# **Final Report Form Cover Sheet**

Funder receiving report: Woodcock Foundation

Name of organization completing report: Western Transportation Institute, Montana State University

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Have there been any changes to your organization's IRS 501 (c)(3) not-for-profit status since your request for this grant? No

Project name or brief project description: "Pilot study: DNA profiling to identify individuals using wildlife crossings"

Check one: General operating \_\_\_\_ Project support \_\_X\_\_\_

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Check one: This is a Final Report

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### I. NARRATIVE

#### Introduction

Roads obstruct animal movements, fragment critical habitats, and reduce landscape connectivity. This can result in higher mortality, lower reproduction, and ultimately smaller populations and lower population viability. Up until now, most research assessing mitigation crossing effectiveness in Banff and elsewhere has been focused at the level of individuals and not populations. Healthy functioning ecosystems require viable wildlife populations. Thus, it is critical to know the performance of wildlife crossings at the population level. This will enable us to more accurately assess the demographic consequences of roads and the utility of wildlife crossings in enhancing population viability.

Crossings that serve as habitat or landscape connectors should allow for the following: (1) movement within populations and genetic interchange; (2) biological requirements of finding food, cover and mates; (3) dispersal from maternal ranges and recolonization after long absences; (4) redistribution of populations in response to environmental changes and natural disasters; and (5) long term maintenance of metapopulations, community stability, and ecosystem processes. These five biological functions represent a range of connectivity from basic (1,2) to functional (3,4,5). Individual level needs can be met and measured by regular monitoring of animal use of the crossings. However, verifying that population-level needs are met will require information on breeding movements, dispersal and type of genetic mixing occurring (close or distantly related individuals).

Obtaining this information from fragmentation-sensitive species that typically occur in relatively low densities and have low reproductive rates is problematic. Thus, demonstrating crossings provide for population level benefits (adult male and female movement across roads; dispersal, survival and reproduction of young) can require 15-20 years of intensive monitoring of radio-marked grizzly bears alone. Molecular techniques now make it possible to identify individual animals, their sex, and genetic relatedness. Therefore, DNA/hair sampling can be a powerful, non-invasive technique that could provide critical information regarding genetic interchange facilitated by crossings in a relatively short period of time, without ever having to capture or see the animal.

This <u>pilot study</u> funded by the Woodcock Foundation in 2004, is testing and developing a simple, non-invasive, cost-effective method to identify animals using wildlife crossing structures. Target species are mammals, *coyote-sized and larger* with emphasis on *large carnivores*, particularly *grizzly and black bears*. This innovative approach, if used with DNA-based population sampling, has the potential to mark a major advancement in methods to scientifically understand levels of connectivity occurring at wildlife crossings and assessing their conservation benefits.

#### Methods & Results

#### A. Project Development and Operational

*Parks Canada Environmental Screening Report.* - Prior to beginning the pilot project Parks Canada required that we prepared an "environmental screening report", which basically describes in detail the objectives of the study, how we will carry it out, and what potential environmental impacts we might foresee. The report was completed in mid-May 2004 and was open for public comment for a two-week period. There were no public concerns raised during the comment period, therefore we were given permission by Parks Canada to begin setting up the hair-sampling system at two wildlife underpasses selected for testing the technique.

*Wildlife Underpass Selection*. - We placed the hair-sampling system at two wildlife underpasses (Healy, Duthil). These underpasses were selected because they historically have had a relatively high level of carnivore use and low human use<sup>1</sup>. We also placed a third hair-sampling system at the Wolverine Overpass, however, without a videocamera monitoring system. Thus, hair capture success rates of the system could only be measured at the two underpasses with videocameras.

*Hair-sampling System.* - The initial prototype for DNA/hair-sampling consisted of stringing one strand of barbed-wire between steel posts on the level section of an underpass (see "System *Flexibility for Outsmarting*"). Page-wire fencing was installed behind the steel posts on either side of the crossing structure to ensure that animals contact the barbed-wire as they pass through. Barbed-wire was strung at a height of 50 cm above ground to ensure contact with different sized animals. Hair that was snared on the barbs as the animal passed through the underpass (or overpass) was collected daily and used for DNA analysis. No bait or scent lures were used at the onset of the study (see "Attracting Cats").

*Videocamera System.* - Two videocameras are used to record the 24-hr behaviour of animals as they approached the hair-sampling system at the underpasses from either direction. The cameras were equipped with an infrared light source to record animals that approach the underpass at night. Videocameras were operated with C-cell batteries. Use of videocameras enabled us to evaluate the efficacy of the technique by determining the DNA/hair capture success rate. This is measured by determining the number of times hair-samples are obtained compared to the total number of underpass visits by that species.

*Gradual Approach for Animal Habituation.* - The hair-sampling system was installed in stages to allow time for wildlife to adapt to placement of videocameras, lights, and barbed-wire across the underpass. Videocameras were in place during the entire procedure to allow continuous monitoring of all stages of the installation process. The steel support posts were first put in place. Next, we installed the page wire barriers behind the support posts. Videocamera boxes and posts to secure lights were installed. The final step involved stringing barbed-wire across the central portion of the underpass. The staged installation process occurred over a period of roughly two weeks.

*Hair-Sampling System Monitoring.* – We checked videocameras and barbed-wire every day to ensure proper functioning and to collect hair samples. No more than two people attend the wildlife underpass to check track pads, barbed-wire, and videotapes once the system is functioning.

#### **B. Monitoring Results**

*Monitoring Periods and Wildlife Underpasses.* – We began monitoring our prototype hairsampling system at the Healy Underpass on 22 June 2004. Once the Healy system was in place, we began plans to install an identical system at the Duthil Underpass. The hair-sampling system at Duthil Underpass was activated on 10 July 2004. Monitoring of the Wolverine Overpass began without videocameras on 28 June 2004. We continued to collect hair samples and monitor animal responses at the three crossings until 7 October 2004.

*System Flexibility for Outsmarting.* – Devising an effective DNA/hair-sampling technique involves time for "research and development". Several designs were tested and adjustments made. In addition experimenting with different hair-sampling designs, testing also requires sufficient time because wildlife need to adapt to the system within the underpasses being tested. We describe below the incremental changes in DNA/hair-sampling design we have used to date. These only were implemented at the two wildlife underpasses with video monitoring.

(1) <u>One strand of barbed-wire</u>. We initially began hair-sampling with a system consisting of one strand of barbed-wire stretched across the Healy Underpass at a height of 50 cm above the ground. This system of hair-sampling (single barbed-wire 50 cm above ground) has been used extensively by wildlife biologist in North American for DNA-based population surveys<sup>2,3,4,5</sup>. Monitoring of animal movement, behaviour, and hair-sampling success soon led us to devise a more efficient system, as some animals were able to pass over or under the barbed-wire without much problem. One large grizzly bear did not hesitate to crawl under the barbed-wire, quite adeptly, as it moved through the Healy Underpass, surprisingly leaving no hair behind on the barbs.

(2) Two strands. – From videomonitoring, it was evident we consistently missed some important hair-sampling opportunities because animals were able to go under the 50 cm strand. Therefore, we added a second strand at 35 cm height above the ground. This seemed to help sample more hair of passing animals, but still some bears were able to clearly jump over the top 50 cm strand.

(3) <u>Sticky string</u>. – After some doubt about the effectiveness of barbed-wire as a hair-sampling device, we investigated other methods currently in use and how they might be applied to our unique situation and goal. Other proven methods of hair-sampling include adhesive tape and glue boards. Glues used for preparing boards to trap rodents can be obtained in bulk. These are unique adhesive formulations scientifically designed to capture rodents over a wide range of temperatures. We inquired about obtaining bulk glue to brush onto string stretched across the underpass, in place of the barbed-wire. We contacted Laurent Sirois at Atlantic Paste and Glue Co. Inc. in Quebec, and explained what we needed. Laurent had a product, consisting of 1/8"-wide plastic webbing or string covered in the trapping glue. He offered to let us test the "sticky string" to see if it would work for sampling animal hairs in Banff. On 13 July 2004, we strung sticky string at the Duthil Underpass and wrapped existing barbed-wire with sticky string at Healy Underpass. Eventually we removed the barbed-wire strand at Healy Underpass and used only sticky string as a means of capturing hairs at both underpasses.

(4) <u>Two strands/Wide opening</u>. - The last hair-sampling setup consisted of two strands of sticky string at 35 cm and 75 cm above the ground at both underpasses. This was devised after observing bears jumping over and moving under the strands we had placed earlier. The primary focus of the study is to sample hairs from bears, and we believed that by placing one strand high (too high to jump over) and one low (too low to go under), but with about 40-50 cm between, it would provide a large opening but would still prove difficult for bears to pass through without eventually rubbing against either of the strands of sticky string. The next day after installing this two-strand/wide opening setup, a black bear ran through the Duthil Underpass, jumped between the strands and left hairs on the top strand.

Our experience with the use of sticky string and barbed-wire has demonstrated that the former is more effective at sampling hairs of animals.

Attracting Cats. – Since we began monitoring, cougars used the three wildlife crossings a total of 17 times (Table 1). Cougars easily jump over the strands, however, lynx detection surveys carried out by the U.S. Forest Service have been effective in attracting and obtaining hair samples by using a rubpad (patch of carpet) and cat lure<sup>6</sup>. At the three crossings we installed the same lynx rubpads and cat lure for sampling hairs of passing cougars and lynx. Monitoring by videocameras and trackpads showed that no cats or other animals were attracted to the rubpads.

*Underpass Monitoring*. – We have amassed so far a total of 461 video recordings of animals attempting to use or using the two underpasses during the entire study period (June-Oct). These recordings are from both cameras, therefore are duplicative and greater than the actual number of wildlife using the underpasses during that time. Nine wildlife species coyote-size and larger were detected a total of 742 times using the three wildlife crossings set up for hair-sampling since 22 June 2004 (*Appendix, Table 1*). This is for the total period, thus covering the incremental changes in DNA/hair-sampling design (see *System Flexibility for Outsmarting* above).

**DNA/hair-sampling and Success Rate.** During entire study period (including the early period of "research & development" of hair-sampling design), we have collected a total of 167 DNA samples from the three wildlife crossings (*Appendix, Table 2*). Fifty-seven known samples have been collected from elk and 24 known samples from deer. DNA/hair samples were obtained from black bears 9 times; grizzly bears 4 times, moose 8 times, and "suspected cougar" 2 times. Samples were collected from "unknown" species 29 times and "unknown ungulates" 26 times.

At this writing, we believe the "two-strand/wide-opening" DNA/hair-sampling system is most effective for grizzly and black bears, our focal species. Thus, we measured a preliminary hair-capture success rate once the system was in place (10 Aug-7 Oct). Hair-capture success was a respectable 54% for black bears (*Appendix, Table 3*). One grizzly bear used one underpass after August and avoided the strings. Grizzly bears move out of the valley up to higher elevations after early July, thus their low number of visits to the two wildlife underpasses (see *Ongoing and Future* below). Hair-capture success was high for elk (62%) and moose (100%, 2 samples from 2 visits). Low samples and hair-capture success rates were obtained from cougar and deer, as there were few visits after August by cougars, and deer easily jumped over the sticky strings when passing through.

The hair-capture success rate was 54% for black bears. However, two of the 13 black bear passes when hair was not captured occurred when a cub was able to avoid the strings; hair was obtained from the mother each time. The system is likely to be less effective in capturing hairs from cubs; being smaller they can manage to avoid the strings. However, when using the hair sampling system with routine track-pad monitoring, DNA/hair may not always be collected from cubs, but equally important genetic information (mother, offspring presence) will usually be obtained through observation of tracks on the track pads.

#### C. Ongoing and future

*Data collection*. –We terminated collection of DNA/hair samples at Healy and Duthil Underpasses on 7 October 2004. This concluded the field data collection segment of this pilot project for 2004.

*DNA analysis*. – All DNA samples collected from the project between June and October were sent to Dr David Paetkau at Wildlife Genetics International, Nelson, British Columbia, for molecular analysis. The results from the analysis will become available in January 2005. These results will be used to refine preliminary estimates of hair-capture success rates after the first year (*Appendix, Table 3*).

**2005** *Woodcock proposal* – We submitted a 2005 grant proposal to the Woodcock Foundation for continued support of long-term monitoring and DNA-based approaches for restoring landscape connectivity across transportation corridors. The proposed project will continue research, monitoring and transfer of science-based information that will result in a range of applications useful to transportation planning, practice and policy in areas where road networks and landscape conservation concerns collide. There are four main strategies, but the most critical being to conclude 2004 study of the DNA-based hair sampling technique by measuring a hair-capture success rate. The investigation requires an additional year because more time is needed to fully test the technique. Several designs have been tested and adjustments made (see *System Flexibility for Outsmarting* above). Testing requires more time because it must encompass a period of adaptation of wildlife to the system within the underpasses tested. Beginning testing earlier in the year (May) will allow the technique to be tested for grizzly bears as most of their underpass use occurs from May to early July.

#### **D.** Acknowledgements

We thank the Woodcock Foundation for providing key financial support to undertake this study and enable future funds to be leveraged with other foundations and Parks Canada. We sincerely thank Jeremy Guth for his work in generating support from other foundations and within Parks Canada. This work could not have been possible without supporting grants from the Wilburforce Foundation and the U.S. Humane Society.

This project has benefited from the generous assistance of several individuals in preparing the hair-sampling and video monitoring systems – M Gibeau, J Woods, T Hurd & T Davidson (Parks Canada), G Mowat & D Paetkau (Wildlife Genetics International), L Ruggiero & J Malloy (US Forest Service), L Sirois (Atlantic Paste & Glue), and N Dodd (Arizona Game & Fish).

#### E. Project partners

Woodcock Foundation, New York, New York
Wilburforce Foundation, Seattle, Washington
Friends of Banff, Banff, Alberta
Humane Society of the United States, Washington, DC
Parks Canada, Banff National Park, Banff, Alberta
Parks Canada, Ecological Integrity Branch, Ottawa, Ontario

### F. References

- <sup>1</sup> Clevenger, A.P., B. Chruszcz, K. Gunson, and J. Wierzchowski. 2002. Roads and wildlife in the Canadian Rocky Mountain Parks Movements, mortality and mitigation. Final report to Parks Canada. Banff, Alberta, Canada.
- <sup>2</sup> Boulanger, J. et al. 2002. A meta-analysis of grizzly bear DNA mark-recapture projects in British Columbia. Ursus 13.
- <sup>3</sup> Mowat, G. and Strobeck, C. 2000. Estimating population size of grizzly bears using hair capture, DNA profiling, and mark-recapture analysis. Journal of Wildlife Management 64, 183-193.
- <sup>4</sup> Poole, K. et al. 2001. DNA-based population estimate for grizzly bears in northern British Columbia. Wildlife Biology 7, 105-115.
- <sup>5</sup> Woods, J. et al. 1999. Genetic tagging of free-ranging black and brown bears. Wildlife Society Bulletin 27, 616-627.
- <sup>6</sup> McDaniel, G.W. et al. 2000. Efficacy of lures and hair snares to detect lynx. Wildlife Society Bulletin 28, 119-123.

# **II. FINANCIAL**

## Financial sheet

Grantee:

Dr Anthony P Clevenger

Western Transportation Institute, Montana State University

Title:

"Pilot study: DNA profiling to identify individuals using wildlife crossings"

Disbursement of funds (up until Sep 2004):

Description	Proposed	Project-to-date	Balance
Salary	4,800.00	500.00	4,300.00
Subcontracts*	24,450.00	12,990.00	11,460.00
Supplies	2,500.00	0.00	2,500.00
Travel	1,500.00	0.00	1,500.00
Administrative	1,750.00	600.00	1,150.00
	35,000.00	14,090.00	20,910.00

\*Includes Banff research assistants (Bryan Chruszcz, Kari Gunson), laboratory analyses and consulting (Dr D Paetkau, Wildlife Genetics International, Nelson, British Columbia).

### Planned disbursement of remaining funds to end of contract (Dec 2004):

Description	Balance	Planned Disbursement
Salary	4,300.00	8,300.00
Subcontracts*	11,460.00	11,460.00
Supplies	2,500.00	0.00
Travel	1,500.00	0.00
Administrative	1,150.00	1,150.00
	20,910.00	20,910.00

\*Includes Banff research assistants (Bryan Chruszcz, Kari Gunson), laboratory analyses and consulting (Dr D Paetkau, Wildlife Genetics International, Nelson, British Columbia).

# APPENDIX

		CROSSED		τοται	DID N	OT CROSS		τοτλ	MAY HAVE CROS		SED TOTAL		GRAND
SPECIES	DUTHIL U	JP HEALY UP	WOP	TOTAL	DUTHIL UP	HEALY UF	P WOP			HEALY UP	WOP	TOTAL	TOTAL
Black Bear	17	2	0	19	0	6	0	6	0	0	0	0	25
Cougar	1	2	12	15	0	2	0	2	0	0	0	0	17
Coyote	2	2	0	4	0	4	0	4	0	1	1	2	10
Deer	117	50	190	357	54	38	4	96	5	2	2	9	462
Elk	70	63	5	138	47	9	0	56	0	0	0	0	194
Grizzly													
Bear	0	5	1	6	2	0	0	2	0	0	0	0	8
Moose	0	4	1	5	0	0	0	0	0	0	0	0	5
Bighorn													
Sheep	0	0	0	0	0	1	0	1	0	0	0	0	1
Wolf	0	0	6	6	0	14	0	14	0	0	0	0	20
TOTAL	207	128	215	550	103	74	4	181	5	4	3	12	742

**Table 1.** Summary of wildlife detections at the three crossing structures used in the DNA profiling study, June – October 2004. This data includes the most accurate information from the track pads and video monitoring equipment.

	DUTHIL UP	HEALY UP	WOLVERINE OP	TOTAL
Bird Feathers	0	4	1	5
Black Bear	8	1	0	9
<b>Suspected Cougar</b>	0	0	2	2
<b>Suspected Coyote</b>	0	1	1	2
Deer	9	9	6	24
Suspected Deer	0	0	1	1
Elk	47	10	0	57
Suspected Elk	0	0	0	0
Grizzly Bear	0	4	0	4
Moose	0	8	0	8
Ungulate	20	3	3	26
Unknown	3	3	23	29
TOTAL	87	44	37	167

**Table 2.** Summary of the number of video recordings of wildlife at the three wildlife crossing structures monitored with video cameras, June – October 2004.

**Table 3.** Summary of the DNA/hair capture success rate by species, obtained from Healy and Duthil underpasses. Hair capture success rate is measured from the time the "two-strand/wide-opening" DNA/hair-sampling system was in place (10 August to 7 October 2004).

Species	Animal passages	DNA/hair sample collected	DNA/hair capture success rate
Black bear	13	7	0.54
Grizzly bear	1	0	0.00
Cougar	1	0	0.00
Coyote	2	1	0.50
Elk	66	41	0.62
Deer	78	4	0.50
Moose	2	2	1.00