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Wolf Monitoring Protocol for Denali National Park and Preserve, Yukon-Charley Rivers National Preserve and Wrangell-St. Elias National Park and Preserve

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ON THE COVER
John Burch and Tom Meier collecting data and samples from an anesthetized wolf in Denali National Park and Preserve.
Photograph by helicopter pilot Troy Cambier

Wolf Monitoring Protocol for Denali National Park and Preserve, Yukon-Charley Rivers National Preserve and Wrangell-St. Elias National Park and Preserve, Alaska

Natural Resource Report NPS/CAMN/NRR—2009/168

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Executive Summary

This paper introduces a wolf monitoring protocol for the Central Alaska Network. This network consists of three units of the U. S. National Park system: Denali National Park and Preserve, Yukon-Charley Rivers National Preserve, and Wrangell-St. Elias National Park and Preserve. The Central Alaska Network has identified Fauna Distribution and Abundance as one of its top three vital signs for tracking major drivers of ecosystem change. Wolves (*Canis lupus*) occur in all three network parks. The primary objectives of wolf monitoring will be to track the distribution and abundance of wolves using the Standard Operating Procedures, found in the appendices to this paper. Additional data obtained in the monitoring process will have great value for wildlife management and research.

Wolves are a difficult species to monitor. They are inconspicuous, and live at low density in a structured population of territorial packs. Effective monitoring of wolf populations needs to address not only their numbers, but the spatial structure of the population. Radio-telemetry is the most effective means of studying whole populations of pack-dwelling wolves. This allows for year-round tracking of wolves, including the identification of den and rendezvous sites, the characterization of pack movements across park boundaries, and information on the incidence and causes of wolf mortality. By capturing and examining wolves, morphological, genetic and disease information is obtained that is not available by other means.

Wolves occupy habitat on a coarse scale. A 10,000 square kilometer monitoring area conforms to this scale and provides a reasonably sized sample of the local wolf population. The actual survey area will vary from year to year, depending on the movements of collared packs. The initial survey from a fixed-wing aircraft provides a "snapshot" of wolf numbers and sizes of packs in the area. Full implementation of the monitoring protocol requires that two to three wolves from each pack be radio-collared, involving aerial darting by helicopter. Aerial surveys and radio-tracking have special considerations for equipment, permitting and training. A limited array of samples is collected from each wolf during collaring.

Oversight of this program involves data management, analysis and reporting, tracking of personnel certifications and training, and the operational requirements of personnel time, facilities and equipment. As the program evolves over time, documented changes will be made to the narrative of this paper, the field data forms and the Standard Operating Procedures in the Appendices.

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This protocol was informally reviewed by Dr. Layne Adams and Dr. L. David Mech of the U. S. Geological Survey. Drs. Mech and Adams were also the principal investigators for the Denali Wolf Project at the time of its founding in 1986. Al Lovaas and John Dalle-Molle of the National Park Service were instrumental in initiating and funding this work. Over the last 23 years, Denali's park superintendants and staff, and more recently the staff of the Central Alaska Network, have facilitated the project in innumerable ways. Finally, wildlife research and monitoring in Alaska are uniquely dependent on the skilled helicopter and airplane pilots who make the work possible and safe.

1 - Introduction: Background and Objectives

1.1 Issue being Addressed and Rationale for Monitoring Wolf Populations

This narrative is designed to introduce a wolf monitoring protocol for the Central Alaska Network (CAKN), which contains three units of the U. S. National Park system: Denali National Park and Preserve (DENA); Yukon-Charley Rivers National Preserve (YUCH); and Wrangell-St. Elias National Park and Preserve (WRST). This narrative is accompanied by six standard operating procedures (SOPs) that describe in detail the techniques to be used in monitoring wolves. An additional three SOPs describing techniques of genetic analysis, while part of the overall monitoring plan, are not included here in order to concentrate on the techniques directly related to monitoring wolf numbers and distribution. Further supplemental materials that contribute to the protocol are found in Appendix C.

CAKN has adopted a holistic view of network ecosystems and will track the major physical drivers of ecosystem change and the responses of the two major components of the biota, plants and animals. Thus, CAKN has identified Fauna Distribution and Abundance as one of its top three vital signs. In general, CAKN wants to know where fauna are distributed across the landscape and to track changes in both their distribution and abundance. The Fauna Distribution and Abundance vital sign includes monitoring efforts for a suite of vertebrate species spanning the significant elevation gradient found in CAKN parks, and also including species of specific interest within each park. Wolves (*Canis lupus*) occur in all three network parks and are one of six keystone large mammal species in interior Alaska. Wolves are of great importance to people, from both consumptive and non-consumptive viewpoints, and to the ecosystem as a whole. As a top predator, wolves may play a key role in influencing ungulate populations, and as a result may influence vegetation patterns (Gasaway et al. 1983, Miller et al. 2001, Ripple and Beschta 2003). The effects of wolves on ungulate populations may be important determinants of ungulate availability for subsistence harvest on National Park Service (NPS) park and preserve lands in Alaska, and harvest by the general public on NPS preserve lands (National Park Service 2003).

Wolves are a species specifically identified in the enabling legislation and management objectives of all three CAKN parks (U. S. Congress 1980). Wolves are of great importance to park visitors because of the unique opportunities to view wolves in Alaskan parks. While the primary objectives of monitoring will be to track the distribution and abundance of wolves, a variety of accessory data will be obtained in the monitoring process that are likely to have great value for wildlife management and research. The body of data on wolf populations in Alaska parks is of great value in developing scientific models of predator/prey systems. In heavily visited portions of the parks, managers may want to know the locations of active wolf dens and rendezvous sites so that they can be protected from disturbance. When intensive wolf harvest or wolf control take place near parks, it is vital to know the patterns of travel of park packs, in order to determine whether they are being significantly impacted by activities outside of the parks. Data on the genetic and morphological characteristics of wolves, obtained as a sidelight to wolf capture, will be important in evaluating long-term changes in wolf populations in Alaska.

1.2 Historical Development of Wolf Monitoring

Scientific studies of wolves began with ground-based observational studies, including the landmark research of Adolph Murie in Denali (Murie 1944). Snow-tracking of wolves with aircraft provided the next great step in understanding wolf populations, as packs could be tracked and observed to study their travels, predation patterns, and behavior (Burkholder 1959, Mech 1966, Haber 1977). Without a systematic means of locating wolf packs, only a few packs could be intensively studied by snow-tracking. Only with the development of radio-telemetry was it possible to track wolves on a population-wide basis (Mech and Frenzel 1971, Mech et al. 1998, Theberge and Theberge 2004). Radio-telemetry studies remain the standard technique for monitoring and research on wolf populations (Boitani 2003, Mech and Barber 2002).

1.3 Measurable Objectives

- Locate wolf packs in the designated survey area by winter snow tracking.
- Determine the makeup (numbers, colors, age structure) of surveyed wolf packs.
- Capture and radio-collar representatives of each wolf pack identified in the study area.
- Obtain morphological measurements from captured wolves.
- Obtain genotypic data (mitochondrial and microsatellite DNA analyses) from captured wolves.
- Obtain immunological (disease exposure) data from captured wolves.
- Define territories of collared wolf packs by radio-tracking.
- Define pack territory mosaic, perform additional track searches and captures to fill gaps.
- Determine pack sizes for collared packs in early winter and late winter each year.
- Calculate wolf density over the survey area for early winter and late winter each year.
- Determine number of pups in each pack in spring (if possible) and early fall.
- Perform annual capture efforts to maintain coverage of radio collars in the population.
- Detect pack extinction and pack formation events in the population.
- Detect changes in wolf density over time.
- Detect changes in wolf pack size over time.
- Detect changes in wolf territory size over time.
- Detect changes in the morphological and genetic makeup of the wolf population over time.

2 - Sampling Design

2.1 Rationale for Selecting this Sampling Design Over Others

In most habitats, wolves are a difficult species to monitor. They are inconspicuous, and live at low density in a structured population of territorial packs. In Alaska, wolves live at particularly low densities, and in most areas human access is limited to aerial, rather than ground travel. If wolf pack territories were regular and predictable, wolf surveys would be much easier, but it is seldom possible to predict the arrangement of pack territories from studying the terrain. Wolf pack territories overlap one another and change over time. Effective monitoring of wolf populations needs to address not only their numbers, but the spatial structure of the population. Radio-telemetry is the most effective means of studying whole populations of wolves (Boitani 2003, Mech and Barber 2002). Population estimates obtained by radio-telemetry should include all wolf packs in the survey area, and thus are a census of the population, not a survey or an index to wolf numbers. Disadvantages of radio-telemetry include its cost, and its level of intrusiveness through aircraft use, animal capture and collaring. Radio-telemetry studies of wolf packs tend to ignore the transient part of the wolf population, made up of wolves that have dispersed away from their packs. In nearly 20 years of wolf research at Denali, only one territorial, solitary-living wolf has been found in the park. Nearly all "lone wolves" are wolves that are in transition between one pack-living situation and another. The larger pack sizes seen in early winter represent the high point of numbers of wolves traveling together in most packs, with the addition of that year's pups increasing the pack size prior to its diminution by dispersal and mortality. There is probably a greater proportion of dispersing wolves left uncounted in the population in late winter than early winter. It should be specified that our wolf population estimates are for pack-dwelling wolves, with the understanding that the numerical and biological significance of wolves that are temporarily between packs is not expected to be great.

Other methods of counting wolves include track surveys and genetic studies. Track surveys (Burkholder 1959, Ballard et al. 1995), based on local knowledge and targeted searches, can locate wolf packs readily, providing estimates of pack sizes and the number of packs in an area. More rigorous, random-transect methods (Becker et al. 1998) can estimate wolf numbers with confidence intervals over large areas, but are themselves expensive, and are highly dependent on favorable snow conditions immediately followed by favorable flying conditions, and on the experience of pilots and observers. A wolf monitoring program based on snow-track surveys would not be able to generate population estimates on a predictable basis. Snow-track surveys provide only a snapshot of a wolf population, rather than a picture of its structure. Alaskan wolf packs frequently travel outside their usual territories, so that the pattern of pack occupancy seen in a one-time survey may be unrepresentative of the usual state of the population. If park staff have some prior knowledge of wolf activity in a limited area, targeted snow-tracking, to determine pack sizes, may be more useful than random track surveys.

Genetic sampling has been suggested as a means of surveying wolf populations (Person and Russell 1999, Darimont and Pacquet 2002, Creel et al. 2003). It is difficult to extract DNA from carnivore feces (Talbot et al. 2004), and collection of scat or hair samples across large areas of interior Alaska would not be practical. Techniques for estimating wolf population size from genetics-based mark/recapture techniques are not a reasonable option in this area. Genetics sampling of captured wolves is, however, likely to provide a wealth of information on population structure and long-term demographic patterns.

The additional benefits of a survey method that entails year-round tracking of wolves include the identification of den and rendezvous sites, the characterization of pack movements across park boundaries, and information on the incidence and causes of wolf mortality. By capturing and examining wolves, we obtain morphological, genetic and disease information that is not available by other means.

2.2 Site Selection

An area of up to 10,000 square kilometers will be selected in each park where wolf monitoring is to take place. An area of this size is likely to encompass the territories of about 10 wolf packs, monitoring a large proportion of the resident wolf packs in the park. Monitoring a cluster of wolf packs makes it easier to delineate pack territories, and provides economies of scale that make monitoring more efficient. It is desirable that wolf monitoring be co-located with monitoring efforts for other large mammal species, when practical. The area chosen for wolf monitoring should be designed to fit the management needs of the individual park. Monitoring areas with a compact, more nearly circular shape will facilitate wolf monitoring because they will include more packs which abut one another, rather than extending outside the designated survey area (Burch 2001).

2.3 Population being Monitored

The selection of a fairly large (10,000 square kilometer) area in which to concentrate wolf monitoring efforts is designed to conform to the fairly coarse scale with which wolves occupy their habitat, and to provide a reasonably sized sample of the local wolf population. The sizes of individual wolf packs vary stochastically, depending on the behavior and fates of individual wolves. Monitoring must take place at the level of a cluster of packs in order to get a reasonable picture of population-level processes. The scale of monitoring proposed here attempts to strike a balance between the need for a large, representative sample of a wolf population and the practical and budgetary constraints that argue for a more limited effort. Because prey populations, weather conditions and management regimes vary widely across Alaska, no monitoring effort will be representative of wolf populations across the state, or across the CAKN monitoring network.

The monitoring area will initially be surveyed by snow-tracking from fixed-wing aircraft, in order to locate and identify wolf packs for collaring. The initial survey provides a "snapshot" of wolf packs in the area, and the sizes of wolf packs present, even if no further work is done. Obtaining the pack sizes of several wolf packs can provide an index to wolf numbers, at fairly low cost. Wolf densities depend on pack size and on the number of wolf packs occupying an area (Mech et al.1998), so observations of pack size alone provide only a rough index of wolf numbers. Full implementation of the monitoring protocol requires that all wolf packs within the designated monitoring area be located and radio-collared. Two to three wolves from each pack will be collared. Establishing a wolf monitoring effort is likely to be an iterative process, where the territories of collared wolf packs are determined, gaps are identified where uncollared packs may live, and further collaring efforts are made to fill in the gaps. Dominant, breeding adults will be targeted for capture when possible, because they are less likely to disperse away from the pack and require further capture to replace collars. Inevitably some collared wolves will disperse, and these can be used to help characterize the transient portion of the population that is not accounted for by collaring pack-dwelling wolves. Estimates of wolf numbers and wolf density will be over the aggregate area of all pack territories in the survey area. Thus, the actual survey area will vary from year to year, depending on the movements of collared packs. This biologically-based survey area is more realistic than choosing an arbitrary boundary and estimating wolf numbers within an area that contains various fractions of the territories of many wolf packs.

2.4 Sampling Frequency and Replication

Collared wolves will be located approximately twice per month, with additional monitoring flights added in early and late winter to get pack counts. Each year, early winter and late winter estimates of wolf numbers and wolf density will be made. Ongoing protocol development includes the addition of Global Positioning System (GPS) collars programmed to take one location per day and upload the data through the ARGOS satellite system. The use of GPS collars will greatly facilitate the determination of pack territories, so that aerial tracking flights may be reduced and concentrated on those periods of the year when packs may be sighted and counted. The mortality and dispersal of collared wolves requires that collars in the various packs must be replaced on an ongoing basis. Capture of wolves for radio-collaring will be done annually in early and late winter, with limited capture efforts in the remainder of the year as needed. More frequent capture operations help prevent data gaps caused by loss of contact with packs, and help eliminate the need to re-locate lost packs by snow tracking.

2.5 Detectable Level of Change

Wolf densities in DENA have been found to fluctuate over at least a threefold range, depending on prey numbers, weather and other factors (Mech et al.1998). Thus, it is challenging to identify truly unusual changes in wolf demography. The mortality data from collared wolves, along with genetic, disease, and behavioral information gathered in the course of monitoring, can provide clues to the causes of population changes. Methods will be developed to characterize long-term trends in wolf populations. A history of wolf monitoring data (since 1986 in DENA, and since 1993 in YUCH) will be included in the analyses for a considerable head start in developing such methods.

3 - Field Methods

3.1 Preparation and Equipment Setup

A number of procurement and regulatory steps will be taken in advance of wolf monitoring efforts. Radio frequencies for radio collars will be cleared through the National Wildlife Telemetry Frequency Management Database, and the National Park Service's Radio Coordinator. Minimum equipment for a wolf monitoring study will include radio collars (at least two per pack in the intended study area), at least one telemetry receiver, antennae, cables, wing strut brackets, and switchbox for aerial telemetry, a dart rifle, darts, internal and external dart charges and drugs, and a well-stocked capture kit. A complete list of capture and monitoring equipment and a list of suppliers can be found in SOP #1. Under the Controlled Substances Act, only a person with a Drug Enforcement Agency (DEA) registration number may purchase Telazol and other controlled substances. Appropriate Personal Protective Equipment (PPE) for fixed-wing and helicopter special-use flights must be obtained for field workers.

All aircraft and pilots must be carded for the mission and procured according to NPS, United States Department of Interior (USDI) Aviation Management Directorate (AMD), and USDI Interagency Helicopter Operations Guide (IHOG) (U. S. Department of the Interior 2006), regulations. Animal handling personnel must become certified for the Aerial Capture, Eradication and Tagging of Animals (ACETA) (U. S. Department of the Interior 1994), as well as obtaining appropriate training in animal anesthesia, handling techniques and aircraft safety. Flight plans and flight following according to Federal Aviation Administration (FAA) and NPS regulations are important for safe operations. Clothing and equipment specifications are determined by NPS, ACETA and IHOG guidelines. Permits for animal capture must be obtained from the Alaska Department of Fish and Game (ADF&G), as needed. Monitoring staff must perform internal and inter-agency scoping to assure that the project meets the criteria for a categorical exclusion under the National Environmental Policy Act.

3.2 Location, Capture, and Radio-Tagging of Wolves

Typically, a wolf capture operation involves two or three aircraft working together. Fixed-wing aircraft locate wolf packs by aerial snow-tracking, and communicate their location to a capture helicopter. The helicopter captures wolves by aerial darting. A series of standard measurements and specimens are collected from each wolf, a radio-collar is attached, and the wolf is released. Details of wolf capture and handling are described in SOP #1, Techniques for Capturing and Tagging Wolves.

3.3 Disposition and Analysis of Specimens

A limited array of specimens is collected from each captured wolf, including blood and tissue samples. Initial protocol development will include gathering redundant specimens for genetic analysis, in order to determine the best collection technique to optimize DNA extraction in the laboratory. Mammalian red blood cells are anucleated and do not contain DNA, thus solid tissue samples tend to yield more DNA than blood samples. Serum samples will be submitted to ADF&G and/or the Washington Animal Disease Diagnostic Laboratory (WADDL) for disease screening. Genetic and serum samples will be sub-sampled and archived at United States Geological Survey (USGS) in Anchorage, the University of Alaska Museum, and ADF&G in Fairbanks for future analysis. Details of specimen collection and handling can be found in SOP

#1. Details of DNA extraction and analysis of microsatellite and mitochondrial DNA are in SOP's #5, #6, and #7 of the wolf monitoring protocol, not included in this publication.

3.4 Radio-Tracking and Collection of Wolf Location Data

Collared wolves are located from fixed-wing aircraft approximately twice per month. Details of equipment needs and radio-tracking techniques are found in SOP #2, Techniques for Radio-Tracking of Collared Wolves. Initially, wolf location data will be recorded on standard data sheets (Appendix A and SOP#2). The introduction of telemetry receivers which incorporate global positioning units and data loggers in the same unit may eventually allow the gathering and storage of location data without hand transcription.

4 - Data Management

4.1 Database Design

Wolf location data from DENA and YUCH was originally entered and maintained in dBase III+ (.dbf) files (see documentation files wolfloc.doc, dwfld.doc, Appendix C). Data has since been consolidated into Excel spreadsheets (see documentation files dwfld.doc, wolfstat.doc, Appendix C). Microsoft Access databases for storage of wolf monitoring data will be developed in accordance with the CAKN's Data Management Plan. Historic wolf monitoring data will be incorporated into these databases. Procedures for data handling and quality assurance/quality control for all monitoring protocols implemented by the CAKN are detailed in the program's Data Management Plan and in the SOPs for this protocol.

4.2 Data Entry, Verification and Editing

Prompt entry of wolf locations into a database is important to the integrity of the data. When data is entered soon after a flight, it can be pasted from the database into an appropriate map program and visually inspected to ensure that locations are as the observer remembers them, while the flight is still fresh in the observer's memory. Most gross errors in the transcription of coordinates can be detected in this way. Entry of field data is facilitated by the development of data entry screens that resemble field data collection sheets. Procedures for documentation of data entry and archiving of data will be developed in accordance with the Network Data Management Plan.

4.3 Data Validation

Wolf location data is gathered for individual collared wolves, but analyzed on the basis of pack territories, so an important step in data analysis is reviewing the pack designations of collared wolves, to ensure that data are grouped correctly. Wolves disperse away from their packs, start new packs or join existing packs, and their pack designations must be recorded appropriately.

A final step in data validation is the selection of telemetry locations that should be included in pack territory calculations. Anomalous data can result from pre-dispersal movements by wolves preparing to leave their packs, and from extraterritorial forays by individual wolves and packs. Forays by collared wolves and wolf packs far outside their normal range, and outside the scope of the monitoring effort, can greatly change the estimate of wolf density, because the wayward wolves are almost surely encroaching on the territories of non-collared wolf packs. Our initial approach will be to generate minimum convex polygon territories, leaving out those locations that represent obvious forays into the territories of other packs. However, more systematic means of identifying anomalous locations should be investigated (Breck and Biggins 1997).

4.4 Metadata Procedures and Data Archival Procedures

Procedures for documenting data management and for archiving data from the wolf monitoring program will be developed in coordination with the CAKN data manager, and described in SOP #4, Data Management.

5 - Analysis and Reporting

5.1 Determining Pack Home Ranges, Wolf Density Estimation

A home range program (Weinstein et al. 1997, Hooge and Eichenlaub 2000) will be used to generate minimum convex polygon territories (Odum and Kuenzler 1955) for each monitored wolf pack. Various studies have estimated that 30 to 100 independent, year-round locations are needed to adequately describe a wolf pack territory (Fritts and Mech 1981, Scott and Shackleton 1982, Bekoff and Mech 1984, Fuller and Snow 1988). The large pack territories of wolves in Alaska probably require a relatively greater number of radio-locations per year to adequately define them. Burch (2001) found that there was no "magic number" of radio locations guaranteed to adequately describe a wolf pack territory, but that monitoring a number of adjacent packs in a block allows territories to be defined with fewer locations. He recommends combining data over years if one year's sampling effort is insufficient to define territories. The crucial question is whether pack territories are sufficiently defined to ensure that other, undetected wolf packs don't exist within the mosaic of packs being monitored. Weather and budget constraints seldom allow the collection of more than 30 independent locations of collared wolves per year. The combination of location data from two years has been commonly used when wolf densities are estimated annually (Adams, Mech, and Stahlnecker 1997, Burch 2001) As an initial approach to analyzing CAKN wolf monitoring data and estimating wolf densities, we will use location data for each pack for the previous two years to calculate minimum convex polygon territories, leaving out locations that represent obvious forays into other packs' territories. Increased use of GPS collars, which typically provide ≥ 300 locations per year, will solve the problem of sample size in delineating pack territories. The area of the wolf population estimate will be the aggregate of the territories of all monitored packs in the area. Details of determining the population estimate can be found in SOP #3, Data Analysis and Reporting.

Wolf packs can usually be seen and counted during the period of snow cover in Alaska, roughly from October through April. Most wolf packs show some decrease in size during this period, as members die or disperse away from the pack. Monitoring flights will concentrate on getting visual observations of packs during early winter, in order to estimate pack size as of October 1, and late winter, to estimate pack size as of March 15. Population estimates will be generated simply by summing the sizes of the packs in the survey area. Density estimates will be calculated by dividing the population estimate by the area of the mosaic of pack territories. Density estimates will be converted to wolves per 1000 square kilometers, a common measure of wolf density (Fuller, 1989).

5.2 Reporting the Results of Wolf Monitoring

Reports generated by the wolf monitoring effort, and the vital signs monitoring program in general, will be designed to be promptly produced, appropriate to their target audience, widely available, and visually accessible. Concise summaries will be a part of each report produced. Reports will conform to guidelines set by the CAKN and the Alaska Region Inventory and Monitoring Plan. Graphical methods, maps, and other visual aids will be used to make results readily understood.

An annual administrative report and workplan will be produced in the fall of each year. The report will account for the expenditure of funds and personnel time in Full Time Equivalents (FTEs), and describe the objectives, tasks, accomplishments, and products of the monitoring effort during the previous fiscal year. The report is designed to improve communication within each park, the CAKN, the Alaska Region, and the National Monitoring Program. Its intended audience includes park superintendents, network staff, regional coordinators, and NPS-wide program managers. The report will be written by the park biologists implementing the wolf monitoring protocol, and reviewed and approved by CAKN, Regional Office and NPS-wide program managers.

An annual report will be prepared for publication each year, reporting on wolf monitoring activities during the previous biological year (May 1 - April 30). It will contain early winter and late winter population estimates for the previous winter. The annual report will provide a summary of all wolf monitoring activities and data for the year, and describe the current condition of the wolf population in the areas monitored. Any changes in monitoring protocols will be documented. The intended audience for the annual report includes park resource managers, network staff, and external scientists. The report will be written by the park biologists implementing the wolf monitoring protocol, and peer-reviewed within the CAKN.

In addition to the two scheduled annual reports, results of the wolf monitoring effort will be presented at the biennial CAKN Vital Signs Monitoring Conference, and at other symposia, conferences, and workshops. Wolf monitoring data will be provided to park interpretive staff for written and oral presentation to visitors. Significant findings will be reported in scientific journals and popular publications.

6 - Personnel Requirements and Training

Wildlife biologists from individual parks will serve as principle investigators for wolf monitoring in their parks. Capture efforts will often be conducted in cooperation with biologists from other network parks. Radio-tracking efforts will be conducted by pilots with wildlife biologists or wildlife technicians as observers, or by pilots alone. Personnel needs for wolf monitoring are not great enough to require additional park staff. The greatest need for specialized expertise will come in the capture and radio-collaring of wolves. NPS personnel involved in capture operations must have ACETA certification, proper training in animal immobilization and handling, and appropriate aircraft safety and management training under IHOG regulations.

After wolves are collared, monitoring flights are conducted about twice per month. It is desirable for data to be gathered by an NPS observer, but data is sometimes gathered by pilots operating alone. When NPS personnel take part in telemetry flights, they must have appropriate aircraft safety training as specified by AMD, and wear PPE suitable for Special Use flights under AMD guidelines.

The selection of aircraft and pilots is of great importance for the safe and economical capture and monitoring of wolves. Aerial snow-tracking requires considerable experience and expertise to perform efficiently, and the piloting of helicopters for wolf capture is among the most demanding jobs required of pilots in Alaska.

In addition to park biologists and pilots, important personnel in the wolf monitoring effort include collaborators at U. S. Geological Survey in Anchorage for genetic analyses, veterinarians with the ADF&G for wolf necropsies, the CAKN database manager and park Geographic Information Systems (GIS) specialists for help with data management and analysis.

7 - Operational Requirements

7.1 Annual Workload and Field Schedule

Wolf capture operations are usually carried out in early winter (November) and late winter (March), when snow cover and daylight conditions are suitable. It is reasonable to expect that initial capture of wolves for a large (10 wolf pack) monitoring effort will require a week's effort, using one helicopter and two or three fixed-wing aircraft. Costs of aircraft use for wolf capture commonly run $1500 - $2500 per wolf.

In the initial stages of a wolf monitoring effort, it is to be expected that not all packs in the targeted survey area will be found and collared on the first attempt. As packs are radio-tracked and their territories determined, gaps may be discovered between the collared packs, where other, uncollared packs of wolves may live. A followup round of wolf capture is undertaken to fill in the gaps.

Wolf monitoring flights are scheduled roughly twice per month, and commonly take most of a day to complete. If NPS biologists serve as observers on all monitoring flights, the effort is likely to require about 24 work days per year. Commonly, however, pilots radio-track wolves and report data without NPS or other observers in the aircraft.

7.2 Facility and Equipment Needs

No specialized facilities are required for the wolf monitoring effort. Laboratory analyses for genetic and disease screening are carried out by cooperators (USGS, UAF, ADF&G, and WADDL). Equipment required for capture and monitoring of wolves is detailed in SOP #1. A complete kit for field capture of wolves can be expected to cost about $2,500. Equipment to perform radio-tracking from a fixed-wing aircraft totals about $4000. At present, adequate field equipment is already present within the Network. The costs of wolf monitoring lie primarily in aircraft costs, and secondarily in expendable materials like radio collars and drugs.

7.3 Startup Costs and Budget Considerations

A wolf monitoring effort can be implemented in CAKN without the need for additional personnel or facilities. Field work, data management and reporting will be handled by existing park staff. Because wolf monitoring programs are already in place in two parks, startup costs and training needs are minimal. Costs estimated in Table 1 below are for establishing and maintaining a wolf monitoring effort covering 10 wolf packs in each park, but costs will be highly dependent on the scope of the survey area, the frequency of monitoring flights, and mortality of collared wolves.

Table 1. Cost estimates for establishing and maintaining wolf monitoring for 10 packs.

Estimated Costs	DENA	YUCH	WRST
Travel Expenses	2,000	2,000	2,000
Startup Equipment Costs	0	0	8,000
Annual equipment/supplies	10,000	10,000	10,000
Annual aircraft use budget	80,000	80,000	80,000
Total	92,000	92,000	100,000

7.4 Procedure for Revising the Protocol and Archiving Previous Versions of the Protocol

Over time, revisions to both the Protocol Narrative and to specific SOPs are to be expected. Careful documentation of changes to the protocol, and a library of previous protocol versions are essential for maintaining consistency in data collection, and for appropriate treatment of the data during data summary and analysis. The MS Access database for each monitoring component contains a field that identifies which version of the protocol was being used when the data were collected.

The rationale for dividing a sampling protocol into a Protocol Narrative with supporting SOPs is based on the following:

- The Protocol Narrative is a general overview of the protocol that gives the history and justification for doing the work and an overview of the sampling methods, but that does not provide all of the methodological details. The Protocol Narrative will only be revised if major changes are made to the protocol.
- The SOPs, in contrast, are very specific step-by-step instructions for performing a given task. They are expected to be revised more frequently than the protocol narrative.
- When a SOP is revised, in most cases, it is not necessary to revise the Protocol Narrative to reflect the specific changes made to the SOP.
- All versions of the Protocol Narrative and SOPs will be archived in a Protocol Library.

The steps for changing the protocol (either the Protocol Narrative or the SOPs) are outlined in SOP #9, “Revising the Protocol”. Each SOP contains a Revision History Log that should be filled out each time a SOP is revised to explain why the change was made, and to assign a new Version Number to the revised SOP. The new version of the SOP and/or Protocol Narrative should then be archived in the Long Term Environmental Monitoring (LTEM) Protocol Library under the appropriate folder.

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Appendix A. Field Data Forms

Wolf Capture Sheet
Aging Wolves by Tooth Wear (back of Wolf Capture Sheet)
Sample Wolf Radiotracking Form
DENA Wolf Necropsy Sheet

WOLF CAPTURE DATA PARK ____________ WOLF # ____________________
CAKN *capsheet.doc* RECAP?_____ LASTCAP__________ OLDFREQ ___________

CAPTURE DATE____/____/________ WEIGHT________ SEX ___ AGE(EST)____________

LOCATION, GENERAL____

COORDINATES (WGS84): LAT ______°______.______' N, LONG ______°______.______'W

PACK________________ COLOR/MARKINGS____

RADIO SERIAL #____________ MAGNET OFF?___ VERIFIED?____ FREQ_____________

VHF GPS3 GPS4 ARGOS#______________ SET OF HOLES USED ______ (from transmitter)

CAPTURE METHOD______________ SHOOTER / PILOT____________/______________

DRUG_________ DOSE___________ DART LOCATION__________ DART TIME__________

GENERAL BODY CONDITION____

TEAT / TESTIS LENGTH_________cm WIDTH_________cm TEAT COLOR______________

VULVA APPEARANCE / DISCHARGE____

CANINE LENGTH UL_____ LL_____ UR_____ LR_____ CHECK MOUTH FOR STICK___

CANINE WEAR, UL__________UR__________ ERUPTION: UL__________UR__________
None, Rounded, Blunt, or Flat tips Forward, Down, or Back

UPPER CARNASSIAL WEAR: NONE CAPPED CUSPS PITTED CUSPS PITTED CUSPS & VALLEYS HEAVY

TOOTH WEAR: UPPER INCISORS______________ LOWER INCISORS_______________

GENERAL TOOTH CONDITION, STAINING____

____

PUP CHARACTERS: RADIUS-ULNA BUMP ___________ HAIR COAT________________

SPECIMENS	DRUG BOOST	TEMP	PULSE	0_2	RESP	TIME
blood: Longmires _______						
serum (red) _______						
EDTA (purple) _______						
tissue: hair _______						
cheek swab _______						
other _______						

REMARKS____

PHOTOS ____

ASSOCIATES: (#, COLORS)__________________ (COLLARED WOLVES)_____________

OBSERVERS________________________________SPOTTER__________________________
revised 02/09

AGING WOLVES BY TOOTH WEAR

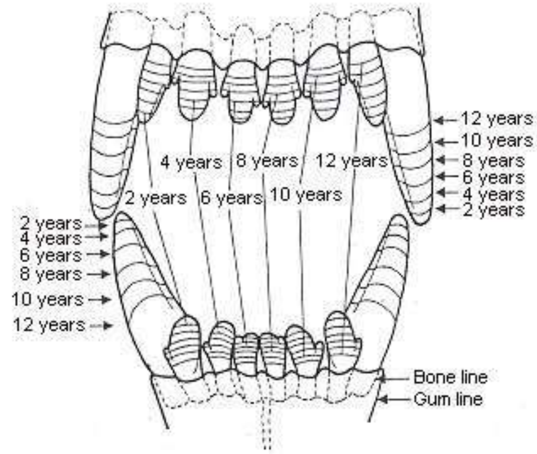

Fig. 1. Progressive wear on wolf incisors and canines in 2-year increments from ≤1 to ≥12 years of age. Wear on incisors typically progresses beyond the lobes on the first 2 upper and lower incisors at 8 years of age, leaving approximately 5 mm of enamel. At 10 years of age, 2-4 mm of enamel remain on the first and second incisors. Length of canines is reduced 30-50% with 10-16 mm of enamel remaining. Beyond 12 years of age, incisors may be worn to the roots, with a few peg-like stumps projecting above the gum line, or the gums may cover the roots. Length of canines is reduced ≥50% with ≤10 mm of enamel remaining.

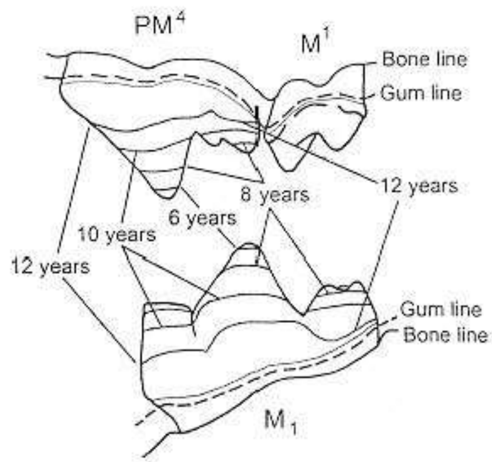

Fig. 2. Progressive wear on wolf carnassials (upper premolar 4 and lower molar 1) in 2-year increments from ≤6 to ≥12 years of age. Wear is visible on tips of major prominences at 5 years of age and profiles flatten slightly by 6 years. Deep wear on the posterior cusp of the lower carnassial after 10 years of age results from occlusion with the first upper molar, not the upper carnassial.

Source:

Gipson, P.S., W.B. Ballard, R.M. Nowak, and L.D. Mech. 2000. Accuracy and precision of estimating age of gray wolves by tooth wear. Journal of Wildlife Management 64: 752-758.

Sample Wolf Radiotracking Form

RADIO TRACKING FORM - WOLF **PILOT/OBS___________________/_______________________PLANE______________DATE____________________Page ______ of ______**

Form Revised: October 12, 2004 **PARK/PROJECT_____________________________________ WEATHER____**

														KILLS				
	CapDate	**FREQ**	NO.	**PACK**	A/S	LOCATION	GNO	TIME	MO	VI	AD/PUPS	MOR?	BEH	SPP.	AG/SX	%EAT	GPS	COMMENTS/ Wolf Colors
1	0304	**0.121**	0408	**LAZY RIVER**	GFyrl													
2	1003	**1.230**	0318	**LAZY RIVER**	GF													
3	0901	**2.459**	0105	**SALAD FORK**	BM													
4	1003	**1.110**	0103	**SALAD FORK**	GF													
5	0902	**2.668**	0215	**SQUIRREL R**	BM													
6	0304	**2.540**	0411	**SQUIRREL R**	GF yrl													
7	0204	**2.666**	0402	**SLIME CR**	GF													GPS collar
8	0304	**2.250**	0409	**SLIME CR**	WF													
9	0304	**0.230**	0410	**BIG HILL**	GF													
10	0303	**0.240**	0322	**BIG HILL**	SWM													
11	1003	**2.650**	0320	**FISH MTN**	BM													
12	1004	**1.890**	0403	**FISH MTN**	BF pup													
13	1004	**1.780**	0455	**FISH MTN**	BM													
14	0302	**1.869**	0208	**LONER**	GF													
15	0302	**2.368**	0209	**SCHIST CR**	GM													
16	0902	**2.740**	0210	**SCHIST CR**	BF													

Please note colors of wolves observed in comments (B=black, G=grey, W=white, DG=dark grey).
LOCATION = new location (general)
GNO = group number TIME = 24 hour
Mode = 1 active, 2 mortality, 3 capture, 4 not heard
Visual = 1 individual (see radioed animal)
2 group (see group only and hear radio)
3 no visual

GPS collars: 40 PPM = No Fix &Active, 60 PPM = Fix & Active, 80 PPM = Mortality
AD/PUPS =# of adults/number of pups
one number indicates adults only
MOR? =more wolves not seen? (Y or N)
BEHavior =# doing each (e.g. 7R, 2S)
R resting, T traveling, F feeding,
I interacting, S standing, O other

Map all wolf mort locations
KILLS: SPecies = M moose, C caribou, S sheep, O other
AG/SX (AGE/SEX) = AD adult, YOY (calf or lamb); M or F (male or female)
Provide evidence in comments, e.g. antlers, horns, etc.
% EATen = how much has been consumed
GPS = LAT & LON in decimal minutes
COMMENTS = description of actions, numbers and colors of wolves, den site, etc.

<u>DENA WOLF NECROPSY RECORD</u>

Necropsy No:___________
Wolf No:_______________
Frequency:_____________
Pack:__________________

Date died:____________________ Date examined:_________________
Last heard alive:_____________ First heard in mort:___________
Township:_______ Range:_______ Section:__________ Datum:__________
Lat:______deg._____._____min.N Long:________deg._____.______min.W

Manner taken:____________Weight: whole__________skinned__________
Age________________ Sex______________ Color____________________
Body length__________ Tail length__________ Chest girth __________

<u>Specimens examined:</u>

General condition____
Subcutaneous fat____
Omental and kidney fat___________________________________
Xiphoid fat: ____________ grams
Femur marrow: appearance______________ % fat______________
Stomach contents____
Parasites____
Tooth wear____
____ M_1 wear class________
Canine lengths: UL________UR_________LL_________LR_________
Testis or teat length_________________width________________

<u>Specimens collected:</u>

Entire Carcass ____
Skull____________Mandible____________Front ankle____________
Reproductive tract: Collected_____________________________
Analysis____
Tissues: Heart______Liver______Kidney______Muscle_______
Femur/femur marrow:____________ Radio Collar: _____________
Blood: whole____
serum____
Urine____
Other____

Comments:____
____
____
____
____
____
____

Carcass Retrieved by: ____
Hunter or trapper: _______________________ Reward paid?__________
Necropsy by: ____

Appendix B. Standard Operating Procedures

Standard Operating Procedure (SOP) # 1 Techniques for Capturing and Tagging Wolves

Standard Operating Procedure (SOP) # 2 Techniques for Radio-Tracking of Collared Wolves

Standard Operating Procedure (SOP) # 3 Data Analysis and Reporting

Standard Operating Procedure (SOP) # 4 Data Management

Standard Operating Procedures (SOPs) #5, #6, #7

Standard Operating Procedure (SOP) # 8 Handling GPS/ARGOS Wolf Location Data

Standard Operating Procedure (SOP) # 9 Revising the Protocol

Standard Operating Procedure (SOP) # 1 Techniques for Capturing and Tagging Wolves

Version 1.01 (July 2009)

Revision History Log:

Prev. Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #
1.00	July 2009	Thomas Meier	Minor corrections	Preparation for publication	1.01

This Standard Operating Procedure explains the procedure for locating, capturing and tagging wolves for radio-telemetry monitoring. This SOP also describes the measurements and specimens that are collected from captured wolves, and the disposition of specimens. The steps for changing the protocol are outlined in SOP #9, "Revising the Protocol".

I. Preparation and Equipment Procurement

Once it has been decided to implement a wolf monitoring program, a number of procurement and regulatory steps must be taken, well in advance of any capture efforts. Radio frequencies for radio collars must be cleared through the National Wildlife Telemetry Frequency Management Database, and the National Park Service's Radio Coordinator. Approved frequencies will be spaced 15 kilohertz apart. For GPS/ARGOS collars, ARGOS identification codes must be obtained from the ARGOS representative in the USA, CLS America (www.clsamerica.com). Details of collar design are arranged with the manufacturer to fit the needs of the project. Minimum equipment for a wolf monitoring study should include radio collars (at least two per pack in the intended study area), at least one telemetry receiver, antennae, cables, wing strut brackets, and switchbox for aerial telemetry, a dart rifle, darts, internal and external dart charges and drugs, and a well-stocked capture kit (see list below).

Under the Controlled Substances Act, only a person with a Drug Enforcement Agency (DEA) registration number may purchase Telazol and other controlled substances. Appropriate Personal Protective Equipment (PPE) for fixed-wing and helicopter special-use flights must be obtained for field workers.

Permits for animal capture must be obtained from the Alaska Department of Fish and Game, as needed. Monitoring staff must perform internal and inter-agency scoping to assure that the project meets the criteria for a categorical exclusion under the National Environmental Policy Act.

II. Aerial Tracking and Wolf Capture

Procedures:

1. *Choosing a survey area.* For each park in the monitoring network, an appropriate survey area for wolves will be selected. When possible, it is desirable that wolf survey areas be co-located with the monitoring of other vital signs, including large mammal surveys and vegetation monitoring. The wolf survey area will be chosen according to the particular management and research needs of a park/preserve. A desirable survey area is approximately 10,000 square kilometers, a size which is likely to include from 4 to 10 wolf packs, depending on terrain and prey availability. The actual survey area will be determined by the territories of the wolf packs that comprise it. Elongation or subdivision of the targeted survey area increase the chances that actual pack territories will straddle the boundary of the targeted area. An area that is more nearly round in shape can be more efficiently and predictably surveyed.

2. *Timing and logistics of capture work.* Once a wolf survey area has been established, wolf packs are located and radio-collared, and their territories are determined by radio-tracking. Often, another round of pack location and capture will need to be performed in order to fill in the gaps in the pack territory mosaic. Track searches are best performed several days after a fresh snowfall, at a time of year when the angle and duration of sunlight are adequate. Most wolf surveys and wolf capture efforts take place in November and March. Because of increased difficulty and danger of field work and survival at extremely low temperatures, capture operations are usually not attempted at temperatures lower than -30º F (-34º C). Both a helicopter and pilot approved and carded by AMD for animal capture work must be scheduled, and usually two or more fixed-wing tracking aircraft are scheduled to locate wolves for capture. All aircraft and pilots must be carded for the mission and procured according to NPS, AMD, and IHOG (US Department of the Interior, 2006) regulations. Flight plans and flight following according to Federal Aviation Administration (FAA) and NPS regulations are important for safe operations. Clothing and equipment specifications are determined by NPS, ACETA (US Department of the Interior, 1994), and IHOG guidelines. Radio communication is essential for flight plans, flight following, and communication between the various aircraft involved in an operation.

3. *Aerial tracking and identification of animals for capture.* The monitoring area will initially be surveyed by snow-tracking from fixed-wing aircraft, in order to locate and identify resident wolf packs for collaring. The initial survey provides a "snapshot" of wolf packs in the area, and the sizes of wolf packs present, even if no further work is done. Obtaining the pack sizes of several wolf packs can provide an index to wolf numbers, at fairly low cost. Aerial tracking in winter is the standard wolf survey method in Alaska, used in early studies (e.g. Burkholder 1959), in line-transect methods (Ballard et al. 1995, Becker et al. 1998) and as a method of locating wolves for radio-collaring (Mech et al. 1998). Prior to 1986, aerial snow-tracking surveys were the only method of estimating wolf numbers and movements in Denali (Haber 1977, Dalle-Molle and Van Horn 1985). Tracking is best performed from small, slow-flying aircraft such as the Piper PA-18 Supercub or Christen Husky. Wolf tracks must be differentiated from those of other species and followed, sometimes for many miles, until the wolves are overtaken. Aerial snow-tracking is a specialized skill that is best developed by

long practice. The tracking abilities of the pilot are more important than those of the observer, so careful selection of survey pilots is vital to the success of the survey.

The location of wolf packs in the targeted survey area can be greatly aided by prior experience and local knowledge. The tendency of wolf packs to make forays and seasonal migrations further complicates the task of collaring all of the wolf packs in a given area. Often, an initial round of radio-collaring and tracking to determine pack territories must be followed by subsequent tracking and collaring efforts to fill in the gaps in the pack territory mosaic. It is to be expected that all areas that harbor prey populations in Alaska are occupied by wolf packs at some time.

When wolf tracks are encountered, they are followed until the wolves that made them are sighted. Experienced trackers can readily determine the species, direction of travel, and even the numbers of animals that have made a track. Nonetheless, following the tracks of a wolf pack, often through forested areas, through the tracks of other species, and through windblown terrain, can be exceedingly difficult. Old snow, poor light, wolves traveling in the tracks of moose or caribou, or backtracking in older wolf tracks can make this process almost impossible. Good snow and light conditions cannot be overemphasized. Once a wolf pack is sighted, the number of wolves present, their colors, sizes, and behavior are noted, and the wolves are kept in sight while a capture aircraft is summoned. Wolves that are unused to aircraft or have been hunted from aircraft may scatter and flee on approach. It is sometimes vital to quickly assess a wolf pack from the air and follow the leading, fastest-fleeing animals, more likely to be the breeding members of the pack, rather than those who loiter or fall behind, more likely to be juveniles. Monitoring efforts are greatly aided by collaring the dominant, breeding members of a pack rather than subordinates who are likely to disperse away from the pack. The goal is to maintain two or three radio collars per pack, and the breeding male and female should be included in this number if possible.

4. *Helicopter operations.* When wolves have been located and identified for capture, the helicopter is summoned. The spotter plane ascends to a higher altitude to direct the helicopter and monitor the wolves. The spotter plane is also useful for notifying the helicopter about other aircraft and low-level hazards in the area. Commonly used helicopters for capture work include the Bell Jet Ranger, Hughes 500, and Robinson 44 aircraft. The individual aircraft must have a current card from AMD for aerial capture work, as must the pilot. If additional handling personnel are present on board the capture helicopter, they are usually set off, along with excess baggage as needed, to lighten the aircraft and increase maneuverability during darting. The capture pilot and gunner then pursue the wolves, as directed by the spotter plane. Descriptions of the appearance, location and behavior of wolves is communicated by radio as animals are selected for capture. Safe and efficient piloting of capture helicopters is an uncommon skill and, as with the selection of tracking pilots, selection of helicopter pilots for wolf capture must be done carefully, with emphasis placed on experience.

5. *Darting and drugging.* Wolves are anesthetized by darting with Palmer Cap-Chur® equipment (Palmer Chemical and Equipment Company, Douglasville, GA) including a Extra Long Range Projector, brown (extra low velocity) or green (low velocity) propellant charges and 5 ml. darts. The dart is propelled with a 22 caliber blank charge inserted into an adapter

that is loaded into the breech of the gun behind the dart. The aluminum, 32 gauge Palmer darts are equipped with ¾-inch barbed needles and cloth tails. Internally, they contain a lubricated rubber plunger, an internal explosive charge specifically made for 4 to 10 ml. Cap-Chur® darts, and a space for the drug in front of the plunger. Darts are assembled and loaded before departure, with 400-600 mg. of Telazol® (Tiletamine-Zolazepam) hydrated to 5 ml. with sterile water. Other drug combinations may be tested and used as they become available. A test-tube rack provides a convenient platform for dart assembly and loading. Telazol is a "tolerant" drug with a high therapeutic index (Kreeger 1999), with little likelihood of overdose, so a standard dose can be used without the need for estimating body mass. Any darts unused at the end of the day must be unloaded to prevent corrosion. Telazol may be stored in original bottles and used for up to 2 weeks after it is hydrated. During capture operations, loaded darts are stored in an ammunition can, with a handwarmer if necessary, to keep them secure and safe from freezing. A cleaning rod or a set of shorter push rods must be kept available for removing unfired darts from the projector barrel. It is useful to have several blank adapters for the dart rifle on hand, pre-loaded with powder charges, so that fired blank charges need not be removed and replaced in flight. Communication between pilot and shooter requires a functional intercom capable of being locked into a "hot mike" position for the shooter's microphone. When the door is removed, the shooter is held in the helicopter by a shooting harness while he/she leans out the door to fire the dart gun. An alternative is the use of a shooting door, with a window that opens to allow darting without removal of the door. The pilot maneuvers the helicopter to match the speed of the fleeing wolf, usually within 15 meters of the animal, and the shooter attempts to fire a dart into the muscle of the wolf's hindquarters. The relatively small size and maneuverability of wolves make them difficult targets, and pilot and shooter experience are necessary for successful and safe capture. The most common cause of capture-related death among wolves in Denali, representing fewer than 2 % of capture attempts, has been from internal injury caused by a dart striking the wolf's chest or abdomen. Considering misses and other dart losses, one must prepare three darts for each wolf intended for capture. When a wolf has been successfully darted, the helicopter may move away and let the spotter aircraft monitor the animal during drug induction, or the helicopter may be used to keep the wolf in a desirable area while drugs take effect. The time of initial darting, the dose of drug in the dart, and the position of the dart hit on the animal should be recorded on the capture form (see attached sample data form). If a wolf has been successfully darted with a deep intramuscular injection of Telazol, the effects of the drug (staggering, head-bobbing, collapse) are usually seen within 5 minutes, and the wolf is usually immobilized within 10 minutes. If no effects are seen within 10 minutes after darting, it may be assumed that the injection was unsuccessful and another dart used. With the help of a fixed-wing spotter aircraft to keep track of the various wolves in a pack, more than one wolf can be darted and handled in a single operation. In this case, the helicopter can be used to transport wolves to a single location for processing.

Care must be taken in selecting the area where wolves are darted, and if possible the area where the wolf travels after being darted. Steep terrain and open water present hazards for the animal and for handling personnel, and availability of helicopter landing areas can determine the distance that handling personnel must travel to the anesthetized animal. Careful monitoring of darted wolves by spotter planes, along with monitoring and judicious herding by the helicopter, can protect the wolf and increase the efficiency of the operation. A wolf that shows the effects of the drug but does not become completely anesthetized may require the administration of a second dart with 250 mg. of ketamine in a 3 ml. dart. A wolf that is anesthetized enough to be approached but not enough for complete handling may be boosted with ketamine (5 mg. per kg. of body weight), desirable because it is shorter lasting than Telazol, or with additional Telazol. Personnel administering drugs to wolves and handling drugged wolves should have current certification under the NPS ACETA program, as well as training in wildlife chemical immobilization. The presence of a veterinarian during capture operations is desirable but not necessary.

III. Wolf Handling and Specimen Collection

Procedures:

1. *Initial evaluation and body temperature check.* Wolves successfully drugged with Telazol typically show a desirable level of muscle relaxation and anesthesia. The combination of tiletamine (a dissociative anesthetic) with zolazepam (a tranquilizer) produces anesthesia with less risk of muscle hypertonicity, overheating, and seizure than drug combinations that were used in the past. Disadvantages of Telazol include sometimes excessive salivation and a down-time that may exceed two hours, longer than is necessary to process a wolf. It is to be expected that better drug combinations will be developed in the future. The use of a drug or drug combination for which an antagonist (antidote) is available would be especially desirable. No antagonist is known for the tiletamine component of Telazol, so wolves are allowed to metabolize both drugs and recover gradually.

   A drugged wolf should be approached with caution, until it is determined that anesthesia is sufficient to preclude flight and biting. Leg and ear movements indicate that a wolf is not sufficiently sedated for handling. If the wolf has collapsed on uneven ground or in an awkward position, it should be moved to a flat spot and laid out either on its side or on its sternum. Verification that the airway is clear and the wolf is breathing should be done immediately. The authors once handled a wolf that had choked during anesthesia, and saved it with a Heimlich maneuver.

   Upon reaching a drugged wolf, handlers should locate the dart and remove it. Barbed needles are used to prevent the dart from bouncing out of the animal before injection is complete, to help with the verification of a successful dart hit, and to retain the dart for re-use. Removal of the barbed needle can be accomplished by pushing the dart sideways so the barb clears the entrance hole, or if necessary by making a very small incision with a scalpel. Used darts should be stored in a labeled Nalgene bottle or other secure container. Processing kits should also contain a small sharps container for safe storage of disposable hypodermic needles and other hazardous items.

   The eyes of drugged wolves should be protected from sunlight, foreign objects, and drying. Antibiotic eye ointment may be applied to each eye, and the eyes covered with a cloth or blindfold.

   The most dangerous complication of drugging with Telazol, as with other commonly used drugs for anesthetizing carnivores, is hyperthermia (overheating) caused by the exertion and excitement of capture, coupled with the loss of thermoregulatory ability in anesthesia. Among the first actions to be performed on a drugged wolf, therefore, is taking body temperature. A battery-powered, digital thermometer commonly available for human use is adequate for monitoring the body temperature of drugged wolves. LCD readouts may become sluggish or inoperative in very cold weather, so spare thermometers should be carried. Temperature is taken rectally, with a thermometer or probe lubricated with K-Y Jelly® and inserted at least two inches to obtain core body temperature. Normal body temperature for wolves is about 101º F (38.3º C). If the observed temperature is greater than 105º F (40.5º C), attempts should be made to cool the wolf by packing the groin area with snow or placing the wolf in a sternally recumbent position with its underside exposed to the snow. Coolant packs, sold in drug stores, can be wrapped in cloth and placed in the groin area. The lack of thermoregulatory ability in anesthetized wolves can also lead to hypothermia (reduced body temperature), especially under conditions where a wolf has become wet. Body temperatures lower than 98º F (36.7º C) are cause for concern, and handlers should take action to warm the wolf. Warming strategies can include placing the wolf on a foam pad, drying its fur, covering it with a blanket, or using warming packs commonly sold for sporting or therapeutic use. If the latter are used, they should be wrapped in cloth and placed in the groin area. Both warming and cooling of a wolf may be slow processes, taking 30 minutes or more, and temperature should be monitored and recorded every 5 minutes until it has reached the desirable range. Starting the helicopter and warming the wolf inside of the passenger compartment has been used successfully in severe cases of hypothermia.

2. _Vital signs monitoring._ The most reliable way to evaluate the status of an anesthetized animal is to monitor its vital signs, particularly temperature, pulse, and respiration. Most importantly, handlers should immediately verify that the airway is clear and an animal is breathing when it is first handled. Second in importance is verification that body temperature is in the desirable range of 98-105 º F (36.7-40.5º C). These are not absolute thresholds, and an animal whose body temperature is approaching the limits of this range should be treated pre-emptively, particularly if serial temperature measurements have shown a trend toward hyperthermia or hypothermia. Once it has been determined that the animal is safely anesthetized and handleable, quantitative measures of respiration and pulse are made and recorded. Respiration can be observed visually, and pulse detected with a stethoscope or manually, on the left side of the animal between the fourth and fifth ribs. Pulse and respiration can be counted for ¼ or ½ minute and multiplied to get a rate per minute. A desirable respiration range is 12-20 breaths per minute. A desirable heart rate is 50 to 120 beats per minute. If available, a pulse oximeter can be used to measure blood oxygen content, but pulse oximeter displays may not work well below 15° F. Oxygen saturation can be improved by rotating the position of drugged wolves every 5 or 10 minutes. Pulse and respiration that diverge from normal ranges can indicate potentially serious problems in anesthetized animals. A thorough course in wildlife chemical immobilization will include the proper use of drugs such as Atropine and Dopram to regulate abnormal pulse or respiration, but these conditions are rarely an issue with drugged wolves. Capillary refill time, an index of cardiac function, can be measured by pressing on a non-pigmented part of the gum, releasing pressure, and determining the time to restoration of color. A capillary refill time of less than 2 seconds generally implies adequate blood pressure. A slower refill time indicates low blood pressure or other circulatory dysfunction. All vital signs, along with any booster drugs or other drugs administered, should

be recorded along with the time (military or 2400 hour time) on the table provided on the capture sheet.

3. *General condition and appearance.* The general body condition of captured wolves should be briefly described. A wolf in good condition should have a layer of fat over the pelvis and ribs, making them palpable but not sharply so. On a wolf in excellent condition, the pelvis and ribs may not be felt beneath a heavy layer of fat. Wolves in fair condition, and poor condition, will show decreasing fat levels and more sharply palpable bones.

   Drugged wolves should be examined for old or new injuries, including broken bones, abrasions, and abnormalities of the skin and fur. Digital photographs should be taken of any injuries or abnormalities. A skin disorder of unknown origin has been seen in interior Alaska wolves, causing the loss of guard hairs on the flanks, a ridge of hair on top of the back, and a "dusty" appearance to the undercoat (Beckmen et al., 2009). Infestations of the dog louse *Trichodectes canis* have been observed in interior Alaska wolves, including wolves caught in or near Denali (Wolstead et al., 2009) Louse infestations are characterized by hair loss between the shoulder blades and in the groin area, and a characteristic dishrag-like odor. Suspected ectoparasites and skin scrapings from affected areas can be preserved in alcohol in an empty blood tube. Fine-toothed combs can be used to scrape ectoparasites onto a sheet of butcher paper, for transfer into alcohol. Wolves infected with lice or other parasites can be treated with Ivermectin (eprinomectin). Based on the distribution of lice in interior Alaska, it appears likely that infestations are introduced from dog populations, and thus not a natural feature of the environment (Wolstad et al., 2009). A potentially fatal circumstance that is common in trapped wolves but seen even in free-ranging wolves is a stick lodged crosswise in the top of the mouth between the carnassial teeth. Such a wedged stick can cause massive infection, and the palates of all wolves that are handled should be examined. A wedged stick can be removed with a Leatherman® tool or other pliers. New or old wounds can be treated with the application of Nolvasan® antibiotic ointment or nitrofurazone spray. Injured wolves may be given a long-lasting antibiotic such as Flo-Cillin® or Dual-Cillin®, with a concentration of 150,000 units per ml, at a dose of 1.5 ml per 10 kg of body weight, administered subcutaneously.

   The general coat appearance should be noted on the capture form, along with a description of color and markings. Digital photographs, taken at every capture, provide a valuable record of the conformation, color, and condition of captured wolves. Juvenile wolves, less than 12 months in age, usually show a "fluffy" appearance because of their incompletely developed guard hairs, and this should be noted in the "Juvenile characters: Hair Coat" section. Other morphological indicators of youth are a foreshortened face and overall body shape, and a bump at the distal end of the radius and ulna, caused by the epiphysial growth plate, that can be felt by palpating the ankle. The sex and approximate age of the wolf should be recorded on the data sheet. Objective aging of wolves is often impossible, but some general guidelines include: Canines incompletely erupted, uppers shorter than lowers: juvenile. Canines completely erupted, all teeth sharp and white with no apparent wear: yearling. Increasing levels of tooth wear, often combined with larger body size and evidence of reproductive adulthood: 2-10 years (Gipson et al. 2000, Appendix A). The removal of a tooth for aging is not performed on wolves, because accurate aging of cementum annuli are possible only from canine teeth.

4. *Tooth examination.* The evaluation of tooth eruption and wear are important for estimating the age of a wolf, and several lines of the data sheet are devoted to tooth measurements and observations. Canine teeth are measured with a caliper, in millimeters, from the gum line to the tip of the tooth. A subjective description of canine wear (none, rounded, blunt, flat) and canine eruption (tips directed forward, down, or back) is recorded. The third upper premolar or carnassial tooth, the largest upper tooth, is evaluated for wear and one of five descriptions circled: no wear, capped cusps (enamel is worn off), pitted cusps (darker dentine layer is visible on the tips), pitted cusps and valleys (dentine is visible into the valleys between the cusps), or heavy (entire tooth is worn down and rounded). Incisor tooth wear is recorded for both top and bottom incisors. Diagrams of tooth wear on the canine, incisor, and carnassial teeth of wolves found in Gipson et al. (2000), attached to this protocol (Appendix A), can be copied onto the back side of capture sheets for quick reference to wear classes and age estimates. Finally, a subjective evaluation of overall tooth condition, along with notes on broken teeth, discoloration, and any abnormalities in the dentition are recorded in the space provided on the data sheet. Digital photography of teeth can provide a valuable record of tooth wear.

5. *Body measurements.* Wolves should be weighed on an accurate scale with units (pounds or kilograms) recorded as needed. Handlers may elect to use a weighing cloth, or simply tie the wolf's legs together and suspend it from the scale. The location of a wolf at a fresh kill site, or the observation of a distended belly from eating, should be noted on the capture sheet so that weights can be interpreted in light of a possible full stomach. A minimal set of standard body measurements may be useful in documenting long-term changes in the morphology of wolves that could result from hybridization and other factors. The most valuable measurements for long-term evaluation of morphological change are likely to be measurements of the skulls of dead wolves.

6. *Identification, collaring and tagging.* Each captured wolf is assigned a unique number, based on the year and the order in which it was captured. Thus, the first wolf captured in 2005 will be named 0501. Preceding this with the species and park, as in WOLF DENA 0501, should provide a unique identifier even when data or specimens from many areas are combined. The data sheet provides spaces for indicating a recapture, and for recording the previous capture date and the previous radio collar frequency for recaptured wolves. Capture date should be recorded in the format month/day/year. A general description of the wolf's capture location (distance from some landmark) is recorded, along with the latitude and longitude of the capture site in degrees, minutes, and decimal minutes (datum WGS84). The pack affiliation, sex, and age of the wolf are recorded. Age classes are Pup, Yearling, or Adult. For adult wolves, an estimated age in years or a qualitative estimate of age should be recorded. Few wild wolves reach the age of 10, and those that do have usually turned white, and appear very old.

   The standard conventional VHF radio collar used for monitoring will be the Telonics MOD-500 or MOD-515 collar as configured for wolves. The MOD-515 collar configuration, with multiple small batteries replacing one large battery, was developed by Telonics in order to provide a more durable and reliable collar for use on wolves. Among the most important specifications to be arranged with radio-collar manufacturers is the placement of all antennae within the collar material itself, to preven antennae from being chewed off. The frequency and serial number of the collar are recorded on the data sheet. The magnet, taped to the collar to keep it from broadcasting, is removed to start radio transmission. A telemetry receiver is used to verify the operation and frequency of the collar. Collars are affixed snugly around a wolf's neck, leaving enough room to insert a hand between the collar and the neck. In winter, heavy fur often makes a collar appear tight when it is adjusted initially. A collar that cannot be pulled off over a wolf's head is probably not too loose, but ideally, collars should be tight enough that they do not flop around excessively when the animal moves. Collars placed on yearling and pup wolves should be left an inch or two looser to allow for growth. On Telonics VHF collars, the 4^{th} or 5^{th} set of holes from the transmitter (collar circumference 17-18 in.) is commonly used on female wolves, and the 5^{th} or 6^{th} set (collar circumference 18-19 in.) on males. Space is provided on the capture form to record the set of holes used in attaching the collar. Belting material that extends beyond the collar attachment should be trimmed off so that it doesn't attract chewing. With the radio frequency currently being used (164 to 169 MHz), the antenna extends nearly to the end of the belting, so that care must be taken not to cut the antenna off when the belting is trimmed away. The end of the antenna will extend down from the cut end of the collar, providing a much less attractive target for chewing than the loose belting would. Before capture efforts begin, each collar should be marked in waterproof pen with the radio frequency and agency information including a contact phone number. If the frequency of a collar is also recorded on the distal end of the collar belting, the cut-off end provides a useful record of the collar that was used. A nut driver for tightening nuts and a sheet metal shears for cutting collar material are needed.

Standard VHF wolf collars have a lifespan of 3-5 years. Collar life can be doubled by purchasing collars with a duty cycle of 12 (daytime) hours on and 12 hours off, but this precludes tracking of wolves early or late in the day, which can be a problem in summer. A limited number of wolves will also be collared with GPS/ARGOS collars, which determine an animal's location using the GPS satellite system, and upload the data through the ARGOS satellite system. Collars recently deployed on wolves have been programmed to take one GPS location per day. The lifespan of these collars is expected to be about 30 months. At the present time, Denali has a combination of Telonics' model Gen 3 and Gen 4 GPS/ARGOS collars on wolves in the park. As with the VHF collars, all antennae are placed within the collar belting to prevent chewing. On the GPS/ARGOS collars, this means that both the VHF and ARGOS antennae must be protected. Each GPS collar carries a duty-cycled conventional VHS transmitter so that it may also be tracked from aircraft in the usual way.
Radio collars can be slipped off or chewed off, so it has been useful in some studies to mark wolves with eartags, tattoos, or PIT tags (Biomark Inc, Boise ID) for a backup identification. The use of tattoos is probably not practical under the harsh conditions and time constraints that apply during wolf capture, and most wolves have darkly pigmented lips. If the recognition of a wolf that has lost its collar is deemed important enough, wolves should be marked with eartags, or with PIT tags placed subcutaneously between the shoulder blades. It is worth noting that genetic analysis should establish the identity of a wolf that has lost its collar, because the variability of microsatellite DNA makes it highly unlikely that any two

wolves would share the same microsatellite genotype. When the purpose of marking wolves is primarily to keep track of packs for population monitoring, the identity of particular wolves may not be of critical importance.

7. *Reproductive evaluation.* Reproductive observations can be useful in judging the age and status of a captured wolf. Testis measurements show a seasonal cycle (Kreeger, 2003), but can be used to differentiate between reproductively mature males and juveniles (Mech et al. 1998). The length and width of nipples in female wolves undergo a permanent increase with the first period of pregnancy or pseudopregnancy, so that nipple measurements can be used to differentiate reproductively mature from immature females, as well as females that have nursed pups from those that have not (Mech et al. 1993). Much wolf capture takes place in March, when female wolves are likely to be in proestrus or estrus. The presence of vaginal discharge or bloody urine indicates reproductive cycling, but not necessarily reproductive status, because it has been found that virtually all female wolves aged 22 months or greater show estrus and pseudopregnancy, even if they do not breed or produce pups (Packard, 2003). Swelling and coloration of the vulva, and the presence of any discharge, should be recorded as indicated on the data sheet. The measurements of testes (length and width) in male wolves, or inguinal (rearmost) nipples (length and width) in females should be taken in millimeters and recorded on the data sheet.

8. *Specimen collection.* Blood is drawn from the cephalic (foreleg) vein, using either a large (20 ml) syringe with an 18 gauge needle, or a Vac-U-Tainer® system (Becton, Dickinson Inc). For disease antibody screening, whole blood is placed in two serum separation tubes (red and gray stopper) for subsequent centrifugation, freezing and analysis. Tubes containing EDTA anticoagulant (purple stoppers) are used to store blood for hematology and genetic analysis. All blood tubes should be gently upended several times to mix the contents. In addition, we will be collecting several other specimens for genetic analysis during a protocol development stage, to determine which gives the best results for DNA extraction. These will include whole blood placed in empty tubes (red stopper) with a small amount of Longmire's Solution (Longmire et al., 1988), buccal swabs (cotton swab rubbed on the lining of the mouth), and a tuft of hairs (with roots attached. The last two tissue samples will placed in small manila envelopes for storage. The only other specimens likely to be collected are any ectoparasites, stored in empty blood tubes with alcohol. Care must be taken to label all specimens with the animal number, because multiple wolves are typically captured in a day.

   In cold weather, blood samples must be kept from freezing, which causes cell destruction. Tubes should be labeled with the animal number and stored in a protected location (e.g. a cooler with a hot water bottle or hand warmer in it to prevent freezing) until returning from the field. Excessive heat can also damage blood cells, so the ideal storage temperature is cool but not frozen.

   It is convenient to prepare specimen collection packs in ziplock bags, with the supplies needed for one wolf. These might include one 20 ml syringe for blood drawing, two 18 gauge (green), needles 1 inch in length, two 10 ml serum separation tubes, one 10 ml blood tube ("red top") containing Longmire's Solution, one 3 ml EDTA (purple top) tube, a small manila envelope containing two cotton swabs for buccal sampling, and a small manila envelope for a hair sample.

9. *Recovery of drugged wolves.* Processing of drugged wolves commonly takes about 1 hour. At the end of that time, wolves drugged with Telazol may still be fully anesthetized, although many are clearly waking up. Capture personnel will not remain with the wolf during recovery from the drug. Any benefits of watching a recovering wolf are counteracted by the disturbing effects of human and aircraft presence during the recovery period. Care should be taken that recovering wolves are not left near open water or steep terrain, because recovery from anesthesia is a gradual process, and recovering wolves may be able to locomote but not able to avoid hazardous situations. Wolves located near snowmobile trails or lakes suitable for aircraft landing should be placed in an inconspicuous location to prevent their being discovered, moving them with the helicopter if needed. A wolf left alone will usually recover in place, rather than attempting to move before it is capable of coordinated locomotion. A drugged wolf is not likely to be attacked by members of its own pack, but the presence of wolves from other packs in the immediate area may be cause to move a drugged wolf or remain with it during recovery. Bears are not likely to be active during the time of year when wolves are captured. Recovering wolves are monitored by fixed-wing spotter aircraft later in the day, or first thing the next day, to confirm their recovery.

10. *Personnel, photos, remarks.* Spaces on the data sheet allow the recording of the helicopter pilot and the shooter, handling personnel and other observers, a record of photographs taken, and other remarks. The remarks section can be used to record any injuries or abnormalities not noted elsewhere on the data sheet, unusual coat color or physical conformation, identity and activity of packmates during the capture operation, other wolves captured at the same time, significant weather conditions, the condition of the wolf on release, and any other observations that may be useful in interpreting laboratory results or the subsequent behavior of the wolf.

11. *Specimen handling and disposition.* When capture personnel return to the laboratory, serum separation tubes are centrifuged for 15 minutes, and the serum poured into screw-topped plastic CryoTubes® (Nalge Nunc Inernational) and frozen. The original blood collection tube with its plug of cellular material (blood clot) is kept as a genetic sample. Whole blood in Longmire's buffer collected for genetic analyses is frozen. Hair and buccal swab specimens, also collected for genetic anaysis, are dried in manila envelopes and stored at room temperature. Both blood and tissue samples are collected to ensure quality DNA extraction. EDTA (purple-topped) tubes are stored in a refrigerator, not frozen, if they will be used for hematology analysis (e.g. a wolf with an apparent infection). Otherwise, blood from EDTA tubes can be frozen for a genetic specimen. The final containers for all specimens are labeled in Sharpie® permanent marker, on the sides of each tube, with the species, park unit, animal number, capture date, sex, and tissue type (whole blood, blood clot, serum, hair, or tissue, etc.).

    None of the specimens collected require rapid handling or express shipping. Genetic specimens are sent to the National Park Service in Fairbanks or to the USGS Alaska Science Center Molecular Ecology Laboratory in Anchorage. Details of DNA extraction, microsatellite DNA analysis and mitochondrial DNA analysis can be found in Standard Operating Procedures 5, 6, and 7, respectively (not included in this publication). Specimens will be sub-sampled by NPS and deposited at various agencies. Redundancy of specimens will allow long- term storage by USGS, ADF&G, WADDL, and the University of Alaska Museum Frozen Tissue Collection, so that archival specimens are available for future analyses. Frozen serum specimens will be sent to ADF&G and WADDL for disease analysis (Brand et al. 1995, Zarnke and Ballard 1987, Zarnke et al. 2001). In cases where a systemic infection or other blood abnormality is suspected, hematology analysis of whole blood preserved in EDTA can be performed by a veterinary laboratory.

12. *Daily equipment maintenance*. At the end of the day, processing kits should be cleaned out and dried, refuse disposed of and contents replenished. Capture guns are cleaned, and spent charges removed from gun inserts. Used darts are disassembled, the spent charges discarded, and the barrels, dart needles and plungers cleaned in hot, soapy water. Used darts can contain pressurized gases and drug residue, and should be disassembled with care. Particular care should be taken to remove contamination from the inside of dart needle barrels. A paper clip wrapped in toilet paper or a small diameter pipe cleaner are useful tools for cleaning needle barrels. A test-tube brush is used to scrub the interior of dart barrels. Dart tails should not be soaked in water, but simply brushed off and fluffed out for re-use. Telazol is removed from unused darts and stored in original containers, being usable for two weeks after hydration. Unused darts are washed to remove potentially corrosive drug solutions from aluminum surfaces. Data sheets are checked for completeness, photocopied and stored securely.

13. *Human safety and legal issues.* Human safety should have the highest priority in conducting animal capture operations. Many aircraft safety issues are mitigated by adherence to the IHOG guidelines for helicopter operation, including training in aviation safety and the use of appropriate Personal Protective Equipment (PPE) for special-use helicopter operations. An exemption, arranged between NPS and AMD, allows deviation from some PPE specifications for winter flying, recognizing the importance of warm clothing for survival in Alaska in winter. Thus, all-leather shoes need not be worn for winter flying. Nevertheless, survival in an aircraft accident will be maximized by the avoidance of synthetic fabrics and the selection of Nomex®, Kevlar®, wool or cotton clothing.

    Selection of capable, experienced pilots is perhaps the most important safety consideration in wolf capture work. Wolf capture involves mountain flying and low-level operations, often in cold weather, and requires skills (snow tracking, helicopter darting) that not only require a great deal of experience to perfect, but also require innate abilities that are not necessarily teachable. As long as a pool of experienced pilots is available in Alaska, and they are available under AMD and NPS procurement guidelines, they should be used for this work. Attempting to locate and capture wolves with inexperienced pilots is likely to at least double the cost of the operation, and to cause increased risk to pilots and crew.

    Safe handling of capture equipment and drugs is ensured by thorough training in ACETA procedures and wildlife immobilization techniques. The use of dart rifles, rather than net guns, for wolf capture results in a safer operation for both humans and wildlife, while allowing capture in a greater variety of terrain. Careful, deliberate attention to the details of safe aircraft and firearms operation are required for safe wildlife capture. Secure handling and storage of dart rifles and darts should preclude the chance of accidental injection with anesthetizing drugs. Nonetheless, capture operations must plan for the event of accidental exposure to these drugs. The dose of Telazol used in wolf capture is not likely to be lethal to a human. But the cyclohexane drugs, which include phencyclidine, ketamine, and tiletamine, are notorious for causing behavioral abnormalities in humans. These may include agitation, incoordination, aggression, disorientation, hallucination, self-mutilation and other abnormal behavior. Convulsions occasionally occur. In the event of accidental exposure to Telazol or ketamine, subjects should be kept in a quiet environment, watched (never left alone), and restrained if necessary. The zolazepam component of Telazol should help to mitigate the effects of the tiletamine. The administration of 10 mg. of diazepam (Valium®), intramuscularly or slowly intravenously, will help control convulsions and calm the patient. Humans exposed to immobilizing drugs should be transported to an emergency center as soon as possible. Physicians unfamiliar with Telazol should be informed that the effects are similar to a combination of ketamine and diazepam. Procurement and storage of Telazol, a Schedule III controlled substance, is governed by Drug Enforcement Authority regulations.

Radio communications are an important safety factor in this and all aircraft operations. Filing and closing flight plans with FAA, obtaining weather reports, flight following with park dispatch, and most importantly, coordination between the various aircraft involved in the operation, are all accomplished with appropriate radio equipment. Radio equipment for aircraft is specified under the contracts and rental agreements with AMD. Handheld radios, both on aircraft and NPS frequencies, should be carried by personnel on the ground so that they can communicate with aircraft and with park dispatch. Radio frequencies for coordination between aircraft are arranged at the beginning of the operation.

Other hazards that may be associated with wolf capture include extreme weather conditions (causing danger of frostbite, hypothermia, and increased chance of injury from tools and other equipment), terrain hazards including falls and avalanche danger, injuries sustained while lifting and moving wolves, and bites from incompletely anesthetized wolves. These dangers can be mitigated by suspending capture operations when weather is too harsh, by avoiding wolf capture in overly steep terrain, by wearing proper clothing, and by thoughtful behavior even when circumstances are rushed or stressful.

14. *Data management*. The wolf capture data sheet (found in Appendix A) should be printed on Rite-in the Rain® paper, with the guide to aging wolves by tooth wear printed on the back side. Graphite pencils or waterproof pens should be used to record data. The data sheet should be examined before leaving a recovering animal, to check for measurements or observations that may have been missed. If enough personnel are present, designating one person to record data can facilitate the consistency and accuracy of the process. This is especially true when more than one wolf is being processed together. The capture location should be waypointed with a GPS unit, and the coordinates recorded on the data sheet as soon as practical.

    Wolf capture data will be entered and stored in a Microsoft Access® database, linked to other databases maintained by CAKN for storing wolf monitoring data. Data is available for wolf captures in DENA dating back to 1986, and in YUCH dating back to 1993. Details of data management for long-term wolf monitoring are contained in SOP #4.

IV. Addresses and Contact Information

Genetic analysis:	Dr. Sandra Talbot USGS/BRD Alaska Science Center Molecular Ecology laboratory Lab 102 and 103 1011 E. Tudor Road Anchorage, AK 9503 http://www.absc.usgs.gov/research/programs/technical.htm#genetics	(907) 786-3582
Wolf necropsy:	Dr. Kimberly Beckmen Wildlife Veterinarian Alaska Department of Fish & Game Division of Wildlife Conservation 1300 College Road Fairbanks, AK 99701 Kimberley_beckmen@alaska.gov	(907) 459-7257
Telemetry Equipment:	Telonics, Inc. 932 E. Impala Ave. Mesa, AZ 85204-6699 www.telonics.com	(602) 892-4444
	Advanced Telemetry Systems 470 First Avenue North Box 398 Isanti, MN 55040 www.atstrack.com	(763) 444-9267
	Communications Specialists Inc. 426 West Taft Ave. Orange, California 92865-4224 http://www.com-spec.com/r1000/r1000.htm	(800) 854-0547
Capture Equipment, Eartags and Applicators:	Nasco - Modesto 4825 Stoddard Road P.O. Box 3837 Modesto, California 95352-3837 www.enasco.com	(800) 558 -595

Flight suits, PPE	Gibson & Barnes 1675 Pioneer Way El Cajon, CA 92020 www.flightsuits.com	(800) 440-5904
Aircraft Procurement & Aircraft Safety Training	USDI Aviation Management Directorate 4405 Lear Court Anchorage, AK 99502-1032 www.amd.nbc.gov	(907) 271-6061
Syringes, medical supplies	Alaska Scientific 664 East Dowling Road Anchorage, AK 99518 http://www.alaskascientific.net/	(907) 563-2758

V. Recommended Equipment List

radio collars, VHF (Telonics) mod-500 or mod-515 configured for wolves (Cast-1, 1.5 inch wide CLM collar, internal antenna, S6A mortality sensor, 44bpm active, 88 bpm mortality with 10 hour delay, adjustable from 15 to 22 inches circumference.	~$300@
radio collars (Telonics), refurbishment of used VHF collars	~$200@
radio collars, GPS/ARGOS (Telonics) TGW-4580 150 sec rep rate, 32 bits. All antennae internal. GPS: 1 location per day @ 1600 UTC (0700 AST) ARGOS: 2, 4-hour on times every 6 days/ Power 800 mW. Partial Pages and all accessory data collection disabled. VHS transmitter on 12 hours beginning @1500 GMT (6am - 6 pm AST or 7am - 7pm ADT)	~$3000@
Radio collars (Telonics) refurbishment of used GPS/ARGOS collars	~1500@
telemetry receiver (Advanced Telemetry Systems)	
Model R4000 (4 Megaherz frequency range)	2750.00
Model R4500 (receiver, GPS, data logger)	5800.00
telemetry receiver (Communications Specialists)	
R-1000 Telemetry Receiver (handheld scanner)	695.00
aircraft tracking antenna, RA-2A (Telonics) (pair)	220.00
aircraft antenna brackets, TAB-3 (Telonics) (pair)	540.00
antenna cables, 17 foot (Telonics) (pair)	70.00
antenna switchbox (Advanced Telemetry Systems)	95.00

antenna switchbox (TAC-2, Telonics)	125.00
3-element folding handheld antenna (Advanced Telemetry Systems)	120.00
2-element flexible (rubber ducky) antenna (RA-14K, Telonics)	212.00
Palmer Cap-Chur Extra Long Range Projector	350.00
extra long range blank adapter (several recommended)	27.70@
Cap-Chur syringe, 5 cc complete, with 3/4 " barbed needle	9.00@
external charges, extra low velocity (brown), box of 50	12.00
internal charges, 4-10 cc, box of 50	23.00
plunger lubricant	4.00
positioning rod for dart assembly and disassembly, barrel clearing (3 needed)	2.00@
syringe brush	1.50
shooting harness for helicopter	~100.00
nut driver for collar bolts	5.00
sheet metal shears for cutting collars	10.00
calipers (metric vernier, stainless or plastic)	20.00
scale, Pelouze, 100 kg capacity, 1 kg accuracy	200.00
rope for weighing	1.00
blindfold/muzzle	10.00
small tarp for processing wolves	5.00
ensolite pad	20.00
backpack for capture gear	50.00
Nalgene bottle for used darts	10.00
small sharps container	10.00
digital fever thermometer	20.00
warming packs for hypothermic wolf	10.00@
20 ml syringes, Luer Lock, (Monoject brand), case of 200	200.00
10 ml syringes, Luer Lock, case of 100	25.00
3 ml syringes, Luer Lock, case of 100	13.00
18 gauge needles, (green) Luer-Lok, 1 inch length, package of 100	12.00
20 gauge needles, (pink), Luer-Lok, 1 inch length, package of 100	12.00
Vacutainer® serum separation tubes (red/gray tops), 9.5 ml draw, package of 100	50.00
empty vacutainer tubes, (red tops)10 ml draw, package of 100	25.00
EDTA blood collection tubes (purple top), 3 ml draw, package of 100	25.00
NUNC™ CryoTube® Vials, 2 ml, self standing, screw top, model 375418 (450 pack)	175.00
Flo-Cillin, 100 ml vial	20.00
Telazol, 500 mg vial	25.00@
Sterile Water, 30 ml vial	3.00@
ketamine, 10 ml (100 mg) vial	20.00
Nolvasan antibiotic ointment, 7 oz jar	20.00
nitrofurazone antibiotic spray, 7 oz spray bottle	10.00
centrifuge	400.00
small manila envelopes	10.00
surgical gloves	5.00

disinfectant wipes	5.00
alcohol for specimen preservation	2.00

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Standard Operating Procedure (SOP) # 2 Techniques for Radio-Tracking of Collared Wolves

Version 1.01 (July 2009)

Revision History Log:

Prev. Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #
1.00	July 2009	Thomas Meier	Minor corrections	Preparation for publication	1.01

This Standard Operating Procedure explains the procedure for radio-tracking collared wolves, recording and storing wolf location data, entering data into the wolf location database, and verification of data. Further information on the use of radio telemetry for wildlife studies can be found in Gilmer et al. (1981) and Mech (1983). This SOP also outlines procedures for responding to wolf collar mortality signals. The steps for changing the protocol are outlined in SOP #9, “Revising the Protocol”.

I. Preparation and Equipment Procurement

Procedures:

1. *Procuring equipment for aerial telemetry.* Complete radio-tracking systems are available from a number of companies in the United States, Canada and elsewhere. This protocol will recommend the use of particular brands and models of equipment with which the authors are most familiar, with some alternatives mentioned.

   Great technological advances are being made in the development of radio telemetry equipment for wildlife. "Standard" telemetry receivers that received widespread use for twenty or more years are no longer being manufactured, and new models are taking their place. For example, the Telonics TR-2 receiver and TS-1 scanner, manufactured by the same company as the radio collars recommended in this protocol, are no longer made. They have been replaced by the TR-5 scanning receiver, a unit that the authors have only minimal familiarity with. This procedure will recommend the use of scanning receivers made by Advanced Telemetry Systems, while anticipating the use of receiver/data logger units that may replace the manual recording of data in the near future.

   A highly functional tracking system for small aircraft consists of the following:

basic system:

Model R4000 telemetry receiver (Advanced Telemetry Systems, Isanti MN)	2750.00
RA-2A directional tracking antenna (pair) (Telonics, Inc, Mesa AZ)	220.00
TAB-3 aircraft antenna brackets for Supercub (pair) (Telonics)	540.00

RW-3-17 coaxial antenna cables, 17 foot (Telonics) (pair)	70.00
RW-2 coaxial cable (to attach switchbox to receiver) (Telonics)	15.00
TAC-2 antenna switchbox (Telonics)	125.00
1/4 " mono phone cable (#42-2381) and splitter (#42-2545) (Radio Shack)	10.00

<u>alternatives:</u>

Model R8000 telemetry receiver (8 MHz range) (Advanced Telemetry Systems)	3200.00
Model R4500 receiver, GPS, and data logger (Advanced Telemetry Systems)	5800.00
TR-5 scanning receiver (Telonics)	2520.00
R-1000 scanning receiver (Communications Specialists)	695.00
antenna switchbox (Advanced Telemetry Systems)	95.00

<u>antennae for handheld, ground tracking:</u>

Model 13863, 3-element folding handheld antenna (Advanced Telemetry Systems)	120.00
RA-14K 2-element flexible "rubber ducky" antenna (Telonics)	212.00

2. *Alternatives.* The basic system listed at the top will give efficient, durable performance for aerial radio-tracking. For an extra fee, Advanced Telemetry Systems will install a signal attenuation switch on the R4000 receiver, which improves its performance for close-in tracking. Superior antenna performance, both in sensitivity to distant signals and directionality for close-in tracking, can be obtained with an alternative antenna mount that changes the orientation of the antennae and moves them away from the metal of the wing strut, by using a wooden dowel that projects forward of the wing. This attachment method has been approved by the Federal Aviation Administration and AMD) (Drawings No. OAS-2-8-87, OAS-2-9-87, OAS-2-10-87). All antenna attachments must be performed and logged by a certified aircraft mechanic.

   Alternate choices for a telemetry receiver include the ATS R8000 receiver, which covers a wider range of frequencies for use on multiple projects, the ATS R4500 receiver/GPS/data logger, which records location information internally, the Telonics TR-5 scanning receiver, which can also be configured for data acquisition, and the Communications Specialists R-1000 scanning receiver, a very compact and inexpensive unit that is excellent for handheld tracking and as a backup unit. Some users prefer the antenna switchbox manufactured by ATS, which has proved to be more durable than the Telonics unit, but is somewhat larger.

   For ground-based radio-tracking, as when retrieving a collar in mortality mode, a handheld antenna that can be conveniently and safely carried in a vehicle or backpack as needed. The flexible and folding antennae listed above are both good choices.

II. Aircraft and pilot procurement and flight preparation

Procedures:

1. *Aircraft and pilot selection.* Aircraft and pilots used for radio-tracking by the National Park Service (NPS) in Alaska must be procured in accordance with the directives of AMD. This

may include the use of aircraft owned by NPS or AMD, or privately owned aircraft obtained on a contract or rental agreement. Information about aircraft procurement is available from AMD.

Experience has shown that aerial radio-tracking is performed most efficiently when the pilot operates the switchbox, so that the aircraft can be oriented in response to changes in signal strength. This requires a period of training and practice for pilots. Expertise at radio-tracking, along with the many challenges of safe flying in the remote and mountainous terrain of Alaska and the harsh weather conditions often encountered, provide strong arguments for careful selection of pilots for this work. Aerial radio telemetry and Alaskan aviation in general are relatively high-risk activities (Sasse 2003, Conway and Moran 2002), and careful selection of aircraft and pilots are necessary to minimize risk.

The standard aircraft for radio telemetry in Alaska is the Piper PA-18 Supercub. Other aircraft such as the Christen Husky and the Bellanca Scout have similar capabilities. The tandem seating, allowing full visibility by pilot and passenger when circling in either direction, the ability to fly slowly for effective observations, and the ability to land in a great variety of locations are factors that have made these aircraft desirable for wildlife work. Occasionally, when long distances must be traveled for telemetry work, or during very short mid-winter days, a faster aircraft such as the Cessna 185 or Found Bushhawk might be favored.

2. *Necessary equipment.* Aircraft used for aerial telemetry and other wildlife work must carry at least one Global Positioning System (GPS) receiver, with an external antenna mounted on the aircraft. Suitable units include the Garmin GPS 150 (panel mounted) and the Garmin GPS 296 (moving map) receivers. Appropriate radio equipment is specified in AMD contracts and FAA regulations, so that aircraft can communicate flight plans, obtain weather information, and notify other aircraft of their presence in certain areas. Radio equipment carried in fixed-wing aircraft is not usually able to communicate on park radio systems. Appropriate equipment can be specified so that this communication can be made, but the very small aircraft used for radio telemetry have limited space for radio equipment. A valuable safety device for communication in remote areas is the satellite telephone, which is becoming increasingly common in Alaska.

   Aerial radio-tracking usually involves descending lower than 500 feet above ground level, and therefore qualifies as a "special use" activity under AMD regulations. NPS personnel on radio-tracking flights are required to wear Personal Protective Equipment (PPE), including a Nomex flight suit, all-leather boots and an approved helmet. An exception for winter flying has been arranged between NPS and AMD, that allows warm clothing and footwear that would not otherwise meet PPE standards to be worn on special-use flights, because of their survival value in cold weather.

III. Preparing data sheets and programming telemetry receivers

Procedures:

1. *Preparing data sheets.* A suitable data sheet for use in an aircraft should be prepared in advance of tracking flights. Appendix A shows a sample radio-tracking form similar to those used at DENA and YUCH. This form is maintained as an Excel file, and could be integrated into the Microsoft Access database used for storing wolf data.

   A variety of data sheet designs may be used, but certain features have proven to be necessary or useful. Each page of a multi-page data sheet should have spaces to record the pilot, observer, aircraft tail number, date, page number and total pages used on that flight. Each animal to be tracked should be listed on the data sheet by its unique animal number, the frequency of its radio collar, and any other identifying data that are helpful in searching for the animal. For wolves, these additional data include the wolf's age, sex, color, pack affiliation and most recent capture date. A serial number on the data sheet provides a useful shorthand when information about particular animals are discussed between aircraft on the radio. The first line of the sample data sheet is wolf number 0408, captured in March of 2004, a gray female yearling with a radio frequency of 160.121 (the first two digits of the radio frequency are left off the data sheet for brevity and security). It is sometimes useful to put the most recent location of an animal on the data sheet, to facilitate searching.

2. *Programming scanning receivers*. Prior to a flight, the radio frequencies of all animals that will be sought are entered into the telemetry receiver's memory bank. Figure 1 shows the control panel of an ATS R4000 telemetry receiver. The R4000 has four memory banks, so that frequencies can be divided into geographic areas to avoid scanning for collars that are not nearby. With the receiver turned on and the bypass switch in bypass mode, a memory bank (A,B,C,D) is selected, the last four digits of the desired frequency are dialed in with the four frequency selectors, and the ADD/DELETE switch is pressed momentarily to the ADD position to place the frequency in the memory bank. The procedure is repeated for each desired frequency. Newer telemetry receivers like the ATS R4500 can be programmed by direct hookup to a computer database. Another advantageous feature of the R4500 is the availability of animal identity notes on the LCD readout, so that the pack affiliation, sex, and age of an animal can be seen when the frequency is addressed. This feature is also available on the inexpensive Communications Specialists R-1000 receiver.

   The receiver needs to be charged according to manufacturer's instructions, before the flight. The operator's manual for the R4000 receiver can be found in the Supplements folder for this monitoring protocol.

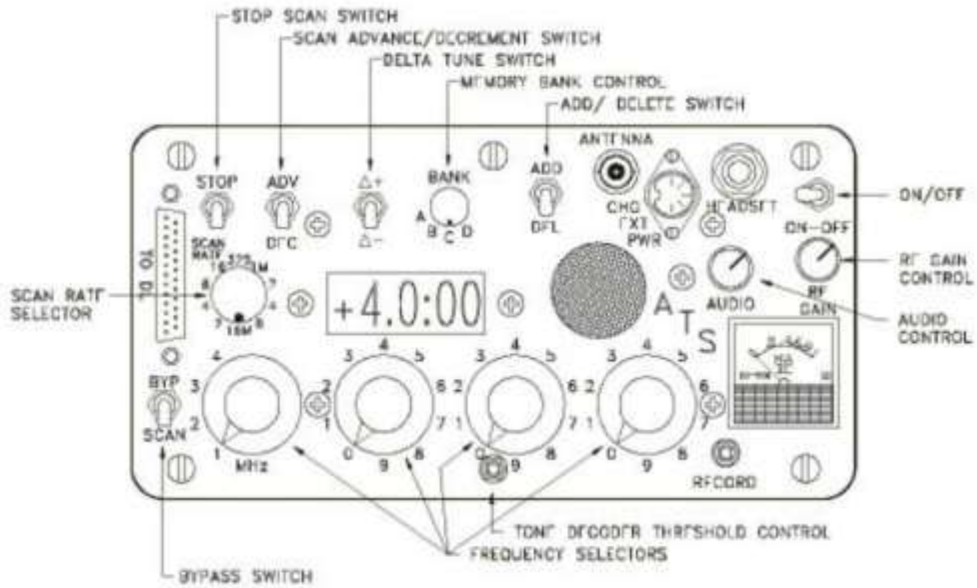

Figure 1. Controls of Advanced Telemetry Systems R-4000 telemetry receiver/scanner. (Advanced Telemetry Systems, 2004)

IV. Aerial Radio-tracking

Procedures:

1. *System setup.* Before beginning an aerial radio-tracking flight, the telemetry receiver must be connected to the aircraft's intercom system, and to the antenna system. Coaxial cables from the two wing-mounted antennae connect to a switchbox capable of selecting either the right or left antenna or both at once. A single coaxial cable runs from the switchbox to the antenna connector on the telemetry receiver. The audio output (labeled "headset") on the receiver is connected to the aircraft intercom system with a ¼ inch phone cable, either plugged directly into the intercom (if a dedicated telemetry input is provided) or through a splitter, available at Radio Shack.

2. *Homing in on a radio collar.* The aircraft proceeds to the area where the first collared animal is thought to be, with the antenna switchbox in the "both" position, so that signals from both sides of the aircraft can be heard. The RF gain switch on the receiver should be turned all the way up, and the attenuation switch, if present, should be in the off position. The volume knob is adjusted for a comfortable level. When a signal is heard, the scanner is stopped on that frequency, then the switchbox is used to listen to each antenna separately, and the aircraft is turned in the direction of the louder signal. When the aircraft is headed directly toward the source of the signal, the volume from the left and right antenna are equal, or the signal may be lost altogether because neither antenna is pointed toward the signal source. As the radio collared animal is approached, the RF gain knob is turned progressively lower, and when the signal is quite loud the attenuation switch (if present) is turned on if needed. The lower gain level makes it easier to detect slight differences in signal strength between the two antennae.

If the collar is passed, there will be an abrupt decrease in signal strength. The aircraft is maneuvered around the signal while using the switchbox to select between the antennae. When the aircraft is circling the collared animal, the signal from the inside wing is consistently louder, all around the circle. In open country, particularly in winter, the collared animal and its packmates may already have been sighted by this time. If the wolf is concealed by vegetation, it may never be seen, but the accuracy of the tracking system should allow its location to be estimated to within 100 meters or less. Once the pilot and observer have seen the animal or determined its location, the site is waypointed with the aircraft's GPS receiver. Care must be taken to mark the waypoint when the aircraft is directly over the animal location.

3. *Data recording.* Once a collared wolf has been located, that animal's line on the data sheet is filled out. Where the coding of information on the data sheet isn't intuitively obvious, a key at the bottom of the sheet contains guidelines for recording data (see Appendix A). The columns on the data sheet:

LOCATION: A verbal description of the wolf's location (drainage, distance from landmark).
GNO: Group number. If desired, individual locations with one or more collared wolves can be asssigned a unique group number to help facilitate the identification of unique locations for home range calculations.
TIME: 24 hour Alaska time.
MO: mode, either 1 (active), 2 (mortality mode), 3 (capture), 4 (not heard).
VI: visual. Either 1 (collared animal seen), 2 (group seen but not the collar), or 3 (no visual).
AD/PUPS: Number of adults and pups seen, as in 2/4 (2 adults, 4 pups).
MOR?: Could there be more wolves present than were seen? (1 = Yes, 2 = No).
BEH: R = resting, T = traveling, F = feeding, I = interacting, S = standing, O = other.
    Record the number of wolves engaged in each activity, as in 7R, 2S.
KILLS: Data on prey kills found with collared wolves. Covers the next three columns.
    SPP: The species of the kill. M = moose, C = caribou, S = sheep, O = other.
    AG/SX: Age (AD = adult, YRL = yearling, YOY = calf or lamb), Sex (M or F) of kill.
    %EAT: The percent of the carcass that appears to have been consumed already.
GPS: The latitude and longitude of the collar location, as determined by the aircraft's GPS.
    Latitude and longitude are recorded in degrees, minutes, and decimal minutes.
    Latitude and longitude are recorded in the datum WGS84.
    (At present, the coordinates are read from the aircraft GPS and recorded by hand on the data sheet. Waypoints may also be downloaded from the GPS to a computer and pasted into a database. Newer technology like the ATS R4500 receiver will allow location data to be stored in the telemetry receiver itself, eliminating manual transcription and its potential for errors.)
COMMENTS: description of activity, numbers and colors of wolves, den sites etc.

If associated animals are on adjacent rows of the data sheet, ditto marks can be used to indicate that data is the same. Associated animals should be noted in the comments column. Space at the bottom of the data sheet is used to record more extensive comments.

4. *Continuing the flight.* Once a collared animal has been located, its frequency is deleted from the memory bank. Typically, two or three memory banks on the R4000 receiver are used for different geographic areas, with missing or dispersing wolves being programmed into more than one bank. When searching for the next wolf to locate, the attenuation switch (if present) is turned off, the RF gain switch returned to its maximum position, the antenna switchbox is returned to the "both" position, and the aircraft regains altitude. Because VHF telemetry signals are more or less "line of sight", the range at which collars can be heard varies greatly, and increases with increased altitude of the tracking aircraft. Practical range can vary from 60 kilometers or more when an animal is on a mountaintop with nothing between it and the aircraft to less than a kilometer when an animal is in a canyon or behind a mountain.

5. *Timing of radio-tracking efforts.* A primary goal of wolf monitoring is the determination of pack sizes. A population estimate will be made based on early winter (October-November) pack sizes and another based on late winter (March-April) pack sizes. Typically, wolf packs decrease in size over the winter because of mortality and dispersal of wolves. Mortality and dispersal are not restricted to the winter period, but pack counts are usually not possible in summer because wolves are less visible and packs are less likely to be traveling together. In fall and early winter, it is often possible to differentiate pups from adults, so that pup production for that year (actually, pup production and survival to fall) can be estimated. By mid-winter, pups usually cannot be differentiated from adults from the air. When wolves are radio-tracked in winter, observers try to get accurate counts of wolf pack members on repeated occasions, until confident that wolf numbers for each pack have been determined.

   The goal of radio-monitoring efforts is to get about two locations per month for each collared animal. In order for consecutive locations to be considered independent from one another, they should be at least several days apart in time. Spreading the radio-tracking effort out over the whole year ensures that the data represent the whole seasonal pattern of movements by a wolf pack. Typically, wolf packs occupy larger areas in winter than in summer, when they are likely to be tied to a den site by relatively immobile pups. In fall, when pups become capable of traveling with adults, packs expand their movements to include their entire territory. Some packs make extensive forays away from their usual territories in winter, or may even migrate, so that the winter territory does not include the den area. Such moves are probably caused by significant shortages of prey animals in the territory. Extraterritorial movements by packs increase the chances of intraspecific strife and killing by neighboring packs.

   Individual parks may tailor monitoring efforts in order to answer specific research or management questions. Examples include the identification of den sites so that they can be protected from disturbance; the identification of movements outside of park boundaries that can expose wolves to greater danger from humans; movement and food habit studies with implications for subsistence harvest regulations; survival and mortality data for population modeling; behavioral research; and food habits and kill rate data. Once the investment has been made to place radio collars on wolves, it is desirable to get the maximum benefit from the effort for monitoring, research, and management goals.

6. *Developing and using wolf location databases.* Microsoft Access databases will be developed for storing wolf location data, to replace dBase and Excel files that have been used

for nearly 20 years to store wolf location data from DENA and YUCH. Documentation and guidelines for data entry will be developed along with the new databases. Some data entered in a single column on the data sheet will need to be parsed into separate fields of the database, for instance the various numbers of wolves engaged in activity categories, the numbers of adults and pups, the colors of adults and pups. Standard pack abbreviations are used so that pack territories can be analyzed. Data entry is easiest when the one-screen data entry format is arranged similarly to the field data sheet.

Wolf telemetry data consists of locations of individual collared animals, which may or may not belong to a pack and may change pack affiliation. However, data analysis will deal primarily with pack territories. Therefore, it is important that a wolf's pack affiliation be correctly recorded and entered, and changed if the wolf disperses or otherwise changes pack affiliation. Pack territory calculations are sensitive to the number of unique locations that they are based on, so a group number may be assigned to each radio-location so that multiple collars at a single site can be identified and not counted as independent locations.

7. *Verification of wolf location data.* Once databases are in place, data entry should take place as soon after a telemetry flight as possible. Until we have the capability of storing wolf location information in nonvolatile memory in the telemetry receiver, we rely on hand-copying and manual data entry. Such operations inevitably allow for errors to enter into the data. An important step in data verification is to take the animal location data, after it has been entered into the database, and place it on a map using ArcGIS or other mapping software, so that the flight observer can verify that the data as entered represent the correct locations. This is an easy procedure that is capable of picking up nearly all significant errors in the recording of latitude and longitude coordinates

V. Response to wolf mortalities.

Radio-tracking of collared wolves inevitably requires responding to "mortality" signals (collars that are beeping at twice the normal pulse rate because they have not moved for 10 hours). Experience has shown that wolves very seldom remain still long enough for their collars to enter mortality mode while still attached to a living wolf. It can be expected that any collar in mortality mode represents a dead wolf, a collar that has come off of the wolf, or rarely, a collar with a malfunctioning motion sensor. Although the fates of individual wolves are not a central part of data gathering for this population-level monitoring protocol, they can provide valuable information for interpreting population-level changes.

In Alaska, investigating wolf mortality signals usually requires a helicopter to reach the site. Antenna mounting systems are available for all models of helicopters commonly used for wildlife work, so that the helicopter can home in on the signal. Alternatively, if accurate coordinates were obtained when the mortality signal was first tracked, the helicopter may simply land near those coordinates, allowing the investigators to home in on the collar on the ground. A portable handheld telemetry antenna is needed for this tracking. Folding, 3-element antennae and flexible 2-element units, manufactured by ATS and Telonics, respectively, are convenient to carry and effective. As with aerial telemetry, it is necessary to decrease the gain setting on a receiver as a signal is approached, in order to maintain directionality. At a high gain setting, the signal may seem to be of equal strength at all points of the compass. Decreasing the gain and moving to a different vantage point are the best strategies for overcoming a lack of obvious directionality in a signal. Once the tracker is close to the collar, it may be easiest to disconnect the antenna and simply walk about with the receiver, searching for the loudest signal until the a dead wolf or a chewed-off collar are stumbled upon. A signal coming from under snow presents unique difficulties, because a great deal of shoveling may be required to reach the collar. Suspending the receiver, without antenna, over the surface of the snow and moving it about can be very useful in pinpointing where to dig.

Chewed-off radio collars are a common source of mortality signals. Packs of wolves have been observed to develop the habit of chewing their packmates' collars off, sometimes within a few days after the collar is attached (Thiel and Fritts, 1983, Mech et al. 1998). It should not be assumed, however, that a chewed-up collar was necessarily chewed off a living wolf, as some dead wolves are consumed by wolves or other species, and their collars may be chewed and carried off. Wolves occasionally are able to slip radio collars off over their heads. This usually happens soon after collaring, particularly when collars are left oversized on a young animal to allow for growth.

Most collars that are heard in mortality mode will represent an actual wolf mortality. Prompt investigation of mortalities makes it more likely that data will not be lost due to decomposition or scavenging. When a relatively intact carcass can be obtained, necropsy should be performed by a pathology lab, such as the ADF&G laboratory in Fairbanks. Ideally, a carcass should be refrigerated but not frozen, to preserve histology, but field conditions do not usually allow a choice of storage temperature. If transportation of the carcass is impractical or the remains are too decomposed for necropsy, a field necropsy may be performed. A thumb-sized chunk of skeletal muscle should be collected from each necropsied wolf for genetic samples. Each sample should be placed in two Ziplock® bags and labeled in Sharpie® permanent marker with the collection date, sex, animal id number, park unit, pack id, species and tissue type. These samples should be kept frozen and transported to NPS in Fairbanks for sub-sampling.

A full discussion of necropsy procedures is beyond the scope of this protocol, but some general guidelines follow. A sample necropsy data form is found in Appendix A. The most common cause of death for wolves in protected areas of Alaska is being killed by other wolves. Wolves are seldom attacked by packmates; rather they are killed by the members of neighboring packs. The location of a dead wolf may in itself provide a clue to its fate, if wolves from other packs are in the area, or if the wolf was outside its normal territory when it was killed. Wolves killed by neighboring wolf packs wolves commonly don't show extensive external injuries, blood, or torn skin, although massive damage exists under the skin. If a carcass isn't frozen, skinning out the neck and forequarters may reveal the damage caused by a wolf attack. The skulls of wolf-killed wolves commonly are perforated by tooth holes, particularly on the rostrum. Wolf-killed wolves are seldom eaten by their attackers. The attacks seem to be related to territoriality, not predation. But dead wolves are often eaten by scavengers, which may include other wolves. Another clue to the fate of a wolf that may remain even when little is left of the carcass is bone marrow. By cracking open a femur, humerus or other large leg bone, and examining the color and consistency of the marrow, an idea of the nutritional state of the wolf can be learned. Red, gelatinous or watery bone marrow (or, after drying, a bone with almost nothing in it) indicates a starving wolf. Pink, waxy marrow indicates a better-nourished wolf. The simplest investigation of a wolf mortality should include photographing the site and collection of the skull and a large leg bone. If illegal activity may have been involved, the site should be left alone until it can be examined by law enforcement personnel.

VI. Addresses and Contact Information

Wolf necropsy:	Dr. Kimberly Beckmen Wildlife Veterinarian Alaska Department of Fish & Game Division of Wildlife Conservation 1300 College Road Fairbanks, AK 99701 Kimberley_beckmen@alaska.gov	(907) 459-7257
Telemetry Equipment:	Telonics, Inc. 932 E. Impala Ave. Mesa, AZ 85204-6699 www.telonics.com	(602) 892-4444
	Advanced Telemetry Systems 470 First Avenue North Box 398 Isanti, MN 55040 www.atstrack.com	(763) 444-9267
	Communications Specialists Inc. 426 West Taft Ave. Orange, California 92865-4224 http://www.com-spec.com/r1000/r1000.htm	(800) 854-0547
Flight suits, PPE	Flight Suits/Gibson & Barnes 1675 Pioneer Way El Cajon, CA 92020 www.flightsuits.com	(800) 440-5904
Aircraft Procurement & Aircraft Safety Training	USDI Aviation Management Directorate 4405 Lear Court Anchorage, AK 99502-1032 www.amd.nbc.gov	(907) 271-6061

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Standard Operating Procedure (SOP) # 3 Data Analysis and Reporting

Version 1.01 (July 2009)

Revision History Log:

Prev. Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #
1.00	July 2009	Thomas Meier	Minor corrections	Preparation for publication	1.01
			Updated Figure 2		

This Standard Operating Procedure explains the procedures for calculating wolf population size and density estimates and other population parameters, and procedures for reporting wolf monitoring data. The steps for changing the protocol are outlined in SOP #9, "Revising the Protocol".

I. Estimating Wolf Population Parameters from Radio-Tracking Data

Procedures:

1. *<u>Data sorting and validation.</u>* Wolf radio-tracking data are made up of locations of individual collared animals. Wolf population parameters are estimated based on the territories and sizes of wolf packs. Therefore, a crucial step in data analysis is the assignment of pack status to a wolf. Most wolves are identified as members of a particular pack at the time of capture, and for many wolves that pack assignment doesn't change during the time that they are monitored. But wolves may disperse away from their packs and live alone for a time, start new packs or be incorporated into existing packs. Wolf packs themselves have finite lifespans, and may split in two, or disappear altogether through the dispersal or death of their members.

   Before beginning to analyze wolf population data, the database of wolf locations should be reviewed to ensure that pack abbreviations are consistent, and that the pack assignments for individual wolves are appropriate to that animal. By sorting the database by pack affiliation, wolf ID and date, a clear picture can be gotten of the entire data set, and the pack status of each animal reviewed over the course of the monitoring period.

   A further complication of wolf population analysis is the identification of locations that should not be included in the analysis. These can result from a number of causes. Mistakes in the recording of wolf locations should have been detected in the data verification process described in SOP#2, Techniques for Radio-Tracking of Collared Wolves. If wolf locations are examined with an appropriate map program, the locations can be visually inspected for gross errors while the flight is still fresh in the observer's mind.

   Other anomalous data can result from pre-dispersal movements by wolves preparing to leave their packs, and from extraterritorial forays by individual wolves and packs. Assuming that virtually all suitable habitat in interior Alaska is occupied by wolves, the mosaic of wolf pack territories in the monitoring area is just part of a larger mosaic of wolf packs that surrounds it on all sides, except where major mountain ranges or water bodies prevent wolves from living. Forays by wolf packs outside their usual territories thus do not change the density of wolves overall. Rather, they just change the degree of overlap between wolf pack territories. But in any monitoring effort, wolf densities are estimated over some defined area, and rare, long-range movements of wolf packs outside their normal range can greatly affect our estimate of the size of their territory and the perimeter of our monitoring area. The identification of which locations we choose to consider valid, and which we choose to leave out of our analysis, therefore becomes an important decision.

It has been common in wolf research and monitoring efforts for extraterritorial forays to be identified on an ad hoc basis, by researchers who were familiar with the wolf packs being studied and knew where they belonged and where they didn't. Clearly, a more systematic way of defining extraterritorial forays is needed. This question does not affect the process of data collection and should be re-examined, and the backlog of data re-analyzed, as the monitoring effort progresses. Our initial approach will be to use minimum convex polygons which exclude data points representing obvious extraterritorial movements. However, more systematic means of identifying anomalous locations should be investigated (Breck and Biggins 1997). The use of kernel estimators for territorial delination, as an alternative to minimum convex polygons, will also be investigated (Worton 1989).

2. *Determining pack territories from radio-tracking data.* When data have been validated and grouped by pack affiliation, a home range program (Weinstein et al. 1997, Hooge and Eichenlaub 2000) is used to generate minimum convex polygon territories (Odum and Kuenzler 1955) for each monitored wolf pack. One program that is compatible with newer versions of ArcGIS is Hawth's Tools (www.spatialecology.com). A 95% minimum convex polygon, using the 95% of the data that generate the smallest territory for each pack, may be used in an attempt to exclude extraterritorial forays. Although the minimum convex polygon may not provide as much biological insight as more complex calculations of wolf habitat use, it lends itself to wolf density estimation over a mosaic of pack territories, and is adequate for our purposes. Various studies have estimated that 30 to 100 independent, year-round locations are needed to adequately describe a wolf pack territory (Fritts and Mech 1981, Scott and Shackleton 1980, Bekoff and Mech 1984, Fuller and Snow 1988). The large pack territories of wolves in Alaska probably require a relatively greater number of radio-locations to adequately define them. Burch (2001) found that there was no "magic number" of radio locations guaranteed to adequately describe a wolf pack territory, but that monitoring a number of adjacent packs in a block allows territories to be defined with fewer locations. He recommends combining data over years if one year's sampling effort is insufficient to define territories. The crucial question is whether pack territories are sufficiently defined to ensure that other, undetected wolf packs don't exist within the mosaic of packs being monitored. Weather and budget constraints seldom allow the collection of more than 30 independent locations of collared wolves per year. The combination of location data from two years has been commonly used when wolf densities are estimated annually (Adams et al. 1997, Burch 2001.). As an initial approach to analyzing CAKN wolf monitoring data and estimating wolf densities, we will use location data for each pack for the previous two years to calculate

minimum convex polygon territories. Increased use of GPS collars, which typically provide ≥300 locations per year, will solve the problem of sample size in delineating pack territories.

3. ***Determining the area of the population estimate.*** In order to estimate wolf density over a number of pack territories, we will generate a population estimating area by combining the territories of all monitored packs in an area. Ideally, these should form a contiguous block with considerable overlap between the packs and only minor gaps between them, so that we can be confident that all packs in the area are being counted. Figure 1 shows such a block of packs, from Denali National Park and Preserve, in spring 2009. In this example, a significant gap between defined pack territories can be seen. Aerial surveys suggested that no other packs were to be found in this area, and it was assumed that all wolf packs within the survey area were being counted. This method of estimating wolf densities results in a different survey area being used for each year's estimate, but is probably more biologically sound than choosing an arbitrary boundary and estimating wolf numbers within an area that contains various fractions of the territories of many wolf packs.

4. *Early and late winter wolf population and density estimates.* Wolf packs can usually be seen and counted during the period of snow cover in Alaska, roughly from October through April. Most wolf packs show some decrease in size during this period, as members die or disperse away from the pack. Monitoring flights will concentrate on getting pack size estimates for each pack during the early winter (October-November) and late winter (March-April) periods. Separate population estimates for early and late winter will be generated simply by summing the sizes of the packs in the survey area. Density estimates will be calculated by dividing the population estimate by the area of the (not convex) polygon that combines all of the pack territories, and converted to wolves per 1000 square kilometers, a common measure of wolf density (Fuller, 1989). Early- and late-winter density estimates during the same winter will use the same aggregate pack area, normally calculated in late winter using the previous two years' location data. Wolves that do not live in packs will not be included in these estimates. In nearly 20 years of wolf research at Denali, only one territorial, solitary-living wolf has been found in the park. Nearly all "lone wolves" are wolves that are in transition between one pack-living situation and another. The larger pack sizes seen in early winter represent the high point of numbers of wolves traveling together in most packs, with the addition of that year's pups increasing the pack size prior to its diminution by dispersal and mortality. There is probably a greater proportion of dispersing wolves left uncounted in the population in late winter than early winter. It should be specified that our wolf population estimates are for pack-dwelling wolves, with the understanding that the numerical and biological significance of wolves that are temporarily between packs is not expected to be great.

5. *Other wolf population parameters.* As the numbers and distribution of wolves are monitored, other data become available. The sample of radio-collared wolves is not expected to be a random sample of the population, because we are trying to mark the dominant, breeding members of packs, and in any case different classes of wolves are unequally vulnerable to capture. Nevertheless, the survival and mortality of our sample of collared wolves provides valuable life history data on this species in areas where they are relatively protected from human harvest. Trends in survival and mortality over time may provide valuable clues for interpreting changes in numbers and distribution of wolves. The causes of death for collared

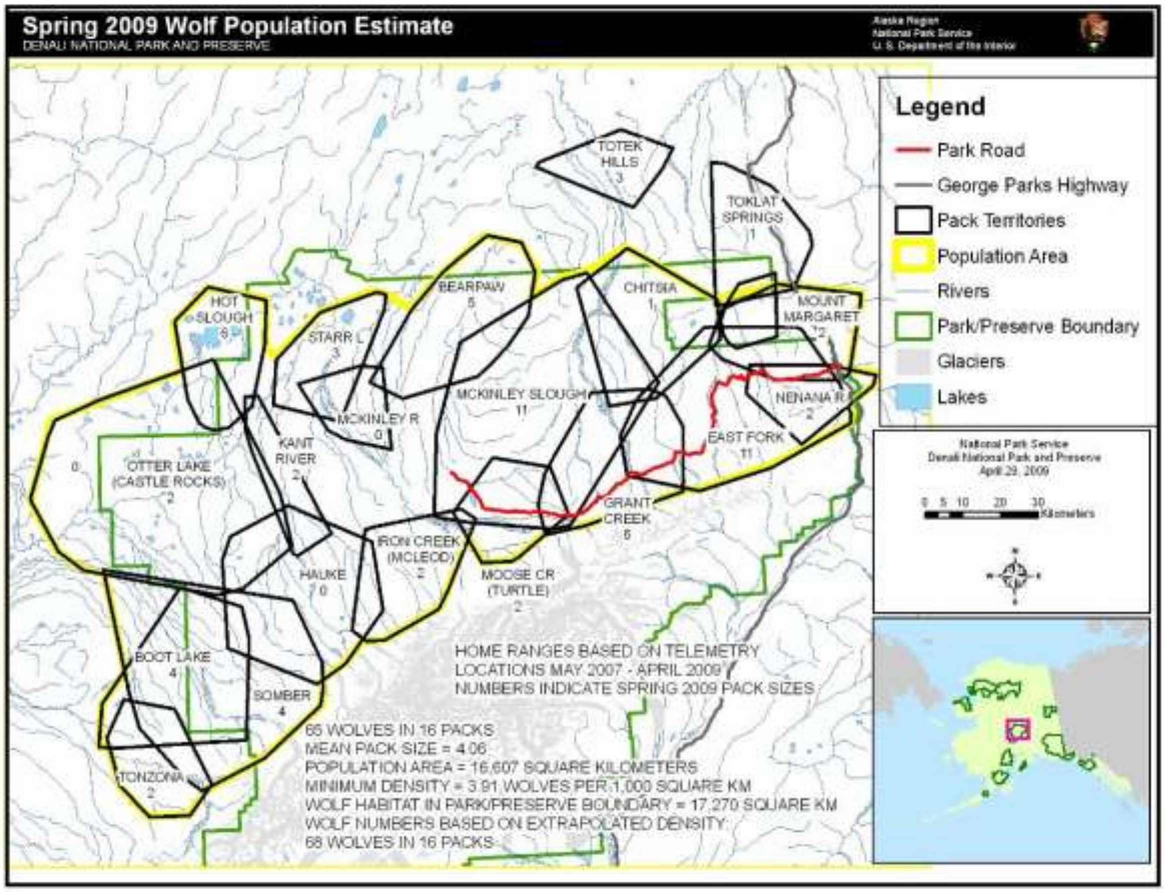

Figure 1. Spring 2009 wolf population estimate for Denali National Park and Preserve (Denali National Park and Preserve, 2009)

wolves, obtained by examining and retrieving wolf carcasses, are similarly valuable for understanding population dynamics. Annual estimates of pup recruitment are available from early winter pack observations. Trends in the morphology, disease profile, and genetic makeup of wolves, from data gathered as a sidelight to capture and population monitoring, may prove to be important measures of long-term impacts on the species in Alaska

6. *Analysis of wolf population changes over time.* Wolf population density in Denali has been found to change on an irregular cycle, in response to prey numbers and availability and a variety of other causes (Mech et al. 1998; Adams, Mech, and Stahlnecker 1997; Denali National Park and Preserve 2009). A two- or three-fold change in wolf density over several years may thus be of little significance, and large changes can be expected to be countered by trends in the opposite direction soon afterward. These variations make it difficult to identify changes that may have biological significance, or that could serve as warnings that the system has somehow changed. Further analysis of archived wildlife population data may allow models of predator numbers, prey numbers and weather to be developed, allowing some predictions of system behavior. Overlaying genetic and radio-telemetry data may assist in clarifying breeding events, migration and dispersal patterns and levels or relatedness within and among packs (Lehman et al. 1992, Meier et al. 1995, Smith et al. 1997). But it will remain a challenge to interpret changes in wolf numbers and distribution, and to identify truly unusual events. Statistical methods for identifying long-term trends in the wolf population have yet to be developed, and investigation of these methods should form a part of the monitoring program.

II. Reporting of Wolf Monitoring Data

Procedures:

1. *General features of monitoring reports.* Reports generated by the wolf monitoring effort, and the vital signs monitoring program in general, will be designed to be promptly produced, appropriate to their target audience, widely available, and visually accessible. Concise summaries will be a part of each report produced. Reports will conform to guidelines set by the Central Alaska Monitoring Network and the Alaska Region Inventory and Monitoring Plan. Graphical methods, maps, and other visual aids will be used to make results readily understood.

2. *Annual administrative report and workplan.* This report is designed to be incorporated into the annual Report to Congress on the nationwide Vital Signs Monitoring Program. It will be produced in the fall of each year. The report will account for the expenditure of funds and FTEs, and describe the objectives, tasks, accomplishments, and products of the monitoring effort during the previous fiscal year. The report is designed to improve communication within each park, the Central Alaska Network, the Alaska Region, and the National Monitoring Program. Its intended audience includes park superintendents, network staff, regional coordinators, and Servicewide program managers. The report will be written by the park biologists implementing the wolf monitoring protocol, and reviewed and approved by CAKN, Regional Office and Servicewide Program managers.

3. *Annual report.* An annual report will be prepared for publication each March, reporting on wolf monitoring activities during the previous phenological year (October-September). Thus, it will contain early winter and late winter population estimates for the previous winter. The annual report will provide a summary of all wolf monitoring activities and data for the year, and describe the current condition of the wolf population in the areas monitored. Any changes in monitoring protocols will be documented. The intended audience for the annual report includes park resource managers, network staff, and external scientists. The report will be written by the park biologists implementing the wolf monitoring protocol, and peer-reviewed within the Central Alaska Monitoring Network.

4. *Other reporting.* In addition to the two scheduled annual reports, results of the wolf monitoring effort will be presented at the biennial CAKN Vital Signs Monitoring Conference, and at other symposia, conferences, and workshops. Wolf monitoring data will be provided to park interpretive staff for written and oral presentation to visitors. Significant findings will be reported in scientific journals and popular publications.

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Standard Operating Procedure (SOP) # 4 Data Management

Version 1.0 (August 2009)

Revision History Log:

Prev. Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

Note: This SOP, which is under development, will document the Microsoft Access database for storage of wolf monitoring data and provide instructions for the maintenance and distribution of monitoring data associated with the Wolf Monitoring Protocol for the Central Alaska Monitoring Network. Procedures for data handling and quality assurance/quality control for all monitoring protocols implemented by the Central Alaska Network are detailed in the program's Data Management Plan. The Access databases for wolf monitoring data are currently (as of August 2009) being constructed and documented. These databases will be designed to accommodate all wolf monitoring data collected since 1986 in DENA and 1993 in YUCH.

Standard Operating Procedures (SOPs) # 5, # 6, #7 Genetic Analyses

Standard Operating Procedure (SOP) # 5
Techniques for Assessing Genetic Variation of Mammalian Populations Using Nuclear-DNA Microsatellite Genotyping and Mitochondrial-DNA Sequencing at USGS, Biological Resources Division, Alaska Science Center, Molecular Ecology Laboratory (USGS-BRD-ASCMEL), Anchorage, Alaska

Standard Operating Procedure (SOP) # 6
Techniques for Assessing Genetic Variation of Mammalian Populations with Nuclear-DNA Microsatellite Genotyping and Mitochondrial-DNA Sequencing at USGS, Biological Resources Division, Alaska Science Center, Molecular Ecology Laboratory (USGS-BRD-ASCMEL), Anchorage, Alaska

Standard Operating Procedure (SOP) # 7
Techniques for Assessing Genetic Variation of Mammalian Populations with Nuclear-DNA Microsatellite Genotyping and Mitochondrial-DNA Sequencing at USGS, Biological Resources Division, Alaska Science Center, Molecular Ecology Laboratory, Anchorage, Alaska

Due to length, complexity, and frequency of updates, these SOPs have been omitted from this publication. Please contact the authors for current information.

Standard Operating Procedure (SOP) # 8 Handling GPS/ARGOS Wolf Location Data

Version 1.0 (August 2009)

Revision History Log:

Prev. Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

Wolf monitoring projects in CAKN currently use two types of radio collars. The first type, used in Denali National Park and Preserve since 1986 and Yukon-Charley Rivers National Preserve since 1993, is the Telonics VHF transmitter mounted on a flexible collar, which can be homed in on from an aircraft so that the wolf can be located and observed. The second type, used since 2003, is the Telonics GPS/ARGOS collar, which in addition to the conventional VHF transmitter contains systems that determine the collar's location using the GPS satellite system, store location data inside the collar, and periodically upload location information through the ARGOS satellite system.

The GPS system consists of approximately 30 medium Earth orbit satellites at an elevation of 20,200 km that transmit precisely timed radio signals. GPS receivers use the signals from four or more satellites to calculate their location. GPS/ARGOS animal collars determine their location at some pre-programmed interval and time (in the case of the wolf collars used in CAKN monitoring, locations are determined daily at 0700 Alaska Standard Time). The collars have sufficient data storage capability to store all locations gathered during the lifespan of the collar, typically 2.5 to 3 years. In addition, the collars are programmed to upload location data every 6 days to ARGOS satellites located 850 km above the earth. ARGOS satellites receive and store the data, and relay it to a system of ground antennae. ARGOS processing centers (in the U.S., CLS America in Largo, MD) collect and process incoming data and distribute them to users via daily email messages.

Location data from Telonics GPS/ARGOS collars (currently the Telonics Gen 4 collar system) is encrypted to conserve bandwidth, and must be decoded using Telonics' proprietary Telonics Data Converter© (TDC) software. CAKN has contracted with Dr. Edward DeBevec of the University of Alaska, Fairbanks, to produce a web-based platform incorporating TDC and other functions, so that ARGOS data can easily be decoded, checked for anomalous or redundant values, tagged with wolf numbers and other identifying information, and stored in an Excel spreadsheet. Using this website and a GIS program such as ArcGIS©, wolf locations can be placed on a map and examined within minutes of the reception of the email containing the data.

ARGOS location data is received as text in an email message. Multiple ARGOS emails received from CLS America can be strung together for processing by marking them in Lotus Notes and forwarding them to create a single message. Text data is exported using the File/Export function in Lotus Notes, changing the Word Wrap Within Documents setting (in a popup box) to "Wrap Words at 100 Characters Per Line," and exporting the file.

On the CAKN/UAF web platform, the input file is specified, and dropdown boxes used to select the park unit of interest and to choose between one location per day (appropriate for wolf collars) or multiple locations per day (for collars currently deployed on moose). Other filtering parameters can also be entered, such as inclusive dates and bracketing coordinates to help detect data errors. When the "Submit" button is pushed, the location data appears on screen. To produce an Excel file, data fields can be selected in the box at the bottom of the page and the "Download" button pressed. From the Excel file, locations can be viewed in ArcGIS by using the "Tools/Add XY data" function.

An administrative page on the CAKN/UAF website is used to maintain the list of active ARGOS identification codes, designating the animal ID, species, park, location (e.g. wolf pack), capture date, collar ID, and model of Telonics GPS/ARGOS collar. The collar model (Gen 3 or Gen 4) must be specified because different data encryption and decoding algorithms are used on the older and newer model collars. By including the capture date in the administrative database, a complete history of collar use and re-use is maintained, so that data from any time period can be processed. Dr. Debevec continues to update and modify the web-based processing tool to accommodate the needs of individual projects.

GPS/ARGOS wolf collars provide up to 365 locations per year for collared animals, greatly increasing the sample size of locations that can be used to calculate wolf pack territories. Wolves must still be located from aircraft using VHF telemetry, in order to observe wolf packs and get pack sizes, pup counts, den locations, physical descriptions of wolves, and other data that can only be obtained from direct visual observation. At present, the cost of Telonics Mod-515 VHF collars used on wolves is approximately $400.00. Telonics Gen-4 VHF/ARGOS collars cost About $3000.00, plus about $300.00 per year for ARGOS data charges. Combining conventional telemetry with GPS/ARGOS location data allows for monitoring the status and movements of wolf packs efficiently while minimizing cost and the intrusion of aircraft overflights.

Telonics GPS/ARGOS wildlife systems: http://telonics.com/products/gps4/

CLS America ARGOS data services: http://www.clsamerica.com/

CAKN/UAF web-based ARGOS data processing site:
http://nuna.uaf.edu:8080/argos/submit_argos_data.php

Standard Operating Procedure (SOP) # 9 Revising the Protocol

Version 1.0 (October 2004)

Revision History Log:

Prev. Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #

This Standard Operating Procedure explains how to make changes to the Wolf Monitoring Protocol Narrative for the Central Alaska Monitoring Network, and accompanying SOPs, and tracking these changes. Observers asked to edit the Protocol Narrative or any one of the SOPs need to follow this outlined procedure in order to eliminate confusion in how data is collected and analyzed. All observers should be familiar with this SOP in order to identify and use the most current methodologies.

Procedures:

1. The Wolf Monitoring Protocol Narrative for the Central Alaska Monitoring Network (CAKN) and accompanying SOPs have attempted to incorporate the most sound methodologies for collecting and analyzing wolf population data. However, all protocols, regardless of how sound, require editing as new and different information becomes available. Required edits should be made in a timely manner and appropriate reviews undertaken.

2. All edits require review for clarity and technical soundness. Small changes or additions to existing methods will be reviewed in-house by CAKN staff. However, if a complete change in methods is sought, than an outside review is required. Regional and National staff of the National Park Service with familiarity in large mammal research and data analysis will be utilized as reviewers. Also, experts in wolf research and statistical methodologies outside of the Park Service will be utilized in the review process.

3. Document edits and protocol versioning in the Revision History Log that accompanies the Protocol Narrative and each SOP. Log changes in the Protocol Narrative or SOP being edited only. Version numbers increase incrementally by hundredths (e.g. version 1.01, version 1.02, …etc) for minor changes. Major revisions should be designated with the next whole number (e.g., version 2.0, 3.0, 4.0 …). Record the previous version number, the date of revision, and the author of the revision. Identify paragraphs and pages where changes are made, and the reason for making the changes along with the new version number.

4. Inform the Data Manager about changes to the Protocol Narrative or SOP so the new version number can be incorporated in the Metadata of the project database. The database may have

to be edited by the Data Manager to accompany changes in the Protocol Narrative and SOPs. Post new versions on the inter-net and forward copies to all individuals with a previous version of the affected Protocol Narrative or SOP.

Appendix C. Supplemental Materials

wolfstat.doc: documentation for storing data on the capture and fate of individual wolves in wolfstat.dbf, later converted to wolfstat.xls (2003).

dwfld.doc: documentation for storing data on locations of collared wolves in dwfld****.dbf, later converted to dwfld****.xls (2000).

WOLFSTAT.WPD: Description of WOLFSTAT.DBF

L. Adams, 13 February 2003.

X:	TEMPORARY FIELD USED TO SELECT CASES AS NEEDED
WOLFNO:	WOLF NUMBER
RCAP:	RECAPTURE? 1 OR 0 (1 ONLY IF WOLF IS RECAPTURED AFTER A PERIOD THAT ITS STATUS IS NOT KNOWN [CHEWED OR FAILED COLLAR??]; IF A COLLAR FAILS BUT THE WOLF IS SEEN REPEATEDLY THEN RECOLLARED, I DID NOT CREATE A NEW ENTRY FOR THE WOLF WITH RCAP = 1)
CAPDATE:	ORIGINAL CAPTURE DATE (E.G 20020316)
MO:	MONTH OF CAPTURE (E.G 3)
SEX:	M OR F
AGE:	AGE ESTIMATE AT ORIGINAL CAPTURE (PUP, YRLG,AD2,AD3,OLD)
EAGE:	ESTIMATED AGE IN MONTHS
BYEAR:	ESTIMATED BIRTH YEAR
COLOR:	PELT COLOR
EARTAGS:	COLOR AND EARTAG NUMBERS (BLU L746,R747) UPDATE AS NEEDED WITH RECAPTURES
SERIAL:	RADIOCOLLAR SERIAL NUMBER; UPDATE AS NEEDED WITH RECAPTURES
FREQ:	RADIOCOLLAR FREQUENCY, UPDATE AS NEEDED WITH RECAPTURES
PACKHIS:	CHRONO. LIST OF PACKS
CURPAKC:	CURRENT PACK
PACKNOTES:	NOTES ABOUT PACK CHANGES, FOUNDINGS ETC.
STUDY:	IN STUDY AREA? Y OR N
COD:	CAUSE OF DEATH CODES
MISSING:	MISSING?? 1 OR 0. ANY ANIMAL FOR WHICH THE COLLAR WAS NOT RETRIEVED IS LISTED AS MISSING HERE, SO THESE INCLUDE DISPERSALS, HARVEST WITH NO COLLAR RETURNED, RADIOFAILURES, ETC.)
STATUS:	DESCRIPTION OF ANIMAL'S STATUS
DOD:	ESTIMATED DATE OF DEATH (20020602; MIDPOINT BETWEEN LAST ALIVE AND FIRST MORT)
AGEDEATH:	AGE AT DEATH IN MONTHS (TOM'S FIELD, I DIDN'T UPDATE)
FATEI,II,III:	COMMENTS ABOUT FATE OF WOLF
RD85-RD01:	RADIODAYS PER YEAR FOR SURVIVAL CALCULATIONS
TRD:	TOTAL RADIODAYS

DWFLD.DBF: Instructions for annual wolf radiotracking data**bases

L. Adams, 27 November 2000.

All wolf radiotracking data is entered in DBASE files with the name DWFLD**.DBF (DWFLD stands for DENALI WOLF FLIGHT DATA and ** representing the year (eg. 99,00, etc). Each year's data begins on 1 May of the year designated and ends on 30 April of the following year (DWFLD99.DBF begins 1 May 1999 and ends 30 April 2000).

1. To create a new database, open the current database (eg. DWFLD00.DBF), go to the command window and type the following commands:

    COPY STRUCTURE TO [NEW FILE NAME]
    USE [NEW FILE NAME] and begin data entry.

2. Whenever you are manipuating a database (sorting, generating fields, etc.) always have keep a copy of the original file and do all the manipulating in a duplicate file. When you are done, rename the original file with a recognizable name (egTemp00.dbf) and hold on to it only until you are sure that the manipulated file is correct and complete, then rename the manipulated file to the appropriate field name (eg. DWFLD00.dbf)
To create a temporary file:

    Open the file you want to work with.
    USE [ORIGINAL FILE NAME (eg dwfld00.dbf)]
    COPY TO [NEW FILE NAME (eg temp00.dbf)]
    USE [NEW FILE NAME] When finished, change file names accordingly in Windows Explorer.

3. When database is complete, sort on date as follows:

    USE [ORIGINAL FILE NAME]
    SORT ON DATE TO [TEMPORARY FILE NAME]

Then in Windows Explorer, rename the original file to a temporary name; and rename the sorted file appropriately (DWFLD**.DBF).

4. Use standard wolf pack abbreviations only!!!! See attached list (packabs.doc).

5. Each observation with coordinates must have a group number. If the pilot/observers did not provide them, then they need to be added at the time of data entry. Observations without coordinates do not get group numbers (GNO = -9).

6. -9 = missing data throughout.

FIELDS (Num, 8.0 = Numeric field, 8 spaces wide with 0 decimal places; Char,10) = Character field 10 spaces wide):
DATE (Num, 8.0): date formatted as '20000506' for 6 May 2000
TAIL (Num, 3.0): last 2 digits of aircraft tail number (eg. 4627Y = '27', 83404 = '04')
OBS (Char, 3): initials of observer
WOLFNO (Char, 5): animal number (eg. '9634') This is a character field so that zeroes show up for animals initially captured in 2000 and beyond (eg. '0089').
ASSOCITS (Char, 15): animal numbers of other radiocollared wolves in observation (eg.'9634,0089').
PACK (Char,11): use specific pack name abbreviations only! It is very important that only a standard set of abbreviations are used because this field is used to sort locations by pack.
GNO (Num, 2.0): Group number. Every observation with coordinates must be assigned a group number. These can be recorded as the data is collected or added in later at data entry. For animals that are heard, but no specific location is provided, enter -9.
TIME (Num, 5.0): Military time (eg 1434 for 2:34pm)
MO (Num, 3.0): Mode 1 = active, 2 = mortality, 3 = capture, 4= listened for but not heard.
VI (Num, 3.0): Visual observation? 1 = saw individual, 2 = saw group and heard signal, but did not see all collared individuals at location, 3 = no visual.
TOT (Num, 3.0): Total number of wolves observed
MORE (Num, 2.0): There could have been more wolves?? 1 = yes, 2 = no.
NR (Num,2.0): Number of wolves resting
NT (Num,2.0): Number of wolves traveling
NF (Num,2.0): Number of wolves feeding
NI (Num,2.0): Number of wolves interacting
NS (Num,2.0): Number of wolves standing
NO (Num,2.0): Number of wolves exhibiting some other behavior (should be described in remarks)
KILSP (Char, 3): Species of kill; M = moose, C = caribou, S = sheep, O = other
KILAGE (Char, 3): age of kill: AD = adult, YOY = young of the year (should be substantiated in remarks)
KILSEX (Char, 3): sex of kill: M or F (should be substantiated in the remarks)
KILEAT (Num, 3.0): % eaten
GENERAL (Char, 42): General description of location. Specific locations (eg 2mi SW of VABM Edge; 3mi upstream EF Bridge) are preferred over very general locations (eg Slippery Creek, Herron, etc.)
COLOR (Char, 25): Any reference to color of wolves observed is recorded here
PUPS (Char, 25): Any reference to the number of pups and their colors is recorded here
REMARKS (Char, 70): additional explanation on colors or pups, info on kills, any other comments recorded here.
LAT (Num, 10.4): Latitude in decimal degrees (eg. 63.1426). This field is derived from data entry fields listed below.
LON (Num, 10.4): Longitude in negative decimal degrees (eg -151.0645). This field is derived from data entry fields listed below.
DATE ENTRY FIELDS FOR COORDINATES:
LAT_DEG,LAT_MIN,LON_DEG,LON_MIN (CURRENTLY USED) or
DLAT,DLON or
ZONE, EASTING,NORTHING.
ID (Num, 12.0): Id number for each observation with coordinates derived from date, tail, and gno as follows:
REPLACE ID WITH DATE*10000+TAIL*100+GNO FOR GNO > 0

Current data entry and derivation of location coordinates:
FIELDS:
LAT_DEG (Num, 10.0): Degrees of latitude entered here
LAT_MIN (Num, 10.4): Decimal minutes of latitude entered here
LON_DEG (Num, 10.0): Degrees of longitude entered here
LON_MIN (Num , 10.4): Decimal minutes of longitude entered here.

To generate LAT and LON use the following commands:
For LAT: REPLACE LAT WITH LAT_DEG+(LAT_MIN/60) FOR LAT_DEG > 0
For LON: REPLACE LON WITH (LON_DEG+(LON_MIN/60))*-1 FOR LON_DEG > 0

The Department of the Interior protects and manages the nation's natural resources and cultural heritage; provides scientific and other information about those resources; and honors its special responsibilities to American Indians, Alaska Natives, and affiliated Island Communities.

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