOW information specialists who will provide footage/photos to media for media use. Filming coordinated by Decision Response Team. CDOW retains right to review all footage/photos prior to release whether provided by CDOW or private media. Decision Response Team will notify Regional Public Information Specialist.	Follow W-20 Guidelines
Interview of Project Personnel	Requests for interviews of project personnel will be relayed to the Decision Response Team whenever possible. Interviews will occur to minimize interrupting routine project activities. As a minimum, the RL and the AWM will be notified of the request to conduct the interview. Decision Response Team will notify Regional Public Information Specialist.	Follow W-20 Guidelines with the exception that questions pertaining to research project objectives, research results, and research protocols will be deferred to the Decision Response Team for accurate answers.

E. DOCUMENT COOPERATORS AWARENESS OF FRONT-RANGE COUGAR RESEARCH PROJECT

We recommend that representatives of cooperating entities, such as, Boulder County Parks and Open Space, Jefferson County Open Space, and City of Boulder Open Space and Mountain Parks be made aware of these protocols and the CDOW approved research study plan that will guide this project.

______________________________________ _____________

Bruce L. McCloskey, Director — Date
Colorado Division of Wildlife
Approval to Implement These Protocols for
Front Range Cougar Research Project

APPENDIX II

CAPTURING AND HANDLING PROCEDURES FOR FREE-RANGING COUGARS

Modified by MWA on 1/18/2007: Puma changed to cougar and schedule for monitoring cage traps modified after consultation with CDOW veterinarian L. Wolfe.

Delivery of anesthetic drugs via projectile syringe or jab pole, cage traps, or foot snare may be used to capture cougars. All of these techniques have proven effective and safe for capturing cougars under field conditions commonly encountered in Colorado. This document is intended to serve as a comprehensive reference for future cougar studies to avoid unnecessary complexity in study protocols submitted for ACUC review.

CAPTURE TECHNIQUES

Trained hound pursuit

As described in Shaw (1979), an experienced houndsman with trained dogs is used to track and tree or bay each cougar. Field anesthesia is determined under the supervision of the attending veterinarian. Anesthetic drugs will be administered intramuscularly (preferably the caudal thigh) via projectile syringe using a gas-powered projector. For capture, cougars will be anesthetized with Telazol® (6-9 mg/kg) and xylazine HCl (1.8-2 mg/kg) or ketamine (10-11mg/kg) and xylazine HCl (1.8-2mg/kg) or ketamine (2 mg/kg) and medetomidine (0.075 mg/kg) (Shaw 1979, Logan et al. 1986, Kreeger 1996). See drug dosages below (Table 1, Appendix II).

If the cougar is treed, then people and dogs should be removed from the immediate area to give the animal a chance to descend before becoming completely anesthetized. If the cougar remains in the tree until almost completely anesthetized, then someone wearing climbing gear will climb to the cougar and attach either a chest harness (preferred) or hind leg noose (e.g. bovine hobbles) to 2 legs and quickly lower the animal to the ground. If possible, other personnel will hold a taunt net, 3 by 3 meters square, below to break the cougar's fall should it slip before a harness or rope can be secured. If there aren't enough people to hold the net, anchor the net about 2 m above the ground and on adjacent trees or branches using ropes & carabiners.

Occasionally cougars will jump from the tree immediately after being darted. If there is snow cover, the cougar should be tracked with the dogs on leads. Attention should be given to changes to the cougar's gait and direction of travel. When anesthesia is effective, the cougar's tracks will weave and show signs of stumbling. Usually the cougar can be found laying or sitting on top of the snow. If after 15 minutes, it appears that the cougar is traveling normally, then dogs can be released on the cougar's tracks again to encourage it to tree. If the ground is bare, then at least one non-aggressive dog can be released on the cougar's trail to drive it to bay. If the cougar is radio-collared, radio-telemetry can be used to track the cougar.

Upon first approach of an apparently anesthetized cougar, a 4-5 foot stick will be used to gently prod the paws and muzzle of the animal; if there is no response (i.e. snarling or biting), then assume anesthesia is sufficient for handling. Once anesthetized apply an antibiotic or mineral oil based eye ointment and a blindfold to reduce visual stimuli and protect the eyes from bright sun light and debris.

Vital signs should be monitored in the anesthetized cougar. Normal signs: pulse ≈ 70—80 bpm, respiration ≈ 20 bpm, capillary refill time ≤2 sec., rectal temperature ≈ 101°F average, range = 95—104°F (Wildlife Restraint Handbook, 1996, California Dep. of Fish and Game, Wildlife Investigation Laboratory, Sacramento, Kreeger et al. 2002). In temperatures near or at sub-freezing wrap the anesthetized cougar in a thermal blanket. In hot temperatures, the cougar should be treated with water on the head, abdomen, and inguinal area. Cougars that receive lacerations during capture will be given antibiotics. When the cougar is being sampled it should be moved from one side to the other or in sternal recumbency about every 10 minutes to prevent hypostasis in the downside lung.

When sampling procedures are completed, the blind-fold and leg restraints (e.g. hobbles or snare cable) will be quickly removed, and the cougar will be allowed to recover from the sedation either naturally or with the aid of an antagonist. When prescribed, yohimbine HCl (0.125 mg/kg IV) will be used to antagonize xylazine sedation and atipamezole (0.3 mg/kg) will be used to antagonize medetomidine sedation.

Cage trapping

A cage-type trap for live capture of bears was developed by Beck (1993). The trap measures 1.8m long and 1.0m high and wide. The frame is constructed of angle iron, and all side and top panels are wire mesh of 1.9cm mesh size. The floor is 16-gauge steel. A spring-powered, solid aluminum door is mounted on a full-length hinge at one end. A full-length latching mechanism holds the door closed. The door is triggered via a treadle pedal mounted on the floor 1.0m from the door. A standard garage door coil spring provides the closing power. Along one side of the trap is a hinged panel measuring 1.8m by 0.3m. Vertical bars placed on 0.3m centers behind this panel. Swinging the window up allows access through the barred area for administering immobilizing drugs by jabpole. Each trap weighs approximately 236kg.

In the first study in which these traps were used, there was only one injury to a bear in 134 captures. An adult male broke a canine tooth while in the trap. Of the limited number of times these trap have been used for cougars, no known injuries have occurred to date (T. Beck, pers. comm.).

A cage trap designed specifically for the capture of cougar has been used to manage cougar human conflicts in California since the late 1980s (Shuler 1992). A similar cage trap was used to safely capture cougar for research on cougar human interactions in San Diego County, California (Sweanor et al. *in prep*.). The cage trap for that study measured 48 in. tall, 40 in. wide, and 10 ft. long. It was built on a frame of 1 ½ in. angle iron with 2 in. by 4 in. grid horse panel made of 3/16 in. welded steel rod for the walls, floor, door, and roof. It weighed about 250 lb (113kg).

A cage trap was designed by Don Hunter (USGS) and Colorado State University's Mechanical Engineering Department. The trap was designed to be smaller, lighter, collapsible, and safer than what was previously available. A counter-weighted door drops closed slowly and quietly so as not to injure any members of a family group caught in the doorway. In addition, there are air-pressured cylinders that slow the door even further and a rubber bumper along the edge in case a tail is caught in the way of the closing door. The trap is 3.5 ft. tall, 3.5 ft. wide and 6 ft. long, constructed of 2 in. by 4 in. grid pattern steel horse panel with 0.225 in. rod.

A cage trap will be baited with a deer carcass that will be tied to the end mesh panel opposite the door of the trap. The trap will be checked as early as possible the following morning or immediately after a capture occurs if fitted with a transmitter to be triggered upon closing of the door. The researchers should monitor the trap as soon as possible after sunrise every morning to minimize time in the trap and to avoid human interference from recreational activities. Normally, when a cougar has claimed a bait at a cage trap, it is caught fairly soon after night-fall. Researchers can work the cougar with a spot-light, head lamps, and lantern. Cougars will be immobilized with a jabpole or syringe as described above. Drug dosages and animal handling will be as described above.

Foothold Snares

Foot-hold snares are an effective, relatively safe technique for capturing cougars particularly in areas not conducive to using trained hounds (Logan et al. 1999). The snares are constructed to minimize injuries to the cougar. The snare, also called the Aldrich foot snare, was originally designed for the capture of bears. It has been modified to use for cougars. The spring activated snare secures a 3/16 inch steel cable around the foot of the cougar, closing tight with the action of a small piece of angle iron fashioned into a sliding lock mechanism. The snares have been modified considerably over the years for cougars by incorporating a large spring to diminish force applied to the foot and a shock absorption device into the cable. The inside of the loop is wrapped with duct tape to minimize the surface abrasion on the skin of the foot. An in-line swivel is placed in the cable to avoid torsion of the foot and potential bone fracture. A short lead is attached to the snare to further minimize stress to the leg. The lead is then secured to a multi-branched flexible bush with a double off-set hook drag made of 5/8 in. rebar steel. It can also be secured to a tree 4 inches or greater in diameter with 3/16 in. or ¼ in. steel cable clamped and stapled to the base of the tree so the cougar can not climb the tree with the snare. Branches of the tree are lopped of with a saw or an axe about 8 ft. up the tree so the cougar can not hang itself from a branch by the snare cable. An area of 5 meters or more is cleared around the snare site to eliminate potential leg fractures resulting from a fulcrum situation in conjunction to an adjacent tree (Duggins Wroe, pers. comm.) or torque on the leg bones caused by revolutionary twisting of the cable when the swivel is isolated by the foot-loop cable becoming wrapped around stout vegetation. Details on how to safely structure the snare and to choose and prepare snare sites are in Logan et al. 1999.

Modifications have been made to avoid capturing non-target animals. The concealed 10 inch loop of cable is positioned over the trigger of the spring. The trigger has a 4 inch plastic trap pan adhered to the top surface. The pan and trigger are positioned over a hole dug in the ground and filled with a 12x12x4 inch piece of high density foam. This foam prevents smaller animals from triggering the snare. Large branches are angled over the snare to force ungulates to step over or go around the snare. The duct tape on the loop keeps it from closing too tightly and usually allows smaller-footed animals such as ungulates, coyotes and bobcats to slip free. The loop size is set smaller than for a black bear, there is, however, a possibility of catching a smaller-footed black bear (Duggins Wroe, pers. comm.). Bears will be drugged and released if caught. Any other non-target animals caught will be examined and treated for injuries and released with snare poles.

Preferred sites will have limber bushes with multiple basal stems to securely anchor the snare drag, and a safety area with a circumference 5 m or more around the anchor point. The snares will not be set near cliff or water, and potentially dangerous vegetation will be cleared from the safety area. Snares will be checked as quickly as possible after sunrise every morning to reduce stress and possibility of hyperthermia. Snares will be checked at least twice a day and will not be reset on extremely hot days (Logan et al. 1999, Logan and Sweanor 2001). Logan et al. (1999) found snaring to be a relatively safe technique for capturing cougars. Life-threatening injuries occurred in 5 of 209 captures. The majority of these injuries were fractures to ulna and/or radius of the snared leg.
Adult cougars will be immobilized with anesthetics delivered by jabpole or CO_2 pistol and projectile syringe as described above. Capture operations will be halted if ambient temperature falls below 0°F or rises above 90°F.

Delivery of anesthetic drugs via projectile syringe

In situations where pursuit by hounds is not possible and snaring or trapping is difficult due to the high abundance of non-target animals, a lure may be used to bring a cougar in close proximity to dart with a projectile syringe using a gas-powered projector. Lures may include a fresh kill made by the target animal, a deer carcass placed out as bait, or a predator call. A hound on a lead will be available to track the animal once it has been darted. The caudal thigh is the preferred target for the dart. The anesthetic choice is at the discretion of the attending veterinarian.

Hand capture of cubs

Nursling cougar cubs can be safely captured by hand or with a catch-pole at nurseries when they are 4 to 10 weeks old (Logan and Sweanor 2001). Cubs usually weigh less than 10 kg, and can be examined and tagged without the need for anesthetics. Nurseries can be located when VHF-collared mothers are present, or by using GPS data from GPS-collared mothers. Wait for a time when the mother is away from the nursery, as determined by VHF-radio-telemetry, in order to capture the cubs. Cubs should be handled with clean leather gloves. They can be picked up by the nape of the neck. A catch-pole may be necessary to extract cubs from holes and crevices. Cubs should be contained together, or in pairs, in new burlap bags to allow ample air circulation. The cubs should be moved about 100 m from the nursery to minimize human activity, disturbance, and odors at the nursery. Individual cubs that are being examined can be held in a separate burlap bag. Once the cubs are processed, they should be returned to the exact nursery, and the researchers should leave the area immediately.

Throughout this process a receiver tuned to the frequency of the radio-collared mother should be constantly monitored. If it appears that the mother is returning, the cubs should be put back in the nursery immediately, and researchers should vacate the area.

INJURIES AND EUTHANASIA

If an animal is seriously injured (e.g. fractured or broken appendage, vertebrae, pelvis, or jaw, severe dislocation, laceration or any other injury that compromises its ability to survive and/or causes severe pain or distress) during capture or recovery, then it will be quickly and humanely euthanized. Cougar will be deeply anesthetized with ketamine or Telazol® and xylazine (IV or IM) and euthanized via rapid IV KCl administration (400-800 mEq). Alternatively, if an injured cougar cannot be handled then euthanasia will be a gunshot to the head or neck with a 0.22 caliber magnum rifle or pistol.

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DRUG DOSAGE FOR COUGARS

Table 1: Drug dosage by weight for cougars as recommended by CDOW veterinarian L. Wolfe.

		Dosage mg/kg	Conc mg/ml	Cougar Dose (ml) by animal weight (kg)									
				10	**20**	**30**	**40**	**50**	**60**	**70**	**80**	**100**	**110**
ANTIBIOTICS													
Oxytetracycline		3	200	0.2	0.3	0.5	0.6	0.8	0.9	1.1	1.2	1.5	1.7
Penicillin G	SID	20000	300000	0.7	1.3	2.0	2.7	3.3	4.0	4.7	5.3	6.7	7.3
PAINKILLERS													
Ketoprofen	SID	2	100	0.2	0.4	0.6	0.8	1.0	1.2	1.4	1.6	2.0	2.2
ANESTHETICS													
ketamine (+ med) 200		2	200	0.1	0.2	0.3	0.4	0.5	0.6	0.7	0.8	1.0	1.1
medatomidine 20		0.1	20	0.1	0.1	0.2	0.2	0.3	0.3	0.4	0.4	0.5	0.6
tolazoline		4	100	0.4	0.8	1.2	1.6	2.0	2.4	2.8	3.2	4.0	4.4
atipamazole		0.3	5	0.6	1.2	1.8	2.4	3.0	3.6	4.2	4.8	6.0	6.6
Dopram		1	20	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	5.0	5.5
Atropine		0.03	0.5	0.6	1.2	1.8	2.4	3.0	3.6	4.2	4.8	6.0	6.6
MISC													
fluids maint ml/ day		60	1	600.0	1200.0	1800.0	2400.0	3000.0	3600.0	4200.0	4800.0	6000.0	6600.0

Colorado Division of Wildlife
July 2006 – June 2007

WILDLIFE RESEARCH REPORT

State of Colorado : Division of Wildlife
Cost Center 3430 : Mammals Research
Work Package No. 7210 : Customer Services/Research Support
Task No. 1 : Library Services

Federal Aid Project: N/A :

Period Covered: July 1, 2006 – June 30, 2007

Author: D. J. Freddy

Personnel: J. A. Boss.

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ABSTRACT

After providing 17 years of professional library services for the entire Colorado Division of Wildlife, research librarian Jackie Boss retired in April 2007. Consequently, the standard detailed listing of accomplishments, publications, and services provided by the library was not available for this FY 06-07 report. We anticipate hiring a replacement librarian in Fall 2007 to continue and expand the services provided by the research library to the entire Colorado Division of Wildlife.

WILDLIFE RESEARCH REPORT

COLORADO DIVISION OF WILDLIFE RESEARCH LIBRARY SERVICES

D. J. FREDDY, J. A. BOSS

P.N. OBJECTIVE

Provide an effective support program of library services at minimal cost through centralization and enhancement of accountability for Colorado Division of Wildlife (CDOW) employees, cooperators, wildlife educators, and the public.

SEGMENT OBJECTIVES

1. Continue to improve and modernize library services by implementing the SirsiDynix Horizon library automation system via an Application Service Provider (ASP) model (project began in June 2002). By joining the Automation System Colorado Consortium (ASCC) we were able to take advantage of a LSTA grant written by the Colorado State Library staff, which facilitated the implementation of this system.

2. Continue to develop, improve, and implement the CDOW Research Center Library web-site (started in June 2004) by implementing the SirsiDynix horizon system online to serve broader spectrum of patrons of the CDOW Research Center Library.

3. Continue to attend ASCC meetings and participate in SirsiDynix Horizon online classes to enhance utilization of the SirsiDynix system.

SUMMARY OF LIBRARY SERVICES

The library continued to provide the following services:

- Maintain and Build Electronic Catalogs of all Research Library Holdings
- Acquire Publications for the Research Center Library
- Receive Publications Donated to the Research Center Library
- Acquire AV Materials for the Research Center Library
- Acquire Theses, Dissertations, Documents and Books through Interlibrary Loan
- Conduct Literature Searches and Deliver Information to Employees
- Archive CDOW Published Manuscripts

Prepared by ____

D. J. Freddy, for Jacqueline A. Boss, Librarian