The Intermountain Journal of Sciences (IJS) is a regional peer-reviewed journal that encourages scientists, educators and students to submit their research, management applications, or viewpoints concerning the sciences applicable to the intermountain region. Original manuscripts dealing with biological, environmental, health and human development, mathematics, molecular-cellular, pharmaceutical, physical and social sciences are welcome.

Co-sponsors/publishers include the Montana Academy of Sciences, the Montana Chapters of The Wildlife Society and The American Fisheries Society. It is the intent of the governing bodies of the co-sponsoring organizations that this journal replace and standardize printed proceedings from the respective annual meetings. Format and style should follow the *Guidelines for Meeting Abstracts Submitted to the Intermountain Journal of Sciences, 1st revision 2016.** It is the policy of the editorial board that abstracts from presentations at annual meetings be published in the last issue of *IJS* for that year of the annual meeting. Submission of manuscripts for review and publication without regard to membership is encouraged.

Baseline funding is provided by the co-sponsoring organizations. Long-term funding will be derived from page charges assessed manuscript authors at $60/page, sponsoring organizations at $40/page for annual meeting abstracts and annual subscriptions: student - $6, regular member - $15, patron member - $25, international member - $25 and library - $25. One time subscriptions are: life member - $150 and sustaining subscriber - $2,500.

The intent of the co-sponsors and editorial board is that *IJS* be expanded to a quarterly journal. Achieving that objective depends upon numbers of acceptable manuscripts received and available funding. The editorial board's policy is that contributing authors be assured of publication within 12 months of acceptance of their manuscript. It is also intended that *IJS* be converted to an eJournal.

The organizational staff is voluntary and consists of an editorial board, an editor-in-chief, a managing editor, associate editors, a business manager and a panel of referees. The editorial board is responsible for establishing policy and the chair of the editorial board serves as liaison to the editor-in-chief and managing editor. The editor-in-chief is responsible for determining acceptability and level of revision of manuscripts based on referees' comments and recommendation of an associate editor. The managing editor serves as supervisor for layout and printing and liaison to the sponsoring organizations. Associate editors and referees are selected on the basis of their field and specific area of knowledge and expertise.

Associate editors and referees judge submitted manuscripts on originality, technical accuracy, interpretation and contribution to the scientific literature. Format and style should follow the *Guidelines for Manuscripts Submitted to the Intermountain Journal of Sciences, Dusek 1995, 2nd revision 2016.** Organization may vary to accommodate the content of the article, although the text is expected to elucidate application of results.

Financial Statement (1/01/19 - 12/31/19)

Balance 01/01/19	**$4,132.85**

Income:

Subscriptions:

Regular Member	$75.00
Library Subscriptions	$175.00
Patron Member	$50.00
International Member	$25.00
Subscriptions Total	**$325.00**

MSU Library Services	$250.00
Page Charges	$6,260.00
Reprints and PDFs	$100.00
Back Issues	$5.00
Total Income	**$6,940.00**

Expenses:

Layout and Printing	$5,862.96
Postage	$315.31
P. O. Box Rental	$154.00
Administrative and Bank Fees	$303.72
Reprints & PDFs	$150.00
Storage	$377.00
Website Hosting	$110.00
MSU Library Service (2019 Archiving Fee IJS)	$975.00
Total Expenses	**$8,247.99**

Balance 12/31/19	**$2,824.86**

Fred Nelson, Business Manager

EDITORIAL REVIEW POLICY

The *Intermountain Journal of Sciences* (IJS) is a fully refereed journal.

Manuscripts are submitted to the Editor-in-Chief (EIC) for initial consideration for publication in the IJS. This review shall include, but not be limited to, appropriateness for publication in IJS, correct formatting and inclusion of a letter of submittal by the author with information about the manuscript as stated in the "Guidelines for manuscripts submitted to the *Intermountain Journal of Sciences*" (Dusek 1995, 2007) available on the IJS website, www.intermountainjournal.org under the Publish tab. This cover letter must also include a statement by the author that this paper has not been submitted for publication or published elsewhere. The EIC notes the date of receipt of the manuscript and assigns it a reference number, IJS-xxxx. The EIC forwards a letter of manuscript receipt and the reference number to the corresponding author. The corresponding author is the author who signed the submittal letter.

Three hard or digital copies of the submitted manuscript, with copies of the "Guidelines and checklist for IJS referees" attached are forwarded to the appropriate Associate Editor. The Associate Editor retains one copy of the manuscript and guidelines for his/her review, and submits a similar package to each of two other reviewers. A minimum of two reviewers, including the Associate Editor, is recommended for each manuscript. The two reviewers are instructed to return the manuscript and their comments to the Associate Editor. The Associate Editor then returns all manuscript copies and reviewer comments plus a recommendation for publication, with or without revisions, or rejection of the manuscript to the EIC. This initial review process is limited to 30 days.

The EIC then reviews the recommendations and all comments and notifies the corresponding author of the results of the review and the publication decision.

ACCEPTANCE

For accepted manuscripts, each copy of the manuscript containing comments thereon and other comments are returned to the corresponding author. Revised manuscripts are to be returned to the EIC in hard copy and four copies if further review is required. These copies can be submitted in digital form by email. The revised manuscript shall be returned to the EIC within 14 days of notification. Review of the revised manuscript by the Associate Editor and reviewers shall be completed and returned to the EIC within 14 days. An accepted manuscript will then be forwarded to the Managing Editor (ME) for final processing.

REJECTION

Each manuscript that is rejected for publication is returned by the EIC to the corresponding author along with the reasons for rejection. The author is also advised that the manuscript may be resubmitted, provided all major criticisms and comments have been addressed in the resubmitted manuscript. The resubmitted manuscript may be returned to the initial review process if deemed appropriate by the EIC. If the manuscript is rejected a second time by either the EIC or the Associate Editor and reviewers, no further consideration will be given for publication of the manuscript in IJS. The corresponding author will be notified of this decision.

REVIEWER ANONYMITY

The identity of all reviewers shall remain anonymous to the authors, called a blind review process. All criticisms or comments by authors shall be directed to the EIC; they may be referred to the ME or the Editorial Board by the EIC for resolution.

Manuscripts Submitted by Editors

Each manuscript submitted by an Associate Editor shall be reviewed by the EIC and a minimum of two other reviewers with expertise in the subject being addressed. Each manuscript submitted by the EIC shall be forwarded with the necessary review materials to the ME or chairman of the editorial board, who will serve as the EIC for that manuscript.

Abstracts

Only abstracts submitted from the annual meetings of the sponsoring organizations will be published in IJS. Other submissions of abstracts shall be considered on a case-by-case basis by the Editorial Board. Sponsoring organizations shall collect abstracts, review them for subject accuracy, format them in Microsoft Word and email them to Rick Douglass, the EIC (RDouglass@mtech.edu), on or before November 1. Each abstract shall be reviewed by the EIC to assure proper grammar, compliance with IJS Guidelines and for publication in the December issue of IJS. The Guidelines for Submitting Meeting Abstracts (Presentation or Poster) are available as a pdf on the IJS website under the Publish tab.

Commentary

Submissions concerning management applications or viewpoints concerning current scientific or social issues of interest to the Intermountain region will be considered for publication in the "Commentary" Section. This section will feature concise, well-written manuscripts limited to 1,500 words. Commentaries will be limited to one per issue.

Submissions will be peer reviewed and page charges will be calculated at the same rate as for regular articles.

Literature Cited

Dusek, Gary L. 1995, revised 2007. Guidelines for manuscripts submitted to the *Intermountain Journal of Sciences*.Int. J. Sci. 1(1):61-70. Revised guidelines are available on the Intermountain Journal of Sciences web site: (www.intermountainjournal.org)

Age-Mass Relationships for Beavers in Montana

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Abstract

Beavers (*Castor* spp.) are receiving increased attention due to their impact on ecosystems and potential for use in stream restoration. Beaver research and relocation projects are especially common in the western United States, and professionals using live-captured animals for projects will benefit from a reliable technique for aging live-caught beavers. The only reliable technique for aging live beavers without sedation is to estimate age based on mass, but estimates of the age-mass relationship for beavers vary regionally and are not adequately quantified in the western United States. We collected beaver carcasses and skulls from trappers throughout southwest Montana to estimate the age-mass relationship using a robust sample collected from a large geographic area. We weighed beaver carcasses and extracted molar teeth from the mandibles to estimate age by counting cementum annuli on cross-sections of the teeth. We collected 193 beaver carcasses and hanging weights from nine major river drainages in Montana. Multiple regression analysis indicated the top prediction equation was mass = 9.4611 + 8.2234 × log(age) + drainage, indicating drainage-level differences in the average mass of beavers. Beavers from the Ruby, Jefferson, and Yellowstone River drainages were larger than those from the other river drainages in Montana. We could reliably separate beavers into four age classes: kits, yearlings, two-year-olds, and adults (>3 years). Our results are useful for researchers who need to estimate beaver age to understand population dynamics and age-specific life history characteristics, as well as restoration practitioners who need to determine colony compositions and recruitment rates to evaluate the success of restoration projects.

Key words: age-weight curve, beavers, Castor canadensis, Montana

Introduction

Humans and wildlife depend on riparian areas and wetlands to enhance landscape-scale water storage capacity and bolster the resilience and connectivity of ecosystems. An extensive body of scientific literature recognizes the habitat-modifying activities of beavers (*Castor canadensis*) as instrumental in the creation, expansion, and maintenance of healthy and productive riparian and wetland areas (Naiman et al. 1988, Collen and Gibson 2001, Wright et al. 2002). As a result, beavers are increasingly used as a tool for habitat restoration, especially in the western United States where water resources are strained by increasing demand and ongoing drought (Baker 2003, Barnett et al. 2008, Hidalgo et al. 2009, Pollock et al. 2017). Projects aimed at recovering beaver populations in areas of their historic range are increasing in popularity and scope, and research directed towards understanding beaver population dynamics, habitat selection, and influence on ecosystems will be important in the future management of this species.

Beavers are territorial mammals that live in well-defended colonies generally composed of a mating pair of adults, kits, and yearlings (Muller-Schwarze 2011). In large colonies located in good habitat, sub-adult beavers between two and four years of age may also be present (McTaggart and Nelson 2003, Muller-Schwarze 2011). The presence of extra adults in the colony means researchers and restoration practitioners relying on live-captured beavers for their work will be capturing a wide range of beaver age classes in a given colony. Researchers may want to estimate the age of captured beavers to evaluate colony size and composition and to study age-specific processes such as dispersal

and breeding. Restoration practitioners looking to translocate beavers may wish to selectively remove beavers from a source colony without disrupting the breeding pair or translocating vulnerable kits. Additionally, restoration practitioners may want to monitor colonies established as part of a restoration effort to evaluate age composition of the colony and recruitment rates.

There are few reliable techniques for aging live beavers, and most require sedation or heavy restraint in order to safely gather measurements (Patric and Webb 1960, Layne 2003). Aging beavers via inspection of cementum annuli on cross-sections of teeth is the preferred method to age beavers, but is not possible with live animals (Van Nostrand and Stephenson 1964, Novak 1987). Many authors have proposed using the body mass of captured beavers to differentiate age classes (Bradt 1939, Hammond 1943, Patric and Webb 1960, Payne 1979, Van Deelen 1991, McTaggart 2002, Layne 2003). However, regional variation in growth rates driven by differing food resources and climatic conditions can cause the relationship between the age of beavers and body mass to vary widely across study areas (Table 1).

Calibration of the age-mass relationship for beavers has been especially lacking in the western United States, where the few studies conducted relied on small samples of beavers from one or two drainages (Townsend 1953, Van Deelen 1991). Accurate estimation of the age-mass relationship for beavers will improve current and future research projects in southwest Montana and similar habitats within the Greater Yellowstone Ecosystem (GYE). Additionally, beaver reintroduction programs, which are most common in the western United States, will benefit from a reliable technique for aging live-captured beavers to determine colony composition and select appropriate individuals for release at restoration sites.

To address the shortcomings in field-based age estimation for beavers, we initiated a study to estimate the age-mass relationship for beavers in southwest Montana. Specifically, our objectives were to: 1) provide a region-specific calibration of the age-mass relationship for beavers inhabiting willow- and cottonwood-dominated streams and rivers, and 2) evaluate drainage-level variation in age-mass relationships for beavers in southwestern Montana. Our goal was to provide researchers and managers with an accurate and efficient tool for estimating the age of live-captured beavers in the field.

METHODS

We collected beaver carcasses or skulls from recreational trappers throughout southwest Montana during Fall 2015−Spring

Table 1. Estimated age-mass (kg) relationships for beavers from various projects in North America, 1943–2018.

Authors	Location	Kits (0-1 yr)	Yearlings (1-2 yr)	Two-year-olds (2-3 yr)	Adults (> 3 yr)
Ritter and McNew, *this study*	Southwest Montana	< 7.8	7.8–14.3	14.3–17.8	> 17.8
Hammond (1943)	North Dakota	—	4.1–11.3	11.8–20.8	15.9–27.2
Townsend (1953)	Montana	3.6–5.4	9.1–11.8	> 13.6	—
Beer (1955)	Minnesota	< 4.5	5.4–11.8	> 13.6	> 13.6
Patric and Webb (1960)	New York	< 6.8	6.8–10.8	10.9–16.0	> 16
Brooks et al. (1980)	Massachusetts	< 6	6–11	11–15	> 15
Van Deelen (1991)	Western Montana	< 6.5	6.5–10.5	10.5–14.5	> 14.5
McTaggart (2002)	Central Illinois	3.2−11.4	10−19.1	15−23.6	> 15.5

2017. To make age-mass calibrations regional, we limited the sample to within 500 km of Montana State University (Bozeman, MT). We contacted trappers with the assistance of biologists with the Montana Department of Fish, Wildlife and Parks (MFWP), local game damage specialists, the Montana Trapper's Association newsletter and e- mail list, and e-mail lists for conservation and outdoor recreation groups in the area. We asked trappers to submit whole beaver carcasses, skulls, or mandibles and record the mass of each beaver they caught with the pelt on. Many trappers also provided the sex of each beaver. We obtained the general location of harvested beavers and grouped them by major river drainage (Fig. 1).

We processed all samples at the MFWP Wildlife Disease Lab in Bozeman, MT. We separated the lower mandible from the skull of each beaver and extracted a molar tooth for use in age determination by cementum annuli. To extract the teeth, we soaked mandibles in water kept just below boiling for approximately three minutes. We then wrapped the mandibles in cloth and struck them with a hammer, targeting the thickest part of the mandible where the ridges coming off the condylar process and angular process meet. We then extracted teeth from the broken mandible parts. We soaked teeth in a 70% ethanol solution and then in Nolvasan Solution (Zoetis, Inc.; 0.8% concentration) for 30 seconds before drying the teeth on a paper towel and depositing them in uniquely marked coin envelopes. We submitted teeth to Matson's Laboratory (Manhattan, MT USA) for aging via inspection of cementum annuli. The lab returned a best estimate of age in years for each beaver tooth sample assuming a common birth date of 1 June each year.
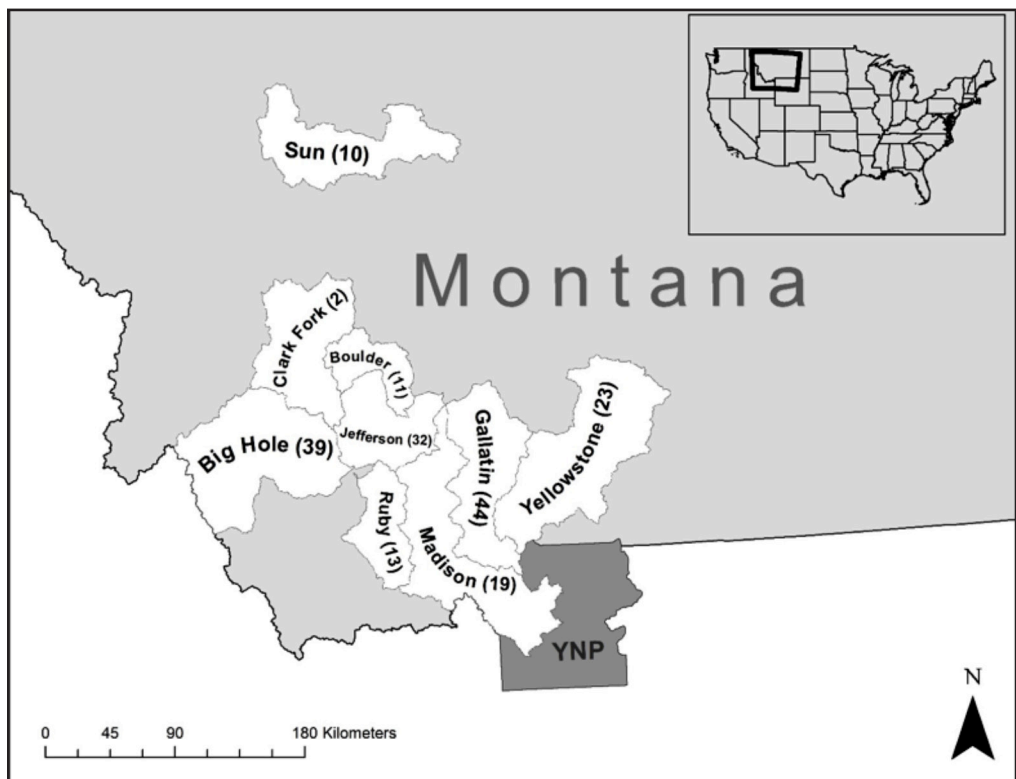

Figure 1. Major river drainages in southwest Montana, USA, where we obtained carcasses and skulls from trappers during 2015–2017 to estimate the age-mass relationship for beavers. The number of beavers submitted from each drainage are denoted in parentheses. No beavers were captured from within Yellowstone National Park (YNP).

We used multiple linear regression to evaluate the relationship between age determined from cementum annuli and hanging mass of carcasses. We constructed and analyzed models using R Statistical Computing Software (R Version 3.3.2, www.r-project.org, accessed 11 Feb 2018). The residuals of the independent variable for age were not normally distributed so we log- transformed the variable and examined residual plots to determine if the assumption of homoscedasticity was reasonably met with transformation. We fit and evaluated four linear models using mass as the response and tested a main effect of age in years, an additive effect of drainage, an interaction between age and drainage, and an intercept-only null model. We ranked models using Akaike's Information Criterion adjusted for small sample sizes (AIC$_c$; Burnham and Anderson 2002). We considered all models ≤ 2 AIC$_c$ from the top model to be parsimonious. Goodness of fit was evaluated for each competing model using adjusted R-squared. We used the coefficients from the top model to predict the age of beavers based on the entire range of beaver masses observed in the study area, and examined the means, standard deviations, and ranges of beaver masses representing each year of age to evaluate support for separation of ages based on the predictions from the top-ranked model. We used a Student's t-test to evaluate differences in mean mass between subsequent ages.

Results

We obtained teeth and hanging weights from 174 beavers collected by 13 different trappers in southwest Montana during Fall 2015−Spring 2017. Beavers were taken from nine major river drainages (Figure 1). Not all trappers reported locations of trapped beavers to individual stream, but the sample of beavers were harvested from a minimum of 27 different streams. Due to low pelt prices, few trappers were targeting beavers over the two years of the study. Although we directly contacted > 25 trappers, the majority of the samples came from eight individuals. We acquired an additional 19

beavers opportunistically during beaver activity surveys and live-capture trapping efforts for a related study on settlement site habitat selection (Ritter 2018), bringing the total sample size to 193 beavers. Trappers recorded sex for 101 beavers (45 males, 56 females); masses were similar between the sexes (males = 16.6 ± 0.83 SE kg, females = 16.2 ± 0.83 kg) so we did not include sex as a covariate in age-mass models.

An initial screening of the mass distributions by age-class suggested an asymptotic relationship between age and mass, with beavers experiencing rapid growth early in life and slower growth as they age (Fig. 2). Beavers typically ranged from 1−8 years old and weighed 2.3−31.3 kg, although one captured beaver was estimated via cementum annuli to be 11 years old. The distribution of ages was strongly skewed towards younger beavers between one and three years of age. Two-year-old beavers were the most common age making up 34% of the sample (Table 2).

The top prediction equation using data pooled over all drainages was mass = $9.5911 + 7.9375 \times \log(\text{age})$ (adjusted R^2 = 0.63, SE = 3.74, N = 193). There was little model uncertainty among the candidate set of models (Table 3). The top-ranked model contained an additive effect of river drainage and accounted for 69% of the candidate set support. The 2nd-ranked model was not considered parsimonious but accounted for 31% of the model support and contained an interaction effect between drainage and age. Confidence intervals on the coefficient estimates for the top model indicated drainage-level differences in the mass of beavers across all ages (Table 4). Beavers in the Yellowstone, Ruby, and Jefferson River drainages were larger overall than those in other drainages (Fig. 3). Beavers from the Big Hole, Boulder, Sun, Madison, and Gallatin River drainages were all similar in size.

Although beaver masses varied within age-classes, we were able to reliably separate one- and two-year-old beavers by mass, with reduced confidence in the separation of two- and three- year-old
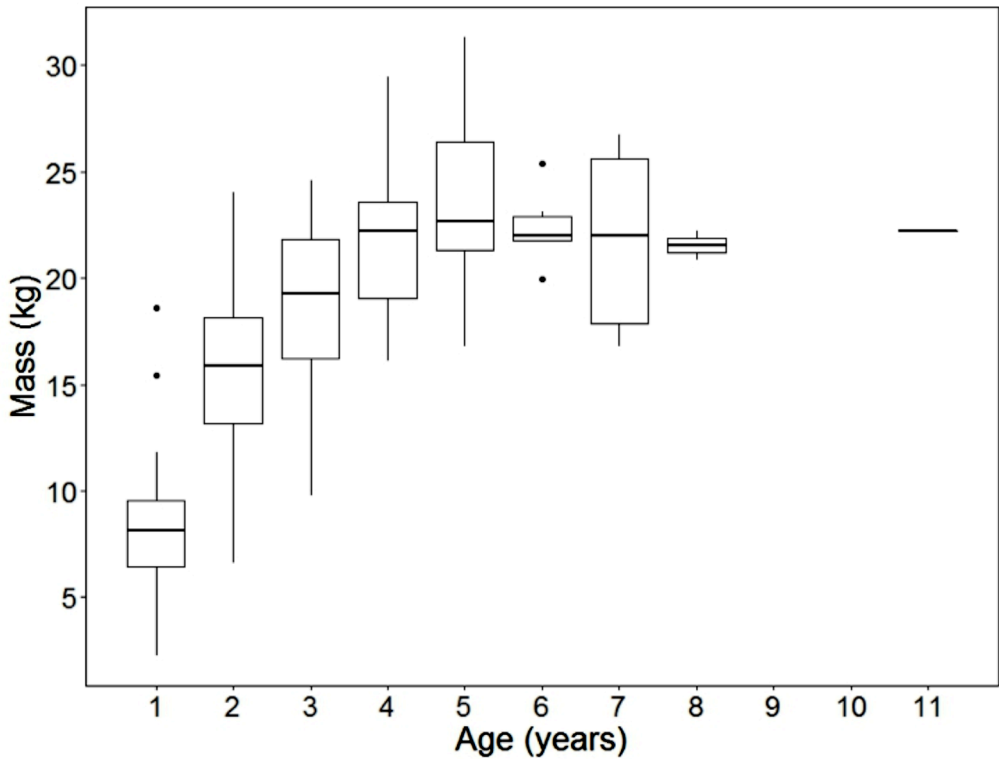
Figure 2. Relationship between age and mass for 193 beaver carcasses obtained during fall 2015−spring 2017 in southwest Montana, USA. Ages were determined through inspection of cementum annuli on molars extracted from the lower mandibles.

Table 2. Distribution of ages and masses (kg) from 193 beaver carcasses collected throughout southwest Montana, USA, during fall 2015−spring 2017.

Age (years)	Number of samples	Mean mass (95% CI)	Range	P [a]
1	46	8.4 (7.5–9.3)	2.3–18.6	—
2	66	15.9 (15.0–16.9)	6.6–24.0	< 0.001
3	33	18.9 (17.6–20.1)	9.8–24.6	< 0.001
4	18	21.8 (20.1–23.6)	16.1–29.5	0.0092
5	11	23.7 (21.0–26.4)	16.8–31.3	0.26
6	6	22.4 (20.9–23.9)	20.0–25.4	0.41
7	10	21.8 (19.5–24.1)	16.8–26.7	0.68
8	2	—	20.9 and 22.2	—
9	0	—	—	—
10	0	—	—	—
11	1	—	22.2	—

[a] *P*-value result of Welch's t-test comparing mean beaver mass between each age-class and the mean mass of the previous age-class.

beavers as well as three- and four-year-old beavers (Table 2). Identification of beaver ages beyond four years was not possible using mass. The model with just the effect of mass had an adjusted R^2 value of 0.63 and was used to offer overall recommended mass ranges for beavers in southwest Montana (Table 1). However, this model was poorly

Table 3. Model selection results testing the influence of age (years) on mass for beavers in southwest Montana, USA, 2015–2017.

Model	K	AIC$_c$	ΔAIC$_c$	w_i	Cum w_i
age + drainage	10	1033.10	0.00	0.69	0.69
age × drainage	17	1034.68	1.57	0.31	1.00
age	3	1051.24	18.14	0.00	1.00
~ 1 (null model)	2	1237.79	204.69	0.00	1.00

Table 4. Recommended mass ranges (kg) for beavers in eight major river drainages in southwest Montana, USA.

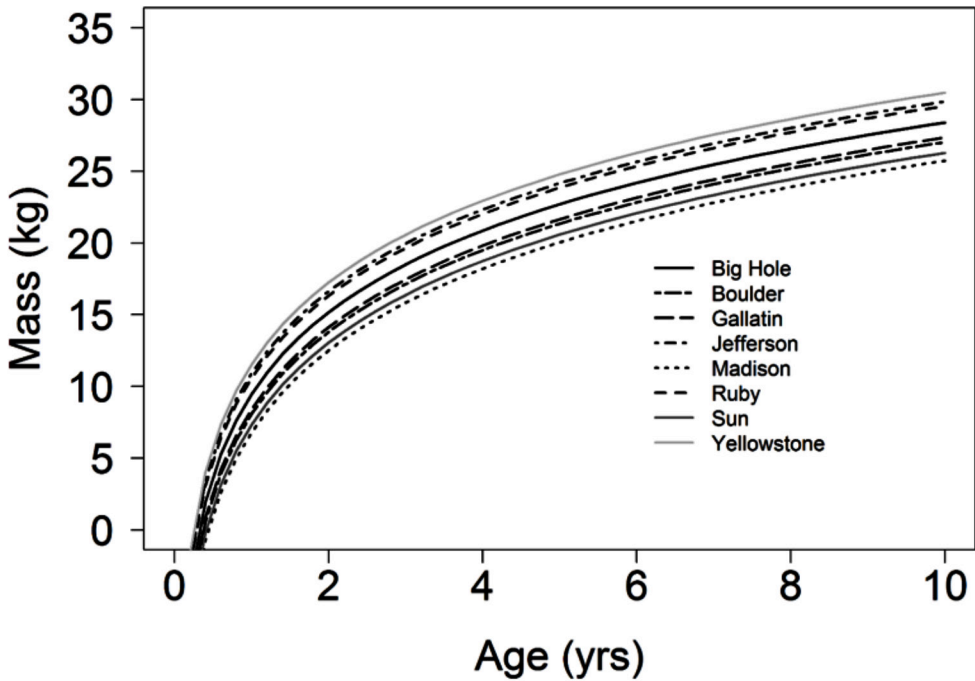| Drainage	Kits	Yearlings	Two-year-olds	Adults
Madison	< 5	5–11.5	11.5–15.5	> 15.5
Sun	< 5.5	5.5–12	12–16	> 16
Boulder	< 6.5	6.5–13	13–16.5	> 16.5
Gallatin	< 6.5	6.5–13.5	13.5–17	> 17
Big Hole	< 7.5	7.5–14.5	14.5–18	> 18
Ruby	< 9	9–15.5	15.5–19	> 19
Jefferson	< 9	9–16	16–19.5	> 19.5
Yellowstone	< 9.5	9.5–16.5	16.5–20	> 20

Figure 3. Estimated growth curves for beavers (n = 193) from eight major river drainage in southwest Montana, USA, 2015–2017.

supported in the candidate set, and the retention of drainage as a variable in the top model suggests mass ranges specific to individual river drainages are more accurate (adjusted $R^2 = 0.67$; Table 4).

DISCUSSION

As expected, our results suggest beavers grow rapidly in the first 1–3 years of life then growth rates slow beyond three years of age. We could not reliably separate kits from one-year- old beavers using mass, but other authors have recommended kits vary from 3.2–11.4 kg (Table 1). Our top model predicted beavers less than one year old are generally less than 5 kg, which is within the range of other studies. The reliability of our estimates of mass ranges decreased as beavers got older (Table 2). While separation of one- and two-year-old beavers was highly reliable, separation of two- and three-year-old beavers as well as three- and four-year-old beavers was only moderately reliable. Our results are consistent with other studies that have found age determination difficult for live-captured beavers beyond three years of age (Layne 2003).

The age distribution of our sample was skewed towards younger animals, and it is unclear if this represents an accurate age distribution for southwest Montana beavers overall. The age distribution of beavers gathered from trappers may not represent actual age distributions in a given area. Larson (1967) suggested beavers harvested by trappers may be skewed toward larger animals as trappers target beavers with more valuable furs. However, Novak (1977) found no bias in age distribution from trapper harvests in Ontario, Canada. McTaggart (2002) observed a similar age distribution as ours in Illinois where beavers were trapped with a more systematic protocol. Larson (1967) also noted a similar age distribution in Maryland from trapper-submitted beaver carcasses, but noted a drop in the number of two-year-old beavers which he attributed to those beavers being missed by trappers due to dispersal. Unlike Larson's study, we found two- year-old beavers were the most common age submitted by trappers. Due

to low market prices for beaver furs during our study, a majority of the beavers in our sample were trapped due to property damage complaints. Stream sections where beavers must be trapped due to property damage are commonly recurring issues where it is likely young, naïve beavers are repeatedly moving into the sites that appear to be open habitat. It is therefore possible our sample was biased towards younger animals which are more likely to have recently dispersed and settled in areas where they are not tolerated by humans.

Beavers in the Yellowstone, Ruby, and Jefferson River drainages were larger than in other drainages. It is unclear why there were dissimilarities, but there are notable differences in environmental conditions among drainages. A large proportion of the beavers in the Yellowstone, Ruby, and Jefferson River drainages came from colonies in or near spring creeks. Beavers in spring creeks may take advantage of stable water temperatures that enhance plant growth and limit ice cover which allows access to quality forage for a longer portion of the year compared to other drainages. Year-round access to forage may allow beavers in the Yellowstone, Ruby, and Jefferson River drainages to maintain or put on weight in the winter, while beavers in other drainages may maintain or lose weight due to being ice-bound during the winter.

We compared our overall growth curve to those of Payne (1979) and Van Deelen (1991) and found beavers grew at a faster rate in our study area (Fig. 4). Payne (1979) examined beavers in Newfoundland but did not report on the food source or winter conditions associated with beaver habitat in his study area, making comparisons difficult. Van Deelen (1991) collected beavers in western Montana and incorporated data from Jackson (1990) in the same study area. Beavers in his study area live under similar climatic and habitat conditions to our study, with mountain streams flowing through willow-dominated riparian areas. Unlike Van Deelen (1991) a large portion of our sample came from spring creeks which may explain why our data resulted in faster estimated
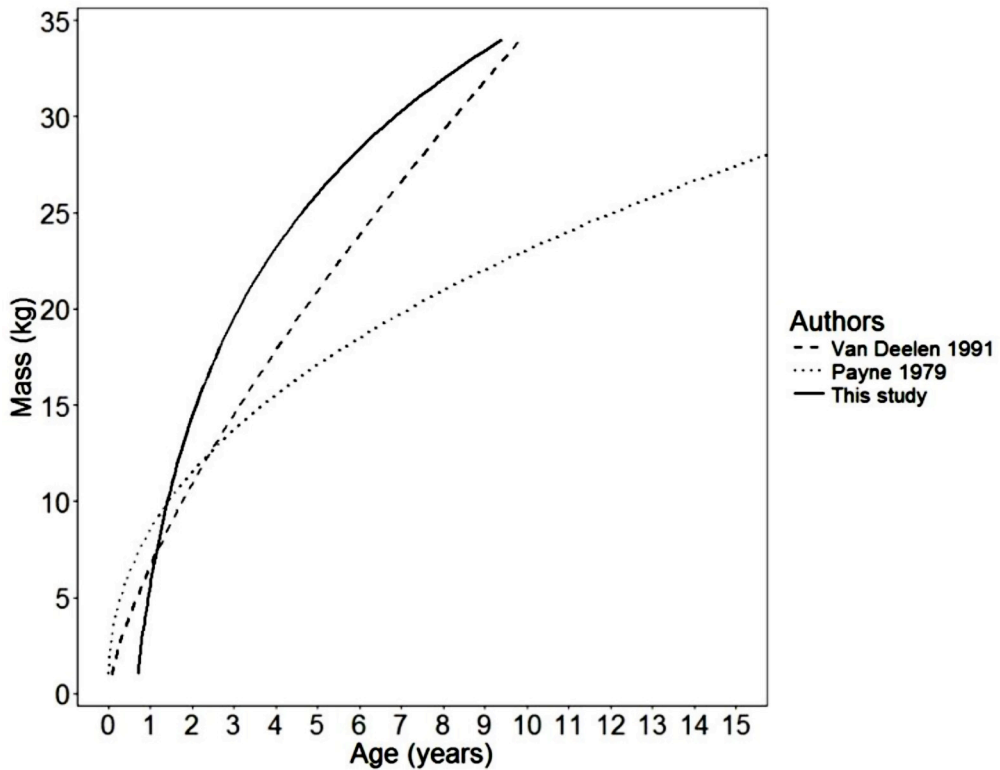
Figure 4. Estimates of the age-mass relationship for beavers from three studies in North America.

growth rates.

This study provides regionally calibrated growth curves allowing for estimation of the age of beavers by mass. Researchers, trappers, wildlife managers, and stream restoration practitioners can use the results of this project to more reliably age live-captured beavers. While our growth estimates were calculated from a relatively large sample size, there is still wide variation in beaver mass across ages which is likely due to differences in habitat conditions among individual colonies in a given drainage that allows members of some colonies to grow faster than others. We recommend future researchers acquire colony-specific locations for beavers if trappers are willing to provide such information. Colony membership information would allow for statistical analyses that account for within versus cross-colony variation. The accuracy of age

estimation may be increased if researchers gather other morphological measurements on captured beavers such as zygomatic breadth and tail dimensions (Patric and Webb 1960, Larson and Van Norstrand 1968, Layne 2003).

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Diets of Desert Cottontail on Prairie Dog Colonies in Western South Dakota

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Abstract

Fecal pellets of desert cottontail (*Sylvaligus audubonii*) were collected during 1981 in May, June, July, August and September for dietary analysis to determine composition of forage plants. Four plants made up 70 percent of the total diet. Forage plants, in order of significance, were western wheat grass (*Pascopyrum smithii*), fescue (*Festuca* spp), squirretail (*Sitanion hystrix*), and plains muhly (*Muhlenbergia cuspidata*). The most common forb in diets was scarlet globemallow (*Sphaeralcea coccinea*) and the shrub, plains pricklypear (*Opuntia polyacantha*). Grasses in the diet ranged from 65 percent to 88 percent while forbs and shrubs ranged from 11 percent to 31 percent, 1 percent to 6 percent, respectively. Botanical composition in the plant community varied throughout the season.

Key words: food selection, forage, lagomorphs, rabbits, rangeland

Introduction

Dietary information of the desert cottontail (*Sylvaligus audubonii*) related to diets of other wildlife and large herbivores is integral to understanding forage use and sustaining our natural resources on rangelands. The most common wild herbivores in Conata Basin and Badlands area in western South Dakota are lagomorphs (*Lagomorpha*), black-tailed prairie dog (*Cynomys ludovicianus*), pronghorn (*Antilocapra americana*), mule deer (*Odocoileus hemionus*) and livestock that compete for the same available forage resources. Bison (*Bison bison*) are forage competitors and are present in the Badlands National Park only. Dietary information for the desert cottontail (*Sylvaligus audubonii*) is essential for resource managers to quantitative asses the dietary overlap of the herbivores to determine compliance with desired plans for sustaining or increasing vegetation conditions and to implement guidelines.

Lagomorphs are often associated with disturbed, grazed or subclimax plant communities (Flinders and Hansen 1975, Hansen and Gold 1977). Flinders and Hansen (1975) found that desert cottontails are associated with livestock grazing at a moderate level on the shortgrass prairie. Desert cottontails commonly inhabit prairie dog towns on western rangelands (Hansen and Gold 1977, Sharps and Uresk, 1990). Dietary information for black-tailed prairie dogs and cattle (*Bos taurus*) has been document for the Conata Basin and Badlands National Park since the 1970's (Fagerstone et al. 1977, Summers and Linder 1978, Fagerstone et al. 1981, Uresk 1984, Uresk 1986). However, dietary habits of the desert cottontail have not been evaluated for this region.

The purpose of this study was to determine the seasonal diets of desert cottontails associated with prairie dog towns, livestock and Bison grazing that have free choice of forage plants on the rangeland in western South Dakota.

Study Area

The study area was located on the Buffalo Gap National Grasslands and Badlands National Park about 13 km south

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of Wall in west central South Dakota. The climate is characterized by cold winters and hot summers and is classified as semiarid continental. Mean annual temperature was 10^0 C and ranged from -5^0 C in January to 26^0 C in July based on data gathered over 12 years from the weather station at Cedar Pass Visitor Center in Badlands National Park. Average annual precipitation was 40 cm. During the growing season, precipitation occurs as scattered thundershowers.

Gently undulating grasslands on the National Grasslands and Badlands National Park area made up the major portion of the study area. Soils of this area are derived from sedimentary deposits of clay, silt, gravel, and volcanic ash (Raymond and King 1976). Grasses which dominated the graminoid vegetation included blue grama (*Bouteloua gracilis*), buffalograss (*Buchloe dactyloides*), needleleaf sedge (*Carex eleocharis*), and western wheatgrass (*Pascopyrum smithii*). Major forbs were scarlet globemallow (*Sphaeralcea coccinea*), bigbract verbena (*Verbena bracteata*), Patagonia Indianwheat (*Plantago patagonica*), and prairie dogweed (*Dyssodia papposa*) (Uresk 1984).

Large free-ranging herbivores (*Bison bison*), pronghorn (*Antilocapra americana*) and mule deer (*Odocoileus hemionus*) graze the study area year round. In addition, the National Grasslands are grazed by cattle from mid-May to the end of October.

METHODS

Eighteen study sites were established on 15 prairie dog colonies ranging in area from about 12 ha to 283 ha. Three replications, each with six permanent sites (0.2 ha) were established. All cottontail pellets were initially cleared from each site prior to the two day sampling period. Fresh fecal pellets of the desert cottontail were collected over a two-day period during May, June, July, August, and September in 1981. The six sites collected for fecal pellets were combined within each of the three replications.

All fecal material from each site was dried at 60 degrees centigrade for 48 hours

and ground through a Wiley mill (1-mm screen) to insure thorough mixing. Five microscope slides were prepared monthly from composited fecal pellets from each of the three replicated sites. All fecal material was washed over a 0.1 mm screen (Sparks and Malechek 1968). Fecal material was cleared of chlorophyll and other compounds with Hertwig's solution. Hoyer's solution was used as a mounting medium (Baumgartner and Martin 1939), and the slides were dried for approximately 72 hours at 60 degrees centigrade.

Twenty microscope fields per slide were systematically located, magnified 100 times, and all recognizable plant fragments were recorded. Frequency of occurrence was determined by dividing the number of microscopic fields in which a given species occurred by the total number of fields observed X 100 (Curtis and McIntosh 1959) and converted to percent relative density (Sparks and Malechek 1968). Test slides were prepared for samples of known plant species to measure the ability of technical staff to properly and consistently identify plant fragments. Testing was applied according to the procedures outlined by Rogers and Uresk (1974). All values were summarized by means for each of the three replications for analyses. All plants >1 percent were selected for analyses and results.

RESULTS

Definite changes in composition of forage plants occurred in diets of the desert cottontail throughout the 5 month growing season (Table 1). A total of 47 plant species were identified in the diet of cottontails. Twelve genera and/or species comprising >1 percent, accounted for 86 percent of the diet in May; twelve plants made up 96 percent in June; eleven forage plants provided 94 percent of the diet in July. However, only ten forage plants accounted for 88 percent composition in August, and eight plants provided 81 percent of the diet in September.

The proportion of grasses in the diets of cottontails increased from 72 percent

Table 1. Average relative density of desert cottontail diets (%) with standard errors (in parentheses) for common plant species (>1%) over five months in 1981 and average composition by category in western South Dakota.

Category and Species	May	June	July	Aug	Sept	Average
Grasses and sedges						
Western wheatgrass *Pascopyrum smithii*	48.5(18.6)	57.4(8.3)	52.8(16.1)	56.8(4.7)	49.4(22.8)	53.0(6.0
Brome *Bromus spp.*	0.8(0.4)	2.0(1.6)	0.5(0.5)	0.0(0.0)	5.3(4.3)	1.7(0.9)
Buffalograss *Bouteloua dactyloides*	1.6(0.4)	0.4(0.4)	3.5(3.5)	0.4(0.2)	0.0(0.0)	1.2(0.7)
Sedge *Carex spp.*	2.1(1.7)	2.0(0.6)	0.4(0.3)	0.0(0.0)	0.8(0.8)	1.1(0.4)
Fescue *Festuca spp.*	6.5(1.8)	10.8(4.0)	7.9(4.5)	5.1(4.1)	0.1(0.1)	6.1(1.6)
Plains muhly *Muhlenbergia cuspidata*	4.2(2.1)	6.3(4.0)	8.7(4.1)	4.9(3.6)	3.5(2.0)	5.6(1.3)
Squirreltai *Sitanion hystrix*	4.1(1.3)	2.3(1.4)	7.0(1.2)	9.6(4.2)	5.4(2.3)	5.7(1.1) l
Sand dropseed *Sporobolus cryptandrus*	2.0(0.5)	1.4(0.2)	1.1(0.7)	1.0(1.0)	0.0(0.0)	1.1(0.3)
Needlegrass *Stipa spp.*	1.9(1.4)	2.7(1.5)	3.8(3.6)	2.2(0.5)	0.1(0.1)	2.1(0.8)
Grasses & Sedges Total[1]	**72**	**86**	**88**	**88**	**65**	**79**
Forbs						
Aster *Aster spp.*	0.0(0.0)	0.2(0.2)	0.3(0.3)	0.0(0.0)	5.0(5.0)	1.1(1.0).08
Lambsquarters *Chenopodium album*	2.9(2.1)	0.8(0.8)	0.3(0.3)	1.2(1.2)	5.6(5.1)	2.2(1.1)
Kochia *Kochia scoparia*	0.2(0.2)	2.1(1.1)	3.1(1.6)	3.9(2.0)	0.6(0.2)	2.0(0.6)
Stickseed *Lappula redowskii*	5.3(4.0)	3.3(3.1)	0.3(0.3)	0.7(0.7)	0.0(0.0)	1.9(1.1)
Woolly plantain *Plantago patagonica*	0.9(0.9)	1.6(1.6)	1.5(1.1)	1.0(1.0)	0.5(0.5)	1.1(1.1)
Scarlet globemallow *Sphaeralcea coccinea*	1.9(1.6)	3.7(0.6)	3.8(1.4)	3.0(1.7)	4.1(3.4)	3.3(0.8)
Forbs Total[1]	**22**	**13**	**11**	**11**	**31**	**17**
Shrubs						
Plains pricklypear *Opuntia polyacantha*	5.1(3.8)	0.6(0.3)	1.2(0.7)	0.7(0.7)	2.8(2.7)	2.1(0.9)
Shrubs Total[1]	**6**	**1**	**1**	**1**	**4**	**4**

[1] Total diet composition of all plants within category.

in May to a high of 88 percent in July and August, then declined to a low of 65 percent in September (Table 1). Western wheatgrass was the most common species in the diet and ranged from 49 percent to 57 percent. Other grasses common in the diet included fescue (*Festuca* spp) (less than 1% to 11%), plains muhly (4% to 9%) and squirreltail (*Sitanion hystrix*) (2% to 10%). Fescue was utilized consistently from May to August and was present in trace amounts in September. Plains muhly (*Muhlenbergia cuspidata*) was more common in June and July in the diet. Squirreltail was greatest in August. Other grass species within the cottontail diet comprised less than 1percent of the total diet during the 5 month growing season.

Forbs in the diet declined from 22 percent in May to 11 percent in July and August (Table 1). However, forbs provided 31 percent of the desert cottontail's diet in September. Scarlet globemallow was the most important forb in the diet followed by lambsquarters (*Chenopodium album*) and kochia (*Kochia scoparia*). Other forbs were minor in the desert cottontail diet. Shrubs ranged from 1 percent to 6 percent of the diet (Table 1). Plains pricklypear (*Opuntia polyacantha*) varied from 1 percent to 5 percent within the cottontail diet. The greatest amount of shrubs in the diets of cottontails occurred in May. Over the five months (May-September), four grasses, western wheatgrass, fescue, plains muhly and squirreltail provided 70 percent of the desert cottontail's diet.

DISCUSSION

Cottontails consumed a wide variety of plants. As in other studies, the desert cottontail in this study consumed mostly grasses and sedges (Hansen and Gold 1977, MacCraken and Hansen 1984, deCalesta 1979, Johnson 1979). Forbs were important in September, and a similar trend was observed by MacCracken and Hansen (1984). Monthly changes in diet composition is expected to be related to moisture content for metabolic water, nutrition requirements, and changes in

maturity of the various plant species to meet the sustainable requirements of the desert cottontail.

Many of the forage species consumed by the desert cottontail in this study were similar to those consumed by desert cottontail on prairie dog towns in Colorado (Hansen and Gold 1977). Sedges were dominant in the earlier studies whereas western wheatgrass was predominant in this study. Desert cottontails are commonly associated with prairie dog towns (Sharps and Uresk 1990, Hansen and Gold 1977).

Although desert cottontails on prairie dog towns consumed a variety of plant species, grasses specifically western wheatgrass were clearly predominant. Forbs were seasonally important, with scarlet globemallow the most common in the diet. Scarlet globemallow was also common in diets (10%) of the black-tailed prairie dog and cattle on the study area (Uresk 1984, Uresk 1986). Shrubs were less important than either grasses or forbs. Dietary analysis revealed clear preferences of the cottontail for certain foods especially as the season progressed. Management for desert cottontail populations is strongly related to four grass species that made up 70 percent of the diet that includes western wheatgrass, fescue, plains muhly and squirreltail.

The study suggests that forage competition for plant species is expected to occur among prairie dogs, cattle, bison, pronghorn, and deer that are common to the area. Information obtained from this study is important for resource managers to be in compliance with desired management plans that will include both plant and animal sustainability and to implement guidelines.

AKNOWLEDGEMENTS

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The Relationship Between Deuterium Excess and Uranium Groundwater Concentrations in Whitehall, Montana

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Abstract

Uranium can become elevated in the environment and pose a human health risk to water resources. Therefore, it is important to understand the origin, transport and concentration of uranium in the environment. With this exploratory study we propose that deuterium (δ^2H) excess may be a viable way to study uranium groundwater concentrations that originate from the surface. The Boulder Batholith of southwestern Montana has been associated with varying levels of uranium. The Whitehall, MT area includes the Boulder Batholith as a geologic unit. We collected 23 water samples and compiled data from a study done by the United States Geological Survey (USGS) within the area of Whitehall, MT. With a linear regression we show that there is a significant negative relationship between deuterium excess and uranium water concentrations. As expected we showed that there is a significant positive, linear relationship between uranium water concentrations and nitrate water concentrations. As expected we also showed that there is a significant positive, linear relationship between calcium carbonate and uranium groundwater concentrations for the compiled USGS data. The results of a Wilcoxon rank sum test showed that median uranium concentration for wells close to ponds is 14.5 $\mu g/L$ higher for our data added to the USGS data. These results may be consistent with a model that involves insoluble uranium being transported in suspension and settling out in ponds where an influx of nitrates and/or carbonates react to produce higher concentrations of water-soluble forms of uranium. We suggest that the relationship found between uranium concentration and deuterium excess in water samples may be related to the higher rates of evaporation in ponds. Thus, in this specific hydrologic environment of valley ponds close to the Boulder Batholith source, deuterium excess may serve to help predict levels of uranium concentrations.

Key Words: groundwater, uranium concentrations, water isotopes, carbonates, nitrates, Jefferson County

Introduction

Uranium (U) exists in varying levels in the soil, air and water of the natural environment. When uranium levels become elevated in water it can pose certain environmental problems. When uranium is dissolved in drinking water, at and above $30\mu g/L$, it poses a significant health risk to humans. These health risks include a slight increased risk of cancer, and kidney complications for drinking water at and above $30\mu g/L$ (CDC 2016). This is important for area populations that rely on groundwater as a main drinking water source. The Whitehall, MT area of Jefferson County mainly relies on groundwater for the population's residential and municipal drinking water needs. A past water quality study done by the United States Geological Survey (USGS) showed that Jefferson County, as well as surrounding counties in Southwestern MT, have groundwater extents with dissolved uranium present (Caldwell R. et al. 2013).

The potential for uranium to enter and become elevated in the groundwater can depend on many different factors. Granitic batholiths can often, but not always, contain detectable levels of uranium. The Boulder Batholith in Southwestern Montana is one geologic unit known to contain varying levels of uranium (Roberts, et al. 1951). The USGS conducted a three-year-long study to assess the uranium (and other radionuclides) groundwater concentrations of wells within Jefferson County, MT and surrounding areas that contain the Boulder Batholith as a geologic unit (Caldwell, et al. 2013). The Elkhorn volcanic range is related to the Boulder Batholith and may be a source of uranium for this area (Caldwell R. et al. 2013, Roberts et al. 1951, Smedes 1966). The alluvium and Boulder Batholith below the surface were also identified as potential sources of uranium for this area.

In this exploratory study, we attempt to use deuterium excess to study uranium concentrations in the Whitehall area. This exploratory study is based on environmental data and therefore not definitive research. There is also no specific known past research into the use of deuterium excess to study groundwater concentrations of uranium. However, the use of stable water isotopes to study chemical groundwater concentrations has been researched in the past (Sidle 1998). This is because the heavier water isotopes become enriched or depleted in natural waters relative to the lighter water isotopes. This process is referred to as isotope fractionation. One way that the heavier water isotopes become enriched in natural waters is through the preferential evaporation of the lighter water isotopes. Ponds for example have a higher ratio of stable water isotopes in comparison to rainwater and streams due to the preferential evaporation of the heavier water isotopes. Because, deuterium excess is calculated from both the stable water isotopes of oxygen (^{18}O) and deuterium (^2H or δD) from isotopic data it may be a more sensitive measure of evaporation (Frohlich et al. 2002, Keesari et al. 2017). Past studies have attempted to understand uranium groundwater concentrations using stable water isotopes to produce mixing models based on the relative age of water sources as well as an attempt to produce a direct relationship to ^{18}O (Helling 2000, Lonschinski et al. 2010, Kumar et al. 2014, Pant et al. 2017). In this exploratory study deuterium excess is used to attempt to understand uranium groundwater concentrations based on evaporation.

Assessing nitrate (NO_3^-) and nitrite (NO_2^-) concentrations for this study is important as they have the potential to increase the solubility of uranium minerals through oxidative dissolution (Gronowski, A. 2013, Nolan et al. 2015). Carbonates may also play a role in reacting with insoluble uranium minerals and producing more water-soluble complexes (Goodwin 1981, Abdelouas et al. 1998, Chau et al. 2011, Gronowski, A. 2013).

The purpose of this study was to study the spatial distribution of uranium groundwater concentrations in the Whitehall area with stable water isotopes as well as nitrate and carbonate concentrations. The study was completed as an undergraduate research project and funded by the University of Montana Western Student Senate through a learning grant. The future application of this study is a basis for a better understanding of the variables that affect uranium groundwater concentrations. This study is primarily an exploratory investigation into the uranium concentrations of wells in a specific area of the Boulder Batholith of Whitehall, Montana.

Study Area

This study was done in the area of Whitehall, MT. The approximately 61km^2 study area included 18 groundwater samples, 2 snow samples, and 3 surface water samples. These samples come from private and municipal wells as well as public access sites. The elevation within the study area ranges from 1200m to 1400m with most of the sample points located in the valley of Whitehall, MT area. Whitehall, MT and the surrounding area is classified

as having a semi-arid-climate. The average annual precipitation in Whitehall, MT is 24.13cm. The common native plants in the study area includes Big Sagebrush (*Artemisia tridentata*), Blue-bunch Wheatgrass (*Pseudoroegneria spicata*), Broom Snakeweed (*Gutierrezia sarothrae*) and Fringed Sagewort (*Artemisia frigida*). The sampling points are shown below over a geologic map of the area in Figure 1.

Geologic Setting

The main geologic feature in the area that is pertinent to this study is the Elkhorn Mountain range, which is mostly composed of Tertiary through Cretaceous aged extrusive igneous rocks. These extrusive and intrusive igneous rocks of the Elkhorn Range are closely related to the Boulder Batholith (Roberts et al. 1951, Smedes 1966). The generation of magma of the Boulder Batholith was caused by the subduction of the Farlon oceanic plate under the North American plate during the Cretaceous. The extrusive igneous rocks of this area resulted from a conduit of the mainly quartz monzonite Boulder Batholith. These volcanic igneous rocks of the Elkhorn Range intruded through older Mesoproterozoic to Mesozoic sedimentary and metamorphic rocks of the range (Roberts et al. 1951, Smedes 1966). The Boulder Batholith has been known to contain elevated levels of radionuclides (Caldwell R. et al. 2013). The valley of Whitehall area is mainly composed of undifferentiated gravel or alluvium of the Quaternary. This Quaternary as well as Tertiary sediment is the source of many of the wells in the valley of Whitehall area. There are also many wells in this area that draw from fractures in igneous or metamorphic source rock. The depth of the wells in this study area ranged from 8m to 107m.
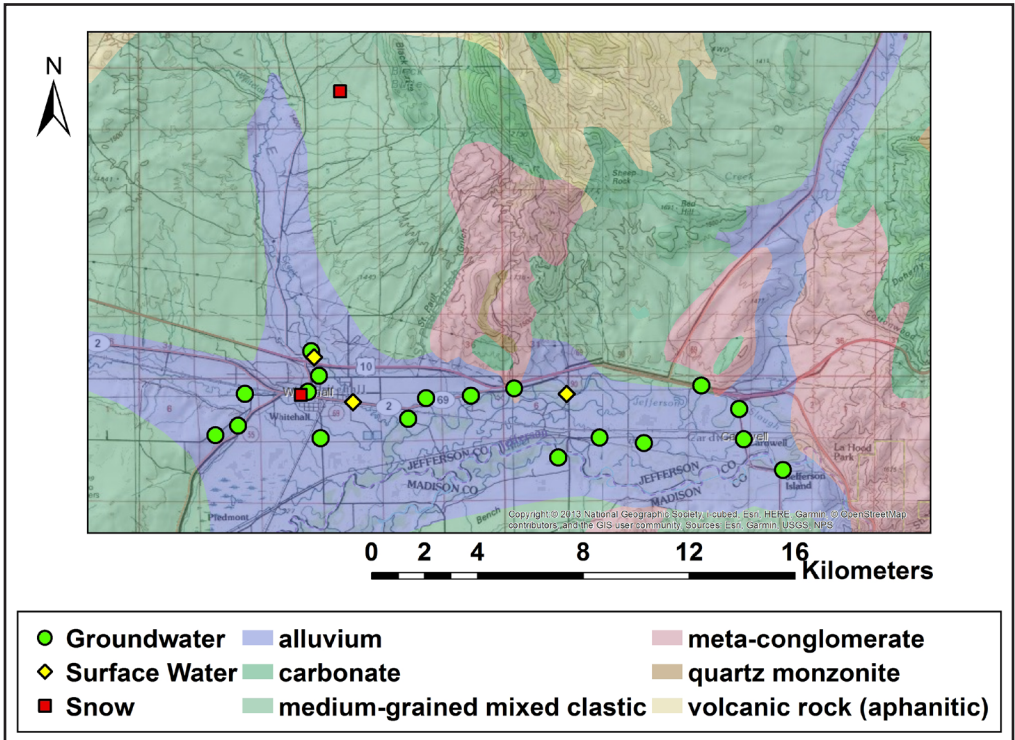

Figure 1. Geologic map of the Whitehall, MT area with the general sampling locations for the field data we collected. These points are generalized and do not reflect definitive sampling locations for this study (ESRI ArcMap 10.6, 2017).

Uranium Groundwater Concentration Factors

The main variables that affect uranium groundwater concentrations from the surface include origin, liberation, transport, and accumulation. The process that liberates uranium from the origin is often the same process that allows it to become mobile (Gronowski, A. 2013). The accumulation phase simply identifies the point at which uranium enters the groundwater aquifer and begins to accumulate over time. The element uranium exists in the natural environment as an isotopic mix of ^{234}U, ^{235}U and ^{238}U. The uranium element has six valence electrons and is a highly reactive metal in the natural environment. The two most common oxidative states of uranium that are found in the environment are the U(VI) and U(IV) valence forms (Gronowski, A 2013). These two valence forms that uranium exists in the natural environment serve as the base for numerous uranium minerals, and complexes. The uranium minerals associated with the Boulder Batholith in this area have been studied in the past and include pitchblende, torbernite, rutherfordine and others (Roberts and Gude, 1953). These uranium complexes and minerals have various solubility characteristics (Závodská et al. 2008). It should be noted that many other uranium complexes can form in the natural environment (Erikson et al. 1990, Závodská et al. 2008, Chau et al. 2011, Nielsen, E. et al. 2013,). The uranium complexes that form through reactions with carbonates and nitrates have been identified as important on how uranium becomes water soluble (Gronowski A. 2013). It should be noted that any one of these uranium minerals can become transported in suspension (Gronowski, A 2013).

METHODS AND MATERIALS

The study area was first established as an area within the valley from Whitehall to Cardwell. The sample points for this study consist of 18 groundwater, two snow, and three stream samples. One snow sample was new snow that precipitated the day of collection and the second was not. The materials used in this study included general field equipment such as a Garmin handheld GPS unit, data sheets, a pH and TDS meter, a YSI meter, and 65 sterile Nalgene bottles of 40ml, 300ml, and 500ml.

The 23 sample sites included the collection of a GPS point, water sample pH, temperature, total dissolved solids (TDS), and a general site description. The water sampling procedures of this study followed the standards outlined by the USGS (U.S. Geological Survey 2017). The 23 water samples were stored in a refrigerator for no longer than ten days before being sent to the lab in Butte for analysis. The 23 water samples were sent to the Montana Bureau of Mines and Geology lab in Butte, Montana for the analysis of water isotope ratios, total nitrate and nitrite concentrations in milligrams per liter, and uranium in micrograms per liter. The isotopic data is represented in standard percent per million as and denoted as δ. An example of the equations used to calculate deuterium (D) excess is shown below.

$$(\delta \text{ or } ‰ \text{ of deuterium}) = \left(\frac{\frac{^2H}{H}(Sample)}{\frac{^2H}{H}(VSMOW\ Std.)} - 1 \right) \cdot 1000, (D\ excess = \delta D - 8\ \delta^{18}O)$$

(Kendell et al. 2001, Lee et al. 2003, Shama 2018)

Statistical Analysis

The data used for this study was analyzed statistically using Microsoft Excel 2016 and MATLAB 2018. The descriptive statistics were calculated in Excel and the statistical tests were conducted in MATLAB. Uranium groundwater concentrations and δD excess for this study were run through a regression. The data for the uranium water concentrations and the nitrate/nitrite ground and surface-water concentrations were also run through a regression with one outlier. The 8 nitrate/nitrite samples below the detection limit were run as 0 for both nitrate/nitrite and uranium. The data from the USGS study for uranium groundwater concentrations and hardness as calcium carbonate was also run through a regression. The uranium water concentration data was categorized by pond distance based on the mean distance (>168m or <168m) and run through a Wilcoxon Rank-Sum (WRS) test as the data was not from a normal distribution. Ponds were identified as approximately larger than a 100-meter perimeter using Google Earth version 9.2.93.1. The data for all tests used a 0.05 level of significance ($\propto$ =0.05).

RESULTS

The uranium water concentrations for all samples had a maximum of 44.4$\mu g/L$ and a minimum of <0.2$\mu g/L$. The total nitrite and nitrate for all samples had a maximum of 10.3 (mg/L) and a minimum of <0.2(mg/L). The complete list of parameters analyzed including pH, total dissolved solids (TDS), temp (Celsius), δ O^{18}, δ D (or ‰), uranium ($\mu g/L$) and total nitrite and nitrate (mg/L), can be found in Appendix A.

The isotopic data for this study is a plot of the δ^{18}O, and δ^2H (δD) for the groundwater, surface water, and snow samples(Fig. 2).

Scatter plots for the uranium water levels vs the δ^{2}H (δD) excess levels, and the uranium water concentrations and total nitrate/nitrite water levels for the Whitehall are depected in Figure 3 and Figure 4.

There is a significant negative linear relationship between uranium water concentrations and δD excess levels (t_{18} = - 3.18, p = 0.0058, R^2 = 0.38). The linear regression line for this data is y = 11.65 - 3.540 x. This data includes the 18 groundwater samples from the Whitehall area.

There is a significant positive relationship between uranium water concentrations and total nitrate nitrite concentrations (t_{20} = 3.663, p = 0.00178, R^2 = 0.42). An outlier was removed for this regression. The outlier was discounted due to a potential influx of nitrates at the site where it was collected. The linear regression line for this uranium and nitrate scatter plot is y = 5.49 + 9.24x (Fig. 5).

The results for the linear regression of the USGS groundwater data showed that there is a significant positive relationship between uranium water concentrations and hardness as calcium carbonate concentrations (t_{66} = 6.295, p = 2.846e^{-8}, R^2 = 0.38). The linear regression line for this USGS data is y = -6.03 + 0.107x. This regression consists of data compiled from the USGS 2007-2010 study as calcium carbonate was not measured in this study.

Table 1. The descriptive statistics for the uranium and total nitrate and nitrite concentrations of the surface water and groundwater samples for the Whitehall, MT area (2019).

Sample Source (U)	Mean ($\mu g/L$)	SD	Sample Size
Groundwater Samples	14.9	12.6	18
Surface Water Samples	14.6	1.76	3

Sample Source (N)	Mean (mg/L)	SD	Sample Size
Groundwater Samples	2.18	3.008	18
Surface Water Samples	.48	0.0316	3
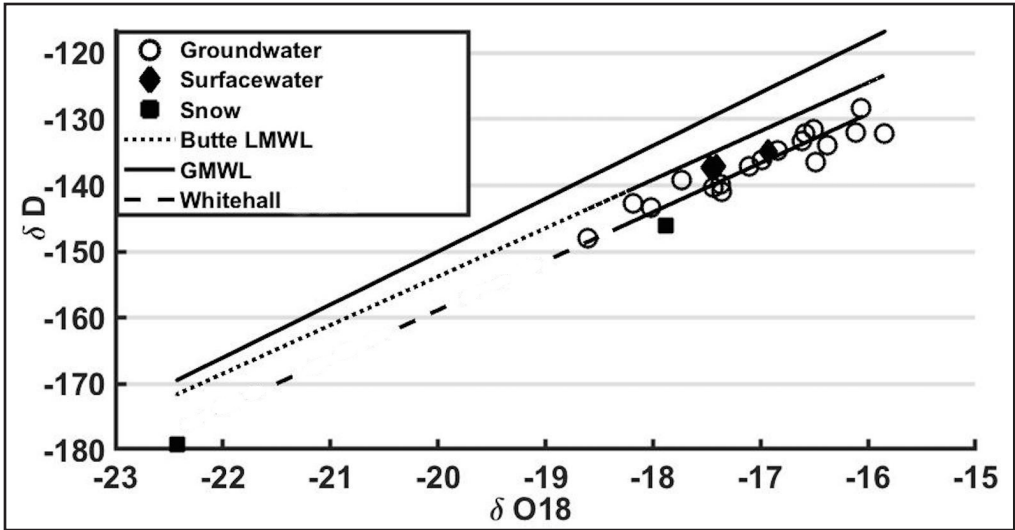
Figure 2. Isotopic data collected in our study which is displayed with the LMWL for the Butte, MT area based on past research (Gammons et al. 2006) and the GMWL (Craig H. 1961), (2019). The dotted line depicts the LMWL for the Butte area for comparison ($\delta^2 H =$ 7.32 δO^{18} -7.32). The GMWL is also depicted in this figure as the solid line for comparison ($\delta^2 H = 8 \delta O^{18} + 10$).
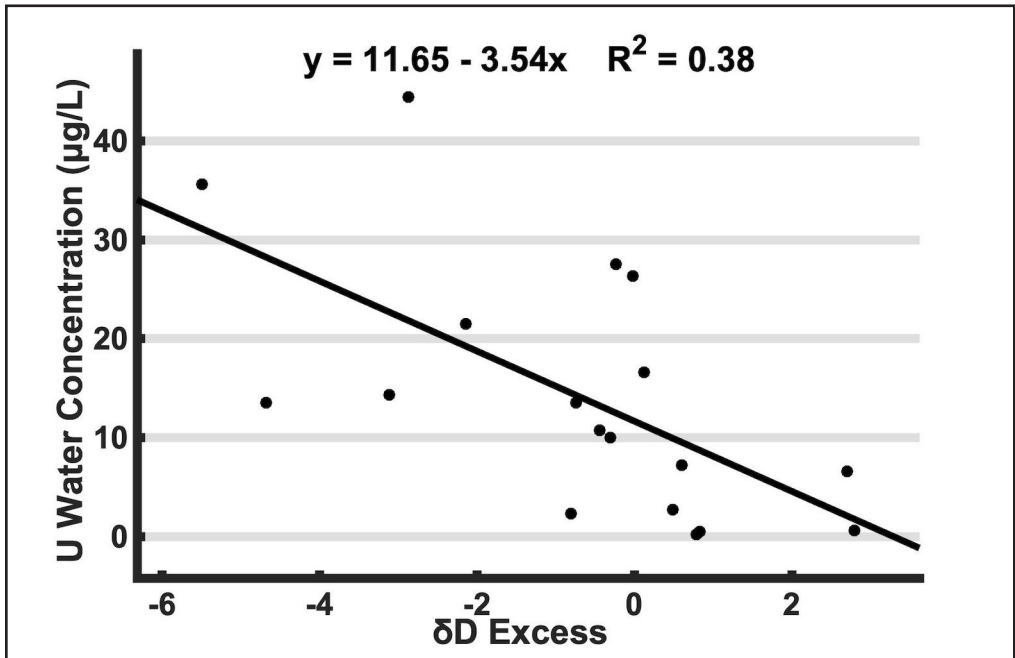
Figure 3. Scatter plot for the uranium water concentrations and deuterium excess of the 18 groundwater samples collected in the Whitehall, MT area (2019).

The figure referenced in this section is a boxplot that depicts the median uranium water concentrations for wells close to ponds (≤168m, 8 samples) and the median uranium water concentrations for wells far from ponds (>168m, 12 samples).(Fig. 6)

The results of the Wilcoxon Rank-Sum test showed that the median uranium concentration for wells close to ponds was significantly higher than the median uranium
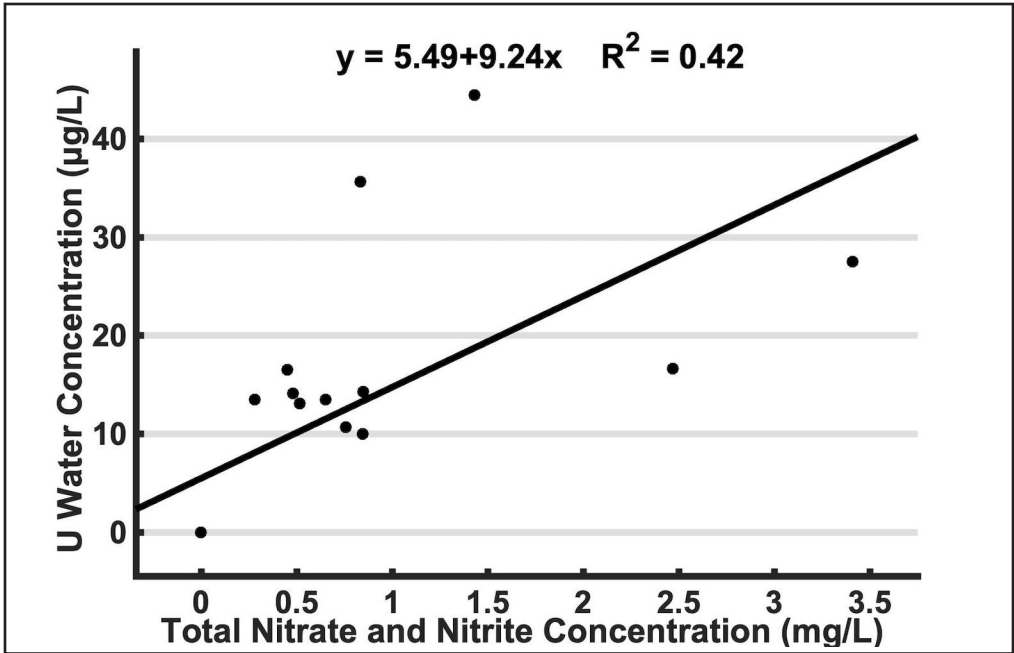
Figure 4. Scatter plot for the uranium water concentrations and total nitrate and nitrite concentrations of 20 surface water and groundwater samples collected in the Whitehall, MT area (2019). One groundwater sample had an abnormally high nitrate concentration (10.3mg/L compared a mean of 2.18mg/L for all other samples) and was not included in this linear regression and scatter plot. The outlier was discounted due to a potential influx of nitrates at the site where it was collected. The 8 samples below the detection limit of 0.02 were run as 0 for both uranium water concentrations and nitrate/nitrite concentrations for this test.
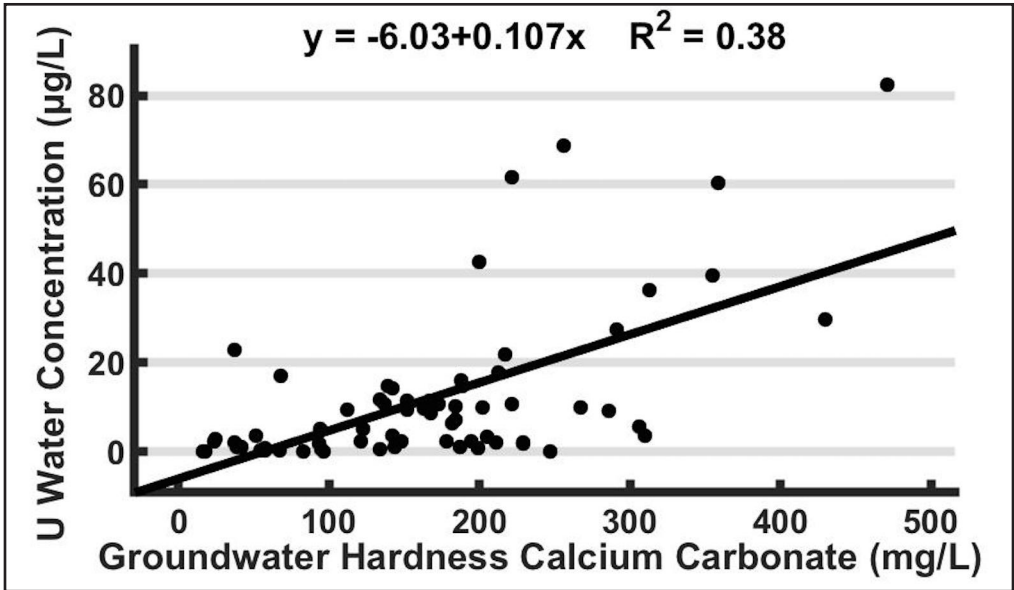
Figure 5. Scatter plot for the uranium water concentrations and calcium carbonate hardness of 66 of the groundwater samples collected in Jefferson County, MT by the United States Geological Survey from 2007 to 2010.
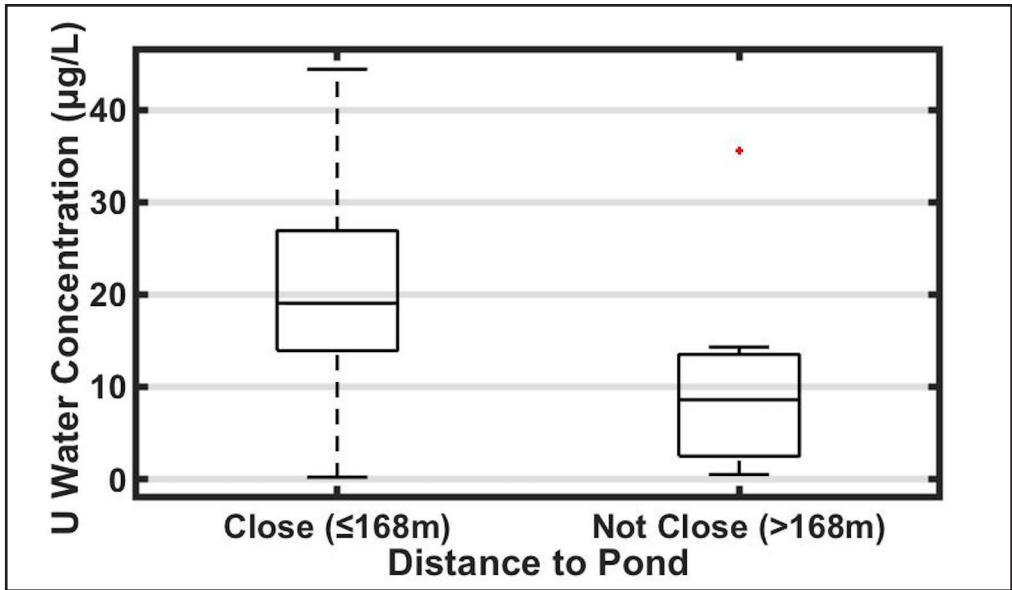
Figure 6. Box plot of median uranium groundwater concentrations for wells close to (≤168m) and not close to ponds (>168m). This distance was based on the average distance from the ponds to the wells for this data. This figure displays data collected for this study and data collected by the United States Geological Survey from 2007 to 2010. The data was collected from the Whitehall, MT area (2019).

groundwater concentration for wells not close to ponds ($WRS_{8,12}$, $W = 111.5$, $p = 0.0338$). The wells close to ponds were 14.5 $\mu g/L$ higher for uranium than wells far from ponds.

Discussion

The understanding of the transport, source, and concentration of uranium in the environment is important for populations that rely on groundwater as a main source of drinking water. Our exploratory study might show that there is some influx of uranium into the groundwater aquifer from the surface. We were able to show this based on the significant value from comparing the deuterium excess and the uranium water concentrations through a linear regression. This relationship can be related to the differing isotopic ratios of water influx from the surface into the groundwater aquifer. The study done by researchers in Punjab Pakistan was not able to find a relationship between ^{18}O and uranium groundwater concentrations (Pant et al. 2017). It may be that deuterium excess

might serve as a more suitable comparison to understanding uranium water concentrations for this area because of its close relationship with evaporation (Frohlich et al. 2002, Lee et al. 2003, Keesari et al. 2017). Or there is less connectivity from the surface to the groundwater in their study area. A relationship between uranium in groundwater and an enrichment in the heavier water isotopes recharging the groundwater may be possible if we can relate it to evaporation. However, it should be noted that the isotopic data suggests that recharge to the aquifer during the short sampling period of this study was affected less by evaporation than the average LMWL which may be why our data falls below it (Gammons et al. 2006). It's also likely that deuterium excess may not be a good fit because it is typically calculated from meteoric water instead of groundwater and there is little past research to support a relationship (Lee et al. 2003, Penga et al. 2012, Pant et al. 2017). It should also be noted that water isotope data is typically collected over a longer period than the time period of this study.

The positive relationship between uranium water concentration and total nitrate (NO_3^-) and nitrite (NO_2^-) concentration has been found by researchers at the University of Nebraska (Nolan et al. 2015) and has also been linked to nitrate dependent microbes that may oxidize uranium into a more water soluble form U(VI) (Senko et al. 2005). These authors speculate that insoluble or less soluble uranium minerals are being oxidized while the nitrates and nitrites are being reduced. The positive relationship between uranium concentrations and groundwater hardness as calcium carbonate ($CaCO_3$) has also been studied as a potential mode for uranium to become more water soluble (Goodwin 1981, Erikson et al. 1990, Gronowski, A. 2013).

The significantly higher median for uranium groundwater concentration of wells close to (≤168m) ponds compared to wells not close to ponds (>168m), may indicate how uranium is being transported on the surface for the Whitehall area. This might suggest that there are uranium minerals being transported in suspension from the source on the surface. The insoluble uranium compounds then settle out in the valley ponds. The Piedmont Pond in the Whitehall area may fit this model due to its relatively elevated uranium concentration of 23.1$\mu g/L$, as well as being relatively enriched in the stable water isotopes and the presence of calcium carbonate (182.7mg/L). The Piedmont Pond does not however appear to be affected by an influx of nitrates or nitrites (<0.02mg/L) (MBMG et al. 2012).

The higher median for uranium groundwater concentration of wells close to (≤168m) ponds compared to wells not close to ponds (>168m), suggests that insoluble uranium minerals are being transported to the valley ponds. The Piedmont Pond in the Whitehall area has been measured by the Montana Bureau of Mines and Geology (MBMG) and they found a relatively elevated uranium concentration of 23.1$\mu g/L$, relatively enriched stable water isotopes (δD: -104.0, $\delta^{18}O$: -10.5) and the presence of calcium carbonate (182.7 mg/L). The Piedmont Pond does not however appear to

be affected by an influx of nitrates or nitrites (<0.02mg/L), (MBMG et al. 2012).

We then may relate the significant result of pond distance to wells back to our first three significant test results displayed. The ponds might have a higher ratio of the stable water isotopes compared to their mainly snow melt source, and other surface water bodies due to evaporation (Penga et al. 2012). We see this for the Piedmont pond in the Whitehall area which is relatively enriched in the stable water isotopes (MBMG et al. 2012). This might relate to decreasing deuterium excess (which is calculated from the isotopic data) being related to increasing uranium concentrations. If we assume that some amount of insoluble uranium is settling out in the ponds, we might also assume that it becomes soluble at some point to enter the groundwater aquifer. An influx of nitrates, nitrites and calcium carbonate into the ponds may react with the insoluble uranium compounds and allow them to become water soluble. It's also likely that the relationship between uranium and ponds has nothing to do with insoluble uranium settling out in the ponds as they often have little to no surface flow influx. It may be more related to the ponds being a sink for nitrate/nitrites, and carbonates which might increase the solubility of uranium.

We can now propose a potential model for an influx of uranium from the surface into groundwater of the Whitehall area.

1. The uranium minerals may be liberated from the Elkhorn volcanic rock of the Boulder Batholith on the surface.
2. The uranium minerals are transported from the source to the valley
3. An influx of nitrates, nitrites and carbonates react with the insoluble uranium compounds and form water soluble uranium compounds.
4. The water-soluble uranium compounds then enter the groundwater aquifer from the valley ponds which can be directly related **to** deuterium excess.

This model is based on an exploratory study based on environmental data and therefore not definitive research. We suggest that future research related to this exploratory study include samples from the ponds in the Whitehall, MT area. The analysis of calcium carbonate, nitrate, and uranium concentration as well as water isotope ratios for future samples to test our model presented in this study. Future research related to this study should also include water isotope data collected over a longer period, and groundwater samples to compare to the pond data in the Whitehall, MT area. The model presented in this exploratory study may also apply to further research in other study areas where uranium is present in the groundwater. The model presented might only account for one part of how uranium enters the groundwater in this area. Other sources of uranium discussed most likely play a part in how uranium enters the groundwater and this study should be viewed solely as an exploratory analysis.

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Appendix of Results A.

Sample #	Temp C°	pH	TDS (ppm)	Uranium µg/L	NO3/NO2⁻ N mg/L	$\delta^{18}O$ (‰)	$\delta^{2}H$ (‰)
1	10	7.17	164	2.67	< 0.2	-16.5	-131.5
2	11	7.1	165	2.29	< 0.2	-17.4	-139.8
3	11	7.15	166	16.6	2.47	-16.1	-128.4
4	9	7.25	169	27.5	3.41	-17.0	-136.2
5	11	7.31	370	13.5	0.281	-16.5	-136.5
6	13	7.56	202	< 0.2	< 0.2	-18.0	-143.4
7	12	7.41	186	26.3	< 0.2	-16.8	-134.7
8	12	7.2	175	44.4	1.43	-16.4	-133.9
9	11	7.23	170	0.503	< 0.2	-18.6	-148.0
10	11	7.18	150	35.6	0.834	-15.8	-132.2
11	11	7.21	14	NA	NA	-17.9	-146.1
12	11	7.22	131	7.18	< 0.2	-16.6	-132.1
13	11	7.21	143	21.5	10.3	-17.4	-141.0
14	16	7.56	301	0.565	< 0.2	-18.2	-142.7
15	17	7.16	206	10	0.847	-17.1	-137.1
16	17	7.11	189	6.54	< 0.2	-17.7	-139.2
17	17	7.12	153	10.7	0.758	-16.6	-133.4
18	17	7.18	123	13.5	0.652	-17.4	-140.3
19	17	7.14	205	14.3	0.848	-16.1	-132.0
20	17	7.18	127	13.08	0.516	-17.5	-137.3
21	17	7.16	141	14.1	0.481	-17.4	-137.2
22	17	7.26	186	16.5	0.453	-16.9	-134.8
23	17	7.35	10	NA	NA	-22.4	-179.1

Evaluating the Accuracy of Bloodstain Pattern Analysis Using Hemodynamic Factors

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Abstract

This comparative survey explores the relationship between the discipline of bloodstain pattern analysis (BPA) and hemodynamic blood properties, such as viscosity and hematocrit. In BPA, forensic scientists study the phase change of blood when in contact with air, but little forensic literature connects blood biomechanics, such as hematocrit levels, to BPA. Red blood cell count, or hematocrit, in females (37-48% of blood volume) is slightly lower than males (45-52% of blood volume) from menstrual red blood cell loss, etc. Strong evidence suggests that erythrocytes influence blood viscosity because of their high concentration (4-6 x 10^6 RBC/mm^3 or 40-45% of blood volume in healthy individuals). When whole blood is altered by a disorder/disease or alcohol intake, hematocrit levels can be affected as well. With this knowledge, there is reason to believe that blood viscosity changes with individualistic hematocrit levels. Therefore, it is hypothesized that traditional BPA can produce inaccurate results. Intravenous blood samples were drawn from nine volunteers (all women, including eight with blood alterations and one healthy control) into collection tubes containing ethylenediaminetetraacetic acid. Each sample was tested for viscosity using a Cannon-Fenske viscometer and hematocrit levels using a ZipCombo centrifuge. Each sample was used to make several bloodstains at varying degrees of impact (10°, 30°, 60°, and 90°). ANOVA ($\alpha = 0.05$) and Tukey HSD statistics were used to compare angle of impact variables against each other within the nine participants. This survey connects hemodynamic properties to angle of impact tests in BPA by significantly showing how bloodstains can be misinterpreted. By examining blood viscosity among several individuals, this research assesses the accuracy of BPA by comparing experimental and expected bloodstain angles and creates a predictive framework for analyzing bloodstains created by physiologically altered blood. In conclusion, hemodynamic factors among individuals were found to influence traditional BPA methodology and future research is recommended to better understand hemodynamic properties and fulfill the recommendations made by the 2009 National Academy of Sciences (NAS) Report.

Key words: angle, blood, forensic science, hematocrit, hemodynamics, pattern analysis, viscosity

Introduction

The purpose of this paper is to present a comparative survey among a variety of blood disorder patients capturing hemodynamic factors of blood outside of an individual's body. There is significant research on blood as an internal biological fluid, however there is little research on the influence of hemodynamic factors as blood exits the human body (Nordqvist 2017). This phenomenon is only seen in forensic science literature where bloodstain pattern analysis (BPA) is used to reconstruct crime scenes using blood outside of the human body. BPA is the interpretation of bloodstains at a crime scene in order to reconstruct and/or recreate the actions that caused the bloodshed and pattern. Forensic technologists examine the size, shape, distribution, and location of the bloodstains to form an opinion about what happened at the crime scene.

Additionally, analysts can determine an area of convergence (two-dimensional analysis) or area of origin (three-dimensional analysis) to better understand where the bloodstains originated from at a scene. BPA is a multidisciplinary approach that uses biology, physics, and mathematics to assist the analyst in crime scene reconstruction, corroborating witness statements, and

including or excluding potential perpetrators from the scene investigation. Analysts aim to categorize the bloodstains at a crime scene by gathering information from the spatter patterns, transfers, voids, and other marks that occurred after bloodshed. BPA differs greatly from the testing of blood which is left for serologists and DNA analysts (Koen and Goetz 2017).

Whole blood is composed of cellular elements that include erythrocytes or red blood cells (RBCs), leukocytes or white blood cells (WBCs) and thrombocytes (platelets). These cellular elements are suspended in an aqueous polymer solution called the plasma, which acts as a delivery system for the cellular components in blood. Plasma is composed of electrolytes, hormones, antibodies, enzymes, and other proteins in small concentrations (Bodnar et al. 2011). There is evidence that RBCs influence the mechanical properties of blood because of their high concentration (4-6 x 10^6 RBCs/mm^3 or 40-45% of blood volume in healthy individuals) compared to the other cellular elements (Bodnar et al. 2011). The red blood cell count, or hematocrit, in females (37-48% of blood volume) is slightly lower than males (45-52% of blood volume) because of menstrual red cell loss (Shiel 2017). Low hematocrit levels are seen in individuals with abnormal hemoglobin, bone marrow complications, and anemia, while high hematocrit levels are seen in individuals living at high altitudes and in chronic smokers (Shiel 2017). Dehydration can also cause red blood cell levels to appear extremely high, although the individual may have physiologically normal blood.

When asked in a legal setting, a bloodstain pattern analyst generally describes blood as a Non-Newtonian viscoelastic fluid. Non-Newtonian fluids, such as paint, ketchup, toothpaste and blood have large particles that have limited time mobility when a force is applied quickly, resulting in the formation of a solid. A common example of this phenomenon is corn starch and water, in which a force can quickly change the liquid into a solid. Shear rates are used to measure the rate of change of velocity of two adjacent layers of fluid. They ultimately measure how the fluid is "worked" in the environment in which it is placed. Because blood encounters several types of vessels within the human body, it experiences shear stress, and therefore shear rates can be determined based on the viscosity of the blood. Viscosity, in centistokes (cSt), is the measure of a liquid's ability to resist deformation by force or tension. A fluid is shear-thinning if the viscosity decreases as the shear rate increases. Some shear-thinning fluids are known as pseudo-plastics which are used in industrial and biological processes. Shear-thinning, or a decrease in viscosity, is directly related to blood dynamics and the interactions of RBCs (Lanotte 2016). Conversely, shear thickening occurs when the viscosity of the fluid and the shear rate mutually increase. Plasma exhibits Newtonian fluid properties, in which the viscous stress from flow is dependent on the local strain rate, but whole blood follows non-Newtonian characteristics, especially at low shear rates.

In blood, shear rates occur when the velocity of one layer of fluid is different from the velocity of an adjacent layer. Blood acts as a non-Newtonian fluid because of three phenomena; RBCs tend to form three-dimensional microstructures while having the ability to breakup and/or align with the field of flow (Bodnar et al. 2011). Each of these phenomena can be influenced by changes in shear rates of blood throughout an individual's daily life. At low shear rates, whole blood tends to have very little strain from vessel walls making it viscous or thick. Conversely, high shear rates coerce blood into becoming less viscous allowing it to flow easily through narrowed blood vessels. Recently, Bodnar et al. (2011) proposed a model that can predict the viscoelastic response of blood while inside the human body. They explained that RBCs behave like viscoelastic fluids in venous blood flow because they are elastic membranes filled with fluid, but in stenosed vessels shear rates can follow non-Newtonian properties (Bodnar et al. 2011).

Research suggests that when whole blood is altered by a disorder/disease or alcohol intake, RBCs can be affected in several ways. Hypo- and hyperthyroidism can cause anemia and erythrocytosis leukopenia; two alterations to RBC count (Dorgalesh et al. 2013). Levothyroxine is a common oral medication for treating hypothyroidism and thyroid cancers, but it does not directly influence RBC count in individuals. Evidence shows that Thalassemia I and II increase reactive oxygen species in an individual causing RBC oxidative damage (Ko . 1997). This suggests RBC count and blood viscosity is altered in Thalassemic patients. Studies also show that diabetic patients experience a decrease in RBC deformability, causing RBC rupture from an overall loss of RBC fluidity (Ernst and Matrai 1987). High alcohol consumption reduces thiamine and folate absorption in the intestines. Both of these nutrients are used in RBC production and alcohol dependent individuals (74 drinks/week) can experience bone marrow abnormalities leading to a decrease in RBC production (National Institute on Alcohol Abuse and Alcoholism). In alcoholic anemia, an individual can even experience a decrease in platelet production leading to the inability to form clots, which may also lead to a stroke. It should be noted that the quantity and type of alcoholic beverage consumed, increase the variability in coagulant properties of platelets. Von Willebrand's disease (VWD) is a rare blood clotting disorder that alters platelet activity. It can be classified into three types with decreasing blood clotting activity with each type (Sadler et al. 2006). Prothrombin 20210 mutation or Factor II mutation is a genetic disorder that causes dangerous blood clots to form more frequently than individuals without the mutation (National Blood Clot Alliance). Both VWD and Prothrombin 20210 mutations do not alter red blood cell count, rather the genetic composition of an individual's platelets. Finally, research suggests that blood thinners, such as heparin, bind to thrombin and induce a conformational change in the enzyme to prevent blood clotting (Machovich 1975, Mellanby 1934). Xarelto® is a more expensive alternative to heparin blood thinners, but both have not shown to directly influence the RBC count in individuals.

The conditions stated above lie on a spectrum of direct and indirect influence on blood dynamics. Some disorders directly influence RBC production or degradation, such as hypothyroidism and Thalassemia, while others only affect platelet genotype. Previous research suggests that blood viscosity is dependent on hematocrit levels in individuals, and therefore blood viscosity could be moderately individualistic. As mentioned above, platelet count, activity, diet, and clotting factors can influence blood viscosity. This phenomenon happens because the slightest alteration to blood components in an individual can lead to applied shear stress and a change in blood viscosity. For example, if an individual is dehydrated and exercises for a long period of time their blood would appear more viscous; the dehydration and continuous exercise would increase the level of red blood cells in the blood (to increase oxygen delivery to muscles, tissues, and organs) and the blood vessels would begin to apply shear stress (decrease in blood viscosity) to allow the blood to travel more easily through the body. This example shows how easily blood viscosity can change in an individual throughout their daily life, but it is difficult to monitor this physiological change inside of the body for research.

Blood behaves to specific scientific principles, both inside and outside the human body, and therefore BPA experts can examine stains to draw conclusions as to how blood was shed at a crime scene. Analysts classify bloodstain patterns as either spatter patterns, transfers, voids, other marks, or a combination of these. Bloodstains assist investigators in reconstructing the sequence of events that occurred during or after bloodshed. Analysts are forced to recognize and interpret bloodstain patterns to determine how they were created (National Forensic Science Technology Center). Bloodstain pattern

analysis has proven to be one of the most legitimate pieces of evidence for crime scene reconstruction, but other forms of evidence, such as hair and impression evidence, have received the legal "chopping block". Interestingly, BPA is a traditional forensic identification science that uses subjective assumptions and guesswork. The field of forensic science is amid a paradigm shift to sound scientific foundations and justifiable protocols, especially after the National Academy of Sciences (NAS) Report of 2009 (Saks and Koehler 2005). Forensic scientists must consider revising their methods with more defensible and empirical foundations to remain legitimate in a legal setting. With further research, reliability and accuracy of BPA can increase the ways that bloodstains can be applied judicially.

Currently, there is a gap in scientific literature because blood possesses enigmatic properties once it has left the human body. The specific influence of viscosity and hematocrit on BPA has been researched extensively (Aplin et al. 2019, Kim et al. 2016), but hemodynamic factors are unrecognized in populations with physiologically altered blood. Kim S. et al. (2016) attempted to use standard values of hematocrit in bloodstain trajectory reconstruction but found two systematic errors: one on the blood viscosity, which depends on the blood hematocrit, and the second on the estimated impact diameter (Kim et al. 2016). These uncertainties found in BPA hemodynamics support the recommendations made by the 2009 NAS Report, which mentioned that "the uncertainties associated with BPA are enormous" (Committee on Identifying the Needs of the Forensic Sciences Community, National Research Council 2009). Kim S. et al. states that employing generalized correlations or hematocrit levels to BPA can lead to errors in determining the area of origin of the bloodshed. This research will address this finding and attempt to apply generalized correlations and hematocrit/viscosity levels to BPA in a population of physiologically altered blood patients.

Attinger et al. (2013) provide a comparative review of fluid dynamics of blood, current BPA research, and new BPA methodologies. Their comprehensive analysis indicates that BPA would benefit from a more scientific foundation and joint research between BPA and fluid dynamics is needed to increase BPA reliability (Attinger et al. 2013). More recently, Aplin et al. (2019) analyzed BPA at six varying hematocrit levels and found that differences in the hematocrit and viscosity values did not affect the accuracy of the forensic analysis ($p > 0.05$) (Aplin et al. 2019). This is consistent with previous studies that state hematocrit values of a blood source do not influence its origin. The problem with the proposed methodologies is that the researchers do not attempt to make generalized correlations between hematocrit values and physiological blood alterations, which could be individualistic of a person at the crime scene. This project will assess this concept and analyze impact patterns when the contributing blood source is known to be of a physiologically abnormal sample.

The purpose of this paper is to present a comparative survey among a variety of blood disorder patients capturing hemodynamic factors of blood outside of an individual's body. The work presented here significantly extends the preliminary results obtained by the primary investigator on 12 October 2018 where blood viscosity was compared among dehydrated and hydrated individuals. At first glance, the dehydrated blood was more viscous and appeared to alter BPA accuracy. The preliminary research continued to analyze the blood samples by creating traditional bloodstains using an angle of impact test and string method. Upon analysis of detailed angles of impact, the dehydrated blood samples deviated up to 10 degrees from the position at which the sample was dropped. Although this phenomenon was only examined in one participant, it left reason to believe that blood viscosity could influence the accuracy of BPA. The NAS Report of 2009 recommended that sound scientific research be performed on BPA to decrease its level of inaccuracy. By attempting to examine

blood viscosity and hematocrit levels among several individuals, this present research assesses the accuracy of BPA by comparing experimental and expected angles of impact of bloodstains.

METHODS

Sample Collection

Intravenous blood samples were drawn from nine volunteers (all women, including eight with blood disorders/alterations and one healthy control) at the University of Providence lab into tubes containing ethylenediaminetetraacetic acid as an anticoagulant. Female participants were selected because hematocrit levels vary greatly between sexes and this variable needed to be controlled to analyze how hematocrit and BPA are related. The participants were in their 60s, 40s, 30s, 20s and teens to display a varying age demographic among the sample. Participants were hand selected by the primary investigator to ensure each subject had a blood alteration. Among them were patients with Thalassemia and blood clotting disorders, such as VWD. Other participants were found based off research that suggested physiological conditions that alter blood viscosity (indirectly caused from RBCs), such as diabetic and hypothyroid patients and those on regular blood thinners. Additionally, a participant was asked to consume two alcoholic drinks (28 g) before having their blood drawn. In a separate draw, the same participant consumed four alcoholic drinks (56 g) and had their blood drawn once more. In both cases, the blood was analyzed to see whether low or high alcohol consumption influences BPA. Additionally, the blood alcohol content (BAC) was recorded to ensure the participant had levels of legal and illegal alcohol consumption, in regards to operating a motor vehicle.

Hematocrit Testing of Blood Samples

A ZipCombo centrifuge, heparinized centrifuge tubes, and EZ Reader were purchased from LW Scientific©. Blood samples were taken up into heparinized microcapillary tubes and capped at both ends using a thick clay. For each participant, the microcapillary tube was centrifuged at 12,000 rpm (7500g) for 3 min. After the allotted time, the capillary tubes were placed into an EZ Reader and red blood cell count was determined by the percent of erythrocytes separated from cellular solution.

Viscosity Testing of Blood Samples

A Cannon Fenske viscometer with model number 2700 was obtained from the University of Providence lab. It was cleaned with distilled water and left to dry before testing the viscosity of three standards. The standards chosen for viscometer calibration were toluene (0.6800 cSt), distilled water (1.0038 cSt), and absolute ethanol (1.5200 cSt). The three standards were used to measure the kinematics of the specific glassware used. Kinematic viscosity is measured using centiStokes (cSt) and can be used to quantify unknown viscosities, such as blood. To ensure that temperature did not alter viscosity, each standard was chilled to 20 ^{0}C using an ice bath. Additionally, the glass viscometer was placed in a 20^{0}C-water bath using a ring stand. With this system, both the sample and the viscometer were at a constant 20^{0}C before running the liquid through the glassware to determine viscosity. The viscometer was leveled before each test to ensure that each sample flowed properly though the viscometer. Each standard was loaded into the viscometer in separate trials and the time for each to pass from line "C" to line "E" was recorded (Fig. 1).
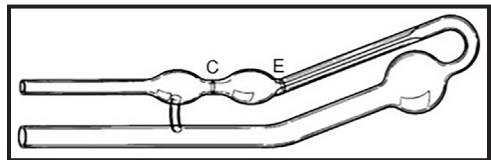
Figure 1. Cannon Fenske viscometer obtained from the University of Providence lab. Viscosity (in centistokes) was measured by timing a fluid between lines C and E. Image credit: Sigma Aldrich.

After chilling to 20°C, blood samples were loaded into the viscometer and timed in triplicate similarly to the standards. The same viscometer was used for standards and blood samples. Between each sample, the viscometer was washed thoroughly with acetone and left to dry to ensure no water influenced the viscosity testing. The time, in seconds, was plotted (Fig. 2) against the "known" viscosity of the standard to create a polynomial curve ($R^2 = 1$) that would be used to determine the unknown viscosities of the blood samples (Engineering Tool Box).

Bloodstain Pattern Analysis

Traditional BPA was tested for accuracy by comparing experimental angles of impact to expected or dropped angles of impact (the angle at which the clipboard was fixed). Expected angles were formed using a clipboard and protractor set-up, in which a volunteer held the clipboard at the following angles: 10°, 30°, 60°, and 90°. A plastic pipette was used to drop the blood samples onto cardstock paper. Five drops were made for each of the four tested angles (Fig 3). Stains were made for each participant for a total of 180 bloodstains that would be used

for comparison of each other. The cardstock paper was attached to the clipboard and set to the angle of impact before blood patterns were made. Drops were made approximately 0.3048 m (1 ft) above the paper and a new paper was used for each angle. Patterns were left to dry overnight before analysis and upon drying, the width and length of each stain was measured. Experimental angles of impact were obtained using the following equation:

$$\Theta \text{angle of impact} = \sin^{-1}\left(\frac{w}{l}\right)$$

where w and l are the width and length of the blood droplets, respectively. The length of the pattern runs parallel with the tail of the blood drop, while the width runs perpendicular with the tail. In BPA, the length of the stain is measured by omitting the length of the bloodstain tail. The measured angles were averaged between the five stains on each paper. Experimental angles of impact were compared both physically and statistically to expected angles. Spines, satellite drops, and individualistic formations were also documented for each blood pattern.
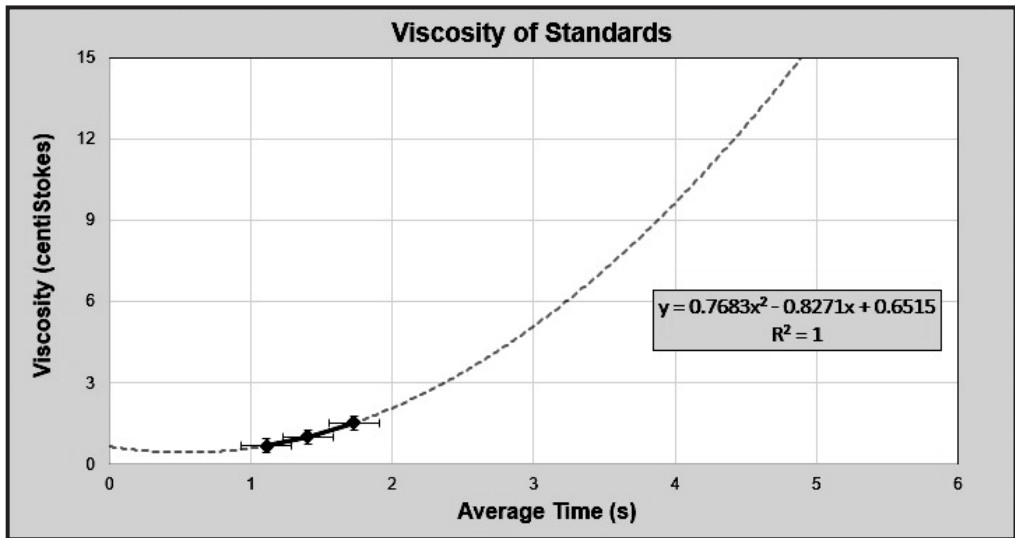
Figure 2. Polynomial curve of standards run through the Cannon Fenske viscometer. Known viscosities of the three standards are as follows: toluene (0.6800 cSt), distilled water (1.0038 cSt), and absolute ethanol (1.5200 cSt). Standard error was calculated to be ±0.25 s. for time calculations.
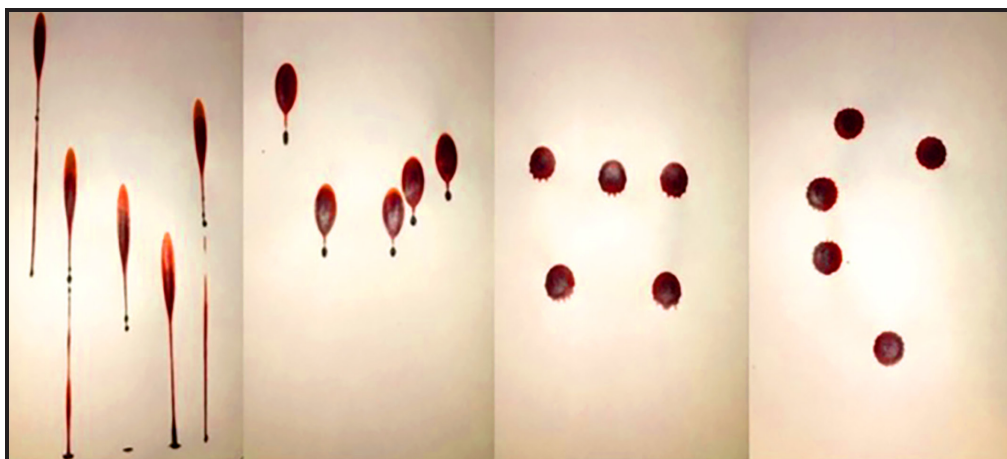
Figure 3. Traditional BPA at 10°, 30°, 60°, and 90° for the control blood sample. Satellite stains were documented if present. Five stains were made at each angle.

Statistical Analysis of Blood Alterations and Angle of Impact

Each blood sample was compared using the following variables: hematocrit, viscosity (in cSt), and angle of impact. Viscosity and hematocrit levels ranged between participants, but statistically the blood samples were considered similar, and therefore were expected to produce bloodstains with similar measurements. Consequently, the average angles of impact between participants were statistically analyzed using analysis of variance (ANOVA). A confidence interval of 95 percent ($\alpha=0.05$) was used. The F-crit and p-values were used to find whether average angles of impact between blood alterations were significantly different and a Tukey Post Hoc test compared each participant between each other for individualism. Finally, a t-test was used to statistically compare expected angles to experimental angles for each participant. Both statistical analyses were used to evaluate the accuracy of BPA regarding hemodynamic influences.

RESULTS

Viscosity and Hematocrit Levels Among Blood-Altered Participants

After averaging the time each blood sample took to cross between lines C and E on the Cannon Fenske viscometer, blood viscosity for each participant was found by using the polynomial curve created by the viscometer standards (Fig. 4). Blood viscosity between the participants varied between 10.29 and 21.22 cSt. The diabetic blood sample appeared to be the most viscous, while the participant with hypothyroidism showed the least viscous blood. The participant with low and high BAC levels showed similar viscosities between the two samples (12.55 and 12.86 cSt, respectively). Because the alcohol intake participant was the same individual for both tests, it is expected that the blood viscosity should not differentiate greatly. The participant had a BAC of 0.02 mg/L on the first blood draw and on the second draw, BAC increased to 0.11 mg/L. The reason for this, was to see how legal and illegal BAC levels (while operating a motor vehicle) could influence blood viscosity. In Figure 4, the viscosities of the alcohol intake participant (with low and high BACs) were similar, signifying that alcohol intake does not greatly alter blood viscosity in one individual. A polynomial trendline has been displayed on Figure 4 to show an increasing viscosity with each blood alteration. Upon testing blood samples for hematocrit levels (% RBC by volume), a figure was made to find the correlation between average blood viscosity and hematocrit (Fig. 5). Patients with hypothyroidism and those on
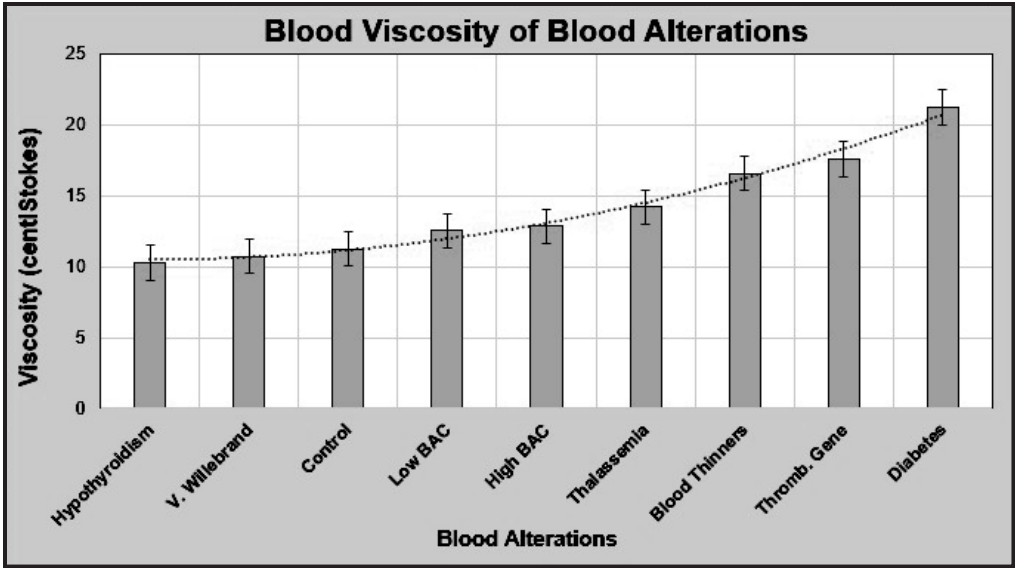
Figure 4. Varying blood viscosities among patients with altered blood. Viscosity ranges from 10.29 to 21.22 cSt. The estimated polynomial trendline shows increasing viscosity levels among the population of physiologically altered blood samples. The hypothyroid patient presents low blood viscosity, while the diabetic patient demonstrates high blood viscosity. Standard error was calculated to be ±1.25 cSt.
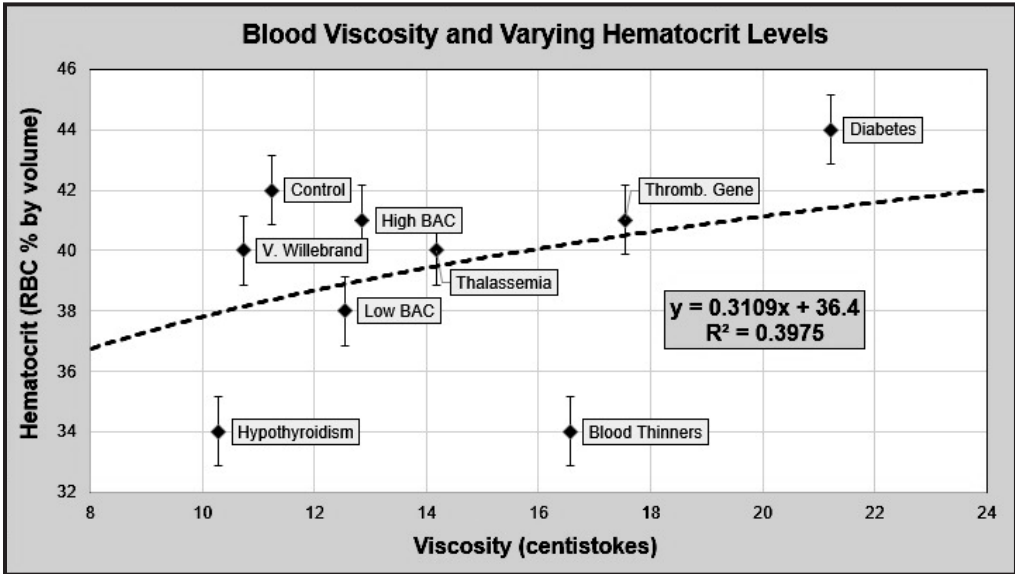
Figure 5. Logarithmic trendline ($R^2 = 0.3975$) shows that blood viscosity and hematocrit levels increase directly proportional to each other. The trendline includes all participants even though the hypothyroid and blood thinner participant appeared far from the trend. Standard error was found to be ± 1% of RBC by volume in this population. Most participants appear above the trendline, but the low alcohol intake patient appears slightly below.

regular blood thinners possessed the lowest hematocrit levels (34% RBC of blood volume). Alternatively, high hematocrit levels were found in the control and diabetic patients (42% and 44% of blood volume). Several of the hematocrit levels lie between 38 percent and 41 percent, and in most, a decreased RBC count was present. In Figure

5, a logarithmic trendline was created, showing that blood viscosity generally increases with increasing hematocrit levels ($R^2 = 0.3975$). In the participants with alterations that typically decrease red blood cell count, such as Thalassemia and Thrombin mutations, data appeared below the trendline. Interestingly, the low BAC participant also appeared below the trendline; the blood viscosity for this participant did not alter between alcohol consumptions, but hematocrit levels increased with alcohol intake. It is expected that alcohol consumption can reduce red blood cell count in heavy drinkers, but the participant was not known to consume alcohol regularly, and therefore red blood cell production may increase with alcohol consumption symptoms (increased heart rate and breathing). The remaining participants appeared slightly above or near the trendline. This provides forensic bloodstain analysts with an estimated trend among individuals with physiologically altered blood in which they can estimate BPA based on hematocrit and viscosity of the blood sample.

Variability Among Blood Alteration Relationships

For each experimental angle, five stains were measured and averaged for each participant. These averages were then analyzed through ANOVA statistics for each tested angle. For the 10°, 30°, and 60° stains, participants were found to produce significantly different stain measurements ($p = 9 \times 10^{-14}$, $p = 9.92 \times 10^{-16}$, and $p = 0.0006$, respectively). In 90° stains, all participants produced similar bloodstains ($p = 0.8929$), and therefore could not be used for individualism. To determine which participant(s) cause variability among the sample, a Tukey HSD comparison test was completed. In this test, 28 relationships among the eight blood-altered participants were cross-examined (one vs. two, one vs. three, one vs. four, etc.). For each relationship, a p-value ($\alpha = 0.05$) was assigned to determine the significance between the two cross-examined participants. A Tukey Post Hoc chart was

created, in which the grey boxes signify relationships that were significantly similar to each other and white areas represent those that were different (Table 1). In the 10° and 30° samples, over 50 percent of participant relationships were found to be different, which signifies individualism and can be used by BPA experts for reconstruction. In 60° stains, only three of the 28 relationships were significantly different from each other showing little individualism among this population in 60° stains. The 90° ANOVA showed that none of the samples were significantly different and all relationships in the Tukey HSD test were deemed similar. Most of the relationships showing 10° and 30° variability came from the relationships paired with the low and high BAC participant. The chart shows similarity in all relationships of the 90° stains, which is due to their spherical nature and less variability among the circular stains. Individualistic properties in bloodstains appear as the angle of impact becomes smaller; subjectivity increases as an analyst must distinguish between the stain body and tail in elongated stains. Spines, satellite drops, and individualistic formations of each bloodstain were analyzed, but no specific pattern was recognized, and therefore these features are not included in the data between blood alterations. Through this analysis, it can be concluded that smaller angles of impact appear to be more individualistic among a population with physiologically altered blood.

Deviations From Expected Angles of Impact

For each experimental angle, the average angle of impact was compared to the angle that each stain was dropped at to test for BPA accuracy among participant blood samples. For example, if a stain was formed by dropping the blood at a 10° angle, the experimental angle that is calculated upon measuring the stain should be significantly close to 10°. A one sample t-test ($\alpha = 0.05$) was used to compare the average experimental angle of impact to each known expected angle. The control sample was removed from this analysis because

Table 1. Tukey Post Hoc plot showing significant differences existing between physiological blood alterations at measured angles (10°, 30°, 60° and 90°.)*

10 degree bloodstains

	Low BAC	Thalassemia	High BAC	V. Willebrand	Hypothyroidism	Diabetes	Thromb. Gene	Thinners
Low BAC								
Thalassemia								
High BAC								
V. Willebrand								
Hypothyroidism								
Diabetes								
Thromb. Gene								
Thinners								

30 degree bloodstains

	Low BAC	Thalassemia	High BAC	V. Willebrand	Hypothyroidism	Diabetes	Thromb. Gene	Thinners
Low BAC								
Thalassemia								
High BAC								
V. Willebrand								
Hypothyroidism								
Diabetes								
Thromb. Gene								
Thinners								

* Dark gray boxes indicate similarities between physiological blood alterations at four traditional BPA angles. Tukey Post Hoc analysis shows more significant differences between blood alterations as the angle of impact decreases or becomes more parallel to the surface it impacts.

Table 1. (continued)

60 degree bloodstains

	Low BAC	Thalassemia	High BAC	V. Willebrand	Hypothyroidism	Diabetes	Thromb. Gene	Thinners
Low BAC								
Thalassemia								
High BAC								
V. Willebrand								
Hypothyroidism								
Diabetes								
Thromb. Gene								
Thinners								

90 degree bloodstains

	Low BAC	Thalassemia	High BAC	V. Willebrand	Hypothyroidism	Diabetes	Thromb. Gene	Thinners
Low BAC								
Thalassemia								
High BAC								
V. Willebrand								
Hypothyroidism								
Diabetes								
Thromb. Gene								
Thinners								

* Dark gray boxes indicate similarities between physiological blood alterations at four traditional BPA angles. Tukey Post Hoc analysis shows more significant differences between blood alterations as the angle of impact decreases or becomes more parallel to the surface it impacts.

it was proven to be significantly similar to all expected angles. Using the remaining eight participants, a stacked bar graph was produced to display the significance between experimental and expected angles of impact for each participant (Fig. 6). The black bars signify experimental angles that were significantly different than expected angles, while the gray bars show when experimental and expected angles of impact were significantly similar. Figure 6 shows that, for each tested angle, over 50 percent of participants produced bloodstains that were significantly different from the angle that the stain was dropped. Interestingly, seven of the eight blood-altered participants deviated significantly from the 30° angle of impact. The hypothyroid and diabetic blood samples deviated significantly from almost every expected angle of impact, showing that BPA may be negatively influenced among these patients. Conversely, the low and high BAC bloodstains showed significant similarity to expected angles of impact, suggesting that BAC may not influence BPA. Therefore, the results among the eight tested individuals

may be used to assist BPA or challenge its techniques.

DISCUSSION

BPA has historically been used as an efficient method for crime scene reconstruction. Various BPA methods have been introduced to the forensic science field, but few analyze the relationships between angles of impact and hemodynamic factors among physiologically altered blood. Theoretically, it was expected that patients with blood alterations may cause inaccuracies in BPA. Hence, in the present study, BPA was evaluated for accuracy when individual hematocrit levels and blood viscosity change between physiological alterations. According to the results presented above, hemodynamic factors have been shown to cause inaccuracy in BPA among the tested population. In ≥ 50 percent of the tested population, hemodynamics caused significant deviations from four angles of impact (Fig. 6). As provided in (Table 2), a one sample t-test demonstrates the cases where hemodynamics may play a
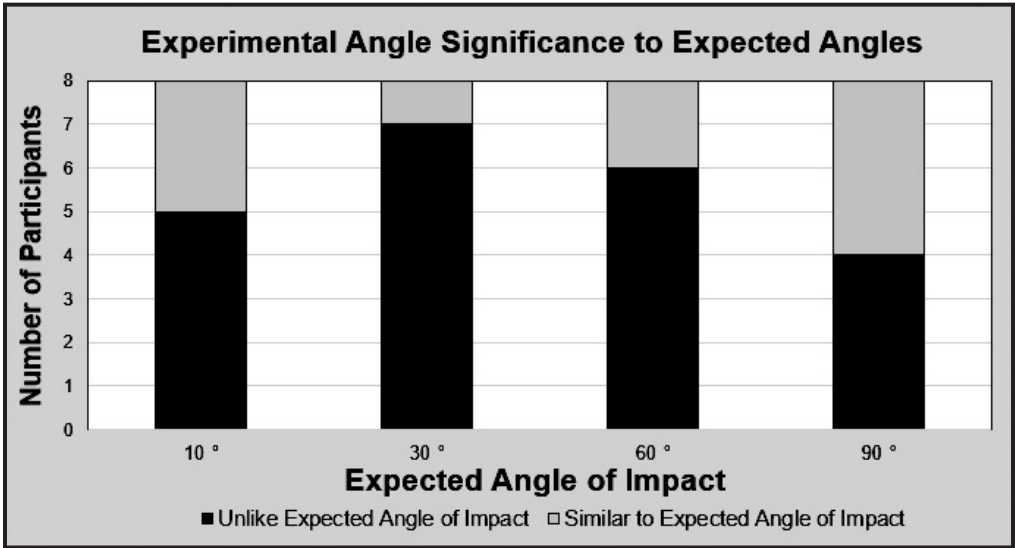
Figure 6. One sample t-test omitting the control sample and placed into a staked bar graph. Black bars show significant differences between the experimentally measured angles of impact and expected angles that blood samples were dropped. Gray bars signify when expected and experimental angles of impact were significantly similar to each other. In almost all expected angles, the diabetic and hypothyroid patient showed significant deviations from expected angles, showing individualism among these participants from the population.

Table 2. One sample t-Test for accuracy of experimental angles of impact among blood-altered participants.

Participant	10° p-value	30° p-value	60° p-value	90° p-value
Low Alcohol Intake (BAC=0.02)	0.0006	0.56	0.0285	0.0705
High Alcohol Intake (BAC=0.11)	0.0006	0.0131	0.219	0.0758
Thalassemia	0.8985	0.0002	0.0251	0.0192
Diabetes (Type 1)	0.7971	0.0001	0.0003	0.0193
Hypothyroidism	0.0106	0.0435	0.4753	0.0219
Von Willebrand's	0.1112	0.0001	0.0202	0.0194
Prothrombin Gene Mutation	0.0398	0.0001	0.0087	0.2001
Regular Blood Thinners	0.0395	0.0002	0.0357	0.0721

crucial role in BPA. Those with a significant p-value (<0.05) displayed inaccurate BPA and should be noted for future research. Individuals that produced erroneous bloodstain measurements may compromise area of origin measurements or the overall crime scene reconstruction.

Additionally, participant individualism was found to increase as angle of impact became more parallel to the surface that they strike. As seen in Table 1, the trend appears to follow previous research which shows inaccuracies in measuring stains with long tails. Consequently, BPA techniques and protocols must compensate for this observation. This is particularly important in forensic science because crime scene reconstruction relies heavily on BPA accuracy. If an analyst is aware that a suspect or victim may have a blood disorder/disease or has consumed alcohol, they can use this to more accurately measure bloodstains. Inaccurately measured bloodstains can three-dimensionally alter the area of origin measurement for all measured bloodstains and produce invalid crime scene reconstruction.

Findings of this study show that hemodynamic factors, such as viscosity and hematocrit levels have the potential to increase the accuracy of BPA. Therefore, it

is recommended that the efficiency of this finding is displayed in larger populations of patients with altered-blood. All participants were female, and therefore this finding must be tested among a male population, where hematocrit levels may be higher. Samples should also be cross-examined among a smaller age range. In this survey, a wide variety of ages were tested, in which hematocrit levels could have been altered based on stage of development.

Calibrated BPA equipment should also be used for future research to provide stable conditions for blood depositing. Blood samples dropped for BPA should be of a similar volume and angles of impact should be accurately determined with power-driven mechanisms. Intravenous viscometers would additionally prevent blood from decreasing in temperature once outside of the body, contributing to the results found in this study without chilling all of the samples. Finally, viscometer standards should be closer to that of blood, to create an accurate standard curve. The results obtained in this study must be reinforced with additional testing to determine how the area of origin can be individualistically influenced by physiological blood disorders/diseases. Based on the findings of this study and by considering new BPA variables, it

is suggested that a standard BPA protocol/ technique, that considers hemodynamics, may be useful in forensic science and could potentially reinforce expert witnesses in a court of law.

Conclusion

The results of this study emphasize that forensic analysts may have to consider hemodynamic factors, such as viscosity and hematocrit levels, when attempting crime scene reconstruction. Altogether, this survey has begun to produce a standard method for taking these factors into consideration, especially when bloodstains have originated from an individual with a known physiological blood alteration. This could be of interest for bloodstain pattern analysts in which BPA can be used more effectively by an expert witness and more importantly to individualize suspects and victims.

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57ⁿᵈ Annual Conference 2019

Building a Conservation Ethic in the New Outdoor Economy

February 26 - March 1, 2019
Delta Hotels Marriott – Helena Colonial
Helena, Montana

Liz Bradley, President Elect 2018-19
Montana Chapter of The Wildlife Society

Introduction

Our Greetings! This conference has always been a highlight of the year for me as a wildlife professional. It's an opportunity for all of us to get together in one place, catch up, and share what we've been learning. Whether we are current professionals, retirees seasoned with wisdom or the future generation of wildlifers, it's a great time to visit and celebrate one another's research and accomplishments. We all have something to learn from and help cultivate in each other. I'm honored to be your incoming president and to work with an amazing team to carry on The Wildlife Society tradition by helping put together this conference. I hope you will all enjoy it and ideally, draw a little inspiration from it, too.

Our conference theme this year is "Building a Conservation Ethic in the New Outdoor Economy." It has been 20 years since we explored recreation as a conference topic. I believe it is more relevant now than ever, with a growing recreation economy in Montana. Most of us are outdoor enthusiasts and were likely drawn to wildlife work because of our love of the outdoors. If you're like me, you are happiest with the stars as a roof over your heads. Here's a little more about the theme:

> The outdoor recreation industry is growing faster than ever in Montana. More people are flocking to parts of Montana because of our incredible natural beauty and outdoor opportunities. Wild places that may have been a secret a decade ago are more accessible to people than ever from information sharing through social media and new technologies that make it easier to get there and people are recreating in new and diverse ways. As growing pressures on wildlife and habitat continue to build, we need to foster and grow our constituencies. On the one hand we need more people to get outside and care about wildlife and wild places. But are we adequately connecting and helping build a conservation ethic in this growing sector of the public that are already out there? This conference will explore the changing face of recreation as it relates to conservation and the challenges and opportunities therein.

I was drawn to this topic not only because of its impacts on wildlife conservation but also its relevance to our Montana culture and our own outdoor ethics. We have five outstanding plenary speakers and an excellent banquet speaker who will explore this topic. On Tuesday evening after the welcome reception we will have Movie Night and show four films that explore the wildlife/recreation interface.

The Board has worked hard this year to encourage student participation in the conference by providing more grants for student travel for more colleges, continuing to support our MSU and UM student chapters, and running a student artwork contest (see cover!). Overall, we have a great selection of workshops, talks, speakers, awards, raffles, student participation and social opportunities. Welcome to the 57th Annual Conference!

MONTANA'S NEW OFFICE OF OUTDOOR RECREATION: ECONOMICS AND STEWARDSHIP

Rachel VandeVoort, Director, MT Governor's Office of Outdoor Recreation

Outdoor recreation is not only intrinsic to the Montana lifestyle and a bedrock reason why so many of us call the Big Sky home, it is also one of the most crucial parts of the state's economy. According to the Outdoor Industry of America's most recent figures, outdoor recreation is now the single largest sector of Montana's economy. It generates over $7 billion per year in consumer spending and supports over 70,000 jobs that pay more than $2 billion worth of wages annually. Those figures don't even account for indirect economic effects, such as the thousands of startups and small businesses that choose to locate here because of the state's outdoor recreation opportunities. In many ways, America is in the midst of an outdoor recreation renaissance, a trend driven by a growing appetite to explore and connect with nature combined with rapid advances in innovation and awareness. The National Park Service and its array of sites are enjoying all-time high popularity. State parks across the U.S. are experiencing similar record crowds, including throughout Montana. The evolution of gear and activities is driving more people outdoors in search of unique experiences.The new Office of Outdoor Recreation could play several critical roles in Montana's outdoor recreation economy: from helping protect public lands to helping properly fund the state and federal agencies responsible for the stewardship of our land, water and wildlife. It could also help rural communities gain access to resources to fully capture the benefits of outdoor recreation, from building trail systems to creating better outdoor infrastructure. Protecting and enhancing this infrastructure is essential to conserving our most vital and sustainable resources and we need to reframe the way we look at its impact on the future of Montana's economy.

INTERSECTING THE RECREATING PUBLIC WITH WILDLIFE AND HABITAT CONSERVATION: IT'S ABOUT PASSION, DATA AND ADVOCACY

Lauri A. Hanauska-Brown, Wildlife Division, Montana Fish, Wildlife and Parks, Helena

Campaigns such as Hooked on Fishing, Nature RX, Every Kid in a Park and Families Afield urge Americans to get outside to fish, hunt, camp or just observe. Some campaigns focus on increasing access to the outdoors and some even make it less expensive to recreate by offering loaner gear or waived park entrance fees. While increasing the general public's interest in the outdoors is good for them and economies of outdoor industries it does present resource managers with a challenge of balancing the needs of wildlife and habitat with increased human presence. Many recreationists already have a vested interest in the outdoors and may be cognizant of the potential impacts of their activities while people entirely new to outdoor recreation may not. It behooves organizations and agencies to engage all these recreationists as part of the larger conservation army in attempts to limit their impact and put their time outdoors to use. Attentive recreationists can help fill data gaps with new mobile applications such as Survey 1,2,3 and iNaturalist that make it easy for them to report the wildlife or even the weeds they see. Submitting information that is used to inform species distribution models and management strategies can result in recreationists feeling like they are part of the monitoring and conservation team. Citizens who volunteer their time to check osprey nests along the Yellowstone River, hair snares and camera traps for carnivores, and the 180,000 hunters who share information on their game harvest or wildlife observations

each year are part of a team of recreationists vested in wildlife management and conservation. Specific programs like Climbers for Bat Conservation encourage recreationists with a passion for climbing to learn more about the animals they observe during their time on the rock. Climbers in Colorado have become advocates who contribute to bat conservation not only by collecting data but also by bringing climbers resistant to sharing their knowledge into the discussion. Being part of the team creates a sense of stewardship as seen when these same birders, skiers, hunters and climbers show up at public meetings where wildlife management, public land access, and habitat conservation are being discussed. Encouraging recreationists to add their voices to specific campaigns is another way to put their passions to use and secure their investment in the outdoors. Local movements like the Montana Outdoor Heritage Project or federal legislative proposals such as the Recovering America's Wildlife Act provide platforms for recreationists to support legislation and funding that will help conserve the places, access and species they love. Engaging all recreationists through our shared passions, information sharing, and advocacy will hopefully lead to a contagious enthusiasm for conservation and responsible use of the resources they enjoy while outside.

WHAT'S OLD IS NEW AGAIN: PROTECTING THE CORE TENETS OF CONSERVATION, NORTH AMERICAN MODEL AND ETHICS IS THE PATH TO GROWTH

Ryan Busse, Chair of the Board of Directors, Backcountry Hunters & Anglers

Hunters and anglers are the traditional "constituency" or "beneficiary" of conservation policy and public lands use. For many years the general assumption has been that this constituency is getting older, more insular and less relevant. This has forced a myriad of questions about the way we manage our resources. Who has the political clout to influence this management? Forces that wish to privatize or monetize land, water and wildlife have seized on this from multiple directions. They have mobilized fear of "a disappearing lifestyle" to encourage segments of outdoorspeople to tear at traditionally proven management policies such as the North American Model. They have embraced media and personalities within it to chip away at ethics, poke at professional management and open political chasms. By doing this they have driven potential new hunters and anglers from our ranks and have repelled growing allied constituency groups in the outdoor sports arena. BHA and other examples I will offer tell another story – a hopeful and exciting story about how the core principles of wildlife professionals are laying the groundwork for dynamic new advocacy and an era of increased participation in the outdoor world. In this new era, ethics, wild places, core principles of wildlife management and the managers themselves are cornerstones of success. It's an era where new people, especially young people, join movements because of wild food and ethical activities which connect them to nature. Where old, divisive assumptions about politics and policies are dissolved. Where people are eager to be participants in improving and protecting resources, not just using them. It's an era where the hard work and dedication of professionals in conservation help drive growth of an org like BHA, which now counts over 30,000 members and is growing every day. Where new hunters and non-hunters join because of a shared vision of the importance of wildness in the lives of everyone.By staying the course and holding to bedrock, fundamental principles, wildlife and resource managers have helped invigorate a new age of activism.

Conservation and Recreation in the Greater Yellowstone Ecosystem

Caroline Byrd, Executive Director, Greater Yellowstone Coalition

Participation in outdoor recreation is increasing and communities of the Greater Yellowstone Ecosystem (GYE) are growing. Increasing recreational pressure is putting demands on resource strapped land management agencies and driving wedges between historically aligned interests around how to balance conservation values and recreational access. The GYE is unique in that it still hosts all the native carnivores and migratory ungulates and is comprised of vast wild landscapes. At the same time, outdoor recreation is important to the social and economic fabric of many places in the west, and threats like climate change and public land transfer highlight the need for a unified conservation and recreation movement. Given this context, we conducted an inventory of outdoor recreational use to better understand the benefits and challenges associated with recreation in the GYE. Following the inventory, we convened a symposium of diverse interests to begin to grapple with the growing tension between outdoor recreation and conservation. People we interviewed said they value wildlife, natural history, and solitude in the places they recreate. Yet we found important areas for wildlife and quiet, wild places intersect with hotspots of recreation infrastructure and demand. There are a variety of challenges ahead in building solutions that balance conservation and recreation, but a common theme at the symposium was interest in finding ways to build a new ethic for recreation in the GYE. At GYC, we think of it as Leave No Trace 2.0, and envision shared commitment to practices that inform how we recreate in the places we love, to ensure they stay wild and healthy for generations to come. Cultural shifts take a long time, but a few key questions surfaced throughout our work that should be considered in any efforts moving forward:

What does a new ethic need to look like (i.e. what are some best recreation practices) and what would it do?

What communities have historically been underrepresented in outdoor recreation and conservation and how do we ensure that building a shared vision for recreation in the GYE is inclusive?

If we are to build a new ethic, how do we collectively change our expectations (especially our expectations of public land management) in ways that ensure our recreational pursuits and conservation values align?

Welcoming Conflict and Cultivating Ethics

Hal Herring, Contributing Editor, Field and Stream Magazine

We're going to see lots of pressures in the years to come. We must welcome conflict, and we must be open minded enough to know that we might find ourselves on one side of it at one time, and another some other time. If peregrine falcons need nesting cliffs to be closed to rock climbing during certain months, then by all means, close them, but be prepared to be specific about the need, and the places. Have the staff to carefully define the need. Blanket closures smack of laziness, and of unacceptable fiat. Make the case. Welcome dissent. Conflict is exhausting, and federal agencies, often with overworked employees, sometimes have little patience with it, but patience for conflict is necessary. Fiats will be met with resistance, and resistance can include anger enough to taint our politics. We have to meet with shared goals. We need, for so-called non-consumptive users (ie. non-hunters and fishermen) to create a way for them to bring money to support the lands and wildlife that we all love. A "backpack

tax" - whatever. And no, it can't be just voluntary. Conflict is great, but when one side of the conflict is footing all the bills, then the argument of the other side is simply not going to be heard with the same gravity it might deserve.We're going to see lots of pressures in the years to come. We must welcome conflict, and we must be open minded enough to know that we might find ourselves on one side of it at one time, and another some other time. If peregrine falcons need nesting cliffs to be closed to rock climbing during certain months, then by all means, close them, but be prepared to be specific about the need, and the places. Have the staff to carefully define the need. Blanket closures smack of laziness, and of unacceptable fiat. Make the case. Welcome dissent. Conflict is exhausting, and federal agencies, often with overworked employees, sometimes have little patience with it, but patience for conflict is necessary. Fiats will be met with resistance, and resistance can include anger enough to taint our politics. We have to meet with shared goals. We need, for so-called non-consumptive users (ie. non-hunters and fishermen) to create a way for them to bring money to support the lands and wildlife that we all love. A "backpack tax" - whatever. And no, it can't be just voluntary. Conflict is great, but when one side of the conflict is footing all the bills, then the argument of the other side is simply not going to be heard with the same gravity it might deserve.

Alphabetical By Presenter's Name
* Denotes Presenter
** Indicates Student Presentation

CHRONIC WASTING DISEASE IN MONTANA

Emily Almberg*, Wildlife Health Program, Montana Fish, Wildlife, & Parks, Bozeman
John Thornburg, Wildlife Health Program, Montana Fish, Wildlife, & Parks, Bozeman
John Vore, Wildlife Division, Montana Fish, Wildlife, & Parks, Helena
Jennifer Ramsey, Wildlife Health Program, Montana Fish, Wildlife, & Parks, Bozeman
Keri Carson, Wildlife Health Program, Montana Fish, Wildlife, & Parks, Bozeman
Justin Gude, Research and Technical Services, Montana Fish, Wildlife, & Parks, Helena

In the fall of 2018, Montana Fish Wildlife and Parks (MFWP) conducted monitoring and surveillance for chronic wasting disease (CWD) along the northern Hi-Line, around Philipsburg, and south of Billings, MT. MFWP detected 26 new cases of CWD from over 1941 samples tested from mule deer (*Odocoileus hemionus*), white-tailed deer (*Odocoileus virginianus*), elk (*Cervus elaphus*), and moose (Alces alces). Of the new detections, 21 were along the Hi-Line in every county from Liberty County east to the North Dakota border, and 5 were detected within the CWD-positive area south of Billings. Prevalence of CWD in the northern CWD-positive hunt districts on the Hi-Line averaged 2% (95% CI: 1-3%) in mule deer, and 1% (95% CI: 0-3%) in white-tailed deer. South of Billings, CWD prevalence was estimated to be 2% (95% CI: 1-3%) in mule deer and 1% (95% CI: 0-3%) in white-tailed deer. Prevalence varied across hunt districts within the CWD positive areas; for example, among neighboring hunt districts south of Billings, CWD prevalence in mule deer ranged from <1% (95% CI: 0-3%, HD 502) to 6% (95% CI: 3-12%, HD 510). MFWP continues to plan for long-term CWD management in positive areas. In 2019, MFWP will consolidate "CWD positive areas" and "Transport Restriction Zones" into the single moniker "CWD Management Zones," and the southern portion of MFWP Region 7 will be included in the southern CWD Management Zone in anticipation of finding CWD positive cervids in that area. CWD surveillance/monitoring during fall 2019 will be focused in southeastern MT, around Philipsburg, and along the Hi-Line.

ESTABLISHING RANGE FOR MONTANA'S RAREST BAT SPECIES: THE NORTHERN MYOTIS

Dan Bachen*, Zoology, Montana Natural Heritage Program, Helena
Braden Burkholder, Information Systems and Services, MT Natural Heritage Program, Helena
Heather Harris, Wildlife Division, Montana Fish, Wildlife & Parks, Glasgow
Mike McGrath, Ecological Services, U.S. Fish and Wildlife Service, Helena, MT
Brandi Skone, Wildlife Division, Montana Fish, Wildlife & Parks, Miles City

In 1978 a single Northern Myotis (*Myotis septentrionalis*) was found overwintering in a coal mine along the lower Missouri River. For 38 years this was the only confirmed detection of the species within the state. In response to population declines caused by White-Nose Syndrome across the eastern and central US, the species was listed as threatened by the US Fish and Wildlife Service in 2015. To better understand the species distribution within Montana, we began targeted surveys in 2015 using mist nets along the Missouri, Yellowstone,

Tongue, Powder, and Little Missouri rivers and forested areas in southeastern Montana. Through August 2018, we have conducted 55 surveys and captured 20 individuals at seven sites, expanding the known range to include forests along the Missouri and Yellowstone rivers upstream from the North Dakota border to Poplar and Glendive. Initial surveys following established protocols where nets are placed over water to capture drinking animals captured few Northern Myotis. To increase efficacy, we developed a novel technique targeting flyways within forested areas, which increased capture success at new and previously surveyed sites. Using this capture data, we predicted suitable habitat within and outside of known range with a presence-only model implemented in Maxent. The model indicates moderate to highly suitable habitat exists along the Missouri river upstream of the current known range. Future surveys should target these areas, as well as areas lacking survey effort adjacent to known populations in Wyoming to delineate the species range within the state.

MISSION OF THE MONTANA WILD BISON RESTORATION COALITION

James A. Bailey*, Coordinator, Montana Wild Bison Restoration Coalition, Belgrade

Domestication is the major threat to persistence of wild plains bison on native range in the USA. It is the opposite of wild. For Montana bison (*Bison bison*), there is a legal definition and a biological definition of "wild". But there are no wild bison, year-round in Montana. Although Olaus Murie recommended bison restoration in eastern Montana in 1937, the state has been unable to establish a public, wild herd. The landscape on and near the Charles M. Russell (CMR) National Wildlife Refuge is the best location for restoring wild plains bison in the USA. The Montana Wild Bison Restoration Coalition was formed to disseminate information on issues and opportunities for bison restoration in Montana and to promote bison on and near the CMR Refuge (see mtwildbison.org). The Coalition supports at least 1,000 bison on at least 100 square miles of diverse habitat.

MONTANA'S STATE OF THE BIRD (MONITORING)

Allison J Puchniak Begley*, Wildlife Division, Montana Fish, Wildlife & Parks, Helena
Lauri Hanauska-Brown, Wildlife Division, Montana Fish, Wildlife & Parks, Helena

The State of Montana has a long history of monitoring birds and has some of the most robust data on life history, status and trend. However, inventory and monitoring of some species remains challenging with current methodologies. There are 433 bird species in the state with 283 documented breeders, and 233 overwintering residents. Broadscale landbird monitoring, species or taxa specific surveys and local assessments have all been used to provide a picture of the status of each bird species in the state and inform conservation of sensitive species. Montana Fish, Wildlife & Parks is currently reviewing available avian trend, density and occupancy data from broadscale monitoring programs such as the USGS Breeding Bird Survey (BBS) and Integrated Monitoring in Bird Conservation Regions (IMBCR). BBS provides long-term trend data and IMBCR provides information on population density and species occupancy estimates. Individualized surveys provide increased focus on species of concern or those species or habitats that are underrepresented or inadequately surveyed by large-scale programs. These monitoring programs vary in their ability to provide information necessary for status assessments across species due to difficulties accessing some areas and habitats at biologically relevant time periods and detecting cryptic or rare species. By reviewing these data and incorporating known status and habitat types provided by the Montana Natural Heritage Program, FWP plans to highlight the status of bird species of interest and outline needs for monitoring in the state. Given the different products and methodologies, this summary will also serve to recognize the combined value of these programs, as each program provides different types of biological information to resource managers.

Motus: Developing the Intermountain West Collaborative

William Blake*, Avian Science, MPG Ranch, Florence, MT

Kate Stone, Avian Science, MPG Ranch, Florence, MT

Migration is essential to thousands of species throughout most animal groups. Yet, we still do not know the migration strategies in most species, including winter grounds, migration patterns, and stopover sites. While larger wildlife performs well with tracking devices, most migratory wildlife is too small. The Motus Wildlife Tracking Network helps to fill that void by using small transmitters called nanotags and coordinating the placement of automated receiving units. Researchers deploy nanotags that operate on a single VHF frequency, but with different signal bursts allowing identification of individual organisms. Motus stations then detect the signals in the field, and data are uploaded and shared with collaborators. This community approach to science allows small organisms to be tracked long distances as they pass multiple Motus stations. The Motus network is most active in eastern North America, but also extends to wintering grounds of many western migratory species. In 2018, MPG Ranch established the Intermountain West Collaborative Motus project in the Bitterroot Valley, MT. We are currently expanding the network to gain coverage in more of western Montana and Idaho. We have worked with partners to deploy nanotags on Gray Catbirds (*Dumetella carolinensis*)and several bat species and will expand nanotag work in 2019. Already, we have detected one of our catbirds at a station in Texas. We hope this talk will encourage a dialogue with Montana researchers about potential sites for Motus stations and how they might deploy nanotags to make use of the network.

Avian Response to Old-Growth Maintenance Logging in the Swan River State Forest

Leah Breidinger*, Trust Lands, MT Dept of Natural Resources and Conservation, Kalispell

Ross Baty, Forest Management Bureau, MT Dept of Natural Resources and Conservation, Missoula

Logging in old-growth forests and how to maintain this age class on the landscape have long been concerns in the northwestern United States. Old-growth maintenance silvicultural treatment is a tool the Montana Department of Natural Resources and Conservation (DNRC) implements to retain old-growth attributes, remove encroaching shade-tolerant trees, and create small canopy gaps. Our objectives were to examine how these treatments effect avian diversity and density of associated bird species. We used a Before-After/Control-Impact Pairs (BACIP) study design and paired old-growth stands proposed for harvest (treatment) with untreated stands (control) based on habitat similarity. Vegetation was also measured in study stands. Logging created small openings, reduced basal area by 40%, and reduced overstory canopy cover by 31%. No large changes in bird species composition or diversity were detected. However, relative densities of evening grosbeaks (*Coccothraustes vespertinus*) (P=0.03) and golden-crowned kinglets (Regulus satrapa) (P=0.09) decreased significantly following harvest, while pileated woodpecker (Drycopus pileatus) (P=0.07) and dark-eyed junco (*Junco hyemalis*) (P=0.06) densities increased significantly. Removal of insect-infested trees likely displaced evening grosbeaks and golden-crowned kinglets, while new openings and added logging slash likely attracted dark-eyed juncos and pileated woodpeckers. Old-growth associated birds continued to occupy treatment stands with some species increasing in density and others decreasing under the landscape conditions we observed. We did not evaluate avian survival or reproductive success, which would provide beneficial metrics for further interpretation of results.

Estimating Natal Origins of Juvenile Northern Goshawks using Stable Hydrogen Isotopes

Brian Busby*, Raptor View Research Institute, Missoula, MT
Robert Domenech, Raptor View Research Institute, Missoula, MT
Adam Shreading, Raptor View Research Institute, Missoula, MT

From 2004 to 2007, we collected hatch-year feathers from 44 juvenile Northern Goshawks (*Accipiter gentilis*) captured at our Rocky Mountain Front banding station near Lincoln, Montana. Due to the relative scarcity and secretive nature of goshawks, little is understood about their migration patterns or the geographic origins of birds encountered at raptor migration count and banding sites. Most studies suggest goshawks are partial migrants, often moving <100km, but select band returns and radio and satellite telemetry have shown that some individuals occasionally travel thousands of kilometers. We performed a stable hydrogen isotope analysis on the feathers we collected from young goshawks to determine their predicted natal origin. We found that 69% of goshawks had predicted natal origins relatively close to our capture site, 24% from areas in northwestern Canada and eastern Alaska, and 7% somewhere significantly south, east or west of our capture site. We did not find any significant patterns with sex and passage date or latitudinal origin, nor did we find a meaningful relationship between latitudinal origin and passage date. Our findings support the current understandings of goshawk migration and dispersal, with most individuals traveling short distances from their natal grounds and a few outliers traveling great distances, not always in a southerly direction.

Miners Gone Birdy! The Importance of Citizen Science and Speciation in Waterfowl Protection at the Berkeley Pit, Butte, Montana

Stella Capoccia*, Biological Sciences, Montana Tech, Butte
Gary Swant, GoBirdMontana, Deer Lodge, MT
Mark Mariano, Rampart Solutions, Butte, MT
Lindsay Torpey, Biological Sciences, Montana Tech, Butte
Justine Desjardin, Biological Sciences, Montana Tech, Butte

This work demonstrates how citizen science is improving the waterfowl protection plan at the Berkeley Pit. On November 28th, 2016, tens of thousands of Snow and Ross's geese (*Chen caerulescens* and *Chen rossii*) landed on the Pit during their migration south; roughly 3000 died during the event. The occurrence initiated a change in the waterfowl protection plan at the Pit. For the 20 years prior, avian monitoring took place multiple times per day and mitigation action occurred for every bird observed on the Pit water. Records were taken on bird type, numbers, time of day, and hazing action. Little was known about species, activity patterns, and variation in seasonality which made it challenging to understand the complexities of mitigation actions. In 2017, we trained the mining personnel to identify the waterfowl known to the region so new records include species and sometimes sex and age class. We now have a growing body of data that shows species and corresponding differences in bird activity. We are using these data to understand how these differences vary in fall versus spring migrations and how to match mitigation and bird type. This undertaking exemplifies how utilizing citizen science improves protection efforts. The importance of this initiative is supported by research by Belt and Krausman (2012) who emphasize that well-trained citizens - in this case, miners - generate data and results on par with scientists. Ultimately, we demonstrate how the use of citizen science will result in improved strategies for waterfowl protection at the Berkeley Pit.

Grizzly Bear Recovery and Delisting: Progress Report

Hilary Cooley*, Grizzly Bear Recovery Program, U.S. Fish and Wildlife Service, Missoula, MT

The USFWS listed grizzly bears (*Ursus arctos horribilis*) as a Threatened species in the lower-48 States in 1975 and subsequently designated six recovery zones. Due to the success of conservation efforts and collaboration among a variety of stakeholders, two of these populations (Greater Yellowstone Ecosystem, Northern Continental Divide Ecosystem) have achieved recovery goals and the USFWS is working towards delisting. Conservation success, however, brings significant management challenges as both populations are expanding into human-dominated landscapes. We review population status, management issues, and litigation challenges. We also present updates on recovery and management in the small Cabinet-Yaak and Selkirk Ecosystems and the currently unoccupied North Cascades and Bitterroot Ecosystems.

** Weasely Recognized or Stoatally Camouflaged: Quantifying Coat Color of a Cryptic Predator

Brandon M. Davis*, Wildlife Biology Program, University of Montana, Missoula
L. Scott Mills, Wildlife Biology Program, University of Montana, Missoula

At least 21 species of birds and mammals across the globe undergo seasonal changes in coloration, molting white in winter to match snow cover to reduce predation risk from visually hunting predators. As snow duration decreases, animals in white winter coats become more conspicuous against snowless ground. For example, camouflage mismatched snowshoe hares (*Lepus americanus*) suffer increased mortality compared to camouflaged hares. Yet, the generality of this climate-induced camouflage mismatch across species is unknown. Given the adaptive value of seasonal camouflage against local snow duration, we hypothesize that sympatric color molting species would show convergent coat color phenology. Therefore, we documented coat color phenology of three sympatric coat color changing species in Montana; short-tailed weasels (*Mustela erminea*), long-tailed weasels (*Mustela frenata*), and snowshoe hares. We used a non-invasive sampling framework consisting of remote cameras and bait tubes to quantify coat color phenology for all 3 species, including molt initiation, rate, and completion. Over a 2-year period we deployed >50 remote cameras over >6000 trap nights. We detected >3000 photographic hare events and >1000 photographic weasel events. Although we are currently completing our analysis it appears that there may be phenology differences between hares and weasels. We conclude that camera trapping is a useful tool for quantifying phenology of sympatric coat color changing species, contributing to the growing knowledge base to determine the potential scope for evolutionary rescue to climate change in wildlife populations.

Eating Their Greens: Relationships of NDVI with Elk Forage and Nutritional Condition

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Kelly Proffitt, Wildlife Division, Montana Fish, Wildlife & Parks, Bozeman
Justin Gude, Wildlife Division, Montana Fish, Wildlife & Parks, Helena
Steve Brown, U.S. Forest Service, Northern Region, U.S. Department of Agriculture, Missoula, MT

The distribution and availability of nutritional resources strongly influence elk (*Cervus elaphus canadensis*) movements and behaviors and are important for females to meet nutritional requirements of lactation, mass gain, and pregnancy. Past studies demonstrated that

elk during the summer select strongly for areas with high values of the Normalized Difference Vegetation Index (NDVI), a remotely-sensed indicator of net primary productivity. There is, however, considerable uncertainty regarding relationships between NDVI, forage availability and quality, and elk nutrition. To fill these knowledge gaps and provide recommendations for managing elk habitat in western Montana, we evaluated the relationships of NDVI with ground-sampled vegetation and nutritional condition measurements from elk in 4 populations (Elkhorn, North Sapphire, and East Fork and West Fork of the Bitterroot). We sampled 510 vegetation plots in summer and 172 elk in fall and winter during 2011-2017. We found some evidence that NDVI was associated with forage quality, forage abundance, and body fat, but not with pregnancy. These results indicate that managing areas for greater levels of NDVI may increase the availability of summer forage and improve the ability of elk to gain body fat; however, NDVI alone is insufficient to fully characterize summer forage. We suggest that combining forest management treatments on public lands with other strategies, such as restricting availability of high-quality forage on private lands, increasing hunter access on private lands, or altering harvest regulations, may provide a more holistic approach to encouraging elk to remain on public lands during the summer and hunting seasons.

EXAMPLE OF MULTI-AGENCY DATA MANAGEMENT TO ADAPTIVELY MANAGE CENTENNIAL VALLEY ARCTIC GRAYLING

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Montana Arctic grayling (*Thymallus arcticus*) declined to about 4% of their perceived historic distribution by the 1990s, which led to formal consideration for listing under the Endangered Species Act. Over the past 70 years numerous hypotheses were posited regarding drivers of the grayling population, including 1) reduction and alteration of spawning habitat, 2) predation by, and competition with, non-native fishes, and 3) limited winter habitat. A multi-agency adaptive management project was initiated to elucidate the relative effect of hypothesized drivers of grayling abundance to direct future management of this population. Data are considered a trust resource. If data are not organized, maintained and accessible, the status, trends, and processes over temporal and spatial scales cannot be addressed. Data are collected by the US Fish & Wildlife Service (USFWS) and Montana Fish, Wildlife & Parks (FWP) and each agency manages it differently. These data include netting, trapping, electrofishing, stream temperature, fish demographics, tagging, spawning habitat, overwintering, beaver dams, and angler harvest data on hybrid cutthroat. FWP Fish Information System (FIS) is a centralized database, commonly referred to as "Godzilla", which houses most fish data for FWP. USFWS does not have a centralized database system but data are entered into relational databases and stored on local servers. The Centennial Valley Arctic grayling database was created to house similarly formatted data from Godzilla and USFWS. The collation of data presented unique challenges. Data are assembled and fed into the models annually, biases are slowly disappearing, and the grayling population is being adaptively managed.

** Annual Elk Calf Survival Following Increased Mountain Lion Harvest

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From 2011 to 2014, we evaluated the factors driving the survival and recruitment of elk (*Cervus elaphus canadensis*) calves in the upper Bitterroot Valley of Montana. After three years, we identified mountain lions as a major source of elk calf mortality and determined that increasing elk calf survival in the southern Bitterroot might have positive effects on elk abundance and recruitment in the area. In 2012, wildlife managers increased harvest opportunity for mountain lions in the study area, which resulted in increased harvest of both male and female mountain lions. To evaluate possible changes in calf survival and cause-specific mortality several years after increased opportunity for mountain lion harvest ended, we monitored 248 elk calves via radio telemetry from 2016-2018 in the study area to determine the timing and cause of death. We estimated period-specific rates of seasonal mortality and cause-specific mortality using data collected prior to ($n = 202$), during ($n = 84$), and after ($n = 248$) increased mountain lion harvest opportunity. Estimated calf survival rates will be compared among the 3 periods of mountain lion harvest opportunity in the upper Bitterroot study area. Possible changes in rates of cause-specific mortality will also be compared among the periods. We will also present the results of an evaluation of the associations between calf survival and (1) spatiotemporal covariates and (2) characteristics of individual calves.

Using GPS Technology on a Migratory Songbird to Determine Overwintering, Migration Route and Connectivity

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Little is known about the connectivity of migratory songbirds in part because mark-recapture methods result in few observations. With rapidly improving technology, tracking devices are becoming more relevant for small animals. We studied migration ecology of Gray Catbirds (*Dumetella carolinensis*) that breed in the Intermountain West. We retrieved 6 geolocator units and, more recently, 12 GPS pinpoint tags that provide information on overwintering location, migration route, and timing of arrival and departure. Our results indicate that catbirds winter in northeastern Mexico and those locations appear to be distinct from wintering locations of eastern Gray Catbird populations. Additionally, our results show that catbirds may have a "second migration" in the spring, moving further south in Mexico at a time when we would expect them to return northward.

** Sugar Content as a Driver of Resource Partitioning Between Foraging Bears and Tribal Harvest of Huckleberries on the Flathead Indian Reservation

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Huckleberries (*Vaccinium* spp.) facilitate important ecological relationships for Native Americans within the Flathead Indian Reservation of Montana and throughout the Pacific Northwest. Huckleberries provide cultural resources such as traditional foods and customs, social elements and economic products. Huckleberries are also an important food source for bears, another culturally important animal for the Confederated Salish and Kootenai Tribes (CSKT) along with many other Native American Tribes. Phenological data on huckleberries is extremely limited. The opportunity for traditional ecological knowledge (TEK) to be utilized to expand knowledge was a key focus for this project. To gain a better understanding of the role that huckleberries play in cultural and ecological networks, ecological characteristics of huckleberries across a range of habitats on the reservation were examined in addition to interviews with enrolled tribal members and tribal elders about historic and modern use of huckleberries. Recorded phenology data at 10 sites at different elevations across the reservation was used to develop a baseline understanding of the time of flowering and berry production. We evaluated the relationship between site productivity of huckleberries and sugar content (measured in brix%) of berries at peak ripeness and compared those metrics with bear use, measured by the amount of bear sign at each site. Bear (*Ursus* spp.) sign was most prevalent at the more remote locations with higher brix% ($R2=0.82$, $p=0.012$) and plentiful berries. Finally, community interviews were conducted with adult tribal members about the importance of huckleberries for the tribal community and the people's understanding of the niche bears maintain concerning huckleberries. This research contributes to collaborative studies in Northwest Montana focusing on huckleberries as a food source for bears in the face of climate change, as well as supporting CSKT in asserting traditional food sovereignty.

Using UAVS, Infrared Cameras and Machine Learning to Count Greater Sage-Grouse

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UAVs have the potential to increase data quality and improve efficiency of wildlife monitoring. However, little information exists on how data collected from UAVs compares to traditional visual based surveys. Here we deployed a UAV autonomously over Greater Sage-grouse (*Centrocercus urophasianus*) leks in Phillips County, Montana to compare traditional field-based lek counts to manual and automated UAV video counts. We conducted 28 paired observations of sage-grouse using traditional visual methods and UAV mounted thermal infrared video. For analysis, we manually counted the sage-grouse in the videos, comparing counts of a trained and untrained observer. We completed field counts before each flight to compare counting methods. Results of our analysis show good agreement between trained and untrained observer manual video counts but less agreement between manual video counts and standard field counts. Next, we built an algorithm to identify, track, count, and size the sage-grouse from the videos. While the algorithm worked exceptionally well tracking and counting, more refinement is necessary to detect all the sage-grouse. The algorithm was also able to

detect a size difference between birds which could signify males versus females. However, methods would have to be developed to validate the size difference detected. Overall, our results show that drones in combination with autonomous methods show enormous potential for counting sage-grouse. Furthermore, counting sage-grouse in the spring is difficult and inconsistent given field conditions. UAVs are tools that can help us overcome tough field conditions and give us visual access to otherwise inaccessible leks.

INTERPRETING AND ADAPTING MONITORING EFFORTS OF A REINTRODUCED POPULATION OF SWIFT FOX IN NORTHEASTERN MONTANA

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Swift fox (*Vulpes velox*) were designated extirpated in Montana in 1969. However, reintroductions initiated in Canada from 1983-1997, reestablished a self-sustaining population in southern Alberta and Saskatchewan and northeastern Montana. Swift foxes are listed as a threatened species in Canada. In Montana, at the northern edge of the species range, swift fox are considered a furbearer, with a limited harvest and a Species of Concern level 3, potentially at risk but abundant in some areas. The swift fox population in Canada and Montana is interdependent and therefore it is important for agencies to collaborate on monitoring efforts that are measured against management and/or recovery goals which may differ across jurisdictional and international boundaries. Four international population surveys have been conducted on the Canada/northeast Montana population in 2000/01, 2005/06, 2014/15, and 2018. Monitoring abundance and distribution of swift fox are key components of the Montana Swift Fox Conservation Strategy. Swift fox are elusive and nocturnal, therefore, conducting surveys for the species is both difficult and expensive. Monitoring data are derived from international population surveys, from harvested fox and anecdotal observations. We will review and compare results of each international survey which has advanced our knowledge of this reintroduced population and has helped guide management of swift fox in northeastern Montana.

** PRELIMINARY RESULTS ON THE DRIVERS OF MOOSE CALVING SUCCESS AND THE IMPACTS OF MOUNTAIN PINE BEETLE EPIDEMICS ON RESOURCE SELECTION

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Understanding factors that influence moose (*Alces alces*) calving success and habitat selection are fundamental to the effective conservation and management of the species. The two primary objectives of this study are to evaluate the effects of willow nutrition on calving success of moose and the effect of mountain pine beetle (*Dendroctonus ponderosae*) epidemics on moose habitat selection and movement. Preliminary research by Colorado Parks and Wildlife has identified differences in calving success between two spatially proximate moose populations with similar geographic and biotic features. As moose diet in this

geographic area has been well documented and a majority is comprised of willow (*Salix* spp.), we investigate the degree to which differences in willow forage quality explain differences in calving success. Both moose populations in our study are surrounded by forests that have been impacted by mountain pine beetles. These epidemics affect forest structure and the availability of thermal cover; however, the impact on wildlife is poorly understood. Thermal cover is thought to be especially important for moose in Colorado at the southern extent of their range. This suggests that moose will display stronger selection against impacted areas in this geographic region. We present a preliminary analysis of moose habitat use and predictions of habitat selection based on the date of mountain pine beetle impact.

Limited Permit Mule Deer Buck Hunting Restrictions in Eastern Montana - Historical Information, Analysis and Public Expectation of Hunting District 652

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Hunting District 652 is a limited permit mule deer (*Odocoileus hemionus*) buck only area, found along the east side of Fort Peck Lake in FWP Administrative Region 6. The district was initially established in 1987 to meet public desire to produce "more and bigger mule deer bucks". Population and harvest objectives were established within the Mule Deer Adaptive Harvest Management (2001) guidelines. Mule deer demographics in this district are monitored annually through post-season aerial survey methods. Hunting and harvest metrics are monitored through an annual survey of successful permit applicants via a mailed questionnaire, along with request for a front incisor for cementum age analysis. We reviewed these data to assess whether management objectives set for HD 652 are being met for buck:doe ratios, density of deer and age structure of harvested bucks. These data were compared to similar mule deer buck data from other general hunting season districts collected at the Havre Check Station. During most years since its inception, management objectives set forth for hunting district 652 are met. The general deer season structure is also producing similar metrics during those years. Wildlife managers considering adapting a limited permit mule deer hunting district should have a clear picture of the public's desired outcome for such a season structure, as they relate to measurable management objectives and the likelihood of meeting public expectation.

** Optimal Use of Wildlife Monitoring Resources

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Resources for monitoring wildlife populations are limited and their availability changes over time. The data collected using these resources is critical for making good conservation and management decisions. Determining the optimal way to allocate monitoring resources for data collection based on the amount of information the data provides for conservation and management is a responsible and efficient use of public resources. We develop a method for determining the most optimal scenarios for data collection which simultaneously minimizes cost and maximizes the precision of the abundance estimate. To accomplish this, we developed a new metric which describes the relationship between data collection cost and estimate precision in a single value, the information gain ratio. We used data collected by the Idaho Department of Fish and Game on the statewide mule deer population of Idaho to

develop our method for determining the optimal allocation of monitoring resources. Using the information gain ratio, we characterize the relationship between cost and precision relative to the specific attributes of each mule deer population management unit. Our method allowed us to generate a set of data collection scenarios that were adapted to the specific characteristics of each unit, changed with the availability of monitoring resources, and are easily comparable via the predicted values of the information gain ratio. The collection scenarios detail the type and amount of each data type to collect for the optimal use of monitoring resources. Our optimization method is adaptable across species, scales, data types, and population models.

** Methods to Estimate Recruitment for Social Species with Limited Data

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Population dynamics for social species depend on hierarchical demography that links local, group-level processes to overall population growth. Many social species cooperatively breed, thus recruitment of offspring is affected by both population- and group-level processes. Traditional methods to estimate recruitment for a species that cooperatively breeds may be insufficient because they do not account for hierarchical demography. Furthermore, for a species that is broad-ranging, rare, or elusive, recruitment data may be too difficult or costly to collect. Our objective was to develop a method to estimate recruitment for a social species that accounts for hierarchical demography and does not rely on recruitment data. We developed an integrated population model (IPM), with both population- and group-level processes, to estimate recruitment in a social species. We were able to estimate recruitment from the IPM without data because changes in abundance are a function of survival and recruitment and we had data for survival. We tested the model using simulated datasets under five scenarios without recruitment data. For all five scenarios we ran models with and without the group-level process to determine if hierarchical demography improved estimation. Simulations demonstrated that the model performed well under most scenarios and provided unbiased estimates of recruitment. We found that explicitly incorporating hierarchical demography was important for estimating recruitment in social species. This model can easily be adjusted to estimate recruitment for any social species. Further, by removing the group-level process this model can be used to estimate recruitment for a non-social species when data are lacking.

** Recruitment of Gray Wolves in Montana

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Recruitment is an important vital rate driving population growth of large mammals. Although large mammals are thought to be regulated by extrinsic factors, cooperative breeding may result in intrinsic factors driving population dynamics, specifically recruitment. The majority of studies evaluating the effects of intrinsic and extrinsic factors on recruitment have not been conducted in a social species that cooperatively breeds, and those that have did not explicitly account for the effects of social structure. We evaluated how intrinsic and extrinsic factors affected variation in recruitment for gray wolves (*Canis lupus*)in Montana using an integrated population model. We hypothesized that variation in recruitment was driven by intrinsic factors such as pack size or population size. Alternatively, we hypothesized

that extrinsic factors drive variation in recruitment and predicted that winter severity, forest cover, road density, or harvest would explain the most variation in recruitment. We found that the main driver of recruitment in wolves was primarily intrinsic factors, specifically pack size. Mean number of pups recruited per pack appeared to vary little over time despite changes in management practices and decreased survival of yearling and adults following harvest implementation. Although recruitment does not appear to compensate for changes in survival, the population has remained relatively stable, suggesting that current harvest rates are sustainable. Recruitment in a large bodied, cooperatively breeding species appears to be driven primarily by intrinsic factors.

** Short-Term Response of Snowshoe Hares to Western Larch Restoration and Seasonal Needle Drop

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Old-growth western larch (*Larix occidentalis*) has been degraded throughout much of its historic range due to extensive timber harvest and fire suppression. We examined the effects of a restoration treatment of western larch on snowshoe hares (*Lepus americanus*), a denizen of the boreal forest serving as a focal animal species to indicate the health of the restored ecosystem. In western Montana, we implemented a restoration treatment using "doughnut thinning" to accelerate development of old-growth attributes in larch stands and simultaneously examined the short-term effects on snowshoe hare density, survival and movement. Although typical forest management activities tend to have adverse effects on hares especially in the short-term, we found that the restoration treatment did not affect hare density or survival in the short-term. In addition, despite significant decreases in cover coinciding with the larch needle drop, we found evidence of year-round immigration into larch stands by hares suggesting larch stands are suitable year-round hare habitat. Taken together, our findings suggest that a larch restoration treatment designed to accelerate the development of old-growth attributes can be implemented so as to have no measurable short-term detrimental effects on hares.

Project WAFLS: Predicting Responses of Short-Eared Owl Population Size, Distribution and Habitat Use in a Changing Climate

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The Short-eared Owl (*Asio flammeus*) is an open-country, ground-nesting species found in marshes, grasslands, shrublands, and tundra across North America and around the world. Evidence suggests that Short-eared Owl populations are experiencing long-term, range-wide, substantial declines in North America, but sufficient monitoring data is lacking to quantify any possible trend. Complicating trend analysis efforts for this species is the expected annual variation in breeding densities, believed to be associated with prey availability. We present evidence from four years of surveys, starting in the Intermountain West, but now encompassing eight western states, confirming annual variation in both density and distribution of Short-eared Owls. Furthermore, we have identified the landscapes features where owls are least susceptible to this variation. Lastly, we present climate-informed

projections for the future viability of the species within the region. The results of our work will directly inform the prioritization of actions to help conserve this often neglected species.

** Complementing Visual Surveys with Wildlife Cameras for Long-Term Wetland Monitoring

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Freshwater wetlands support high biodiversity, but are also subject to increased drying under projected climate patterns. Recently, the use of automated tools for monitoring has become more feasible. Using automated tools to complement traditional visual surveys increases observation time at surveyed sites possibly capturing different species, recording within-season dynamics, and expanding our understanding of wetland biodiversity in a changing environment. In 2017 and 2018, we placed wildlife cameras at 6 seasonal and 6 permanent wetlands in Grand Teton National Park for a week each in June and August; we also completed a single visual survey of amphibian species during each of these time intervals. We compared the difference in the number of species detected by each method over the summer to assess temporal changes in wetlands with varying hydrology and evaluate effectiveness of each method for monitoring. Based on preliminary results, changes in species richness over the summer were more related to wetland hydrology than the survey method. Although we found the two methods captured a similar change in richness over time, the species observed were complementary. Cameras added six species from four additional taxa to the four amphibian species detected during visual surveys. Cameras also captured seasonal water-level patterns, which may be used to ground-truth climate-based drying models. Further analysis, including cost/benefit assessments, will help us understand the utility of cameras for monitoring wetlands. Automated surveillance tools allow us to sample cryptic species over large spatial and temporal scales, providing an important contribution to biodiversity monitoring.

** Time-To-Event Density Estimation of Low Density Species With Remote Cameras

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Abundance estimates can inform management policies and are used to address a variety of wildlife research questions, but reliable estimates of abundance can be difficult and expensive to obtain. For low-density, difficult to detect species, such as cougars (*Puma concolor*), the costs and intensive field effort required to estimate abundance can make working at broad spatial and temporal scales impractical. Remote cameras have proven effective in detecting these species, but the widely applied methods of estimating abundance from remote cameras rely on some portion of the population being marked or uniquely identifiable, limiting their utility to populations with naturally occurring marks and populations that have been collared or tagged. Methods to estimate the abundance of unmarked populations with remote cameras have been proposed, but none have been widely adopted. Working with Idaho Department of Fish and Game, we used the time-to-event model (Moeller et al. 2018) to estimate the density of two cougar populations in Idaho. The time-to-event model uses observed encounter

rates at randomly or systematically placed cameras to estimate the abundance of unmarked populations. Obtaining reasonable abundance estimates for cougars from the time-to-event model shows that remote cameras may lower the costs of abundance monitoring for low density, difficult to detect species and make monitoring programs using remote camera grids applicable to a broader array of species. Future work will compare estimates of cougar abundance from the time-to-event model to estimates obtained from concurrent genetic spatial capture recapture estimates.

** Is Habitat Constraining Bighorn Sheep Distributions and Restoration: A Case Study in the Greater Yellowstone Ecosystem

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We used GPS location data from 65 bighorn sheep (*Ovis canadensis*) in the Madison Mountain Range, located in the northwestern extent of the Greater Yellowstone Ecosystem (GYE), to develop and validate winter and summer resource selection function (RSF) habitat models. Two isolated populations of bighorn sheep occurred within the range, but had demonstrated little expansion into apparent habitat even during years of population growth. We hypothesized that habitat was not the primary factor limiting these populations and that the Madison Range could support a substantially higher abundance and broader distribution of bighorn sheep. We evaluated remotely-sensed landscape covariates expected to influence bighorn sheep resource selection and compared sets of biologically-plausible seasonal models with AICc. We validated our results using temporally- and spatially-independent GPS data and predicted potential habitat throughout the Madison Range. Our results indicate that approximately 82% of winter and 42% of summer habitat was unoccupied. Predicted winter habitat occurred in a non-contiguous distribution primarily along the low-elevation, southwest-facing aspects within the Madison Valley, and predicted summer habitat was concentrated along the high elevation ridgelines associated with steep slopes and reduced canopy cover. By linking our winter RSF to population estimates for the Taylor-Hilgard, we predicted the Madison Range may be capable of supporting 2 to 4 times the number of bighorn sheep currently estimated within the range. Our results support our hypothesis and suggest that a strategy focused on systematically restoring a metapopulation may greatly enhance the potential for bighorn sheep restoration within the Madison Mountain Range.

Invasive and Pest Species Information at the Montana Natural Heritage Program

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The Montana Natural Heritage Program (MTNHP) has centralized information on Montana's native species and habitats since its inception in 1985. In October of 2017, MTNHP started managing information on Aquatic Invasive Species, Noxious Weeds, Forest

Pests, Agricultural Pests, other non-native species, and biocontrol species that have been introduced to control invasives. The MTNHP now manages information on over 500,000 observations of more than 700 invasive and pest species and over 50,000 structured survey locations for invasive and pest species and makes that information available on its websites. This presentation will provide an overview of how biologists and resource managers can access information on surveys, observations, predicted habitat suitability models, and descriptive field guide information for invasive and pest species and native species on the MTNHP's Montana Field Guide, Species Snapshot, and Map Viewer websites.

MONTANA'S SPATIOTEMPORAL MODEL FOR ESTIMATING GAINS AND LOSSES OF GREATER SAGE-GROUSE HABITAT IN A MITIGATION FRAMEWORK: TIME IS OF THE ESSENCE

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Sagebrush-grassland ecosystems support upwards of 300 species, including several charismatic and high-profile big game and upland bird species. For example, habitat conservation for Greater Sage-grouse (*Centrocercus urophasianus*, hereafter GRSG) directly benefits habitat for mule deer and pronghorn. Big game hunting in counties that contain designated GRSG habitat contributes over $113.5 million annually to Montana's economy. Montana has a deep habitat conservation ethic, which is foundational to maintaining Montana's Outdoor Economy. To address the GRSG conservation needs, Montana and a multi-agency, multi-disciplinary, citizen-based stakeholder group developed a Habitat Quantification Tool (HQT model) to quantify GRSG functional habitat gains and/or losses in a mitigation framework. Montana's HQT model is a geospatial raster model. It differs from other state GRSG HQT models because it incorporates time when estimating changes in available functional habitat caused by development or conservation projects. Using biophysical GBSG habitat attributes, the HQT model first establishes a baseline of existing functional habitat. For individual projects, the HQT model assesses functional acres lost or gained due to development or conservation, respectively, by comparing the baseline to post-project condition. Individual HQT model results are aggregated and summed at regional and statewide scales to determine whether Montana is meeting its goal of no net loss of GRSG habitat. The Sage Grouse Habitat Conservation Program (Program) operates and maintains the HQT model. Using an adaptive management framework, the Program will assess the accuracy of HQT model results. The Program will revise the HQT model through time based on new available science.

FREE LUNCH, MAY CONTAIN LEAD: SCAVENGING SHOT SMALL MAMMALS

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Scavengers are subsidized by hunting remains worldwide. While most studies focus on carcasses of large mammals, shot small mammals likely provide a significant food subsidy as

well. Millions of small mammals are shot each year for damage control and recreation, many being left in the field. Despite this high prevalence of carrion, and the potential for scavengers to ingest residual lead from bullet fragments, the fate of these carcasses is largely unknown. We deployed remote cameras to observe which scavengers consumed shot ground squirrels (*Sciuridae* spp.) and black-tailed prairie dogs (*Cynomys ludovicianus*) in 8 locations across Montana, USA. At least 5 species of mammals and 9 species of birds scavenged, including burrowing owls (*Athene cunicularia*). Scavengers fully consumed 67% of carcasses and partially consumed 9%. Carcasses lasted an average of 24.5 hours before the first scavenger arrived. Of carcasses that were scavenged, mammals ate 16% compared to 84% for birds, with corvids and raptors consuming an equal number of carcasses. Common ravens (*Corvus corax*) and black-billed magpies (*Pica hudsonia*) visited the most carcasses and were often the first to arrive. Overall, our results indicate that a diverse scavenger community consumes shot ground squirrels and black-tailed prairie dogs, and consequently, may be exposed to lead from bullet fragments.

Recent Trends in Ungulate Forage Inferred from Remote Sensing Data Sets

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Forage quality, quantity, and phenology play a large role in regulating the habitat use and population dynamics of wildlife populations. Assessing how forage has changed over time can help inform management decisions. Using several freely available remotely sensed and climate-based datasets, we calculated forage-related metrics such as the timing and duration of spring greenup on a pixel-by-pixel basis across Montana. We conducted trend analysis from 2000-2014, identifying areas of consistent change within the state and summarizing trends on a more local scale. We also assessed variability and consistency of estimates within and across datasets. In general, mountainous regions and adjacent lower elevations display somewhat opposing trends, highlighting the importance of understanding the changes for conservation of migratory ungulates which rely on greenscape gradients to optimize nutritional gain.

**Effects of Rangeland Management on the Ecology of Sharp-Tailed Grouse in Mixed-Grass Prairies

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Temperate grasslands, including mixed-grass prairies, suffer from the greatest levels of habitat loss and degradation of any ecosystem, which can have cascading negative effects. Grazing by livestock is the predominant land use across western North America and directly affects the structure, composition, and productivity of native grasslands. While certain grazing regimes can negatively affect wildlife habitat, properly managed grazing can be compatible with wildlife conservation and is preferable to other land uses that destroy or fragment native grasslands. With large home ranges and differing requirements for nesting and winter habitat, sharp-tailed grouse (*Tympanuchus phasianellus*) require large and complex areas of habitat, making them an ideal indicator species for grassland habitats. Poor range management has been implicated in the decline of sharp-tailed grouse throughout North America, but the effects of specific grazing regimes on grouse populations have not been studied. A better understanding of the ecological impacts of grazing is required to develop effective conservation strategies. We monitored 203 radio-collared sharp-tailed grouse in

eastern Montana to assess the effects of grazing management on grouse ecology and evaluate the effectiveness of specific grazing regimes for improving wildlife habitat. In three years of study, we found that choice of grazing system had no effect on space use and important vital rates (nest survival and adult survival) for sharp-tailed grouse. We observed relatively weak effects of stocking rate and stocking density, but overall the performance of the sharp-tailed grouse population at our study area was not affected by grazing system.

Elephant Friendly Tea: An Example of Wildlife Science-Based Commercialization to Save an Endangered Species

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In the U.S. alone, >84 billion servings of tea are consumed per year, totaling $12.5 billion in annual sales. Almost none of these consumers realize that tea is a death crop helping to drive Asian elephants extinct. Building on our applied population ecology research and local outreach in the India-Bhutan region, we have: a) identified tea production practices that drive elephant mortality; b) identified specific actions to reverse those impacts; c) and incentivized those conservation-relevant tea farming practices through a novel "Elephant Friendly Tea" (EFT) Certification. Through our program tea estate owners who implement EFT actions receive a price premium for their tea, which is then sold under a Certified EFT logo; in turn, global tea consumers have a direct opportunity to support science-based elephant conservation with every cup of tea. We expect EFT to be game-changing for arresting the decline of Asian elephants because – unlike traditional conservation approaches – it both implements incentive-based conservation actions on and around the private agricultural lands where most elephants are killed, and it will create profits that we will fully invest into research and conservation actions across the elephant's range. Critical partners to development of EFT include local villagers and tea professionals, the non-profit certification group Wildlife Friendly Enterprise Network, and University of Montana (who will manage the "EFT Elephant Research and Conservation Fund" created by EFT sales). We believe this model has great potential to address seemingly intractable conservation problems in Montana by developing meaningful win-win wildlife friendly enterprises.

A Winter Energetics Model for Bobcats in a Deep Snow Environment

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Understanding basic energetic requirements of wildlife species is critical to evaluate how individuals persist in their current environments as well as to forecast responses to changed climates or habitats. Indeed, northern range limits are often thought to reflect harsh abiotic conditions that exceed the capacity of individuals to stay in energetic balance. Bobcats (*Lynx rufus*) occur across much of North America; at northern latitudes, they face winter challenges such as deep snows, cold temperatures, and possible food scarcity. We developed an energetics model from field data on movements, body mass, and observed diet of bobcats in mountains of northwest Montana, then evaluated overwinter prey requirements that would enable bobcats to stay in energy balance in this difficult environment. Our model indicated average daily energy expenditures were ~1.41 × basal metabolic rate. For 90 days from December-February, a 10.5 kg bobcat consuming prey items in proportion with the observed diet for bobcats in this area would need about 2.1 kg of deer (Odocoileus spp.), 7 snowshoe hares (*Lepus americanus*), 155 red squirrels (*Tamiasciurus hudsonicus*), 9 woodrats

(*Neotoma cinerea*), and 250 small rodents (*Cricetidae*). Bobcats have considerable flexibility in diet, movements, and both timing and duration of daily activity to adjust their energetic expenditures in winter.

Regional Differences in Winter Diets of Bobcats in Their Northern Range

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When generalist predators have wide geographic ranges, diets may differ dramatically, largely as a result of differing prey communities. Bobcats (*Lynx rufus*) are widely distributed across southern North America, with their northern range edge occurring in southern Canada and in the northern US states. Within this northern range, bobcats are exposed to cold and snowy winters and a limited number of prey species, conditions that are atypical for most of the range of bobcats. We examined winter diets of bobcats in high elevation and very snowy forests in northwest Montana to determine how these generalist predators managed in these harsh conditions in comparison to elsewhere in the northern range. Bobcats consumed 5 major prey types: red squirrels (*Tamiasciurus hudsonicus*) and Cricetid rodents comprised >78% of the dietary biomass, whereas the larger snowshoe hares (*Lepus americanus*), deer (*Odocoileus* spp.), and grouse were consumed much less often. The standardized niche breadth of bobcat diets was 0.29; bobcats from across the northern range also routinely ate multiple prey species, although eastern bobcats appear to consume more lagomorphs than do western bobcats. These results indicate that bobcats remain generalists in difficult winter conditions while preying primarily on small-bodied prey, although bobcats have highly variable diets across their northern range.

An Improved Understanding of Population Dynamics Using Count Data: Insights From Elk in Western Montana

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Understanding the dynamics of ungulate populations is a crucial goal for managers given their ecological and economic importance. In particular, the ability to evaluate the evidence for potential drivers of variation in population trajectories is important for informed management. However, the routine use of age ratio data (e.g., juveniles:100 adult females) to evaluate variation in population dynamics is hindered by a lack of statistical power and difficult interpretation. Here, we show that the use of a population model fueled by count, classification and harvest data can dramatically improve the understanding of population dynamics compared to a model using age ratio data by: 1) increasing the power to assess potential sources of variation in key vital rates, and 2) providing easily interpretable vital rates (e.g., per capita recruitment rates and population growth rates) that are useful to managers. Using a time series of spring count data (2004 to 2016) and fall harvest data from hunting districts in western Montana, we constructed a population model to assess the effects of a series of environmental covariates and indices of predator abundance on the per capita recruitment rates of elk (*Cervus elaphus canadensis*) calves. Results from this modeling approach suggest per capita recruitment rates decline in association with wet springs, dry summers and severe winters, and in interactions between predator communities and the environment. In contrast, the analysis of age ratio data failed to detect these relationships. We

recommend using count data and a population modeling approach rather than interpreting estimated age ratio data as a substantial improvement in understanding population dynamics.

Estimating Carnivore Density Using Spatial Capture-Recapture Models: Sampling Designs and Potential Pitfalls

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Spatial capture-recapture (SCR) models have improved the ability to estimate densities of rare and elusive animals. However, SCR models have seldom been validated even as model formulations diversify to incorporate new sampling methods and/or additional sources of information. The relationship between encounter probabilities, sources of additional information, and the reliability of density estimates, is poorly understood but crucial to assessing reliability of SCR-based estimates. We used a simulation-based approach that incorporated prior empirical work on mountain lions (*Puma concolor*) in western Montana to assess the accuracy and precision of density estimates from SCR models using direct search effort. Our simulations focused on understanding the consequences of: 1) variable probabilities of encounter generated from different levels of search effort, and 2) including additional spatial information from collars. Overall, we found that although low search effort resulted in sparse datasets and highly biased and imprecise estimates of density (relative bias, RB = 0.71, coefficient of variation, CV = 1.16), a combination of increased effort and/or additional information generated unbiased and precise density estimates (e.g., moderate effort and 4 collars, RB = -0.004, CV = 0.19). This work suggests that reliable density estimates can be generated by multiple sampling designs such that additional spatial information from collars can be used to supplement direct search effort when resources are limited, or by increasing search effort when collaring is impractical. Using the open-source code for our simulation-based approach, we further offer recommendations on sampling designs for SCR-based density estimation.

Adventure Scientists: Igniting the Outdoor Community to Collect Game-Changing Data

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Outdoor adventurers have the skills and inspiration to spend hours, days, and weeks every year in regions that are inaccessible to the rest of the population. While carrying out their already-planned excursions or planning new ones expressly for this purpose, they can collect data and samples of critical importance to scientists and conservationists working to address today's challenges to human health and the environment. Adventure Scientists has gained extensive experience and developed repeatable protocols for designing projects and training these highly-skilled volunteers to collect research-grade data. These data are being used by governments, businesses, and others to shape policies and practices that benefit the environment. Scientists get otherwise unobtainable data while adventurers enrich their experience of the outdoors and increase their knowledge of the challenges facing the natural world. Together they have a greater impact than either could alone. This spring, serving a partnership with the Montana Department of Transportation, Adventure Scientists will begin recruiting volunteer teams of cyclists throughout the state to create a comprehensive year-round portrait of wildlife-vehicle collisions on more than 8,400 miles of state and federal

highways. Armed with the results, decision makers then can identify key migration corridors, uncover road crossing "hot spots," and design improved road projects such as overpasses and culverts to prevent collisions and allow humans and wildlife to move freely and safely across a shared environment.

** Estimating Forage Quality and Abundance to Better Understand Mule Deer Resource Selection

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To identify the environmental factors limiting growth of herbivore populations, researchers need to understand how benefits (like forage quality and abundance) and costs (like risk of predation) vary across a population's range, and how individuals select habitat with respect to those resources. For mule deer (*Odocoileus hemionus*) living in northwest Montana, predicting forage quality and abundance over large extents is difficult, since much of their habitat use occurs within dense conifer forest where remote-sensed metrics (like NDVI) are of limited use. Therefore, we are developing a landscape forage model using field-collected vegetation data to estimate how digestible energy per area varies across 3 mule deer population ranges. Preliminary evidence has shown that forage resource availability varies with landcover-type. With this forage model, and along with previously developed predator resource selection functions (RSFs), we will develop an RSF to assess how mule deer make tradeoffs between nutrition and predation risk. This will allow us to identify the mechanisms driving mule deer resource selection, and will guide more effective management of mule deer populations and habitat in western Montana.

Greater Sage-Grouse Response to Bentonite Mining

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The greater sage-grouse (*Centrocercus urophasianus*) has undergone range contraction and population decline because of anthropogenic land surface disturbances; yet, there is little information on the effects of mining on sage-grouse populations. In the Bighorn Basin of Montana and Wyoming, bentonite mining is a growing source of surface disturbance that contributes to loss of sagebrush habitat. We evaluated the response of sage-grouse to active and reclaimed bentonite mining, relative to nesting, brood-rearing, adult breeding, and adult winter habitat, through resource selection and habitat-specific mortality risk analyses, based on female sage-grouse monitored with telemetry from 2011-2015. A greater proportion of our monitored sample was exposed to mining disturbance during winter (65%) than during other seasons (range = 25%-34%). We observed avoidance of all mining disturbance for selection of nesting habitat, adult breeding habitat, and adult winter habitat. Evidence was inconclusive for avoidance of mining for brood-rearing habitat. We also observed increased adult breeding season mortality risk associated with active mining disturbance but observed no effect on nest success. Evidence was inconclusive for increased mortality risk associated with broods and adults during winter. Stakeholders in the Bighorn Basin should be flexible and proactive to minimize the negative effects of bentonite mining on sage-grouse habitat use and demographic rates. Stakeholders should prioritize the conservation of winter habitats because of the influence on a greater proportion of the population and they should strive to perfect mining reclamation to return disturbed sites back to pre-disturbance conditions to minimize long-term effects.

Integrated Carnivore-Ungulate Management: A Case Study in West-Central Montana

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In response to poor recruitment and declining ungulate population trends in west-central Montana, wildlife managers implemented an integrated carnivore-ungulate management program designed to reduce carnivore densities via harvest prescriptions in efforts to increase elk (*Cervus elaphus canadensis*) recruitment and abundance. However, the ability of wildlife managers to use carnivore harvest management regulations as a tool to reduce carnivore population densities and increase ungulate recruitment is unknown. The management objective in this case was a moderate reduction in carnivore densities that sustained carnivore populations and associated recreational opportunities, while also reducing predation pressure on ungulate populations. We assessed the efficacy of this integrated carnivore-ungulate management program by evaluating: 1) the effects of a harvest management prescription on mountain lion (*Puma concolor*) population density using a before-after-control-treatment study design, and 2) patterns in elk juvenile recruitment before and after implementation of the mountain lion harvest treatment. We found that 4-years after the management program was implemented, mountain lion population abundance declined by 26% (90% CI = [0.60, -0.05]) within the harvest treatment area and remained stable within the control area. The per-capita recruitment rate of elk was low and stable in the treatment area prior to the mountain lion harvest prescription (e.g., mean = 0.18, [0.14, 0.22]), increased substantially in the year following the implementation of the harvest prescription (mean = 0.32, [0.24, 0.41]) prior to declining to 0.23 ([0.16, 0.29]) at present, which contrasted with a moderate increase in per capita recruitment rates in the control area. Together these results suggest that the mountain lion harvest treatment moderately reduced mountain lion abundances within the treatment area, as intended, although the effect on elk population dynamics was short-lived. Broadly, this integrated management program achieved carnivore and ungulate population objectives. We recommend wildlife managers applying integrated carnivore-ungulate management programs develop carnivore and ungulate monitoring programs that assess the efficacy of management programs and provide information regarding future management prescriptions designed to achieve carnivore and ungulate population objectives.

** Assessing Habitat Quality for Four Grassland Songbird Species of Concern in Northern Mixed-Grass Prairie

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During the past 40 years grassland bird populations have declined faster than any other avian guild in North America. In northern Montana, four species are experiencing severe population declines, Baird's sparrow (*Centronyx bairdii*), chestnut-collared longspur

(*Calcarius ornatus*), McCown's longspur (*Rynchophanes mccownii*), and Sprague's pipit (*Anthus spragueii*). In 2017 and 2018, we evaluated abundance and nest density of these species in relation to local vegetative conditions with the goal of identifying important breeding season habitat conditions to inform management. We conducted fixed-radius point-counts at 100 sites to estimate local abundance, rope drag surveys to estimate nest density, and vegetation surveys to estimate vegetation structure and composition across grassland habitats in Phillips County, MT. Point-counts and rope drag surveys were carried out with replicated visits to allow estimation of species-specific detection probabilities. Habitat conditions were measured at the plot level (9 ha) to provide information at scales relevant for land managers. The abundance of Baird's sparrows was positively associated with residual grass cover and litter cover. Chestnut-collared longspur abundance was negatively associated with residual grass and shrub cover and had a quadratic relationship with biomass. Plot-level abundance of McCown's longspurs was negatively associated with both shrub cover and biomass. Sprague's pipit abundance exhibited a quadratic relationship with biomass. Limited sample size only allowed inference of nest density for chestnut-collared longspurs which was negatively associated with plot scale biomass.

** Application of a Novel Nest Density Estimator: An Example Using Sagebrush-Steppe Songbirds

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Studies show nest density is an important demographic rate for shorter-lived species, such as sagebrush-steppe songbirds. A recent analytical approach, temporal nest density estimator (TNDE), was developed to estimate the nest density of waterfowl species using data routinely collected to assess nest success while accounting for detection and availability of nests. To understand the general applicability of TNDE to species in other avian orders, we evaluated the performance of TNDE on a songbird species, Brewer's sparrow (*Spizella breweri*). We assessed the TNDE by comparing estimates of nest detection rate and nest density from TNDE to distance sampling methods for 43 Brewer's sparrow nests monitored in 2015. The TNDE method produced similar but more precise nest detection and density estimates than the distance sampling method. Now that TNDE has been validated, we plan on converting it to a Bayesian framework and using it in conjunction with fine resolution remote sensing data to determine nest-site selection for three sagebrush-steppe songbirds: Brewer's sparrows, McCown's longspurs (*Rhynchophanes mccownii*) and vesper sparrows (*Pooecetes gramineus*). These three species are representative of sagebrush specialists, grassland specialists, and generalists that use both, respectively. This study will allow us to explore the capability of TNDE and remote sensing data to assist in habitat selection studies. These methods may allow more accurate nest density estimates on broader scales with less effort, which will aid in identifying priority areas for conservation and management.

Young and Restless: A Guide to Getting Your Own Colony as a Juvenile Beaver

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Interest in using beavers (*Castor* spp.) as an efficient riparian restoration tool has increased dramatically in recent decades. The ultimate goal of most beaver-mediated habitat restoration projects is to establish a self-sustaining population of beavers that can occupy

new habitats over time. Therefore, dispersal is a key process to understand when evaluating the potential for project locations to be settled by beavers. We radio-marked juvenile beavers in the Gallatin and Madison River drainages in southwest Montana to investigate dispersal characteristics, survival, and settlement site selection. Our goal was to improve identification of restoration sites with the highest probability of colonization by beavers. The annual probability of dispersal in our study area was low ($0.26 \pm 0.24\text{-}0.29$), and distances and timing of dispersal were highly variable. The probability of a beaver dispersing decreased as local colony density increased, suggesting delayed dispersal. Few radio-marked beavers started new colonies in previously unoccupied habitat during our study. Instead, most dispersers settled in stream segments already modified by previous beaver activity. The low number of new settlement sites and evidence of delayed dispersal suggests most of the suitable habitat in our study area was occupied. We recommend land and wildlife managers assess the density and distribution of beaver colonies around proposed restoration sites prior to project design to evaluate the potential for dispersal into the project area. Beaver dispersal is an important factor in the success of riparian restoration projects, and our work highlights aspects of dispersal that may influence beaver settlement in this context.

** Population Demographics, Breeding Ecology and Responses to Grazing of Montana Sagebrush Steppe Songbirds

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Sagebrush steppe is one of the most threatened ecosystems in North America. Avian adult density estimates are often used to assess conservation actions given the relative ease in collecting data to inform these estimates. However, information on how conservation actions influence life histories such as nest density and nest success are lacking, despite the fact that life histories inform abundance. We investigated songbird adult densities, nest densities, and nest success over multiple breeding seasons in central Montana. Our goal is to understand the relationships among adult abundance, nest density, and nest success, as well as how land management practices, in the form of grazing, influence those patterns. Two grazing systems were compared in our study: a system using a combination of rest and deferment (hereafter rest-rotation) and traditional grazing. For the purposes of our study, we define rest-rotation grazing as changing the timing of grazing in pastures each year, with some pastures alternately rested every few years. Traditional grazing is defined as grazing a pasture at the same annual season each year or all season. Recently, rest-rotation systems have been used as a conservation management tool by the Natural Resource Conservation Service-Sage Grouse Initiative. Their goal is to encourage private landowners to graze their livestock more sustainably to maintain or improve rangeland productivity, while also benefiting habitat for greater sage-grouse (*Centrocercus urophasianus*). We explore the effects of rest-rotation compared to traditional grazing on songbird population demographics during the breeding season.

Demographic Fragmentation of a Protected Wolverine Population Bisected by a Major Transportation Corridor

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Roads fragment terrestrial ecosystems around the globe, but the effects of this fragmentation on biodiversity remain poorly understood. Wolverines (*Gulo gulo*) are snow-dependent carnivores that occur at low densities and they exhibit low genetic diversity at the southern extent of their range where they are snow-limited, rare and fragmented by human development. Therefore, understanding the effect of roads on population connectivity is crucial to effective wolverine management in a changing climate. We examined whether the Trans-Canada Highway, Canada's largest east-west transportation corridor, affects wolverine movement and gene flow in the Canadian Rocky Mountains. We used noninvasive genetic sampling methods to collect DNA samples (i.e. hair, scat) from Banff, Kootenay, and Yoho National Parks and provincial lands in British Columbia and then used population and individual-based genetic analyses to quantify genetic structure of the wolverine population across the highway in the national parks complex. We collected 2586 DNA samples between 2010 and 2013 from which we identified 49 unique individuals (29 males, 20 females). We detected equal numbers of males and females that crossed the highway (4 males, 4 females); however, dispersal and gene flow were affected differently in the two sexes by the transportation corridor. We detected weak population structure in males and relatively strong genetic differentiation in females spanning the highway. Our results demonstrate that sex-biased dispersal across a major highway can lead to genetic isolation and demographic fragmentation in a protected carnivore population, highlighting the urgent need to maintain connectivity for wildlife species over an expanding road network.

How Far Have They Slithered? Genetic Variation Among Garter Snakes in Western Montana

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Snake gene flow across wide geographic regions is poorly understood. Limited sampling opportunities and the challenges posed by microsatellite analysis often prevent researchers from assessing the impact of topographic barriers and the influence of human settlement on the genetics of snake populations. Last year we demonstrated that SNPs significantly improve our understanding of Isolation by Distance for both species of garter snake (*Thamnophis* spp) in Western Montana. Since then we have initially analyzed larger sample sets of the Wandering Garter Snake (*Thamnophis elegans vagrans*) (*N*=192) and Common Garter (*Thamnophis sirtalis*) (*N*=160). These samples were obtained at 77 sites during the 2014-2017 seasons. With these samples we hope to initially understand gene flow on both sides of the Continental Divide. Moreover, herpetologists traditionally assumed that the Continental Divide instigated a subspecies barrier for Common Garters. We hope to determine the efficacy of this assumption in the initial analysis of our data.

** Improving Estimates of Wolf Abundance in Montana

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Estimating wolf (*Canis lupus*) abundance is a key component of wolf management in Montana. Montana Fish, Wildlife and Parks (MFWP) has successfully implemented a Patch Occupancy Model (POM) to estimate area occupied and resulting wolf abundance for the past decade. Estimates of abundance, however, depend on assumptions that territory size is fixed, consistent statewide, and includes minimal overlap; additionally, these assumptions are based on data collected pre-harvest. In reality, territories vary spatiotemporally, and this variability may be even greater under harvest. This variability in turn could affect precision and accuracy of abundance estimates. Furthermore, MFWP requires tools to both keep POM calibrated into the future, and to predict how territorial behavior might change in response to changing environmental conditions or management actions. Critically, these tools must be useful with limited data because intensive monitoring efforts are no longer sustainable. We developed theoretical models of territorial behavior towards accomplishing this goal. Results demonstrate, for example, that territories are expected to be on average smaller where prey are more clumped and abundant, and larger where human influence is greater. Predictions from our models are supported empirically. This provides evidence for how territories will vary based on ungulate populations and human use, which in turn can help guide understanding of the effects of management decisions, e.g., degree of harvest pressure. We are currently parameterizing the models with field data and developing empirical models to contrast with the theoretical models. Altogether, this work will help keep POM calibrated into the future with limited data.

Montana's Greater Sage-Grouse Conservation Strategy All Hands Balancing Conservation and Development Across All Lands

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The Greater Sage-grouse (*Centrocercus urophasianus* or GRSG) inhabits sagebrush grassland habitats exclusively and shares habitat with up to 300 other species. Montana is a key stronghold among the western states. Montana's emphasis on habitat conservation on public and private lands is foundational to supporting and growing Montana outdoor economy. The Montana Sage Grouse Conservation Strategy, implemented by the Montana Sage Grouse Habitat Conservation Program (Program), blends voluntary incentives with regulatory oversight through the permitting process and a new requirement to mitigate for impacts to designated habitat. The Program implements the Greater Sage-Grouse Stewardship Act and Executive Order 12-2015. Key pillars include: 1. a consultation process to guide development; 2. grants to maintain, enhance, restore, and expand habitat; and 3. mitigation to offset impacts of development. Taking an "all lands, all hands" approach, the Program works collaboratively with private landowners, state agencies, and federal land management agencies. Since 2015, the Program has reviewed nearly 1500 proposed development projects

in designated GRSG habitat on public and private lands. Approximately 43,148 acres have been placed in perpetual conservation easements through Sage Grouse Stewardship Fund grants. With the goal of no net loss-net gain preferred, the mitigation system incorporates market-based incentives and a Habitat Quantification Tool to quantify gains and losses of functional habitat due to conservation or development projects, respectively. Successful conservation depends on proactive stewardship of the remaining intact sagebrush landscape. Montana is well-positioned for the range-wide conservation assessment set for 2020.

IDENTIFICATION OF ALTERNATE BAT HIBERNACULA OUTSIDE OF CAVES AND MINES IN EASTERN MONTANA

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Pseudogymnoascus destructans, the fungus responsible for White-nose Syndrome (WNS) and millions of bat deaths in North America, was recently detected in Wyoming and South Dakota near the eastern Montana border. Given the nature of the fungus to thrive in cold and humid environments, bats are most vulnerable to infection in their winter hibernacula. Outside of caves and mines, we have limited knowledge of bat hibernacula in Montana. However, from long-term statewide acoustic monitoring stations we know that some bats are wintering in locations where cave and mine features are limited. In anticipation of WNS and its potential impacts, we were interested in identifying hibernacula and associated characteristics. In October 2018, we attempted to capture and transmitter bats to identify alternate roosts in eastern Montana. We targeted 4 sites, captured 12 bats, placed transmitters on 10, and successfully identified 9 different roost sites. We will characterize microsite (i.e. temperature, humidity, etc.) and macrosite (i.e. roost structure, nearby water, etc.) features of each roost and assess bat use through winter by placing acoustic detectors near roost sites. Although our results are limited, this is the first documentation of alternate winter hibernacula in eastern Montana. Information on specific hibernacula and associated habitats will help inform bat conservation activities. Additionally, findings will assist in site selection for future WNS surveillance efforts and continued efforts will facilitate future exploration of how hibernacula type influences WNS spread and impacts.

ASSESSING OCCUPANCY FOR MONTANA SPECIES OF GREATEST INVENTORY NEED

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Montana non-game species classified as Species of Greatest Inventory Need (SGIN) are not sufficiently monitored by standard ecological monitoring programs, typically because they

are locally rare or cryptic, thus evading detection. Yet, these species are often those for which understanding conservation status is most important. We are developing a tool to characterize the survey effort needed to attain estimates of occupied suitable habitat, while accounting for species characteristics and logistical constraints, and incorporating flexible tools recently presented in the scientific literature. Concurrent with tool development, we have used this approach to examine survey designs for assessing suitable habitat occupancy by Great Gray Owls (*Strix nebulosa*). We identified a survey strategy with sufficient power to estimate occupancy of this inconspicuous species using a combination of automated recording units and ground call-playback surveys that can be employed in the coming years. Understanding effort required to obtain occupancy estimates with acceptable bias and precision can support survey design for species conservation work as well as informing whether existing monitoring datasets have sufficient power to address information needs. We aim for this tool to inform ongoing survey efforts for Species of Greatest Inventory Need and to facilitate survey planning for species without survey protocols underway.

Love Triangles Caught on Camera! Exposing the Intimate Relationships Between Hunters, Gut Piles and Scavengers

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As big game hunting season draws to a close, hunters celebrate filling their freezers and the satisfaction of time spent outside. While they enjoy venison backstraps fresh off the grill, scavengers are feasting on a gut pile left in the woods. The U.S. Fish and Wildlife Service estimates that hunters leave 1.5 billion pounds of carrion annually in the field. However, little is known about how scavengers consume this carrion and what role this food source may play in scavenger populations. We asked hunters in the Bitterroot and Missoula Valleys to set up game cameras on gut piles after successful harvests of deer, elk, and moose. Scavengers ranging from Golden Eagles (*Aquila chrysaetos*) to gray wolves (*Canis lupus*) visited the gut piles. Scavengers continued to visit gut pile locations long after obvious food sources were consumed, suggesting the "life" of a harvest goes beyond a matter of weeks. We'll discuss how this project adds to other citizen-science efforts to understand scavenging species in western Montana, and to our ability as scientists to engage with hunters on tough topics like ammunition choices, carnivore management and public land issues. The high level of interest in this project suggests that hunters are able and willing to contribute to scientific research. We expect to continue and expand this project during the 2019 hunting season.

** Effects of Contamination from Oil Extraction on Amphibian Abundance, Survival and Size

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Saline wastewaters (brine) that contain sodium-chloride salts (NaCl) and heavy metals are a common byproduct of oil extraction. Wetlands and streams are commonly contaminated by brines, but there is sparse information on its effects on freshwater vertebrates. Amphibians are especially sensitive to increased salinity because of their porous skin and primarily-aquatic lifecycle. Therefore, we investigated the influence of brines and pure NaCl on three widespread amphibian species; barred tiger salamanders (*Ambystoma mavortium)*, leopard frogs (*Rana pipiens)* and boreal chorus frogs (*Pseudacris maculata*) in the Prairie Pothole Region of Montana and North Dakota. We determined abundance of larvae in 33 wetlands

that spanned a gradient of contamination. We also used lethal concentration 50 experiments to determine the influences of brine and pure NaCl contamination on survival and size. Abundance and survival of chorus frog larvae declined most rapidly in response to increased concentration of brines and NaCl, followed by leopard frogs and tiger salamander larvae. However, survival of larvae was lower when exposed to brine compared to pure NaCl. For larvae that survived experimental exposures, mass and length were lower for larvae exposed to higher concentrations of brine and NaCl. Our results suggest that brine contamination has reduced survival and abundance of larvae, brine is more toxic to larvae than pure NaCl, and that sublethal concentrations of brine and NaCl negatively influence growth of larvae. The persistence and negative influence of salts in freshwater ecosystems underscores the critical need for tools to restore landscapes affected by brine and NaCl contamination.

** Influence of Pre-Breeding Body Condition on Reproductive Metrics of Greater Sage-Grouse

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Many species are subject to carry-over effects, where habitat quality experienced long before breeding may influence subsequent performance and overall fitness of an individual. Pre-breeding body condition has been shown to affect reproductive metrics including: breeding propensity, timing of nest initiation, clutch size, and offspring quality. Although the effects of pre-breeding body condition on reproduction have been well-studied in waterfowl, we know little about carry-over effects for gallinaceous birds. During on-going research in the Centennial Valley in southwestern Montana, we tracked 237 female Sage-grouse (*Centrocercus urophasianus*) for 5 years and monitored nesting activity and broods until 30 days of age. Based on preliminary regression analyses, we did not detect an influence of pre-breeding body condition on breeding propensity, given that most individuals nested (91%, 187/206) if they survived long enough to breed. We did not observe differences in timing of nest initiation based on variation in pre-breeding body condition, but this timing differed substantially among years. We found pre-breeding body condition positively influenced clutch size and offspring weight, however there was substantial unexplained variation. Although we did not find pre-breeding body condition strongly influenced reproduction in this population, other metrics, such as habitat characteristics and the previous season's weather events, may provide insights about the role of carry-over effects in sage-grouse. We are currently investigating other drivers to understand the importance of winter habitat quality on reproduction and subsequently help guide management decisions.

Estimating Occupancy and Activity of Montana Bat Species Prior to the Arrival of White-Nose Syndrome

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The spread of white-nose syndrome (WNS) across the eastern United States has raised conservation concerns for bats and provided motivation for monitoring efforts. Currently, WNS has not been detected in Montana and understanding the impacts of WNS on bats in

western states requires accurate baseline population assessments and continued monitoring after the disease arrives. Within an occupancy model framework, we analyzed mist netting and acoustic records for eight bat species in Montana to estimate baseline distributions prior to the arrival of WNS. We created distribution maps from this model that explain the heterogeneity in occupancy for each species using covariates for forest cover, elevation, ruggedness, and average degree days. We also developed a model for overall bat activity using additional acoustic data. Even after accounting for nightly weather conditions, patterns in overall activity were highly variable across years and detector locations. These analyses can help inform future surveillance efforts for early detection of WNS and future bat monitoring efforts in Montana. We developed tools to help visualize estimates from both analyses when selecting locations for WNS surveillance so managers can focus efforts on locations with high estimated probabilities of occupancy for susceptible species and consistent bat activity. Based on the model estimates, we also provided recommendations for future acoustic monitoring aimed at identifying how bat species are impacted by WNS when in arrives in Montana.

** PRECIPITATION AND REPRODUCTIVE EFFORT ALTER SURVIVAL OF TURKEY HENS IN THE NORTHERN BLACK HILLS, SD

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Tradeoffs between survival and reproduction are predicted by life history theory. Understanding how reproductive tradeoffs act in concert with abiotic elements to impact survival is crucial for effective management and conservation of wildlife populations, particularly for at-risk or harvested species. During 2016 - 2018, we radio-tracked and collected survival data on 140 Merriam's wild turkey (M. g. *merriami*) hens in the northern Black Hills, South Dakota. We evaluated associations between hen survival and nest incubation, brood rearing, and precipitation. Consistent with life-history theory and the moisture-facilitated nest-depredation hypothesis, increased time spent incubating and daily precipitation amount were associated with reduced hen survival, but the magnitude of the precipitation association depended on incubation status. Seasonal survival was lowest during spring and winter, highest during summer, and intermediate during fall. A hen that did not incubate a nest was predicted to have a higher rate of annual survival (0.54, 95% CI = 0.46 - 0.61) than a hen that incubated a single nest (0.44, 95% CI = 0.33- 0.55); this prediction is based on precipitation data collected in 2017 and assumes the hen began incubation on the median date. We estimated that annual survival for both nesting and non-nesting hens was lower in the northern Black Hills compared to previous estimates from the southern Black Hills. Management options to improve hen survival are limited, but we recommend actions that would reduce hunter-induced hen mortality of this important game species in the northern Black Hills.

THE U.S. FISH AND WILDLIFE PROPOSAL TO DELIST THE CANADA LYNX

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In 2000, the U.S. Fish and Wildlife Service listed the contiguous U.S. distinct population segment (DPS) of the Canada lynx (*Lynx canadensis*) as threatened under the Endangered Species Act (ESA) because of the inadequacy, at that time, of regulatory mechanisms in federal management plans. Since then, federal agencies have formally amended most

management plans in the DPS's range to adopt science based conservation measures for lynx or continued to implement conservation agreements in collaboration with the Service. State and Tribal agencies have also worked to conserve lynx populations and important habitats. Research and monitoring conducted since the DPS was listed suggest that resident lynx are naturally rarer and populations smaller in much of the west than previously thought, but they are more abundant in Minnesota and Maine. The 1999-2006 release of lynx into southwestern Colorado has established a resident population there. The Service recently completed a peer reviewed species status assessment (SSA) for the DPS that evaluated the available scientific information and incorporated the opinions of recognized lynx experts to assess the status and viability of DPS populations. Although the threat for which the DPS was listed has been addressed by improved regulatory mechanisms, the SSA recognized that continued climate warming is likely to reduce lynx populations and distribution in the Lower 48 over the long term. However, the Service concluded that the DPS is not at risk of extirpation in the reasonably foreseeable future and is therefore proposing to delist the DPS.

POSTER ABSTRACTS

Alphabetical By Presenter's Name
* Denotes Presenter
** Indicates Student Presentation

** LURE EFFECTIVENESS IN DETECTING SMALL MAMMALS, SPECIFICALLY THE NORTHERN BOG LEMMING, ON TRAIL CAMERAS (POSTER)

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Mark Hebblewhite, Wildlife Biology Program, University of Montana , Missoula
Mike Mitchell, Montana Cooperative Wildlife Research Unit, US Geological Survey, Missoula
Kristi DuBois, Non-Game Biologist (retired), Montana Fish, Wildlife & Parks, Missoula

Fens and bogs are unique wetlands that support a diversity of small mammals and other rare species. One such species is the northern bog lemming (*Synaptomys borealis*). This species is being considered for listing under the Endangered Species Act so determining their presence is helpful for management. Northern bog lemmings are difficult to trap and when they are caught, experience high mortality rates. Since they are so hard to capture and study, it is difficult to determine presence/absence of this species. This study used a non-invasive trail camera method to detect northern bog lemmings in Finley Fen and Meadow Creek in western Montana. We tested six different types of lure/scent to see if any had better detection rates than muskrat lure which is often used in small mammal studies but is not always readily available and is expensive. The six lures tested were; muskrat lure as the control, almond extract, vanilla extract, strawberry extract, clove oil, and lemongrass oil. Cameras were placed in both fen sites for approximately three weeks and were checked every week. Under each remote camera we placed 6 by 6-inch pieces of plywood with a metric ruler on the sides of the board for size reference. During each check boards received new scent and cameras new memory cards. During the last check before the cameras were removed, the boards were switched between camera sets so that each camera had approximately one week with a different lure under it. Preliminary results suggest that the muskrat lure still produced more photos of small mammals over the time period the cameras were deployed, followed closely in Finley fen by almond extract and strawberry extract. Northern bog lemmings were confirmed in 7 different pictures in Finley Fen, two of which were on almond extract boards.

Bog lemmings weren't detected in Meadow creek, although it was a known bog lemming site as one was trapped there in 1992. The small detection rate for northern bog lemmings indicated that a larger sample size may be needed, or other lure types tested to definitively detect northern bog lemmings in a survey.

** Daily and Seasonal Space Use and Habitat Selection of Greater Sage-Grouse in Carbon County, Montana (Poster)

Erin Birtwistle*, Ecosystem Science and Management, University of Wyoming, Laramie, WY
Aaron Pratt, Ecosystem Science and Management, University of Wyoming, Laramie, WY
Jeffrey Beck, Ecosystem Science and Management, University of Wyoming, Laramie, WY

Greater sage-grouse (*Centrocercus urophasianus*; hereafter 'sage-grouse') are the focus of much research and conservation efforts owing to their obligate relationship with sagebrush (*Artemisia* spp.) and dramatic population declines over the last 50 years. Research suggests female survival, followed by chick, then nest survival, have the greatest impact on population growth, and to sustain sage-grouse populations, focus should be on increasing these vital rates. In addition, recent research has shown habitat partitioning occurs between broodless (i.e., females without a brood) and brood-rearing females such that broodless females have lower mortality risk than females with chicks. Moreover, greater nest attentiveness and incubation constancy can increase nest success. Our study was initiated in spring 2018 in Carbon County, Montana to identify seasonal habitat use and address the most important parameters influencing population growth. Our first objective, to aid the Bureau of Land Management in maintaining sustainable sage-grouse populations on public lands, will focus on natural and anthropogenic landscape features influencing habitat selection during nesting, brood-rearing, summer, and winter life stages. Our second objective is to compare landscape and microhabitat characteristics between brood-rearing and broodless females. Our third objective is to examine nest attentiveness and microhabitat selected during incubation recesses. We captured 39 adult and yearling females in spring 2018 at 6 leks, attached solar-powered, rump-mounted transmitters and monitored 40 nests and 17 broods. In summer 2018, we measured vegetation characteristics (e.g., shrub, grass, and forb cover) at 133 grouse-use and 108 random locations for nests, incubation recesses, and brood and broodless females. Understanding female sage-grouse habitat use during all life stages will aid in directing management on public lands to conserve populations and increase population growth.

** Swainson's Thrush Stopover Habitat In The Bitterroot Valley (Poster)

Shannon Byrne*, Scientific Research Class, Florence-Carlton High School, Florence, MT
Vanessa Haflich, Teacher, Florence-Carlton High School, Florence, MT
Kate Stone, Avian Science, MPG Ranch, Florence, MT

The migratory songbird Swainson's thrush (*Catharus ustulatus*) is a common breeding songbird across North America. We think their populations are in decline due to habitat loss. During fall migration, they travel in large numbers through the Bitterroot Valley. We wanted to study the stopover habitats they use at this time. We used acoustic data from 2017, collected by an autonomous recording unit, at 16 recording stations placed throughout the Bitterroot Valley. The dates ranged from the second week of July to the first week in October. These stations recorded the Nocturnal Flight Calls (NFCs) of songbird migrants, including the Swainson's Thrush. We looked at Swainson's Thrush NFCs that occurred sixty minutes before sunrise because calls given at this time might indicate that birds were close to landing

and stopping over. We used Google Maps to assess the features and vegetation near each station that would offer the habitat we deemed important for migrating birds. We found two sites that had higher Swainson's Thrush calls than other sites: one site north of Darby near the Bitterroot River, and one in the mountains northwest of Florence. These two settings are very similar because they are both located at approximately 3,800 feet, which could indicate an ideal elevation for migration. Additionally, both sites have heavy coniferous forest cover, an important habitat characteristic for Swainson's Thrush. There is not much research available regarding stopover habitat of the Swainson's thrush specifically in Montana. Through this research, we hope to better determine what sites are important to the Swainson's thrush and hopefully be able to preserve said sites to aid the bird in their annual migration.

Conservation Design and Decision Support in the Northern Great Plains Joint Venture (Poster)

Daniel Casey*, Northern Great Plains Joint Venture, Ducks Unlimited, Billings, MT
Kevin Ellison, Northern Great Plains Program, World Wildlife Fund, Bozeman, MT

The Northern Great Plains Joint Venture (NGPJV) covers portions of four states, including 22 counties in eastern Montana. This geography contains some of the most intact, contiguous grassland habitat on the continent, comprising more than 75% of the landscape. It also supports a high diversity of grassland obligate bird species, several of which have seen population declines of 75-90% over the past 50 years. The NGPJV partnership is focused on addressing these declines through the maintenance and management of intact prairie habitats, especially on private working lands. We have developed conservation design and decision support tools to inform voluntary and incentive-based (e.g. Farm Bill) programs and practices, through our Conservation Delivery Network of private lands biologists. Our Conservation Guidance Directory database uses the "plowprint" analysis developed by World Wildlife Fund as a measure of net landscape change (conversion to cropland); using these data in conjunction with soil classes defines both threat and conservation opportunity for each of the counties in the NGPJV. We are then using priority grassland species models (Baird's Sparrow (*Ammodramus bairdii*), Chestnut-collared Longspur (*Calcarius ornatus*), McCown's Longspur (*Rhynchophanes mccownii*), Lark Bunting (*Calamospiza melanocorys*) and Sprague's Pipit (*Anthus spragueii*) to set spatial priorities for habitat restoration, enhancement and protection in a local and regional context. "Conservation Road Show" events at 26 Montana Conservation District offices are helping us address local concerns as we provide technical and financial assistance to our partners. We are using the tool to direct conservation grant money and will be building a web platform to broaden its use in 2019 and beyond.

** Effect of Wildfire Smoke on Nocturnal Bird Migration (Poster)

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Vanessa Haflich, Teacher, Florence-Carlton School, Florence, MT
Kate Stone, Avian Science, MPG Ranch, Florence, MT

Many migratory songbirds travel at night, producing unique nocturnal flight calls (NFCs) for orientation and communication with other individuals. During the fall of 2017, western Montana experienced many wildfires. Preliminary data suggested smoke density may influence bird migration, either by causing more communication from lack of visibility or a decrease in calls due to harmful anatomical effects. The relationship between smoke and NFCs at two locations in western Montana was studied. NFCs were recorded using

autonomous recording units placed at Florence-Carlton High School (FCHS) and east Lolo. Total NFCs were quantified for all species as well as a few individual species: Swainson's Thrush (*Catharus ustulatus*), Savannah Sparrow (*Passerculus sandwichensis*), Chipping Sparrow (*Spizella passerina*) and Wilson's Warbler (*Cardellina pusilla*). Smoke density data was obtained from the Montana Department of Environmental Quality (MT DEQ) in the respective towns. To look at the relationship between smoke and bird activity, Spearman's correlation tests and general linear models that included air quality, time, and location variables to predict the number of NFCs for all birds and individual species were constructed. These variables were analyzed at both nightly and hourly time scales. However, analyses supported only a weak relationship between NFCs and air quality. While analyzing the data differently may result in stronger correlations, it is likely these migration patterns are most influenced by other factors, as smoke may only temporarily alter migration activity. By comparing the frequency of NFCs with smoke density across multiple seasons and locations, a better understanding of the impact wildfire smoke has on migration is hoped to be gained.

** Research in Progress: Assessing the Impact of Increasing Wolf and Grizzly Bear Populations on the Habitat Selection and Foraging Patterns of Cougars in the Southern Greater Yellowstone Ecosystem (SGYE) (Poster)

Jennifer A. Feltner*, Wildlife Biology Program, University of Montana, Missoula

Since the early 2000's, recovering wolf (*Canis lupus*) and grizzly bear (*Ursus arctos horribilis*) populations in the SGYE north of Jackson, Wyoming have been reviving long absent competitive interactions amongst species of the large carnivore guild, potentially leading to behavioral shifts by subordinates such as cougars that can have population and community-level consequences. Research efforts are needed to clarify the responses of cougars following wolf and grizzly bear recovery and resultant impacts to prey populations. In the SGYE, management and monitoring of large mammals is complex. Multiple federal and state agencies, as well as non-profit organizations collect data and conduct research on these species, and anthropogenic impacts ranging from hunting to recreation to supplemental feeding of elk (*Cervus elaphus canadensis*) also play strong roles in the system. However, datasets on the populations, movements and food habits of wolves, cougars, and grizzly bears, as well as their primary prey, elk, from 2001 to the present exist. The aim of this study is to assess the impact of competition from recovering wolves and grizzly bears on cougars by investigating key factors driving cougar habitat selection and foraging patterns, including prey availability, risk of dominant competitor encounter, human activities and other environmental factors. Sixteen years of location data from cougars, wolves, grizzly bears and elk and predation data from cougars shared by my collaborators are currently being analyzed for this study. This project will advance understanding of how competition shapes the behavior of cougars, highlighting potential fitness impacts to cougars and subsequent behavioral shifts that could in turn impact prey species.

** Habitat Selection, Space Use and Demography of Greater Sage-Grouse in North-Central Montana (Poster)

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Mike Borgreen, Glasgow Field Office, Bureau of Land Management, Glasgow, MT
John Carlson, Billings Field Office, Bureau of Land Management, Billings, MT
Lance McNew, Department of Animal and Range Sciences, Montana State Univ., Bozeman

The greater sage-grouse (*Centrocercus urophasianus)* is a sentinel species of sagebrush ecosystems and requires large tracts of intact habitat. Despite the 2015 not warranted for listing decision by the U.S. Fish and Wildlife Service, the sage-grouse is still considered a species of conservation concern. Critical to the not warranted decision is the ongoing development of state-level habitat strategies and sage-grouse focused federal land management plans; both of which require the identification of important seasonal habitats. Recently, federal agencies have identified information gaps associated with sage-grouse habitats in the North-central Sagebrush Focal Area, an important sage-grouse core area in Valley County, MT. In 2018, we began a 3-year study of sage-grouse space use and demography in this area to identify important seasonal habitats in the region. Our primary objectives are to 1) identify conditions influencing seasonal habitat use, 2) evaluate movements and seasonal migration patterns, and 3) evaluate demographic associations with biotic and abiotic habitat conditions and disease risk. During April-May 2018, we captured 45 female sage-grouse and outfitted them with global positioning system (GPS) platform transmitting terminals (PTTs). The transmitters allow us to gather resolute information pertaining to fecundity, female survival, and space use, in relation to seasonal habitat conditions measured both remotely and in the field. We will present preliminary results on demography and space use from our first season.

** Predicting Habitat Suitability for Dusky Grouse in Montana (Poster)

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Claire Gower, Wildlife Division, Montana Fish, Wildlife & Parks, Bozeman
Lance McNew, Department of Animal and Range Sciences, Montana State University, Bozeman

Dusky grouse (*Dendragapus obscurus*), are a forest grouse species found throughout western Montana. Despite being a game species, there has been inconsistent monitoring and few surveys in recent years limiting effective management and knowledge of their ecology in Montana. Previous research indicates that dusky grouse use several different habitat types including conifer forest in the winter and shrub/steppe and grassland communities along the edge of coniferous forests during the breeding season. Our objective was to create a state-wide map predicting relative suitability for dusky grouse occurrence that could be used to identify locations for future surveys. We obtained dusky grouse observations collected during April-June, 2009-2018 from the Integrated Monitoring in Bird Conservation (IMBCR) program and extracted habitat information for detected/not-detected locations using remotely-sensed data. We evaluated relative habitat use with resource selection functions calibrated using generalized linear mixed models. Candidate models representing hypothesized relationships among grouse detections/non-detections and habitat conditions (e.g. forest type and coverage, relative elevation, distance to road) were compared using multi-model inference based on information theory. Preliminary results indicate that relative use for dusky grouse was higher in areas with higher proportions of mixed conifer forest, especially areas with higher proportions of douglas fir and that relative use was lower in grasslands and decreased as the distance to the edge of conifer forest increased.

** Using Ecological Site Condition to Evaluate Habitat Selection by Sharp-Tailed Grouse Broods (Poster)

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Megan Milligan, Dept of Animal and Range Sciences, Montana State University, Bozeman
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Jeff Mosley, Department of Animal and Range Sciences, Montana State University, Bozeman

Brood survival, an important vital rate affecting population viability of sharp-tailed grouse (*Tympanuchus phasianellus*), is largely determined by the selection of brood-rearing habitats by females. Both the quantity and quality of brood-rearing habitat are influenced by land management decisions, and therefore, improper rangeland management can lead to habitat degradation and have a negative effect on sharp-tail grouse populations. Many land management decisions affecting brood habitats (e.g., livestock stocking rates, prescribed burning) are based on metrics including the type and condition of ecological sites. However, associations between brood habitat use and these common rangeland assessment metrics have not been evaluated. We developed a method of delineating ecological sites and assessing vegetation condition by comparing current vegetation to the climax communities across our study area in eastern Montana and western North Dakota. We then evaluated selection ratios of radio-marked brood hens in relation to ecological sites and their relative condition. Our results should provide useful information on brood habitat selection relative to habitat assessment frameworks used by rangeland managers and have implications for the management of sharp-tailed grouse brood habitats in the northern mixed-grass prairie.

Invasive and Pest Species Information at the Montana Natural Heritage Program (Poster and Presentation)

Bryce Maxell*, Montana Natural Heritage Program, University of Montana, Helena
Dave Ratz, Montana Natural Heritage Program, University of Montana, Helena
Andrea Pipp, Montana Natural Heritage Program, University of Montana, Helena
Dan Bachen, Montana Natural Heritage Program, University of Montana, Helena

The Montana Natural Heritage Program (MTNHP) has centralized information on Montana's native species and habitats since its inception in 1985. In October of 2017, MTNHP started managing information on Aquatic Invasive Species, Noxious Weeds, Forest Pests, Agricultural Pests, other non-native species and biocontrol species that have been introduced to control invasives. The MTNHP now manages information on over 500,000 observations of more than 700 invasive and pest species and over 50,000 structured survey locations for invasive and pest species and makes that information available on its websites. This presentation will provide an overview of how biologists and resource managers can access information on surveys, observations, predicted habitat suitability models, and descriptive field guide information for invasive and pest species and native species on the MTNHP's Montana Field Guide, Species Snapshot, and Map Viewer websites.

** New Methods for Monitoring Mountain Goat Populations (Poster)

Molly McDevitt*, Wildlife Biology Program, University of Montana, Missoula
Paul Lukacs, Wildlife Biology Program, University of Montana, Missoula
Frances Cassirer, Wildlife Research Unit, Idaho Department of Fish and Game, Lewiston
Shane Roberts, Wildlife Research Unit, Idaho Department of Fish and Game, Boise

Estimating the distribution and abundance of wildlife populations is a critical component to the conservation of wild species. Precise estimates of these parameters enable biologists to productively manage and preserve the animal populations they oversee. Methods for monitoring high density species populations are frequently and continuously being improved upon. However, identifying effective methods for monitoring low density species populations; e.g. mountain lion (*Puma concolor*), wolf (*Canis lupus*), mountain goat (*Oreamnos americanus*) and wolverine (*Gulo gulo*), has proved challenging due to small population sizes and difficulty of access to low density species habitat. Current approaches for surveying low density species (e.g. aerial surveys, baited camera traps, DNA sampling) often provide insufficient information and application of statistical models to these data can be complex. In this study, and in partnership with Idaho Department of Fish and Game, I will test disparate non-invasive, ground-based methods for monitoring mountain goat populations in Idaho, USA. This project will compare and contrast findings from three field and analytical methods: camera trapping techniques, single-observer ground surveys, and double-observer ground surveys. Here, I describe the field methods and statistical models being tested and developed in research project. Additionally, I will present findings from the first field season (June-August 2018) of testing these three methods. By improving occupancy and abundance estimates for mountain goat populations, biologists can begin to make smarter conservation decisions around mountain goat management.

** Effects of Mountain Pine Beetle on Bat Activity in Western Montana (Poster)

Monique Metza*, Ecology Department, Montana State University, Bozeman
Shannon Hilty, Ecology Department, Montana State University, Bozeman
Dr. Andrea Litt, Ecology Department, Montana State University, Bozeman

Bat activity throughout forests is likely influenced by stand structure, which can be altered by disturbance (e.g., fire, silviculture, and pests). The mountain pine beetle (MPB, *Dendroctonus ponderosae* Hopkins) is a major forest pest in the western United States that has caused tree mortality in millions of hectares of lodgepole (*Pinus contorta*) and ponderosa pine (*Pinus ponderosa*) forests. This disturbance can increase coarse woody debris (CWD) and open canopies through fallen snags. Our objectives were to 1) determine whether CWD can be used as a proxy to characterize severity of the MPB effects, and 2) assess how bat activity changes with MPB severity within lodgepole and ponderosa forests in western Montana. We measured CWD, assessed severity of MPB, and deployed acoustic detectors to evaluate bat activity during the early active season, prior to activity of newly volant pups. Ponderosa-dominated sites had lower average volumes of CWD compared to lodgepole sites. Overall, the amount of CWD did increase with MPB severity. We recorded 8.7 bat passes per night on average (95% CI = 3.2 to 14.2 bat passes/night). However, ponderosa forests (95% CI = 8.4 to 38.6 bat passes/evening) had 20.8 more bat passes per night on average than lodgepole (1.13 to 15.1). Variation in bat activity was not explained by MPB severity alone; future analyses will explore the influence of additional forest characteristics to help us understand how to manage forests in a way that is beneficial for bats.

** Effects of Flushing on Sharp-Tailed Grouse Nest Survival (Poster)

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Lorelle Berkeley, Wildlife Division, Montana Fish, Wildlife & Parks, Helena
Lance McNew, Department of Animal and Range Sciences, Montana State University, Bozeman

Intensive demographic studies of prairie grouse provide valuable information to guide management recommendations. However, field techniques are frequently invasive, often necessitating concentrated capture efforts and frequent flushing of females from nests, which could potentially bias estimates of nest survival by altering either bird or predator behavior. Researcher-induced biases in vital rate estimation has serious implications when those estimates are used to inform management. As part of a larger study on the effects of grazing management on sharp-tailed grouse, we monitored 102 radio-marked female sharp-tailed grouse (*Tympanuchus phasianellus*) in eastern Montana for two years to better understand the effects of flushing on nest survival. A randomly selected subset of radio-marked females were flushed from nests 1-2 times by researchers using standard protocols for game bird nesting studies, while the remainder were never flushed during the nesting season. Daily nest survival was significantly reduced for birds that were flushed from the nest, but the effect was mediated by the amount of precipitation received during the nesting period. A significant negative effect was only observed during periods with little precipitation, with reduced nest survival due almost entirely to predation rather than nest abandonment. Overall, our results suggest that research activities can introduce bias into demographic estimates, but that the effect depends on weather conditions.

Nest Success, Behavior and Disturbance of Two Nightjar Species in Western Montana (Poster)

Mary Scofield*, Avian Science, MPG Ranch, Florence, MT
Kate Stone, Avian Science, MPG Ranch, Florence, MT

The Common Poorwill (*Phalaenoptilus nutallii*) and Common Nighthawk (*Chordeiles minor*) are two nocturnal species breeding in western Montana. These ground-nesting insectivores deploy similar reproductive strategies, but have unique behavioral adaptations for nest success. From 2015-2018, we used cameras and monitoring to document phenology, disturbance, and success at 20 Common Poorwill and 14 Common Nighthawk nests. Poorwills arrived from mid-April to May, and laid their first eggs in May to early June. Nighthawks arrived a full month later, and initiated nests soon after arrival. Both sexes of poorwills incubate, brood and feed chicks. Poorwills often had two nests per breeding season. In contrast, just female nighthawks performed nesting duties, and their late arrival allowed for only one nest in a season. We documented disturbances from six different intruder types, from insects like grasshoppers, to rodents like chipmunks. We observed nine behavioral responses to intruders, including flushing off the nest, defensive posturing, and standing over or beside the nest. Poorwill and nighthawk nests were disturbed most by humans, unknown intruders, and rodents. Poorwills were most likely to flush off of nests upon disturbance, while nighthawks often defended the nest with aggressive posturing. Of the nests that we could determine fate, we confirmed failure at eight poorwill (40%) and two nighthawk (12.5%) nests. Most poorwill nests failed when nestlings were left unattended by adults. We don't know what caused nighthawk nest failure. Future monitoring may give more understanding of how nest disturbances impact nesting success in both poorwills and nighthawks.

Habitat and Land-Use Effects on Scavenging Rates and Potential Brucellosis Transmission in Southwest Montana (Poster)

Kimberly Szcodronski*, Northern Rocky Mountain Science Center, U.S.Geological Survey, Bozeman, MT
Paul Cross, Northern Rocky Mountain Science Center, U.S. Geological Survey, Bozeman, MT

Brucellosis, a bacterial disease caused by *Brucella abortus*, is a major concern in the Greater Yellowstone Ecosystem due to potential transmission from elk (*Cervus elaphus*) to livestock. B. abortus can lead to abortion in infected animals and is primarily transmitted between elk and livestock when individuals contact infected abortion materials. Therefore, the risk of transmission may be a function of how long abortion materials remain on the landscape. Previous studies suggest the rate of fetus removal by scavengers may vary spatially and that scavengers may play a vital role in the persistence of B. abortus on the landscape and the dynamics of brucellosis transmission. To investigate fetus removal in southwest Montana, we placed bovine fetuses and placentas at 266 sites within suitable elk habitat during the brucellosis transmission risk period from February-June 2017 and 2018. We used remote cameras to quantify the scavenging rate of abortion material, as well as the community of scavengers that participate in fetus removal. Preliminary estimates suggest abortion materials were scavenged at an average of 84 hours (± 8.5 SE) across all habitat types. When comparing habitat types, fetuses were removed quicker in grasslands (55 hours ± 8.2 SE) than in sagebrush steppe (102 hours ± 17.0 SE) and forest (102 hours ± 7.4 SE). Abortion materials were consumed by a variety of scavengers including magpies, ravens, red-tailed hawks, eagles, turkey vultures, skunks, foxes, coyotes, wolves, mountain lions, and black bears. This research will help identify management options aimed at decreasing the risk of brucellosis transmission from elk to livestock in Montana.

Beyond Words: Thought – A Visual Attempt to Inspire Thoughts on the Climate Crisis (Poster)

Mike Thompson *, Wildlife Division, Montana Fish, Wildlife & Parks, Missoula

The climate crisis presents unprecedented challenges to the wildlife profession, as it does for all of humanity. Does the human animal have the capacity to wrap its mind around it? This poster is an attempt to convey and inspire individual thoughts about the climate crisis where words in scientific or popular literature may fail us, and from individual thoughts may arise actions and initiative beyond our experience. For actions beyond our experience are required of those who strive to make a difference, and of others who would hope to support, rather than impede, their efforts. So, we hope that the visual image of this poster will speak to someone for whom words are not as impactful.

Decoding Avian Migration: Cold Fronts and Their Influence on Nocturnal Flight Calls (Poster)

Carrie Voss*, Avian Science, MPG Ranch, Florence, MT
Debbie Leick, Avian Science, MPG Ranch, Florence, MT
Kate Stone, Avian Science, MPG Ranch, Florence, MT

Passerines respond to a variety of environmental cues during migration. Some research suggests that passerine migration may be influenced by weather variables such as the passage of cold fronts. During fall 2017, we used acoustic recorders at 23 stations to document the nocturnal flight calls (NFCs) of migrating passerines in the Bitterroot Valley. We looked at

correlations between the number of NFCs and the daily change of two weather variables that may indicate an oncoming cold front: temperature and barometric pressure. Cold fronts tend to correlate with migration waves and an increase in nocturnal flights calls. In the future, we intend to include other environmental variables including light pollution, cloud cover, wind direction and precipitation.

Keeping the Crown of the Continent Connected: A Report on Highway 2 Near Glacier National Park (Poster)

John Waller*, Science and Resources Management, Glacier National Park, West Glacier, MT
Tabitha Graves, Glacier Field Station, U.S. Geological Survey, West Glacier, MT

The US Highway 2 corridor separates Glacier National Park from the Bob Marshall Wilderness complex to the south. With increasing vehicle traffic, recreation, and high train traffic, resource managers in the region are concerned that Highway 2 is slowly becoming a barrier to north-south wildlife movement in the Crown of the Continent Ecosystem, and thus, this corridor has been identified as a priority for wildlife connectivity planning. While there have been a number of efforts to understand wildlife connectivity across this corridor, they have tended to be narrowly focused and temporally disjointed. Over the last year, an interagency group of local researchers and managers met in two workshops to evaluate existing research and data sources, identify knowledge gaps, and establish a research framework to increase understanding of wildlife use of the US2 corridor. The long-term goal is to identify explicit management options for preserving trans-highway movements, seasonal migrations, and dispersal movements of animals, plants and ecological processes. This report builds on previous efforts to understand and plan for terrestrial wildlife connectivity across this inter-jurisdictional corridor by beginning a multi-agency conversation for collaborative research and management.

Montana Academy of Sciences

2019 Annual Meeting

April 5-6, 2019
Student Union, Montana Tech
Butte, Montana

Chrissie Carpenter, President, Montana Academy of Sciences
James N. Barron, Executive Director, Montana Academy of Sciences

Introduction

The Montana Academy of Sciences (MAS) was incorporated on the 20th day of March, 1961, as a non-profit, educational organization. The objectives of the Montana Academy of Sciences are to encourage interest and participation in the sciences and to promote public understanding of science and its contribution to society. The Academy accomplishes its objectives by conducting meetings of those interested in sciences and the education of scientists, by publishing contributions to scientific knowledge, by supporting research, by making awards to recognize accomplishments in science, by administering gifts and contributions to accomplish these aims, by assigning and cooperating with affiliated and other organizations with similar objectives, and by engaging in such other activities as deemed necessary to accomplish its objectives.

We held our 2019 Annual Meeting at Montana Tech in Butte, MT. on April 5 and 6. Over 90 registrants participated, viewing 24 contributed oral presentations and 27 poster presentations over the day and a half meeting. We present the abstracts from our meeting here so that the readers of the Intermountain Journal of Sciences can see the quality and types of science supported by MAS. Please mark your calendars for our next meeting, Saturday, April 4, 2020 in Butte. Finally, the Board of Directors of MAS would like to thank the sponsors of our 2019 Annual Meeting:

Dr. John Cech, President, Carroll College, Helena

Dr. Beverly Hartline, Vice Chancellor for Research, Montana Technological University, Butte
  The Department of Biological and Physical Sciences, Montana State University, Billings
  College of Humanities and Sciences, University of Montana, Missoula

Dr. Matt Redinger, Provost, University of Providence, Great Falls

Presentation Abstracts

*Indicates Presenter

Comparing the Habitat Connectivity of American Marten and Fisher in the Northern Rocky Mountains, USA

Rex Koenig*, Hellgate High School, Missoula, MT

The connectivity of a species is the ability of its individuals to move, and importantly, to move between populations. This project predicts and compares the connectivity of American marten (*Martes americana*), as well as fisher (*Pekania pennanti*) in western Montana and northern Idaho, to determine the presence of wildlife corridors that facilitate the connectivity of both. MaxEnt was used to construct a niche-model for American marten using marten location data coupled with environmental covariates (topography, precipitation, snow, temperature, vegetation). Based on the assumption that identified areas of use also serve to facilitate connectivity, this niche model was transformed into a resistance surface. Connectivity models were then derived from this. This same process was followed using a published niche-model for fisher. Assuming that both fisher and marten might travel between areas of the highest quality habitat, connectivity nodes were located randomly within areas of high quality habitat and Circuitscape was used to infer connectivity paths. To compare these connectivity maps they were then quantiled into ten grading on the value of connectivity, and added together for a total quantile value of twenty. Areas with the top two quantiles of connectivity were define as mutual connectivity. This study identified seven mutual wildlife corridors for these two species, and three areas of high mutual use. These findings may not only advise land management regarding how to better preserve the connectivities of American marten and fisher, but also the connectivities of other similar species.

Failure to Detect Anti-Viral Activity by Prokaryotic Argonaute Proteins

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Tanner Wiegand, Microbiology and Immunology, Montana State University, Bozeman

Royce A. Wilkinson, Microbiology and Immunology, Montana State University, Bozeman

Dominick Faith, Microbiology and Immunology, Montana State University, Bozeman

Blake Wiedenheft, Microbiology and Immunology, Montana State University, Bozeman

Argonaute (Ago) proteins are present in all three domains of life and are involved in nucleic acid guided silencing and interference pathways. The well-studied Eukaryotic Argonautes (eAgo) form the catalytic core of the RNA interference (RNAi) pathway that is involved in gene silencing, transposon silencing and antiviral defense. Limited research on Prokaryotic Argonautes (pAgo) suggests these proteins are involved in defense against foreign genetic elements such as plasmids. pAgos have been hypothesized to defend against bacteriophage, however no direct evidence has been found so far. Here, we report the failure to detect antiviral defense mediated by pAgos against various Escherichia coli bacteriophages. We overexpressed 8 phylogenetically diverse prokaryotic Argonaute proteins in Escherichia coli BL21 (DE3) and challenged them with 7 bacteriophages spanning the Myo-, Sipho-, and Podoviridae families. No antiviral activity was detected that could be attributed towards the pAgo proteins. However, up to 100,000-fold reduction in viral infectivity was observed for 2 phages that correlated with plasmid-based protein expression. The mechanism through which this antiviral activity is mediated is unknown and subject for future study.

Ligand-Controlled Chemodivergent Suzuki Cross Coupling Using Palladium-N-Heterocyclic Carbene Catalysts.

Emily Reeves*, Chemistry & Biochemistry, Montana State University, Bozeman.

Sharon Neufeldt, Chemistry & Biochemistry, Montana State University, Bozeman

The successful synthesis of complicated organic molecules such as pharmaceuticals, agrochemicals, and organic materials requires an extensive toolbox of synthetic strategies for constructing carbon-carbon bonds. Problems can arise, however, when a molecule contains multiple functional groups that are reactive toward the same transformations. For example, both aryl halides and aryl triflates can undergo palladium-catalyzed Suzuki cross-coupling in the presence of arylboronic acids. Control of selectivity between these two electrophilic sites using different Pd-phosphine catalysts has been described previously, but the scope of such methodology is severely limited. In this presentation, we describe the discovery of an orthogonal pair of Pd-N-heterocyclic carbene (Pd-NHC) catalysts for chemodivergent cross-coupling of aryl chlorides and aryl triflates with phenylboronic acids and demonstrate the synthetic utility of these catalysts with diverse boronic acids and substrates. The mild conditions used in these reactions allows for high functional group tolerance with respect to both substrates and boronic acids. We additionally present experimental and computational evidence that the origin of selectivity using Pd-NHC catalysts is dissimilar to the mechanism of selectivity control using Pd- phosphine catalysts.

Identification of Cellulolytic Hot Spring Organisms Through Bioorthogonal Labeling

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Zackary Jay, Chemistry and Biochemistry, Montana State University, Bozeman

Viola Krukenberg, Chemistry and Biochemistry, Montana State University, Bozeman

Rachel Spietz, Chemistry and Biochemistry, Montana State University, Bozeman

Roland Hatzenpichler, Chemistry and Biochemistry, Montana State University, Bozeman

Bioprocessing of cellulose from plant waste into ethanol has been a focus of the renewable energy field for several decades. The recent developments of genome sequencing technologies have reinvigorated the topic. Identification of genes and the potential to degrade cellulose has been the major outcome thus far. High-throughput techniques to link taxonomy and in situ function of the organisms responsible for these processes have, however, been lacking. Bioorthogonal non-canonical amino acid tagging is a nondestructive method to fluorescently label active, protein-synthesizing cells that can later be separated from a community using fluorescent activated cell sorting (BONCAT-FACS). Biomass collected from a Yellowstone National Park hot spring was incubated with four cellulose substrates to identify organisms that were preferentially active in the presence, but not the absence of cellulose substrates. A short timeframe (2 weeks) bioorthogonal labeling incubation and a longer enrichment (6 weeks) incubation were compared for differences in community composition to highlight the competition of an incubation that is typically missed with enrichment schemes. Over 14 days of incubation the community began to shift to a simplified population comprised of Aquificae, Deinococcus-Thermus, or Chloroflexi. However, at early time points, the active community was comprised by a multitude of uncultured and cultured organisms including Fervidibacteria, Aigarchaeota, or Thermotogae which were eventually outcompeted by dominating organisms in the later time points. BONCAT-FACS outlines a novel approach of using single-cell bioorthogonal labeling to profile a microbial community's activity regarding biotechnology industry interest.

Ecosystem Engineering by Net-Spinning Caddisflies in Rocky Mountain Streams

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Lindsey K. Albertson, Ecology, Montana State University, Bozeman
Molly McLaughlin, San Francisco state university, San Francisco, CA
Zachary Maguire, Ecology, Montana State University, Bozeman
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Melinda D. Daniels, Stroud Water Research Center, Avondale
Leonard S. Sklar, Geography, Planning and Environment, Concordia, Montreal

Ecosystem engineering is a ubiquitous process by which organisms modify physical habitat characteristics and influence patterns of biological organization. Often, modification of physical environments by ecosystem engineers can facilitate other organisms by creating habitats for species that would otherwise be absent. Despite the potentially wide reaching consequences of facilitation through ecosystem engineering, many facets remain poorly understood in stream ecosystems. Here we present a synthesis of ecosystem engineering by hydropsychid caddisflies (*Hydropsychidae*) and describe how they affect stream macroinvertebrate communities. Hydropsychid caddisflies are a globally distributed group of net-spinning insects that live in stream gravel beds and have high abundances across western Montana. Hydropsychid caddisflies act as ecosystem engineers because their silk structures alter sediment transport conditions and local flow patterns of streambeds. Using lab experiments and field surveys, we show that ecosystem engineering by caddisflies changes physical templates of streambeds and influences other members of the macroinvertebrate community. We found that caddisfly nets increase the shear stress required to initiate gravel movement for gravels up to 70 mm. Additionally, we found that caddisfly silk structures substantially alter local flow at the streambed surface by reducing velocity by 70%. Furthermore, we have found that the presence of net-spinning caddisflies markedly increases local abundances and biomass of other stream macroinvertebrates across environmental gradients. Taken together, these findings indicate that caddisflies impart substantial physical changes to streambed habitats that have ecologically significant consequences for stream macroinvertebrate communities. Our findings indicate that caddisflies influence ecological processes from physical habitats to biological community structure and could act as important controls of Rocky Mountain stream ecosystems

Assessing the Effects of Competition and Seasonality on Stress, Immunity and Number of Scars in Deermice

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Amy Kuenzi, Biology, Montana Tech of the University of Montana, Butte
Angela Luis, Ecosystem and Conservation Sciences, University of Montana, Missoula

Infectious wildlife diseases are becoming more common, causing population declines and species extinctions. Ecological and environmental stressors can influence disease spread in wildlife, through effects on parasite transmissibility (regulated by host immunity), and contact rates. Glucocorticoids (GCs) are hormones that mediate physiological and behavioral responses to stressors, and thus, can influence immunity and behavior in wildlife. Sin Nombre virus (SNV) is carried by deermice (*Peromyscus maniculatus*), and in western Montana grasslands, deermice compete with voles (*Microtus* spp.) and shrews (*Sorex* spp.). Because voles are dominant over deermice, they could increase SNV prevalence in deermice via stress-induced immunosuppression and/or alteration in contact rates, while shrews may have

a lesser effect. Our objectives were to investigate whether voles and/or shrews could increase SNV prevalence in deermice through changes in stress, immunity and/or contact rates, and to examine seasonal changes in these same measures. We live-trapped small mammals over 2 years in western Montana grasslands. We evaluated deermice for scar numbers (proxy for contact rates), demography, and body condition scores (BCSs). Deermouse blood was evaluated for white blood cell counts/differentials, and SNV antibodies, and feces for fecal corticosterone metabolites (FCMs) to measure stress (baseline and stress-induced). Using mixed effect regression trees, we found that higher vole density was inconsistently associated with lower BCSs and scar numbers. Higher shrew density was consistently associated with lower stress-induced FCMs, but inconsistently with lower BCSs, and higher scar numbers. Neutrophil: lymphocyte (N:L) ratios were highest in spring/summer, and lowest in fall/ winter. Due to low SNV prevalence, we could not evaluate effects on infection. Interspecific competition may influence disease spread via effects on chronic stress (i.e. lower stress-induced FCMs and BCSs), and scar numbers. Higher N:L ratios, suggestive of chronic stress, over spring/summer may provide an ideal time for SNV transmission. Our findings may extend to other diseases

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Evaluating the Accuracy of Bloodstain Pattern Analysis Using Hemodynamic Properties

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Mykal Gernaat, Forensic Science, University of Providence, Great Falls, MT
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Meredith Hecker, Mathematics, University of Providence, Great Falls, MT

This comparative survey explores the relationships between the discipline of bloodstain pattern analysis (BPA) and hemodynamic blood properties, such as viscosity and hematocrit. In BPA, forensic scientists study the phase change of blood when in contact with air, but little forensic literature connects blood biomechanics, such as hematocrit levels to BPA. Red blood cell count, or hematocrit, in females (37-48% of blood volume) is slightly lower than males (45-52% of blood volume) from menstrual red cell loss, etc. Strong evidence suggests that erythrocytes influence blood viscosity because of their high concentration (4-6 x 10^6 RBC/mm3 or 40-45% of blood volume in healthy individuals). When whole blood is altered by a disorder/disease or alcohol intake, hematocrit levels can be affected as well. With this knowledge, there is reason to believe that blood viscosity changes with individualistic hematocrit levels. Therefore, it is hypothesized that traditional BPA angle of impact tests can produce inaccurate results. Intravenous blood samples were drawn from 9 volunteers (all women, including 8 with blood disorders/alterations and 1 healthy control) at the University of Providence into collection tubes containing ethylenediaminetetraacetic acid as an anticoagulant. Each sample was tested for viscosity using a Canon-Fenske viscometer and for hematocrit levels using an LW Scientific ZipCombo centrifuge. Finally, each sample was used to make several blood stains at varying degrees of impact (10°, 30°, 60°, and 90°). A MANOVA was used to compare viscosity, hematocrit, and angle of impact variables against each other within the 8 participants. This survey connects hemodynamic properties to angle of impact tests in BPA by significantly showing how bloodstains can be inaccurately misinterpreted. By examining blood viscosity among several individuals, this research assesses the accuracy of BPA by comparing experimental and expected bloodstain angles and creating individualistic standards for future forensic methods.

Generation of a High Throughput Screening System for Small Molecules that can Rescue Axonopathy

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David McGee, Biological and Physical Sciences, Montana State University, Billings
Lynn George, Biological and Physical Sciences, Montana State University, Billings
David Butler, Biological and Physical Sciences, Montana State University, Billings

Familial Dysautonomia (FD) is a neurological disease with both developmental and degenerative aspects including axonopathy of peripheral nerves. FD results from a point mutation in the ELP1 gene, causing reduced levels of the corresponding protein that functions in assembling a highly conserved, six-subunit complex known as Elongator. Elongator catalyzes the chemical modification of transfer RNAs needed for the translation of codon-biased transcripts that preferentially use AA- or AG-ending codons. Like FD, axonopathies are a common feature of many other neurological diseases including amyotrophic lateral sclerosis (ALS), and Alzheimer's. Despite their prevalence in neurodegenerative diseases, the discovery of therapeutics for treating axonopathies has been impeded by the difficult and costly nature of culturing primary neurons. Therefore, an experimental model that can withstand the manipulation required for a high throughput small molecule screen is essential for drug discovery. Candida albicans shows a pronounced polarized growth phenotype that is distinct among other yeasts. To determine whether this phenotype is dependent on Elongator, as is polarized growth in neurons, we made a C. *albicans* Elp1 knockout. Importantly, this knockout exhibits a severely compromised growth habit. Our long-term goal is to use this knockout to develop a high throughput screen for small molecules that can rescue normal polarized growth. Molecules that rescue in our yeast model system, will then be tested in vitro for the ability to rescue axon elongation in Elp1 deficient neurons.

A Logistic Regression Implementation of the "ABCD" Method for Identifying Malignant Melanoma

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Stephen Muir, Mathematics, University of Providence, Great Falls, MT

This project investigated a way to more accurately assess the probability that a skin mole is malignant or benign using the ABCD classification system used in healthcare. To determine the probability that a mole was malignant or benign, numerical values were calculated for each classification. The numerical value for A (asymmetry) was calculated by drawing an estimated half-way point through the image and then filling in each half with simple geometric shapes whose areas could be easily calculated in Microsoft Word. Once the area of each half was found, the absolute value of the difference between the area of the two halves was the numerical value assigned to A. For B (border irregularity), the perimeter and area of each mole was put into the formula $B=(P)2/(4\pi T)$, where T stands for area. C (color) was estimated on a scale of 1-5, where 1 was light, uniform color distribution and 5 was uneven, splotchy, and dark color distribution. For D (diameter), the archive used (ISIC Archive) to obtain the images had the diameter included in the metadata of each image. In total, 45 training images were used, and 5 different test images were used to cross-validate the results obtained. Both a quadratic logistic regression model and linear logistic regression model were used to see how accurate both models were in predicting the probability that a mole was malignant or benign. The results produced showed that the quadratic model was more accurate than the linear; however, both models had a high rate of accurate predictions. The quadratic model accurately predicted 44 out of 45 of the training data sets and 5 out of 5 of

the test data sets. The linear model accurately predicted 41 out of 45 of the training data sets and 4 out of 5 of the test data sets.

LIGAND K-EDGE STUDY OF M(PDTC)L COMPLEXES

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Siana Wiles, Biological and Physical Science, Montana State University, Billings
Dr. Matt Queen, Biological and Physical Science, Montana State University, Billings

Carbon tetrachloride is a known carcinogen that can cause cancer related illnesses when it is dechlorinated by Cytochrome P450, located in the human liver. Environmental reduction of carbon tetrachloride can produce lesser chlorinated intermediates, such as the trichloromethyl radical, which is harmful to living organisms. [Cu(PDTC)L]- has shown to be able to dechlorinate carbon tetrachloride so that it will produce CO_2 and chloride, which are environmentally safe compared to lesser chlorinated intermediates. The mechanism between [Cu(PDTC)L]- and carbon tetrachloride is poorly understood. This study links the dechlorination reactivity of a series of [M(PDTC)L]- complexes (L = Cl-, CN-, and PPh3 and M = Cu and Ni) to the individual electronic structures of each complex by using sulfur k-edge. We find that reactivity of a given [Cu(PDTC)L]- species is linked to the lability of L in a given solvent rather than to the relative covalency of the M-S bond in each species.

ELECTRICAL PROPERTIES OF CARBON NANOFLUIDS

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Electrically conductive inks have wide applications in wearable electronics, sensors, and inkjet printed circuits. The goal of this project was to make quantitative measurements on the electrical properties of aqueous suspensions of carbon nanomaterials. To create the electrically conductive ink, Sodium Dodecyl Sulfate (SDS) was used to stabilize the MWNTs in solution. Coats of ink were applied to standard printer paper to measure the sheet resistance of the ink using a Signatone S-302 4-point probe. The resistivity of the ink was determined with an apparatus that I designed and built, it used a series of inter-locking vinyl tubes and solid copper rods as the electrodes. An Agilent 3458A 81/2 Digital Multimeter was used to capture a resistance measurement which was then used along with the dimensions of the vinyl tubes to calculate the resistivity of the ink. By measuring the resistivity of a 5 wt% sample of saltwater and then comparing it to the values found in a table, it was concluded that the apparatus was accurate within a reasonable margin of error.

CHICKEN TAIL VERTEBRAL FUSION SHEDS LIGHT ON A HUMAN BACKBONE DISEASE

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Susan C. Chapman, Biological Sciences, Clemson University, Clemson, SC.

Ankylosing spondylitis (AS) is an inflammatory disease that causes vertebral fusion, affecting approximately 0.6% of the U.S. population. Genome-wide association studies have shown that over 90% of AS patients harbor the HLA-B27 allele, which is associated with the histocompatibility complex. Still, underlying cellular mechanisms of AS are little understood, and a trigger has not been identified. Since AS mechanisms are difficult to study in humans, we require a suitable animal model. The chicken naturally fuses four of its last tail vertebrae during post-hatching growth, forming a compound bone called the pygostyle.

As part of this fusion process, cartilaginous intervertebral discs remodel to become bone, and the nuclei pulposi at the centers of the discs completely degrade. How do chicken discs mineralize and does this phenomenon follow a similar path as human AS? We have observed several analogous features between chicken and AS vertebral fusion, including tissue bands bridging fusing vertebrae and fat deposits at the base of fusing vertebrae. Also, we observe an infiltration of neutrophil-like cells, suggesting that chicken vertebral fusion, like in AS, involves the immune system. Histology stains indicate that mineralization is present, but osteoid and osteoclasts are absent; fusion does not occur by endochondral ossification. We hypothesize this phenomenon represents the unusual transchondral ossification, where chondrocytes transform into osteoblasts. Currently, we're testing for cell death using TUNEL assays. Cell deaths would suggest that chondrocytes are dying not transforming. A possible trigger scenario involves transdifferentiation of disc chondrocytes into osteoblasts. Dying notochordal cells may cause the release of Complement 3 which facilitates transchondral ossification by recruiting neutrophil cells and promotes transdifferentiation of chondrocytes to osteoblasts. Finally, this study provides insights into AS mechanisms and elevates the chicken as an animal model.

Design, Build and Testing of Resonant Air Compressor

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Emily Maynard , Mechanical Engineering, Montana Tech, Butte
Dr. Peter Lucon, Mechanical Engineering, Montana tech, Butte

Mechanical resonance of a system is achieved when the oscillation amplitude of a system is maximized and the resultant velocity and input force are in phase. In theory, a vacuum pump running at resonance should be more efficient when compared to an off-the-shelf air compressor. The focus of this project was to build a resonant compressor, obtain efficiency values from that compressor, and compare those values to that of an off-the-shelf compressor. An off-the-shelf air compressor was tested by pulling a vacuum of a bell jar while collecting data for every 100 mmHg of vacuum that was pulled to the vacuum pressure of 400 mmHg. The values collected were the current, voltage, power, and time it took to reach each vacuum value. In building the resonant air compressor various pieces were printed using 3D printing technology, machining various parts out of steel, and purchasing steel springs. In the design of the new compressor, the piston and valving from the off-the-shelf compressor were used. A copper wire was coiled around a 3D printed bobbin and the compressor was assembled. Alternating current was sent through the copper wire and the current, voltage, and power values were all collected during the same vacuum test as previously defined. These values were then compared to the values collected from the off-the-shelf air compressor.

Zic Transcription Factors that Influence Convergent Extension

Jocelyn Waggoner*, Cell Biology and Neuroscience, Montana State Univeristy, Bozeman

My research is focused on a specific time frame during embryonic development called gastrulation, in which intricate cell movements give rise to the ectoderm, mesoderm and endoderm layers of the embryo. The mesoderm layer undergoes movements called convergent extension, which allow cells to intercalate and move inside the embryo. The Zic family of zinc-finger proteins have a crucial role in gastrulation and neurulation. Based on findings from our lab (K. See and Merzdorf, in preparation) and others (Cast et al., 2012), I hypothesize that zic genes are required for convergent extension during gastrulation. Since there are 5 different, but very similar, zic genes, I will test each of these genes for a role in convergent

extension during gastrulation. A knockdown method for each of the 5 zic genes in embryos of the model organism Xenopus laevis will be combined with a method called Keller Explants to allow analysis of convergent extension. In my experiments, I have ruled out zic 2 as a regulator of convergent extension. My current hypothesis is that zic3 is the most likely candidate because of its known roles in neural tube formation and other reasons I will explain below. I am also analyzing zic1,4 and 5 for convergent extension regulation

Is Time Resolved Infrared Radiometry an Alternative of Positron Annihilation Lifetime Spectroscopy for Defect Detection in Metals: An Experimental Approach and a Model Development

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Brahmananda Pramanik, Mechanical Engineering, Montana Technological University, Butte

Time resolved infrared radiometry (TRIR) and positron annihilation lifetime spectroscopy (PALS) are both defect detection methods used in different materials for more than 40 years. Interestingly, both of the methods are similar in many ways, such as non-invasive testing procedures and follow a decay scheme. Both of the methods are successfully applied to metals, polymers, and composites materials. However, the methods are different in application mechanism and principle. TRIR is a thermography technique that measures temperature decay with time of the investigated material whereas PALS uses positronium decay with time of the tested material. In PALS, two-state trapping model is mostly used. Two state trapping model is a positronium trapping model used to describe positron diffusion inside the material. According to the trapping model, high energy positron beam is emitted from radioactive nuclei, diffused into the material, and trapped in a defect caused emissions of comparatively lower energy γ (gamma) rays. We develop a model to describe the similarity between time resolved infrared radiometry and positron annihilation lifetime spectroscopy. In the model, we describe the heat transfer and diffusion mechanism in the investigated material in time resolved infrared radiometry. The model also describes an experimental development for time resolved infrared radiometry research.

Student Engagement While Establishing Classroom Mathematical Practices

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Derek Andrew Williams, Mathematics, Montana State University, Bozeman

There is a significant connection between student engagement and performance achievement. Klem and Connell write, "student engagement has been found to be one of the most robust predictors of student achievement and behavior in school, a conclusion which holds regardless of whether students come from families that are relatively advantaged or disadvantaged economically or socially" (2004, p. 5). However, student engagement is complex, and currently relationships to outcomes such as mathematical understanding and learning are elusive (Fredricks, Blumenfeld, & Paris, 2004; Middleton, Jansen, & Goldin, 2017). This study investigated student engagement while learning through use of an app that collected student engagement reported by participants during a classroom teaching experiment. This paper discusses preliminary results on students' engagement in the process of learning. Though not anticipated, we observed differences between male and female students' engagement while working in mixed-pairs worthy of investigation. In particular, we observed differences between male and female students' engagement while working in

mixed-pairs surrounding important mathematical contributions from female partners. Female students described situations in which they perceived of male partners overlooking valuable contributions towards completing tasks, resulting in dips in engagement. With, regards to data collection, the app and survey effectively gathered information on student engagement, which was triangulated by students' descriptions in recall interviews.

CREATING A RECIPROCATIONG AIR COMPRESSOR USING A SINGLE DEGREE OF FREEDOM SPRING MASS DAMPER RESONANT SYSTEM

Emily Maynard*, Mechanical Engineering, Montana Technological University, Butte

Theoretically, a reciprocating air compressor should work with a single degree of freedom spring mass damper resonant (1D-SMD) design. The following steps were used to demonstrate how the system could be applied to an air compressor. Once a mathematical model was created, the calculated results determined the parameters for the solid model using SolidWorks. The model included a design that was based around an off-the-shelf air compressor. These parts were either 3d printed, ordered or machined and assembled. Finally, the system was tested as an air compressor. Several design iterations were performed to improve the system. The application of this project resulted in a newly built air compressor, which suggests this is an efficient way to compress air.

DEVELOPMENT OF A UAS-BASED ELECTROMAGNETIC INDUCTION SENSOR FOR SUBSURFACE CONDUCTIVITY MAPPING

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The Electromagnetic Induction (EMI) method is a popular and favorable geophysical technique for shallow subsurface exploration because of its high-quality, rapid data acquisition of subsurface conductivity or resistivity. However, EMI based instruments are generally ground-based. Our research aimed to investigate the practical application of an airborne EMI sensor that does not require direct contact with the ground. The objective for our project was to measure the inductive response of a conductive subsurface, and locate anomalously high subsurface zone in conductivity. To address our objective, we designed and constructed a lightweight EMI sensor for an unmanned aircraft system (UAS) based on the principles of EM induction and EM sounding. We successfully tested the instrument by measuring the secondary field response from a metal-cased Unexploded Ordinances (UXO) during its high conductivity. The innovative design and construction of our EMI sensor is original and has shown to be a practical approach for use with an UAS. The EMI sensor consists of a signal wave generator, a power amplifier, concentric transmitter and a receiver coils, and a microcontroller-based data collection and storage system. The micro-controller code for the data collection and storage system was developed in the open-source Arduino Software, Integrated Development Environment (IDE). EMI sensors operate using a transmitter coil to generate a primary magnetic field that penetrates into the subsurface and induces eddy currents in conductive subsurface Earth materials. A secondary magnetic field generated by the eddy current induced in the conductive Earth material will pass through the receiver coil, and generating an electromotive force (emf). By measuring the emf induced in the receiver, we can approximate the conductivity of Earth material.

Sin Nombre Virus Prevalence in Deer Mice Captured in Ranch Buildings in Southeastern Montana

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Sin Nombre Virus (SNV) is a type of Hantavirus that is carried by deer mice (*Peromyscus maniculatus*). When SNV is transmitted to humans it causes a serious, sometimes fatal, illness known as Hantavirus Pulmonary Syndrome (HPS). Most HPS cases are linked to SNV exposure in peridomestic environments, particularly human dwellings and out-buildings such as garages, sheds, or barns. While many studies have examined SNV prevalence and transmission in natural environments, little work has been done in peridomestic environments. The objectives of my study were to determine the prevalence of SNV in deer mice found in the outbuildings of my family's ranch south of Hardin, MT. I used Sherman live traps to capture mice in around ranch buildings for 3 nights each month beginning in August 2018 and continuing until February 2019. Captured mice were ear tagged with sequentially numbered metal fingerling tags and a blood samples were collected. Blood samples were tested for antibodies reactive with SNV recombinant nucleocapsid protein by an enzyme-linked immunosorbent assay (ELISA). During the course of my study I collected 31 blood samples from 25 different mice captured in ranch buildings. The majority of mice I captured were females (64%) and capture were highest in December. None of the mice I captured tested positive for SNV antibodies.

Science Education Exploration

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What inspires college-bound people to enter science-related majors? This question is critical to understanding the complex issue of getting more people to go into STEM fields in order for the USA to remain competitive and to advance our economy and society. The objectives of this study was to: explore what variables have influenced undergraduate student's decision to select a science-related major at Montana Tech; gain an understanding of students' attitudes toward K-12 education; examine the influences from K-12 teachers; and explore the level of external support to choose a science major. I hypothesized that studying these influences, along with prior research, would lead to a greater understanding of students who selected a science major as well as reveal ideas of how to support and recruit a diverse group of students for these majors. My research indicates that over half of our responders felt that their K-12 education prepared them for college and that their favorite subject influenced their chosen major. A new understanding could lead to improved techniques in science education in the Montana school systems, including high school and elementary, to engage and interest the students.

Metagenomic VS 16S DNA Sequencing for Identifying Bacterial Populations

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Next Generation Sequencing (NGS) technology has launched immense growth of DNA sequence databases, paving the way to better categorize the genetic diversity of the biosphere. Bioinformaticians curate and analyze this massive amount of DNA sequence

data for applications that range from science and medicine to forensics and commercial ancestry services. The DNA sequences provided by NGS can be analyzed to profile bacterial communities within environmental niches. Experimental approaches for assessing these communities vary widely between labs. Our goal was to compare two common methods to test for cohesion between these approaches' results. We obtained DNA sequences from both metagenomic ("shotgun") and bacterial 16S gene sequencing for five DNA samples (two soil and three human microbiome). We developed a custom software pipeline in conjunction with the program BLAST, processed hundreds of thousands of DNA sequence reads produced by each sequencing method (metagenomic or 16S) and, for each sample, compared the two resulting bacterial profiles. For all five samples, community taxonomic profiles produced from the two methods were distinctly different. Explanations for these disparities may include sampling bias in databases or the reliability of the 16S gene as a species identifier. These results highlight the importance of establishing a common methodology to accurately infer bacterial communities from DNA sequence data.

COMPARISON OF WORDS AND PHRASES USED FOR CATEGORIES IN RISK ASSESSMENT MATRICES

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Occupational safety has moved from a rule-based practice to a progression making use of risk assessments. A core assessment tool in risk assessment is a risk matrix consisting of two or three categories for assessing a hazard. This project addressed the words and used to define the categories of severity, likelihood and exposure frequency. We identified from literature 16 words for each category and created a 16-page paper survey to obtain numerical ratings of all of the words. After obtaining Institutional Review Board approval, we surveyed senior engineering design courses and an OSH senior course. These courses were selected because the students are likely to be involved in risk assessments during their career. Respondents rated each word on a 100-point rating scale. An initial quality check was performed to identify respondents who made reasonable efforts and understanding to provide a genuine rating. Data from the selected booklets were used to determine the mean and standard deviation of each word. The last phase of the analysis involved Montana Tech Occupational Safety and Health faculty with professional credentials to identify recommended word sets. We had 82 qualified survey results. The faulty meeting resulted in recommended sets of ordered words of 3, 4, 5 and 6 word-sets for severity, likelihood and exposure. For example, an exposure category we recommend a 4-word categorization of very frequent, frequent, infrequent and very infrequent with mean values of 85, 72, 23, and 15, respectively.

POSTER ABSTRACTS

A REVIEW ON PROPHYLAXIS TREATMENTS FOR CHRONIC LYMPHOCYTIC LEUKEMIA INFECTIONS

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Chronic Lymphocytic Leukemia (CLL) is a type of cancer of the blood and bone marrow that degrades the immune system of a person. This is due to the uncontrolled white blood cell (lymphocyte) growth. The disruption of lymphocyte growth affects the response to pathogens, leading to a disruption in the immune system. Patients with CLL are prone to infections due to their immunodeficiency. As of 2019, there have been 20, 720 new cases of CLL in the United States and approximately 3,930 deaths. Recent studies have analyzed infection prevention

methods to increase survival rates for people with CLL. Intravenous Immunoglobulin Prophylaxis (IVIG) has been demonstrated to elevate levels of immunoglobulins, which are as the first line of defense against pathogens. The IVIG entails extracting antibody proteins from the plasma of a healthy donor and injecting those antibodies into a CLL patient. Another treatment for this infection is the antimicrobial drug Fludarabine Prophylaxis (FAMP). FAMP reduces the growth of lymphocytes in the blood system which allows for the immune system to produce regular amounts of immunoglobulins necessary to prevent infections. This review compares the biochemistry of IVIG and FAMP as treatments for infections in patients with CLL and indicates that IVIG is a more effective treatment than FAMP.

A Small RNA That Regulates Pyrimidine and Methionine Metabolism is Necessary for Establishing Coxiella Burnetii's Intracellular Niche During Early Stages of Infection

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Coxiella burnetii is an obligate intracellular gammaproteobacterium and zoonotic agent of Q fever in humans. We previously identified 15 small RNAs (sRNAs) in C. burnetii with differential expression in the large and small cell developmental forms grown axenically and in infected host cells. Here, we describe the function of one of these sRNAs, termed Coxiella burnetii small RNA 12 (CbsR12). CbsR12 is highly expressed in both large- and small-cell variants in vitro, and is the dominant non-tRNA/rRNA/tmRNA transcript in both morphotypes during mammalian tissue culture infection. Through a combination of in vitro and in vivo assays, we have identified several targets of CbsR12. Of these, we have confirmed that CbsR12 binds to and upregulates translation of carA transcripts coding for carbamoyl phosphate synthetase A; an enzyme that catalyzes the first step of pyrimidine biosynthesis. In addition, Cbsr12 binds and downregulates translation of metK transcripts coding for S-adenosyl methionine (SAM) synthase, an essential component of the methionine cycle. Furthermore, we have established that CbsR12 is necessary for full expansion of Coxiella-containing vacuoles (CCVs) and is linked to growth rate in a dose-dependent manner in the early phase of infection of Vero and THP-1 cell lines. This is the first characterization of a trans-acting sRNA of C. burnetii and the first description of a bacterial sRNA that regulates carA and metK expression. This study also illustrates the utility of transposon insertion mutants in elucidating Coxiella's sRNAs and the importance of sRNA regulation in establishment of the intracellular CCV niche.

Bartonella Bacilliformis HFQ Regulates Small RNAs

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Bartonella bacilliformis is a facultative bacterial intracellular pathogen of the human circulatory system, causing Carrion's disease. B. bacilliformis is transmitted between human

hosts by a sand-fly vector, two markedly distinct backgrounds. As such, it presumably requires a high degree of regulation at the genomic level. Hfq, a common RNA-binding protein of bacteria, is present in B. bacilliforms, and likely plays a key role in post-transcriptional regulation. Hfq is involved in regulating many processes by choreographing small RNA (sRNA) binding to mRNA targets for transcriptional regulation via specific base pairing interactions. To better understand the role of Hfq in B. bacilliformis, the hfq gene was cloned into an expression plasmid to generate a His-tagged Hfq fusion protein. The plasmid was then used to transform E. coli, which was then induced with IPTG, to produce high quantities of the Hfq protein. The cells were lysed and the Hfq protein was purified by affinity chromatography using a NiNTA column with His tag specificity. Recombinant Hfq was used to generate rabbit polyclonal anti-Hfq antibodies, and specificity was verified by western blots. The anti-Hfq antibody will be utilized to co-immunoprecipitate Hfq and its sRNA / mRNA targets from B. bacilliformis lysates. The resulting RNAs will then be used to construct a cDNA library, which in turn will be deep sequenced to generate a list of RNAs that interact with Hfq. Results will directly inform us of the genes that are regulated by Hfq in this potentially life-threatening pathogen.

Bioinformatic Analysis of the Flathead Lake Monster Bacteriophage

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The Flathead Lake Monster (FLM) bacteriophage was noted to have an abnormally-long tail upon its discovery. Once its genome was sequenced, this research sought out to identify the 117 FLM gene products using the BLASTp sequence alignment algorithm. This resulted in the discovery of five genes that are considered to be novel to the FLM. A specific gene within the FLM genome called the tape measure gene (TMG) was further analyzed once it was identified based on homology with other phages. Previous literature has suggested that a longer TMG can manifest itself as a longer bacteriophage tail length. This observation led to the hypothesis that a long tail length should yield a correspondingly-long TMG within the FLM. The bioinformatic investigation involved comparing the FLM tail length and FLM tape measure gene length to other phages. The results found that the FLM does not have an abnormally long TMG when compared to how long its tail is, indicating that the FLM is an anomaly when compared to other phages. Future examination of phage mosaicism may yield more information as to why the FLM tail length is abnormally long.

Cancer Risk Due to Cumulation Diagnostic Radiation

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The computed tomography (CT) scan is the best imaging tool in emergency situations. The use of CT scanning has increased drastically in the last few decades due to its helical capability. Advantages of helical CT scans include dramatically shorter examination times, improvement in vascular visibility, and enhancement of the outline of parenchymal organs. Helical scans also permit retrospective reconstruction of three-dimensional structures. Cancer risk related to ionizing radiation from CT scans increases as the cumulative radiation dose increases. While a typical CT scan delivers 10 millisieverts (mSv) of ionizing radiation, a single dose of 100 mSv can lead to solid organ cancer or leukemia. Because children have a higher cell turnover rate, they are more susceptible to developing radiation-induced cancers. We found that brain CT scans have similar carcinogenic potential in men and women, whereas, abdominopelvic CT scans are more likely to induce cancers in women. Body mass

index (BMI) also influences radiation-induced cancer risk, because a higher dose of radiation is needed to penetrate a more massive body.

Coordinated Regulation of Stem Cell Proliferation and Differentiation by RNA-Binding Proteins Cooperating with CCR4-Not Deadenylase Complex in *C. Elegans*

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Uncovering molecular mechanisms regulating the balance of stem cell proliferation and differentiation can help us understand the causative factors for ageing, cancer and various degenerative disorders. Pumilio and FBF (PUF) family RNA-binding proteins are highly conserved regulators of stem cell development. We are investigating how PUF proteins regulate proliferation and differentiation of stem cells using C. elegans as a model organism. FBF-1 and FBF-2, two PUF family proteins in C. elegans, are required for maintaining germline stem cells by translational repression of their target mRNAs (Crittenden et al., 2002). FBF-1 and FBF-2 are very similar in primary sequence and share target mRNAs (Prasad et al., 2016, Kershner et al., 2010 and Porter et al., 2018), but we found that they have differential effects on target mRNAs and maintained stem cell numbers. Our findings suggest that FBF-1 may destabilize target mRNAs while FBF-2 may protect targets from degradation; FBF-1 may slow stem cell proliferation and differentiation whereas FBF-2 may promote stem cell proliferation and differentiation in coordinate fashion. Additionally, we found that FBF-1 activity in stem cells requires CCR4-NOT deadenylase machinery that shortens poly (A) tail of mRNA leading to mRNA instability. By contrast, FBF-2 activity in stem cells shows less dependence on CCR4-NOT, which is related to FBF-2 interaction with its specific cofactor, DLC-1. We propose that FBF-1, but not FBF-2, interacts with CCR4-NOT complex in stem cells. We are going to test this hypothesis by performing proximity ligation assay. In conclusion, our study uncovered mechanisms of PUF proteins controlling stem cell proliferation and differentiation through differential cooperation with CCR4-NOT deadenylase machinery.

Detection of Tetracapsuloides Bryosalmonae in the Beaverhead River Using Environmental DNA Analysis

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Tetracapsuloides bryosalmonae is a Myxozoan parasite that causes proliferative kidney disease (PKD) in Salmonid fish. An outbreak of PKD in the Yellowstone River in 2016 resulted in a significant kill of mountain whitefish (*Prosopium williamsoni*); however, the exact reason for this outbreak remains unclear. Researchers first suspected a recent introduction of the parasite, but the analysis of samples collected from the Yellowstone prior to 2016 revealed that the parasite was present before to this event. This led to the hypothesis that environmental conditions may have exacerbated the effects of infection leading to increased mortality. To obtain more insight into the effects of environmental conditions it would be helpful to compare the spatial and temporal distribution of T. bryosalmonae in the

Yellowstone to other rivers where the parasite is known to occur. This comparison should include other freestone rivers with environmental conditions similar to the Yellowstone, as well as tailwaters with different parameters. We thought the Big Hole and Beaverhead rivers might be suitable for this purpose. The parasite was detected in the Big Hole in 2016, but there is no documented occurrence of the parasite in the Beaverhead. Therefore, we collected eDNA samples from five locations on the Beaverhead river in May, August and September and tested them by PCR using primers specific for T. bryosalmonae 18S rDNA. We detected parasite DNA at one of the five locations in two of the three samples collected from that location. To our knowledge, this is the first report of T. bryosalmonae in the Beaverhead River and it indicates that the Beaverhead may be suitable for a long-term study examining the spatial and temporal distribution of T. bryosalmonae. In addition, the positive samples will provide another source of parasite DNA from Montana that can be used in comparative genetic studies.

DOES AQUAPORIN 3B AFFECT THE NUMBER AND CHARACTERISTICS OF CALCIUM WAVES IN THE NEURAL PLATE OF AFRICAN CLAWED FROG EMBRYOS?

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Early in the development of the nervous system, vertebrate embryos undergo neural tube closure. During this process, the cells in the dorsal part of an embryo, the neural plate, constrict on their outward facing side (apical constriction) to form a tube. The Merzdorf lab has found that expression of the Aquaporin 3b (Aqp3b) protein in African clawed frog (*Xenopus laevis*) embryos is critical for neural tube closure, specifically for apical constriction of the cells of the neural plate. While aqp3b is only expressed in a well-defined line along the outer edge of each side of the neural plate, it affects a pan-neural plate process. Thus, the question my study attempts to answer is how Aqp3b signals to the rest of the neural plate. A likely candidate for the signal is calcium, a common intercellular and cellular signal. I hypothesize that the neural plate in embryos with inhibited Aqp3b expression will have fewer calcium waves and/or calcium waves with different characteristics. To test this hypothesis, the number and characteristics of calcium activity will be compared between control embryos and embryos that have been inhibited from expressing Aqp3b. This is accomplished by injecting a morpholino oligonucleotide, which inhibits Aqp3b expression, and GCaMP6, which is fluorescent in the presence of calcium, into frog embryos at the four-cell stage. I then collect time lapses of calcium activity that occurs during neural tube closure and analyze them for differences in the length, period, intensity, etc. of calcium signaling events using a variety of software. As my project continues, I will continue to collect data and begin to draw a conclusion on it.

EFFECTIVENESS OF TEETH CLEANING TREATS ON DOGS ORAL HYGIENE

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Domestic dogs can transfer bacteria from their mouth to the mouth of a human, the most common being bacteria that cause gingivitis and periodontitis. Even though most humans will not receive any medical consequences from this bacteria, problems may arise if that human has a reduced immune system. The purpose of this experiment was to determine the

effectiveness of oral hygiene treats in reducing bacterial growth in domestic dog mouths (*Canis lupus familiaris*). The saliva samples were taken from dogs at Associated Veterinary Services (AVS), the University of Providence (UP), and the Great Falls Animal Shelter (GFAS), located in Great Falls, Montana. Information such as age, breed, weight, health, food consumption, and oral care for each dog was recorded. There were 30 dogs sampled. A cheek swab was taken from each dog immediately before the canine received an oral hygiene treat and 5 minutes after treat administration. Canine saliva samples were placed on agar plates. Before the samples were placed on the agar plates, each agar plate was divided in half and labeled "before treat" and "after treat". The samples were then incubated for 7 days in a CO_2 incubator set at 37 ºC. After 3, 5, and 7 days, the plates were examined. The preliminary results cannot identify what kinds of bacteria were growing. On day 7, before the treat was given, $x\bar{I}$ = 9.8% growth (SD = 15.705). After the treat was given, $x\bar{I}$ = 10.023% growth (SD = 17.284). The results suggest that there is no difference in the amount of growth between the Before and the After (F = 0.250, P = 0.619, df = 1, 53). The results indicate the oral hygiene treats were ineffective in the short amount of time the treats were given, but further studies are necessary.

Effects of Familiarity on Mate Selection in Guppies

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Guppies (*Poecilia reticulata*) are freshwater fish commonly used in behavioral research. Originating from South America, this species can be purchased locally in the United States. We investigated the effect of familiarity on female mate selection. Our research consisted of individual female guppies each paired with two male guppies, from separate schools of fish, which were bred at MSUB. Preceding each experiment, one female was familiarized with one male in a divided fish tank. Uniform experiments were then conducted by placing a female guppy in a sectioned fish tank with the familiar male and a second unfamiliar male. Both males were placed on either side of the female. Following each trial, the individual female would be familiarized with the male that was not familiarized in the previous trial. Results were concluded by calculating time intervals that a female guppy spent near either male guppy or in the middle of the experimental fish tank. To record live behavior, with least amount of stress, a video recorder was used to record the guppies. To show effects of familiarization on female guppy choosiness, familiarizations were alternated monthly between a selected pair of males and one female. In these data there is a trial that suggests female mate preference may be influenced by familiarity.

Enhanced Reductive Defluorination of 6:2 Fluorotelomer Alcohol using Bio-Electrochemical Systems

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Current research on the biological degradation of poly- and perfluorinated alkyl substances (PFASs) is still challenged by two limitations:

1) Poor feasibility: successful biodegradation is limited to a small number of perfluoroalkyl acid precursors such as fluorotelomer alcohols (FTOHs);
2) Lack of means to precisely study the mechanism: conventional biological methods can

only create an anaerobic (vs. aerobic) environment, which cannot pinpoint whether the mechanism is oxidative or reductive.

This study uses Bio-Electrochemical Systems (BES) to precisely accomplish and enhance reductive PFAS degradation. BES is a platform technology where bacteria directly exchange electrons with solid electrodes to catalyze biochemical reactions. It has great potential for studying and enhancing PFAS degradation because it can:

1) Enhance bio-reactions by utilizing electric power;

2) Precisely separate oxidation and reduction reactions given the fuel-cell-like setup.

This study monitored the reductive defluorination of 6:2 fluorotelomer alcohol (6:2 FTOH) for over two months in BES. The results indicated that BES significantly increased (p-value<0.05) the rate of F- release under -0.55V poised potentials (vs. AgCl), and decreased the accumulation of 6:2 fluorotelomer carboxylic acid (6:2 FTCA), an intermediate degradation product. This study was the first time that reductive PFAS bio-degradation was precisely achieved, and the first report of successful BES-driven PFAS degradation. The findings provided the proof of concept needed to further explore whether enhanced defluorination can be accomplished with BES on more recalcitrant PFAS classes (e.g., perfluoroalkyl acids), and develop a new remediation technology.

EXAMINING THE RELATIONSHIP BETWEEN INJURY AND DOMINANCE IN THE ALPHA FEMALE WOLVES OF YELLOWSTONE NATIONAL PARK

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The social dynamics of wolf packs have been heavily studied, and nowhere as extensively as in Yellowstone National Park (YNP) since the reintroduction of gray wolves in 1995. The objective of this study is to gather data on the impact of injury of alpha female wolves in Yellowstone, specifically the effect of injury on their dominant status. Injury is a serious threat to wild wolves in the park, who may sustain several injuries during their average lifespan of 5 years. Female wolves may only have a few litters of pups in their lifetime, and the alpha female is the primary breeder in a pack. There have been multiple documented cases of alpha females sustaining injuries and subsequently losing their status to another related female. We collected observational data on four established wolf packs in the Northern Range of YNP, focusing primarily on wolves 907F and 969F, two sisters who alternated as dominant females of the Junction Butte Pack from 2017 to 2019. We also analyzed observational data on wolves 870F and 970F, a second pair of sisters who alternated as dominant females of the same pack from 2012 to 2015. The results of this project will add to our understanding of the long-term impacts of injury in alpha females on pack dynamics. Understanding this issue is a key component of wolf conservation in the Greater Yellowstone Ecosystem.

GRAM-POSITIVE BACTERIAL INHIBITION BY A POTENTIAL ANTIMICROBIAL SMALL MOLECULE

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With the advent of antimicrobial molecules in the mid nineteenth century countless people were saved from life threatening infections. With decades of use, antimicrobials put enormous selective pressure on targeted pathogens resulting in resistances forming in many common microbes. These resistant strains of pathogens result in over two million illnesses,

23,000 deaths every year, and billions of dollars in medical expenses. This represents a need to research new molecules that have antimicrobial properties to combat the rise of resistance and prevent undue suffering to those afflicted. A small molecule is under investigation now that displays promising antimicrobial properties in preliminary studies against Candida species and many pathogenic gram-positive bacteria. The gram-positive bacteria that are inhibited by this small molecule at include Staphylococcus aureus, Streptococcus pneumonia, Listeria monocytogenes, Staphylococcus epidermidis, Streptococcus pyogenes, Bacillus cereus and Clostridium difficile. Research is currently focused on S. aureus and C. difficile with disk assays performed on both and plate assays performed on S. aureus to narrow down the minimum inhibitory concentration. C. difficile has a zone of inhibition of 9mm, while S. aureus has zone of inhibition at 10mm. The plate assay for S. aureus has shown a minimum inhibitory concentration to be between 150-100 uM of the small molecule, though further studies are needed. Plans are being made to find the mechanism of action of the small molecule by using mutant strains of S. aureus.

INVESTIGATING THE BINDING OF BH3I-1 DERIVATIVES TO ANTI-APOPTOTIC BCL-2 PROTEINS

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Candida albicans is a leading cause of superficial and life threatening fungal disease. The ability to grow as filamentous cells is an important virulence trait for this fungus. Thus, molecules that block filamentous growth may form the basis for a novel approach to controlling infections by *C. albicans*. We have characterized a small molecule called BH3I-1 that specifically inhibits filamentous growth, but not yeast-form growth. BH3I-1 is not likely to be a good candidate for an anti-fungal drug, as it targets human Bcl-2 proteins and has the potential to induce apoptosis in human cells. The goal of our project is to identify structural derivatives of BH3I-1 that retain anti-fungal activity, but not Bcl-2 binding activity.

KNOCKOUT OF ELP3 GENE IN CANDIDA ALBICANS

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Familial Dysautonomia (FD) is a devastating neurodegenerative childhood disease characterized by diminished polarized growth of autonomic neurons. FD results from a mutation in the ELP1 gene and reduced levels of the corresponding protein ELP1, a scaffolding protein that assembles a multi-subunit complex called Elongator. Elongator functions in the modification of tRNAs that mediate efficient translation of AA- and AG-ending codons. The Elongator complex also includes the ELP3 protein, a catalytic subunit of the complex. We are using hyphal growth in the fungus Candida albicans to model the role of Elongator in polarized growth. In this study C. albicans was genetically modified to knockout the ELP3 gene. As with the ELP1 gene, the absence of the ELP3 is expected to eliminate Elongator function. We have found that knocking out ELP3 mitigates the polarized growth of hyphal filaments under certain growth conditions.

Legacy Effects of Abandoned Insect Ecosystem Engineering Structures on Montana Stream Hydraulics

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Habitat modifications from ecosystem engineering can have profound legacy effects on ecological processes and communities. Our research identifies a hydraulic effect stemming from net-spinning caddisfly (*Hydropsychidae*) retreat structures that are ubiquitous in Montana stream ecosystems and describes the longevity of this effect over ecologically relevant timescales. We used a laboratory experiment to investigate how caddisfly net and retreat structures built in Montana streams influence fluid dynamics at local spatial scales over a two month time period after simulating abandonment by removing the caddisly larvae, leaving the retreat structure intact. We made velocity measurements with accoustic doppler velocimetery around caddisfly silk structures to test how hydropsychid caddisflies influence flow velocity and if any changes to flow velocity are maintained after the structure is abandoned by its caddisfly. We found that caddisfly silk nets reduce flow downstream of the structure by 85% and upstream of the structure by 17%. We also found that caddisfly silk structures without their caddisfly present can persist for over 60 days, suggesting ecologically significant legacy effects of these biotic structures on near bed hydraulics. The legacy of these local changes to hydrology may provide important refugia for less flow-tolerant benthic macroinvertebrate taxa and especially to those with rapid life histories and high turnover rates. Future work could address variation in the magnitude and duration in biotic engineering effects among different silk-producing species, densities of the structures through space or time, and decay rates of the silk structures at different flows that span those observed in Montana streams.

Natural Infection in Honey Bee Hemocytes

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Honey bees (*Apis* spp.) act as the primary pollinators of plants including fruit, nut, and vegetable crops. Since 2006, however, viral and other pathogens have caused honey bee colony losses averaging 33% annually (Lee et al, 2015). In an effort to better understand how the immune system of honey bees works, especially honey bee antiviral responses, I am investigating natural infection in honey bee larvae and in isolated hemocytes, which are macrophage-like immune cells. It was discovered that fruit fly hemocytes remained uninfected in virus-infected larvae, since hemocytes are important in mediating antiviral responses (Tassetto et al., 2017). The goal of this research project was to determine if these cells may also remain free of viruses in otherwise infected honey bee larvae and, thus, play an important role in honey bee antiviral defense. RNA was extracted separately from larval carcasses and hemocytes and cDNA was made, which was then screened for eight different honey bee viruses to determine the presence of viruses in the hemocytes and in the cells that make up the carcass. I found that in naturally infected larvae, hemocytes are generally uninfected. Of 39 larvae, 13 had natural infections. Three of the eight viruses were responsible for these infections. Of these 13 infected larvae, the hemocytes of only one larva was infected with the same virus that had infected the larva. Further experimentation will include testing hemocytes and carcasses from a larger sample of naturally infected larvae and from adult honey bees that will be injected with flock house virus. This will help determine if adult honey bee hemocytes are similarly resistant to viruses as larval hemocytes. Should hemocytes remain uninfected in otherwise infected larvae or adult bees, they may be a useful model for studying infection and the resulting antiviral response in honey bees at a cellular level.

Study of an Atomic Compressive Material Behavior at High Strain Rate Compression in a Simple Aluminum System

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Aluminum alloys are one of the structural materials with novel properties for building functional parts. Aluminum alloys are light metal alloys which have high demand in aerospace and automotive industries. In structural applications, functional parts are designed for over a broad range of strain rates and temperatures. A numerical simulation was performed to characterize an aluminum (Al) system at high strain rate uniaxial compressive loading conditions. We deformed the Al system at room temperature to higher temperatures with zero pressure. Al has face-centered cubic structure and we use four thousand atoms in periodic boundary conditions to perform the analysis. We used a modified embedded atom method, a widely used atomic level semi-empirical model for metals and impurities, for many-body interatomic potentials for monoatomic metal. We found characteristic material behavior for the Al system at different temperature at high strain rate compressive loading.

Studying Codon Bias and Kidney Dysfunction in a Mouse Model for Familial Dysautonomia (Poster)

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Familial dysautonomia (FD) is a debilitating disease primarily known for its damage to the peripheral nervous system. However, kidney failure is the most common cause of death in FD patients. FD results from a mutation in the ELP1 gene, which is part of the 6-subunit complex, Elongator. It is known that some genes preferentially use specific synonymous codons, and Elongator is essential for translating genes that are enriched in either AA- or AG-ending synonymous codons. It has been assumed that kidney disease in FD results from irregular blood pressure and compromised innervation of kidney vasculature. However, here we show that ELP1 is robustly expressed in the kidney collecting duct, suggesting that it may play a direct role in kidney function and therefore the dysfunction in FD. To investigate this hypothesis, we made a mouse model where ELP1 is selectively ablated in the kidney collecting duct. Our data indicate an essential role for ELP1 in normal kidney function. These data suggest that compromised ELP1 levels in the kidneys of FD patients may be a contributing factor to chronic kidney disease. Our ongoing work focuses on identifying kidney-specific, codon-biased genes that are misregulated in the absence of Elongator and thus contribute to kidney dysfunction.

"That's Not Coming Off of There"; An Exploration of Ligand Lability in Carbon Tetrachloride Dechlorination Technologies

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Carbon Tetrachloride (CT) is a carcinogenic industrial solvent, and known environmental contaminant. The Department of Energy's Hanford Site has been identified as a Superfund

Site. CT, is just one of the many environmental concerns. Environmentally CT dechlorinates in a stepwise manner forming lesser chlorinated intermediate complexes that pose a health threat to humans. [Cu(PDTC)L] is a small coordination compound capable of dechlorinating CT into less harmless decomposition products: CO_2 and Cl^-. In this study we draw a correlation between the dechlorination kinetics of CT of [Cu(PDTC)L] and the ligand liability of the L ligand. We use P K-edge X-Ray Absorption Spectroscopy to quanititate the phosphorus covalency of a series of PR_3 transition metal ligands [Cu(PDTC)L] (L= PPh_3, and PCy_3). Our results show that there is no correlation between the covalent character of Cu-P bonds and the kinetics of dechlorination. Instead, we propose that the correlation has a greater correlation with the solubility of a ligand in a given solvent system.

The Effects of Surfactants in Glyphosate-Based Herbicides on the Spotted Salamander

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Salamander populations have been declining worldwide. There are many reasons that are thought to be contributing to this decline, one being the use of agricultural herbicides. Glyphosate-based herbicides are one of the most commonly used herbicides worldwide and their use is continuing to increase. Originally it was thought that glyphosate was the toxic ingredient causing declines in amphibian populations; however, studies are indicating that the surfactants used in these herbicides are what is responsible for the toxicity. The purpose of this study is to evaluate the toxicity (48 h) of a silicon-based surfactant on the spotted salamander (*Ambystoma maculatum*) compared to the surfactant in Glystar Plus®. The two experimental groups will be exposed to a solution containing 41% glyphosate with varying concentrations of a silicon based non-ionic surfactant at 14% and 28% respectively at an application rate of 0.35 mL/m2 added to Glystar Original®. Toxicity will be evaluated by comparing length, girth, weight, liver somatic index, and physical appearances.

The Influence of Antimicrobial Peptides, Cutaneous Microbial Communities and Water Quality on the Susceptibility of Columbia Spotted Frogs To Chytridiomycosis

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Chytridiomycosis is a potentially deadly skin disease found in amphibian populations all over the world. Chytridiomycosis is caused by the fungus *Batrachochytrium dendrobatidis* (Bd) that grows on the skin of amphibians. The fungus can cause a disruption in the homeostatic functions of the amphibian skin and can lead to death. The relationship between an amphibian's antimicrobial skin secretions, cutaneous microbial communities and environmental water quality can affect their resistance and susceptibility to Bd infection.

Columbia spotted frogs (*Lithobates luteiventris*) are native to western Montana and have been affected by chytridiomycosis. Three lakes known to be positive for Bd were used as collection sites where a total of ten frogs were caught on three different occasions throughout the summer for a total of 90 collected samples. Each frog was swabbed for bacterial and fungal samples and placed in a solution that would collect antimicrobial peptides secreted from the frogs' skin. Water samples were collected at each site and analyses were performed to assess various parameters. Early results suggest that AMPs may affect the relative abundance of an important anti-fungal bacteria (*Rhizobacta*) which influenced Bd status. The goal of the ongoing research is to determine what parameters of environmental conditions, antimicrobial skin secretions, and cutaneous microbial communities allow Columbia spotted frogs to be resistant to chytridiomycosis in order to better predict and treat future outbreaks.

The Relationship Between Deuterium Excess and Uranium Groundwater Concentrations in Whitehall, Montana

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Uranium can become elevated in the environment and pose a human health risk to water resources. Therefore, it is important to understand the origin, transport and concentration of uranium in the environment. With this study we propose that deuterium ($\delta(_1^2)H$) excess may be a viable way to understand uranium groundwater contamination that is mainly derived from the surface. We collected 23 water samples and compiled data from a study done by the United States Geological Survey (USGS) within the area of Whitehall, MT. With a linear regression we show that there is a significant relationship between deuterium excess and uranium water concentrations for our data ($t_{19}= -3.015, p=0.0071, R^2=0.32$). We also showed that there is a significant linear relationship between uranium water concentrations and nitrate water concentrations for our data ($t_{19}= 2.573, p=0.0192, R^2=0.27$). In addition we showed that there is a significant linear relationship between calcium carbonate and uranium groundwater concentrations for the compiled USGS data ($t_{66}= 6.295, p=2.846e^{(-8)}, R^2=0.38$). The results of a Wilcoxon rank sum test showed that wells close to ponds have a significantly higher median uranium concentration for our data added to the USGS data ($WRS_{8,12}, W=111.5, p=0.033778$). With this relationship we may infer that an influx of uranium into the aquifer originates at the surface most likely from the Boulder Batholith. These results are also consistent with a model that involves insoluble uranium being transported in suspension and settling out in ponds where an influx of nitrates and/or carbonates react to produce higher concentrations of water-soluble forms of uranium. We suggest that the relationship found between uranium concentration and deuterium excess in water samples may be related to the higher rates of evaporation in ponds. Thus, in this specific hydrologic environment of valley ponds close to the Boulder Batholith source, deuterium excess may serve to help predict levels of uranium concentration.

XFEB, a Direct Target of ZIC1, is Involved in Neural Crest Development

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During early embryonic development, neural crest cells give rise to the peripheral nervous system, melanocytes, bone and craniofacial cartilage. A network of signaling and transcription factors regulate early neural crest development, including Zic1, Pax3, Gbx2, and Xfeb.

Combinations of Zic1 plus Pax3 and Gbx2 plus Pax3 are able to induce ectopic neural crest development. We hypothesized that Xfeb also contributes to neural crest development, as it is present in the same region at the correct time. Besides being a direct downstream target of the transcription factor Zic1, Xfeb was also identified as a potential neural crest gene induced by Zic1in genomic screens. We hypothesize that pax3, Xfeb, gbx2 and zic1 are all part of a gene regulatory network controlling neural crest development. To investigate these relationships, we overexpressed the Xfeb gene using Xfeb sense RNA and inhibited Xfeb expression with morpholino oligonucleotides (MO). We used in situ hybridization to visualize neural crest induction by staining for slug RNA expression, a known neural crest marker. Our results show that embryos injected with Xfeb sense RNA expanded slug expression while those injected with Xfeb MO diminished slug expression. In further experiments, we injected embryos with pax3 sense RNA without and with Xfeb MO. Injection with pax3 sense RNA alone expanded slug expression, while embryos injected with pax3 sense RNA plus Xfeb MO showed a decrease in slug expression. This suggests that Xfeb acts downstream of Pax3 in the neural crest gene regulation network. Our next step will be to determine if upregulation of gbx2 or zic1 will rescue neural crest development in the absence of Xfeb. This research will contribute to our understanding of gene regulatory networks, and how these contribute to early neural crest development.

Zic Transcription Factors that Influence Convergent Extension

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My research is focused on a specific time frame during embryonic development called gastrulation, in which intricate cell movements give rise to the ectoderm, mesoderm and endoderm layers of the embryo. The mesoderm layer undergoes movements called convergent extension, which allow cells to intercalate and move inside the embryo. The Zic family of zinc-finger proteins have a crucial role in gastrulation and neurulation. Based on findings from our lab (K. See and Merzdorf, in preparation) and others (Cast et al., 2012), I hypothesize that zic genes are required for convergent extension during gastrulation. Since there are 5 different, but very similar, zic genes, I will test each of these genes for a role in convergent extension during gastrulation. A knockdown method for each of the 5 zic genes in embryos of the model organism *Xenopus laevis* will be combined with a method called Keller Explants to allow analysis of convergent extension. In my experiments, I have ruled out zic 2 as a regulator of convergent extension. My current hypothesis is that zic3 is the most likely candidate because of its known roles in neural tube formation and other reasons I will explain below. I am also analyzing zic1,4, and 5 for convergent extension regulation.

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CONTENTS

ARTICLES

MEETING ABSTRACTS