



Remote Biopsy Dart Sampling of Brown Bears





Alaska Region Technical Report Series

Natural Resources Technical Report NPS/AR/NRTR-2009-74.

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Front Cover: An adult female brown bear waits for her yearling along the bank of Brooks River, Katmai National Park, Alaska. Photo by Howard Maltby, NPS.

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January 2009

**Alaska Region Natural Resources Technical Report
NPS/AR/NRTR-2009-74**



**National Park Service
U.S. Department of the Interior**

REMOTE BIOPSY DART SAMPLING OF BROWN BEARS

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Abstract

I used biopsy darts to collect skin tissue samples from brown bears (*Ursus arctos*) at Brooks River in Katmai National Park, Alaska, USA, on 154 occasions. The majority of 148 biopsy needles that were recovered retained a visible skin tissue sample that consisted of a small skin punch and a few mm of attached underlying tissue. The proportion of needles that retained a visible skin tissue sample was similar between July when bears were relatively lean and shedding winter coats and September–October when bears had put on considerable weight and had thicker winter coats. Bears typically reacted to biopsy darting by trotting away several steps.

Key Words

Alaska, Brooks River, brown bear, biopsy dart, skin biopsy, remote biopsy, *Ursus arctos*

Remote biopsy dart sampling enables researchers to obtain tissue samples from free-ranging animals without the risks associated with live capture (Karesh et al. 1987). A biopsy dart generally consists of a specially designed biopsy sampling needle mounted in place of a standard dart needle on the body of a projectile syringe. When fired, the biopsy dart hits the animal, cutting a section of skin and underlying tissue, then the dart bounces off the animal or falls to the ground after the animal moves around. The skin tissue sample is retained within the biopsy needle. Biopsy darts have been used to remotely collect skin tissue samples from a variety of free-ranging terrestrial mammals including cougar (*Puma concolor*; R. Beausoleil, Washington Department of Fish and Wildlife, personal communication), lion (*Panthera leo*; Spong et al. 2002), spotted hyena (*Crocuta crocuta*; East et al. 2003), warthog (*Phacochoerus africanus*; Muwanika et al. 2003), pronghorn antelope (*Antilocapra americana*; Carling et al. 2003), impala (*Aepyceros* spp.; Lorenz and Siegismund 2004), African elephant (*Loxodonta* spp.; Roca et al. 2001, Muwanika et al. 2003), and giraffe (*Giraffa camelopardalis*; Huebinger et al. 2002).

Observational sampling of brown bear (*Ursus arctos*) activity at Brooks River in Katmai National Park (KNP), Alaska, USA, has produced several years of detailed records of river use and

behavior for >60 different bears recognized based on observable physical and behavioral characteristics (T. Olson, National Park Service [NPS], unpublished data). To investigate genealogy and relatedness among bears at Brooks River, skin tissue samples were collected from bears there over a 3-year sampling period. It was important to be able to assign observational bear identification numbers to DNA collected so that observational records of individual bear behavior could be evaluated in light of kinship information. It was somewhat unclear to me how well remote biopsy darting would work to collect skin tissue samples from brown bears because of their potentially dense hair coat and thick fat layer. Still, I employed biopsy darting because it could potentially facilitate collection of skin tissue from specific individual bears known from observational sampling. Here I report on the effectiveness of remote biopsy darting to obtain skin tissue samples from bears at Brooks River, and responses of bears to those darting efforts.

STUDY AREA

I conducted biopsy dart sampling at Brooks River in KNP, Alaska, during July and late September through early October of 2005–2007 when brown bears were frequenting the area to feed on migrating and spawning sockeye salmon (*Oncorhynchus nerka*). In recent years, >60 different

independent bears have been documented using the area during these periods (T. Olson, NPS, unpublished data). Brooks River was bordered by mixed white spruce forest (*Picea glauca*) with open marshes near the river mouth. Three elevated public viewing platforms (2 connected via an elevated boardwalk system) were available along the south side of the river to view bears securely separated from the sometimes heavy bear traffic along the river. The viewing platforms and boardwalk system were elevated 2.4–3.0 m above ground level, and during peak visitation were sometimes occupied by ≥ 40 people.

METHODS

I collected skin tissue samples remotely from brown bears via Dan-Inject™ (Dan-Inject ApS, Børkop, Denmark) biopsy needles mounted on Dan-Inject 1.5-ml dart syringes. I primarily used needles that were 20 mm in length ($n = 122$), but to determine whether a shorter needle could collect sufficient skin tissue I also used some 10-mm needles ($n = 32$; both needles were 2 mm in diameter; Fig. 1). The biopsy darts were delivered using a Dan-Inject CO₂-powered rifle (model JM Special with rifled barrel extension). The Dan-Inject rifle was exceptionally quiet when fired, which helped to minimize any noise effects on bear behavior. To facilitate quick aiming, I fitted a red laser sight (Model LLC; BSA Optics, Inc. Fort Lauderdale, FL) to the rifle that projected a red dot onto the target.

I determined dart rifle pressure settings by adding approximately 2 bars of pressure to the settings listed in the manufacturer's standard pressure table (settings based on distance from the target animal). The additional pressure was intended to provide sufficient force for the biopsy dart to potentially bounce away from the animal (Dan-Inject 2004) while also minimizing the risk of dart syringe penetration. I estimated distances to target animals using a laser rangefinder (RX®-II; Leupold & Stevens, Inc., Beaver, OR). Due to presence of vegetative cover and considerations

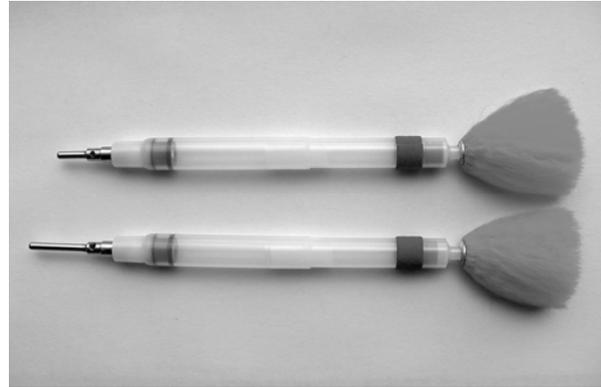


Figure 1. Biopsy darts (10-mm and 20-mm needles) used to remotely collect skin tissue samples from brown bears at Brooks River, Katmai National Park, Alaska, 2005–2007.

regarding safe dart retrieval, I limited sampling to distances of ≤ 15 m.

I used biopsy darts only on bears > 2 years of age, and I fired darts only at a bear's rump when the bear was perpendicular to me. No darts were fired when other bears were present within the potential target animal's likely escape routes or when a potential target animal appeared to be interacting with another bear. In addition, I did not sample bears that were sitting or lying down. I classified the reaction of each bear to darting as minimal (bear continued its ongoing behavior or modified its behavior slightly, such as glancing back toward the dart, flinching, or taking a few steps), moderate (bear modified its behavior in an obvious way beyond minimal reaction, such as trotting away < 15 m before returning to walking pace), or strong (bear moved away at a rapid pace > 15 m).

To avoid frequent close-range encounters with bears, I conducted most sampling from the public bear viewing platforms (sampling was conducted during periods when visitors were absent). Because the platforms were located immediately adjacent to the river, many bears passed close by the structures. I attempted to minimize the downward angle at which I fired darts by positioning myself on the platform access stairs/ramps when possible. At Brooks Falls where bear activity was sometimes especially high, dart

retrieval was often accomplished from the platform using a mechanical gripper (activated via a cord that was attached to the squeeze-handle) that was mounted on the end of an extendable pole. For each dart sample collected, I used a Dan-Inject venting pin to extrude the collected skin tissue from the biopsy needle (the pin was supplied by the manufacturer for this purpose) into a storage vial containing 100% ethanol as soon as possible after the dart was retrieved (Fig. 2).

The proportion of recovered biopsy needles that retained a visible skin tissue sample was lower for 10-mm needles than for 20-mm needles ($\chi^2_1 = 8.11$, $P = 0.004$). Therefore, to evaluate whether biopsy needle tissue retention was similar between July (when bears were relatively lean with shedding winter coats) and September–October (when bears were considerably heavier with heavier winter coats), I limited the χ^2 analysis to 20-mm needles (20-mm needles: $n_{Jul} = 65$, $n_{Sep-Oct} = 53$; 10-mm needles: $n_{Jul} = 16$, $n_{Sep-Oct} = 14$). I also used a χ^2 test to determine whether the proportion of darting events in which bears reacted strongly was similar ($P \geq 0.05$) between adults (believed to be >5 years old) and subadults (believed to be <5 years old).

RESULTS

I used biopsy darts to collect skin tissue samples from brown bears at Brooks River on 154 occasions. The dart was recovered in 148 of 154 cases (96%) and was lost in the river in the remaining 6 cases (4%). Although the dart was designed to bounce back out of the animal (Dan-Inject 2004), this occurred in only 12% ($n = 10$) of darting events in July and 18% ($n = 13$) of darting events during September–October. More typically, the dart fell out within a few seconds after the bear moved around. Reactions of bears to the darts varied. Bears showed moderate reactions in 45% of darting events ($n=70$), and minimal or strong reactions in 29% ($n = 44$) and 26% ($n = 40$) of



Figure 2. A bear skin tissue sample is extruded from a 20-mm biopsy needle into a storage vial, Katmai National Park, Alaska, September 2005.

darting events, respectively. Strong reactions to the darts were more often exhibited by adult bears (minimal to moderate reactions = 76, strong reactions = 36) than by subadults (minimal to moderate reactions = 38, strong reactions = 4; $\chi^2_1 = 8.13$, $P = 0.004$).

The majority of (84%) biopsy needles that were recovered (67% of 30 10-mm needles; 88% of 118 20-mm needles) retained a visible skin tissue sample that usually consisted of a small skin punch and a few mm of attached underlying tissue (Fig. 2). The proportion of 20-mm needles that retained a visible skin tissue sample did not differ between July when bears were relatively lean and shedding winter coats and September–October when bears had put on considerable weight and had thicker winter coats ($\chi^2_1 = 0.03$, $P = 0.869$; July = 88%, Sep–Oct = 89%).

DISCUSSION

Biopsy needles occasionally did not retain a visible skin tissue sample. This occurred more frequently with 10-mm than 20-mm needles. It seems likely that coat length and thickness were related to the difference in sampling success rate between the 2 needle lengths. A longer needle would be more likely to penetrate through the coat. It should also be noted that biopsy darts that didn't strike perpendicular to the skin made

contact with less than maximum force and would have been more prone to glance off without collecting a sample, regardless of needle length.

The remote biopsy dart system used in this study offered several advantages. A built-in CO₂ pressure gauge oriented on the dart rifle facing the shooter, along with a pressure control valve, allowed the pressure setting to be monitored and fine-tuned while aiming. In addition, a red laser sight facilitated quick accurate aiming. The bright pink color of the dart tails made the darts relatively easy to find, and the hollow rear chamber of the dart caused the darts to float tail-end-up when they fell into water (on the very few occasions when darts fell into Brooks River and were recovered, skin tissue samples were still retained within the needle).

Because some bears reacted strongly to the darts by retreating quickly into cover, it is advisable to provide for clear escape routes for target animals when using the darts. Most of the bears sampled during this study were known to researchers from ongoing observational studies, and many of the animals were observed by researchers repeatedly following biopsy dart sampling. There were no apparent adverse effects of the biopsy darts on bears that were sampled aside from reactions to being struck by the darts (the typical reaction was for a bear to trot away several steps). Often bears sampled via biopsy darts were seen near the observation platforms during subsequent sampling sessions, and no behavioral changes were noted (in some cases, bears observed had been previously darted on >1 occasion). Still, bears usually showed some degree of reaction to the darts, and so it is possible that in particular if a bear were repeatedly dart-sampled it could perhaps learn to associate any discomfort from the darts with other aspects of the darting events (e.g., close proximity to a viewing platform, certain human movements, etc.).

Remote biopsy dart sampling proved effective for obtaining skin tissue samples from many individual bears at Brooks River. However, some

bears that were especially wary of people or that were rarely seen near the public viewing platforms were difficult to sample via biopsy darts. Therefore, monitoring of installed hair snares (Beier et al. 2005) using remote video camera systems (VS-04; Penn's Woods Products, Inc., Export, PA), was also used to maximize the likelihood of obtaining DNA from these animals.

ACKNOWLEDGMENTS

I thank H. Maltby, J. Fox, E. Groth, K. Mocnik, S. Graziano, T. Chu, S. Farley, R. Squibb and J. Wendland for field and technical assistance. Three anonymous reviewers provided useful comments on drafts of this paper. This project forms part of a cooperative study being conducted with the U.S. Geological Survey, Alaska Science Center. Field work was conducted under permits provided by the Alaska Department of Fish and Game (05-076, 06-074, and 07-060) and the National Park Service (KATM-2005-SCI-0005, KATM-2006-SCI-0007, and KATM-2007-SCI-0001).

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