Ashley Whipple Dept. of Ecology and Evolutionary Biology Institute of Artic and Alpine Research University of Colorado Boulder, CO 80309 <u>Ashley.Whipple@colorado.edu</u>

Boulder County Nature Association: 2018 Year-End Report

Putting stress to the test: Could feces be the key to conserving the American pika?

### Summary:

The American pika (*Ochotona princeps*) is a small mammal that inhabits rocky slopes across the mountain west. Ongoing pika occupancy surveys within Colorado show current pika occupancy and distribution is very high, but recent projections of pika vulnerability to climate change predict dramatic declines in pika occupancy for Rocky Mountain National Park by 2100<sup>1</sup>. The currently broad distribution of pikas provides us with an opportunity to test the pika forecast by looking for early evidence of population stress. By comparing stress in pika populations that occupy sites with and without subsurface ice, we aim to inform predictions of potential fine scale microclimates and inform managers which populations are most vulnerable to changes in climate today. Using a non-invasive technique to collect fecal samples from the field, we can extract a stress hormone called glucocorticoid metabolites to compare stress between targeted populations. We predict that previous vulnerability predictions may have over looked sub-surface ice as a microclimate that has potential to extend the range and persistence of pika. Subsurface temperatures in rock piles (aka talus) underlain by ice are cooler in summer, warmer in winter, and more stable year-round<sup>2</sup> providing a refuge from external stressors. Our preliminary results show that stress may vary seasonally and is highest in the spring and decreases into the fall.

#### Background:

Climate change is predicted to influence range dynamics for many species over the coming decades, with general trends showing upslope and poleward shifts. Developing methods for evaluating those predictions in the short term, rather than waiting for range adjustments, would speed the refinement of predictive models and conservation efforts. For the American pika (*Ochotona princeps*) in Rocky Mountain National Park, future occupancy predictions look particularly dire. Predictions from the NPS-sponsored Pikas-in-Peril (PiP) study suggest that under moderate carbon-emission scenarios, pikas would be lost from more than half of currently suitable habitats by 2100; under high-emission scenarios, no pikas would remain<sup>1</sup>. However, PiP predictions were based on surface climate projections and did not consider sub-surface microclimates that can extend the range and persistence of this species<sup>3</sup>.

Sub-surface ice can create a favorable microclimate for pikas and has been shown to reduce physiological stress<sup>2</sup>. We predict that sub-surface ice could provide better quality habitat for pika and could prove important for the species to persist in the face of a warming climate in mountainous regions. Animals who utilize poor habitats may be most susceptible to new threats and be the first to show population decline because poor habitat quality could be a source of chronic stress. Chronic stress is associated with lower reproduction, lower immune response, and lower body mass<sup>4</sup>.

To explore the stress response in pika, we conducted two studies; 1) *Temporal study*: evaluate how stress varies between and among individuals during the alpine spring and fall seasons using feces collected from pika territories through time; and 2) *Habitat quality study*: evaluate how stress varies between pikas inhabiting talus with sub-surface ice (good quality habitats) vs pikas inhabiting talus without sub-surface ice (poor quality habitats) using feces collected from pika territories in spring and fall. Results from the temporal study will help determine if stress levels are different during cool versus warm seasons. We expect stress levels to be higher coming out of winter, and lower after the peak growing season. Results from the habitat quality study will help determine whether the suggested effect of subsurface ice is widespread and important for species persistence. We expect stress to be higher in poor quality habitats.

### Methods:

Fecal pellets were found in the field by first locating a pika haypile. Because pikas are highly territorial, it is common for them to define site ownership by depositing pellets at or near their haypile. During the temporal study, pellets were collected from 20 individual pika territories on Niwot Ridge, Colorado. Pellets were collected every two weeks from June to September. After every visit, pellets were cleared from the territory to ensure samples collected within the following two weeks were fresh. For the habitat quality study, we visited eight pika populations residing along the Front Range of Colorado. Four of the populations inhabited talus with sub-surface ice and four inhabited talus without sub-surface ice. Sites with sub-surface ice were paired with a nearby site without sub-surface ice so that environmental factors experienced by pikas were as similar as possible. At each of the eight pika populations, fresh pellets were collected from ~15 individual pika territories in the spring and fall. See figure 1 for a map of study area locations.

Feces were considered fresh if found "perched" on a rock, usually adhered with urine, and were somewhat green in color. To ensure samples were from individual pika territories, pellets were collected at least 60 meters apart from one another. Stress levels decline with time after pellet deposition due to environmental factors such as temperature and precipitation, and differ between individuals by sex and age. To account for this, only large fresh adult pellets were collected in the field, and sex will be determined through genetic analysis of samples using a partnership with a college genetics class at Warren Wilson College. Fresh feces collected in the field were placed in a coin envelope and frozen in a -20°C freezer at the University of Colorado-Boulder until lab analysis occurred.

Fecal glucocorticoid metabolites (a type of stress hormone) were measured for each sample collected. Glucocorticoid metabolite measurements typically reflect an average level of circulating glucocorticoids over time and can be collected without capturing or handling an animal. Briefly, 0.1 grams of fecal pellets were dried and mashed to powder and mixed with ethanol. Liquid was separated from fecal solid to analyze the concentration of glucocorticoid metabolites within a sample using a commercially available corticosterone enzyme immunoassay kit. Quantifying stress concentrations in samples using corticosterone enzyme immunoassay kits was made possible from the BCNA research grant.

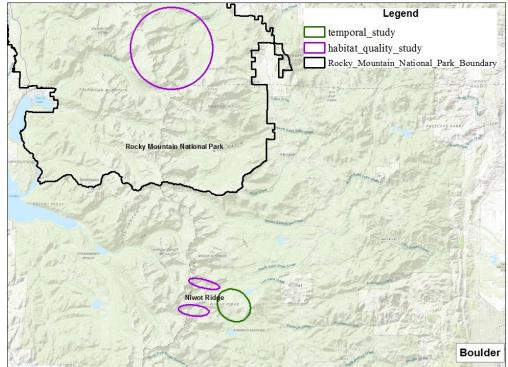


Figure 1: map of area locations of study sites

# Preliminary results:

We collected a total of 100 samples for the temporal study, and 200 samples for the habitat quality study in 2018. Analysis is ongoing for samples collected from the habitat quality study, and we anticipate having all samples analyzed by April 2019. Preliminary results from the temporal study shows variation among individuals was low, indicating that individuals within the same population were not able to differentially modify stress (or are exposed to same level of stressors). Interestingly, the data does show a trend in stress variation among individuals through time (Figure 2). In this study, average stress among individuals is highest in early spring after snow melt, and then decreases until mid-August where after stress begins to increase again. Results coincide with our original prediction that pikas would be more stressed coming out of winter than going into fall, providing evidence that winter causes more physiological stress in pika. We hypothesize that this decrease in stress could be due to an increase in available forage and an increase in food caching ability. Future exploration using mixed effects models will allow us to explore what habitat variables better explain stress in the population of pikas at Niwot Ridge. Some variables of interest are biomass, elevation, and aspect. In the summer of 2019 we plan to revisit sites to collect more data on individual site characteristics such as ratio of graminoids to forbs and forb cover, as these factors have been shown to predict pika occupancy in the state<sup>5</sup>. We suggest that future studies could use this data to then compare differences in stress between years in this particular pika population. Results could help determine which habitat characteristics drive stress levels in pika. To disseminate research findings, an abstract was submitted for a poster presentation at the Biannual Rocky Mountain National Park Research Symposium in March 2019 and a poster will be presented at the Front Range Ecology Symposium at the end of February.

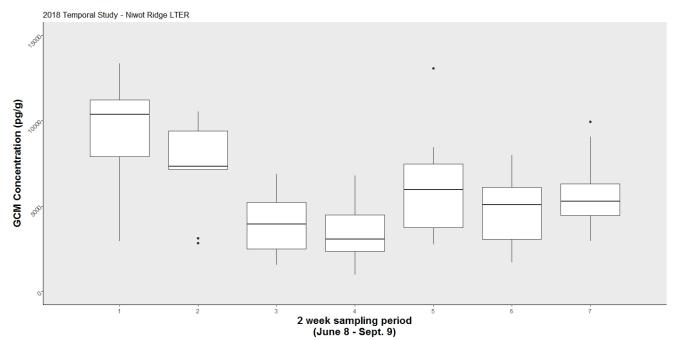


Figure 2: Glucocorticoid metabolite levels measured in American pika (Ochotona princeps) as part of the temporal study. Samples were binned into two-week sampling periods. Sampling period began June 8, 2018 and ended September 9, 2018. Boxes represent medians and the 25% and 75% quartiles (IQR). Whiskers extend 1.5 \* IQR above and below the box. Points are outliers

### Broader impact:

Our research project provided engagement and education to volunteers on local wildlife issues and provided a mentorship opportunity for a local high school student. During July 2018 I mentored a local high school student through the University of Colorado Science Discovery Program. The student accompanied me in the field to learn about ecological research and to practice scientific thinking skills such as hypothesis building and data collection. He also helped with fecal sample analyses in the lab and learned standard laboratory techniques. Lastly, he evaluated stress data collected from the 2017 Niwot Ridge pika occupancy survey and explored if there was a correlation with elevation, slope, or aspect. He presented his findings as a poster in an event open to the local community. Additionally, in collaboration with the Front Range Pika Project, I had the opportunity to train and guide ten volunteers in pika occupancy survey methods and help start a pika citizen science program in Rocky Mountain National Park. Continued engagement in the citizen science program helps volunteers understand why monitoring is important for evaluating pika distribution in the state and how the data is used to inform science.

# References:

<sup>1</sup>Schwalm, D., C.W. Epps, T.J. Rodhouse, W.B. Monahan, J.A. Castillo, C. Ray & M.R. Jeffress (2016) *Global Change Biology* **4**:1572–1584

<sup>2</sup>Wilkening, J., C. Ray & J. Varner (2015) *PLoS ONE* **10**(3): e0119327.

doi:10.1371/journal.pone.0119327

<sup>3</sup>Varner, J. & MD. Dearing (2014) *PLoS ONE* 9(8): e104648. doi:10.1371/journal.pone.0104648
<sup>4</sup>Dantzer, B.; Q.E. Fletcher, R. Boonstrak & M.J. Sheriff (2014) *Conservation Physiology* 2: doi:10.1093/conphys/cou023.

<sup>5</sup>Erb L.P., C. Ray, & R. Guralnick (2014) Ecological Applications 24(3): 429–435