Wyoming Animal Damage Management Board

2005 Project Summaries

Compiled for the ADMB by Hank Uhden

Projects funded from Rabies budget:

1. Project Title: Johnson County Rabies Mitigation Project. Funding: $40,365.00

Synopsis: Preemptive rabies management in outlying targeted areas of Buffalo, Kaycee, and rural Johnson County. Submitted by: Johnson County Predatory Animal District.

2. Project Title: Rabies Education Project. Funding: $10,943.40

Synopsis: Cost assistance with the Wyoming Department of Health for a rabies education project targeted towards health care providers, veterinarians, and the general public. Submitted by: Wyoming Dept. of Health.

3. Project Title: Campbell County Rabies Nuisance Project. Funding: $13,006.50

Synopsis: Funding assistance for a six month contract person to address rabies and nuisance animal problems in Campbell County. Submitted by: Campbell County Predatory Animal District

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FY06 ADMB Projects:

1. Project Title: Wolverine Presence in Wyoming. Funding: $500.00 (left over funding from previous allocation.

Synopsis: Using cameras at baited sites, attempt to photograph wolverine in order to establish their presence at various locations.

2. Project Title: Black Bear/Grizzly Bear/Mountain Lion Livestock Depredation Prevention. Funding: Up To $12,500.00

Synopsis: Funding cost share on a 25:75 match up to a maximum of $12,500 with the Wyoming Game & Fish Commission and Wildlife Services to alleviate black bear, grizzly bear, mountain lion depredation to livestock, bees and beehives in all counties of the state. Submitted by: Wyoming Game & Fish.

3. Project Title: Extended release of Mifepristone for field delivery to coyotes. Funding: $15,000.00

Synopsis: This ongoing research will investigate the treatment efficacy and bio-availability of the compound to terminate the pregnancy in coyotes. Submitted by: University of Wyoming.

4. Project Title: Whiskey Mountain Big Horn Sheep Predation Project. Funding: 11,000.00

Synopsis: Management of coyotes on Whiskey Mountain to enhance Big Horn Sheep lamb survival under a timed and targeted program. Submitted by the Foundation for North American Wild Sheep - Wyoming Chapter.

5. Project Title: Devil’s Canyon Supplemental Big Horn Sheep Transplant. Funding: $8,000.00

Synopsis: Under approval from the 2004 WY Game & Fish Commission and secured funding, the Devil’s Canyon big horn sheep population is to be supplemented with transplanted sheep from Oregon. To increase the success of this effort, predator management will be conducted in a timed and target manner around lambing and wintering areas. Submitted by: Wyoming Game & Fish.

6. Project Title: Fremont County sage grouse/mule deer project. Funding: $10,000.00

Synopsis: Project has three components: 1) Identification of predators affecting sage grouse nesting sites through the use of motion cameras; 2) measure and document differences in nesting success between areas of predator management and non-managed areas; 3) To enhance a struggling mule deer population to desired population levels. Submitted by: Fremont County Predatory Animal Board.

7. Project Title: Increasing speed & potency of Theobromine and Caffeine based toxicants. Funding: $12,124

Synopsis: Indentify inexpensive formulation adjuvants which will increase the speed of action, potency and decrease the cost of the threobromine/caffeine predacide which is being collaboratively developed by the ADMB, CA Dept. of Food & Agriculture, and the National Wildlife Research Center (NWRC). Submitted by: NWRC

8. Project Title: Credible Baseline Wildlife Data Collection. Funding: $13,900.00

Synopsis: Real West Natural Resource Consulting. Second year of post monitoring of the wildlife data collection on two on-the-ground management projects (Weston, and Carbon). Submitted by Real-West upon the request of the ADMB.

9. Project Title: Habitat use and population dynamics of Shira’s moose in NW Wyoming. Funding: $23,600.00

Synopsis: Investigate population dynamics and resource selection patterns of the north Jackson moose herd. Submitted by: UW - Wyoming Cooperative Research Unit.

10. Project Title: Badger Creek - Hanging Woman Fawn Predation Project. Funding: $9,164.00

Synopsis: Coyote management to enhance antelope and deer fawn survival. Project include areas of management and control (no predator management) areas to provide comparative results. Management will include timed and targeted predator management in the project area. Project is being conducted in cooperation with the Game & Fish, who will conduct antelope and deer population census and classification. Submitted by: Sheridan County Predatory Animal Board.

11. Project Title: DNA analysis for control of predatory wolves. Funding: $11,312.00

Synopsis: Analysis of DNA isolated from predator saliva, blood, feces and/or hair can be used to identify the species, sex, and/or individual genotype of the predator. Results of this project should allow development of predator sample collection protocols and enhance management strategies. Submitted by: National Wildlife Research Center.

12. Project Title: Preemptive human/bear conflict mitigation. Funding: $3000.00

Synopsis: This project adopts preventative methods for reversing an overall long-term trend of increasing human/bear conflicts in northwest Wyoming, particularly on private lands in areas like the North and South Forks of the Shoshone River, Teton Village, Wilson, and Dubois. With human safety as the highest priority, this project seeks to minimize human/bear conflicts through (1) minimizing and properly managing unnatural bear attractants; (2) employing bear-resistant waste management systems; (3) managing bears/attractive bear habitat where potentials for conflict and risks to human safety are high; and, (4) employing a public outreach program to reduce knowledge gaps about bears and conflicts. Submitted by: Wyoming Game & Fish.

Johnson County Rabies Mitigation Project

Johnson County Predatory Animal Board
211 Upper French Creek Road
Buffalo, WY 82834
November 27, 2005

Wyoming ADMB
2219 Carey Avenue
Cheyenne, WY 82002

Dear Hank,

I am proud to write of the success of our Rabies Mitigation Project.   Since its start in April it has more than met the goals set, by significantly reducing the troublesome animal calls within the towns of Buffalo and Kaycee, according to local law officials. Population management of potentially rabid animals has been obtained by creating a buffer zone of control around these towns. These zones include greenbelts, wetland areas, a cemetery, a golf course, parks and campgrounds as well as riparian areas of Clear Creek and Powder River.

Our project began with the contracting of a part-time control officer to work with our two full time trappers. A training session was conducted for our employees and board by a local veterinarian covering the proper handling, disposal, sampling and shipping methods.

We were fortunate to have three good employees, all striving to make the project a success. They reported to the board each month of the daily activities and catches. We also had great cooperation with other local and state agencies. The county landfill disposed of the animals at no charge and the Wyoming State Veterinary Lab did the testing at no cost.

The program did not go without surprises and disappointments. An unexpected expense to have all employees have pre-exposure rabies shots, surprised us to the tune of $1215.

We were also disappointed to have some trap tampering and theft. Maybe that's to be expected as you do this kind of work in and around populated areas. We were also asked to leave a property because of the daily early morning activity.

All in all, we were very excited about the projects first few months and satisfied that it met the goals we had set. Enclosed are reports of monthly activity and financial projections and expenses.

Thank you for your continued support of the project.

Dave Hall, Secretary

Johnson County Predatory Animal Board

Johnson County Predatory Animal Board
2005 Rabies Mitigation Project
Tabulation of targeted animals taken by month and employee

Totals by Month

	Skunks	Raccoon	Fox	Fox Pups
April	23	6	1	8
May	12	34	1	0
June	11	19	3	14
July	7	9	1	1
August	7	9	1	0
September	14	7	3	0
October	10	2	5	0
Total	84	86	15	23

Totals by Employee

	Skunks	Raccoon	Fox	Fox Pups
Floyd Cummings	17	12	13	14
Cenny Burnell	10	25	1	8
Randy Tissino	57	49	1	1
Total	84	86	15	23

Johnson County Predatory Animal
Board
2005 Rabies Mitigation Project
Projections / Accual Expenses - April to October

	Projected Expense	April	May	June	July	August	September	October	Total Expense
Contract Labor	7500.00	950.00	1300.00	1250.00	1300.00	1350.00	1300.00	400.00	7850.00
% of County Trappers' Salary	10000.00	1378.85	1341.00	1341.00	1341.00	1341.00	1341.00	1341.00	9424.85
Traps, Lure, and Supplies	1500.00	2900.18							2900.18
Testing and Disposal	1250.00	59.30			18.37			35.53	113.20
Training	500.00	114.90							114.90
Admininstation and Record Keeping	750.00	125.00	125.00	125.00	125.00	125.00	125.00	125.00	875.00
Misc. Expense	1000.00	1215.00							1215.00
Total	22500.00	6743.23	2766.00	2716.00	2784.37	2816.00	2766.00	1901.53	22493.13

Rabies Education Project

Project Title: Rabies Education Project. Funding: $10,943.40
Project Update, December 2005
Submitted by Jamie Snow
November 21, 2005

Overview:

We called the veterinarians and health care providers in the state. Calling was completed on September 2005. We contacted over 50% of the Wyoming practicing veterinarians and about 25% of the family practice physicians within the state. Through the information collected from the surveys we determined areas were both veterinarians and health care providers needed additional information/education about rabies. We plan to recall all of the veterinarians and health care providers in the summer of 2006 to evaluate the success of the educational campaign.

Two educational themes were created:

- "See a Vet, Save your Pet" – emphasizing the need for people to get their pets vaccinated against rabies
- "Look, Don't Touch I Might Have Rabies" – emphasizing how people should keep a safe distance from wildlife and strays because these animals may be infected with rabies.

Two types of posters were created with these two themes and were distributed to all practicing veterinarians in the state, all family practice physicians in the state, County Public Health Nursing Offices and grade schools in Wyoming.

Notebooks were created specifically for veterinarians and physicians. These notebooks are to be used as a resource for veterinarians and health care providers to answer many of the questions they have about rabies. It provides information about when to give rabies post exposure prophylaxis and when not to, what actions to take with an animal that bites or potentially exposes someone to rabies. It also provides information on what to do with a domestic animal that is potentially exposed to rabies. It also gives information about who should receive rabies pre exposure prophylaxis and where and how to get it done.

September was "Rabies Prevention and Awareness Month". Three Power Points were created, a power point for veterinarians, one for school age children and one for health care providers. These power points were distributed to all the Wyoming Regional Veterinary Coordinators, All Hazards Response Coordinators and Public Health Nursing Offices around the state. These individuals gave numerous rabies presentations around Wyoming during the month of September.

A rabies educational curriculum was developed and distributed throughout the state to all Wyoming grade schools. Principals were encouraged to utilize the curriculum, during the month of September, to help educate students about rabies and how to prevent it.

Post cards were distributed to both veterinarians and health care providers in the state with various messages to help educate both groups about different aspects of rabies.

Rabies ads were created and ran through out the month of September emphasizing the two above mentioned themes.

Wyoming Department of Health created a web site about rabies. This web site has updated information about rabies animal cases in the state - thanks to information provided by the Wyoming State Veterinary Laboratory. Please take a look at our web site at: http://wdh.state.wy.us/epiid/rabies.asp

Materials:

Book Cover

Budget:

Intern costs: $3,209.20
Educational materials: $2,500 (design, shipping and printing)
Radio ads: $1,500
Mailing costs: $880

Total spent to date = $8089.20

Campbell County Rabies Nuisance Project

Campbell County Rabies Nuisance Project
Final Report

This project has been very successful here in Campbell County. It was a six month project, with the primary goal being rabies intervention. The project also dealt with nuisance calls which inevitably canceled out a number of the rabies situations. There was a great deal of positive feed back from the citizens of Campbell County. Brandon O'Brian, the APHIS agent in charge of taking the calls, did an excellent job of dealing with individual situations. When time permitted, Brandon also did some work in the Crook and Converse counties.

Throughout the six month period, the following rabid or nuisance animals were taken:

Striped Skunk……………..183
Raccoons………………….. 73
Feral cat……………………52
Coyote……………………..18
Badger……………………... 4
Rattlesnake………………..…4
Bull Snake……………..……1
Mink………………………...3
Muskrat……………………...7
Red Fox…………………..…7
Porcupine………………..….5
Northern Flicker…………....1

Brandon O'Brian brought up a good point in a monthly report, that not only does this project contribute to the public's health and safety, but it also benefits the animal populations themselves, keeping them healthy as well.

This project has also been vital to the public as a source of education to the dangers of rabies. Many calls were attended to that would have otherwise been neglected had it not been for the funding of this project. It is hard to calculate or estimate the number of serious injuries that were avoided due to the implementation of this program.

As president of the Campbell County Predator Board, I would like to extend my gratitude to the ADMB for the continued funding of this project. It has been a success for the last two years, and many benefits have been proven.

Dan Reimler

Campbell County Predator Board

Wolverine Presence Project

WOLVERINE PRESENCE PROJECT
1910 BUSTLE CREEK ROAD
ALTA, WYOMING 83414
(307) 353-2407

WOLVERINE PRESENCE PROJECT

STATUS AND RESULTS

November 2004 through April 2005

(ANNUAL REPORT)

Included: Narative

Economics

WOLVERINE PRESENCE PROJECT
1910 BUSTLE CREEK ROAD
ALTA, WY 83414
(307)353-2407

June 13, 2005

Animal Damage Management Board
2219 Carey Ave.
Cheyenne, WY 82002

Attn: Hank Uhden

Attached please find the ANNUAL REPORT for the Wolverine Presence Project.

Thank you,

Dick Staiger

Richard D. Staiger
Coordinator

WOLVERINE PRESENCE PROJECT

Project Status as of 4-30-05:

Overview:

All cameras were bench tested for function and appeared to be in working order. Testing included freezer test for cold weather operation with Lithium batteries.

Cameras were then field tested in cold weather with very poor results. They would not function properly.

Because of the large amount of volunteer time and effort involved in locating and maintaining cameras with the strong likelihood of no results it was decided not to send the cameras out and the project was not activated for the 2004-2005 season.

Economics:

Total Project Receipts:	\$5,300.00
Total Expenses:	\$2,153.11
Returned to ADMB 5-24-04	\$2,646.89
Project Balance:	\$ 500.00

Respectfully,

Dick Staiger

Richard D. Staiger
6-7-05

Black Bear/Grizzly Bear/Mountain Lion Depredation Prevention

FY06 PROJECT UPDATE
JULY 1, 2005- DECEMBER 1, 2005

Project Title: Grizzly Bear/Black Bear/Mountain Lion/Livestock Depredation Prevention and Control

Brief Synopsis of project: This is a request for the ADMB to fund, through cost share on a 25:75 match basis, up to a maximum of $12,500.00 with the Wyoming Game and Fish Commission to contract with USDA Wildlife Services to alleviate grizzly bear, black bear and mountain lion depredation to livestock, bees and beehives in all counties of the State.

Project Update: During the first quarter of FY06 USDA Wildlife Services billed the WGFD for 800.5 hours of grizzly bear damage investigation and control for a total of $25,616.00, 50 hours of mountain lion damage investigation and control for a total of $1,600.00, and 106.2 hours of black bear damage investigation and control for a total of $3,398.40. Total hours charged during the first quarter of FY06 was 956.7 for a total monetary obligation of $30,614.40. The total monetary obligations for the first quarter of FY06 under the terms of the grant agreement (25/75 match) were: ADMB $7,653.60, WGFD $22,960.80. USDA Wildlife Services charges are itemized in Table 1.

Table 1. Itemized charges by USDA Wildlife Services for work performed for the WGFD to investigate and manage trophy game damage.

FY06 WGFD CONTRACT EXPENDITURES

July 1, 2005- Sept. 30, 2005

Wyoming Game and Fish Tax ID #: 83-02008667

WY G&F Agree #: 06SC04032222
Acctg Code: 5737356316
Agree #: 0573585378
Bill #:

Dates	Name	Species	Hrs	Salary Per Hour	Salary Cost
8/8-9/05	Chuck Bunch/Mike Kimsey	Mtn Lion	4.0	$32.00	$128.00
8/24/2005	Chuck Bunch/LU Sheep Co	Black Bear	4.0	$32.00	$128.00
9/7/2005	Chuck Bunch/North Fork Owl Creek	Griz Bear	8.5	$32.00	$272.00
7/11-13/05	Arnie DeBock/Upper Green River	Griz Bear	27.0	$32.00	$864.00
8/1-5/05	Arnie DeBock/Upper Green River	Griz Bear	43.0	$32.00	$1,376.00
6/28/2005	Jed Edwards/Smith Fork	Mtn Lion	6.0	$32.00	$192.00
9/6/2005	Jed Edwards/Three Forks	Black Bear	8.0	$32.00	$256.00
7/25-8/10/05	Wade Jones/Upper Green River	Griz Bear	98.5	$32.00	$3,152.00
7/20-28/05	Matt Lumley/Porcupine	Black Bear	16.0	$32.00	$512.00
9/8/2005	Matt Lumley/Blind Bull	Black Bear	7.0	$32.00	$224.00
7/13-31/05	Rod Merrell/Upper Green River	Griz Bear	86.5	$32.00	$2,768.00
8/11-28/05	Rod Merrell/Upper Green River	Griz Bear	121.5	$32.00	$3,888.00
9/1-25/05	Rod Merrell/Upper Green River	Griz Bear	122.0	$32.00	$3,904.00
8/15-28/05	Dan Braig(Temp)/Upper Green River	Griz Bear	121.5	$32.00	$3,888.00
9/1-2/05	Dan Braig(Temp)/Upper Green River	Griz Bear	21.5	$32.00	$688.00
7/18/2005	Steve Moyles/Truman Julian	Black Bear	8.0	$32.00	$256.00
7/25/2005	Steve Moyles/Truman Julian	Mtn Lion	8.0	$32.00	$256.00
8/7-15/05	Merrill Nelson/Upper Green River	Griz Bear	72.0	$32.00	$2,304.00
9/2-14/05	Merrill Nelson/Kendal Bear Camp	Griz Bear	63.5	$32.00	$2,032.00
7/29/2005	Kent Officer/Joe Broadbend	Black Bear	4.0	$32.00	$128.00
8/5/2005	Kent Officer/Joe Broadbend	Black Bear	5.2	$32.00	$166.40
7/29-8/19/05	Jim Pehringer/Table Mtn & Bald Ridger	Griz Bear	15.0	$32.00	$480.00
8/2/2005	Mike Peterson/Redland	Black Bear	4.0	$32.00	$128.00
8/11-29/05	Mike Peterson/Harlan	Mtn Lion	25.0	$32.00	$800.00
8/14/2005	Mike Peterson/Turk	Black Bear	3.5	$32.00	$112.00
9/2/2005	Mike Peterson/Curuchet	Black Bear	3.0	$32.00	$96.00
8/29-9/2/05	Mike Peterson/Harlan	Black Bear	18.0	$32.00	$576.00
9/26/2005	Mike Peterson/ Red Fork	Mtn Lion	7.0	$32.00	$224.00
7/26-8/23/05	Tracy Villwok	Black Bear	25.5	$32.00	$816.00
				$32.00	$0.00
	Totals		956.7		$30,614.40

Submitted by: Scott Talbott

Affiliation: Wyoming Game and Fish Department
Mailing Address: 3030 Energy Lane, Suite 100
City: Casper, Wyoming **Zip:** 82604
Phone: 307-473-3404 **Fax:** 307-473-3433
E-mail: Scott.Talbott@wgf.state.wy.us

Extended Release of Mifepristone for field delivery to coyotes

EXTENDED RELEASE OF MIFEPRISTONE (RU 486)
FOR FIELD DELIVERY TO COYOTES

Co-investigators:
Steven W. Horn, Department of Animal Science, University of Wyoming
M. Delwar Hussain, School of Pharmacy, University of Wyoming
Charles E. Stith, USDA, Agricultural Research Service, Laramie, Wyoming
Weiling He, School of Pharmacy and Dept of Animal Science, University of Wyoming

ANNUAL PROGRESS REPORT

12/1/04 – 12/1/05

PROJECT SUMMARY

Research conducted during the past year has centered on developing an effective single dosage formulation of mifepristone (RU 486) to terminate pregnancy in coyotes (*Canis latrans*). Investigations have concentrated on two approaches in order to sustain the release of the compound over time within the coyote: 1.) Development of a solid dispersion formulation to enhance the bioavailability of mifepristone; and, 2.) Development of a sustained release nanoparticle formulation that releases mifepristone within the coyote over a three day period. Previous research has demonstrated the effectiveness of this antiprogestin compound in controlling reproduction in captive coyotes when 10 mg/kg of mifepristone was delivered in oral baits for three consecutive days. This year development of both solid dispersion and sustained release nanoparticle formulations of mifepristone have yielded significant results, 100% and 66% respectively, in terminating the pregnancies of captive coyotes after they consumed a single oral bait. Further, in-vitro and in-vivo preliminary studies with rats (*Ratus ratus*) have shown that bioadhesive nonoparticles of DL-lactide/glycolide copolymers (PLGA) can sustain the release of mifepristone for at least three days. Extensive in-vivo studies with coyotes this fall investigated treatment efficacy and bioavailability of three compound formulations. The results of the bioavailability studies will be applied in the 2006 coyote breeding season beginning in January.

SUMMARY OF 2005 ACOMPLISHMENTS

Thirteen male-female pairs of coyotes were identified and kenneled in a fashion to promote the establishment or reaffirmation of pair bonds. Males were placed with females for 24 hours, every other day beginning on January 24, 2005. The pairings continued until March 1, 2005 when no further signs of estrus were evident. A total of 12 copulatory ties were observed. The 13 mated females were palpated and ultrasounded on March 22 and 24, 2005. Eight females were diagnosed pregnant (the presence of at least two embryonic vesicles). The eight pregnant coyotes were randomly assigned to three experimental groups (two treatment groups of three coyotes each and one control group with two coyotes). All coyotes were treated on April 1, 2005.

Treatment Group 1: Each coyote received a lard bait containing nanoparticles prepared from 1A and 2A PLGA (encapsulating 300 mg equivalent of mifepristone). Two of the three females failed to whelp puppies. One female whelped three puppies on April 19, 2005, after a normal gestation of 62 days

Treatment Group 2: Each coyote received a lard bait containing 600 mg of a solid dispersion of mifepristone and PEG 4000(1:1) (equivalent to 300 mg of mifepristone). All three females failed to whelp puppies.

Control Group: Each coyote received a lard bait containing 600 mg of corn starch. One female whelped three puppies on April 15, 2005 after a normal gestation of 62 days. The other female failed to whelp puppies.

In-vitro studies show that the mifepristone nanoparticles, specially the 1A nanoparticles sustains the release of mifepristone for at least 3 days. In-vivo studies in rats also showed that this bioadhesive nanoparticle can release mifepristone for at least 3 days when administered orally.

The in-vivo efficacy studies in coyotes with both solid dispersion and nanoparticle formulation yielded significant results, albeit small sample sizes.

The solid dispersion of mifepristone and PEG 4000 (1:1) had 100% success for Treatment Group 2 after a single administration of the formulation. The success of this formulation may be due to increased bioavailability. Earlier studies have shown low bioavailability of mifepristone in coyotes (only ~30%) and PEG 4000 may increase the bioavailability by increased dissolution (Hussain, 2002). Also, serum concentrations of mifepristone on 3rd day were higher and more consistent in coyotes after oral administration of the PEG 4000 solid dispersion of mifepristone.

The nanoparticle formulation was successful in two out of three treated coyotes in Treatment Group 1. The formulation consists of mixtures of PLGA 1A and 2A in 2:1 ratio. The mixed nanoparticles in this ratio were based on their in-vitro release profile. The objective was to obtain continuous delivery of mifepristone for at least three days. Based on the results from this current study, the mixed nanoparticle formulation, it is possible that the formulation may have a slow initial release of mifepristone after oral administration. For future studies, the formulation will be changed to a mixture of PLGA 1A nanoparticle and straight mifepristone in a 2:1 ratio. The straight mifepristone and the nanoparticles together should be able to have initial high mifepristone serum concentration and also maintain effective serum concentration of mifepristone for at least three days.

The failure of one of the two control subjects to whelp is possibly the result of a misdiagnosis of pregnancy. This subject showed little mammae development and palpation yielded no results. Ultrasound analysis showed what appeared to be an embryonic vesicle. That possible embryonic vesicle may have inadvertently been counted twice, thus yielding a positive diagnosis of pregnancy.

This past summer, the bioavailability of mifepristone alone, mifepristone-PEG 4000 solid dispersion and PLGA 1A nanoparticles were studied in rats. This study was essential to illustrate the increased bioavailability of mifepristone-PEG 4000 solid dispersion and the sustained release of mifepristone from the nanoparticles. Since it is easier to do the bioavailability study in rats in significant numbers than in coyotes, the data generated from the rat study was used to design the bioavailability study of these formulations in coyotes (fall, 2005). Individual rats in three groups were treated with one of the following treatments: mifepristone alone, mifepristone-PEG 4000 solid dispersion and PLGA 1A nanoparticles. The results showed that mifepristone release was sustained from mifepristone-PLGA-1A nanoparticles. Both in vitro and in vivo studies showed that these nanoparticles can release mifepristone quickly and can sustain the release for at least three days.

This fall nine male coyotes were divided into three groups to study the bioavailability of: 1.) Mifepristone only; 2.) Mifepristone-PEG 4000; and, 3.) Mifepristone-PLGA-1A nanoparticles. Blood serum was drawn from each coyote at 2, 4, 6, 10, 24, and 48 hrs. after delivery of the compound. Each coyote received their respective treatment presented in a 26.5g ball of lard kept at -20 degrees Centigrade until administration. The results are still being analyzed, but it appears that the Mifepristone only group looks very similar to the other two groups (PEG 4000 and nanoparticles) when comparing how rapidly the compound was metabolized within the first 48 hrs. after administration.

In-vivo efficacy of the compounds will be studied in coyotes this winter. Thirteen pairs of coyotes have again been identified and paired for possible breeding. Males will be presented to the females beginning in mid-January through the end of February. Ideally, it is proposed that 113 pregnant coyotes be divided into four experimental groups:

Treatment group 1 (Mifepristone only): Coyotes will receive one single dose of 10 mg/kg of mifepristone in a lard bait.

Treatment group 2 (Mifepristone:PEG 4000 1:1): Coyotes will receive one single dose of Mifepristone:PEG 4000 1:1 solid dispersion equivalent to 10 mg/kg of mifepristone in a lard bait.

Treatment group 3 (Mifepristone nanoparticles): Coyotes will receive one single dose of PLGA 1A nanoparticles and mifepristone (ratio of 2:1) equivalent to 10 mg/kg of mifepristone in a lard bait.

Treatment group 4 (Control): Coyotes will receive one single dose of 600 mg corn starch in a lard bait.

The size of the treatment groups and the final research design will depend on the number of confirmed pregnancies.

Whiskey Mountain Big Horn Sheep Predation Project

Whiskey Mtn Bighorn Sheep Project

This is a brief update on the Whiskey Mtn Bighorn Sheep project. Since the sheep population data are not available at this time, and field work is still ongoing, this is a preliminary report, and as this is the last year of the project, we will provide a more thorough final report after the end of December.

This is a progress report of Wildlife Services activities in the Dubois area funded by FNAWS and the ADMB to protect bighorn sheep. This is the third and final year of the project and the program was conducted similar to previous years. WS role is to conduct field work to remove coyotes from the project area. During the last year WS personnel spent approximately 210 hours on site (does not include travel time to and from the site) using aerial and ground methods to remove a total of 62 coyotes (58 aerial and 2 trapped and 2 shot). WS has flown approximately 42 fixed wing hours (including ferry time from Rock springs) in support of the project. The majority of the coyotes were taken on the winter ranges during the months the sheep use those ranges.

Merrill Nelson worked in the upper country of the project this past summer and noticed very little coyote sign and a fair number of fox.

Aerial control efforts were conducted this past week with 9 coyotes removed (included above), 6 from the north side and 3 from the south side of the highway. Two dead ewes were observed on the south side. DS Merrill Nelson and Warden Cole Thompson were able to verify the first as a coyote kill. The ewe appeared to be in good condition and in her prime. The second ewe was not confirmed but the aerial crew observed that it also appeared to be predation. Twenty wolves in two groups were observed yesterday, in the Spence Moriarty Unit with sheep and elk nearby.

Official counts conducted by the Wyoming Game and Fish were not available at this time. In late November, Merrill Nelson observed 50 ewes, 14 lambs and 16 rams on Sheep Ridge. He also observed about 40 head of sheep on Torrey Rim but was not able to classify.

Once the population data is available from the WG&FD, I would like to meet with representatives of FNAWS and WG&FD to review the project for results and determination of any future activities and to put together a final report. When discussing the future of this project it should be noted there is an effort underway to introduce a bill requesting funding for predator managment from the Legislature. Currently local predator management programs conducted by the county Predatory Animal Boards (PABs) (who often contract with Wildlife Services) are funded primarily by livestock brand inspection fees. A recent determination by the Wyoming Attorney General's Office, that predator management for the protection of wildlife is a responsibility of the County Predatory Animal Boards is significant. This effort to obtain funding from the state to the PABs is in part, to enable them to provide services for protection of wildlife.

A very positive outcome of this effort could be that the bighorn sheep in the Dubois area, in the future, would receive routine predator management from the PAB without having to seek outside funding.

Sorry I am not able to attend your meeting. I enjoy the relationship we have with FNAWS and hope to continue to help you in your efforts "to put sheep on the mountain".

Rod Krischke
State Director
Wyoming Wildlife Services
307 261-5336

Devil's Canyon Supplemental Big Horn Sheep Transplant

To: Animal Damage Management Board

From: James J. Pehringer

Devil's Canyon Bighorn Sheep Project 2006

2006 Budget request (Nov. 2005 – June 2006) - $8000.00

Due to hunting seasons in this area starting September and ending in November we have had little activity in the project area to avoid disruption of public recreation. We will begin aerial activities when ideal conditions are received starting in December. I was able to spend some time in the area during the month of August and found a few coyotes which were removed and suspect a drift of new inhabitants starting November. Predation from the new transients normally do not show up significantly until snow conditions become crusted which enable them to trap bigger game in snow depths and allow them fast and efficient mobility over the top. At this time we will be in the area regularly working to keep this to a minimum. There had been some mountain lion predation identified this fall and with help of the upcoming hunting season we may see relief.

Observations in this are have proven Ewe/lamb numbers to be excellent and the sheep look as healthy as they can be. We are looking forward to this year's transplant and hope we will get more in the future.

We are excited for this year as we are able to use this money during the time of year which we can be most effective in contributing to the success of the project.

I have enclosed last year's budget and activities and have put together a work schedule for this year. This work schedule can and will vary due to the conditions of weather we receive this year. We plan to make the most of every dollar in the best efficient way possible to achieve success for this project.

If anyone has any question about USDA-WS activities in this project at any time please call me at (307) 587-5271.

Budget 2005
Devil's Canyon Bighorn Sheep

- ADMB/G&F Funds $6,200.00
- Wyoming FNAWS $3,000.00 available if needed (did not use).

- Helicopter Time 3.5 hrs. @ $375.00 hr. $1,312.50

- Jack Clucas $70.00 a day and .16 mile $736.64 - 10 days and 229 miles total.

- Fixed Wing 16.05 hrs. @ $85.00 hr. $1,364.25

- Dave Blakeman 7.5 days @ $238.50 day $1,788.75

- Total $5,202.14 (BH County PAB will have to return $ 997.86 to ADMB)

Coyotes taken:

- 1 coyote helicopter.
- 10 coyotes fixed wing.
- 5 coyotes den/firearms
- 13 Denning

Fremont County sage grouse/ mule deer project

FREMONT COUNTY SAGE GROUSE AND MULE DEER PROJECT

FREMONT COUNTY PREDATOR ANIMAL BOARD
Tracy Frye
USDA APHIS Wildlife Services
67 Dodrill Road
Riverton, WY 82501
Ph. 307-857-2015

Dear ADMB,

Here is our year end progress report on our project. We received an initial 3000.00 to start the sage grouse portion of our project. This money was received in May 2005 and was used entirely to purchase the Talon trail cameras we needed. The ADMB board asked us to return at their next meeting in July 2005 to re-submit our project proposal to secure the rest of the needed funding. We received the trail cameras in late May and were able to put them out for a few weeks with the artificial sage grouse nests. As we were not full funded and with the grouse nesting season coming to a close, we decided that this would be considered a “practice” time for learning how to use the cameras effectively and to work out any problems that might come up with using them. Our initial findings during this time were the following: Two artificial nests in the control area were molested by ravens and one nest in the treatment area. The eggs were totally destroyed and eaten in all three cases with ravens photographed at each nest site. No other eggs or nests were molested during this short period. We also used this time to do coyote population census work, make maps of sage grouse nest in both the control and treatment area and make work plans. The Fremont County PAB funded all work during this time in anticipation of being fully funded in the fall of 2005. We received the 10,000.00 yearly funding in September.

Our main focus of work at this time is the reduction of coyotes in the treatment area of the project. Approximately 100 M-44s have been set and some aerial hunting has been done. Approximately 40 coyotes have been taken at this time. We are also helping the Wyoming Game and Fish Department with deer population counts in the area.

We recently met with Wyoming Game and Fish Dept. personnel in Lander to discuss the boundaries of the project. It was mutually agreed that a small change was needed in order to get a better count on mule deer populations. Rod Merrill and I were invited to give a talk about our project in Lander to the Sportsmen for Fish and Wildlife local chapter. They were supportive of our project and would like to be kept informed about our progress. They also stated that there might be some funds available in the future for the project or others like it.

We will be putting out 10 artificial sage grouse nest at the start of the nesting season in the late spring. Our project is really just getting going good, so there isn't a lot more to report on. I would like to again thank the ADMB for funding this project. I look forward to making it a success.

Sincerely,

Tracy Frye
USDA Wildlife Services

Increasing speed & potency of Theobromine and Caffeine based toxicants

United States Department of Agriculture

Animal and Plant Health Inspection Service

Wildlife Services

National Wildlife Research Center
4101 LaPorte Ave
Fort Collins, CO 80521

Phone: (970) 266-6082
Fax: (970) 266-6089
Email: john.j.johnston@usda.gov

December 4, 2005

Animal Damage Management Board
2219 Carey Avenue
Cheyenne, WY 82002-0100

Subject: Mid-term Progress Report Predator Toxicant

Dear ADMB:

I appreciate your funding of my request for INCREASING THE SPEED OF ACTION AND POTENCY OF THE THEOBROMINE AND CAFFEINE BASED PREDACIDE.

The proposal contained two objectives:

Objective 1: Determine potential U.S. EPA registration requirements, costs and time-line for a theobromine:caffeine predator toxicant.

Objective 2: Determine the effect of adding sodium benzoate and calcium citrate on the toxicity of theobromine and caffeine to rats

Objective 3: Determine the effect of adding sodium benzoate and calcium citrate on the toxicity of theobromine and caffeine to coyotes

Progress through 11-2005:

Objective 1: Determine potential U.S. EPA registration requirements, costs and time-line for a theobromine:caffeine predator toxicant.

Registration costs for a theobromine:caffeine predacide have been estimated by reviewing pertinent

EPA registration guidelines as well as the chemical and toxicological properties of theobromine and caffeine as summarized in the scientific literature. Based on this review, we estimate that common sense waivers and literature reviews will reduce the total potential registration costs to $815,280.

- Total cost for all potentially required studies = $4,681,000
- Projected savings
  - Waivers = $2,587,000
  - Literature = $1,279,000
- Projected study costs = $815,280

These Projected EPA registration costs are summarized below:

Data Group	Theobromine	Caffeine	Predacide Formulation	NRWC Studies & Costs	Contract Lab Costs
Product Chemistry	16,020	19,000	29,460	64,480	0
Non-target Wildlife	144,200	144,200	16,400	0	304,800
Toxicology	110,500	26,000	20,500	26,000	131,00
Non-target Plant	0	0	0	0	0
Post-Application Exposure	0	0	6,000	6,000	0
Environmental Fate	40,500	40,500	0	0	1,000
Applicator Exposure	0	0	2,000	2,000	0
Efficacy	0	0	$200,000	0	200,00
TOTAL ($815,280)	**$311,220**	**$229,700**	**$74,360**	**$98,480**	**$716,800**

Objective 2: Determine the effect of adding sodium benzoate and calcium citrate on the toxicity of theobromine and caffeine to rats

Toxicity tests were conducted with Wistar Rats.

Rats were orally gavaged with aqueous solutions of the test mixtures. Test mixtures consisted of theobromine:caffeine (5:1), theobromine:caffeine (5:1) plus various ratios of sodium benzoate, and theobromine:caffeine (5:1) plus various ratios of calcium citrate. The addition of sodium benzoate increased the toxicity of theobromine:caffeine (5:1) by approximately 40% percent. The optimal ratio of sodium benzoate to theobromine:caffeine (5:1) appeared to be 1:1. Calcium citrate had no effect on the toxicity of theobromine:caffeine (5:1).

Pending research:

Objective 3: Determine the effect of adding sodium benzoate and calcium citrate on the toxicity of theobromine and caffeine to coyotes

Coyotes will be transported from NWRC Logan field station and University of Wyoming at Laramie to the Outdoor Pen Facility at NWRC headquarters in Fort Collins. The toxicity of theobromine:caffeine (5:1):sodium benzoate (1:1) to coyotes will be compared with sodium benzoate:caffeine (5:1).

If you require any additional information, please contact me at john.j.johnston@usda.gov or 970-266-6082. I appreciate your support of my research.

Sincerely,

John J. Johnston Ph.D. MBA
Chemistry Project Leader, NWRC

Evaluation of Cocoa- and Coffee-Derived Methylxanthines as Toxicants for the Control of Pest Coyotes

John J. Johnston

USDA/APHIS/Wildlife Services, National Wildlife Research Center,
4101 LaPorte Avenue, Fort Collins, Colorado 80521

Hank & WY ADMB -
Thanks for all
your support on
this project!!
John Johnston

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FOOD CHEMISTRY

Evaluation of Cocoa- and Coffee-Derived Methylxanthines as Toxicants for the Control of Pest Coyotes

JOHN J. JOHNSTON

USDA/APHIS/Wildlife Services, National Wildlife Research Center, 4101 LaPorte Avenue, Fort Collins, Colorado 80521

Methylxanthines were quantified in coffee, tea, and chocolate products. Tarajuilie tea from India, cocoa powder, and cocoa nibs contained the highest levels of methylxanthines. Theobromine, caffeine, and theophylline combined in the ratios observed in tea and chocolate were ingested by coyotes. Although both mixtures induced acute toxicity, the symptoms accompanying the chocolate methylxanthine mimic were preferable. Manipulation of the ratios of methylxanthines in the chocolate mimic led to the identification of a 5:1 theobromine/caffeine mixture as a promising coyote toxicant. This mixture was then administered to coyotes using the coyote lure operative device (CLOD). Mortality occurred in every coyote that ingested any portion of the CLOD contents. These results indicate that mixtures of theobromine and caffeine have the potential to be developed into a selective, effective, and socially acceptable toxicant for the control of pest coyotes.

KEYWORDS: **Methylxanthine; theobromine; caffeine; CLOD; coyote toxicant**

INTRODUCTION

In 2000, U.S. farmers and ranchers reported \$51.6 million in cattle and calf losses from animal predators. Canids (coyotes, dogs, wolves) were responsible for 83.4% of these predatory losses (*1*). That same year, U.S. farmers and ranchers suffered \$16.5 million in sheep and goat losses from animal predators. Canids were responsible for 75.8% of these losses (*2*). Coyotes were responsible for ~80% of livestock predation attributed to canids. Overall, coyotes were responsible for \$44 million in U.S. livestock losses.

Other damage caused by coyotes include collisions with aircraft (*3*, *4*), attacks on pets (*5*) and children (*6*), damage to fruit and vegetable crops (*7*), predation on game species such as elk and deer (*8*), and predation on poultry (*9*). In addition to directly damaging fruit and vegetable crops, coyotes also contribute to crop losses via damage to hose irrigation systems (*10*). Coyotes have also been implicated in the transmission and spread of epizootic rabies in the United States (*11*).

Ranchers and pest control specialists use a variety of control techniques to minimize damage caused by coyotes. These techniques include exclusion (fencing), guard animals, scaring devices, trapping, shooting, and toxicants (*12*). The broad-spectrum mammalian toxicants sodium cyanide and sodium fluoroacetate (Compound 1080) are the only oral toxicants registered for predator control in the United States. In 1998, California voters passed Proposition 4, which severely restricted the use of sodium cyanide and sodium fluoroacetate for the control of livestock predators such as coyotes. As toxicants are a critical component of nearly all integrated pest management strategies (*13–15*), these bans severely restrict the ability of ranchers and pest control specialists to limit losses caused by coyotes. Since the passage of Proposition 4 in California, similar initiatives have been passed in Colorado and Arizona. It is likely that this trend will expand to other states. As the development of pesticide products and subsequent registration with the U.S. Environmental Protection Agency (EPA) typically require more than a decade of work, it behooves the agricultural community to proactively develop new, more selective, and socially acceptable toxicants for pest (especially predatory) coyotes.

Because most predator-induced livestock losses in the United States are due to canids, a predator control substance should exhibit a high degree of toxicity to canids and, ideally, a low toxicity to nontarget animals and humans. The propensity for dogs to overdose on chocolate is documented in the veterinary literature (*16–18*). The methylxanthines theobromine and, to a lesser extent, caffeine are believed to be responsible for the toxicity of chocolate to dogs (**Figure 1**) (*19*, *20*). In addition to chocolate, significant quantities of theobromine and caffeine are found in tea, coffee, and cola beverages. Although it is unknown what levels of caffeine and theobromine are acutely toxic to humans, given our significant and constant exposure to these compounds, toxicity to humans is likely quite low. However, 40 kg (100 pound) dogs have been poisoned by the amounts of these compounds contained in 0.5 pound of cocoa powder or 1 pound of dark chocolate (*18*). These findings suggest that theobromine and caffeine are much more toxic to canids than to humans—a desirable characteristic for any predacide. Theobromine and caffeine oral LD_{50} (median lethality) values for dogs are 200–500 and 140 mg/kg of body weight (BW), respectively (*21*, *20*). Theobromine and caffeine LD_{50} values for rats are 1265 and 355 mg/kg of BW, respectively (*22*, *23*). Additionally, for theobromine, the oral LD_{50} value is 837 mg/kg for mice (*22*). For caffeine, there is a reported LD_{50} value

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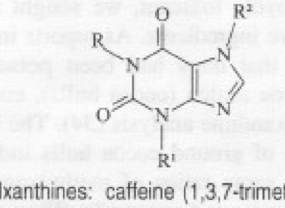

Figure 1. Methylxanthines: caffeine (1,3,7-trimethlxanthine), R = R[1] = R[2] = CH_3; theobromine (3,7-dimethylxanthine), R = H, R[1] = R[2] = CH_3; theophylline (1,3-dimethylxanthine), R = R[1] = CH_3, R[2] = H.

of 246 mg/kg of BW for rabbits (*23*). A comparison of these toxicity values suggests that methylxanthines are more toxic to dogs than to many other animal species.

The enhanced toxicity of these compounds to canids is believed to be related to a metabolic pathway that produces an unidentified methylxanthine metabolite that is unique to dogs. Additionally, unique ratios of *N*-demethylase-derived detoxification enzyme activities have been observed in dogs. These metabolism idiosyncrasies likely contribute to the relatively long half-life of methylxanthines in dogs (*24*). Methylxanthine toxicosis is associated with diuresis and inhibition of cellular calcium reuptake, which increase free calcium concentration and enhance skeletal muscle contractions. The resulting symptoms of methylxanthine toxicosis typically include central nervous system stimulation and tachycardia (*25*).

Although it would be impractical to deliver several pounds of chocolate to coyotes, the methylxanthines theobromine and caffeine are routinely extracted and concentrated from cocoa manufacturing waste and coffee beans, respectively. These compounds can be reformulated to duplicate the methylxanthine ratios in chocolate. This methylxanthine mixture could then be evaluated for its potential as a canid-specific toxicant. Additionally, it may be possible to manipulate the methylxanthine ratios to improve the potency, selectivity, symptoms, and/or cost of the pest coyote toxicant. Furthermore, it is possible that familiarity with coffee, tea (caffeine), and chocolate (theobromine) would minimize public perception of risk and possible opposition to such a product.

The objective of this research was to determine if methylxanthines have potential as active ingredients for a selective toxicant to control pest coyotes. To accomplish this goal, the following experiments were conducted to (1) identify natural products that contain significant quantities of methylxanthines, (2) develop two prototype pest coyote toxicants containing the methylxanthine ratios in the natural products identified in the first step, (3) conduct toxicity tests to determine the optimal combination of methylxanthines for development as an improved toxicant for pest coyotes, and (4) administer the pest coyote toxicant to captive coyotes using a proven field delivery device (*26*−*28*). All study procedures and experimental design were reviewed and approved by the WS/NWRC Institutional Animal Care and Use Committee.

MATERIALS AND METHODS

Materials. Ground (espresso grind) Arabian Mocha Sanani, Ethiopian Fancy, Kenya, Colombia, Costa Rica, Guatemala, Nicaragua las Hermanas, New Guinea, Sulawesi-Kalosi, and Sumatra coffee beans and green (Gunpowder) tea (China) were purchased from Peet's Coffee (Berkeley, CA). Tarajuilie orange pekoe [Thakubari, (northeastern) India], Nonsuch [Nilgiri, (south-central) India), Kambaa Estate Kenya (Africa), and St. Coombs pekoe (Sri Lanka) black teas were purchased from Jolly Good Tea and Gifts (Vader, WA). Cocoa nibs, cocoa powder, bittersweet chocolate bars, and dark chocolate chunks were obtained from Italco Food Products (Denver, CO). Cocoa hulls (cocoa mulch) were obtained from Mirana International Resources (Palos Verde Peninsula, CA). Lard (Morrell Co., Cincinnati, OH), Crisco vegetable (soybean) oil (J. M. Smuckers, Orrville, OH), bacon (Shure Fine International, North Lake, IL), and canned Alpo Prime Cuts dog food (Purina, St. Louis, MO) were purchased from local supermarkets. Acetonitrile (Optima grade) was obtained from Fisher Scientific (Fair Lawn, NJ). Caffeine, theobromine, and theophylline for analytical standards were obtained from Sigma Chemical (St. Louis, MO). Natural caffeine and theobromine extracts were obtained from Pechiney World Trade USA (Stamford, CT).

Coyotes. Adult coyotes (mixed sex) were obtained from the U.S. Department of Agriculture (USDA)/Animal and Plant Health Inspection Service (APHIS)/Wildlife Services (WS)/National Wildlife Research Center (NWRC) field station in Millville, UT, and from the University of Wyoming's predator colony in Laramie, WY. Coyotes were transported to and housed in the USDA/APHIS/WS/NWRC outdoor pen facility in Fort Collins, CO. Coyotes were quarantined for at least 2 weeks prior to toxicity testing. Coyotes were maintained on a daily ration of 350 g of Mazuri Exotic Canine Diet (PMI Nutrition International, Brentwood, MO) and water ad libitum.

Methylxanthine Analyses. In a 50 mL beaker, 10 ± 1 mg of ground coffee beans, tea leaves, chocolate, cocoa powder, cocoa nibs, or cocoa hulls was added to 10 mL of boiling water and covered with a watch glass. Each sample was simmered for 5 min and subsequently permitted to cool at room temperature for 20 min. The aqueous extract was filtered through a 0.45 μm filter for subsequent injection of 10 μL aliquots into a Hewlett-Packard (Palo Alto, CA) high-performance liquid chromatograph equipped with an H-209 4.6 × 250 mm column. Methylxanthines were separated using an acetonitrile/water (11:89) mobile phase at a flow rate of 1 mL/min. Methylxanthines were detected by ultraviolet absorption (245 nm) and quantified versus external standards.

Methylxanthine Dosing. *Test 1.* Caffeine and theobromine were mixed together at a ratio of 1:13 to prepare a methylxanthine chocolate mimic. A methylxanthine tea mimic was prepared by mixing a 20.5:1.3:1 ratio of caffeine/theobromine/theophylline. The mimics (15 g) were individually mixed with ~40 mL of lard/rendered bacon fat/soybean oil (5:1.5:1). Two coyotes were offered the lard mixture containing the chocolate mimic, and two coyotes were presented the lard mixture containing the tea mimic. The mixtures were offered in tared stainless steel bowls. After 3 h, the bowls and unconsumed mimics were removed and weighed to determine consumption.

Test 2. Aqueous suspensions of methylxanthines were prepared by combining 1 part theobromine or theobromine/caffeine mixture with 24 parts water. A variety of theobromine/caffeine ratios were evaluated. Coyotes were sedated with 1 mL of Dormitor (1 mg of medetomidine hydrochloride) (Pfizer Animal Health, Exton, PA) and 0.5 mL of Ketaved (50 mg of ketamine hydrochloride) (Vedco, St. Joseph, MO). Methylxanthine suspensions were administered via oral gavage, followed by 60 mL of water to ensure quantitative delivery of the desired quantity of methylxanthines. For each methylxanthine mixture evaluated, methylxanthines were administered to 16 coyotes; 4 coyotes were dosed at each of 4 different dose levels. After the coyotes had been replaced in their cages, the sedation was reversed by the administration of 1 mL of Antisedan (5 mg of atipamezole hydrochloride) (Pfizer Animal Health). Coyotes were observed intermittently for at least 12 h postdosing.

Toxicity data were analyzed by constructing log dose versus probit mortality curves for each theobromine/caffeine mixture evaluated. The median lethal dose (LD_{50}) and 99% lethal dose (LD_{99}) were calculated for each mixture (*29*).

Test 3. Canned dog food was mixed 2:1 with water and blended to achieve a "pourable" homogenate. The dog food homogenate was blended with corn syrup and the 5:1 theobromine/caffeine mixture to give a final ratio of 3:3:2 (dog food homogenate/corn syrup/theobromine/caffeine mixture). It was hoped that the corn syrup would mask the bitterness of the methylxanthines and that the dog food would serve as an incentive for the coyotes to consume the mixture. Fifty grams of this mixture was added to the 60 mL reservoir of a coyote lure operative device (CLOD) (**Figure 2**), a device that is being developed to deliver a variety of active ingredients to coyotes under field conditions (*30*−*33*). No lure or attractant was placed on the CLOD. A single

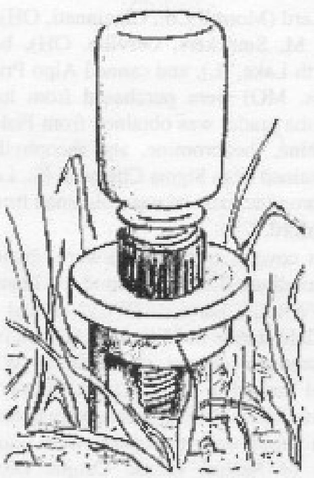

Figure 2. Coyote lure operative device (CLOD). Below-ground stake is not shown. Adapted from Berentsen et al. (*33*).

CLOD was then secured to the floor of each of six coyote pens at 7:00 a.m. Coyotes were observed at 0.5 h intervals. Unconsumed CLODs and CLOD remnants were removed at 10:00 a.m. This experiment was repeated with another six coyotes using a methylxanthine/dog food formulation containing a 4:1 ratio of theobromine/caffeine.

RESULTS AND DISCUSSION

Methylxanthines were quantified in a variety of coffee, tea, and chocolate extracts from samples grown at various locations to evaluate the possibility of a source that was particularly rich in methylxanthines. Caffeine was the only methylxanthine identified in coffee (**Figure 3**). Regardless of the country of origin, coffee samples from around the world contained ~27 mg of caffeine/g of coffee beans (**Figure 4**). Caffeine, theobromine, and theophylline were detected in tea samples from around the world. Methylxanthine levels were highest in Tarajulie tea from India. Caffeine, theobromine, and theophylline ratios in aqueous Tarajulie tea extracts averaged 20.5:1.3:1, respectively (**Figure 4**). Theobromine and caffeine were detected in chocolate products. The highest methylxanthine concentrations were detected in cocoa nibs and cocoa powder. Theobromine and caffeine were detected in these products at a ratio of 13:1, respectively (**Figure 4**).

Test 1. On the basis of these findings, tea and chocolate methylxanthine mimics were prepared by combining caffeine, theobromine, and theophylline at the ratios observed in Tarajulie tea and cocoa powder. Lard/bacon fat/soybean oil-based mixtures of each mimic were consumed by two coyotes each. For each mimic, one coyote vomited shortly after consumption of the mimic-fortified lard mixture. These two coyotes survived. Both coyotes that retained the mimic/lard mixture died within 4 h of consumption. The coyote that retained the tea mimic received a dose of 204 mg of caffeine/kg of BW, 13.3 mg of theobromine/kg of BW, and 9.9 mg of theophylline/kg of BW. This animal died about 1.5 h after the onset of symptoms, which included trembling, increased salivation, and seizures. The coyote that retained the chocolate mimic received 31.6 mg of caffeine/kg of BW and 413 mg of theobromine/kg of BW. This animal died following about 15 s of relatively minor symptoms, which included recumbent posture and labored breathing. On the basis of the symptoms preceding mortality, it was decided to further pursue a toxicant based on the chocolate mimic formulation.

Test 2. Because an extract of cocoa beans or powder would likely be prohibitively expensive to provide the active ingredients for a pest coyote toxicant, we sought an economical source of these active ingredients. As reports in the veterinary literature indicated that dogs had been poisoned following consumption of cocoa mulch (cocoa hulls), cocoa hulls were obtained for methylxanthine analysis (*34*). The HPLC analysis of aqueous extracts of ground cocoa hulls indicated that the hulls contained the same ratios of methylxanthines as were detected in cocoa powder and cocoa nibs (**Figure 4**). Following estimation of the resources required to extract the several hundred grams of methylxanthines required for toxicity testing, kilogram quantities of cocoa hull extracted theobromine and coffee bean extracted caffeine were obtained from a commercial supplier.

Theobromine and caffeine were combined in the proportions observed in the cocoa extracts, 13:1 theobromine/caffeine, to permit an evaluation of the methylxanthine cocoa mimic toxicity to coyotes. Four coyotes were dosed at 450 mg/kg of BW; two coyotes were dosed with a water-based suspension, and two coyotes were dosed with a soybean oil based suspension. The coyotes dosed with the oil-based suspension regurgitated the suspension shortly after dosing; both of these animals exhibited no signs of toxicosis and survived. The animals dosed with the water-based suspension retained the dosing solution. Both animals exhibited relatively mild signs of toxicosis: increased salivation and slight trembling for several minutes. One animal died ~4 h after dosing. The other animal survived. On the basis of these results, it was decided to pursue water-based suspensions for subsequent experiments.

Dose versus response toxicity testing was conducted with water-based solutions using four groups of sedated coyotes (four coyotes per group) that were administered the cocoa mimic at 400, 450, 650, or 850 mg/kg of BW. Premortality symptoms were relatively mild, and mortality ranged from 50 to 100% (**Table 1**). Analysis of log dose versus probit mortality curve yielded LD_{50} [50th percentile (median) lethal dose] and LD_{99} (99th percentile lethal dose) estimates of 424 and 640 mg/kg, respectively.

Assuming that it would be more economical to pursue U.S. EPA registration of a single active ingredient rather than a two active ingredient product, we evaluated the toxicity of theobromine to coyotes. Four groups of sedated coyotes (four coyotes per group) were administered aqueous suspensions of theobromine at 400, 450, 650, or 850 mg/kg of BW. Premortality symptoms were relatively mild, and percent toxicity ranged from 0 to 75% (**Table 1**). Analysis of the log dose versus probit mortality curve yielded LD_{50} and LD_{99} estimates of 516 and 618 mg/kg, respectively.

Although theobromine or the cocoa mimic could be used to render an apparently humane mortality to coyotes, I felt that a more potent mixture of theobromine and caffeine would be needed to deliver a lethal dose under field conditions. Because caffeine is more toxic to canids than theobromine, different theobromine/caffeine ratios were evaluated in an attempt to identify a product that displayed the higher toxicity associated with caffeine while retaining the minimal premortality symptoms associated with theobromine. Coyotes were dosed with theobromine/caffeine mixtures at 600 mg/kg of BW. One coyote each was dosed via oral gavage with an aqueous suspension containing a 1:1, 1:2, 2:1, 4:1, 5:1, or 6:1 ratio of theobromine/caffeine. Coyotes dosed with the 1:1 and 1:2 mixtures exhibited undesirable symptoms of toxicosis and were euthanized. Coyotes dosed with the other mixtures died during the postdosing observation period. For these coyotes, premortality symptoms were considered to be acceptable, although the duration and

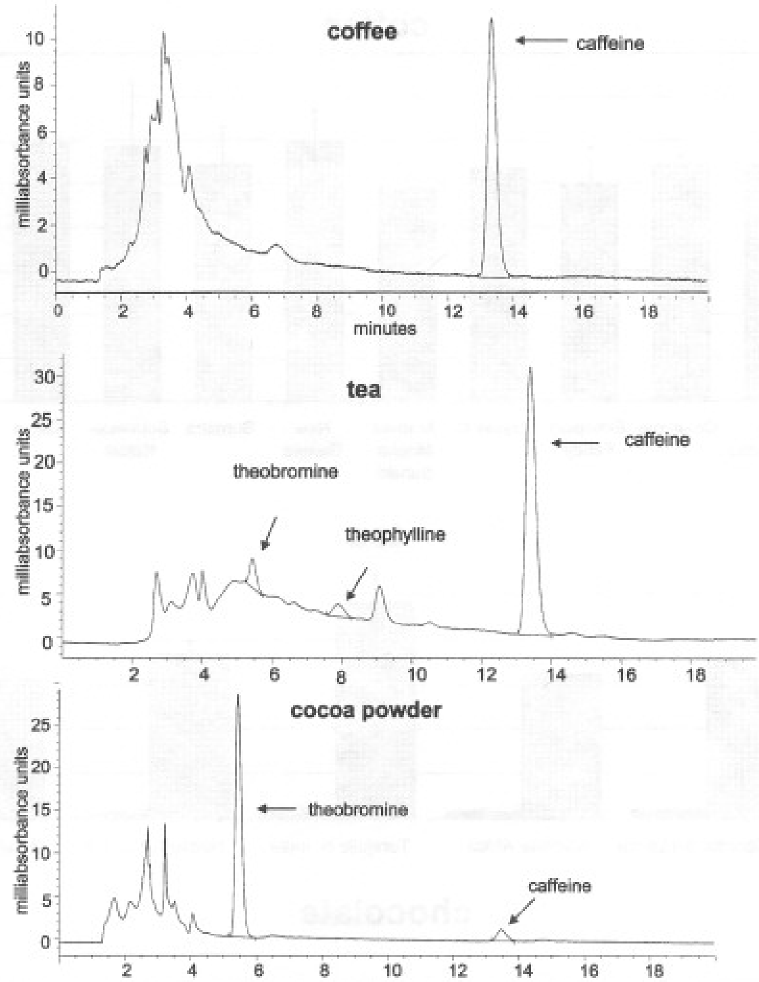

Figure 3. Chromatograms illustrating methylxanthine analyses of coffee (top), tea (Tarajulie) (middle), and cocoa powder (bottom).

magnitude of premortality symptoms generally decreased with increasing proportion of theobromine in the mixture.

On the basis of these findings, dose versus response toxicity testing was conducted with the 5:1 mixture of theobromine/caffeine. Four groups of sedated coyotes (four coyotes per group) were administered aqueous suspensions of theobromine at 250, 350, 450, or 650 mg/kg of BW. Percent toxicity ranged from 0 to 100% (**Table 1**). Premortality symptoms appeared to be nonexistent to minimal. Analysis of the log dose versus probit mortality curve yielded LD_{50} and LD_{99} estimates of 336 and 385 mg/kg, respectively. The increased toxicity of the 5:1 theobromine/caffeine mixture accompanied by minimal premortality symptoms suggested that this ratio of theobromine and caffeine has potential as a natural and socially acceptable toxicant for pest coyotes.

Test 3. To evaluate the potential of a methylxanthine-based pest coyote toxicant in a proven field delivery device, a 5:1 theobromine/caffeine formulation was added to CLODs and offered to six coyotes. All coyotes that bit the CLOD consumed some of the contents. Three of six coyotes consumed the CLOD contents. Consumption ranged from ~50 to 100% of the CLOD contents. Even though one of these coyotes vomited 15 min after consumption, all three coyotes died. Death occurred approximately between 2 and 7 h after the CLODs were offered to the coyotes. Premortality symptoms were extremely minimal and were of several seconds in duration. Estimated doses ranged from 700 to 1200 mg/kg.

Given the absence of undesirable symptoms noted in the coyotes dosed with the 5:1 theobromine/caffeine mixture, the CLOD experiment was repeated with a 4:1 theobromine/caffeine mixture and six coyotes (the higher proportion of caffeine should give a more potent and less expensive product). In this experiment, two coyotes consumed the CLOD contents. One animal consumed the entire CLOD contents and vomited shortly thereafter; this animal died 3 h after the CLOD had been offered. The second animal consumed only 10% of the CLOD contents; it died 8 h after being offered the CLOD. Both animals staggered before becoming recumbent. Labored breathing preceded mortality. Estimated doses ranged from 140 to 1300 mg/kg. Although no formal control experiments were conducted (e.g., coyotes dosed with mixtures containing no theobromine or caffeine), the fact that no coyotes died in the lowest test 2 treatment groups (**Table 1**) indicates that the dosing procedure did not contribute to mortality observed during these experiments. Also, given the limited numbers of coyotes available for this testing, it was not possible to detect sex- or age-related trends in sensitivity to methylxanthines.

To decrease the cost of active ingredients, several modifications could be considered including using smaller (30 mL) CLOD reservoirs with synthetic theobromine and caffeine. This would lower the cost of active ingredients to approximately \$0.40 per delivery device. Also, because methylxanthine toxicosis is mediated by an increase in intracellular calcium concentrations, it is possible that the potency of methylxanthine mixture may be increased by simultaneous administration of inorganic calcium. Alternatively, observations in our laboratory indicate that sodium benzoate increases the solubility and toxicity of caffeine. The addition of relatively inexpensive

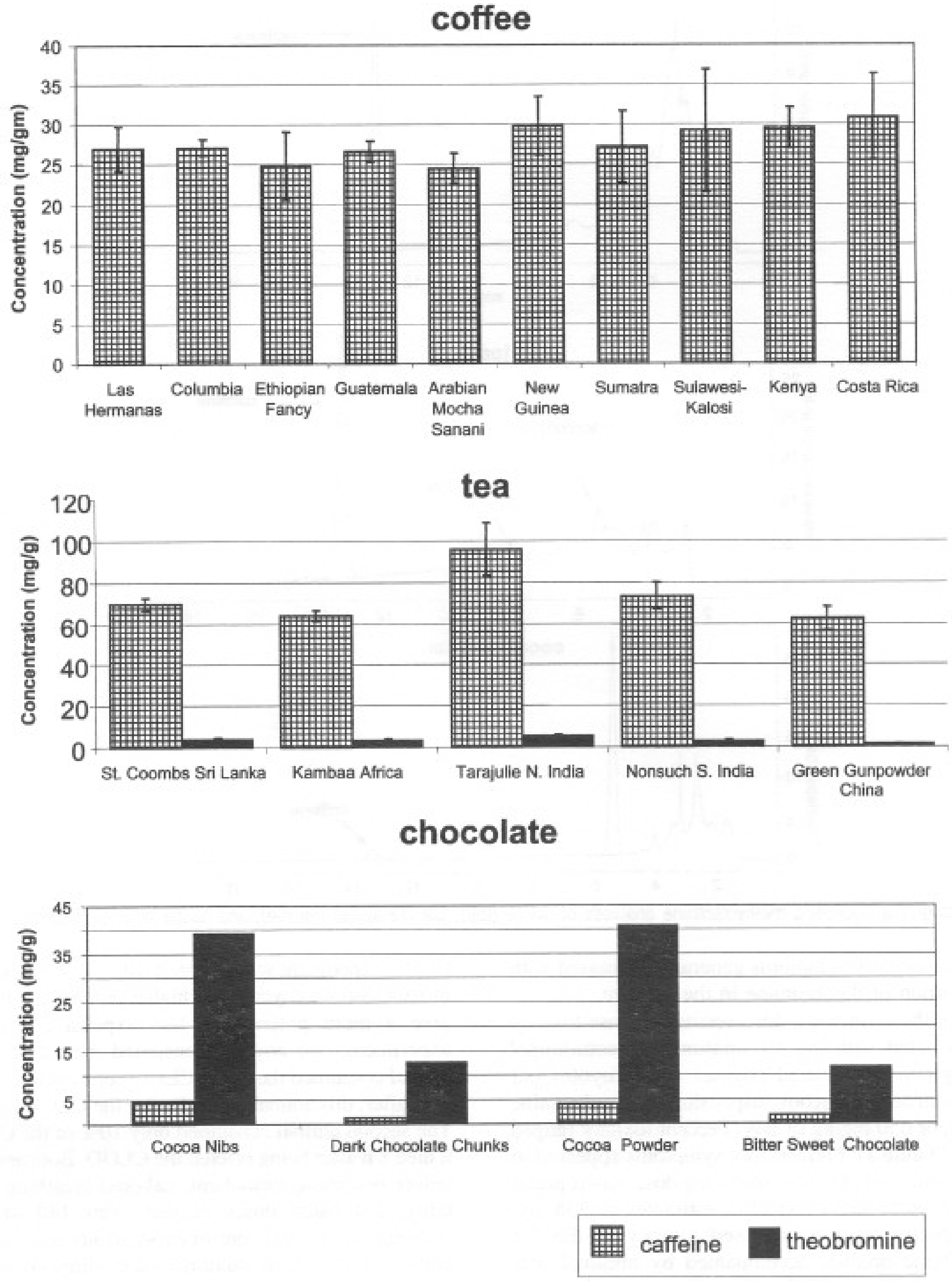

Figure 4. Theobromine and caffeine composition of various coffee (top), tea (middle), and chocolate (bottom) samples.

calcium and/or sodium benzoate to the formulation may permit the use of smaller quantities of the more expensive methylxanthines, which would decrease the cost of the required dose of the pest coyote toxicant.

In 1983, criteria for the selection and development of predator toxicants were summarized by Savarie and Connolly (*35*). These criteria included high effectiveness, acceptable taste and odor, rapid speed of action, low hazards to humans, availability of an antidote, acceptable level of environmental safety, minimal regulatory concerns, low cost, and reasonable availability. The data from this study clearly indicate that methylxanthine mixtures can effectively induce acute toxicity in coyotes and meet the criteria cited by Savarie and Connolly. Savarie and Connolly stated that substances with noxious tastes or odors are likely to be rejected by coyotes; the fact that captive coyotes ingested methylxanthine formulations from CLODs indicated that methylxanthines can be formulated in a manner that is palatable to coyotes. Given the nearly ubiquitous occurrence of caffeine and theobromine in the diets of numerous human societies, it appears that the theobromine and caffeine hazards to humans are minimal. With respect to antidotes, given the low toxicity of theobromine and caffeine to humans, it is unlikely that a human antidote for the methylxanthine pest coyote toxicant would be required. Veterinary clinics are well aware of potential methylxanthine poisoning in dogs, and supportive therapy for inadvertently dosed dogs (vomiting, oral charcoal administration, oral saline solutions) is readily available. This is not a likely scenario for cyanide- or fluoroacetate-poisoned dogs. Environmental safety with respect to effects on nontarget wildlife are likely acceptable for methylxanthines as

Table 1. Toxicity Summary for Methylxanthine-Dosed Coyotes

methylxanthine mixture	dose (mg/kg of BW)	no. exposed	% mortality	LD_{50} (mg/kg)	LD_{99} (mg/kg)
13:1 (T:C)[a]	400	4	50	424	640
	450	4	75		
	650	4	100		
	850	4	100		
1:0 (T:C)	400	4	0	516	618
	450	4	25		
	650	4	50		
	850	4	75		
5:1 (T:C)	250	4	0	336	385
	350	4	75		
	450	4	100		
	650	4	100		

[a] Theobromine/caffeine.

these compounds appear to exhibit increased toxicity to canids compared to most other species tested. Additionally, selectivity can be enhanced by the mode of application. For example, if delivered via the livestock protection collar, only animals that attack livestock would be directly exposed to the toxicant. In field studies, nontarget interest in CLODs has been minimal. Consumption by other species would be minimized because curious nontarget wildlife would need to puncture the polypropylene bottle to consume the contents. At \$0.40 per unit, the cost of the current formulation is higher than optimal. However, the addition of inexpensive synergists might significantly decrease the required amount and associated cost of the active ingredients. Regulatory requirements and associated costs for U.S. EPA registration for any predator toxicant are significant. Additionally, each of the two active ingredients in this product will likely have to be evaluated by the EPA. However, registration criteria focus on efficacy and safety. On the basis of these criteria, a methylxanthine-based pest coyote toxicant should fare well. Finally, given the ability of citizens to effectively regulate the availability of predator control measures through voter initiative options available in many states, potential social acceptability should be considered. Although there will invariably be a segment of society that is uncomfortable with any predator or pest coyote toxicant, most people's high degree of familiarity and comfort with chocolate and coffee should minimize opposition to a pest coyote toxicant based on mixtures of theobromine and caffeine.

This study demonstrated that theobromine and caffeine can be combined to deliver a potent and humane toxicant for coyotes. Such mixtures can be delivered to coyotes via the CLOD. Future research needs include the identification of the optimal quantity of theobromine and caffeine mixture to be included in a CLOD and the subsequent evaluation of this toxicant and delivery system under field conditions. Evaluation of this toxicant in the livestock protection collar could expand the types of delivery devices compatible with this product and further increase the selectivity of this coyote toxicant (*36*). However, the limited volume of the ingested livestock protection collar contents may need to be increased for use with methylxanthine coyote toxicant. This may be accomplished by formulating the collar contents to contain a material that functions as a taste attractant for predatory coyotes. The results from this research clearly demonstrate that theobromine/caffeine mixtures have potential as a pest coyote toxicant that is effective, selective, and potentially more socially acceptable than fluoroacetate or sodium cyanide.

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Credible baseline wildlife data collection

WYOMING ANIMAL DAMAGE MANAGEMENT BOARD

BIG GAME SURVEYS
at
THREE SITES IN WYOMING

Carbon County
Weston County
Rawhide Hills

Year Five - 2004

July 2005

Prepared for
Wyoming Damage Management Board
Department of Agriculture
Cheyenne, WY

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1.0 INTRODUCTION

The Animal Damage Management Board (ADMB) was created by the 1999 Wyoming State Legislature (W.S. 11-6-301-313) for the purposes of mitigating damage caused to livestock, wildlife and crops by predatory animals, predacious birds and depredating animals or for the protection of human health and safety. The departments of Agriculture and Game and Fish serve as co-chairs and give general direction to the board.

The mission of the ADMB is to coordinate and implement an integrated animal damage management program, based on best available science, for the benefit of human and natural resources throughout Wyoming.

In an effort to meet both the purpose and the mission of the ADMB, the board contracted Real West Natural Resource Consulting (Real West) in 2000 to prepare a study plan and collect baseline data for a big game monitoring project. Since then, Real West has repeated the surveys every year from 2001 through 2004. The purpose of this report is to document the field methodology and survey results for the 2004 field season and to provide an analysis on the possible affects of predator control efforts on big game reproduction in these areas.

The primary goal of the big game monitoring project is to "develop and implement a statistically valid monitoring program and data collection protocol to evaluate the effects of predator management on antelope and deer populations in three study areas" (ADMB Request for Proposals, June 2000).

2.0 METHODOLOGY

The 2004 surveys mimicked, as close as possible, the previous surveys. The routes, the timing, and the biologist conducting the surveys were as similar as possible on each of the five years of the surveys. Dates for the surveys each year are listed in Table 2-1.

The pronghorn classification surveys were conducted from a 4-wheel drive vehicle and on foot. Binoculars and a high-powered spotting scope aided in the observations. All pronghorn observations were marked on a map and classifications were recorded in a field notebook. The mule deer survey was conducted from a helicopter and all observations were recorded in a field notebook and marked using a GPS receiver.

In all years, groups of pronghorn/mule deer were classified according to does, fawns and bucks. Following the initial baseline surveys in 2000, efforts were made in subsequent years to also identify yearling bucks. The assumption is that the number of yearling does would be similar to that of the young bucks. Since yearling does do not produce young, a high number of yearling does could skew the fawn:doe ratio.

2.1 Field Surveys

2.1.1 Carbon County Project

Surveys in 2004 repeated the routes covered in previous years. Surveys were conducted on a portion of Hunt Area 63, all of Hunt Area 55 and Hunt Area 108, and the control area (a portion of Hunt Area 61) from Sept. 5 though Sept. 14. Data collection details are presented in Table 2-2.

Table 2-1. Survey dates for each of the project areas for each of the five years of the study.

Year	Survey Dates		
	Carbon County[1]	Weston County	Rawhide Hills
2000	Sept 9 - 17	Sept 21 - 25	Nov. 18
2001	Sept 9 – 16	Aug 31 – Sept 3	Nov. 18
2002	Sept 1 – 13	Aug 27 - 31	Nov. 19
2003	Sept 4 – 13	Aug 27 – 31	Dec. 7
2004	Sept 5 – 14	Sept 1 – Sept 5	Dec. 6

[1]Dates are not continuous; the surveys occurred intermittently during the dates indicated

Table 2-2. Survey Details for the Carbon County Pronghorn Surveys for 2000 - 2004.

	Number of Days for the survey	Hours of Survey	Miles Traveled During Survey
Ferris Mountain (Hunt Area 63)			
2000	Not surveyed	Not surveyed	Not surveyed
2001	2	15	208
2002	2	15	197
2003	2	17.5	220
2004	2	16	204
Hunt Area 55			
2000	2	20	196[1]
2001	2	17	173[1]
2002	2	17	180[1]
2003	2	17	198[1]
2004	2	17	202[1]
Hunt Area 108			
2000	2	15	196[1]
2001	2	15	173[1]
2002	2	14	180[1]
2003	2	15	198[1]
2004	2	14	202[1]
Control Area			
2000	2	18	164
2001	2	15	143
2002	2	16	150
2003	2	16	150
2004	2	14	163

[1] Mileage is half of the combined mileage for HA 55 and 108. Due to proximity of the two areas, surveys were intermixed and separate mileages were not recorded.

Separate mileage counts were not kept for hunt areas 55 and 108 due to their proximity to each other. Along the shared boundary, surveys were conducted back and forth between the two areas. Mileages listed are half of the combined total for the two areas. Slight reductions in mileages and hours was possible after the first year due to improved efficiencies, based on knowledge of the road system and the terrain.

In 2004 weather conditions were generally sunny and mild. Weather conditions were generally sunny and mild. Temperatures ranged from overnight lows in the mid 30s to low 40s to daytime highs ranging from the mid 60s up to nearly 80 degrees. Winds were generally light in the

Table 2-3. Survey details for the Weston County Pronghorn Surveys for 2000 though 2004.

	Number of Days for the survey	Hours of Survey	Miles Traveled During Survey
Treatment Area			
2000	4	34	408
2001	4	29	374
2002	4	27	359
2003	4	28	370
2004	4	26	360
Control Area			
2000	2	18	132
2001	2	15	165
2002	2	14	132
2003	2	14	157
2004	2	14	162

mornings and turning breezy, with winds reaching 10 to 15 mph, during the day. Cloud cover ranged from 0% to 70%. There was no precipitation over the period and roads were dry during the entire survey period.

2.1.2 Weston County Project

Weston County surveys were conducted from Sept 1 through Sept 5, 2004 and they followed the same routes and timing as pervious surveys (Table 2-3). The weather was mild and pleasant. Temperatures ranged from overnight lows in the upper 50s to low 60s, and reaching daytime highs in the mid to upper 80s. Winds were calm to light. A rainstorm, accompanied by thunder and lightning, took place on Sept 2. Lightning ignited sagebrush and grassland on the control area; however rainfall quickly doused the fire and the burned area covered less than 0.25-acre.

2.1.3 Rawhide Hills Project

The mule deer survey was conducted from the air using a Jet Ranger helicopter, contracted through Bighorn Aviation out of Sheridan, Wyoming. Real West coordinated helicopter use with Wyoming Game and Fish Department (WGFD) biologists who were also conducting mule deer surveys during the same time period and in the same region. During the 2004 surveys, WGFD biologist Martin Hicks assisted with the survey as an observer in the helicopter; he also obtained counts for several elk herds that were also observed during the survey.

The Rawhide site was surveyed on December 6, one day earlier than the surveys in 2003, but several weeks later than those conducted in 2000, 2001, and 2002 (Table 2-4). The delay was due to the availability of the helicopter since it was the only one in use for both WGFD and other agency surveys. The flight began at 7:20 a.m. from the Wheatland airport. The survey lasted 3.8 hours, with 0.6 hour for shuttle time from Wheatland to the project area. The pilot followed a similar flight pattern as pervious year, covering the central hilly area first due to concerns that increasing high winds would limit access later in the day. Winds were moderate (10 to 15 mph) at the start of the survey but they increased to 25 mph through the morning. Efforts were made to fly up drainages in the rough terrain around the Rawhide hills while maintaining a grid pattern to ensure that the survey covered the entire area.

All mule deer and coyote observations were recorded in a field notebook. Mule deer age and gender classifications were recorded. The biologist entered each observation as a waypoint in a GPS receiver. Following the flight, the UTM (Universal Transverse Mercator) coordinates were retrieved from the GPS receiver for each observation point and recorded in the field notebook. These points were later identified on a 1:250,000 scale U.S. Geological Survey map.

Table 2-4. Survey details for the Rawhide Hills Mule Deer Surveys for 2000 though 2004.

	2000	2001	2002	2003	2004
Date of Flight	Nov. 18	Nov. 18	Nov. 19	Dec. 7	Dec. 6
Start Time	8 a.m.	10 a.m.	7 a.m.	7:30 a.m.	7:30 a.m.
Flight Duration (hrs)	3.3	3.3	4.3	3.6	3.5

3.0 RESULTS

3.1 Carbon County Project

3.1.1 Project Location

The treatment area, as described in the ADMB project proposal application, includes pronghorn hunt areas 55 and 108. This area covers approximately 570 square miles immediately southwest of Rawlins (Figure 3-2). It is bound on the north by Interstate 80; on the west by Highway 789; on the south by Muddy Creek; and on the east by County Road 401. Concentrated predator control efforts took place in HA 55 between January and April 2001 through 2004. A similar concentration of effort did not occur in HA 108. Some predator control took place but it was primarily accomplished through a private contractor rather than through Wildlife Services. Due to the difference in predator control intensity, the data is separated for the two areas.

The treatment area also includes a portion of HA 63, immediately north of Ferris Mountain (Figure 3-1). This area is bound on the north by Wyoming Highway 220, on the west by U.S. Highway 287, and on the east by Alcova and Pathfinder reservoirs. Data collection in this treatment area began in 2001. Concentrated predator control measures took place January through April 2001 though 2004.

The project control area is located approximately 15 miles north of Rawlins on the edge of the Great Divide Basin of the Red Desert (Figure 3-1). The area covers approximately 244 sq. mi. and is bound on the south by the Mineral X road (Carbon County Road 63), on the west by the Crooks Gap Road (County Road 23N), on the north by County Road 22, and on the east by Bull Springs Rim.

3.1.2 Survey Results

Pronghorn classification results for each of the five years of the project and each of the hunt areas is presented in Table 3-1. Figures 3-3, 3-4, and 3-5 illustrate classification and population number changes each of the five years in all four project areas. Individual analysis of each area is provided below.

The adjusted fawn:doe ratio takes into account the number of yearling does that are too young to reproduce. This ratio assumes the number of yearling does is equal to the number of yearling bucks in the population. That number is subtracted from the total number of does and ratio recalculated.

<u>Hunt Area 63 – Ferris Mountain</u>

The greatest increase in fawn:doe ratios for the Carbon County areas was observed in HA 63, north of Ferris Mountain, where the ratio went from 46 to 70 fawns per 100 does (Table 3-1). While this rate was considerably higher than ratios in 2002 and 2003, it was slightly below that observed in 2001 when it was 79:100.

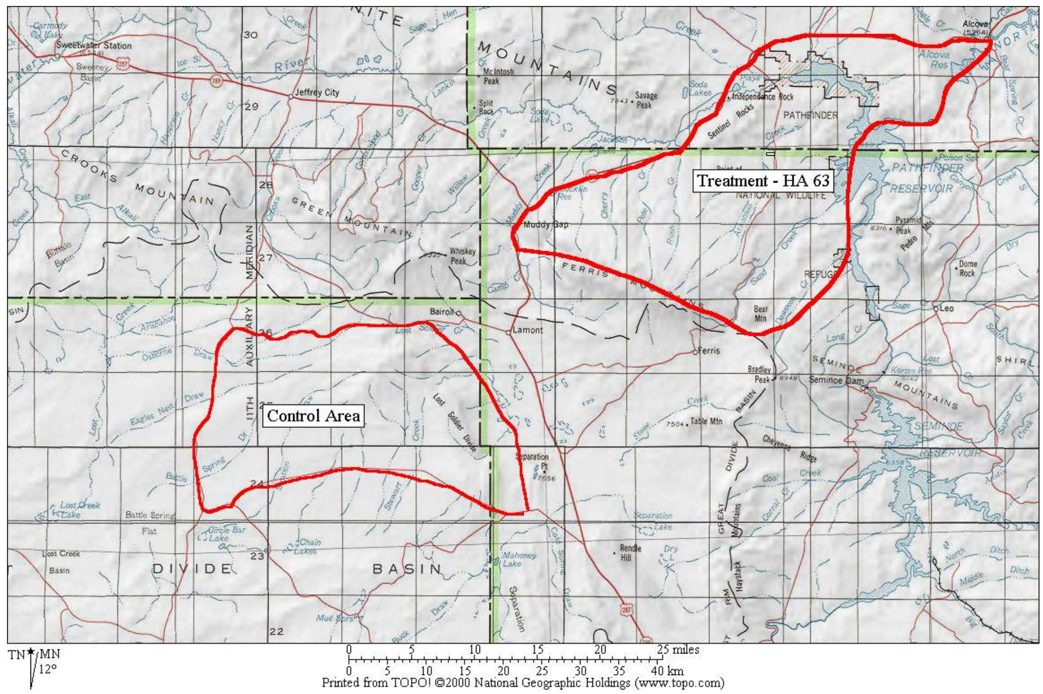


Figure 3-1. Carbon County Project Area showing the Control Area and the Treatment Area north of Ferris Mountain in Hunt Area 63.

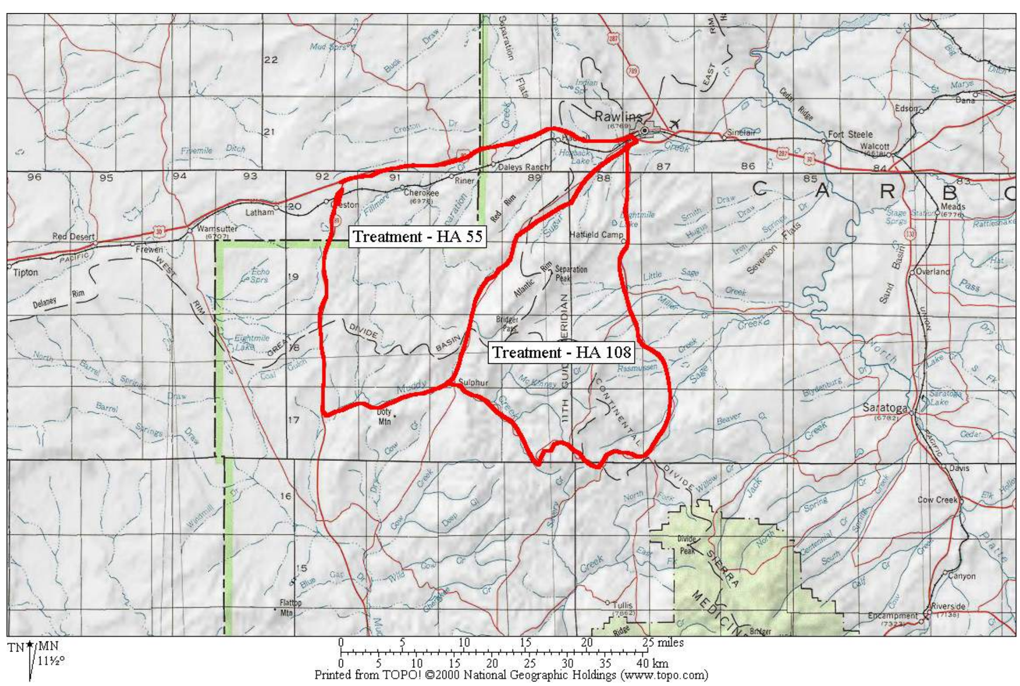


Figure 3-2. Carbon County Project Area showing the Treatment Areas south of Rawlins in Hunt Area 55 and Hunt Area 108.

Table 3-1. Results of Pronghorn Classification Surveys in for the Carbon County Project.

	No. Groups	Total No. Pronghorn	Does	Fawns	Mature Bucks	Yearling Bucks[1]	Fawn:doe ratio	Buck:doe ratio[2]	Adjusted fawn:doe ratio[3]
Treatment Areas									
HA 55									
2000	47	269	147	60	62	--	41:100	42:100	--
2001	63	331	173	90	60	8	52:100	39:100	58:100
2002	91	534	295	136	75	28	46:100	35:100	51:100
2003	88	675	389	175	88	23	45:100	29:100	39:100
2004	76	649	367	200	59	23	54:100	22:100	48:100
HA 108									
2000	52	350	184	101	65	--	55:100	35:100	--
2001	52	454	231	169	42	12	73:100	12:100	68:100
2002	91	540	310	139	77	14	44:100	29:100	47:100
2003	70	719	404	223	67	25	55:100	23:100	49:100
2004	50	373	197	92	46	38	47:100	43:100	27:100
HA 63									
2000	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
2001	37	136	53	42	31	10	79:100	77:100	60:100
2002	60	381	212	93	60	16	43:100	36:100	47:100
2003	83	876	480	223	79	94	46:100	36:100	27:100
2004	114	1152	552	391	120	89	70:100	38:100	58:100
Control Area									
2000	35	130	65	31	34	--	48:100	52:100	--
2001	45	274	147	75	25	14	51:100	36:100	56:100
2002	55	303	154	58	70	21	46:100	35:100	44:100
2003	61	353	182	94	62	15	52:100	48:100	56:100
2004	47	217	94	68	41	14	72:100	59:100	85:100

[1]Data in 2000 did not separate yearling bucks from mature bucks.
[2]Number totals all bucks, including yearlings.
[3]Adjusted ratio excludes yearling does (based on the yearling buck count).

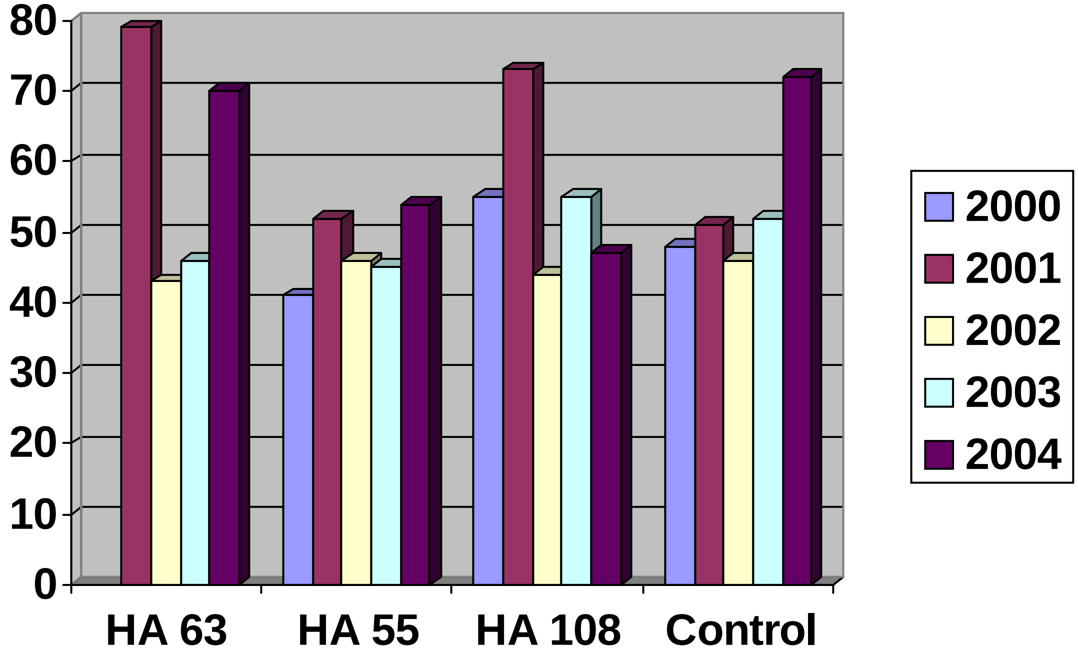


Figure 3-3. Ratios of Fawns per 100 Does on the Carbon County Project Area.

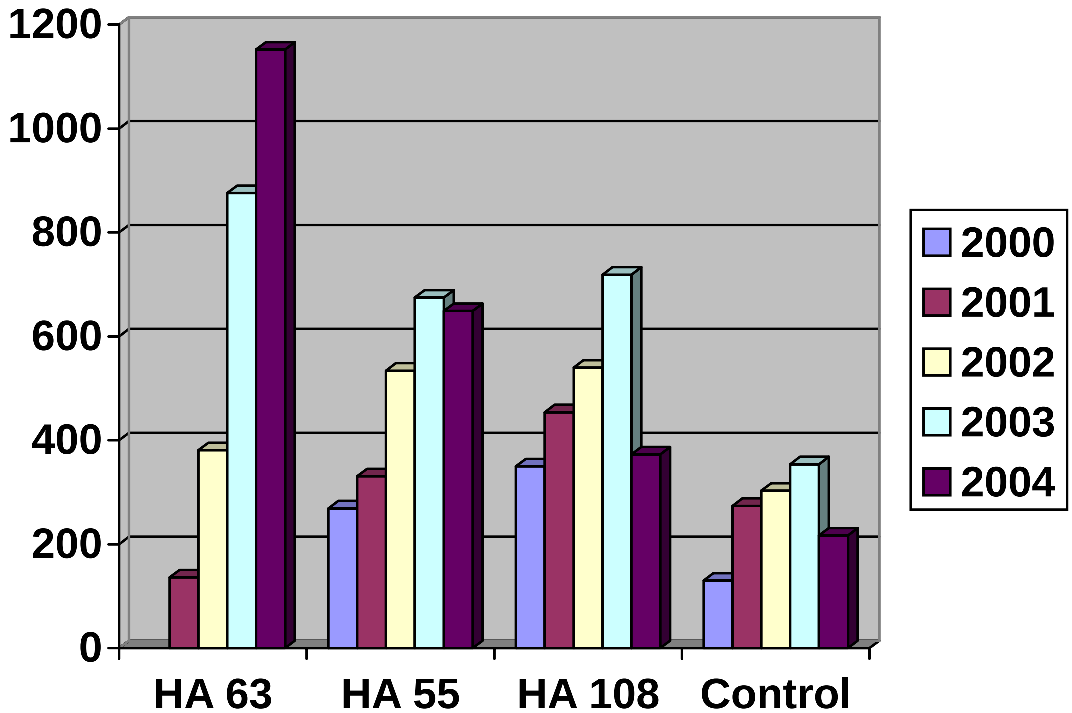


Figure 3-4. Total Pronghorn Observed on the Carbon County Project Area.

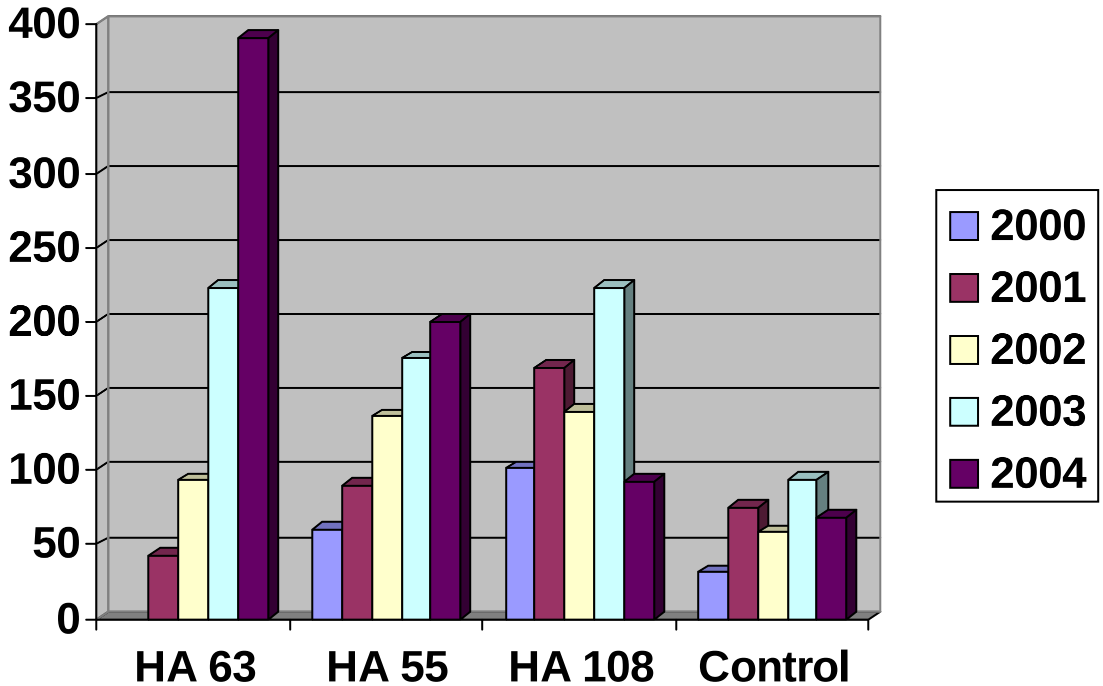


Figure 3-5. Total Number of Fawns Observed on the Carbon County Project Area.

While the ratio was still lightly below 2001 levels, the number of pronghorn observed during the surveys has continued to increase every year. In 2004, 1,152 pronghorn and 114 herd groups were observed. This compares to only 136 pronghorn and 37 groups observed in 2001.

The low observation number in 2001 was at least partially attributable to unfamiliarity of the area and the reduced survey time in the vicinity of Pathfinder Reservoir. It is possible animals in that area were undercounted in 2001. However, that was not the case in subsequent years, at least in the vicinity of the reservoir. That area is especially rich in pronghorn and is also part of Pathfinder National Wildlife Refuge. From 2002 through 2004, this area was carefully surveyed due to the high number of pronghorn. The high densities made surveying tricky due to the difficulty in distinguishing individual groups and not accidentally double-counting some animals. Efforts were made every year to avoid counting pronghorn more than once, especially in this area.

Hunt Area 55

The fawn:doe ratio increased in 2004 from all previous levels to 54 fawns per 100 does. Pronghorn numbers decreased slightly in 2004 by 26 animals and the number of groups observed declined from 88 to 76. These numbers are still considerably higher than those reported in 2000 and 2001.

<u>Hunt Area 108</u>

The fawn:doe ratio showed a decline in 2004 from 2003, although it was still higher than the ratio reported in 2002. At the same time, pronghorn numbers in 2004 were considerably lower than 2003, however they were still slightly higher than the number observed in the baseline year of 2000.

<u>Control Area</u>

The fawn:doe ratio increased substantially in 2004 to 72 fawns per 100 does, compared to 52 fawns per 100 does in 2003. The ratio in 2004 was the highest reported for the five years of the surveys. At the same time, the overall number of pronghorn and groups observed was down in 2004 to 217 animals, compared 353 observed in 2003. The 2004 numbers observed were higher than the first year of the surveys, but were the lowest of all the subsequent four years.

3.2 Weston County Project

3.2.1 Project Location

The project treatment area covers approximately 330 square miles in northeast Wyoming, west of Newcastle (Figure 3-6). The triangular shaped area is bound on the southeast by Highway 450 beginning at Newcastle and the Mush Creek Road. The boundary continues along Mush Creek Road to Highway 116, north on Highway 116 to the Raven County Road; north to the intersection with Highway 116, and then north to Upton; and east along Highway 16 to Newcastle.

The control area covers approximately 236 square miles immediately southwest of the treatment area. The area is bound on the north by the Thunder Basin National Grassland boundary, on the east by Wyoming Highway 116, Wyoming Highway 450 and County Road 7C (Bruce Road); on the south by the Cheyenne River Road (County Road 54); and on the west by County Road 7A, Wyoming Highway 450 and the Keeline Road (Forest Road 930).

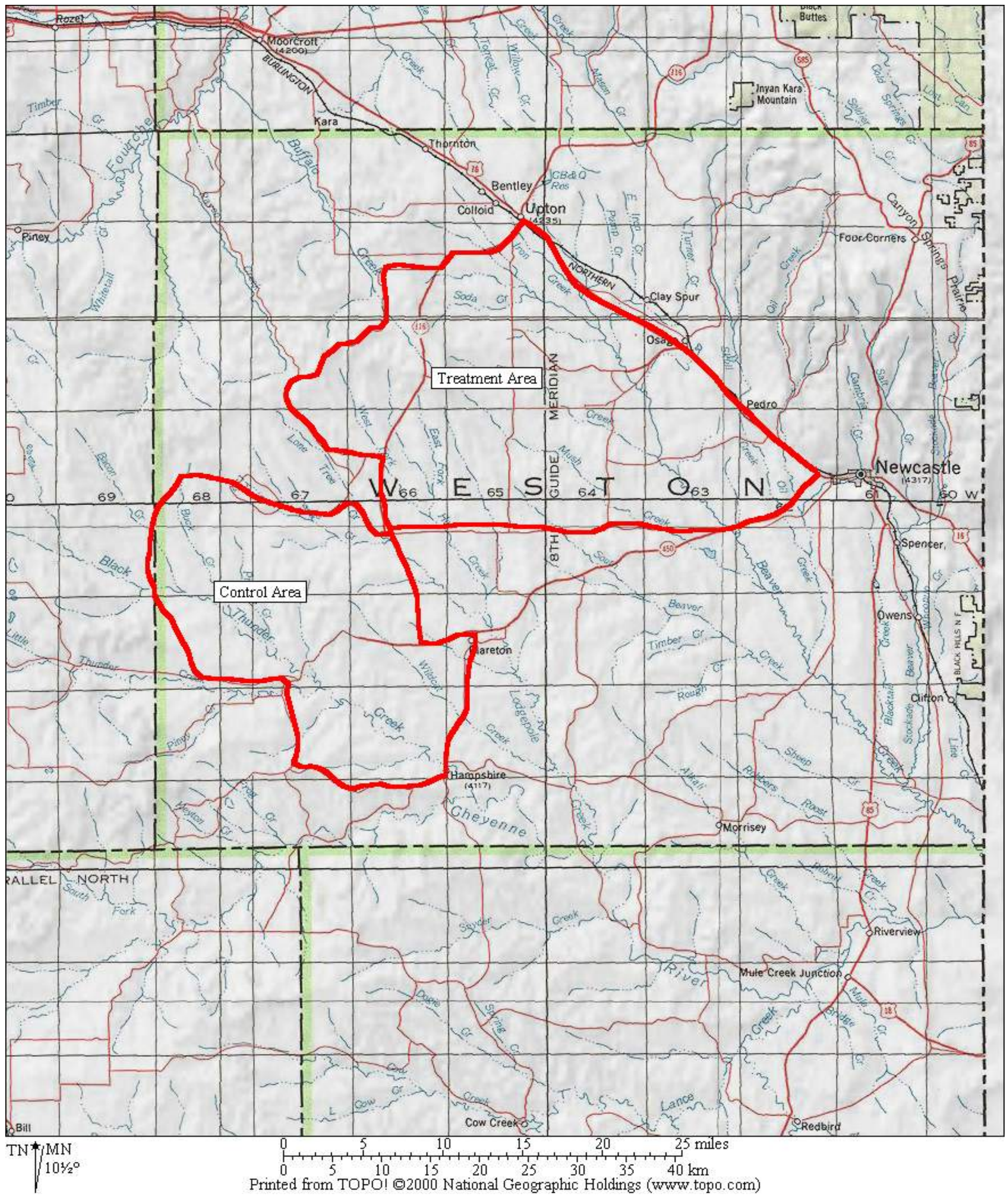


Figure 3-6. Weston County Project Area

3.2.2 Survey Results

The results of the 2004 surveys, compared with previous years are presented in Table 3-2. Comparisons of the five survey years and the fawn:doe ratios, number of pronghorn observed, and number of fawns observed are illustrated in Figures 3-7, 3-8, and 3-9, respectively.

The results indicate the total number of pronghorn observed on the treatment area has increased steadily, reaching a high of 948 pronghorn in 2004, up from a low in 2001 of 426 animals observed. By comparison, the number observed in the control area was fairly steady the first three years, and then it decreased in 2003. That trend reversed in 2004, with 238 animals observed, compared to just 71 the prior year.

In 2004, the number of fawns per 100 does was up considerably in the treatment area, reaching a five-year high of 127 fawns per 100 does. Even when the ratio is adjusted for yearling does, the rate is at a five-year high of 98 fawns per 100 does. By comparison, from 2003 to 2004 the ratio dropped 22 points in the control area. The decrease is even more significant when the ratio is adjusted for yearling does.

Table 3-2. Results of Pronghorn Classification Surveys in for the Weston County Project.

	No. Groups	Total Number	Does	Fawns	Mature Bucks	Yearling Bucks[1]	Fawn:doe ratio	Buck:doe ratio[2]	Adjusted fawn:doe ratio[3]
Treatment Areas									
2000	77	544	256	193	95	--	75:100	37:100	--
2001	64	426	191	125	77	33	65:100	40:100	79:100
2002	101	711	356	239	86	30	67:100	33:100	73:100
2003	113	916	419	327	100	70	78:100	41:100	61:100
2004	101	948	325	414	115	94	127:100	64:100	98:100
Control Area									
2000	27	153	66	56	31	--	85:100	47:100	--
2001	27	143	69	46	25	3	67:100	41:100	70:100
2002	49	128	59	30	29	10	51:100	66:100	61:100
2003	17	71	27	25	14	5	93:100	70:100	74:100
2004	36	238	103	73	26	36	71:100	60:100	36:100

[1]Data in 2000 did not separate yearling bucks from mature bucks.
[2]Number totals all bucks, including yearlings.
[3]Adjusted ratio excludes yearling does (based on the yearling buck count).

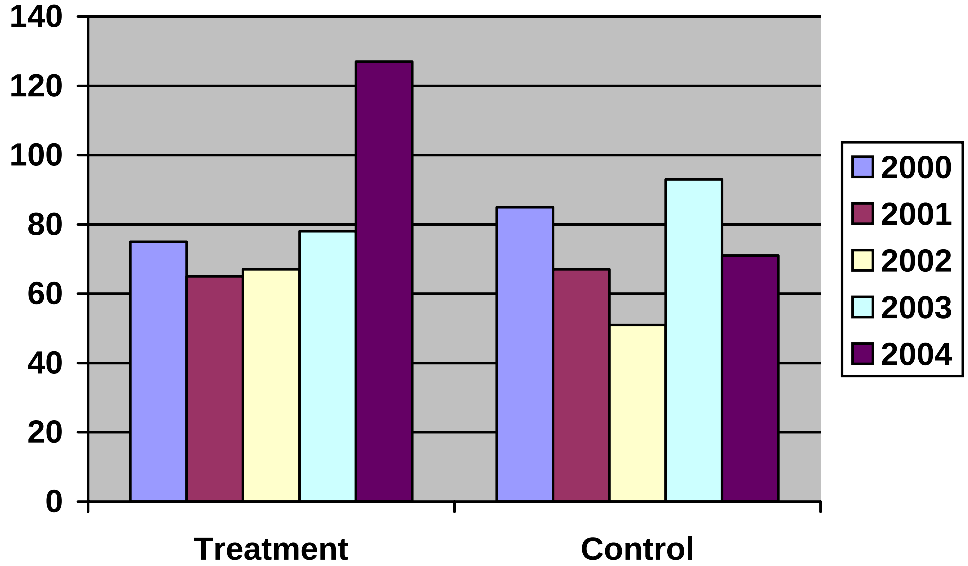


Figure 3-7. Ratios of Fawns per 100 Does on the Weston County Project Area.

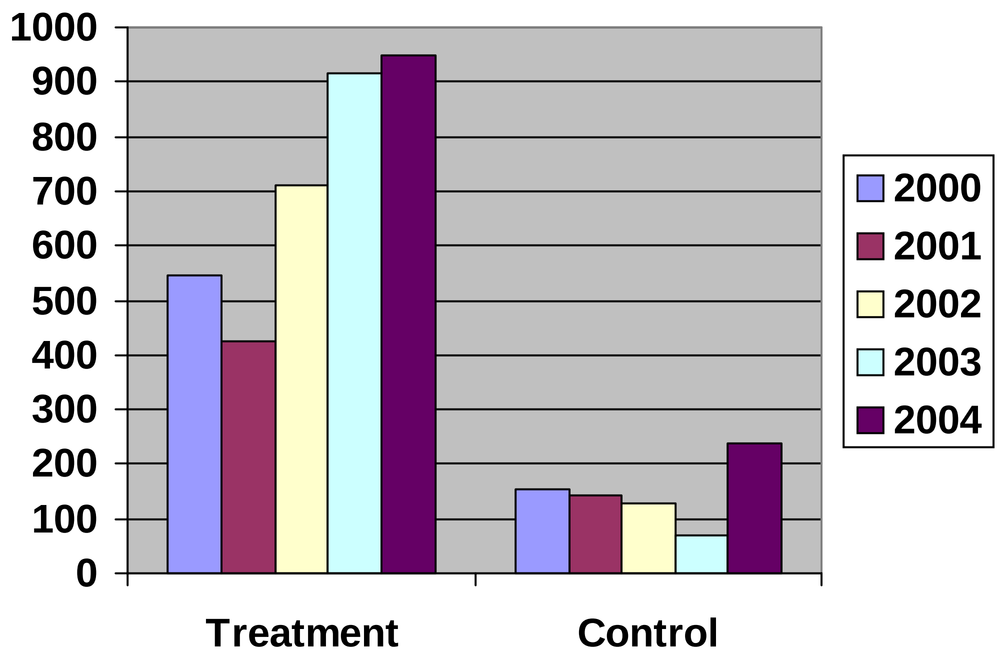


Figure 3-8. Total Pronghorn Observed on the Weston County Project Area.

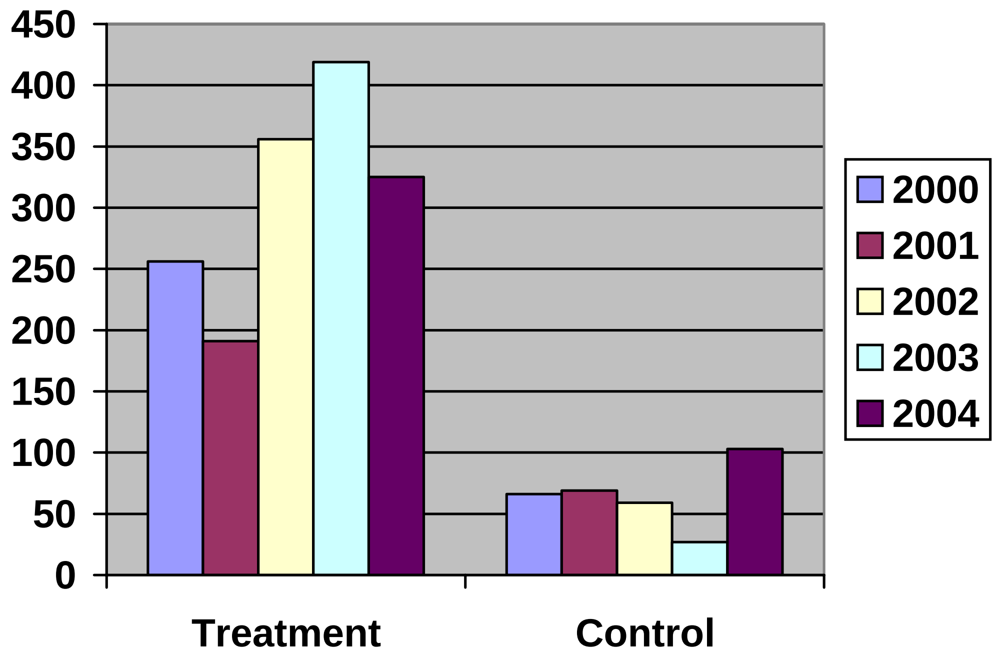


Figure 3-9. Total Number of Fawns Observed on the Weston County Project Area.

3.3 Rawhide Hills Project

3.3.1 Project Location

The project area is located in east-central Wyoming, southwest of Lusk and northeast of Guernsey (Figure 3-10). The treatment area covers approximately 390 square miles and is bound on the north by U.S. Highway 18-20, on the west by Highway 270 and Jireh Road, on the east by U.S. Highway 85, and on the south by a combination of county roads and transmission lines.

The control area is a triangular area covering approximately 160 square miles. It is bound on the east by U.S. Highway 85, on the south by a powerline, and on the northwest by a county road.

3.3.2 Survey Results

The mule deer classification survey was conducted on November 19, 2002. Weather conditions were clear with strong winds and no snow cover. Observation conditions were similar to those of 2001 but were drastically different from those in 2000. In 2000, snow cover was 100 percent except for a few wind-blown ridges. The flight time for 2002 was approximately 4.3 hours, an hour longer than the 2001 and 2000 surveys.

Figure 3-10. Rawhide Hills Project Area.

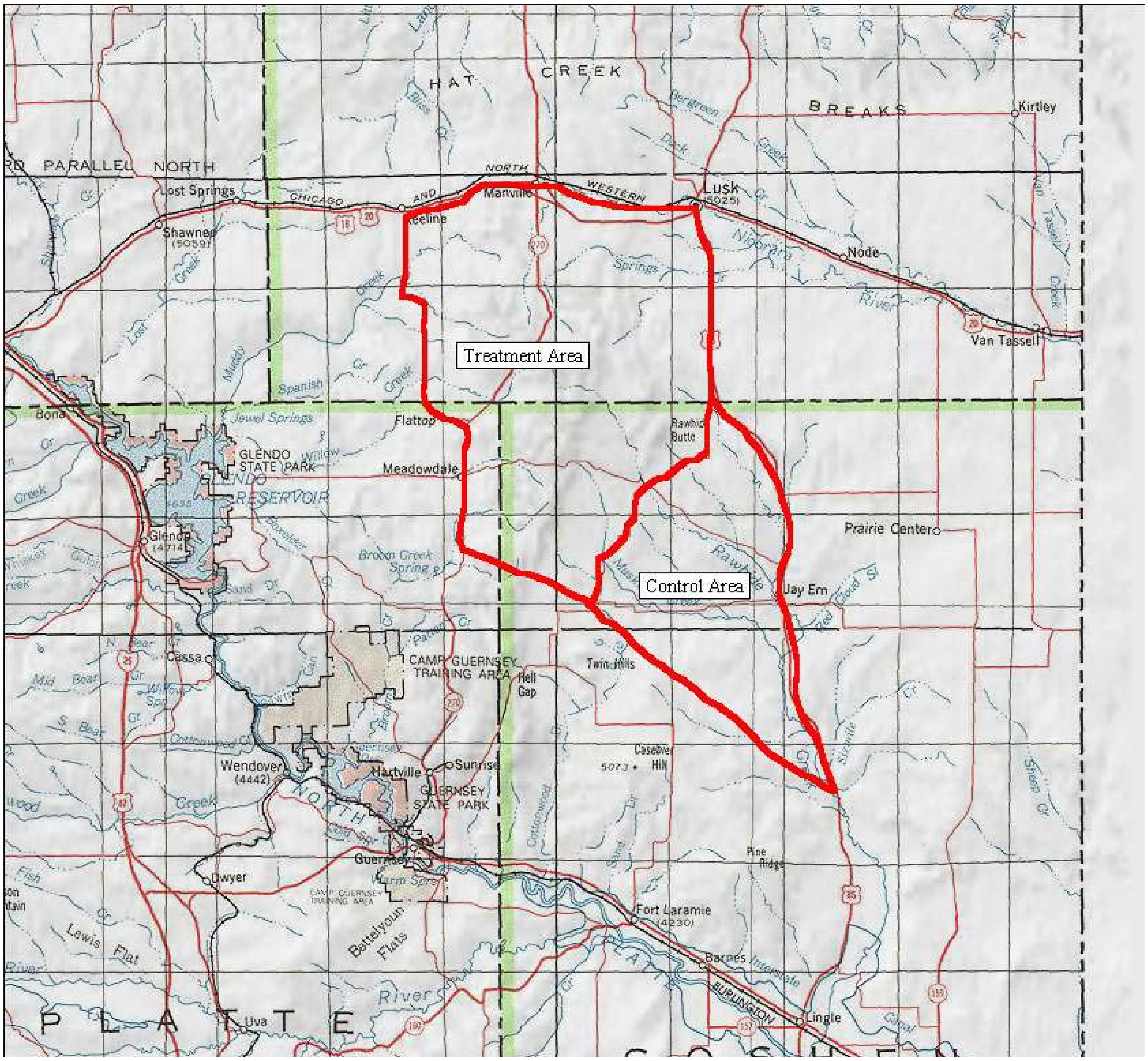


Table 3-3. Results of the Mule Deer Classification Surveys for the Rawhide Hills Project from 2000 through 2004

	No. Groups	Total Number	Does	Fawns	Mature Bucks	Yearling Bucks[1]	Fawn:doe ratio	Buck:doe ratio[2]	Adjusted fawn:doe ratio[3]
Treatment Areas									
2000	15	107	58	28	21	--	48:100	36:100	--
2001	17	58	28	15	12	4	53:100	23:100	39:100
2002	50	300	163	100	25	12	61:100	11:100	79:100
2003	4	31	19	10	1	1	52:100	11:100	47:100
2004	22	110	56	37	12	5	66:100	30:100	57:100
Control Area									
2000	5	37	21	11	5	--	52:100	24:100	--
2001	7	29	16	9	3	1	56:100	25:100	50:100
2002	7	54	31	17	2	4	54:100	12:100	42:100
2003	1	10	3	4	3	0	133:100	100:100	133:100
2004	7	82	40	19	15	8	48:100	57:100	28:100

[1]Data in 2000 did not separate yearling bucks from mature bucks.
[2]Number totals all bucks, including yearlings.
[3]Adjusted ratio excludes yearling does (based on the yearling buck count).

A summary of the data for the five years is presented in Table 3-3 and data comparisons with previous years are illustrated in Figures 3-11, 3-12, and 3-13.

From 2000 to 2002, the fawn:doe ratio steadily improved on the treatment area until declining in 2003, but rebounding in 2004 (Figure 3-11). In the control area, it remained fairly steady the first three years, then increased sharply in 2003 and then it decreased in 2004. The ratio for 2003 should be viewed with caution, however, since it was the ratio found in the only group of deer observed on the control area.

Total numbers of pronghorn observed (Figure 3-12) on the treatment varied considerably from year to year, although the sample size and number observed was consistently low, except in 2002. That year, the number of pronghorn observed was at least three times the number seen in each of the other years.

Figure 3-11. Ratio of Fawns per 100 Does on the Rawhide Hills Project Area

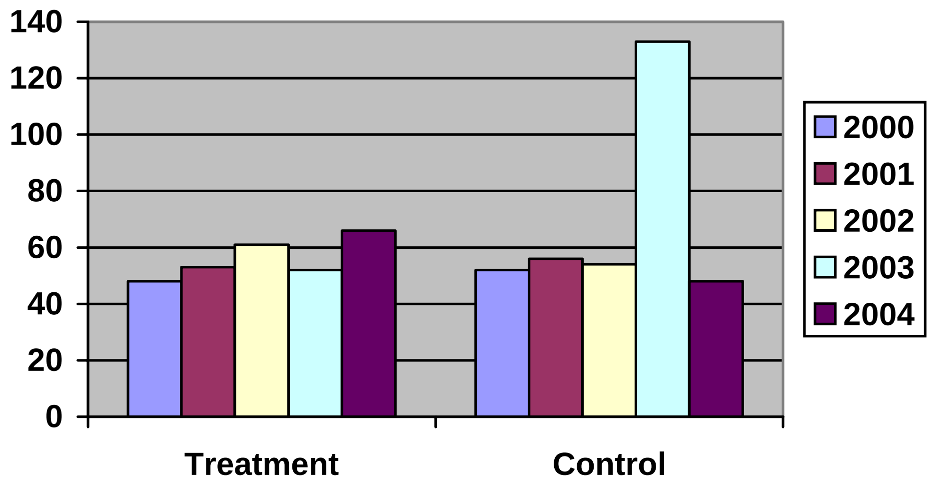
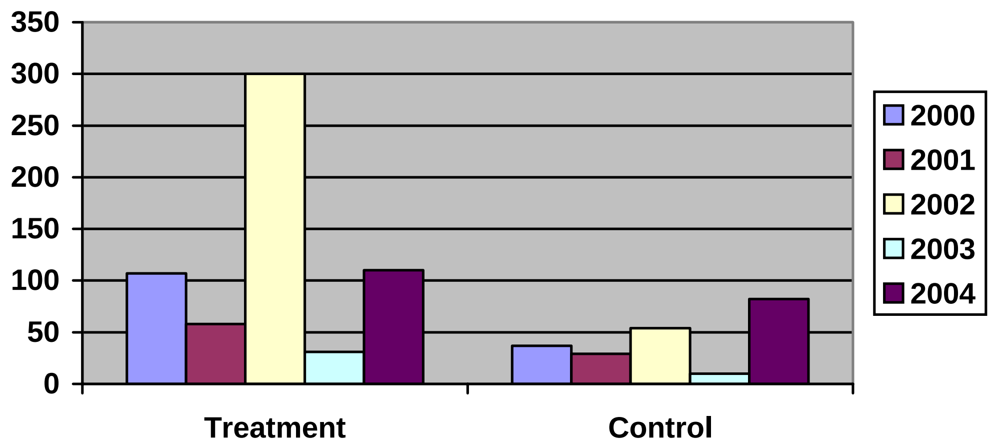


Figure 3-12. Number of Pronghorn Observed on the Rawhide Hills Project Area



4.0 DISCUSSION

This section will consider historical big game data collected by Wyoming Game and Fish Department (WGFD) biologists and compare it with data collected by Real West. Comparisons will also be made between the various Hunt Areas within the Herd Units that include the project areas.

4.1 Historical Records

4.1.1 Carbon County Project

A listing of the Hunt Areas and corresponding WGFD Herd Units is listed in Table 4-1. Hunt Area 55 is included in the Baggs Herd Unit, which also includes part of Hunt Area 53. Hunt Area 108 is in the Iron Springs Herd Unit, which also includes Hunt Area 56 and the rest of Hunt Area 53. The control area is in the northern portion of Hunt Area 61, which is part of the Red Desert Herd Unit along with hunt areas 60 and 64. Hunt Area 63 is the only area included in the North Ferris Herd Unit but the Real West survey area is only the area north of the Ferris Mountains.

Comparisons of fawn:doe ratios and population changes between the Real West and WGFD data are presented in Table 4-2. In this data, the Real West observation numbers are the actual pronghorn observed each year. The WGFD population estimates are based on population sampling and statistical modeling.

<u>Hunt Area 63</u>
Hunt Area 63, the North Ferris Antelope Herd Unit, has an objective of 5,000 animals. In 2001 it was well below that, at 2,600 animals. After three years it had increased by 5% and by 44%

Table 4-1. Hunt Areas and corresponding Herd Units for the Carbon County Project.[1]

Project Area Hunt Area	Herd Unit	Other Hunt Areas within same Herd Unit
Hunt Area 55	Baggs	Part of 53
Hunt Area 108	Iron Springs	Part of 53, 56
Hunt Area 63	North Ferris	No others
Hunt Area 61	Red Desert	60, 64

[1] The hunt areas are always a sub-area within the herd unit.

Table 4-2. Historical Fawn:Doe Ratios and Population Estimates for the Carbon County Pronghorn Project.

		2004	2003	2002	2001	2000
HA55 Real West	Fawn:doe ratio	54	45	46	52	41
	Observation numbers	649	675	534	331	269
Baggs HU	Fawn:doe ratio	55	43	53	33	30
	Population Estimate	9,300	8,700	7,600	6,800	7,600
HA 108 Real West	Fawn:doe ratio	47	55	44	73	55
	Observation numbers	373	719	540	454	350
Iron Springs	Fawn:doe ratio	65	55	48	37	53
	Population Estimate	10,000	9,500	10,100	9,450	10,450
HA 63 Real West	Fawn:doe ratio	70	46	43	79	--
	Observation numbers	1,152	876	381	136	--
North Ferris HU	Fawn:doe ratio	71	68	71	73	52
	Population Estimate	3,925	2,850	2,900	2,600	2,725
HA 61 Real West	Fawn:doe ratio	72	52	46	51	48
	Observation numbers	217	353	303	274	130
Red Desert HU	Fawn:doe ratio	72	54	53	41	55
	Population Estimate	14,650	13,400	14,000	13,350	17,150

after four years. The population estimate for 2004 was 3,925. The fawn:doe ratios for the entire herd unit have increased over the five year period from 52 to 71. The ratios for the sub-area surveyed by Real West decreased in 2002 and 2003 to 43 and 46, respectively, to increasing to 70 fawns per 100 does in 2004. This ratio was still below that observed at the start of the project in 2001.

<u>Hunt Area 55</u>

The population objective for the Baggs Herd Unit, which includes Hunt Area 55, is 9,000 animals. The unit is slightly above that at 9,300 in 2004 and has shown an increase each of the past five years, except for 2001. Real West's data for the smaller hunt area shows a steady increase the first three years, with a slight decrease in observed numbers in 2004, but still well above the number of pronghorn seen in 2000.

The fawn:doe ratio for the Baggs herd unit has increased from 30 to 55 over the five year period, with increases each year except in 2003. Real West has also found a steady increase, although not as great as that of the overall herd unit. Both the herd unit and the Real West data show a similar ratio in 2004 of 55 and 54 fawns per 100 does, respectively.

Hunt Area 108

The population objective for the Iron Springs Herd Unit is 12,000. The 2004 WGFD population estimate is below this at 10,000. This estimate has remained fairly steady over the past five years, with population estimates ranging from a low of 9,450 in 2001 and a high of 10,450 in 2000.

Observation numbers in 2004, at 350 animals, are similar to those reported by Real West in 2000 with 373 animals. During the intervening three years, numbers have ranged from a low of 454 in 2001 to a high of 719 in 2003.

Fawn:doe ratios for the herd unit have fluctuated annually, with a high of 65 in 2004 and a low of 37 in Iron Springs. Ratios observed by Real West have similarly fluctuated but with the highest level in 2001 at 73 and the lowest in 2002 at 44.

Hunt Area 61

The population objective for the Red Desert Herd Unit, which includes the control area, is 15,000. The 2004 population estimate is near this, at 14,650. The estimate was at a high of 17,150 animals in 2000 followed by a decline to 13,350 in 2001. Real West observation numbers show a gradual increase all years, with a decline in 2004.

The fawn:doe ratio for the entire herd unit has ranged from a low of 41 in 2001 to a high of 72 in 2004. The Real West data had a low level of 46 in 2002 and also had a peak in 2004 of 72.

4.1.2 Weston County Project

The treatment area is contained within pronghorn Hunt Area 7, although the hunt area is larger than the treatment area. The northeast corner of the hunt area is outside the treatment area while the southwest corner is included in the control area. Hunt Area 7 is included in the Cheyenne River Antelope Herd Unit, which also includes hunt areas 4, 5, 6, 8, 9, 27, and 29.

The remainder of the control area is contained in Hunt Area 6, which is also part of the Cheyenne River Herd Unit. Approximately two-thirds of the hunt area is outside the control area.

The WGFD population objective for the entire Herd Unit is 38,000 pronghorn. The 2004 population estimate was right at this level with 38,059 pronghorn.

Population trend counts and fawn:doe ratios for the Herd Unit, as compared to the Real West data, are shown in Table 4-3. The population trend of the entire herd unit has increased steadily

Table 4-3. Historical Fawn:Doe Ratios and Population Estimates for the Weston County Pronghorn Project.

		2004	2003	2002	2001	2000
Real West Treatment Area (HA 7)	Fawn:doe ratio	127	78	67	65	75
	Observation numbers	948	916	711	426	544
WGFD HA 7	Fawn:doe ratio	86	97	77	71	81
	Observation numbers	809				
Real West Control Area (Partial HA 6)	Fawn:doe ratio	71	93	51	67	85
	Observation numbers	238	71	128	143	153
WGFD HA 6	Fawn:doe ratio	73	79	60	70	62
	Observation numbers	526				
Cheyenne River Herd Unit (WGFD)	Fawn:doe ratio	85	84	74	67	74
	Population Model Est.	28,406	27,121	25,920	25,143	23,571

over the five-year period. Likewise, the Real West observation numbers in the treatment area have increased steadily. Observation numbers in the control area have fluctuated, however, with a low of only 71 animals observed in 2003 and a high of 238 in 2004.

Fawn:doe ratios for the treatment area fluctuated from 2000 through 2003, but showed a significant increase in 2004. The treatment area also demonstrated fluctuations but no similar increase was observed in 2004. The WGFD data for the entire herd unit show more of a gradual increase in ratios over the five-year period, except for a slight drop in 2001. The herd unit ratio in 2004 was similar to that observed in 2003.

4.1.3 Rawhide Hills Project

The treatment area is included in mule deer Hunt Areas 15 and 16. Both are part of the Goshen Rim Herd Unit, which also includes hunt areas 55 and 57. The control area is contained in Hunt Area 16.

The population objective for the Goshen Rim Herd Unit is 25,000 mule deer. The 2004 population estimate is 6% below this at 23,399 animals. This is slightly above the five-year average of 21,295 mule deer. Over the last five years, the estimated number of mule deer was highest in 2000 and nearly the same in 2004, with lower levels for the three intervening years (Figure 4-4).

Table 4-4. Fawn:Doe Ratios (fawns per 100 does) and observation numbers for the Rawhide Hills Mule Deer Project Area over the past five years.

		Hunt Area 15[2]	Hunt Area 16[3]	Entire Herd Unit[4]	Treatment (HA 15, 16)	Control (HA 16)
2000	Fawns per 100 does	50	63	68	48	52
	Pop Est/No. counted			23,420	107	37
2001	Fawns per 100 does	35	22	49	53	56
	Pop Est/No. counted			19,056	58	29
2002	Fawns per 100 does	57	54	55	61	54
	Pop Est/No. counted			19,956	300	54
2003	Fawns per 100 does	81	66	71	52	133
	Pop Est/No. counted			20,957	31	10
2004	Fawns per 100 does	38	58	56	66	48
	Pop Est/No. counted			23,399	110	82

[1] Data from Wyoming Game and Fish Department Annual Reports and personal communication with WGFD biologists Bob Lanka, Bart Kroger, and Martin Hicks (WGFD 2004, 2003, 2001, 2000, 1999, 1998, 1997, 1996, 1995).
[2] Approximately one-third of Hunt Area 15 is in the treatment area.
[3] Portions of Hunt Area 16 are in the treatment area and in the control area.
[4] Includes Hunt Areas 15, 16, 55 and 57.

Real West has consistently observed low numbers of mule deer in this area. The highest number observed on the treatment area was in 2002 with 300 mule deer being seen during the helicopter survey. That number dropped considerably in 2003 to only 31 animals, the lowest over the five-year study period. The number of deer observed rebounded somewhat in 2004 with 110 animals being observed. Observation numbers in the control area are consistently even lower. The highest number of observations was the 82 observed in 2004; the lowest count was only 10 animals in 2003.

WGFD fawn:doe ratios for Hunt Area 15 fluctuated over the five year period, with a high of 81 in 2003 and a low of 35 in 2001. These high and low years were similar in Hunt Area 16, with a high of 66 in 2003 and a low of 22 in 2001. This trend was also found in the entire Herd Unit with 49 in 2001 and a high in 2003 of 71.

Real West fawn:doe ratios have shown a different pattern. The lowest ratio was the first year of the study, at 48:100 in 2000. The highest level was observed in 2004 at 66:100. Similarly, the control area had a low in 2000 but the highest ratio was in 2003. This ratio of 133:100 is unreliable due to the low sample size; only 10 mule deer were observed on the control area that year.

4.2 Weather Conditions

During this year's growing season (1 April - 30 September) in Wyoming, above normal precipitation extended from Albany County to Yellowstone National Park, with below normal amounts to the NE and SW of this band. This precipitation pattern has also been persistent for the 2004 water year (1 October 2003 to 30 September 2004). Based on this general demarcation, the Carbon County area would have had the above normal precipitation, while the Weston County and Rawhide Hills area would have been below normal amounts (Figure 4-5).

In April 2004 average temperatures across Wyoming ranted from five degrees above normal in the extreme western and northeast corners (which would include Weston County), to near normal temperatures over a large portion of central Wyoming. Precipitation was normal or above normal across all but the northeast part of the state where hardly any precipitation fell. Statewide, Snow Water Equivalent (SWE) was about 54 percent of average, compared to 76 percent in 2003.

In May 2004 Wyoming, on average, experienced slightly warmer and significantly drier conditions. Average temperatures ranged from four degrees below normal over portions of northern Wyoming to four degrees above normal over a portion of Laramie County. Precipitation was much below normal across all but the extreme western regions of the state. Soil moisture at the end of May revealed dryness over all but the northwestern region of Wyoming.

In June 2004 Wyoming experienced cooler and significantly wetter and drier conditions. Average temperatures ranged from 6 degrees below normal over portions of Hot Springs County to 2 degrees above normal over portions of extreme western Wyoming. Precipitation was much below normal northward from a line cutting diagonally from SE to NW Wyoming (which would include Weston and Rawhide project areas) and much above normal south of this line (which would include Carbon County). Soil moisture at the end of June revealed dryness over the eastern third of the state, especially over extreme NE Wyoming (to include Weston County).

In July 2004 Wyoming experienced cooler and significantly wetter and drier conditions. Average temperatures ranged from 4 degrees below normal over portions of Platte and Hot Springs Counties to 2 degrees above normal over portions of extreme NE (which includes Weston County), SW, and NW Wyoming. Precipitation was much below normal over SE and SW Wyoming and much above normal over central portions of the state (which includes Carbon

Table 4-5. Annual precipitation and snowfall for the three project areas.

	Carbon County (Muddy Gap)	Weston Area (Upton)	Rawhide Hills (Lusk)
Annual Precipitation (inches)			
30-yr Average	9.94	14.49	15.60
2001	6.72	15.33	11.68
2002	4.63	13.67	11.85
2003	4/48	13.96	11.16
2004	10.89	8.62	7.02
Annual Snowfall (inches)			
30 year average	50.81	43.56	52.79
2001	35.80	37.00	63.50
2002	17.00	31.00	42.40
2003	Not available	41.00	41.00
2004	25.30	42.00	27.20

County). Soil moisture at the end of July was normal over all but the extreme NE and NW Wyoming.

In August 2004 Wyoming experienced cooler and significantly wetter or drier conditions. Average temperatures ranged from near normal over portions of Sublette and Teton Counties to well below normal temperatures over portions of Platte and Goshen Counties (which includes the Rawhide area). Precipitation was much below normal north of a line from SE to NW Wyoming (to includes all project areas) and much above normal from south central and western Wyoming. Soil moisture at the end of August was normal over all but the extreme eastern and northeast regions of the state.

During the month of September, temperatures were within plus or minus two degrees of normal with the extreme western region of Wyoming having generally the coldest readings. Precipitation was generally well above normal over the southern half of the state with below normal amounts over Park and Campbell counties.

4.3 Analysis

4.3.1 Methodology Strengths and Weaknesses

The purpose of this study was to track fawn production in three project areas where predator control measures were being conducted. In each area, an adjacent control area was also monitored to provide a comparison area. The control area had no predator control, aside from incidental take.

As has been stated in previous annual reports for this study, identifying actual cause and effect interactions that explain changes in fawn survival, as expressed in changes in the fawn:doe ratio, are difficult to verify with certainty. Real West identified the following shortfalls in the methodology of this project prior to initiation of field surveys in 2000:

1. The project areas, while generally covering large areas, did not include entire herd units. In all cases, the project areas are subsets of a herd unit, making it more likely that animals move in and out of the area. Herd unit boundaries have been delineated to describe, for the most part, geographical limits of populations within which animals move freely. Changes in population size and composition could be a result of ingress and egress of animals, rather than a result of increased or decreased production.
2. Coyote populations were not monitored either on the treatment areas or the control areas. It is unknown what impact the predator control measures actually had on the coyote population, the percent of the population that was removed, or the level of coyote reproduction in the area.
3. The causes of fawn mortality are unknown and may not be a result of coyote predation. Poor vegetation, harsh winter, and other environmental conditions could also result in fawn mortality.

A concern that had been mentioned in several of the subsequent annual reports is the small sample sizes in some of the survey areas. There are particularly low numbers in the Rawhide Hills project area, as well as the Weston County control area.

Efforts were made through survey methodology to minimize these shortcomings in the protocol. Most notably, the surveys were conducted by the same person using the same routes, and within the same time period each year. While observer bias can always play a factor in how many animals are seen, since the same observer conducted all surveys in all years, this bias is minimized.

It is possible that improved numbers from the first year to the second could be attributed, to a small degree, to improved knowledge of the areas and expectancy of where to concentrate the survey time. For example, in the Carbon County surveys for Hunt Area 63, the area around Pathfinder Reservoir is especially rich with pronghorn. More time was spent in this area after the first year, and likely helped to increase observation numbers in subsequent years. Likewise, on the control area for Carbon County, the pronghorn were particularly concentrated in areas proximal to water sources. While all areas were covered every year, more survey effort was directed in the vicinity of water holes in years 2 though 5 simply because it was known that that was where the animals concentrated.

Each project area also had a control area. These areas were sometimes limited in size due to the inability to eliminate predator control measures over a five-year period in a larger area. In spite of the smaller size of the controls, they provide some comparison since weather conditions are consistent between the control and treatment areas.

4.3.2 Project Area Summaries and Herd Unit Comparisons

It's possible the predator control measures resulted in a sort of refuge where the pronghorn experienced greater security due to the lower number of predators. If this is the case, the population increase could result from ingress of animals from the adjacent area. Consideration of the population estimates of the entire Herd Unit may indicate if this is occurring. Comparisons with Herd Units and Real West results are compared and discussed in the following sections.

4.3.2.1 Carbon County

The three treatment areas in the Carbon County Project Area are considered separately due to the differences among the sites. Hunt Area 63 nearly corresponds with the boundaries of the North Ferris Mountain Herd Unit. The Herd Unit also extends north of Ferris Mountain but the bulk of the Herd Unit includes the Hunt Area 63 survey area. For that reason, of all the survey areas, this one is least likely to have significant ingress and egress of animals.

Concentrated predator control efforts took place in HA 55 but a similar concentration of effort did not occur in HA 108. Some predator control took place but it was primarily accomplished through a private contractor rather than through Wildlife Services. Due to the difference in predator control intensity, the data is separated for the two areas.

Fawn:doe ratios in HA 63 increased steadily over the past three years after an initial decline. More notable than the changes in the ratios was the number of pronghorn observed on the site. It increased steadily over the four years of the surveys.

From 2001 through 2004, the North Ferris Herd Unit increased population by 44% (Table 4-6) at the same time the number of pronghorn observed in the survey area HA 63 increased by 747%. While the entire area increased in numbers, the increase was dramatically greater in the area with predator control.

These comparisons generally provide evidence that some migration in and out of the project areas is occurring but, because herd unit populations also increased in most areas, such movements would not explain all of the increase in numbers on the treatment areas.

Table4-6. Percentage changes in pronghorn observations and population estimates after four and five years in the project areas and each respective herd unit.[1]

Time Frame	HA 55	Baggs HU	HA 108	Iron Springs HU	HA 63	N. Ferris HU	HA 61	Red Desert HU
4 yrs	141%	14%	105%	-9%	544%	5%	172%	-22%
5 yrs	151%	22%	6%	-4%	747%	44%	67%	-15%

[1]Data for Hunt Areas is observation numbers by Real West and data for Herd Units is for population estimates from the WGFD. For each combination, the Hunt Area is contained within the much larger Herd unit.

4.3.2.2 Weston County

Comparisons of fawn:doe ratios for the two survey areas and the entire Herd Unit show some variation in the treatment area, with a significant increase in 2004 (Figure 4-1). This increase in 2004 was not observed on either the control area or the herd unit as a whole.

The number of pronghorn observed on the treatment area has shown a steady increase since 2001. The control area had four years of decline, but increased in 2004. The population of the entire Herd Unit declined the first two years of the survey period , remained steady for two years, and then declined in 2004.

Percentage changes in observation numbers and herd unit population estimates, show the treatment area increased considerably more than the herd unit itself (Table 4-7). This indicates

Figure 4-1. Comparisons of Fawn:Doe ratios on the Weston County Project Area.

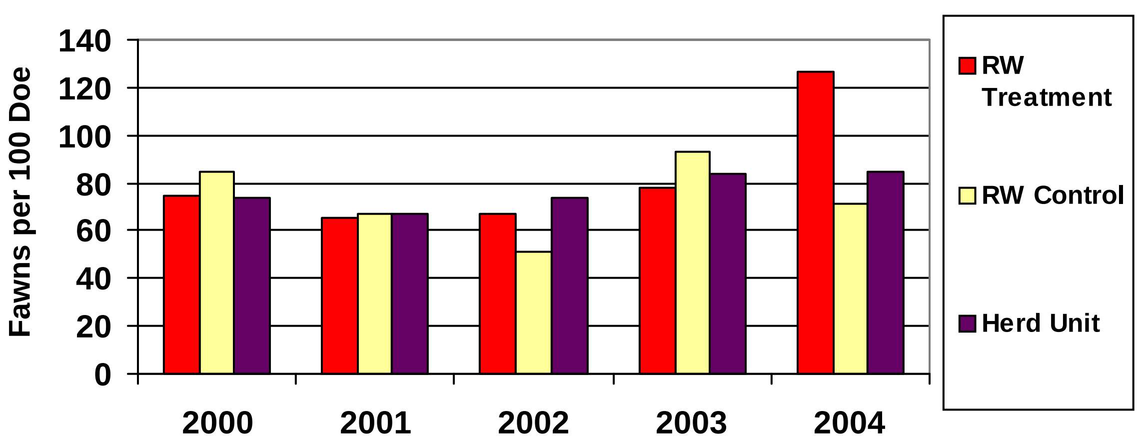


Table 4-7. Percentage changes in pronghorn observations and population estimates after four and five years in the project areas and herd unit for the Weston County Project.[1]

Time Frame	Treatment	Control	Herd Unit
After 4 yrs	68%	-53%	15%
After 5 yrs	74%	56%	21%

[1]Data for treatment and control areas is observation numbers by Real West and data for Herd Units is for population estimates from the WGFD.

that, while some migration into the treatment area occurred, it is not the sole explanation for the improved populations. The control area showed more variability, most likely due to the low sample size.

4.3.2.3 Rawhide Hills

Due to low samples sizes, comparisons of data in the Rawhide Hills area should be considered with caution. The fawn:doe ratios on the treatment area have varied only slightly over the five year period, with the highest ratio in 2004. The control area showed similar slight variations over the period, with a large increase in 2003 but this number is skewed because only a single group of mule deer was observed on the control area. Herd Unit ratios have also fluctuated over the period, with a low in 2001 and a high in 2003.

Population estimates for the herd unit and Real West observation numbers (Table 4-8) show a decline in the treatment area after fours years of predator control, but an increase over the five year period. At the same time, the Herd Unit also showed a slight decline over four years but returned to nearly the same population at the end of the five-year period.

Table 4-8. Percentage changes in pronghorn observations and population estimates after four and five years in the project areas and herd unit for the Rawhide Hills Project.[1]

Time Frame	Treatment	Control	Herd Unit
After 4 yrs	-71%	-73%	-11%
After 5 yrs	+3%	+122%	0%

[1]Data for treatment and control areas is observation numbers by Real West and data for Herd Units is for population estimates from the WGFD.

4.4 Conclusions

The weather over the five years of the survey period was a mixed bag for wildlife. Severe drought conditions persisted for the entire period. At the same time, winters were relatively mild. The expected result of this combination is that fawn production would be lowered due to poorer nutrition of the does as a result of reduced forage availability. However, those fawns that survive into the winter were more likely to make it to yearling age because of the mild winters.

Percentage of the population comprised of yearling bucks provides information on survival of fawns to the next year. While female yearlings are not distinguished in the field, bucks are more readily identified.

The percentage of yearling bucks in the Carbon County project areas show rates up to 10.2 and 10.7 percent for the Hunt Area 108 (2004) and Ferris Mountain (2003), respectively (Figure 4-9). In all areas, including the control area, the percentage of yearling bucks in the population increased over the five-year period. The same was true of both the treatment and control areas for Weston County, with the control area showing highest percentage of yearling bucks. In the Rawhide hills, the percentage of bucks declined in the treatment area, while increasing in the control area.

Changes in fawn:doe ratios over four and five years (Table 4-10) show improvements after four years in all but Hunt Area 108 and Ferris Mountain areas of the Carbon County Project area. By the fifth year, changes from the start of the study showed a decline in Hunt Area 109 and in the control areas of both Weston County and Rawhide Hills. All other areas showed an increase in

Table 4-9. Percentages of the surveyed populations comprised of yearling bucks in each of the three project areas.

	2001	2002	2003	2004
Carbon County				
Treatment: Hunt Area 55	2.4	5.2	3.5	3.5
Treatment: Hunt Area 108	2.6	2.6	3.5	10.2
Treatment: Ferris Mountain	7.3	4.2	10.7	7.7
Control Area	5.0	6.9	4.2	6.5
Weston County				
Treatment Area	7.7	4.8	6.2	9.9
Control Area	2.0	7.8	7.0	15.1
Rawhide Hills				
Treatment Area	6.9	4.0	3.2	4.5
Control Area	3.4	7.4	0	9.8

Table 4-10. Summary of Fawn:Doe Ratio Changes for 2000 - 2004 in the Three Project Areas.

	2000 - 01	2001 - 02	2002 - 03	2003 – 04	Yr 1 to 4 and 1 to 5	Yr 1 and 5
Carbon County						
HA 55	+11	-6	-1	+9	+4	+13
Ferris Mountain Treatment	--	-36	+3	+24	-30	+9
Hunt Area 108	+18	-29	+11	-8	Same	-8
Control Area	+3	-6	+7	+20	+4	+24
Weston County						
Treatment Area	-10	+2	+11	+49	+3	+52
Control Area	-18	-16	+42	+22	+8	-6
Rawhide Hills						
Treatment Area	+5	+8	-9	+14	+4	+18
Control Area	+4	+2	+78	-85	+81	-4

[1] Adjusted ratio is the number of fawns per 100 does, subtracting the number of yearling bucks from the number of does.
[2] Extremely small sample size.

ratio.

Population changes after four and five years (Table 4-11) show increases in all four survey areas of Carbon County with the most significant change in the Ferris Mountain area. Improvements were also observed in the both the treatment and control areas of Weston County, with the greatest increase in the treatment area. Increases in the Rawhide Hills areas were observed after five years, but not after four years. As mentioned previously, drawing any conclusions on that project area should be done cautiously due to the low observation numbers.

In assessing impacts of predators to big game populations it must be determined what factor(s) are limiting the population. Often winter range quality and quantity are limiting the population, regardless of quality of vegetation during spring, summer, and fall. If the big game population is near the carrying capacity of the area, it is likely that predation is not resulting in a reduced population. If predators didn't reduce numbers, other habitat limitations would.

However, if the big game population is well below the area's carrying capacity, predation on fawns could be additive. In other words, if a predator didn't kill the fawn, it would likely live to reproductive age. The result is that predation can have a significant impact on the big game population.

Ballard et al (2001) provides a literature review of deer and predator relationships. The authors of the review concluded that predator control was useful in improving deer populations in some

Table 4-11. Summary of Population Changes for 2000 - 2004 in the Three Project Areas.

	Yr 1 to 4	Yr 1 and 5
Carbon County		
HA 55	141	151
Ferris Mountain Treatment	544	747
Hunt Area 108	108	6
Control Area	172	67
Weston County		
Treatment Area	68	74
Control Area	-53	56
Rawhide Hills		
Treatment Area	-71	3
Control Area	-73	122

[1] Adjusted ratio is the number of fawns per 100 does, subtracting the number of yearling bucks from the number of does.
[2] Extremely small sample size.

cases, while in other cases it was not.

They found the following similarities among those cases where predator control appeared to help the deer:

- Predator control was implemented when the deer populations were below habitat carrying capacity;
- Predation was identified as a limiting factor;
- Control efforts reduced predator populations enough to yield results (e.g., at least 70% of the coyote population was removed);
- Control efforts were timed to be most effective (just prior to predator or prey reproduction); and,
- Control took place at a focused scale (generally a study area <259 mi^2).

Conversely, there were similarities where predator control was not effective or could not be measured at improving mule deer populations. These included:

- When mule deer populations were at or near habitat carrying capacity;
- When predation was not a key limiting factor;
- Where control failed to reduce predator populations sufficiently to be effective; and,
- Where control efforts were on large-scale areas.

They found one failure in much of the research has been a lack of an adequate experimental design. Often small sample sizes limited usefulness of studies because of their low statistical power to actually detect significant differences.

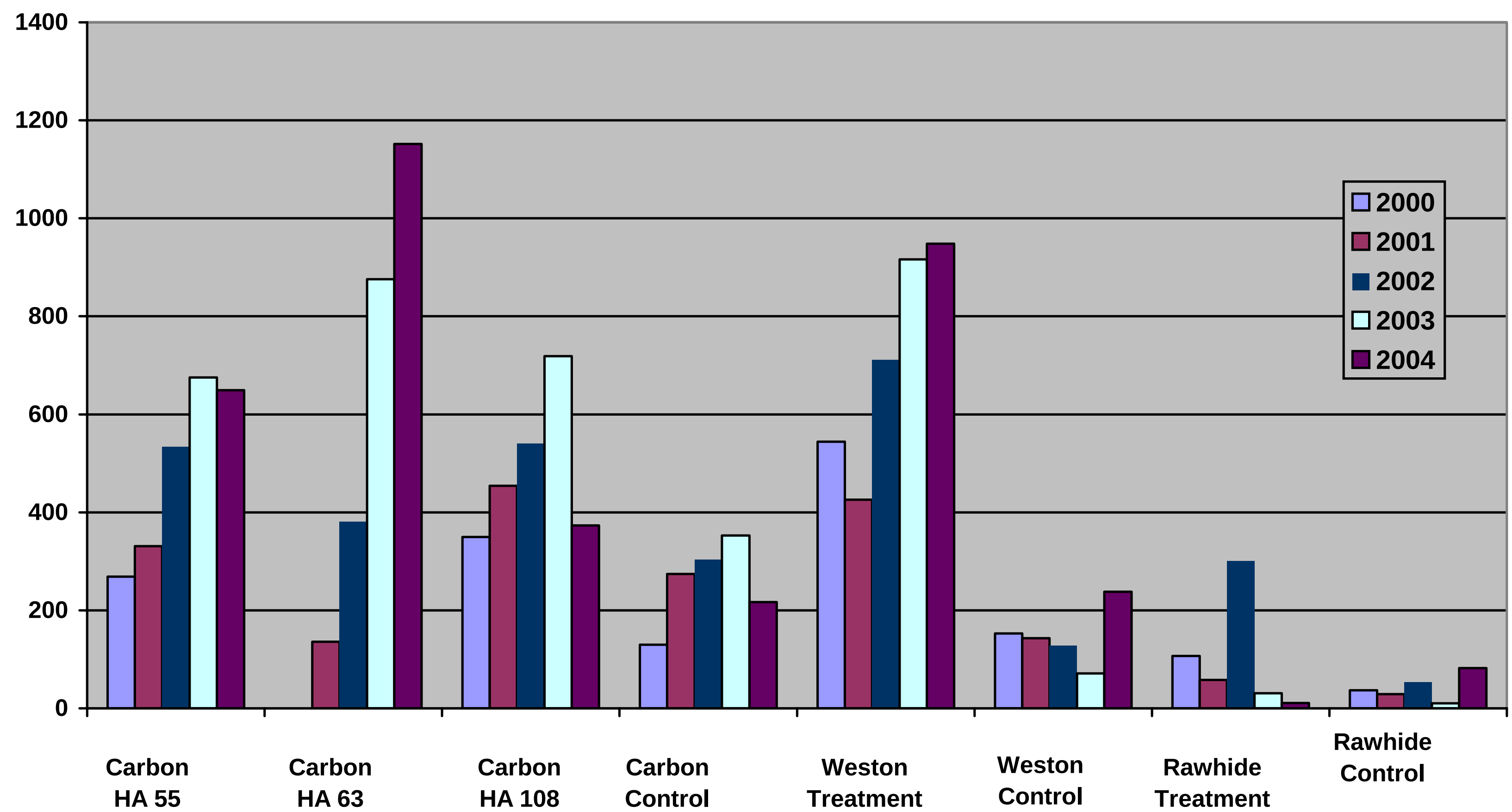
The authors of the paper concluded that "seasonal or long-term changes in fawn:doe ratios can provide an indication of when most losses are occurring, but cannot be used to determine causes for changes. Simple changes in fawn:doe ratios cannot be used to determine whether predation is a limiting factor. Only through intensive studies can predation be identified as a significant mortality factor."

The implications from this research on this project are that any increases in fawn survival on all project areas when compared to control areas should be viewed with caution. The sample sizes are small, as are the study areas. No doubt migration in and out of each area by big game and predators occurs.

However, the results of these surveys over the five-year period indicate that it is likely predator control, in association with mild winters, helped improve big game populations through improved fawn survival. When a big game population is depressed and seemingly unable to rebound several years after a harsh winter, predation could a factor in keeping the population down. Temporary control measures to increase fawn production could help in population recovery. Once big game numbers near the habitat carrying capacity, predation is less likely to have as noticeable an impact on the population.

The result of this study suggests intensive predator control may result in increased, observed, preseason fawn:doe ratios and result in increases in pronghorn numbers. The areas with predator control showed population increases that were, in most areas, greater than those observed within the entire herd unit (Figure 4-2).

Figure 4-2. Total Pronghorn or Mule Deer Observed in Three Study Areas Over Five Years.



5.0 PERSONS CONTACTED

The following individuals were contacted for information contained in this report:

Individual	Company/Agency	Reason for Contact
Dr. Archie Reeve	Adjunct Professor University of Wyoming	Survey technique analysis
Dr. Fred Lindsey	Associate Professor University of Wyoming, Wyoming Cooperative Research Unit	Survey technique analysis
Rod Merrel	Wyoming Wildlife Services	Predator control information
Lee Denney	Rancher, Rawhide project area	Predator control information
Alan Todd	Rancher, Weston County Predatory Animal Board	Predator control information
Greg Hiatt	Wyoming Game and Fish Dept., Rawlins District	Pronghorn classifications
Greg Anderson	Wyoming Game and Fish Dept., Newcastle area	Pronghorn classifications
Bob Lanka	Wyoming Game and Fish Dept., Laramie District	Mule deer classifications
Bill Rudd	Wyoming Game and Fish Dept., Rock Springs Dist.	Helicopter observation success rates
Daryl Lutz	Wyoming Game and Fish Dept., Casper Dist.	Predator control effects; helicopter coordination
Mark Nelson	Wyoming Game and Fish Dept., Cheyenne area	Helicopter survey results
Bart Kroger	Wyoming Game and Fish Dept., Douglas area	Helicopter coordination
Mark Zornes	Wyoming Game and Fish Dept., Wheatland area	Helicopter coordination
Martin Hicks	Wyoming Game and Fish Dept., Wheatland area	Helicopter coordination; Mule deer classifications
Joe Sandrini	Wyoming Game and Fish Dept., Newcastle area	Pronghorn classifications

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Habitat use and population dynamics of Shira's Moose in NW Wyoming

FY 2006 ADMB Progress Report

Project Title: Resource Selection and Population Dynamics of Shira's Moose (*Alces alces shirasi*) in Northwest Wyoming

Submitted by: Scott A. Becker, Master of Science Candidate, U.S. Geological Survey, Wyoming Cooperative Fish and Wildlife Research Unit, Department of Zoology and Physiology, University of Wyoming

Project Description: In the fall of 2004, funding was secured from the Teton County Conservation District, Wyoming Animal Damage Management Board, Wyoming Cooperative Fish and Wildlife Research Unit - University of Wyoming, Wyoming Department of Transportation, Wyoming Game and Fish Department, and the Wyoming Governor's Big Game License Coalition/Wildlife Heritage Foundation of Wyoming to expand the Jackson moose study and fund a graduate student at the University of Wyoming. The current phase of the study began in January 2005 when a graduate student was selected for the position. There are 3 primary research goals associated with this project: (1) investigate resource selection, seasonal distribution, and movement patterns of adult female moose in the Buffalo Valley to better understand the relationships between moose and their habitat requirements, (2) assess potential causes for recent population declines by estimating population parameters and measuring physiological health indices, and (3) estimate the timing and location of adult female moose movements associated with U.S. Highway 26/287 and use this information to build a model that will be used to predict important moose crossing locations.

Project Summary (as of 12-1-05): Moose captures were initiated in February 2005 and 27 males and 20 females were captured over a 3-day period. All males were fitted with VHF radio transmitters to monitor survival status and females were fitted with global positioning system (GPS) radio transmitters. The GPS collars will collect a high quantity and quality of location data that will be used to assess resource selection patterns and movements associated with highway 26/287, in addition to survival status. Furthermore, 18 adult females fitted with VHF radio collars from the pilot study will be used to assess female survival rates.

All moose captured in 2005 were found to be negative for 6 disease antigens, they had a low prevalence of endoparasites (only detected in 11 moose), low to moderate tick loads, and most moose appeared to be in relatively good condition based on mid-winter rump fat depths.

Eighteen of 20 (90%) adult female moose captured were pregnant, and we were unable to determine pregnancy rate of the 18 cows from the pilot study. Calf production surveys were initiated in early June to determine number of calves born to radiocollared females. Of the 20 cows with GPS collars, 17 were still alive in June and 16 were known to be pregnant; however, only 3 calves were observed. Of the 18 cows with VHF radio transmitters, 15 were still alive and 11 were observed with calves. A high percentage of calf deaths occur within 6-8 weeks post-parturition, so cow/calf surveys were initiated on July 20 to document early calf survival. All collared females were surveyed to assure that no calves were missed during calf production surveys in June. Of the 14 calves observed in June, only 7 remained, and no additional cows were observed with calves, suggesting initial surveys were accurate in determining calf presence.

To date, there have been 12 radiocollared moose mortalities - 3 were capture related (1 F, 2 M), 3 were legal harvests (3 M), 2 were unknown causes (2 F), 1 was a possible cougar predation (F), 1 was possible wolf predation (F), 1 bear or wolf predation (F), and 1 appeared to have died of natural causes (F). In addition, 1 calf of a radiocollared female got hung up in a fence and had to be put down and 3 adult moose (1 M, 1 F, 1 Unk - all uncollared) died as a result of vehicle collisions on highway 26/287.

Monthly fixed-wing surveys to document survival status is ongoing and field work will begin once again during January 2006. In February 2006, females with GPS radio collars will be recaptured to measure physiological health parameters and to download data from the transmitters. In addition, a sample of males will be captured to maintain sample size at 25 and the remaining VHF transmitters will be placed on additional females in the area. Analysis of the first year of data will begin once captures are complete.

FY05-06 Budget:

INCOME			
Source		**FY 2005**	**FY 2006**
Animal Damage Management Board		23600	23600
Teton County Conservation District		12000	
University of Wyoming (Coop Unit)		12000	
Wyoming Dept. of Transportation		30000	10000
Wyoming Game and Fish Dept. - M&O		14400	35000
WY Governor's Big Game License Coalition		10000	15000
	Total	**102000**	**83600**

EXPENSES			
VHF Radio Collars		10000	
GPS Radio Collars		30100	
Laboratory Analysis		4000	2000
Immobilization Drugs		3000	3000
Helicopter (capture - $450/hr)		10800	10000
Helicopter (calving flights - $530/hr) - early June		10000	10000
Helicopter (calf survival flights - $530/hr) - late July			10000
Fixed Wing (ferry time - $145/hr)		3500	3500
Fixed Wing (telemetry - $220/hr)		7900	7900
Graduate Student Stipend and Tuition		10000	20000
UW Coop Research Unit Overhead		5000	4200
Vehicle Expense and Mileage		3700	9000
Travel and Per Diem		2000	2000
Computer Facility Support		1000	1000
Misc. Equipment and Maintenance		1000	1000
	Total	**102000**	**83600**

RESOURCE SELECTION AND POPULATION DYNAMICS OF SHIRA'S MOOSE (*Alces alces shirasi*) IN NORTHWEST WYOMING

STUDY PLAN

By

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November 2005

Graduate Committee	Funding Agency Representatives Present
Stanley Anderson (Chair in Memoriam)	Cody Beers (WYDOT)
Wayne Hubert (Co-Chair)	Doug Brimeyer (WGFD)
Fred Lindzey	Michael Patritch (WYDOT)
Tom Thurow	Hank Uhden (ADMB)
Chuck Anderson	

It is generally believed the first modern moose (*Alces alces*) arrived in North America via the Bering land bridge approximately 10,000–14,000 years ago (Peterson 1955, Bubenik 1998a, Bowyer et al. 2003, Hundertmark et al. 2003) when large continental ice sheets covered most of the northern portion of the continent. With the retreat of the continental glaciers, moose began to inhabit portions of North America once covered with ice, although the exact patterns of dispersal and speciation are still heavily debated (Peterson 1955, Bubenik 1998a, Bowyer et al. 2003, Hundertmark et al. 2003). Nevertheless, 4 distinct subspecies of moose evolved in North America, each strongly associated with the distribution of boreal forests. *A.a. americana* occurs from Ontario eastward to the Atlantic Ocean; *A.a. andersoni* occurs from western Ontario and northern Michigan to British Columbia and the Yukon Territory; *A.a. gigas* occurs in Alaska and western Yukon Territory; and *A.a. shirasi* occurs in the mountains of Colorado, Utah, Wyoming, Idaho, Montana, southern Alberta, and southeastern British Columbia (Bubenik 1998a).

Moose were quite rare in Wyoming prior to the mid 1800s. Osborne Russell trapped and traveled extensively throughout the Jackson Valley and what is now Yellowstone National Park (YNP) from 1834 to 1843 and never mentioned observing a moose in his journal (Haines 1955), although detailed descriptions of other wildlife species were noted. Sporadic observations of moose occurred in YNP during the late 1800s (Houston 1968), but it is believed the population did not begin to increase until after the establishment of YNP in 1872 (Peterson 1955, Denniston 1956). By the late 1890s and early 1900s, observations of moose in the Jackson Valley were on the rise (Peterson 1955) and Houston (1968) believed the establishment of a self-sustaining moose population in the Jackson area occurred shortly before 1912 via emigrations from YNP.

Moose management in Wyoming began shortly after moose appeared in the state (Blair 1987). In 1882, the first hunting seasons for big game were set from 1 August through 15 November in an attempt to protect species from overexploitation. Protection intensified in 1899 when the Wyoming legislature enacted laws eliminating moose hunting due to very low densities. This continued until 1912 when moose populations were deemed large enough to sustain limited harvest. Populations continued to increase in Wyoming through the 20th century and currently, through natural emigration or translocation, moose occupy almost all available habitats in the state.

During the late 1800s and early 1900s, government sponsored predator removal programs greatly reduced large carnivore populations in northwest Wyoming. Gray wolves (*Canis lupus*) were extirpated from the state by the 1930s (Wyoming Game and Fish Department [WGFD] 2003). Within the last 30 years, grizzly bear (*Ursus arctos*) densities have increased (Haroldson 2005) and increases in mountain lion (*Puma concolor*) populations are also suspected. Wolves were reintroduced into YNP in 1995 and 1996 and were first observed in the Jackson area during the winter of 1997/98 (WGFD 2003). As of 2004, the Teton Pack (numbering 13 wolves – 5 adults and 8 pups) is the only wolf pack that inhabits the Jackson Valley, and the Yellowstone Delta Pack (numbering 19 wolves – 15 adults and 4 pups) resides along the Yellowstone River in the northeast portion of the study area (U.S. Fish and Wildlife Service [USFWS] 2005).

There have been 4 primary studies of moose in the Jackson Hole area during the last 50 years. The first significant study was conducted by Denniston (1956) who investigated the ecology, behavior, and population dynamics of Shira's moose in the Jackson Valley. Altmann (1959, 1960) then studied group dynamics and social organization of the Jackson moose. From 1963 to 1967, Houston (1968) studied habitat relationships, energetics, and population dynamics of moose in the Snake River drainage to address "life history requirements and potential mechanisms of population regulation." More recently, Berger et al. (1999, 2001) investigated moose population dynamics and habitat relationships.

During these previous studies, predator populations were either extirpated from the valley or existed in low densities. Nonetheless, fluctuations in moose populations were observed, and both Denniston (1956) and Houston (1968) felt that willow conditions may have limited moose populations during their studies. Also, Berger et al. (2001) reported detrimental browsing in this region during spring and summer 1998 and suspected moose had degraded their habitat due to relatively high densities.

WGFD (1975-2004) was also concerned about overutilization of willows on winter range during the 1970s. They felt moose population declines observed in the early 1980s were a result of winter range overuse, not only from moose, but also from an increasing elk (*Cervus elaphus*) population. An increase in elk hunting permits, habitat improvement projects on moose and elk winter range, a reduction in the number of moose permits issued, and a series of mild winters led to an increase in moose numbers by the late 1980s (WGFD 1975-2004). From the early 1990s through 2004, moose populations have continued to fluctuate. As of 2004, however, population trend counts are at the lowest levels since the late 1970s and calf:cow ratios are the lowest in 30 years (Figure 1).

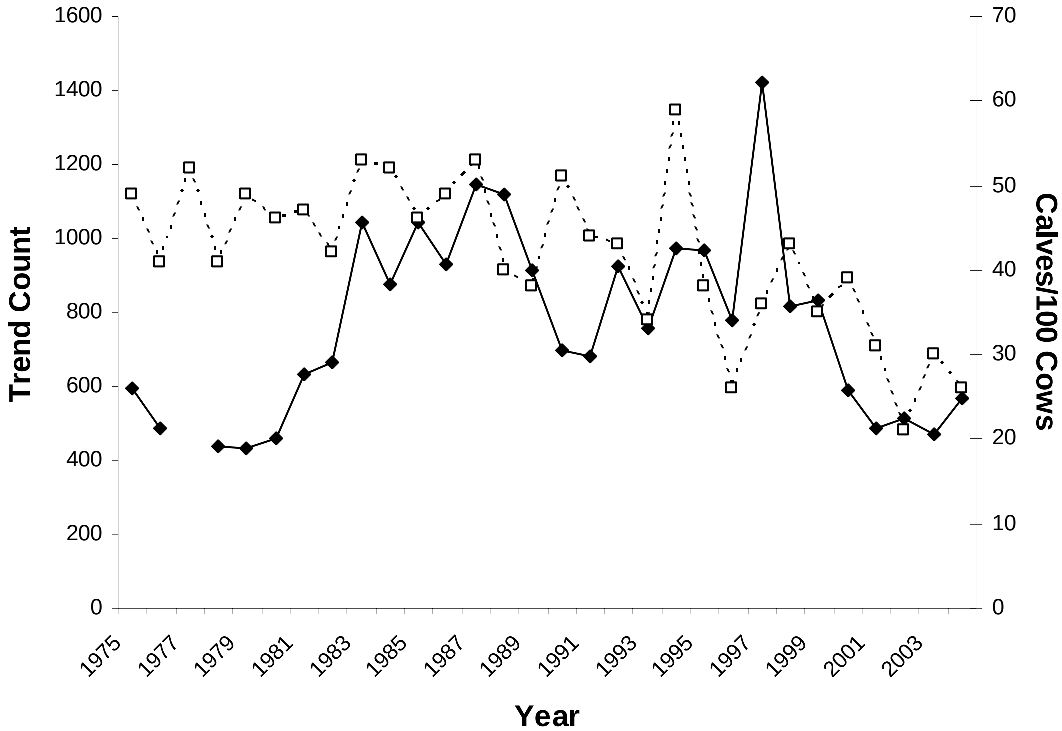

Figure 1. Jackson Moose Herd Unit (M103) population trend counts (solid line) and calf:cow ratios (dotted line), 1975-2004. No trend count data collected in 1977.

This study was initiated to investigate potential causes for the recent population decline observed in the north Jackson moose herd. My research goal will be to assess moose physiological health, survival, reproductive rates, and moose resource selection to infer the potential mechanisms limiting the Jackson moose population. In addition, I will estimate the timing and location of moose movements associated with highway 26/287 which bisects crucial moose winter range in the Buffalo Valley. The remainder of this study plan is divided into 3 chapters describing (1) moose resource selection, distribution, and movement patterns, (2) population dynamics, and (3) movements associated with highway 26/287. To minimize redundancy, the study area, capture methods, and moose monitoring techniques are presented in Chapter I only.

CHAPTER I:

RESOURCE SELECTION, SEASONAL DISTRIBUTION, AND MOVEMENT PATTERNS OF ADULT FEMALE MOOSE IN THE BUFFALO VALLEY USING GLOBAL POSITIONING SYSTEM (GPS) TECHNOLOGY

INTRODUCTION

Telfer (1984; as cited in Peek 1998) characterized primary moose habitats into 5 broad vegetation classes: (1) boreal coniferous forest, (2) mixed coniferous/deciduous forest, (3) large delta floodplains, (4) tundra and sub-alpine shrub zones, and (5) stream valley shrubs including riparian areas. These 5 classes contain both permanent and transitory habitats that are important for moose species throughout their range. Stable habitats are characterized by plant communities that do not succeed to different vegetation types (Peek 1998), such as delta floodplain and stream valley shrub habitats. Although these habitats are transitory in the sense that stream alterations and occasional flooding changes the structure of the system, the plant community structure rarely varies. In Wyoming, however, Houston (1968) noted that some stream valley communities will succeed to coniferous forest types if undisturbed. Large delta floodplains and stream valley shrub habitats consist of deciduous shrubs, which provide a high quantity of forage biomass that is able to support populations of migratory and non-migratory moose (Houston 1968, Peek 1998; Bowyer et al. 2003). These habitats can be found throughout the Rocky Mountain region, through western Canada, and into Alaska (Peek 1998).

Transitory habitats are those created by fire, insect outbreaks, blowdowns, and/or commercial timber harvest which form a mosaic of plant successional stages (Peterson 1955, Peek et al. 1976, Peek 1998, Bowyer et al. 2003) and are most notable in the boreal coniferous and mixed forest types. Of these disturbances, fire is particularly important in creating the early seral shrub communities that provide a variety of forage, both in quality and quantity (Peek et al. 1976, Pierce and Peek 1984, Bangs et al. 1985, Regelin et al. 1987, Peek 1998, Bowyer et al. 2003). On the Kenai Peninsula, Alaska, Oldemeyer and Regelin (1987) observed that shrub densities were highest in burned areas that were 8, 13, and 30 years old, but varied greatly depending on soil type. In addition, nutritive value of spring and summer forage was greater 12 years post-fire compared to 32 years post-fire, while winter forage quality remained unchanged between the 2 seral stages (Regelin et al. 1987). Furthermore, moose densities (Loranger et al. 1991) and moose twinning rates (Franzmann and Schwartz 1985) were greater in early compared to later successional stages. The benefits of increased biomass and nutritive quality of spring and summer forage created by fires declines after 25 years (Regelin et al. 1987) resulting in decreases in population density (Loranger et al. 1991) and reproductive output (Franzmann and Schwartz 1985). Peek (1998) cautioned that although habitat conditions improve post-fire, low density populations are less likely to benefit if the population is limited by predation.

Although most moose populations are highly associated with transitory habitats, some populations in the Rocky Mountain region utilize mature conifer forests more than other available habitats. This may be due in part to the reduced abundance of riparian areas (Matchett 1985, Peek et al. 1987, Peek 1998), an abundant understory of forage in mature forests (Pierce 1984, Pierce and Peek 1984, Peek et al. 1987), or interspecific competition (Tyers and Irby 1995, Tyers 2003). In northwest Montana, where major understory forage species or large willow (*Salix spp.*) complexes are absent, moose utilized logged sites where shrub density is greatest throughout most of the year, then shifted to densely timbered sites during late winter (Matchett 1985). In north-central Idaho, moose use of old-growth forests was greater than expected in all seasons except summer (Pierce and Peek 1984). Understory species within the old-growth forests comprised approximately 90% of moose diets in all seasons (Pierce 1984). In YNP, moose were not found to utilize early seral vegetation possibly due to competition for browse with elk (Tyers and Irby 1995, Tyers 2003). Moose either used willow stands at higher elevations than elk winter range or small patches of willow or aspen within elk winter range until browse availability was depleted or deep snows forced them to mature conifer stands.

Moose winter ranges are typically shrub-dominated habitats that occur naturally along floodplains or riparian areas, or they may be created via natural (e.g., fire) or man-made (e.g., timber harvest) means. As snow depths increase or snow crusts form, however, the use of mature forests with substantial overstories becomes increasingly important (Houston 1968, Peek et al. 1976, Pierce and Peek 1984, Pierce 1984, Matchett 1985, Hundertmark et al. 1990, Renecker and Hudson 1992, Tyers and Irby 1995, Bowyer et al. 2003). Renecker and Hudson (1992) suggested that moose can be highly selective during early winter, choosing areas that maximize energy intake (i.e., high biomass) while reducing energy expenditure (i.e., search time). However, as winter progresses, deep snows and/or snow crusts limit forage availability. Therefore, moose seek out areas with substantial overstories where snow depths may be less (Bowyer et al. 2003) and forage can be found protruding from the snow (Peek 1998). In areas where overstory cover does not exist, moose seek out microsites that offer the best combination of decreased snow depths and forage availability (Peek 1998). In Wyoming, Anderson (1994) noted that moose left narrow riparian bottoms when snow depths increased and selected upland slopes holding less snow.

The size of an individual moose home range depends on many factors, but one of the primary factors is habitat quality as it relates to the nutritional demands of the animal (Hundertmark 1998). Many moose within a population have distinct summer and winter home ranges to meet their basic life requirements while other moose remain on the same range year-round. Within a home range, moose may display an area of core use where an important component of the habitat may be found that satisfies the animals' needs. Comparisons between home range sizes among studies are difficult to ascertain due to the method of analysis used (Gallerani Lawson and Rodgers 1997) and the seasonal breakdowns of home range size. In general, however, Hundertmark (1998) found that south of 60 degrees north latitude, mean seasonal home range size never exceeded 51.2 km^2, but north of this latitude, seasonal home range size increased sharply. In Wyoming, Houston (1968) noted that mean summer and winter home range size rarely exceeded 3.9 km^2 and movements between ranges were typically 8 to 16 km with some moose moving more than 32 km.

Analysis of seasonal distribution, movement, and resource selection patterns is the primary focus of this study and will aid in understanding the ecology of moose in northwest Wyoming. The spatial and temporal dynamics of moose distribution and movement are one of the most basic components to understanding the ecology of moose, and by analyzing seasonal resource selection patterns, I hope to better understand the "why" of moose movement and distribution. Resource selection patterns will also assist state and federal agencies in developing effective land management strategies for moose in northwest Wyoming.

GOALS, HYPOTHESES, AND OBJECTIVES

Research Goal

In this study, I propose to investigate resource selection, seasonal distribution, and movement patterns of adult female moose in the Buffalo Valley to better understand the relationships between moose and their habitat requirements.

Hypothesis

H_{o1}: Moose will utilize vegetation types in proportion to availability within the study area.

H_{a1}: Moose will utilize vegetation types disproportionate to availability with riparian areas receiving the highest amount of use relative to availability in summer and winter.

H_{o2}: Home range size of individual adult female moose will not differ between seasons or years.

H_{a2}: Home range size of individual adult female moose will be greater in summer than winter and will differ between years.

H_{o3}: Timing of movements between seasonal ranges will not differ between individual moose or between years.

H_{a3}: Timing of movements between seasonal ranges will differ between years

Objectives

1) Create a detailed model of summer and winter resource selection patterns for adult female moose in the Buffalo Valley
2) Determine size and distribution of seasonal home ranges, estimate timing of movements between seasonal ranges, and location of migration routes and/or travel corridors

STUDY AREA

The study area is located in northwestern Wyoming and lies within the central portion of the Greater Yellowstone Ecosystem (GYE). It covers approximately 5195 km^2 (Smith 1994) and is primarily located in Teton County, Wyoming (Figure 2). The majority of this area is on the west side of the continental divide and encompasses the headwaters of the Snake River drainage south to its' confluence with the Gros Ventre River (GVR). Major drainages of the Snake River include Pacific Creek, the Buffalo Fork River (BFR), and the GVR. The segment of the study area on the east side of the continental divide includes a portion of the Yellowstone River drainage north to Yellowstone Lake. Houston (1968), Cole (1969), and Smith and Robbins (1994) describe additional physiographic features of this area where elevations range from 1,866 m in the southern end of the Jackson Hole valley to 4,195 m at the peak of the Grand Teton.

Approximately 97% of the study area is public land administered by the National Park Service (NPS) and the United States Forest Service (USFS). It includes most of

Grand Teton National Park (GTNP), the southern portion of YNP, and the northern section of the Bridger-Teton National Forest (BTNF). The Teton Wilderness encompasses the majority of USFS land between the BFR and YNP. The Blackrock area, described by Holm (1998), is the primary multiple use area south of the BFR. About 3% of the remaining area is private or state-owned lands.

Lower elevations, and many south-facing slopes at higher elevations, are dominated by big sagebrush (*Artemisia tridentata*). In the southern end of the valley, big sagebrush interspersed with bitterbrush (*Purshia tridentata*) is common. Mid-elevations are characterized by large stands of lodgepole pine (*Pinus contorta*) intermixed with small stands of Douglas fir (*Psuedotsugia menziesii*) and aspen (*Populous tremuloides*). Engelmann spruce (*Picea* engalmanni) and subalpine fir (*Abies lasiocarpa*) are found on north slopes and at more mesic sites. Engelmann spruce, subalpine fir, and lodgepole pine intermixed with smaller stands of whitebark pine (*Pinus albicaulis*), limber pine (*Pinus fexilis*), and aspen dominate higher elevations. Open forest parks and subalpine meadows dominated by grasses and forbs occur in all elevational gradients. Riparian areas are dominated by willow intermixed with narrowleaf cottonwood (*Populus angustifolia*) (Wigglesworth and Wachob 2004). Primary moose winter ranges are located in riparian areas to mid elevation habitats and summer ranges encompass all available habitats.

One of the largest elk herds in North America also inhabits the study area. Antelope (*Antilocapra americana*) migrate to the Jackson Valley in the summer and populations of bison (*Bison bison*) and mule deer (*Odocoileus hemionus*) occur year-

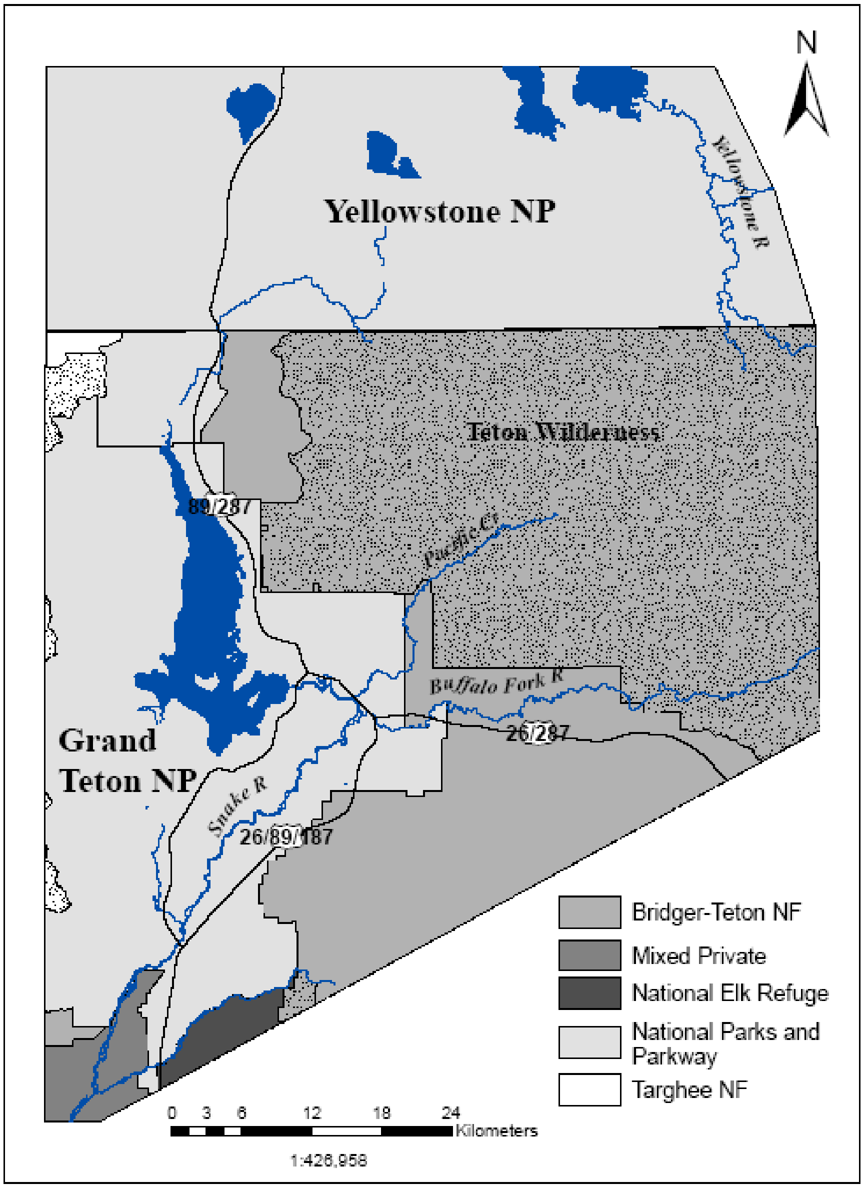

Figure 2. Study area located in northwest Wyoming.

round. Carnivores that inhabit the area include black bear (*Ursus americanus*), grizzly bear, mountain lion, wolves, and the full suite of forest meso-carnivores.

Mean annual temperatures from all weather stations in the Snake River drainage over a 30-year period (1975-2004) averaged 2.1°C, with the lowest mean temperature occurring in January (-10.2°C) and the highest occurring in July (14.9°C). Precipitation varies throughout the study area with the northern portion receiving the greatest amounts (Boyce 1989). The Snake River drainage averages 56.2 cm of precipitation each year primarily as winter snowfall.

The Jackson Moose Herd Unit (M103) includes WGFD hunt areas 7, 14, 15, 17-19, 28, and 32. Hunt areas 7, 14, and 32 in the Teton Wilderness have been closed for the 2005 and 2006 hunting seasons. As of 2004, the population estimate for the Jackson Moose Herd Unit was 2,797 individuals and the population objective for the entire herd is 3,600 moose (WGFD 1975-2004).

Jackson is the primary human population center in the region (population 8,650), and the economy is driven by tourist activities. Major highways in the valley are few, but heavily traveled, especially during the summer months. In 2004, there were 2,360,373 recreational visitors to GTNP with the majority of visits occurring from May thru September. US highway 26/287 bisects the Buffalo Valley west of GTNP before it splits at Moran. US highway 26/89/187 is the major thoroughfare between Jackson and Moran and US highway 89/287 is the main travel corridor between Moran and YNP. The Blackrock area, south of the BFR, contains the majority of primary and secondary dirt roads in the area. Much of the remaining area is accessible only on foot or horseback.

METHODS

Moose Capture and Handling

Moose will be captured using ground and helicopter darting techniques in February 2005. A CO_2-powered, adjustable dart rifle (Dan-Inject North America, Fort Collins, Colorado, USA) will be used for ground captures and a .22-caliber blank dart rifle (Model 193, Pneu-dart, Williamsport, Pennsylvania, USA) will be used during helicopter captures (Kreeger et al. *in press*). All moose will be immobilized with 10 mg thiafentanil (A-3080®, Wildlife Pharmaceuticals, Fort Collins, Colorado, USA) and antagonized with 300 mg naltrexone (Trexonil®, Wildlife Pharmaceuticals, Fort Collins, Colorado, USA) administered one-half intramuscularly and one-half subcutaneously (Kreeger et al. *in press*). Once immobilized, moose will be approached by ground crews who will ensure that the animal remains in a sternally recumbent position.

Sex, morphometric measurements, ultrasonic rump fat measurements (Stephenson et al. 1993, Stephenson et al. 1998), and body condition scores (Franzmann 1977) will be recorded. Age will be estimated from incisor-tooth wear and photographs of incisors will be taken of all animals for reference. Blood, hair, fecal, and ear swab samples will be collected for (1) internal and external parasite analyses, (2) disease analyses, (3) trace mineral analyses, (4) serum chemical analyses, (5) hematological analyses, and (6) pregnancy status of females. In addition, a 30 second tick count will be performed along the spine to evaluate tick loads (T. Kreeger, Wyoming Game and Fish Department, Wheatland, Wyoming, USA, pers. commun.). Numbered, aluminum ear tags will be attached to each ear of all captured moose. Twenty-five to 30 males >2 years-of-age will be fitted with VHF radio transmitter collars (Advanced Telemetry Systems [ATS], Isanti, Minnesota, USA) and 20 females >2 years-of-age will be fitted with GPS radio transmitter collars (Telonics, Inc., Mesa, Arizona, USA). Oxytetracycline antibiotics (OxyCure 200, Vedco Inc., St. Joseph, Missouri, USA) will be administered to combat infection and Ivermectin (Ivomec, Merial Ltd, Iselin, New Jersey, USA) will be given to control internal and external parasites.

In addition, 10-20 females fitted with GPS radio transmitters will be recaptured during February 2006 to download GPS data and to resample physiological parameters. Additional males and females will be captured to maintain the original sample size of 25-30 and 20, respectively.

Transmitters

Twenty adult female moose in the Buffalo Valley will be monitored using Telonics TGW-3700 GPS radio transmitters with store-on-board technology (Telonics, Inc. 2004). These collars are preprogrammed to obtain a GPS location every hour from 15 November to 15 June and every 5 hours from 16 June to 14 November. GPS locations will be collected continuously until 1 March 2007 when the transmitters are scheduled to "drop-off" the moose via a preprogrammed breakaway device. GPS location data will be downloaded from the collars using the Telonics Data Download Utility (Telonics, Inc. 2004). The data will be managed via Microsoft Access and Excel programs. ArcView 3.3 (Environmental Systems Research Institute [ESRI], Redlands, California, USA) and ArcGIS 9.1 (ESRI, Redlands, California, USA) will be used to examine and analyze all location data.

Twenty-five to 30 adult male moose and 18 adult female moose (from the pilot study) will be monitored using ATS M2710 radio transmitters (ATS, Isanti, Minnesota, USA). VHF radio transmitters will be attached with a cotton spacer that will rot away in 2-3 years.

Resource Selection

To obtain accurate resource selection estimates for a population, Millspaugh and Marzluff (2001) suggest a sample size of ≥20 animals per season be monitored. A sample of 20 adult female moose will be captured and fitted with GPS radio transmitters. Location data collected from the GPS collars will be used to assess resource selection patterns by comparing resource use and resource availability at the individual home range level and at the study area level. Logistic regression (Hosmer and Lemeshow 1989) will be used to create a predictive model of summer and winter use in Geographic Information Systems (GIS) using certain habitat variables based on the first year of data. This model will be validated using the second year of location data and a final model will be created using all location data available. Habitat variables that have been identified as being important to moose and that will be used in the model include slope (Matchett 1985, Langley 1993), aspect (Matchett 1985, Langley 1993), elevation (Stevens 1970, Matchett 1985, Langley 1993), distance to water (Peek 1998), vegetation type (Houston 1968, Pierce 1984, Pierce and Peek 1984, Matchett 1985, Peek et al. 1987, Langley 1993, Van Dyke et al. 1985, Tyers 2003), percent canopy cover (Houston 1968, Stevens 1970, Peek et al. 1987, Peek 1998, Bowyer et al. 2003), and a winter severity index (Houston 1968). Slope, aspect, and elevation data will be obtained from Digital Elevation Models (DEMs) created for Wyoming at a 1:100,000 scale (Wyoming Geographic Information Science Center [WYGISC], Laramie, Wyoming, USA). Water layers will come from a statewide hydrography layer (1:100,000, WYGISC, Laramie, Wyoming, USA). Vegetation type and percent canopy cover layers will be obtained from the Grizzly Bear Cumulative Effects Model (Mattson and Despain 1985) that was developed for the GYE. A GIS layer of the most recent areas that have burned will be obtained from the Bridger-Teton National Forest. Vegetation types will be categorized into 6 classes based on primary cover type – non-vegetated, burned areas, open meadow/sagebrush, shrub dominated/riparian, deciduous forest, and coniferous forest. A winter severity index will be calculated, however, a specific method has not yet been identified. Research into this technique is ongoing.

Selection ratios will be calculated for each moose and for all moose combined to determine selectivity for or against vegetation types by season. For all home range analyses, the minimum convex hull method (Getz and Wilmers 2004) will be used to delineate seasonal home ranges for each moose and selection ratios with 95% confidence intervals will be calculated for each season. Comparisons of selection ratios will be performed to test for statistical differences ($P < 0.05$) in selectivity for each animal. In addition, comparisons will be made between animals, between seasons, and between years.

Study area analysis of selection ratios will permit population level inferences of habitat use by season. The study area that will be used for this analysis will be defined by the movement patterns of radio-collared moose; it will not be the geographic boundary that defined the study area previously. A minimum convex polygon will be created to encompass all moose locations and selection ratios with 95% confidence intervals calculated for each season. Comparisons of selection ratios will be performed to test for statistical differences in selectivity by season and between years.

A logistic regression model will be developed after the first year to create a predictive model of use based on resource selection function (RSF) values (Manly et al. 2002). Use will be defined as actual moose locations and availability will be derived from an equal number of systematic points with a random start generated by GIS. If a certain habitat variable is not significant ($P \geq 0.05$), that variable will be removed from the model and the model will be refit. Once a final, working model is determined, a predictive map of habitat use will be created in ArcGIS (ESRI, Redlands, California, USA) using RSF values scaled from 0 to 1.

Validation of the model will occur after retrieval of the radio transmitters in March 2007 using the BIN technique (T. McDonald, West Inc., Laramie, Wyoming, USA, pers. commun.) where a histogram will be created based on the RSF values of the predictive model. Each bar of the histogram will become a BIN and within each BIN RSF values will be averaged and totaled. The expected number of moose locations will then be calculated for each BIN. Then the second year of location data will be overlaid on the model and the observed number of moose locations within each BIN will be calculated. Finally, the number of observed locations within each BIN will be plotted on the expected number of locations and the slope of the line will be calculated. I will assume no year effect if the slope of the line is not significantly different from 1.0. If the slope is significantly different from 1.0, then a new predictive model of use will be created using the K-fold cross validation technique (Kutner et al. 2004; page 375). This will be accomplished by randomly selecting 75-80% of all moose locations (first and second year combined) to build the regression model and using the remaining 20-25% for model validation.

NOTE – due to issues with sampling independence, the use vs. availability analysis may be changed so that each moose is considered its' own sample unit and location data will be considered the sub sample. Using this technique, all location data can be used to examine resource selection and no data will be thrown out to adjust for independence.

Seasonal Home Ranges and Migration Routes

Seasonal home ranges and migrations of individual moose will be calculated using the minimum convex hull method (Getz and Wilmers 2004). This method provides improved estimates of core use areas and corridors than do kernel estimators as the quantity and quality of location data increases (e.g., GPS locations; Getz and Wilmers 2004). Locations from each animal will be separated into 2 seasons (summer and winter) and analysis of home ranges will occur via this seasonal separation. Migration between seasonal ranges will be assumed to have occurred when sequential moose relocations fall outside of the previously used seasonal range. Moose will be assumed to have arrived at its subsequent seasonal range when relocations fall within that seasonal home range. Migration routes will be denoted by those locations that fall outside of seasonal ranges and distances between seasonal ranges will be measured by calculating the straight-line distance between the home ranges.

If summer and winter home ranges overlap, moose will be considered non-migratory (Houston 1968, Langley 1993) seasonal ranges will be delineated by calculating the mean season dates for migratory moose. Non-migratory moose locations that fall within the mean seasonal range date distributions of migratory moose will be calculated as separate summer and winter ranges. Locations that fall outside of the mean seasonal date range distribution will be removed from seasonal range analyses and will be considered migration periods. This will permit meaningful comparisons between seasonal home range size of migratory and non-migratory moose. Annual home range sizes will also be compared between the 2 groups of moose.

CHAPTER II:

MOOSE POPULATION DYNAMICS

INTRODUCTION

Moose populations are not static; rather they fluctuate around an equilibrium that is also in constant change, based on density dependent and density independent factors. Density dependent factors work to regulate population growth while density independent factors may limit population growth. Population regulation can be defined as "density dependent factors that keep a population within its normal density range" (Ballard and Van Ballenberghe 1998). Limiting factors are defined as "those that alter the rate of population increase" (Ballard and Van Ballenberghe 1998). At any given time a population is influenced by numerous combinations of factors making inferences as to the mechanism(s) that limits or regulates populations very difficult to ascertain (Van Ballenberghe and Ballard 1998).

The primary factors that influence moose population size are reproduction and mortality. Throughout North America, annual adult female pregnancy rates averaged 84.2% (Boer 1992) and most adult females produce 1-2 calves per year (Schwartz 1998). Franzmann and Schwartz (1985) suspected that twinning rates were an indication of the nutritional status of the population. Schwartz (1998) suggested populations below carrying capacity had twinning rates as high as 90%, populations near carrying capacity had twinning rates between 5-25%, and populations above carrying capacity experienced twinning rates of less than 5%.

Data suggests that established Shira's moose populations experience lower reproductive output than other subspecies of moose, but the cause remains elusive (Schwartz 1998). In the Jackson Valley, Houston (1968) reported pregnancy rates of 90% and twinning rates of only 4.5% while Berger et al. (1999) observed pregnancy rates of 75% with 83% of those females producing young. In the Gravelly and Snowcrest Mountains of southwest Montana, Peek (1962) observed only 2 sets of twins during a 3-year study and in the Gallatin Mountains, Stevens (1970) reported twinning rates of only 2.5%.

Shira's moose are capable of increased reproductive output, but examples appear to be limited to expanding and/or translocated populations (i.e., populations well below carrying capacity). In YNP, Bailey (1930; as cited in Houston 1968) noted that between 1894 and 1917 twin moose calves were more common than singletons. In 1948, Denniston (1956) observed 15% twinning rates in the Jackson Valley and Shira's moose translocated to Colorado in 1978 and 1979 were observed with twinning rates of 17% in 1980 (Duvall and Schoonveld 1988).

The largest proportion of annual mortality occurs among calves within the first 6-8 weeks of life with predation accounting for the majority of losses (Franzmann et al. 1980, Ballard et al. 1981, Ballard and Van Ballenberghe 1998). Studies in Alaska, where moose were the primary prey species, have shown that grizzly bear and black bear predation can be significant sources of moose calf mortality (Franzmann et al. 1980, Ballard et al. 1981, Franzmann and Schwartz 1986, Ballard et al. 1990, Osborne et al. 1991, Schwartz and Franzmann 1991, Gasaway et al. 1992, Bertram and Vivion 2002, Bowyer et al. 2003). Grizzly and black bear predation rates ranged between 3-52% and

2-50% of young moose, respectively (Ballard and Van Ballenberghe 1998). Wolf predation on moose calves is most pronounced in the winter (Peterson 1977, Ballard et al. 1987, Gasaway et al. 1992).

In addition to predation, moose calves also die as a result of stillbirths, trampling or accidental smothering by cow, abandonment, starvation, exposure, disease, or accidents (i.e., drowning, impalement, entanglement, etc.; Child 1998). In south-central Alaska, 9% of annual calf mortality (Ballard et al. 1981) and 12% mortality within the first 6 months of life (Ballard et al. 1991) were directly attributed to miscellaneous factors including injuries inflicted by the cow, drowning, and pneumonia. While searching for newborn calves, Ballard et al. (1991) found that only 3 of 221 calves were stillborn or died immediately after birth. In western interior Alaska, Osborne et al. (1991) reported that 5% of moose calves died from causes other than predation and on the Kenai Peninsula, Alaska, Franzmann and Schwartz (1986) determined that 5% of total calf mortalities were attributed to natural abandonment, drowning, or unknown causes. For older aged calves, (6-12 months), Ballard et al. (1991) reported that starvation accounted for 79% of all mortalities.

Adult annual survival rates are generally high, ranging from 75% in a population subjected to intensive predation and hunting (Hauge and Keith 1981) to 92% where wolves and hunting were the primary mortality factors (Bangs et al. 1989). Wolves (Peterson 1977, Bertram and Vivion 2002) and grizzly bears (Boertje et al. 1988, Bertram and Vivion 2002) are the primary predators of adult moose. Wolves generally prey on older aged adults that may (Peterson 1977) or may not (Ballard et al. 1987) be in poor physical condition. Mountain lions may also prey upon juvenile and adult moose

(Kunkel et al. 1999; Anderson and Lindzey 2003, R. Jaffe, Beringia South, Kelly, Wyoming, USA, pers. commun.). Black bears may prey on adult moose but are not considered significant predators (Ballard 1992). Adult moose also die as a result of accidents that may include drowning, falls, entanglement, and sparring to name a few (Child 1998).

Moose are hosts to a variety of pests, parasites, and infectious diseases that may not directly contribute to mortality, but can exacerbate an underlying condition resulting in death. The winter tick (*Dermacantor albipictus*) is perhaps the most common moose parasite throughout much of North America (Lankester and Samuel 1998, Samuel 2004). Moose in poor physical condition may die as a result of large tick infestations due to anemia, damage to and/or loss of winter coat, reduced stored visceral fat, and decreased feeding as a result of increased grooming (Lankester and Samuel 1998, Samuel 2004).

Using the "animal indicator concept" proposed by Franzmann (1985), I will estimate overall body condition of each captured moose via a suite of physiological measurements that can be used to describe the status of the population with respect to carrying capacity of its habitat. Many of these physiological indices ultimately drive reproductive rates, adult and calf survival, and influence recruitment. Thus, if moose are limited by habitat conditions, signs of nutritional stress and decreased reproductive output should be evident. If, on the other hand, predation has increased, reducing moose densities, habitat conditions should be improving and would be reflected in overall health and reproductive parameters.

GOALS, HYPOTHESES, AND OBJECTIVES

Research Goal

In this study, I propose to estimate population parameters and measure physiological health indices of adult moose to determine where the population is with respect to habitat carrying capacity. I will also use this information to assess potential causes for the recent population decline observed in the Jackson moose herd.

Hypothesis

H_{o1}: Physical condition of adult female moose will be intermediate suggesting predation and forage contribute equally to moose population decline

H_{a1}: Physical condition of adult female moose will be relatively good or poor suggesting either predation or forage, respectively, may contribute more strongly to moose population decline

H_{o2}: Adult male and female survival rates will be relatively high suggesting that adult survival is not limiting the population

H_{a2}: Adult male and female survival rates will be relatively low suggesting that adult survival is limiting the population

H_{o3}: Pregnancy and parturition rates of adult female moose will be relatively high suggesting that reproduction is not limiting the population

H_{a3}: Pregnancy and parturition rates of adult female moose will be relatively low suggesting reproduction is limiting the population

H_{o4}: Calf survival will be relatively high suggesting that survival of this age class is not limiting population growth

H_{a4}: Calf survival will be relatively low suggesting that survival of this age class is limiting population growth

Objectives

1) Estimate monthly and annual adult male and female survival rates for moose in the north Jackson moose herd
2) Estimate pregnancy and parturition rates of female moose in the north Jackson moose herd
3) Measure physiological health parameters from blood samples, hair samples, and rump fat measurements
4) Estimate calf survival and recruitment to yearling age class

METHODS

Adult Survival Rates

To obtain accurate survival rate estimates for a population, Powell et al. (2000) and Millspaugh and Marzluff (2001) suggest a sample size of at least 25 animals per cohort and season be monitored. Eighteen adult females are currently being monitored with VHF radio transmitters, 20 additional adult females will be captured and monitored using GPS radio collars, and 25-30 adult males will be monitored using VHF collars. Sex specific seasonal and annual survival rates will be evaluated using Kaplan-Meier Analysis techniques (Pollock et al. 1989, Van Ballenberghe and Ballard 1998, Millspaugh and Marzluff 2001). This method accounts for animals that are lost due to radio failure, slipped radio collars, or emigration and allows for staggered entry of new study animals (i.e., all animals do not have to be captured at the same time for accurate survival estimates). To enhance survival rate estimates, hunt areas 7, 14, and 32 in the Jackson Moose Herd Unit have been closed for the duration of the study.

Adult bull and cow survival will be evaluated using ground and monthly fixed-wing radio telemetry techniques. Ground surveys will be used to monitor radio-collared moose for active or inactive signals. Most ground surveys will be conducted from a truck or on foot during the winter months (December to May) when moose are concentrated on winter range. If a moose is observed, general condition, location, and presence or absence of calf (if female) will be recorded. Aerial surveys using fixed-wing aircraft (Mountain Air Research, Inc, Driggs, Idaho, USA) will be used to monitor moose monthly for the duration of the study.

No attempts will be made to obtain specific locations of moose unless a mortality signal is evident. When a mortality signal is received, the location of the signal will be located as soon as possible to determine if it is a faulty radio transmitter, a dropped radio collar, or a mortality. Attempts will be made to observe and/or monitor movements of moose with ground surveys to determine if the animal is still alive. The fixed-wing pilot will attempt to obtain a visual of the animal during each successive flight until an active signal is heard or until the animal has not moved for 2 consecutive flights. If a dropped radio collar is found, it will be retrieved, a GPS location will be taken, and habitat type will be recorded. If cause of the signal is a mortality, attempts will be made to identify the specific cause of mortality using field necropsy techniques. General location data, scavenger/carnivore sign present at carcass, utilization of carcass, general condition of moose via examination of femur marrow categorized into three broad classes (Peterson 1977), presence or absence of body fat, arthritic joints, and/or any other bone abnormalities will be recorded. If available, tissue samples will be collected and analyzed by the Wyoming State Veterinary Laboratory to determine if the moose had any underlying health conditions that may have contributed to death. Two incisors will be collected and sent to the Wyoming Game and Fish Department Forensic Laboratory for age determination. A "kill evaluation and categorization chart" prepared by Toni Ruth (Wildlife Conservation Society, Gardiner, Montana, USA) and Rose Jaffe (Beringia South, Kelly, Wyoming, USA) (Appendix A) will be used to record possible, probable, or positive cougar, wolf, or bear (black and grizzly) involvement. Also, principal investigators of the cougar and wolf projects will be contacted to determine if any of their radio-collared animals were in the area around the time of the suspected mortality event. Photographs of the carcass and kill site will be taken.

Dates of death for moose fitted with GPS radio transmitters will be estimated by downloading the radio transmitters, analyzing the location data, and locating the first known location in a point cluster near the UTMs where the carcass was found. For moose fitted with VHF radio transmitters (or if no point clusters or carcass was found), the date of death will be estimated from the midpoint between the last time the signal was heard on active and the first time the signal was heard on mortality. If the radio transmitter is in good condition after retrieval, it will be placed on another moose as soon as possible to maintain sample size; otherwise, collars will be refurbished and reattached.

Pregnancy and Parturition Rates

Blood samples collected at time of capture will be used to determine pregnancy status of the 20 adult females captured in February 2005 and 10-20 recaptured females in 2006. Samples will be sent to Bio Tracking in Moscow, Idaho and radio immunoassays

(RIA) will be conducted to detect concentrations of the pregnancy hormone progesterone. Progesterone levels greater than 3.0 ng/mL indicate a pregnant animal whereas progesterone levels of 0.5 ng/mL or less indicate a nonpregnant animal (Schwartz 1998). Results of blood RIA pregnancy tests are extremely accurate, but false positive results may occur if an embryo dies shortly before sampling or if the embryo dies after sampling (C. Schwartz, Interagency Grizzly Bear Study Team, Bozeman, Montana, USA, pers. commun.). A 2 x 2 Chi-square contingency analysis will be used to examine pregnancy rate differences between years ($P < 0.05$).

Fecal samples from radio collared female moose will also be collected and analyzed for fecal progesterone concentrations to determine if in utero losses occur prior to parturition. Monfort et al. (1993) and Schwartz et al. (1995) suggest that 3 fecal samples be collected over a 2-3 week period from each individual to reduce the likelihood of false positive results. Female moose will be located using ground telemetry techniques, and once observed, tracks will be backtracked until a fecal sample is located. If backtracking becomes too difficult due to the number of tracks in an area, moose will be observed until defecation occurs. Fecal samples will be collected in March and April of each year and frozen until analyzed. Fecal samples will be sent to Dan Baker (Colorado Division of Wildlife, Fort Collins, Colorado, USA) or Steve Monfort (Smithsonian Institution, Washington, D.C., USA) for hormone extraction and analysis.

Parturition rates will be assessed using ground and helicopter surveys (Soloy Bell 47, Savage Air Services, American Falls, Idaho, USA) during the first and second week of June in 2005 and 2006 (Franzmann and Schwartz 1986, Osborne et al. 1991, Van Ballenberghe and Ballard 1998, Bertram and Vivion 2002). A telemetry flight using fixed-wing aircraft will be flown 1-2 days prior to parturition surveys to expedite helicopter surveys. All females will be surveyed for presence or absence of calves to verify blood assay and fecal progesterone pregnancy results acknowledging that some in utero and/or neonate mortalities may occur prior to surveys. Ground surveys of females that were not located during aerial surveys and that are accessible from the ground will be attempted using hand-held radio telemetry equipment. Collar frequency, general location, UTM locations, number of calves observed, habitat type, and comments on behavior of moose will be recorded on the parturition data sheet.

GPS data from female moose will also be used to verify birthing success or failure and timing of birthing events. Female moose restrict their movements for the first few weeks of the calf's life, and then gradually expand their movements as the calf gains strength and coordination (Langley and Pletscher 1994, Bubenik 1998b, Bowyer et al. 1999). Using this knowledge, GPS data can be analyzed using the Animal Movement extension in ArcView to look for the above characteristics in females that were observed with calves. If similarities in movement patterns are found, this can be applied to females that were not observed with calves to examine the possibility that these females also successfully gave birth, but lost the calf prior to or shortly after the parturition surveys. Females that successfully give birth to a calf, but then lose it soon after may show a localized point cluster followed by a long range movement in a short time period (C. Schwartz, Interagency Grizzly Bear Study Team, Bozeman, Montana, USA, pers. commun.; J. Berger, Wildlife Conservation Society, Victor, Idaho, USA, pers. commun.). Movements of females that were not known to be pregnant and were not observed with calves can also be compared to the above scenarios to determine similarities or differences in movement patterns.

Physiological Health Parameters and Disease

Blood samples collected at time of capture will be examined for (1) serum chemical analyses (Vetex, Alfa Wasserman, West Caldwell, New Jersey, USA), (2) hematological analyses (Hemavet 850FS, Drew Scientific, Oxford, Connecticut USA), and (3) disease antigens. Franzmann (1978, 1998) noted calcium (Ca), phosphorous (P), total protein (TP), packed cell volume (PCV; percentage of red blood cells), and hemoglobin (Hb) concentrations have the greatest value in determining physiological health in moose and will be evaluated more critically. However, blood values alone are not good indicators of physical condition in moose (Ballard et al. 1996, Keech et al. 1998). Thus, data will be used in conjunction with other physiological variables to determine overall herd health.

Disease analyses will be conducted by the Wyoming State Veterinary Laboratory by testing blood samples for certain disease antibodies. Moose will be tested for *Brucella*, *Leptospira*, infectious bovine rhinotracheitis virus, bovine viral diarrhea virus, parainfluenza-3 virus, and bovine respiratory syncytial virus. A positive test may suggest that the moose is presently infected with the disease or that it survived the infection and antibodies are in the blood to protect against reinfection (Lankester and Samuel 1998).

Parasitology will be analyzed from fecal samples and ear swabs collected at time of capture by the Wyoming State Veterinary Laboratory. The fecal flotation technique will be used to look for eggs and oocysts of internal parasites and flukes. Ear swabs will be collected to determine mite presence. In addition, a 30 second tick count will be performed along the spine of each moose to quantify low, moderate, or high tick loads at time of capture. Observations of individual moose periodically throughout late winter will also be used to determine the amount of tick parasitism by noting (1) the amount of hair loss, (2) the amount of hair turning white, and (3) the general location of hair loss or white hair (Samuel 2004).

Hair collected from the hump of female moose during capture efforts will be used to examine concentrations of macro- and micro-elements. Low concentrations of some of these trace elements have been linked to decreases in reproduction in moose (Flynn et al. 1977) and elk (C. Whitman, Interagency Grizzly Bear Study Team, Bozeman, Montana, USA, pers. commun.).

Body condition will be assessed by ultrasonic measurements of rump fat depth. The rump region will be scanned using an Omega I Portable Ultrasound unit (E.I. Medical, Loveland, Colorado USA) with a 5 MHz 8-cm linear-array transducer. Prior to scanning, hair on the measurement site will be shaved using hand-held, surgical grade animal clippers and a water-based gel will be applied frequently to the exposed skin to ensure air-free contact between the transducer and the skin. Subcutaneous fat thickness will be measured with electronic calipers to the nearest 0.1 cm at the midpoint between the coxal tuber (hip bone) and the ischial tuber (pin bone) immediately adjacent to the spine (Stephenson et al. 1993, 1998, 2002). When a usable image is observed, it will be frozen on the screen of the ultrasound unit and the measurement will be taken. Images will be saved to a memory card in the event that further analysis is warranted. Due to the time involved and the speed of the capture event, ultrasonic rump fat measurements will not be made on every moose captured. Therefore, moose will be opportunistically sampled as time allows using a single ultrasound device. For winter captures, rump fat depths of 2 cm or greater indicate an animal in good or excellent condition and measurements below 2 cm may indicate an animal under nutritional stress (T. Lohuis, Director, Kenai Moose Research Center, Alaska Department of Fish and Game, Soldotna, Alaska, USA, pers. commun.).

Franzmann (1977) developed a set of condition evaluation criteria (Table 1) that are used to assess condition of individual moose. This technique is more subjective and involves palpating the animal and feeling for certain characteristics which are an indication of the animals condition. Based on the results of the palpations, a score from 10 to 0 will be assigned to the animal. Scores of individual moose will be compared to measurements of rump fat to determine if there is any correlation between the 2 body condition indices.

Regional, seasonal, and age variations in physiological health parameters exist between moose populations (Flynn and Franzmann 1987, Ballard et al. 1996, Keech et al. 1998). Therefore, measurements of physiological condition will be used to identify extremes (low or high) within the population for each measurement taken or sample collected (Franzmann et al. 1987). If physiological measurements of condition fall within normal ranges for moose populations (as measured by studies throughout North America, e.g., Houston 1969, Franzmann and LeResche 1978, Franzmann et al. 1987, Franzmann 1998, Keech et al. 1998), no further analysis will be warranted. However, if mean values for certain physiological variables fall outside normal ranges, further investigation into possible causes may be warranted, although that will fall outside the realm of this study. Due to regional, seasonal, and age differences associated with physiological data,

Table 1. Condition evaluation criteria used to assess general condition of moose (Franzmann 1977).

Score	Description
10	A prime fat moose with thick, firm rump fat by sight. Well fleshed over back and loin. Shoulders round and full.
9	A choice, fat moose with evidence by feel of rump fat. Fleshed over back and loin. Shoulders round and full.
8	A good fat moose with slight evidence by feel of rump fat. Bony structures of back and loin not prominent. Shoulders well fleshed.
7	An "average" moose with no evidence of rump fat, but well fleshed. Bony structures of back and loin evident by feel. Shoulders with some angularity.
6	A moderately fleshed moose beginning to demonstrate one of the following conditions – (a) definitions of neck and shoulders, (b) upper foreleg musculature distinct from chest, or (c) prominent rib cage.
5	When 2 of the characteristics in class 6 are evident.
4	When all 3 of the characteristics in class 6 are evident.
3	When hide fits loosely about the neck and shoulders, head is carried at a lower profile. Walking and running postures appear normal.
2	Malnutrition obvious. Outline of the scapula is evident. Head and neck low and extended. Walks normally, but trots and paces with difficulty and cannot center.
1	Point of no return. Generalized appearance of weakness. Walks with difficulty and cannot trot, pace, or canter.
0	Dead from malnutrition/starvation.

information will be presented as means with standard deviations and only generalized comparisons will be made between results of this study and other studies.

Calf Survival and Recruitment to Yearling Age Class

Calf survival will be assessed using ground and helicopter surveys (Soloy Bell 47, Savage Air Services, American Falls, Idaho, USA) approximately 8-9 weeks post parturition (around the third week in July) in 2005 and 2006. All females will be surveyed for presence or absence of calves to verify parturition surveys conducted in early June. In addition, helicopter surveys flown in conjunction with WGFD winter trend counts in mid-February will be used to assess calf recruitment to yearling age class. Ground surveys of females known to have calves will also be initiated at this time to monitor survival of calves through late winter using hand-held radio telemetry equipment. Survey protocols will follow methods described for parturition surveys.

GPS collar location data may also be used to investigate calf survival and timing of possible calf mortality events. C. Schwartz (Interagency Grizzly Bear Study Team, Bozeman, Montana, USA, pers. commun.) and J. Berger (Wildlife Conservation Society, Victor, Idaho, USA, pers. commun.) have observed long-range movements of female moose shortly after the death of a calf. Using this knowledge, GPS location data can be analyzed using the Animal Movement Extension in ArcView to look for similarities between movement patterns of female moose that were known to have lost a calf. Also, if a female appears to have 2 distinct core use areas within a home range, timing of use of each core area can be analyzed, and if no overlap exists it may be that she lost a calf and moved to another location soon after. Even with the use of GPS technology and the knowledge of female moose behavior, it may be difficult to quantify actual timing of calf mortalities because of variations in individual behavior. Calf survival will be examined using change in ratio analysis (C. Schwartz, Interagency Grizzly Bear Study Team, Bozeman, Montana, USA, pers. commun.) from (1) birth to calf survival surveys, (2) from calf survival surveys to trend count surveys, (3) from trend count surveys to April just prior to moose leaving the winter range, and (4) annually.

CHAPTER III:

MOOSE HABITAT USE AND MOVEMENT RELATIVE TO US HIGHWAY 26/287

INTRODUCTION

Moose-vehicle collisions (MVC) are a significant cause of mortality for some moose populations in North America (Child et al. 1991, Del Frate and Spraker 1991, McDonald 1991, Oosenbrug et al. 1991, Child 1998). In a survey compiled by Child (1998), approximately 3,000 MVC are reported annually in North America; however numbers increase if train collisions and unreported collisions are factored in. The majority of collisions occur (1) at dawn and dusk, (2) under poor light conditions or decreased visibility from encroaching vegetation, (3) on straight, relatively flat stretches of highway, (4) at speeds in excess of 80 km/hr, and (5) because of driver inattention (Child 1998). In Sweden, Seiler (2004) reported that MVC were highest at moderate vehicular speeds (approximately 90 km/h), at moderate traffic volumes (2,000 – 4,000 average vehicles per day), and where moose densities were the highest. In British Columbia, Child et al. (1991) reported that peak MVC occurred from late November through the end of January and Del Frate and Spraker (1991) noted that MVC peaked in December and January on the Kenai Peninsula, Alaska. Lavsund and Sandegren (1991) reported that peak MVC occurred in early summer and autumn in southern Sweden while in northern Sweden, MVC peaked in December and January.

Spatial and temporal patterns of moose movement and activity can be affected by the construction and/or maintenance of transportation corridors. Roadways tend to create successional habitats that are favored by foraging moose (Child 1998). Many roadways, especially in mountainous terrain, follow the path of major waterways that may bisect critical moose winter range (McDonald 1991) limiting the animals' ability to travel between winter ranges. This ultimately poses a hazard to moose and motorists when moose attempt to cross roadways. In areas of deep snow, transportation corridors offer easy travel routes (Modafferi 1991). Maintenance of roadways in winter may lead to increased vehicular speeds, even under poor driving conditions. Del Frate and Spraker (1991) noted that moose-vehicle collisions almost doubled following the Alaska Department of Transportations new winter road maintenance policy which increased plowing and sanding of roads after snowstorms to make them safer for motorists. Salting of roadways to prevent icing results in artificial "licks" that attract moose (Child 1998). Snowplowing and the creation of high snow banks on either side of the roadway limits the ability of moose to escape oncoming traffic.

Habitat parameters associated with roadways can also be analyzed to predict the likelihood of MVC along roadways. Malo et al. (2004) used regression analysis to predict the likelihood of animal-vehicle collisions (AVC) in Spain. The authors reported that the probability of AVC increased with increasing habitat diversity and high forest cover with a reciprocal decrease in agricultural cover and number of buildings. Finder et al. (1999) also used regression analysis to identify important habitat components that could be used to explain white-tailed deer-vehicle collisions (WDC) in Illinois. It appears from these studies that by analyzing variables important to the study animals' habitat, one can accurately predict sections of roadways that have high probabilities of AVC.

Many different techniques have been used to mitigate MVC with varied success (Child 1998). Road signs and public awareness programs are the least costly and are, perhaps, the easiest to implement (Oosenbrug et al. 1991, Del Frate and Spraker 1991), however, their effectiveness has been variable. Vegetation removal and highway lighting can increase a motorist's ability to anticipate potential hazards (Del Frate and Spraker 1991). Lavsund and Sandegren (1991) noted a 20% decrease in MVC by removing vegetation within 20 m of a roadway, but they concluded that the MVC reduction could also be attributed to chance variation. Vegetation removal is also very expensive and must be maintained often (Lavsund and Sandegren 1991). More permanent measures include moose-proof fences with or without one-way gates, underpasses, or overpasses. McDonald (1991) compared MVC prior to (no MVC mitigation techniques) and after highway expansion (with MVC mitigation techniques) in southeast Alaska. He reported that moose-proof fencing, one-way gates, an underpass, and highway lighting reduced MVC by 70% overall and by 95% within the fenced portions of the highway. However, fencing and maintenance of fence structures are very expensive and may only be practical in sections of roadway with high traffic volumes (Schwartz and Bartley 1991). Regression models developed by Seiler (2004) reported that reduced vehicle speed, fencing, and vegetation removal along roadways provided the strongest evidence for reducing MVC. Driver behavior is perhaps the most difficult mitigation measure to reduce MVC (Lavsund and Sandegren 1991).

Identifying important crossing locations and timing of crossing events is crucial to the understanding of animal movements in human dominated environments. I will employ RSF analysis techniques to predict locations of moose crossing events. The model will aid the Wyoming Department of Transportation in identifying areas where moose crossings are likely to occur. By examining these areas, recommendations will be made that can be incorporated into existing roadway design to make this section of highway 26/287 safer for motorists and moose alike.

GOALS, HYPOTHESIS, AND OBJECTIVES

Research Goal

In this study, I propose to estimate the timing and location of adult female moose movements associated with highway 26/287 which bisects the Buffalo Valley moose winter range.

Hypothesis

H_{o1}: Moose crossing events will be evenly distributed throughout the day

H_{a1}: Moose crossing events will occur most frequently at dawn and dusk

H_{o2}: Moose will choose highway crossings randomly

H_{a2}: Moose will select highway crossings relative to vegetation structure and type and highway structure

Objectives

1) Estimate the timing and location of moose movements across highway 26/287

2) Create a detailed model of adult female moose resource selection patterns associated with highway 26/287 and use this to predict important moose crossing locations
3) Evaluate and recommend mitigation that can be incorporated into existing roadway design that may reduce the likelihood of MVC

METHODS

Using location data downloaded from GPS radio transmitters, the timing and location of moose crossings between milepost 3.0 and 9.0 along highway 26/287 will be identified. Habitat features and highway structures will be incorporated into the analysis in an attempt to determine where moose select highway crossings. In addition to the GIS layers used to create the predictive resource selection model, a road layer (1:100,000) depicting all roads in the area will be downloaded from WYGISC and a layer depicting fence types along the highway will be created.

Finder et al. (1999) created a 0.8 km buffer around sections of road to evaluate habitat components that were important in predicting locations of white-tailed deer crossings in Illinois. Thus, to create a study area specific to highway 26/287, a 0.8 km buffer will be applied to the section of highway between milepost 3.0 and 9.0. Analysis techniques used to evaluate resource selection patterns of adult female moose along highway 26/287 and validation of the model will follow methods described above under the Resource Selection section. The only exception is that moose locations that fall outside of the 0.8 km buffer surrounding the highway will not be used in this analysis. In addition, moose crossing data collected by West, Inc. (Laramie, Wyoming, USA) along this same section of roadway will be evaluated and may be used to validate the predictive model if the quality and quantity of data is sufficient. High and low RSF values will be calculated by averaging RSF values across all moose locations that fall within the highway study area. High RSF values will then correspond to values greater than or equal to the average and low RSF values will fall below the average. A chi-square analysis will be used to examine differences between moose crossing areas.

NOTE – if analysis of resource selection is changed to the technique described in the Resource Selection section, techniques used to analyze this section will also change.

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APPENDIX A. Kill Evaluation and Categorization Chart

Carnivore kill:

1) Sign of struggle evident
    a) Scuff or track evidence of chase and struggle
    b) Blood on ground from pursuit
    c) Broken branches
    d) Blood on trees
2) SubQ hemorrhage on hide/carcass
3) Aspirated blood in trachea, mouth, nose

Carnivore	Possible	Probable	Positive
Cougar	Old cougar tracks discernable by shape and stride pattern only Ungulate prey hair in small "clumps" around carcass has appearance of being sheared off or cut near root base Feces covered in "toilet" but dry or chalky in appearance Toilets, scrapes, bed sites not apparent or appear "weathered" Carcass remains in dense cover, small drainage or side draw	Fresh cougar tracks discernable by detailed toe and pad arrangement Carcass remains concealed near tree and/or brush Remains buried/cached with light debris such as plucked hair, small sticks, leaf-needle-soil duff. During winter, caches may be made completely of snow packed on top of carcass. Cougar feces dark, moist, or liquid Toilets and/or scrapes appear recently created Tree scratching at site 2-4 bed sites oriented upslope from carcass and concealed at base of tree, against boulder or rock outcrop Bed sites contain cougar hair Rumen rejected as food, possibly buried/cached Presence of cougar via visual, tracks, or radio location	Canine punctures to back of neck or at throat Hemorrhage to back of neck or at throat near jawline Canine holes measure 4.5 – 5.0 cm apart for top canine punctures and/or 3.0 – 4.0 cm apart for bottom pair Claw marks or claw tracks apparent along neck, shoulders, or back Drag marks to cache/concealment site with ungulate prey hair along drag line and blood Kill not scavenged by bear or wolf Evidence of ambush and/or struggle
Wolf	Old wolf tracks discernable by shape and stride pattern only Carcass remains in open habitat with ≤ canopy cover Feces in open and not covered; feces dry or chalky in appearance, or no feces present at all	Fresh wolf tracks discernable by detailed toe and pad arrangement Carcass remains scattered possibly > 300 m from kill site No bed sites evident If bed sites evident, generally more than 2-4 bed sites oriented in "spoke-wheel" fashion around kill site; bed sites in open on grass or under canopy cover, but not necessarily at base of tree Bed sites contain wolf hair Radio-collared wolves at or in vicinity of kill	Hemorrhage apparent on hide at back of metatarsus and femur areas If any hide or organs left to examine on head or throat – canine punctures to throat on cows and calves No cougar or bear sign at site
Bear	Old bear tracks discernable by shape or as depressions in soil only Carcass may be in open (generally grizzly) or forest cover (generally black) **NOTE**: bears in YNP scavenge carcasses (winter kill, cougar and wolf kills) during spring, so make sure no other carnivore sign is evident at the site. Bear predation in YNP is generally directed at calves and fawns where most of the carcass is entirely consumed	Recent bear tracks that show track details Moist/wet bear scats in vicinity or on outer perimeter of bed sites Bed sites contain bear hair Grizzly or black bear hair on antlers, trees, or brush Carcass buried with large amount of material including large sticks and dirt; area has churned or rototilled appearance indicative of grizzly Carcass in tree cover or draw, but not cached – indicative of black Hide on carcass is inverted over the head and down legs resulting in a "banana-peel" appearance Viscera consumed as food	Bear sign only; no other cougar or wolf sign present Broken neck-rib or occipital condyle/cervical vertebrae Extensive bruising on back of hind quarters, ribs, and/or shoulders Bite marks to spine behind shoulders

Badger Creek – Hanging Woman Fawn Predation Project

January 13, 2006

Dear Hank,

The antelope classification was completed in August and the mule deer classification was completed in late October or early November. I contacted Tim Thomas, the biologist from Sheridan, who did the flying and classification. He flew with Bighorn Airways. We received the bills from both studies, but the information has not been submitted to the Sheridan County PAB.

The first week in December, since we have not had proper flying weather, I had two guys do some ground work. There was no charge for their work. The results were:

one adult male - empty stomach
one adult female - full stomach - mainly rabbit
two young males - both full of mice, some rabbit
one young male - mice in stomach
one three legged coyote unsure of sex - had mange, did not check for stomach contents

We are waiting for proper flying conditions. I have Bob at Bighorn Airways ready to fly at minutes notice. Helicopter is ready and so are we. This is all the information I have at this time.

Cole Benton

Wyoming Game & Fish Dept.

On August 13, 2005, we flew for 2.9 hours in the Bell Jet Ranger helicopoter conducting a pronghorn herd composition or classification survey. The survey was flown along north-south transects at approximately 4 km (2.48 mi) apart. There were two observers and the pilot. We observed the following:

Badger Creek
2 yrl males
30 adult males > 1.5 yrs old
75 adult females > 0.5 yrs old
64 juveniles
171 total pronghorn

Hanging Woman Creek
12 yrl males
32 adult males
56 adult females
36 juveniles
136 total pronghorn

As we discussed at the November 11, 2005 Sheridan Predator Board Meeting, I suggest we do not continue the pronghorn portion of this study since the landowners in the Badger Creek drainage have significantly increased harvest of pronghorn.

On November 15, 2005, we flew the mule classification survey for this study. This flight was in conjunction with regularly schedule deer surveys in hunt area 23. We flew for 3.2 hours in a Bell Jet Ranger along north-south transects at ~4 km (2.48 mi) intervals. There were two observers and the pilot. We observed the following:

Badger Creek
13 yrl males
40 adult males
116 adult females
91 juveniles
261 total mule deer

Hanging Woman Creek
2 yrl males
11 adult males
57 adult females
44 juveniles
114 total mule deer

I have not had time to do look at the data in any manner. Let me know if this is enough information.

TIM

TIM THOMAS
Sheridan Wildlife Biologist
Wyoming Game & Fish Dept
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Sheridan, Wyoming 82801
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DNA Analysis for control of predatory wolves

United States Department of Agriculture

Animal and Plant Health Inspection Service

Wildlife Services

National Wildlife Research Center
4101 LaPorte Ave
Fort Collins, CO 80521

Phone: (970) 266-6082
Fax: (970) 266-6089
Email: john.j.johnston@usda.gov

December 4, 2005

Animal Damage Management Board
2219 Carey Avenue
Cheyenne, WY 82002-0100

Subject: Mid-term Progress Report DNA - Wolves

Dear ADMB:

I appreciate WYADMB's funding of my request for **DNA ANALYSIS FOR CONTROL OF PREDATORY WOLVES**.

Objective 1: To identify practical procedures for obtaining and analyzing DNA collected from predation events in WY

The ultimate application of this research to apply the analysis of DNA isolated from predator saliva, blood, feces and/or hair collected from predated carcasses to identify the species, sex and/or individual genotype of the predator. This information will be applied to the developing an increased understanding wolf predation in WY and will help the Wildlife Services Program develop better techniques to minimize wolf predation and to identify, track and selectively remove predatory wolves. The results of this project should permit us to develop practical predator sample collection protocols and to determine the value of the information provided by DNA analyses for management of politically sensitive predators such as wolves.

Progress through 11-2005:

Objective 1: To identify practical procedures for obtaining and analyzing DNA collected from predation events in WY

- Ph.D. Molecular Geneticist Toni Piaggio has been hired by NWRC. Dr. Piaggio has significant experience applying DNA forensics to wildlife issues for state and federal wildlife agencies.

- 117 wildlife samples have been collected by WY and 36 by MT wildlife services and shipped to NWRC molecular genetics laboratory.
- NWRC molecular genetics laboratory is currently extracting DNA from these samples
- The position description for the molecular genetics technician has been created and approved. The position is slated to be advertised in early December.

If you require any additional information, please contact me at john.j.johnston@usda.gov or 970-266-6082. I appreciate your support of our research.

Sincerely,

John J. Johnston Ph.D. MBA
Chemistry Project Leader, NWRC

Preemptive human/bear conflict mitigation

FY06 PROJECT UPDATE
JULY 1, 2005- DECEMBER 1, 2005

Project Title: Preemptive Human/Bear Conflict Mitigation

Brief Synopsis of project: This project will minimize human/bear conflicts in the North and South Forks of the Shoshone River area through (1) minimizing and properly managing unnatural bear attractants; (2) employing bear-resistant waste management systems; (3) managing bears/attractive bear habitat where potentials for conflict and risks to human safety are high; and (4) employing a public outreach program to reduce knowledge gaps about bears and conflicts.

Project Update: The funds ($3,000) provided by the ADMB have been used in providing several different prototypes of bear-resistant waste containers to local sanitation companies. The sanitation companies will distribute these containers to local residents in the North and South Forks areas to determine which containers are preferred. All containers purchased will continue to be used in the future, but it is necessary to obtain preference of the containers by both sanitation companies and residents using them before purchasing bulk numbers of containers. A coordinator has been hired to organize the program and will be performing a needs assessment in the area for management of other unnatural attractants. Informational presentations and meeting have been set up with county government officials, school administrators, select federal government employees, and residents to begin the education and outreach program for the project.

Submitted by: Dana Courtney, Bear Wise Community Coordinator

Affiliation: Wyoming Game and Fish Department
Mailing Address: 2820 State Hwy 120
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Rabies and stomach contents component of plague project

USDA

United States
Department of
Agriculture

Animal and
Plant Health
Inspection
Service

Wildlife
Services
Western Region

USDA, APHIS, WS
6731 W. Coal Rd.
P.O. Box 59
Casper, WY 82602
307/261-5336
Fax: 307/261-5996

Subject: Rabies and Stomach Contents Component of Plague Project (Jamie Snow, WY. Dept. of Health).

Date: 11/29/2005

To: Hank Uhden, (WY. ADMB).

The progress to this point on the rabies and stomach contents component attached to the Plague Project is not complete at this time. We have collected 100 coyotes each in Niobrara, Weston, and Goshen Counties (300 total taken). The remainder of the study will consist of collecting an additional 100 coyotes from Platte County. This should be completed by March 31st, 2006, barring unfortunate weather conditions for aerial hunting.

We have submitted a total of 150 coyote heads (50 samples from each of the 3 counties completed) for rabies sampling with no positive animals found. There will be an additional 50 submitted for Platte County when it is completed. Also, when Platte County is completed, we will do a cumulative stomach contents analysis that will encompass the total number of coyotes taken during the whole project which will be 400 total.

If you or any members of the ADMB have any questions please don't hesitate to call.

Sincerely,

Craig Acres
Eastern District Supervisor
Cc: Files

Sage Grouse Predation studies

Sage Grouse Field Technician Job Responsibility Summary and Results of the Pinedale Local Work Group Predator Study

From March 23, 2005 to May 17, 2005, I assisted a University of Wyoming, Wildlife Division, Graduate Student, with his project of determining the impact of gas drilling on Sage Grouse in the Pinedale, WY area. My duties included placing data collectors with car batteries at 14 leks, replacing and re-charging the batteries weekly, trapping live Sage Grouse hens to be radio collared, and counting Grouse using the 14 leks during mating season.

Starting June 3, 2005, I assisted with the vegetative inventory for 42 Sage Grouse nest sites that were either hatched successfully or predated prior to hatching. The vegetative inventory was completed within a week and a half. A follow-up on 30 of the 42 nest sites was conducted to verify the original inventory was accurate, was completed approximately 3 weeks later. The follow-up took 2 ½ days.

The predator study started on May 13th with the first camera being set up on an actual (live) Sage Grouse nest near the lek referred to as 4-2. It was decided to remove the camera the nest day due to the fact the nest was within 40yards of a somewhat frequently traveled drillers road and the camera's visibility made it a high risk to be stolen or vandalized.

Two live nests were identified in the lek area referred to as Shelter Cabin. The one nest had two talon cameras set up at the site and the other with one. I went back to the nests when it was determined with radio telemetry equipment that the hens were no longer on the nest. I determined the nest to be successfully hatched. As it turns out all cameras had ended up on the ground due to cattle in the area, rubbing on the rebar that was used in the camera set-up.

On May 14th all remaining cameras (Moltrie) were set out on artificial nests. Monitoring of the cameras (live & artificial) ran for 43 days. All cameras were removed from the field on June 26, 2005.

There were times cameras were not in the field. These times were when the nest was predated and the camera was removed in order to download the pictures on a computer. The download of the pictures was delayed if the predation occurred on a weekend due to accessibility to a computer.

During the predation events, 2754 pictures were taken by thirteen cameras. At times due to grass swaying in the breeze, camera movement, etc., the camera memory would be full and the nest not predated. These pictures, a total a 1416, were downloaded and filed as np or non-predated. The animal species that were photographed at predated nests but not necessarily the predator of the Sage Grouse nest included:

Raven -	418 pictures
Cattle –	203 pictures
Magpie –	26 pictures
Pronghorn -	18 pictures
Songbirds -	11 pictures
Coyote -	4 pictures
WY Ground Squirrel -	3 pictures
Crow –	1 picture

Positive identification of predation based on photographs can only be associated to ravens. Several photos show a raven with an egg in it's beak. In one predation event it can be presumed that cattle crushed several eggs, as cattle were the only animal species recorded for the predation event. The photos from the predation event showed cattle with their heads in the nest bowl numerous times.

In summary this predator study has some merit in the management of Sage Grouse, but how much merit remains to be seen. Larger issues such as the amount of habitat and the quality of habitat need to be addressed by the Pinedale Local Work Group.

It turns out that is somewhat difficult to attract predators to a nest, as with the Shelter Cabin nest (2 live, 2 artificial), that were never predated. This area had cracked eggs as well as dead fish distributed near the nests and still no predation occurred. It is obvious that avian predators are easier to attract than mammalian predators, based on photo documentation.

No photos of mammals (other than cattle) were recorded until fish was distributed in the area of the nest. On the other hand, out of 5 camera sites that had fish used as bait, a mammalian predator (coyote) was recorded at only one site. It can not be determined what the coyote did to the eggs, as ravens and magpies were at the nest site prior to the coyote. Also, three of the five had ravens predating the nests and the other two had no predation.

This nest site was re-stocked with 8 brown chicken eggs and a fish was used as bait. There is documentation on the Talon camera that the coyote visited the artificial nest site. The fish was gone, but eggs remained in the nest bowl – untouched.

Cattle had a moderately negative impact on the study, as the cattle would rub on the rebar being used to hold the cameras upright and resulted in the cameras ending up on the ground or turned away from the nest bowl.

If the predator study is to be continued, I have several recommendations.

1. It may be possible to put cameras on more live nests, as the hens did not flush during the camera set up on the two sites utilized.
2. Since the hens didn't flush during the camera set up, going in on the nest site several times to check the condition of the camera set may be an option.
3. Different bait and scent lures need to be provided to the Technician to use at nest sites in order to increase the possibility of recording a mammalian predation.
4. Use of artificial nests only may get the results being sought after without sacrificing an actual Sage Grouse nest. For several reasons – The cameras can not be set close enough on a live nest to get an unobstructed view and installing a camera too close may flush the hen, resulting in abandonment of the nest. Also, the two live nests used in the study were never predated and the artificial nest had numerous predation events.

Respectively submitted,

Steven W. Huber
Sage Grouse Field Technician

Methods implemented for the Pinedale Local Work Group Predator Study

A study was developed to determine what animal species predate Sage Grouse (*Centocerous europhasianus*) nests and what the general appearance is of the predated eggs/nest. This study will provide general information on predators of Sage Grouse, as well as providing some correlation to 300+ photos of predated Sage Grouse eggs and what the predator of the nest might be.

The evidence to be gathered, will be done with the use of a motion camera set up at nests of actual (live) Sage Grouse incubating the eggs, as well as motion cameras set up on artificial nests. Prior to the study several camera were analyzed for quality of photos, ease of use, dependability, etc. From a group of five different brands of cameras, two were chosen for the study.

The live nest were found with the use of radio telemetry and tracking Sage Grouse hens that had previously been trapped and had radio collars attached by Rusty Kaiser (UW grad student) for his thesis study.

The live nests will have Talon (infra red sensor) cameras set up to record possible predation events. Because of the unknown variables such as alignment of the sagebrush nest site and surrounding sagebrush that would not allow for a clear view of vision for one camera, one of the live nest sites will have two Talon cameras set up to monitor a possible predation event.

The Talon cameras will be set up at a distance of approx. 25' from the nest site. This is the maximum distance that would still provide a picture that could clearly identify the predator species. Setting the camera any closer would run the risk of flushing the hen from her nest. If flushed, there is a possibility of the hen abandoning her nest.

The artificial nests will have one Moultrie camera set up at about 10' from the nest site. Since there was not a concern of flushing a Sage Grouse hen, the camera can be set closer to the nest bowl. Any sagebrush that might obscure the camera was removed.

A total of two live nests will be utilized in the study and initially nine artificial nests constructed. The artificial nests will consist of eight chicken eggs that are brown to resemble closer Sage Grouse eggs, which are a grayish color.

In order to attract predators to the artificial nests, chicken eggs will be distributed around the nest at 25 to 50 yards. Anywhere from four to eight eggs distributed and they will be either broken or have puncture holes, in order to allow mammalian predators to pick up the scent of the eggs. Later in the study dead fish and a scent lure was used in the attempt to attract mammalian predators.

The location of the artificial nests will vary from upland, to bottomland and along dry stream bank areas. As artificial nest become predated the eggs will be replaced until a definite predation pattern is determined for the predator caught on film by the motion cameras. When confident enough pictures of a predator at a certain site have been obtained, the artificial nests will be moved to a new site to hopefully attract a different predator species.

The length of the study will run from the time period typical for Sage Grouse, which is from mid-May to mid-June.

Weston County Wildlife Enhancement Project

WESTON COUNTY
WILDLIFE
ENHANCEMENT PROJECT

2005 YEAR PROJECT REPORT SUBMITTED TO:

Wyoming
Animal Damage Management Board

SUBMITTED BY:

Alan L. Todd
President - Weston County Predatory Animal District
Weston County Wildlife Enhancement Project Administrator
PO Box 795
Upton, Wyoming 82730
(307) 468-2553

Weston County Wildlife Enhancement Project
2005 Project Report

Ground Control Work:

Ground personal conducted no control work in the project area except as called for by livestock owners. No project funds were used in coyote control work. An antelope recruitment study was done on the ground during July 2005.

Aerial Hunting and Survey:

Total flying time expended during the 2005 project 20.2 hours. This time was used to do population surveys done by grid flying to count total numbers of antelope in the area in the late fall of 2004 and summer of 2005. No aerial hunting took place using ADMB Funds. Aerial hunting that was done was on a call basis by livestock owners and paid for by livestock funds.

Removal of coyotes from Treatment Zone:

Fiscal year **2001** coyotes removed by Wildlife Services: 213 Total

Fiscal year **2002** coyotes removed by Wildlife Services: 172 Total
Fiscal year **2002** known coyotes removed by in kind help: 26 Total
Total known coyotes removed from project area FY 2002: 198

Fiscal year **2003** coyotes removed by Wildlife Services: 234 Total
Fiscal year **2003** known coyotes removed by in kind help: 34 Total

Project year **2004** coyotes removed by Wildlife Services: 213 Total
Project year **2004** known coyotes removed by in kind help 41 Total

Project year **2005** survey only

Stomach Content Analysis Over Entire Project Period:

Using the same procedure as outlined in the FY 2001 report, the following information was gathered ending with 2004:

Total Number of coyote stomach contents checked -------------- 196

Contents as follows:

Empty	99
Antelope, Deer	39
Rabbit, mice and gophers, other	42
Livestock	8
Sage Grouse	8

Antelope Counts In the Weston County Wildlife Enhancement Treatment Area taken by Weston County Wildlife Project Personal

November 2000 1388 Total Herd Count (BY AIR)

April 2001 1556 Total Herd Count (BY AIR)

July 2001 Approximately 52% of April 2001 herd count observed, 69.45% recruitment estimated (GROUND COUNT)

November 2001 count was not done to preserve dollars for control work in spring of 2001

May 2002 1738 Total Herd Count (BY AIR)

July 2002 Approximately 59% of April 2002 herd count observed, 83.4% recruitment estimated (GROUND COUNT)

November 2002 2989 Total Herd Count (BY AIR)

May 2003 1912 Total Herd count (BY AIR)

July 2003 352 Mature does observed, 115 Mature Bucks observed, 111 yearling Bucks observed, 111 yearling does tabulated.
421 fawns observed for a recruitment of 119%

May 2004 2134 Total Herd count (By Air)

July 2004 397 Mature Does observed, 231 Mature Bucks observed, 220 yearling bucks observed, 220 yearling does tabulated.
445 Fawns observed with a recruitment of 112%

2005 Project Year

Nov 2004 **4410 Total Herd Count (By Air)**

May 2005 **3019 Total Herd Count (By Air)**

July 2005 **Recruitment Count by ground work**
2088 Total Antelope viewed
- **625 Does**
- **698 Fawns**
- **279 Mature Bucks**
- **243 Yearling Bucks**
- **243 Yearling Does (tabulated)**

2005 Recruitment of 112 %

Antelope survey results:

Grid flying and counting total number of antelope in the project area:

Apr 2001 -- 1556 Head Total Antelope
May 2002 ---1738 Head Total Antelope
Nov 2002 ---2989 Head Total Antelope
May 2003 ---1912 Head Total Antelope
May 2004 ---2134 Head Total Antelope
Nov 2004--4410 Head Total Antelope
May 2005---3019 Head Total Antelope

2005 Project Budget Report
Weston County Wildlife Enhancement Project

Revenue for 2005 Project:

Wyoming ADMB ---------------------------------------	$4175.00
Weston County Predatory Animal District ----------	161.00
In kind donations (time and vehicles) ---------------	250.00
Total Revenue ----------------------------------	**$ 4586.00**

Operational Expenses for 2005 Project:
All Survey work:

Wildlife Services Specialist and vehicle-------------	$ 2240.00
20.2 Hours Airplane ----------------------------------	1616.00
Hazard Duty --	400.00
ATV fuel and time ------------------------------------	80.00
14.25 In kind man-hours and vehicle cost -----------	250.00
Total Expended ------------------------------	**$ 4586.00**

Mountain Lion Studies

Experimental evaluation of population trend and harvest composition in a Wyoming cougar population

Charles R. Anderson, Jr. and Frederick G. Lindzey

Abstract Cougar (*Puma concolor*) management has been hindered by inability to identify population trends. We documented changes in sex and age of harvested cougars during an experimentally induced reduction in population size and subsequent recovery to better understand the relationship between sex–age composition and population trend in exploited populations. The cougar population in the Snowy Range, southeast Wyoming, was reduced by increased harvest (treatment phase) from 58 independent cougars (>1 year old) (90% CI=36–81) in the autumn of 1998 to 20 by the spring of 2000 (mean exploitation rate=43%) and then increased to 46 by spring 2003 following 3 years of reduced harvests (mean exploitation rate=18%). Pretreatment harvest composition was 63% subadults (1.0–2.5 years old), 23% adult males, and 14% adult females (2 seasons; *n*=22). A reduction in subadult harvest, an initial increase followed by a reduction in adult male harvest, and a steady increase in adult female harvest characterized harvest composition trends during the treatment phase. Harvest composition was similar at high and low densities when harvest was light, but proportion of harvested subadult males increased at low density as they replaced adult males removed during the treatment period (high harvest). While sex ratio of harvested cougars alone appears of limited value in identifying population change, when combined with age class the 2 appear to provide an index to population change. Composition of the harvest can be applied to adaptively manage cougar populations where adequate sex and age data are collected from harvested animals.

Key words adaptive management, cougar, exploitation, population trend, *Puma concolor*, sex–age composition

Several authors have noted the need for reliable techniques to adequately monitor cougar population changes (e.g., Shaw 1981, Lindzey 1991, Anderson et al. 1992, Riley 1998). While populations have been monitored with long-term, intensive capture efforts over relatively small areas (Ashman et al. 1983, Anderson et al. 1992, Ross and Jalkotzy 1992, Lindzey et al 1994, Logan and Sweanor 2001), reliable and affordable techniques to monitor population trends for large-scale management programs remain elusive.

Cougar management traditionally has employed harvest levels to achieve specific population objectives with little understanding of the quantitative effect that differing harvest levels have on cougar population demographics. Sex and age classes of cougars exhibit different and relatively predictable movement patterns (Barnhurst 1986). These differences, in turn, presumably expose each group to differing risks of being harvested. This concept has been applied to managing black bear (*Ursus americanus*) populations in many western states

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(Garshelis 1990). Barnhurst (1986) investigated the vulnerability of cougars to sport hunting as a step toward understanding how to interpret harvest data. He proposed that vulnerability to harvest would be related to the frequency at which differing sex- and age-class cougars cross roads because cougars are generally hunted using trailing hounds, typically from roads or trails. The vulnerability index he developed from road-crossing frequencies suggested that transient males were most vulnerable, followed by resident males, transient females, resident females both without young and with young >6 months old, and finally resident females with young ≤6 months old.

Conceptually, the likelihood of a specific sex or age class of cougar being harvested would reflect its relative abundance in the population multiplied by its relative vulnerability. The least-vulnerable individuals should become prominent in the harvest only after the population had been reduced in size by removal of more vulnerable cougars. Our objective was to test the hypothesis that sex and age composition of the harvest would vary predictably with population size in a cougar population primarily hunted using hounds.

Study areas

Experimental population

The Snowy Range, located in southeast Wyoming about 30 km west of Laramie, was a 2,760-km^2 timbered region including a 2,170-km^2 portion of the Medicine Bow National Forest surrounded by private, Bureau of Land Management, and state-owned lands. This terminal mountain range was surrounded by sagebrush (*Artemisia tridentata*) grasslands except on the southern end, where it was connected to contiguous habitat by a 14-km-wide segment of the Medicine Bow Mountains. Cougars occupied about 1,700 km^2 of this area during winter. Wyoming State Highway 230 on the west, United States Interstate 80 on the north, the Laramie River and Sand Creek drainages on the east, and Colorado highways 125 and 127 on the south bounded the Snowy Range. The area was topographically diverse, ranging in elevation from about 2,100 m in the valleys to 3,652 m at Medicine Bow Peak. Vegetation communities were dominated by sagebrush grasslands in the peripheral valleys; lodgepole pine (*Pinus contorta*) stands with interspersed quaking aspen (*Populus tremuloides*), Rocky Mountain juniper (*Juniperus scopulorum*), and limber pine (*Pinus flexilis*) at mid-elevations; and Engelmann spruce (*Picea engelmannii*)–subalpine fir (*Abies lasiocarpa*) forests with occasional limber pine at higher elevations (Alexander et al. 1986). Understory dominants in the mid- and high-elevation communities included huckleberry (*Vaccinium scoparium*), buffalo berry (*Shepherdia canadensis*), serviceberry (*Amelanchier alnifolia*), snowberry (*Symphoricarpos* spp.), and common juniper (*J. communis*). Riparian areas were composed primarily of willow (*Salix* spp.) with interspersed narrowleaf cottonwood (*P. angustifolia*) at low elevations.

Abundant roads provided good access to most cougar habitat in the Snowy Range. Annual harvest was relatively constant during the 5 years before our study, ranging from 9–12 cougars.

Comparison population

The northern portion of the Laramie Range included an isolated mountain range near the cities of Casper and Wheatland in southeast Wyoming and encompassed 2,960 km^2 of timbered habitat. Elevation ranged from 1,620 m in the eastern valleys to 3,132 m at Laramie Peak. Ponderosa pine (*P. ponderosa*) stands dominated low to mid elevations, with lodgepole pine common at mid to high elevations. Low-elevation, nonforested regions and interspersed meadows were vegetated by grasses, forbs, and shrubs. Riparian areas consisted primarily of willow with occasional aspen pockets. Other forest species occurring at low levels included limber pine, subalpine fir, Douglas-fir (*Pseudotsuga menziesii*), and Engelmann spruce.

Annual harvest in Laramie Peak averaged 11 cougars during the 5-year period before harvest treatment, ranging from 7–16 cougars per year. The Wyoming Game and Fish Department changed its management objective from sustained harvest of a stable to increasing population to reducing the population through increased harvest in 1996 and increased harvest quotas from 10 to 34 for the next 7 seasons. Regional Wyoming Game and Fish Department personnel believed the Laramie Peak cougar population was at a relatively high density prior to 1996 based on increased cougar sightings, depredation incidents, and hunter interviews.

Methods

We trailed cougars using hounds and immobilized them upon capture with a mixture of 5 mg/kg

Telazol® (Aveco Co., Inc., Cherry Hill, N.J.) and 1 mg/kg xylazine hydrochloride delivered in a hypodermic dart fired from a CO_2 pistol; we reversed the effects of xylazine hydrochloride using yohimbine hydrochloride (0.15 mg/kg). We tagged independent cougars (>1 year old and solitary) with standard VHF radiocollars (Model 9D, warranty battery life=3 years) and dependent young with 22-g ear-tag transmitters (Model 7PN, warranty battery life=295 days; Advanced Telemetry Systems, Inc., Isanti, Minn.); we equipped transmitters with mortality-sensing options. We also attached a uniquely numbered ear tag to all captured cougars. We recorded sex, age, weight, and morphometric measurements at capture. We estimated age (juvenile <1 year, subadult 1–2.5 years, adult ≥3 years) from tooth wear, canine ridge eruption, spotting progression, and evidence of previous lactation for females (Shaw 1979, Ashman et al. 1983, Lindzey et al. 1989, Laundre et al. 2000) or known birth date for cougars born to radiocollared females based on female denning behavior. We located radiotagged cougars weekly from fixed-wing aircraft between December 1997 and May 2001 and once per month from June 2001–April 2003.

We used radiotelemetry to identify female denning behavior (consecutive locations at the same location), timing of family breakup, and emigration of subadults. We assumed emigration when an individual dispersed from its mother, had not yet exhibited territorial behavior, and we were no longer able to detect its radio signal. We estimated age of juveniles of unknown birth date by applying the growth-curve models developed in the Northern Great Basin (Laundre and Hernandez 2002) after adjusting them for differences detected when comparing model estimates to size of known-age juveniles in the Snowy Range (C. R. Anderson, unpublished data).

Experimental design

We manipulated size of the Snowy Range cougar population using regulated hunter harvest to reduce and then allow recovery of the population; all cougars harvested during the study except 2 were taken using hounds. The cougar-hunting season was open from 1 September–31 March, but most cougar harvest did not occur until mid-November, when snow conditions were adequate for tracking cougars using trained hounds; >90% of cougars harvested in Wyoming were taken using hounds (Wyoming Game and Fish Department 2003). Annual harvest levels were regulated by a quota system in which the season was closed if the quota was met before 31 March. Young (<1 year old) cougars and females with young at side were legally protected from harvest. We concurrently monitored sex and age composition of the population and the harvest and annually tested predictions of harvest composition based on abundance of sex- and age-class cougars in the population and their relative harvest vulnerability (Barnhurst 1986). We predicted that harvest composition would be predominantly subadults (possibly more females) during the pretreatment year (high density, low harvest), shift to adult males during the first year of treatment (from high to moderate density, high harvest), shift from adult males to adult females during the second treatment year (from moderate to low density, high harvest), and return to subadults during the post-treatment period (increasing population, low harvest) where the subadult segment would initially consist primarily of males and eventually consist primarily of females as the population approached pretreatment levels. We examined annual changes in harvest composition of adult males, adult females, and subadults using the Fisher's exact test; we applied 1-tailed tests to compare the first 4 seasons where changes were predicted and 2-tailed tests to examine the recovery period when composition was not expected to change greatly. We also examined the relationship between proportion of adults in the female harvest and estimated harvest rate using simple linear regression analysis, expecting adult female harvest composition to increase with harvest level.

We then compared harvest composition documented in the Snowy Range to that observed in Laramie Peak. Although we did not monitor density in this area, it represented a geographic population (i.e., occupied cougar habitat surrounded by inhospitable, unoccupied landscapes) similar to the Snowy Range, contained a similar amount of cougar habitat, had adequate hunter access to facilitate population reduction, and the population was exposed to harvest levels similar to those we applied in the Snowy Range before and during the treatment period. We assumed that harvest composition from this area would show similar trends to those documented in the Snowy Range if harvest composition changed predictably with population size in harvested populations. We tested for differences in annual harvest composition between populations using the Fisher's exact test (2-tailed). We also determined ages from counts of cementum annuli of harvested adult females in both populations to determine whether age of adult females declined as the population declined following high harvest levels.

Age-class estimates

We assigned harvested and captured cougars to age class based on tooth wear, presence or absence of a canine ridge, evidence of spots or foreleg bars, evidence of previous lactation if female (Anderson and Lindzey 2000), and counts of bands in the cementum of premolars removed from harvested cougars. We first gave priority to evidence of previous lactation in females (subadult: nipples white and ~4–6 mm wide; adult: nipples dark or mottled and ~8–10 mm wide), followed by annuli age (subadult=1–2 yr), canine ridge eruption (absent=subadult), and finally foreleg bars (dark=subadult or young adult) and spots (present=subadult or young adult). To evaluate reliability of our aging techniques, we compared ages estimated from counts of cementum bands to ages estimated with the other criteria for those cougars that were captured and later harvested.

Population estimates

During the first winter (Dec 1997–Apr 1998), we conducted intensive capture efforts in 2 regions of the Snowy Range to obtain an initial density estimate and to create a marked sample for subsequent mark–recapture efforts. We captured cougars in a 439-km^2 area in the southeast region and a 382-km^2 area in the west-central region of the Snowy Range; 90% of cougar harvests in the Snowy Range came from these primarily public land areas (Wyoming Game and Fish Department mountain lion harvest data base, Lander, Wyo.). We estimated density for the 2 areas by summing number of cougars marked and tracks of known, unmarked cougars. We included unmarked cougars only if track characteristics (identified as male or female via planter pad width and stride length; Fjelline and Mansfield 1988) and number and size of young accompanying a female suggested a unique individual and when tracks were located outside traditional use areas of radiocollared cougars identified from previous telemetry locations. The initial density estimates from the 2 areas were then applied to the remainder of cougar habitat in the Snowy Range to estimate population size for the study area. Cougar habitat was delineated using elevations and topography used by radiocollared cougars February–April, 1998.

We applied the Lincoln-Peterson estimator (Pollock et al. 1990) to calculate annual, pre-hunting-season (autumn) population estimates of independent cougars. Post-hunting-season (spring) population estimates were pre-season estimates minus harvest removals and estimated natural mortality from our marked sample. We attempted to meet assumptions of the technique by modifying our sampling design and using information from radiotagged cougars. We addressed geographic closure by recapturing during late autumn and winter months when emigration and immigration were least likely (Ross and Jalkotzy 1992). We addressed the demographic closure assumption by adjusting for deaths based on records from radiocollared cougars and by considering young cougars in our marked sample independent at the mean age family groups became loosely associated (prior to dispersal), and thus available for recapture (e.g., harvest), by the beginning of the recapture period (15 Nov, average date of sufficient snow for hunting). Because cougar captures relied heavily on adequate snow conditions for tracking that varied temporally and spatially, maintaining equal capture effort throughout the study area was not possible and reduced our ability to assure equal capture probabilities across cougars. To minimize potential biases from capture heterogeneity and provide sufficient time to sample the entire study area, we treated the entire winter sampling period (15 Nov–31 Mar) as a single capture effort and counted each individual detected only once in the recapture sample regardless of the number of times they were actually detected. Because captured cougars remained ear-tagged throughout the study but transmitter failures occasionally occurred, we assumed individuals that had established territories prior to transmitter failure and that had been monitored until the previous summer were still in the population and available during the following winter recapture period; on 10 of 12 occasions where transmitters failed, marked residents were subsequently recaptured or harvested.

The capture sample was independent, radiotagged cougars in the population at the beginning of the recapture sampling period (15 Nov) during both treatment and recovery periods. The recapture sample was cougars harvested by hunters during the hunting seasons of the treatment periods, but, because harvests were intentionally reduced during the recovery period (winters of 2000–2001, 2001–2002, and 2002–2003), we augmented the recapture sample by hunting the study area after hunters had finished. During our hunting we tagged and released unmarked cougars, recorded marked cougars recaptured, and recorded presence of individual, unmarked cougars (defined earlier) we were unable to capture. We included cougars marked in the population prior to 15 November each year in our initial capture sample and those captured from 15 November–31 March in our recapture sample. We recorded capture effort as number of hunter days for successful hunters (no data for unsuccessful hunters) and number of days spent tracking and capturing cougars by study personnel. Post-season population estimates were pre-season estimates minus harvest and mortality from other causes estimated from our marked sample during the recapture period. We estimated 90% confidence intervals around pre-season population estimates following Pollock et al. (1990). We estimated autumn sex and age composition of the population by adding unmarked cougars harvested during that year's hunting season to our sample of marked cougars.

Results

We tagged 16 independent and 13 dependent male and 17 independent and 15 dependent female cougars between December 1997 and February 2002. Twenty-one marked, independent cougars were harvested during the treatment and recovery phases of the project, and 9 marked cougars (5 adult males, 4 adult females) were alive at the end of the study. Cougar ages estimated using cementum annuli counts were in agreement with other aging criteria in 14 of 18 comparisons and within 1 year for 3 others (Anderson 2003). We noted that ages of dependent young of known birth date in the Snowy Range were consistently underestimated ($\bar{x}$ = 1.47 mo, SD = 1.26, n = 13) using the Northern Great Basin growth-curve models (Laundre and Hernandez 2002) and therefore added the mean difference to estimate ages for litters of unknown birth date.

Dependent cougars became independent at an average age of 14 months (range = 11–17 months, n = 7); 2 litters became independent following the death of their mother at 14 and 17 months old (1 natural, 1 harvest). Association among family members became progressively looser over the month before independence. Thus, to account for recruitment in our recapture sample, we included marked dependent young as subadults if they were 13 months of age by 15 November each season. Emigration occurred between April and September for 8 of 9 emigrants monitored; 1 subadult male emigrated during January.

Population estimates

We tagged 18 cougars in the study area and identified 6 others from tracks after 60 days of trapping and tracking in the southeast and 45 days in the west-central section of the Snowy Range during winter 1997–1998. We estimated independent cougar density at 3.42/100 km^2 in the southeast (15 cougars/439 km^2 × 100) and 2.35/100 km^2 in the west-central region (9 cougars/383 km^2 × 100). Cougar habitat in the Snowy Range during this period, estimated from characteristics of habitat used by marked cougars February–April 1998, was 1,720 km^2. We estimated 50 independent cougars in the Snowy Range in spring 1998 (45–55 depending on the density estimate applied). A harvest quota of 25 was then set for the next 2 hunting seasons (treatment; 1998–1999 and 1999–2000) to elicit the desired (about 50%) reduction in the Snowy Range cougar population.

Harvests were 25 and 17 cougars for the 2 treatment seasons, resulting in an estimated population of 20 independent cougars by spring 2000 (Table 1). Harvest quotas were then reduced to 6–8 cougars per season to facilitate population recov-

Table 1. Pre (autumn) and post-harvest (spring) cougar population estimates[a] from the Snowy Range, Wyoming, USA, autumn 1998–spring 2003. Note population decline following 2 years of high harvest and population increase following 3 years of light harvest.

Season	n_1	n_2	m_2	$\hat{n}_{pre}$ (90% CI)	No. harvested	% natural mortality	$\hat{n}_{post}$
1998/99	15	25	6	58 (36–81)	25	11	30
1999/00	19	17	8	39 (28–50)	17	9	20
2000/01	15	21	9	34 (26–42)	8	0	26
2001/02	15	25	10	37 (29–44)	6	0	31
2002/03	11	39	7	59 (42–76)	8	9	46

[a] $\hat{n}_{pre} = [(n_1 + 1)(n_2 + 1) / (m_2 + 1)] - 1$, where n_1 = number marked and released in first sample, n_2 = number captured in second sample, and m_2 = number captured in second sample that were marked from first sample. $\hat{n}_{post} = (\hat{n}_{pre} - \text{harvest}) - [(\%\text{ natural mortality})(\hat{n}_{pre} - \text{harvest})]$.

ery. The population increased to an estimated 46 independent cougars by spring 2003 (Table 1). The number of hunter-days totaled 47 and 79 during the 2-year treatment period and 27, 50, and 21 days during the 3-year recovery period; high hunter effort during the second treatment year and the second recovery year were due to excessive time spent hunting by an individual hunter each year (30 and 36 days, respectively). We spent 60, 54, and 68 days tracking and marking cougars to augment the recapture sample during the recovery phase.

Cougar harvest composition in response to manipulation

Cougar harvest ($n=22$) composition during the pretreatment period was composed primarily of subadults (36% F, 27% M) followed by adult males (23%) and finally adult females (14%; Figure 1). As harvest levels increased and the population declined in size, there was an initial increase (40%) followed by a decrease (24%) in proportion of adult males in the harvest and a consistent increase in the proportion of adult females (14 to 24 to 41%). Subadult harvest declined from the pretreatment period (from 63 to 36%) but was consistent during the treatment period (35%) and was primarily composed of females (28 and 29%). Subadult cougars again dominated the harvest after harvest quotas were reduced, but subadult male composition was relatively higher than during pretreatment and treatment periods until the third year of recovery when the population returned to pretreatment levels. Annual harvest composition among adult males, adult females, and subadults differed significantly ($P \leq 0.034$) from the pretreatment period through the first year post-treatment and was similar ($P \geq 0.664$) during the 3-year recovery phase.

We compared harvest records from Laramie Peak, the comparison population, to harvest records from the Snowy Range including the first 3 years of harvest (harvest levels below quota) in Laramie Peak and 2 years of harvest treatment and the first year post-treatment in the Snowy Range. During the 3-year period, harvest declined and pri-

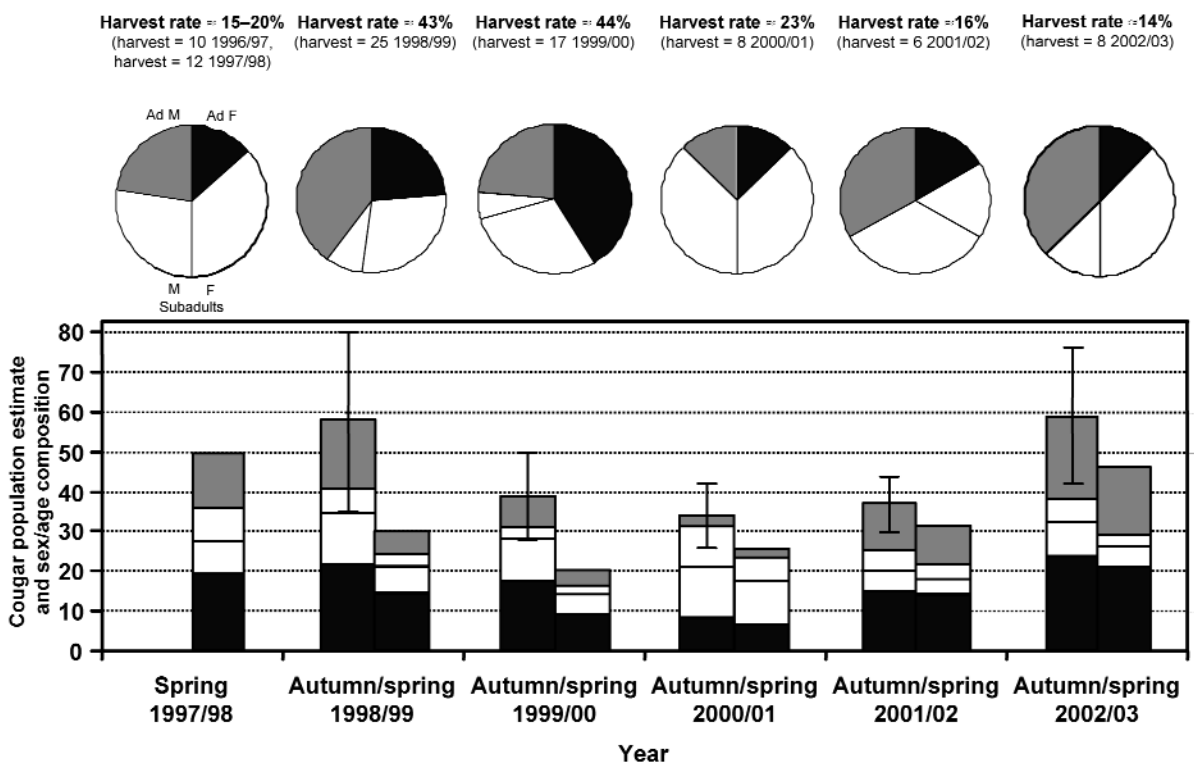

Figure 1. Sex–age composition of cougar harvest (pie charts) from the Snowy Range, Wyoming, relative to population change through increased (1998–2000) and reduced (2000–2003) harvest levels (order of sex–age classes in bar graphs follow pie charts). Harvest composition and rate prior to 1999 represent harvest years 1996–1997 and 1997–1998 combined (first column). The population estimate for spring 1998 was determined from mountain lion density detected from capture and tracking efforts during winter 1997–1998; subsequent population estimates were derived using mark–recapture methods. Error bars represent 90% confidence intervals. Number of cougars known to be in the population each spring were 22, 12, 15, 18, 20, and 34, respectively.

marily consisted of adult males initially, followed by adult females, and finally subadults in both populations (Figure 2); annual harvest composition was similar between populations ($P \geq 0.217$). Mean annuli age of adult females declined following the first treatment year from 6–8 years old to 3–4 years old the second year in both populations. Unlike the Snowy Range, unrestricted harvests continued in Laramie Peak for the next 4 years, resulting in annual oscillations in harvest level and harvests of primarily subadults (Figure 2); adult females averaged 4.3 years of age during this period.

Characteristics of female cougar harvest

We noted that proportion of adults in the female harvest increased with harvest rate, ranging from 20% with a 21% harvest rate to 58% with a harvest rate of about 44% (Figure 1), but this relationship was not statistically significant ($r^2 = 0.40$, $F_{1,6} = 3.32$, $P = 0.13$). Sixteen adult and 19 subadult females were harvested (total harvest = 64) in the Snowy Range during the 2-year treatment and 3-year post-treatment periods. Of 8 marked adult females harvested, 4 were without young, 3 had young at the time, and we suspect the last female may have had young when harvested because we had seen kitten tracks with her 2 months earlier. All harvested females with young were taken during the treatment period (>40% harvest rate).

Discussion

The Snowy Range cougar population recovered in numbers after 2 years of intensive harvest (~43% of independent cougars) followed by 3 years of light harvest (~18% of independent cougars). Recovery of the population was facilitated by immigration of males and recruitment of females from within the population as found in other recovering cougar populations (Lindzey et al. 1992, Logan and Sweanor 2001). Composition of the harvest from pretreatment through the 2 years of heavy harvest supported our predictions based on predicted relative vulnerability of the various sex and age classes. The most vulnerable classes were harvested until their reduced abundance in the population

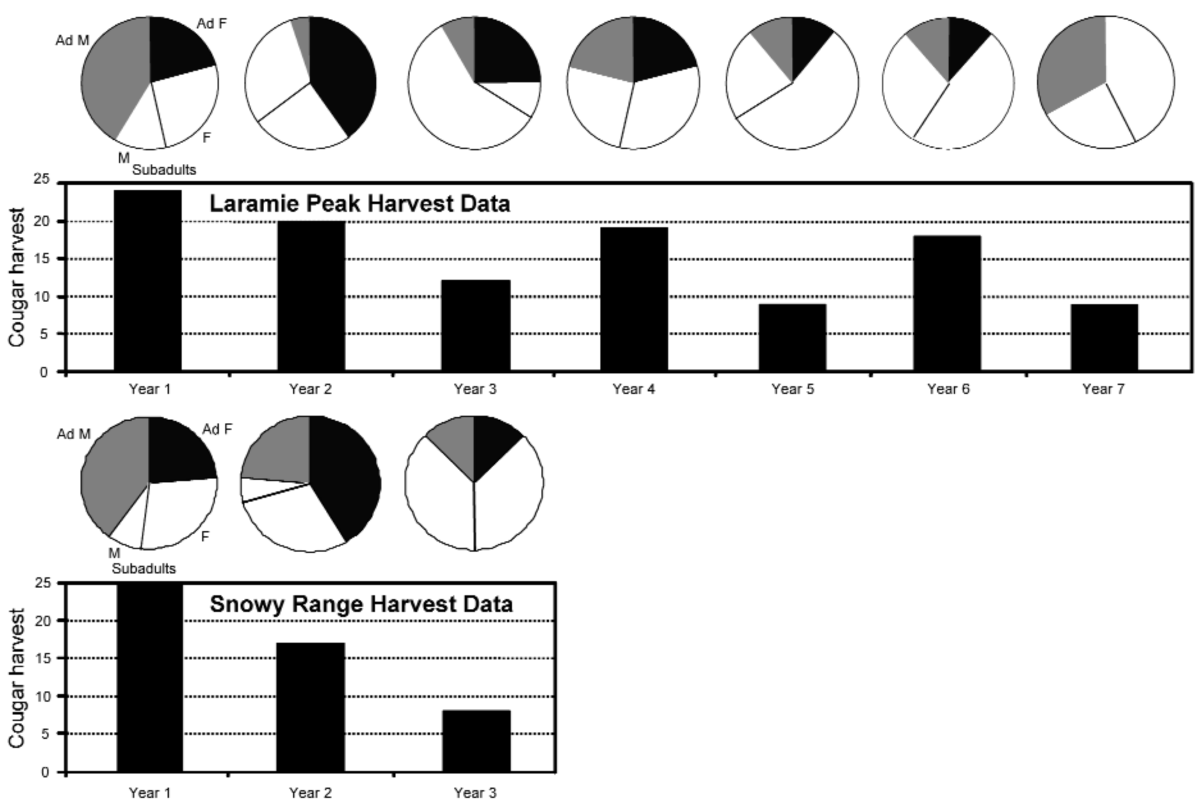

Figure 2. Comparison of total harvests (bar graphs) and harvest composition (sex–age class; pie charts) from Laramie Peak and the Snowy Range in southeast Wyoming. Cougar harvest quotas were not met, except in the Snowy Range during years 1 and 3. Note similarities in harvest levels and composition between populations exposed to similar harvest treatments.

exposed the next most vulnerable class, terminating in a harvest dominated by adult females (Figure 1). The increase in adult females in the harvests coincided with a decrease in size of this hunted population, suggesting that proportion of adult females in harvests may be a useful indicator of trends in other hunted cougar populations. The similarity of composition trends in the Snowy Range and Laramie Peak populations during the initial years of intensive harvest suggests that the intensive harvest in the Laramie Peak population had achieved its goal of reducing population size in this area. Decline in average age of harvested females in both populations further suggested that harvests had similar effects on the 2 populations.

While factors other than composition of hunted cougar populations (e.g., weather patterns, changes in legal access) can influence harvest level, none should result in adult females dominating the harvest if they are not proportionately the most abundant sex or age class present in the population. Experienced cougar hunters often can differentiate males and females from track size, presence of scrapes, or body characteristics if the cougar is seen, but selective hunters tend to harvest males. Further, our experience suggests that hunters tend to be most selective when competition for available cougars is low. When demand exceeds harvest quotas, competition among hunters appears to result in less-selective hunting, and harvest should reflect the relative abundance or vulnerability of sex and age classes. Snow conditions also can affect hunting success (>90% of cougars harvested in Wyoming are hunted using hounds and most require snow cover), but this should influence harvest rate, not the relative vulnerability of the sex and age classes. Access, influenced by weather events or land-ownership patterns, can create ephemeral or more permanent refuges within cougar management areas. In these situations harvests may be maintained by adjacent, unavailable adult females providing young females for the harvest (e.g., Figure 2). We identified areas of suitable cougar habitat in the Laramie Peak area that received no cougar harvest and apparently were functioning as refuges. The similar abundance of subadult females in the pretreatment Snowy Range harvest and post-treatment harvests from Laramie Peak illustrates the contribution of refuges to maintaining harvests and underscores the need to monitor harvest composition over a number of years before drawing inferences about trend in the population from harvest composition. Subadult females in the pretreatment Snowy Range harvest reflected their relative abundance and vulnerability to harvest, while their dominance in later harvests from Laramie Peak apparently reflected their abundance in the portion of the area accessible to hunters. Examination of composition of earlier harvests should help identify whether the harvest reflects a lightly hunted population or one that has been reduced with harvests being supported by young produced by adjacent, unavailable adult females. Prior harvests in the Laramie Peak area were composed of progressively more adult females, suggesting the population had been reduced in size.

Management implications

Cougar managers typically have used harvest level and occasionally sub-quotas typically aimed at protecting females to achieve population objectives, although both imply knowledge of population size. While observations suggest that cougar populations can sustain harvest rates of up to 20–30% (Ashman et al. 1983, Ross and Jalkotzy 1992), the effect of harvests on populations will differ depending on sex and age of cougars removed. Harvest of males, the cohort most easily replaced by immigration, and subadult females, which can be quickly replaced by female young produced in the population, will have less impact on the population than harvest of adult females, which are more difficult to replace. Adult females that die are most often replaced by the population's female progeny and less often by immigrating subadults because most female progeny are philopatric (Lindzey et al. 1989,

Duggin Wroe's dog, Luna, corners male cougar number 610. Photo by Hall Sawyer.

Anderson et al. 1992, Logan and Sweanor 2001).

Monitoring levels of adult females in cougar harvests to index the effect the harvest is having on the population is intuitive. Sensitivity analyses by Martorello and Beausoleil (2003) suggest that cougar populations are most sensitive to survival of this sex and age class. Adult females provide the resiliency in a population that allows it to respond to loss of members. This approach will work well in an adaptive management framework, where harvest composition goals are set to achieve specific population objectives. Hunting programs can simply be modified until harvest composition indicates that desired population and recreation objectives are being met. The proportion of adult females in the Snowy Range harvest when the more vulnerable sex and age classes had been removed and the population was beginning to decline was about 25%, while the population appeared to sustain a harvest composed of 10–15% adult females (Figure 1). The 25% estimate came from a single experiment and should be used with caution in other programs because cougar populations more isolated than the Snowy Range or that contain more refuge areas may respond differently to similar harvest rates of adult females. Also, because harvest from a single management area in a single year may be too small to support inferences, and harvest level may vary because of weather events, combining years or adjacent management areas for analyses may be appropriate.

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GENETIC STRUCTURE OF COUGAR POPULATIONS ACROSS THE WYOMING BASIN: METAPOPULATION OR MEGAPOPULATION

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We examined the genetic structure of 5 Wyoming cougar (*Puma concolor*) populations surrounding the Wyoming Basin, as well as a population from southwestern Colorado. When using 9 microsatellite DNA loci, observed heterozygosity was similar among populations ($H_O = 0.49$–0.59) and intermediate to that of other large carnivores. Estimates of genetic structure ($F_{ST} = 0.028$, $R_{ST} = 0.029$) and number of migrants per generation (*Nm*) suggested high gene flow. *Nm* was lowest between distant populations and highest among adjacent populations. Examination of these data, plus Mantel test results of genetic versus geographic distance ($P \leq 0.01$), suggested both isolation by distance and an effect of habitat matrix. Bayesian assignment to population based on individual genotypes showed that cougars in this region were best described as a single panmictic population. Total effective population size for cougars in this region ranged from 1,797 to 4,532 depending on mutation model and analytical method used. Based on measures of gene flow, extinction risk in the near future appears low. We found no support for the existence of metapopulation structure among cougars in this region.

Key words: central Rocky Mountains, cougar, gene flow, genetic structure, metapopulation, microsatellite DNA, panmixia, *Puma concolor*

Cougars are solitary carnivores exhibiting a polygynous breeding strategy where dominant males typically breed with females that reside within their home range (Murphy 1998). Resident males aggressively defend their territories against male intruders, whereas females allow more overlap with conspecifics, but express mutual avoidance (Logan and Sweanor 2001; Ross and Jalkotzy 1992). Size of female home ranges tends to be large enough to provide sufficient prey for themselves and their young, whereas male home ranges tend to be larger, overlapping those of several females, apparently to maximize their reproductive success (Murphy et al. 1998). Female recruits commonly express philopatric behavior upon independence, but males typically disperse from their natal range (Anderson et al. 1992; Lindzey et al. 1994; Ross and Jalkotzy 1992); movements of >450 km have been documented for subadult males (1998–1999 harvest records, Wyoming Game and Fish Department, Rock Springs—Logan and Sweanor 2001). The purpose of this paper was to assess connectivity among cougar populations by using microsatellite DNA markers.

Conflicting evidence currently exists for whether cougars in North America are panmictic or whether local populations occur in a less connected metapopulation structure. A metapopulation is a population distributed in subpopulations across a set of suitable habitat patches typically isolated in a matrix of unsuitable habitat, in which each subpopulation in each patch has a nontrivial probability of extinction (Gilpin and Hanski 1991). Suitable habitat patches for cougar populations in the western United States typically occur in mountainous regions with some form of overstory canopy, whereas unsuitable habitat consists of open shrub and/or grassland basins separating mountain ranges (e.g., Laing 1988; Logan and Irwin 1985; Williams et al. 1995). Other factors, such as heavy exploitation of the population or human development, may inhibit or alter gene flow, enhancing the potential for metapopulation structure

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of cougar populations. Beier (1996) convincingly demonstrated cougar metapopulation structure from telemetry studies in California, where increased development created small, isolated pockets of occupied cougar habitat. Sweanor et al. (2000), without genetic data, proposed cougar metapopulation structure in New Mexico from estimates of dispersal, emigration, and immigration by using radiocollared cougars, but they also suspected gene flow might be high enough to limit risk of extinction in the near future. Culver et al. (2000) and Sinclair et al. (2001) examined genetic structure of cougar populations in the Western Hemisphere and Utah, respectively. Culver et al. (2000) concluded that North American cougars were a single genetic subpopulation and Sinclair et al. (2001) reported high gene flow across Utah. However, both studies used only small regional samples, which limited insight into whether cougars over large areas exhibit a metapopulation structure.

Wyoming offers an excellent opportunity to assess the existence of metapopulation structure of forest-dwelling species because the Wyoming Basin, running diagonally through the center of the state, separates several terminal mountain ranges dominated by conifer forests with open, basin habitats (Fig. 1) and may be a natural barrier to gene flow among cougar populations. Genetic studies support the Wyoming Basin as a barrier to gene flow in other species, including long-tailed voles (*Microtus longicaudus*—Conroy and Cook 2000), pikas (*Ochotona princeps*—Hafner and Sullivan 1995), and black bears (*Ursus americanus*—D. B. McDonald, University of Wyoming; http://www.uwyo.edu/dbmcd/molmark/lect09/lect9.html). Our objective was to assess genetic structure and gene flow among 5 geographically distinct cougar populations terminating in Wyoming and 1 distant population in southwestern Colorado and to determine whether the structure is consistent with metapopulation dynamics.

Materials and Methods

The Wyoming Game and Fish Department provided tissue samples from 234 cougars harvested in Wyoming during 1996–1998. Fecske (2003) provided 8 cougar blood samples from the Black Hills, South Dakota, collected during 2000–2001; Koloski (2002) provided 15 cougar blood samples collected from southwestern Colorado during 2000–2001; and we collected blood samples from 55 cougars in the Snowy Range in southeastern Wyoming (Fig. 1) during 1997–2001. Cougar capture procedures from the Snowy Range are described in Anderson (2003). Capture protocols were reviewed and approved under the University of Wyoming Animal Care and Use Committee, form A-3216-01, by following the American Society of Mammalogists guidelines (http://www.mammalogy.org/committees/indes.asp).

We genotyped cougars by using microsatellite DNA primers from the domestic cat (Menotti-Raymond and O'Brien 1995; Menotti-Raymond et al. 1999) at 10 loci (FCA008, FCA035, FCA043, FCA057, FCA077, FCA081, FCA082, FCA098, FCA132, and FCA149). Using conditions suggested by Li-Cor, Inc. (2000; Lincoln, Nebraska), an MJ PTC-200 and MJ tetrad Peltier thermal cycler (M. J. Research, Inc., Waltham, Massachusetts) performed 10-μl polymerase chain reactions (PCRs) on 60 ng of template DNA. We included 2 fluorescent primers complementary to a 19- and 20-base-pair extension on the 5′ end of the forward primer in the PCR reaction; the fluorescent primer binds to the amplifying product during the annealing stage of the PCR reaction. We used a Li-Cor 4200-S automated DNA sequencer running 25-cm polyacrylamide gels to visualize PCR amplicons detected by infrared laser fluorescence. Analog gel images were viewed by using GeneImagIR (version 3.0, Li-Cor, Inc.) and SAGAGen2 (version 2.1, Li-Cor, Inc.). To validate allele scores, 30% of our DNA samples were genotyped at least twice; we found no evidence of allelic dropout.

Data analyses.—We examined genetic variability (expected heterozygosity [H_E] or gene diversity—Nei 1987) and structure (θ, the F_{ST} analog of Weir and Cockerham [1984] and R_{ST} following Goodman [1997]) by using program FSTAT (version 2.9.3, Université de Lausanne, Dorigny, Switzerland; http://www.unil.ch/izea/softwares/fstat.html; Goudet 2001). We approximated number of migrants per generation (Nm) by following Slatkin (1995), where N is the effective population size, m is the proportion of migrants per generation, and $Nm = (1/F_{ST} - 1)/4$. Potential departures from Hardy–Weinberg equilibrium were examined by using GENEPOP (version 3.3, Center of Ecology and Functional Evolution, Montpellier, France; http://www.cefe.cnrs-mop.fr/GENEPOP; Raymond and Rousset 1995). Nine of the 10 loci occurred on different chromosomes or different linkage groups on the same chromosome (Menotti-Raymond et al. 1999), and were thus considered independent markers. The 10th locus (FCA098) was not genetically mapped by Menotti-Raymond et al. (1999), and we therefore tested pairwise genotypic linkage disequilibrium between FCA098 and the other 9 loci by using GENEPOP. The alpha levels for all statistical comparisons were adjusted by using a Bonferroni correction for number of populations and/or number of loci, where $P < 0.005$ and $P < 0.0008$ were deemed significant for tests within (10 comparisons) and among (60 comparisons) populations, respectively. Loci that were not in Hardy–Weinberg equilibrium and therefore might be linked to other loci were not included in subsequent analyses (1 of 10 loci).

Because dispersal behavior differs between cougar sexes, we examined potential differences between the sexes in genetic structure and relatedness and examined male-biased dispersal. We applied the model-based clustering method of Pritchard et al. (2000) to infer population structure from individual genotypes for all cougars, female cougars, and male cougars by using program STRUCTURE (version 2.0, University of Chicago, Chicago, Illinois; http://pritch.bsd.uchicago.edu). This approach represents a Bayesian, model-based clustering method that accounts for the presence of Hardy–Weinberg or linkage disequilibrium by introducing population structure and attempts to find the optimal number of clusters (K) that best fits Hardy–Weinberg equilibrium. We assumed individuals may have mixed ancestry (admixture model) and used only genetic information (excluding information on sampling location) to infer population structure. We examined $K = 1$–6 for all cougars and $K = 1$–3 for males and females. We selected a burn-in period of 30,000 iterations and increased the number of independent runs of the Gibbs sampler by 100,000 for each increase in K; this procedure was repeated 3 or 4 times for each K to enhance consistency of estimates. To examine potential differences in relatedness between males and females, we estimated pairwise relatedness (r_{xy}) by applying the method of Lynch and Ritland (1999) with program IDENTIX (version 4.03, Université Montpellier II, Montpellier, France; http://www.univ-montp2.fr/%7Egenetix/identix_ms.pdf.; Belkhir et al. 2002) and approximated 95% confidence intervals applying $SE = SD/\sqrt{n}$. We compared relatedness of female cougars among populations and relatedness between males and females within populations. Comparisons were limited to populations with sample sizes > 10. We also tested for male-biased dispersal by using the assignment t-test described by Goudet et al. (2002) by using program FSTAT.

We used program MISAT (version 1.0, University of California, Berkeley, California; http://mw511.biol.berkeley,edu/software.html;

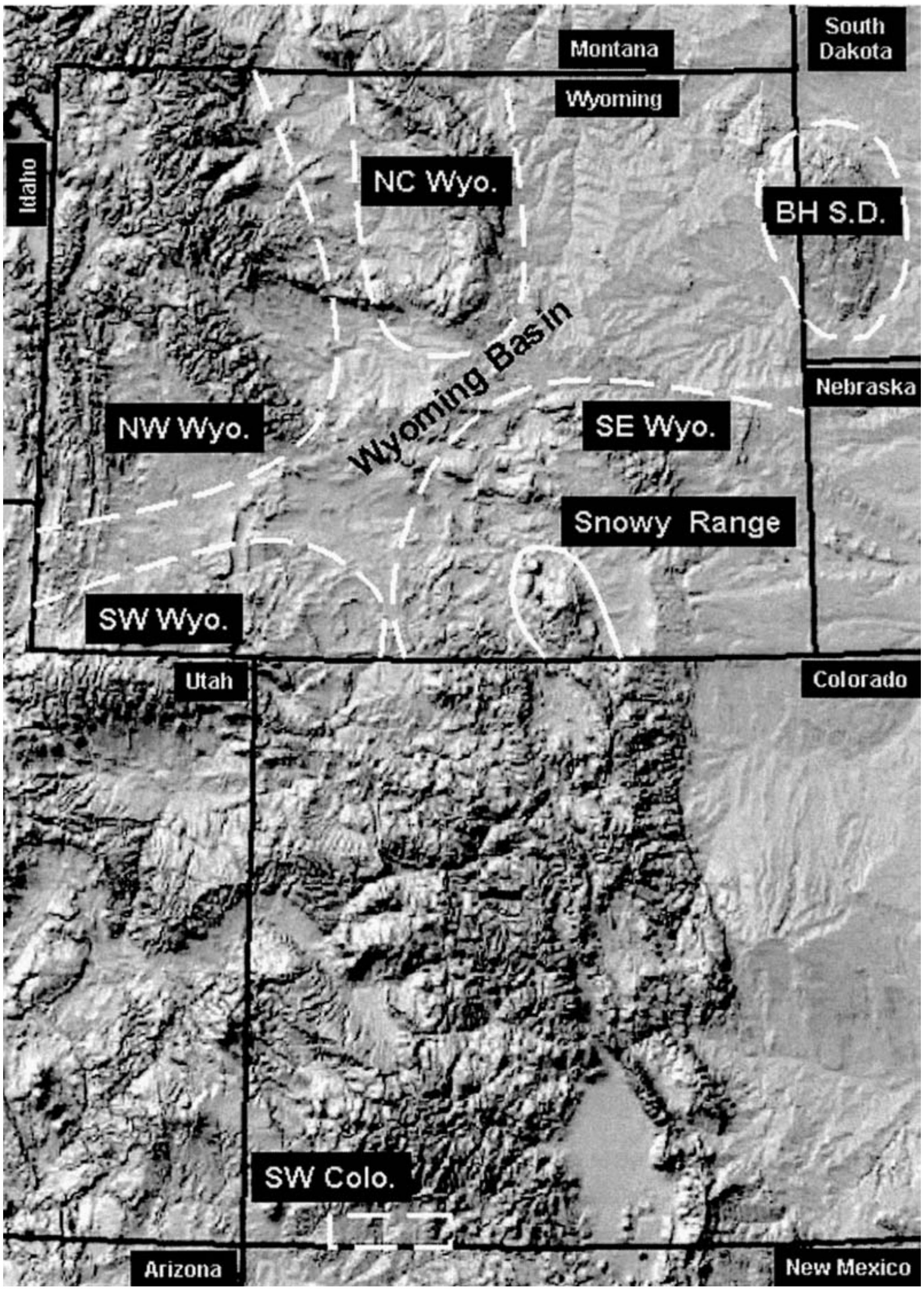

FIG. 1.—Six geographic regions in Wyoming, South Dakota, and Colorado providing cougar DNA samples (dashed lines), and the Snowy Range study site (solid line) in southeastern Wyoming. The Wyoming Basin represents a nonforested region separating mountainous cougar habitats. Coniferous forests dominate mountain ranges (within dashed lines) and sagebrush grasslands characterize basins at lower elevations. BH = Black Hills, NC = north-central, NW = northwest, SE = southeast, and SW = southwest.

Nielsen 1997) to estimate relative effective population size and mutation rate for 5 populations at 9 loci; we excluded the Black Hills population because of small sample size. The program provides a separate maximum likelihood estimate of $4N_e\mu$ (4 times effective population size times mutation rate) for each population–locus combination. To reconcile estimates across loci and populations, we log-transformed the estimates and then used multivariate linear regression to calculate coefficients and estimate relative effective population sizes (assuming constant mutation rate) across populations and loci; we used coefficient standard errors to evaluate differences between populations with 95% confidence intervals. We also reported mean values of $4N_e\mu$ across 9 loci for each population.

Table 1.—Allele size range (base pair [bp] length), number of alleles, and heterozygosities of 312 cougars sampled from Colorado, Wyoming, and South Dakota at 10 microsatellite loci.

Locus	Allele size range (bp)	No. alleles	Observed heterozygosity	Expected heterozygosity
FCA008	148–160	2	0.426	0.448
FCA035	122–136	3	0.571	0.512
FCA043	123–137	5	0.581	0.624
FCA057	146–158	5	0.744	0.679
FCA077	129–133	2	0.222	0.218
FCA081[a]	120–128	4	0.565	0.633
FCA082	239–251	6	0.655	0.691
FCA098	103–119	5	0.738	0.723
FCA132	159–179	5	0.625	0.688
FCA149	112–128	3	0.221	0.227
Mean	—	4	0.535	0.544

[a] Deviated from Hardy–Weinberg equilibrium and was therefore excluded from further analyses.

We estimated effective population size (N_e) for each locus as $N_e = [1/(1 - H_E)^2 - 1]/(8\mu)$ for the model assuming a stepwise mutation process and as $N_e = H_E/4\mu(1 - H_E)$ for the model assuming an infinite alleles mutation process (Lehmann et al. 1998; Nei 1987), where μ is the mutation rate. The stepwise mutation model assumes mutation is a stronger force than genetic drift, whereas the infinite alleles model assumes genetic drift is the dominant force. We also used program MISAT to estimate $4N_e\mu$ across all populations at each locus and then solved for N_e. We estimated N_e by using the average mutation rate from 3 other mammal studies ($\mu = 2.05 \times 10^{-4}$—Rooney et al. 1999).

We examined isolation by distance by comparing pairwise genetic distances and F_{ST} estimates with geographic distances by using the Mantel test (Manly 1991). We also assessed regional phylogenies of 5 cougar populations (excluding the Black Hills where $n = 8$) by constructing neighbor-joining trees from bootstrapped gene frequency data ($\beta = 1{,}000$) by using the SeqBoot, GenDist, Neighbor, and Consense routines in PHYLIP (version 3.5c, University of Washington, Seattle; http://evolution.genetics.washington.edu/phylip.html; Felsenstein 1995). Genetic distances were estimated by using Cavalli–Sforza chord distance, which has been shown to perform well with microsatellite data (Kalinowski 2002) and requires no biological assumptions.

Results

We genotyped 312 cougars from Colorado ($n = 15$), South Dakota ($n = 8$), and Wyoming ($n = 289$) at 10 microsatellite loci. Number of alleles per locus ranged from 2 to 6 and observed heterozygosity varied from 0.221 to 0.744 (overall heterozygosity = 0.535; Table 1). Within-population gene diversity was comparable among populations, ranging from 0.491 to 0.588 (Table 2). Within populations, we found significant deviations from Hardy–Weinberg equilibrium at FCA047 from the southeastern Wyoming population, at FCA081 from the southwestern Colorado population, and at FCA098 from the northwestern Wyoming population ($P < 0.005$). When we examined all populations collectively, we noted that only FCA081 deviated significantly from Hardy–Weinberg equilibrium ($P < 0.0008$). Tests of pairwise genotypic disequilibrium suggested FCA081 and FCA098 were linked ($P < 0.0008$). Because FCA081 deviated from Hardy–Weinberg equilibrium and appeared linked to FCA098, we excluded this locus from further analyses.

Overall F_{ST} and R_{ST} were 0.028 and 0.029, respectively. Pairwise F_{ST} and Nm estimates suggested high gene flow, where effective number of migrants per generation ranged from 2.9 to 30.2 (Table 2). Number of migrants per generation was lowest between the southwestern Colorado cougar population and cougar populations north of the Wyoming Basin ($Nm = 2.9$–3.0) and highest from adjacent cougar populations (e.g., northwestern and north-central Wyoming; $Nm = 10.2$–30.2; Table 2). Inferred population structure from individual genotypes when using program STRUCTURE suggested a single cougar population. Support for a single population was consistent whether the sample included all cougars, only females, or only males (Table 3). Accordingly, relatedness of males and females was similar within populations and did not differ from 0 ($P < 0.05$; Fig. 2), and assignment test results did not support male-biased dispersal ($P = 0.820$). The only hint of female philopatry came from the observation that female cougars from the northwestern and north-central Wyoming populations were less related to cougars in the Snowy Range population than they were to each other (Fig. 2). A neighbor-joining tree based on Cavalli–Sforza distances among the 5 major populations had 98% bootstrap support for a node separating the southeastern and southwestern Wyoming populations, plus the southwestern Colorado population, from the north-central and northwestern Wyoming populations. The 3 southern populations are separated from the 2 northern populations by the treeless expanse of the Wyoming Basin, traditionally considered the dividing line

Table 2.—Pairwise F_{ST} estimates above the diagonal, estimated number of migrants/generation (Nm) between populations below the diagonal, and estimated within-population gene diversity (H_E) along the diagonal from 6 cougar populations sampled at 9 microsatellite loci.

		Cougar population					
Cougar population	n	Northwestern Wyoming	North-central Wyoming	Black Hills South Dakota	Southeastern Wyoming	Southwestern Wyoming	Southwestern Colorado
Northwestern Wyoming	59	0.54	0.008	0.040	0.022	0.017	0.077
North-central Wyoming	59	30.2	0.51	0.024	0.029	0.038	0.076
Black Hills South Dakota	8	6.0	10.3	0.49	0.021	0.051	0.079
Southeastern Wyoming	154	11.1	8.3	11.5	0.55	0.024	0.036
Southwestern Wyoming	17	14.4	6.4	4.6	10.2	0.59	0.048
Southwestern Colorado	15	3.0	3.0	2.9	6.8	4.9	0.53

Table 3.—Inferred number of populations[a] (K) when using 9 microsatellite loci from 6 geographically distinct cougar populations for all cougars ($n = 312$), female cougars ($n = 148$), and male cougars ($n = 164$).

	All cougars		Females		Males	
K	ln $P(X \mid K)$	$P(K \mid X)$	ln $P(X \mid K)$	$P(K \mid X)$	ln $P(X \mid K)$	$P(K \mid X)$
1	−5,417	1.000	−2,593	1.000	−2,887	1.000
2	−5,528	0.000	−2,619	0.000	−2,983	0.000
3	−5,612	0.000	−2,758	0.000	−3,189	0.000
4	−5,936	0.000				
5	−5,992	0.000				
6	−6,117	0.000				

[a] The inferred number of populations was derived from the estimated ln probability of the data [ln $P(X \mid K)$] and the estimated posterior probability of the number of populations [$P(K \mid X)$], where $P(K \mid X) = \exp_{K=1}{}^{\ln P(X \mid K)} / [\exp_{K=1}{}^{\ln P(X \mid K)} + \exp_{K=2}{}^{\ln P(X \mid K)} + \exp_{K=3}{}^{\ln P(X \mid K)} + \cdots]$ (Pritchard et al. 2000).

between the southern and central Rocky Mountains. We also found a significant relationship between pairwise genetic and geographic distances ($r = 0.61, P = 0.011$; Table 4) and an even stronger relationship between pairwise F_{ST} estimates and geographic distances ($r = 0.95, P < 0.001$) supporting an effect of isolation by distance.

Southwestern Wyoming cougars exhibited the largest relative effective population size (N_e), but the confidence interval overlapped those from other populations (Table 5); estimated relative effective population sizes were smallest from the 2 less contiguous populations from terminal mountain ranges in north-central Wyoming and the Snowy Range (Fig. 1). We estimated an effective population size for the central Rockies (applying our estimates of expected heterozygosity from Table 1) of 1,797 when assuming the infinite alleles model and 3,547 when assuming the stepwise mutation model. Solving for effective population size from $4N_e\mu$ averaged over the 9 loci resulted in an estimate of 4,532.

Discussion

Genetic variability of cougars we examined ($H_O = 0.54$) was comparable to that found in other cougar studies in the western United States and intermediate among other large felids and other large carnivores sampled by using microsatellite DNA analyses. Murphy (1998) reported genetic variability of 0.56 from northern Yellowstone cougars, Sinclair et al. (2001) reported 0.47 from Utah cougars, and Culver et al. (2000) reported 0.42–0.52 for cougars sampled from the western United States. Genetic variability of other large felids was estimated to be 0.39 in cheetahs (*Acinonyx jubatus*), 0.66 in African lions (*Panthera leo*—Menotti-Raymond and O'Brien 1995), and 0.77 in leopards (*P. pardus*—Spong et al. 2000). Genetic variation was estimated at 0.30 in Kodiak Island brown bears (*U. arctos*—Paetkau et al. 1998), 0.54 in gray wolves (*Canis lupus*—Roy et al. 1994), and 0.80 in black bears (Paetkau and Strobeck 1994). As suggested by Culver et al. (2000), the moderate level of genetic variability found in western North American cougars may reflect recolonization events following the most recent Pleistocene glaciation.

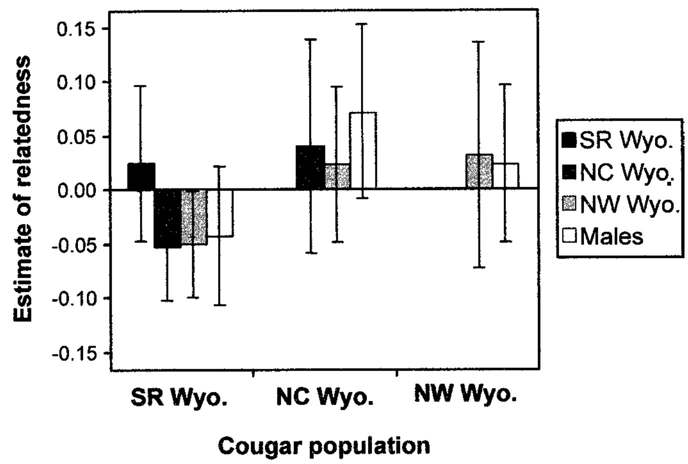

Fig. 2.—Estimated pairwise relatedness (r_{xy}—Lynch and Ritland 1999) of male cougars (white bars) within and female cougars (shaded bars) within and among 3 Wyoming cougar populations (SR = Snowy Range, southeastern Wyoming, NC = north-central, NW = northwest, Wyo. = Wyoming). Error bars represent 95% *CI*. Note similarities between the sexes within populations and slightly negative relatedness between females from the Snowy Range when compared to females from northwestern and north-central Wyoming.

Our findings are similar to those of Sinclair et al. (2001), who reported high gene flow across Utah, and Culver et al. (2000), who suggested North American cougars be reclassified as a single subspecies (*P. concolor couguar*) due to lack of genetic structure. Our low structure and high migration estimates (Table 2) suggest cougar movements are not greatly inhibited by inhospitable habitat (i.e., the Wyoming Basin) or recent development within the Colorado Rocky Mountains; expanses of open habitat across the Wyoming Basin represent distances of about 80–200 km between unconnected, adjacent mountain ranges. These results also were supported by Bayesian simulation methods assigning individuals to a single population regardless of the input pool (e.g., males, females, or both; Table 3). Further, relatedness values were similar within and among cougar populations we surveyed (Fig. 2), and we were unable to detect male-biased dispersal. The only real hint of female philopatry comes from the slightly negative relatedness among females from the Snowy Range compared to elsewhere. However, this difference was not statistically significant (based on overlapping confidence intervals; Fig. 2). We were somewhat surprised to find lack of genetic structure in female cougars and to find that relatedness among females was similar to that among males, despite field-based evidence of a tendency for females to express philopatric behavior and for males to disperse from their natal population (Anderson et al. 1992; Lindzey et al. 1994; Ross and Jalkotzy 1992). This suggests that either high genetic contribution from male immigration is swamping genetic patterns in differential dispersal behavior, or that female dispersal is sufficiently high to preserve genetic cohesiveness, or both. However, the method we used to assess male-biased dispersal provides limited power unless dispersal bias is extreme (80:20—Goudet et al. 2002). One additional factor to consider is postglacial colonization of the area, which

TABLE 4.—Estimated pairwise Cavalli–Sforza chord distances above the diagonal and pairwise geographic distances (km) below the diagonal. Mantel test results ($P = 0.011$) support an effect of isolation by distance and suggest that cougar populations exhibit an equilibrium between migration and genetic drift.

	Population					
Population	Northwestern Wyoming	North-central Wyoming	Black Hills South Dakota	Southeastern Wyoming	Southwestern Wyoming	Southwestern Colorado
Northwestern Wyoming		0.034	0.155	0.060	0.073	0.109
North-central Wyoming	190		0.137	0.070	0.098	0.127
Black Hills South Dakota	550	370		0.113	0.198	0.185
Southeastern Wyoming	450	340	330		0.048	0.074
Southwestern Wyoming	340	360	540	240		0.089
Southwestern Colorado	820	810	820	510	480	

would have an homogenizing effect. In other words, the dominant factor is historical homogeneity, with more recent philopatry not yet evident in genetic data. Also, because isolation by distance is demonstrated, there may be greater geographical structure at a larger geographical scale.

One migrant per generation has been proposed as a necessary minimum to obscure any disruptive effects of genetic drift (Spieth 1974). Mills and Allendorf (1996) investigated this issue further and suggested that more than 1 migrant per generation may be necessary in some cases. Of the 7 criteria they listed (Mills and Allendorf 1996:1516), 2 likely apply to cougar populations, including cases where migrants are closely related to each other or to the local population (Fig. 2) and cases where effective population size is much lower than total population size. Spong et al. (2000) approximated an N_e:N ratio of 0.40 when using cougar data from other studies (Dueck 1990; Harris and Allendorf 1989). As a rule of thumb, Mills and Allendorf (1996) concluded that 1–10 migrants per generation should be sufficient to maintain adequate connectivity while minimizing concerns of local adaptation and outbreeding depression in cases where populations are isolated. The level of gene flow and lack of isolation we observed in the central Rocky Mountains (Table 2) is likely adequate to maintain viable and well-connected cougar populations at the present time.

An effective population size of 500 has been proposed as a minimum to enhance long-term population viability (Franklin 1980). Our estimates of effective population size from cougars in the central Rocky Mountains were well above this minimum

TABLE 5.—Maximum likelihood estimates of $4N_e\mu$ (4 times effective population size times mutation rate) averaged across 9 microsatellite loci and relative effective population size (N_e ratio) and 95% confidence intervals (*CI*) from 4 Wyoming cougar populations and 1 Colorado cougar population.

Population	$4N_e\mu$	N_e ratio[a]	95% *CI*
Southwestern Wyoming	4.54	1.00	0.78–1.22
Southwestern Colorado	4.49	0.96	0.75–1.22
Northwestern Wyoming	3.98	0.85	0.66–1.08
Snowy Range Wyoming	3.66	0.73	0.57–0.93
North-central Wyoming	3.43	0.73	0.57–0.93

[a] Ratio relative to largest effective population subjectively set at 1.00 (southwestern Wyoming).

and ranged from 1,797 to 4,532, depending on the method used and the assumed mutation model. Genetic drift is inversely proportional to N_e, so cougar populations may be similarly driven by both drift and mutation, which was supported by our Mantel test results showing isolation by distance, thereby suggesting that cougars in the central Rocky Mountains exhibit equilibrium between migration and drift. We therefore suggest a provisional estimate of N_e of approximately 2,500, which represents the approximate midpoint of our estimates. Sinclair et al. (2001) reported a much lower effective population size from Utah cougars ($N_e = 571$). When we applied the equations we used assuming the infinite alleles model and the stepwise mutation model (Lehmann et al. 1998; Nei 1987) and using their estimates of expected heterozygosity (Sinclair et al. 2001: table 2, page 261), we calculated N_es of 2,583 when assuming the infinite alleles model and 5,732 when assuming the stepwise mutation model. Although their method was not clearly explained, it was obviously more conservative than ours. However, both studies applied a mutation rate estimated from other mammal species (i.e., Rooney et al. 1999), suggesting these estimates of cougar effective population size be used cautiously until microsatellite mutation rates in cougars are quantified.

Although cougars appear to exhibit metapopulation dynamics in highly developed regions of California (Beier 1996) and possibly in New Mexico (Sweanor et al. 2000), our findings for the central Rocky Mountains are more consistent with a large panmictic cougar population exhibiting rapid and reasonably thorough interchange among subpopulations. The most isolated region we sampled was the Black Hills, which represents the most easterly extension of the Rocky Mountains and is surrounded by grasslands, with the nearest viable cougar populations occurring in the Big Horn Mountains (200 km distant) and the Laramie Mountains (160 km distant) of Wyoming. Historic records suggest the Black Hills population once became greatly reduced or possibly extirpated by the early 1900s (Fecske 2003). Although sample size warrants caution, our findings suggest that extirpation, if it occurred, was brief and genetic cohesiveness was maintained, evidenced by similar estimates of gene flow, heterozygosity, and structure relative to north-central and southeastern Wyoming cougar populations (Table 2). We suspect isolated conifer islands and riparian corridors may have provided movement pathways for immigrants from these areas, which are largely undeveloped and may warrant some protection to maintain connectivity in the future.

Although our results support high gene flow, hints of genetic structure were evident in the slightly negative relatedness of females between cougar populations separated by the Wyoming Basin (Fig. 2). The neighbor-joining tree's split between the southern and northern Wyoming populations coincides with a biogeographic divide between the southern and central Rocky Mountains. Findley and Anderson (1956) pointed out that the Wyoming Basin marks the boundary for morphologically based subspecific breaks in at least 6 mammalian species. Conroy and Cook (2000) found an estimated 350,000-year break in the mitochondrial DNA of the long-tailed vole across this same divide. Our results, although regional in geographic coverage, therefore have implications for a wider region of the North American west. The region we examined represents an area of low human density (among the lowest in the western United States) that could be impacted by future development (e.g., Beier 1996). Thus, periodic monitoring of cougar genetics throughout the western United States to identify changes seems prudent, as does combining results of genetic studies from other regions to determine if cougars are structured at larger geographic scales.

Conclusions

Cougars in the central Rocky Mountains exhibit high gene flow and low structure, presumably because high male dispersal suffices to maintain connectivity between subpopulations. Positive associations between genetic and geographic distances suggest an equilibrium between migration and genetic drift in the historic range of cougars and a lack of significant barriers to gene flow. These attributes are not consistent with metapopulation structure, which requires that subpopulations experience periodic extinctions. Rather, cougars in this region are best considered a large panmictic population. Management and conservation efforts will benefit from periodic monitoring of cougar population structure that will allow detection of fragmentation due to future human development or excess mortality (e.g., disease and exploitation) and determination of whether cougar populations are structured at larger spatial scales. However, because genetic studies will mostly provide insight into past events, periodically assessing status of cougar subpopulations and maintaining habitat corridors sufficient to maintain connectivity will be important to maintain long-term viability.

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Living with Wildlife Foundation – Predator Guides project

Living with Wildlife Foundation

Progress Report on Living with Predators Guides Project

July 2005

The following is a progress report on the *Living with Predators Resource Guides* project which is provided as a requirement for grant funds received from the Wyoming Animal Damage Management Board in 2004.

Three separate guides containing information relevant to co-existing with predators are in the process of being updated and are nearing completion and distribution. The first edition of the guides was produced in 2003 and each of the guides is described below:

- The *Recreating in Bear, Wolf and Mountain Lion Country* guide covers products and techniques which can be used while recreating in wild areas. Information about various types of bear-resistant backpacking containers for food and garbage storage, techniques for hanging gear in the back country, portable electric fencing, outfitters' panniers, and other information is included.

- The *Techniques and Refuse Management Options for Residential Areas, Campgrounds, and Group-Use Facilities* guide covers bear-resistant containers and other products and methods for securing and storing garbage, livestock feed, pet food and other attractants. The guide also contains information on methods for deterring predators from your property, including information about electric fencing.

- The *Predator Behavior and Modification Tools* guide contains information for wildlife professionals who deal with predator conflicts. The guide includes information on tools and non-lethal methods for deterring predators, as well as for trapping and aversively conditioning predators when necessary.

We have added a fourth guide this year that describes electric fencing options and designs for predator exclusion. The guide, entitled “Practical Electric Fencing Guide: Controlling Predators’” is nearly completed and will be made available in August of 2005. The guide contains pictures and specifications for species-specific electric fencing configurations as well as examples of fences that are currently being used to exclude predators.

Funds received from the ADMB grant are being used to update the original three resource guides and to produce the new electric fencing guide. We are also repaginating the original three guides to make it easier and less costly to update them in the future. The new page system will allow for old pages to be discarded and updated pages to be easily inserted. The original format did not allow for this. All four of the guides will be available for distribution in August of 2005.

ADMB funds have allowed us update the guides to include approximately 30 new products which have been developed or we have become aware of during the past year. We have also deleted products which are no longer available. The last part of the update involves incorporation of results of product testing that has been conducted under the Living with Wildlife Foundation, Montana Fish, Wildlife & Parks, Grizzly & Wolf Discovery Center, and Interagency Grizzly Bear Committee Bear-Resistant Products Testing Program. Consumers will now be able to determine which products have been carefully evaluated for their “bear-resistancy” before making a purchasing decision.

Several state agencies printed hard copies of the first edition of the guides for bear managers to carry into the field and to use in visits with landowners who are experiencing conflicts with predators. CD’s of the guides have also been included in homeowner association informational packets and materials distributed at community meetings and workshop materials to assist private landowners in selecting appropriate bear-resistant storage containers for their own use.

We will publish information about the availability of the second edition of the *LWP Resource Guides* in various professional publications and outdoor magazines, and present this information at conferences and professional meetings when possible.

Compilation and production of the *Living with Predators Resource Guides* is viewed as an ongoing project. To maintain the reliability and expand the usefulness of the guides as a reference source, they must updated on a continuing basis to delete discontinued products, to add new products and new predator-proofing techniques, and to include test results for products which have been tested under the bear-resistant product testing program.

We will forward a CD containing the finalized updated guides to you as soon as they are complete.

Thank you again for your support for this vitally important project.