Prey diversity and season mediate trophic overlap between cougars and wolves in anthropogenic ecosystems

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> WDFW Commission Meeting Virtual Friday, December 9, 2022, 1:40 – 2:40 pm

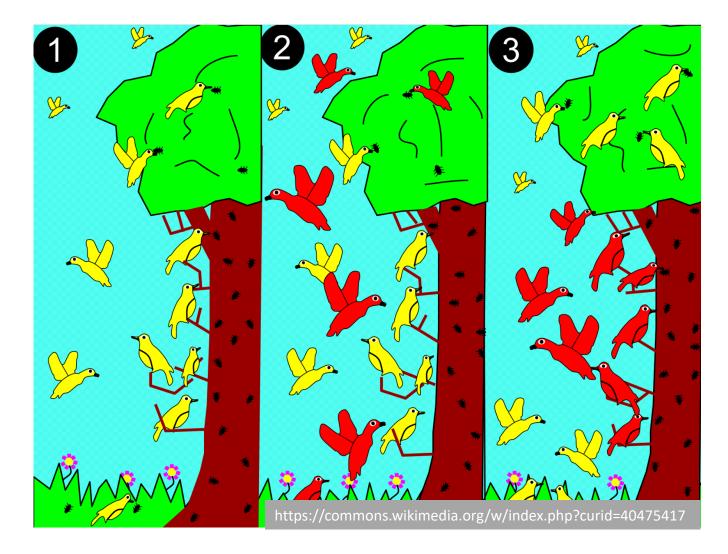
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## Background

- Competitive Exclusion Principle: two species with identical niches cannot coexist indefinitely
- Competition, if present, drives niche partitioning



- Most studies on trophic cascades are single predator/single prey
- Most ignore predator-predator interactions
- Predator-predator interactions may mediate trophic impacts on prey
- Multi-predator landscapes influence prey in divergent ways
- Fewer studies from managed landscapes

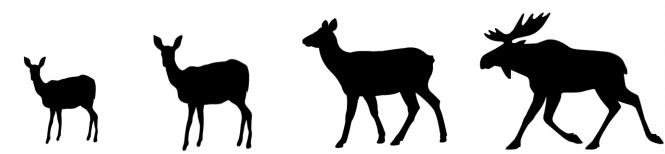
## Background





(Thurber and Peterson 1993, Ripple and Beschta 2003, Ritchie et al. 2012, Hughes et al. 2013)

- Wolves are recolonizing WA after an 80-year absence
- Timeline:
  - **Pre-1900s:** Co-occurrence of cougars and wolves in Washington
  - 1930's: Wolves extirpated by settlers
  - Late 1900's: Cougars remain as sole top predator
  - 2008: Confirmed first wolf breeding pack in 80 years
  - 2021: At least 206 wolves, 33 known packs, 19 successful breeding pairs
- How will the co-occurrence of cougars and wolves and affect Washington's ungulate prey populations?
- White-tailed deer, mule deer, elk, moose





### Introduction



Goal: Investigate indicators of dietary competition between cougars (*Puma concolor*) and wolves (*Canis lupus*)

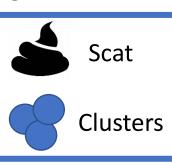
### Objectives:

- Fully characterize diets of wolves and cougars
- Compare dietary overlap between existing cougars and recolonizing wolves
- Explore dietary differences between study areas and season

#### Data:

- Scat metabarcoding (genetic diet analysis)
- GPS cluster investigations





## Introduction

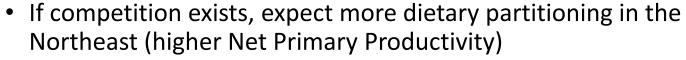
### Hypothesis:

• Increased prey diversity provides greater opportunity for dietary partitioning



### **Predictions:**







• If competition exists, expect more dietary partitioning in summer vs winter (more prey species available in summer)

## Introduction



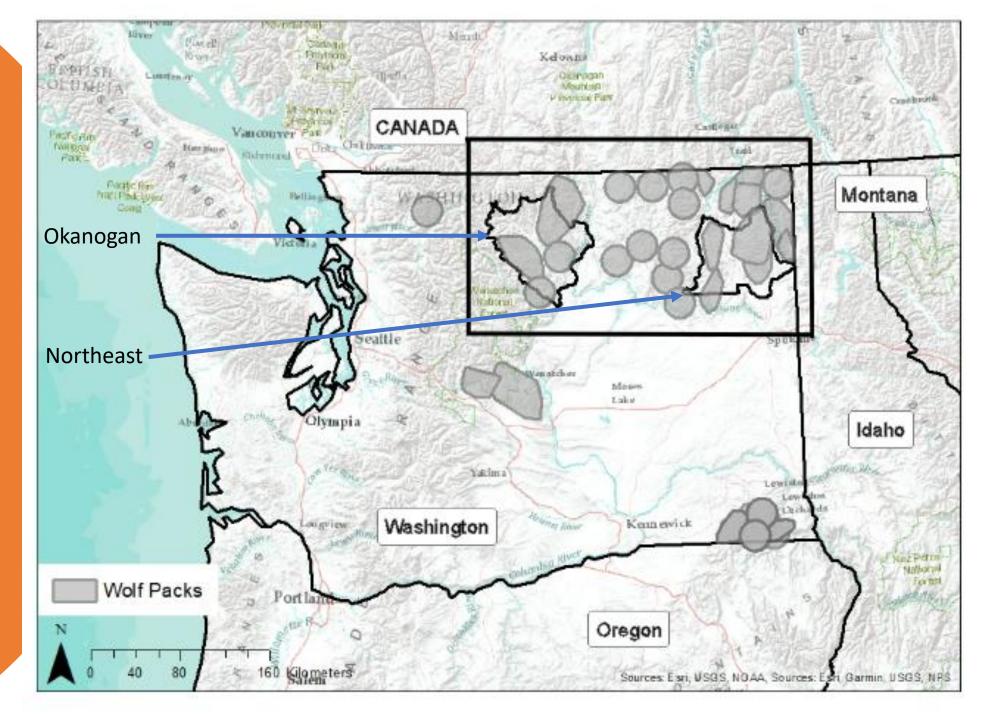
Field Methods







# Field Methods



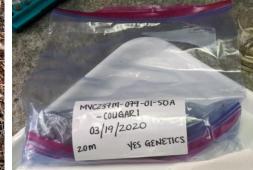












- Cougars and wolves GPS-collared as part of the collaborative Washington Predator-Prey Project
  - 60 cougars (NE: 34, OK: 26)
  - 16 wolves (NE: 14 (4 packs), OK: 5 (2 packs)



- Summer: June 1 December 14
- Winter: December 15 May 31



- At GPS clusters with carcasses
- At GPS clusters without carcasses (i.e., bedsites)
- Opportunistically while hiking to and from GPS cluster sites

Collected whole, stored frozen and double-bagged





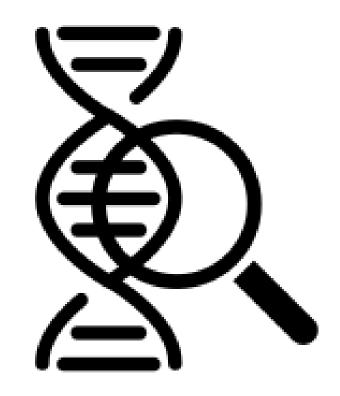


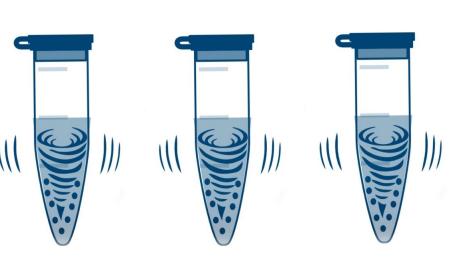


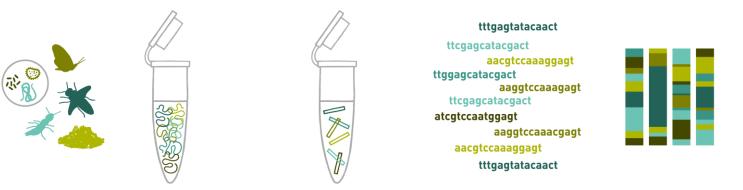
Lab Methods











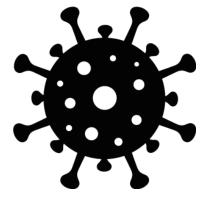
- Why use metabarcoding?
  - Bulk sequencing of multiple species simultaneously
  - Sensitive method detects rare species
  - Works well on new <u>and</u> old scats
  - Ability to identify depositor species *along with* prey species
- Our analysis:
  - 12S region of the vertebrate mitochondrial genome
  - Three replicate samples
  - Follow-up PCR analysis for *Canis* spp. and *Odocoileus* spp.





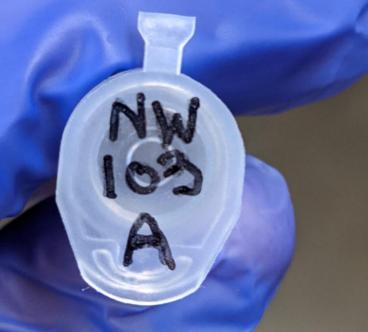
## Lab Methods

- Trained UW undergraduate student in genetic lab methods
- Tubed scats during summer 2021
- Many COVID protocols



- Lab preparation:
  - Thawed scats overnight
  - Selected three scat "subsections"
  - Broke scats subsections open
  - Sampled a lentil-sized piece from the middle of each subsection
  - Three subsamples per centrifuge tube
  - Collected a main (A) and backup
     (B) set of tubed samples





## Lab Methods

### Metabarcoding:

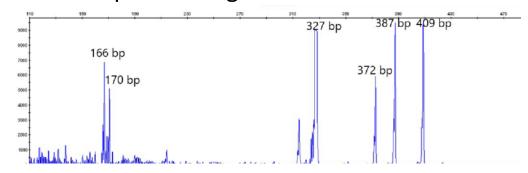
- Levi Genetics Lab Oregon State University
- Removed results where the relative read abundance (RRA) was
   <0.5% of sample</li>
- Required 2 of 3 sample replicates to be successful to include sample results
- Matched species using GenBank (https://www.ncbi.nlm.nih.gov/genbank/)

Analysis to differentiate *Odocoileus virginianus* from *O. hemionus*:

- Collaboration with UW SEFS Genetics Lab to develop primer set
- Primers adapted from previous work (Latch et al. 2008, Lindsay and Belant 2007)
- Primer set also tests for elk (*Cervus canadensis*) and moose (*Alces alces*)
- Combined species and sex ID primers together

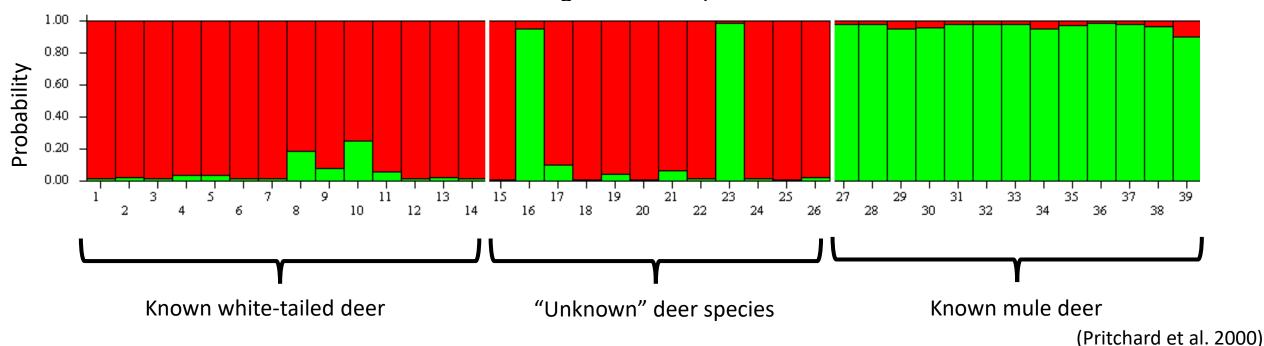
### • Process:

- DNA extraction
- Polymerase Chain Reaction (PCR)
- Fragment analysis

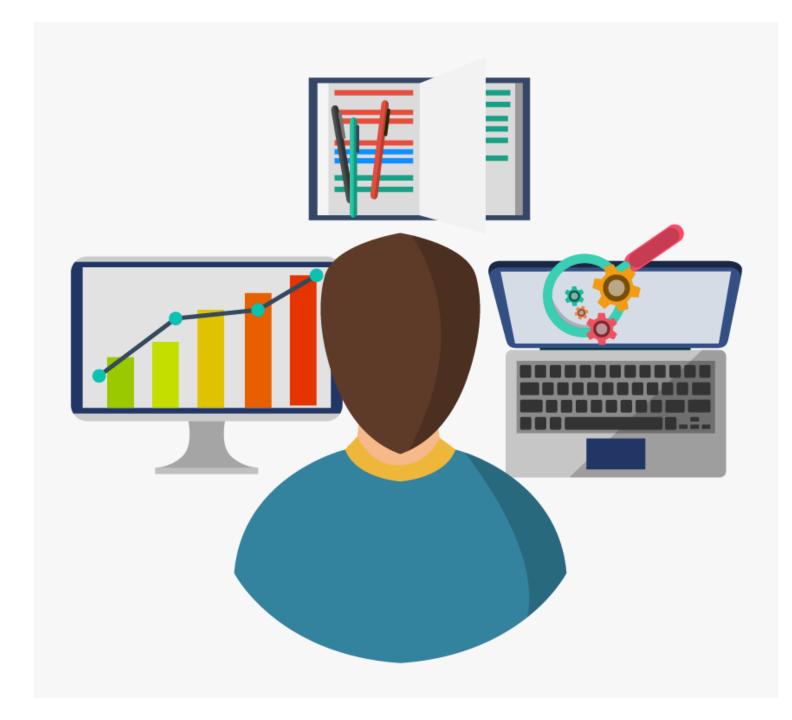


# Cervid Primers – Odocoileus spp. Analysis

- Used Bayesian program Structure to differentiate between unknown target species
- Method showed strong differentiation between Odocoileus spp.
- Differentiation of sex also worked in the genetic multiplex



Statistical Methods



# Statistical Methods

- Analyses
  - Percent frequency of occurrence (%FO) plots (primary prey by carnivore, study area, and season)
  - Pianka's Niche Overlap Index (dietary overlap index prediction testing)
  - Shannon's Diversity Index (species diversity index hypothesis testing)



# Statistical Methods

- % Frequency of Occurrence (%FO): (events / total events) x 100%
- Pianka's Index (range 0 − 1)
  - Metric of dietary overlap between two species or groups j, k
  - O<sub>jk</sub> = Pianka's Index value for dietary overlap between depositor species or groups j and k

$$O_{jk} = \frac{\sum_{i}^{n} P_{ij} \times P_{ik}}{\sqrt{\sum_{i}^{n} P_{ij}^{2} \times \sum_{i}^{n} P_{ik}^{2}}}$$

- $P_{ii}$  = proportion of prey species *i* in the diet of depositor *j*
- $P_{ik}$  = proportion of prey species *i* in the diet of depositor *k*
- Shannon's Diversity Index (H) (range 0 − 1)
  - Metric of dietary diversity within depositor species or groups
  - Related to weighted geometric mean of the proportional abundances of the types

• H = Shannon index value
$$H = -\sum_{i=1}^{n} p_i \ln(p_i)$$

- p<sub>i</sub> = proportion of a prey item found for *i*th prey species
- s = number of prey species categories (Pianka 1973, 1974, Shannon 1948)

# Statistical Methods

### Simplified prey categories for analysis:

- Ungulates
  - White-tailed deer (*Odocoileus virginianus*)
  - Mule deer (*Odocoileus hemionus*)
  - Elk (Cervis canadensis)
  - Moose (Alces alces)
  - Deer Unknown Species
    - randomly assigned to white-tailed deer or mule deer by proportion of known samples in group (carnivore / study area / season)
- Non-Ungulates
  - Bird
  - Carnivore
  - Lagomorph
  - Small mammal
  - Medium mammal
  - Other
- Bootstrapped output dataset 10,000 to calculate results and generate 95% confidence intervals







Results



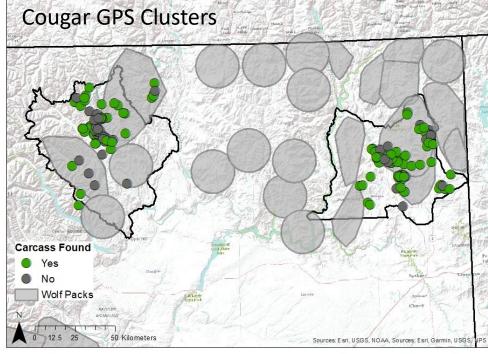
# Scat Collection - Sample Sizes

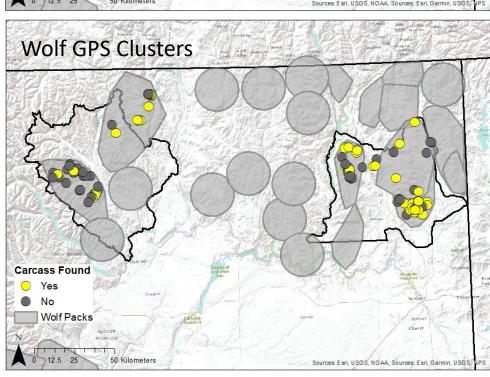
		Cougars			Wolves		
Study Area	Season	Submitted	Amplified	Contained Prey	Submitted	Amplified	Contained Prey
Northeast	Summer	50	41	39	55	56	49
Northeast	Winter	50	40	40	52	51	44
Okanogan	Summer	46	39	37	60	47	38
Okanogan	Winter	55	54	46	32	27	23
	Totals	201	174	162	199	181	154

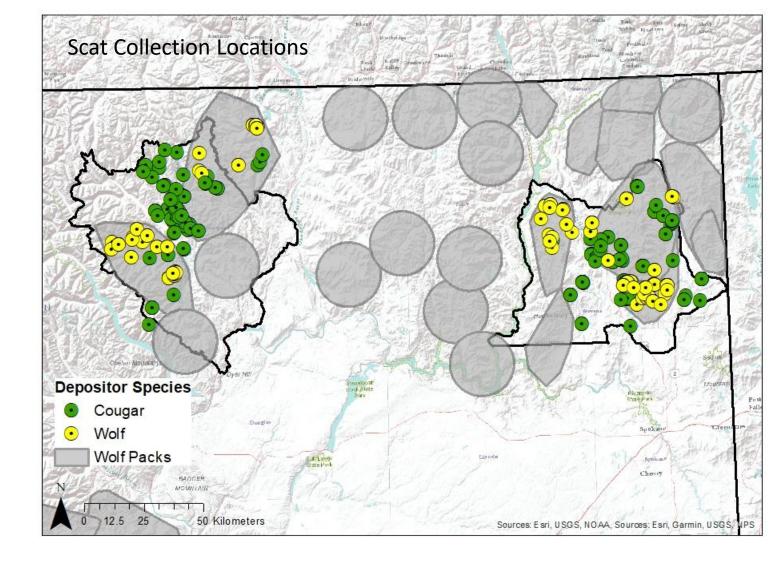
# Scat Collection – Location Types

Species	Feeding Site (GPS Cluster)	Bedsite (GPS Cluster)	Opportunistic	Totals
Cougar	105 (64.8%)	39 (24.1%)	18 (11.1%)	162
Wolf	80 (51.9%)	60 (39.0%)	14 (9.1%)	154
Totals	185 (58.5%)	99 (31.3%)	32 (10.1%)	316

- Scats also collected at carcasses sites may represent carcass item plus any prey eaten immediately prior
- Scats collected at bedsites also allow a wealth of information
- Scats collected opportunistically more similar to traditional scat collection methods







- 400 scats collected total
- Samples collection somewhat reflects wolf and cougar collar data





Scat

Species
Detected
(Cougars)



Cougar\* Livestock Porcupine Moose Mule deer White-tailed deer Deer – Unknown Species

Elk

Beaver Raccoon
Coyote Snowshoe hare
Grouse Unknown Canid
Turkey Unknown Bird
Small Mammal

Bobcat Domestic Cat Shrew Squirrel





Scat

Species
Detected
(Wolves)



Unknown Canid Domestic Dog Dumpsite Magpie Marmot Skunk Elk Moose Mule deer White-tailed deer Deer – Unknown Species

Grouse
Snowshoe hare
Turkey
Livestock

Beaver
Other Bird
Black Bear
Bobcat
Chipmunk
Fish
Flying squirrel
Vole

### Predictions:



If competition exists, expect more dietary partitioning in the Northeast



If competition exists, expect more dietary partitioning in summer vs winter

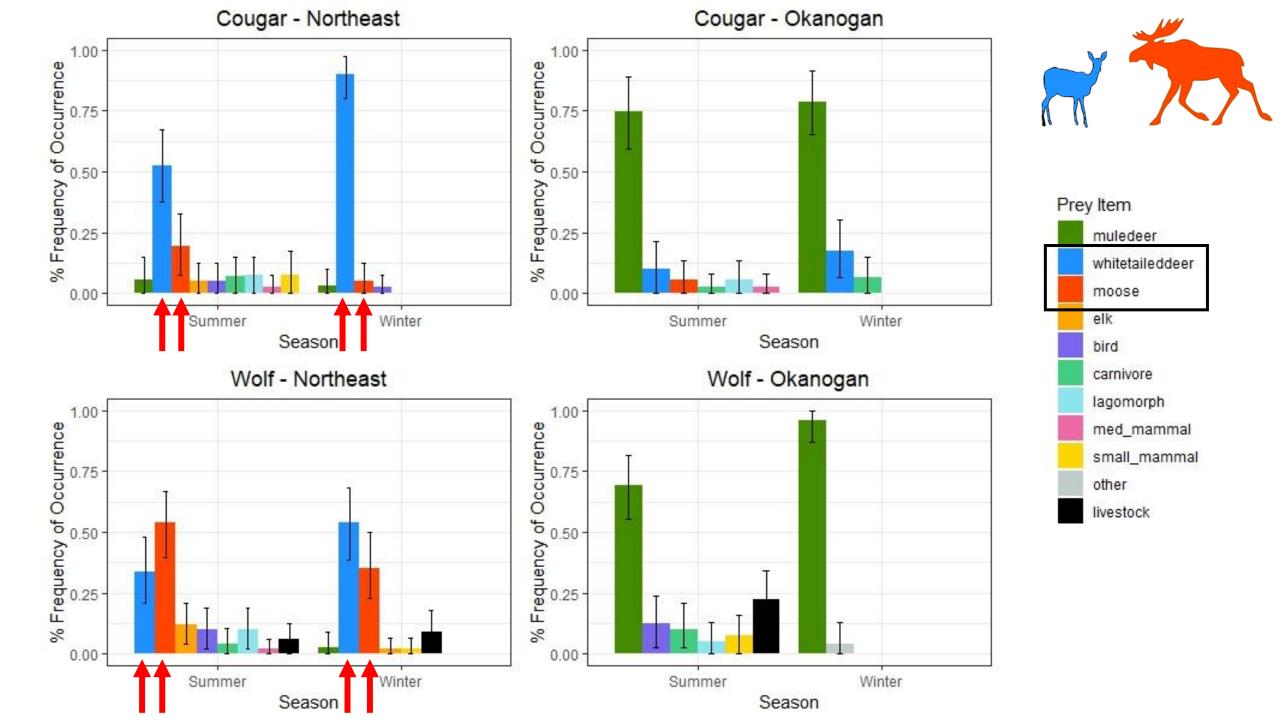
## Results

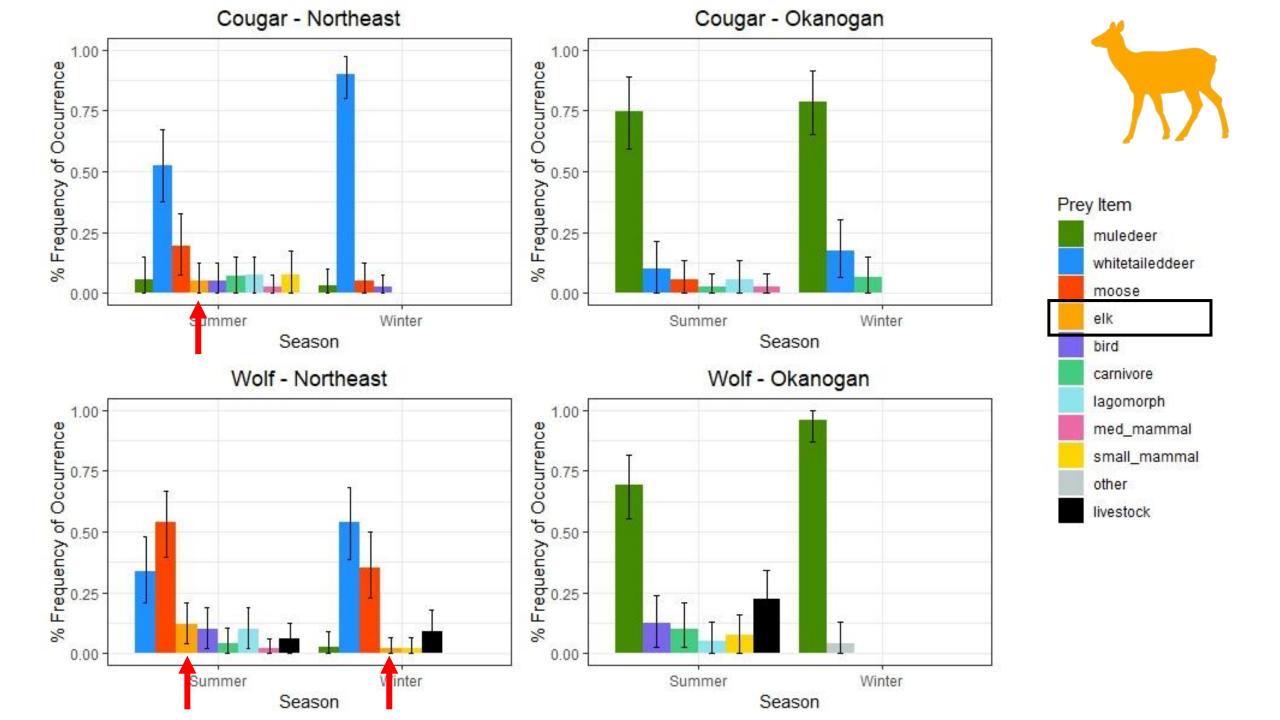


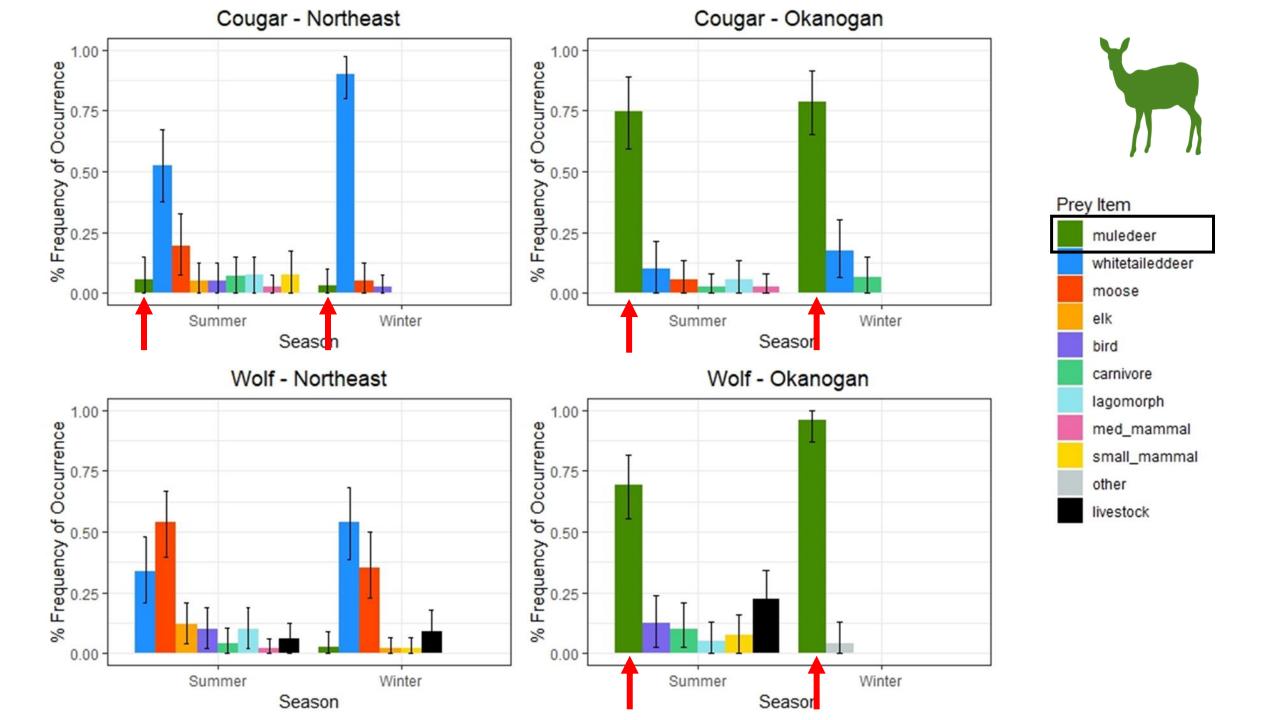


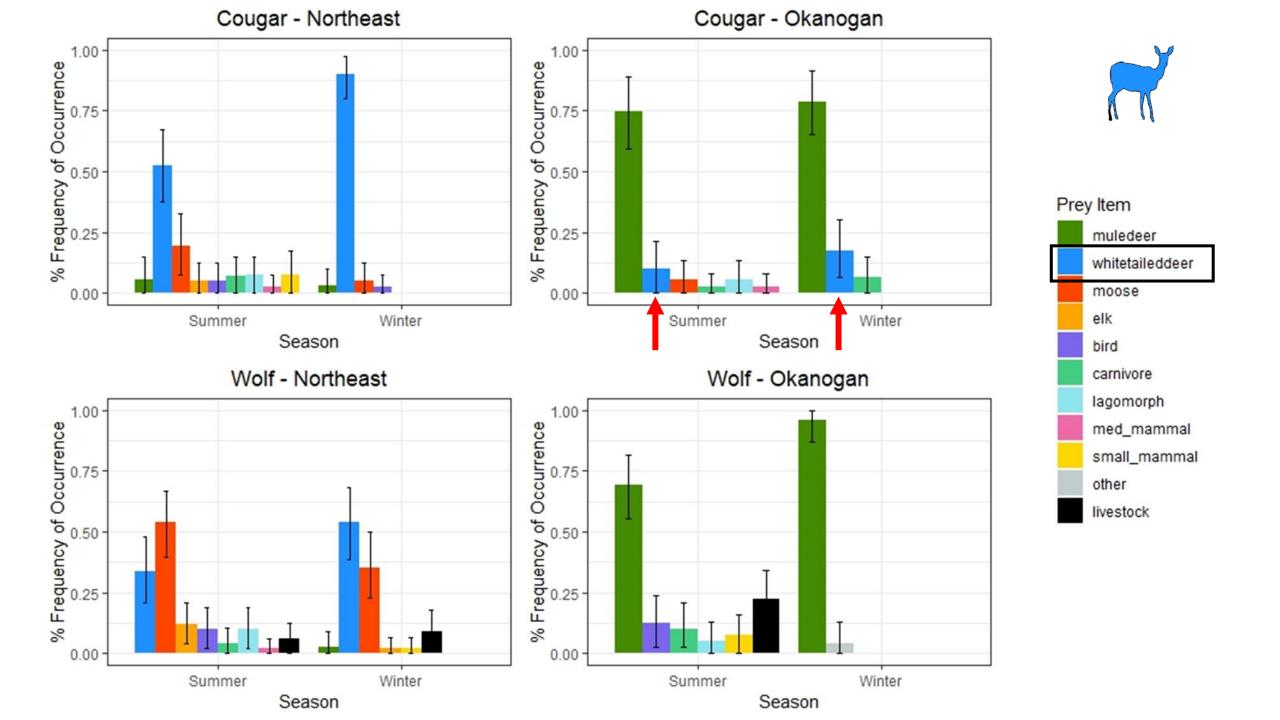


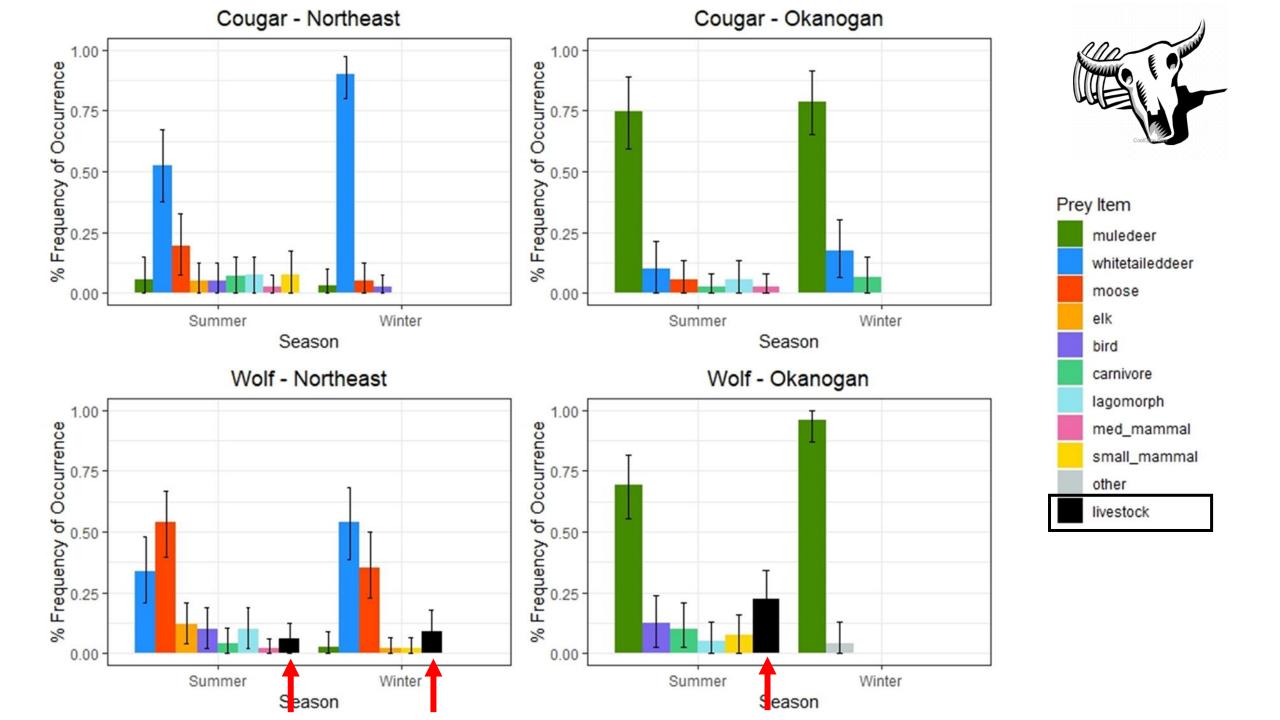




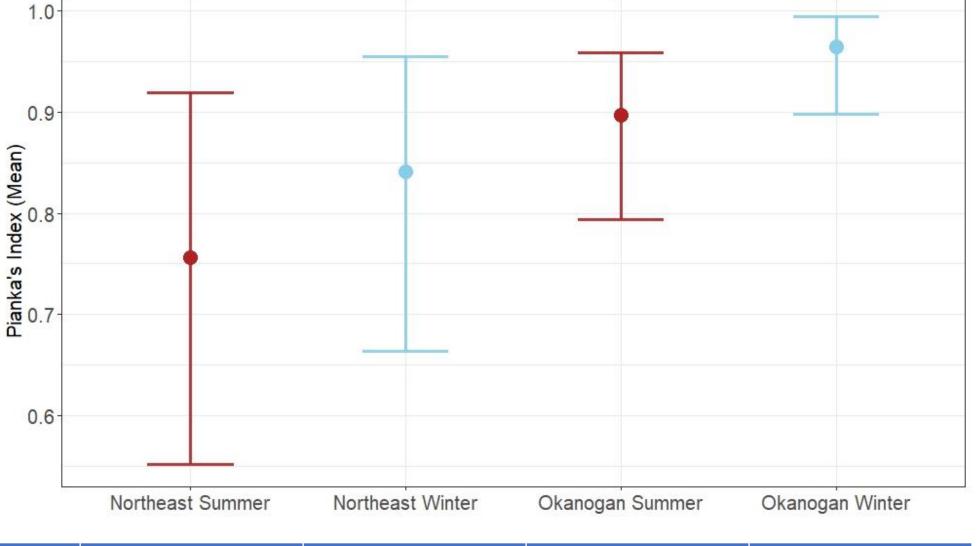








# Pianka's Index



Pianka's Index	Northeast Summer	Northeast Winter	Okanogan Summer	Okanogan Winter
Mean (95% CI)	0.756 (0.552, 0.919)	0.841 (0.664, 0.955)	0.897 (0.794, 0.958)	0.965 (0.899, 0.994)

### Hypothesis:

Increased prey diversity provides greater opportunity for dietary partitioning

### **Predictions:**



 If competition exists, expect more dietary partitioning in the Northeast



 If competition exists, expect more dietary partitioning in summer vs winter

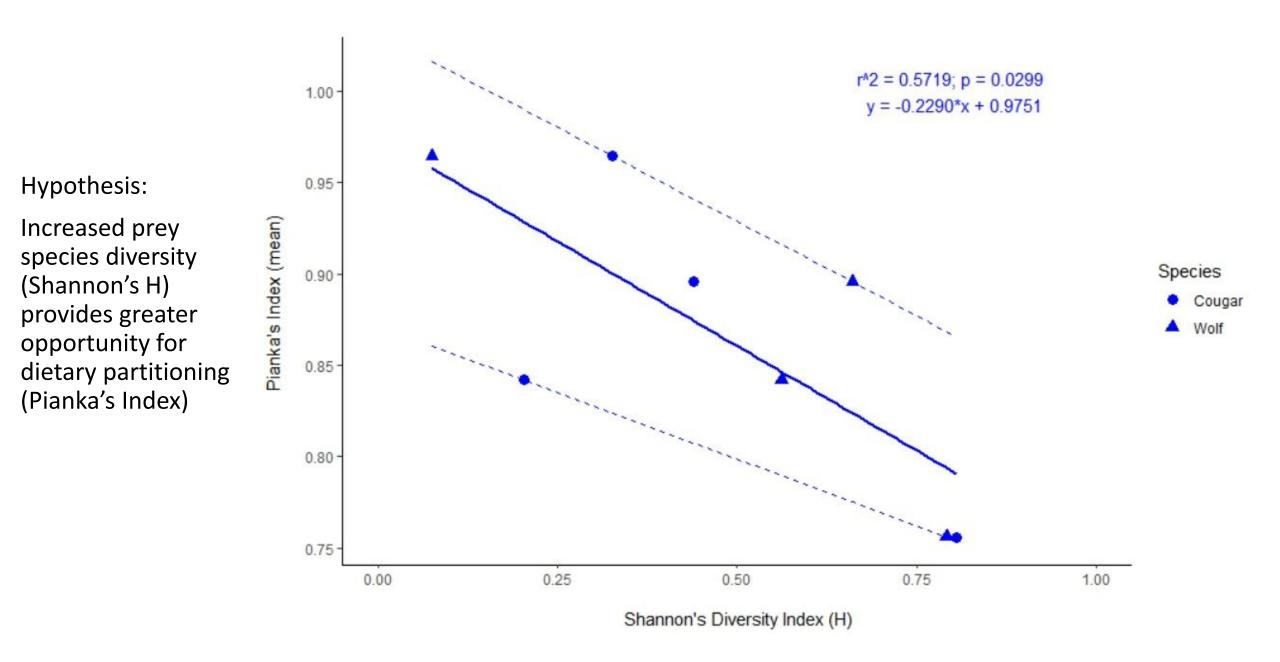
### Results











## Conclusions

- Overview
  - → Prey diversity mediates dietary overlap between wolves and cougars
  - → Season may also play a role in mediating dietary overlap
  - → Areas or times of reduced prey diversity (Okanogan, Winter) have higher dietary overlap in use of of prey species
  - → Areas of higher prey diversity (Northeast) have less dietary overlap in use of prey sex within prey species
- Data consistent with hypothesis inverse relationship between prey species diversity and dietary niche overlap
- Trends indicate that prey species niche partitioning between wolves and cougars occurs:
  - → (i) in the Northeast more than the Okanogan in both seasons
  - $\rightarrow$  (ii) in the summer more than winter in both study areas

## Conclusions

#### Final Remarks:

- Collection of scat makes cluster investigation more efficient and results in higher-quality data, makes use of "bedsite" cluster investigations
- Moose mediate dietary overlap between wolves and cougars in the Northeast seasonally
- White-tailed deer allow greater flexibility of cougar diets in Okanogan –
   Winter as compared to wolves
- Livestock allow greater flexibility of wolf diets in Okanogan Summer
  - → cluster data suggests that livestock use is mainly scavenging

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