

Grizzly Bear Survival in Oil and Gas Operating Areas

Prepared for Petroleum Technology Alliance Canada

> **Final Report** fRI Research Grizzly Bear Program

> > May 29, 2017

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ACKNOWLEDGEMENTS

The collection of scat samples was undertaken by our research team in 2016 and thanks go to Terry Larsen, Cam McClelland, Brent Rutley, Flurina Deagle, Lindsey Proctor, Leonie Brown, Sarah Milligan, Terry Winkler, John Saunders, and Bernie Goski.

We also want to thanks members of our GIS Program (Julie Duval and Daniel Wismer) and Communication Services (Ben Williamson, Terri McHugh, and Fran Hannington) who aided in this project.

REPORT SUMMARY

This project was undertaken to determine if new research techniques and methodologies could be operationalized into grizzly bear monitoring activities at both BMA and watershed scales. The focus of the project was to evaluate scat collection DNA methodologies to investigate population assessment including occupany and survival.

Results from scat collection efforts by researchers showed that it is certainly possible to gather grizzly bear scats in identified watersheds by driving current access features, all weather gravel roads. In addition our research teams, were able to gather other grizzly bear scat samples in this BMA while engaged in other data collection efforts. When viewed together this collection technique showed the value and potential of engaging others who are working in and using grizzly bear habitat to assist with scat collection to support monitoring efforts. We also showed that even with a relatively short collection period (2 months) important data could be collected from scat sampling along motorized access features. DNA extraction rates from scat samples were lower in 2016 than in previous years and we are working with the Norwegian genetic laboratory to understand the reasons for this. Grizzly bear hair remains, in our opinion, the best biological sample for genetic analysis that can be gathered in a non-invasive manner.

Our results did show that scat DNA methodologies allow for the monitoring of survival of individual bears within a BMA and we were also able to understand, from historic GPS location data, that the identified bears were still using a landscape area that is consistent with previously know home ranges. These data sets, when collected on a regular and ongoing basis, will allow managers to speak to questions of grizzly bear displacement and natural resource extraction activities as well as other human uses of the landscape. Similarly these data sets from scat DNA provided evidence of grizzly bear occupancy in watersheds that in 2014 had few or no bears being identify within the watershed. It is recognized that DNA inventory does not locate every bear on the landscape or within a watershed and should managers have specific concerns about either human caused mortality rates or occupancy in key watersheds this DNA scat technique can help to provide greater insights into these questions at a relatively low cost. Our results showed the presence of bears in watersheds were no or few bears were previously identified and that except for one animal these bears were known to reseachers from previous DNA inventory work or from research efforts associated with the capture and handling of bears in this BMA.



Although more work is still required from the laboratory perspective our partnership with the NIBIO laboratory is continuing to put more effort into improving extraction rates and field protocols to maximize genetic data from scat samples. This work is continuing in 2017 as part of the fRI Grizzly Bear Program. As a large scale population inventory tool we believe that current established techinques using scent lures and barb wire are the best approach, however scat sampling can certainly augment and support other monitoring efforts.



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1. INTRODUCTION AND BACKGROUND

Understanding how landscapes are occupied and used by grizzly bears as populations expand is a crucial knowledge gap for provincial grizzly bear recovery efforts. Additionally, having techniques in place to monitor and document occupancy and survival will be important in order to determine when recovery actions have been successful. As part of this effort, wildlife managers need to be able to understand how the management practice of grizzly bear relocations may influence occupancy and population growth. Our ability to identify the spatial distribution of bears on the landscape and track their survival over time would be an important step forward in provincial grizzly bear recovery efforts and would provide important data that will be needed when the next species status review takes place. Combining distribution, survival, and age class through non-invasive genetic sampling represents important new advancements in grizzly bear conservation, recovery and management in Alberta.

This research project will build upon previous research efforts by the FRI Research grizzly bear program to combine advances in DNA scat sampling with recent results from population inventory work conducted in Bear Management Unit 3 in 2014 (Stenhouse et al. 2015). In conjunction with the development of these new assessment and monitoring approaches, we will also gather biological samples to understand the reproductive status of female grizzly bears found within the study area.

2. PROJECT GOALS AND OBJECTIVES

The primary goal of this project is focused on applying research techniques and methodologies (genetic analysis) to determine if these can be operationalized into grizzly bear monitoring activities at both BMA and watershed scales.

Specifically this research and analysis will allow the Department to scientifically evaluate new techniques and methodologies to identify occupied grizzly bear watersheds through the use of DNA scat collection, using population inventory data to identify watersheds to monitor. The research team will also use recently assembled genetic data from relocated grizzly bears moved into the study area over the past 15 years to evaluate what impact this management action may have on grizzly bear population recovery.

Research Objectives:

1. What is the occupancy of grizzly bears in selected watersheds where occupancy was low or zero, as determined by the 2004 population inventory during September – October 2016?

2. How have the bears relocated into BMA 3 between 2004 and 2014 affected the population of bears in this management unit? This will be evaluated from both a survival and a genetic perspective.

3. What is the reproductive status (as determined by reproductive hormone levels and profiles) of female grizzly bears in the selected watersheds?

4. Can the collection of hair samples collected from grizzly bear dens be used to verify fall reproductive status in research animals?



3. METHODS

3.1 Road Transects and Scat Sampling

Grizzly bear scat sampling took place in two sampling periods and centered on driving roads in seven grizzly bear watershed units (GBWUs). During Session 1 (September 28 - October 7th, 2016) one, or at times two, staff members focused sampling efforts on secondary and tertiary gravel roads within GBWUs Y57, Y63, Y65, Y66, Y73, Y79, and Y87. These watershed units were selected for scat sampling by examining differences between the estimated carrying capacity calculated by Nielsen et al (2015 –unpublished data) , and the low number of bears detected in the 2014 BMA 3 hair snag population inventory (Stenhouse et al. 2015(Figure 1, Table 1)). In Session 2, a maximum of five staff members prioritized re-searching the same road routes driven in Session 1, where road conditions permitted. Additionally, during the second sampling period, further search effort was distributed to GBWU Y56, which was found to have relatively high grizzly bear densities in the previous 2014 population inventory (Figure 1, Table 1).

In addition to collecting scat samples, it was important that spatiotemporal measures of search effort were recorded in order to develop and evaluate the efficiency and cost-effectiveness of non-invasive scat based DNA methods and future sampling designs. Therefore, both sample information (date, time, UTM, any relevant notes) and routes searched were documented using the fRI Research GrizzlyScat App. The app is freely available to iOS (Apple) and Android (primarily Samsung devices) smartphone users. For this project, fRI staff simply downloaded the app, registered their information, and initiated the "Log My Route" function when ready to proceed on a sampling search route. This tracking function runs in the background, collecting time and location information (a cellular connection is not required). If scats were encountered, approximately 1cm³ of fecal material was sampled from the center (inside) of each scat and placed into 25mL sampling vials, each containing 10 to 15g of silica desiccant. Scat found on the road was removed from the road surface after sampling, so as to not be re-sampled during the following search session. Each vial was labelled with a unique barcode (Figure 2), which could be scanned from within the app, linking the sample vial to a data form where staff could log any relevant notes and take a photo of the site. Sampling time and location data were captured at the time of bar code scanning. When forays were completed and the app session was terminated, the route and/or scat data was automatically transmitted securely to the fRI Research database when a cellular network connection is in place. Cell service is not required for the app to function; data is simply stored until such a time that cellular service is regained.

3.2 Laboratory Analysis

Scat samples collected between September 28th and October 19th 2016 were sent to the NIBIO (Norwegian Institute of Bioeconomy Research) laboratory for analysis. We have been working with this laboratory since 2012 to validate procedures, standardize field protocols, and to ensure that black bears (a species not found in Europe) could be reliably identified within submitted samples from Alberta. DNA was extracted from the samples, and a species-specific test between grizzly bear and black bear using mitochondrial DNA was performed on all samples. Genotypes, based on twelve markers (G10B, GIOH, G10J, G10L, G10M, G1A, G1D, MU50, G10P, Mu23, Mu51 and Mu59) and one sex-specific marker, was determined for samples testing positive for grizzly bear DNA in the initial test.



The collection of scat to determine the genotype of a grizzly bear has been an ongoing endeavor since 2010. In 2010, we collaborated with the Taberlet lab in France and Wildlife Genetics International (WGI) in British Columbia. Based on knowledge at the time, we used several different methods to collect scat in the field. These methods included dragging a toothpick or a cotton swab across the surface of the scat and storing in paper envelops, or putting 1cm³ of scat in ethanol and later draining the ethanol and drying with silica. We collected replicates of a scat sample and sent a replicated to each lab. Results from both labs were poor. We repeated scat collections in 2011 and sent 10 samples only to WGI and again the results were poor. Meanwhile, the NIBO (at the time, Bioforsk) lab in Norway was achieving 80% success rates with extracting DNA from grizzly bear scats in Scandinavia. In collaboration with NIBIO, we aimed to determine why scat samples from Alberta grizzly bears were experiencing poor extraction rates. NIBIO send 10 swab samples dragged across previously frozen bear scats collected in Sweden to WGI, who then extracted and ran the DNA with much success (9 of 10 samples were successful). The successful extraction of Swedish bear scat samples suggested that unknown properties of scat specific to Alberta grizzly bears were resulting in a low success rate.

In the fall of 2012, we began sending desiccated scat samples in silica to NIBIO. Staff the NIBIO came to our study area in the fall of 2013 and collected fresh scat samples to return to the NIBIO lab. The success rate for the fresh samples was high (9 of 10) while only about half of samples collected throughout the summer 2013 worked (11 out of 20 worked). Since 2013, we have been sending scat samples to the Norway lab and each year has had varying degrees of success. We considered this project to be ongoing with the hopes that we learn new things each year to improve the success rate of genotyping grizzly bears using scat as the DNA source.

4 RESULTS

4.1 Road Transects

During the three weeks of sampling, over 2,100 km of roads were driven within BMA3, collecting scat samples across two sampling sessions in eight GBWUs (Table 2, Figure 3).

4.2 Scat Sampling

In total, 35 scat samples were collected during the three weeks of driving road sampling routes. An additional 32 samples were collected opportunistically outside of the sampling routes during additional fieldwork (GPS cluster visits, capture and collaring efforts) taking part in 2016 within BMA3, resulting in a total of 67 samples sent for DNA analysis at the NIBIO laboratory in Norway in November 2016.

4.3 Genetic Results

Of the 67 scat samples collected within BMA 3 during the 2016 field season, 60 samples (90%) were positive for bear DNA in the initial species test. Of these bear samples, 13 (22%) were found to be black bear, and 41 (68%) were identified as grizzly bear, while bear species could not be distinguished in the remaining 6 samples (Figure 4).

From the 41 positive grizzly bear samples, six samples were successfully genotyped to at least seven markers. The definition of an approved marker is two congruent runs for heterozygotes and three for homozygotes. From these six



samples, five individual bears (two male, three female) were identified. Four of these individuals were known to researchers (11786Bu [F], G007 [F], G152 [M], and G168 [F]; Figure 4) while one (Cuar111 [M]) was identified by a scat sample with no genetic match to any bears previously identified, thus this represents a new animal for our genetic database.

4.4 Survival of Previously Known Bears

Scat samples collected in 2016 revealed the presence of four bears previously known to researchers in BMA3 (Figure 5):

G007- Female, born in 1996. First captured and collared in May 1999. Last collar data locations were from the summer of 2003. Prior to this scat sample, she was last detected in the 2011 hair snag survey.

G152- Male, born in 2009. Captured and collared in July 2013, with the last collar data in June 2014. This bear was additionally detected in 2011, 2013, and 2014 hair snag surveys.

G168- Female. Estimated to be born in 2014, although awaiting lab results on tooth extracted at capture. Captured and collared in May 2016, with the collar currently transmitting data. This bear was also detected in 2014 scat sampling.

11786Bu- Female. Unknown age, as bear is only known from two genetic samples from the 2004 hair snag survey.

4.5 Familial Relatedness

Genetic results from three of the four known bears detected in this scat survey revealed familial relatedness to other bears in our research database. Four bears (Table 3) captured and collared over time by our research program have been found to be the offspring of G007, including G168, also detected in scat as part of this study (Figure 6). Two bears collared in 2012 and 2013, and one male detected in the 2014 hair snag census of BMA3 are the offspring of 11786Bu, who prior to this scat survey, was only ever detected in hair snag studies (Figure 7).

4.6 Hair Sampling at Dens

One component of this project involved the collection of grizzly bear hair at or near den sites to provide samples for reproductive hormone analysis (Cattet et al. in review). The new research results from our work on using reproductive hormone levels to determine pregnancy in female bears and to also use the reproductive hormone profile from hair sampling to identify age class (subadult vs. adult) requires approximately 135 gms. of hair. Our field crews will be visiting grizzly bear den sites in the spring of 2017 (2016 den sites) to determine the amount of hair that can be collected at den sites for this purpose. We currently believe that we will need to conduct directed sampling in selected watershed units in the fall of 2017 using a scent lure and barb wire (with cameras to record individuals) to gather the needed samples for this analysis in 2017. This component of this project will extend into 2017.

5. DISCUSSION

This project aimed to determine if there are alternatives to grizzly bear population trend monitoring, occupancy and survival that could be conducted at a smaller scale and at a reduced cost when compared to large scale population inventory work. Current approaches to understand population trend require the resampling of an entire Bear Management Area (BMA) using new spatially explicit mark-recapture sampling strategies for collecting hair samples



for genetic analysis. These approaches, while recognized goal standard approaches, do not provide any insights as to when the next (repeat) inventory should occur to track population trend. Often managers schedule these repeat inventories based on budget availability and guesswork, not population indicators. This project was initiated to understand if new and innovative techniques, applied in a BMA where an expanding population had been identified, could be used to provide insights into population demographic parameters (occupancy and survival) to guide monitoring efforts and to aid in understanding grizzly bear recovery in provincial recovery zones.

We focused our research efforts on what our previous work on this topic suggested were key elements in applying this new (for Alberta) technique of grizzly bear scat sampling. We wanted to understand if it would be possible to collect bear scat samples along existing road networks without the need for special search effort (e.g. across habitat types). Indeed, driving sampling routes in GBWUs previously known to have low grizzly bear densities still returned scat samples, especially when paired with samples collected opportunistically throughout the field season.

Currently, poor genetic extraction rates of scat to the individual level limits scat-based genetic sampling for the purpose of wide scale grizzly bear population inventories; however, our scat sampling efforts still yielded valuable grizzly bear occupancy data. When paired with our extensive long-term dataset, even the low number of samples genotypes to the level of individual allowed for a limited survival analysis. Considering the cost-effectiveness of scat-based inventory technique relying on searching only roads, we believe this approach holds promise for a long term monitoring tool to be used in conjunction with repeated large-scale BMA hair DNA censuses.

