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MS ENVIRONMENTAL BIOLOGY
CAPSTONE PROJECT

by

Mary E. Strecker

A Project Presented in Partial Fulfillment
of the Requirements for the Degree
Masters of Science
in Environmental Biology

REGIS UNIVERSITY
April, 2022

MS ENVIRONMENTAL BIOLOGY
CAPSTONE PROJECT

by

Mary E. Strecker

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April, 2022

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CHAPTER 1. LITERATURE REVIEW

Causes of Regional Biodiversity Loss: The Case of Alpine Aquatic

Macroinvertebrates

Introduction

Climate change and human-induced habitat loss have caused irreversible damage to biodiversity worldwide and the impacts are going to get worse as humans continue to burn fossil fuels and overexploit natural resources (Forest, 2010; Pörtner et al., 2021). Each geographic region is uniquely affected depending on its location, degree of human influence, elevation, climate, and other characteristics. Mountainous alpine regions have been especially impacted as greenhouse gases accumulate and annual temperatures rise. Typically, these habitats hold some of the most pristine and biodiverse ecosystems on Earth due to their insularity and low amounts of human influence (Hotaling et al., 2017). However, climate change has put significant stress on the cold-adapted aquatic macroinvertebrates that inhabit headwater alpine streams as increasing temperatures reduce glacier and snowpack reserves and the upslope movement of species (Hall & Fagre, 2003; Prowse et al., 2006). The altitudinal gradient of temperatures on mountains leaves alpine zones with harsh environmental conditions that can only be sustained by specialist species that heavily contribute to regional biodiversity (Hauer et al., 1997). Biodiversity is crucial to improving a community's resilience to change by giving it a multitude of avenues to adapt (Singh et al., 2017). Studying the loss of aquatic macroinvertebrate community biodiversity in alpine regions provides insight into the mechanisms that are most influential in biodiversity loss in other ecosystems.

One way to quantify regional biodiversity is by measuring gamma diversity, which is classified as the species richness of a landscape and includes a compilation of diversity metrics in multiple ecosystems within a set region (Arellano & Halffter, 2003). The diversity of a specific site within a region is its alpha diversity while the comparison of diversity between sites has been termed beta diversity; both types contribute to gamma diversity (Babu, 2016). A simplified example: Stream A is on the south side of a mountain while Stream B is on the north side, each containing a biotic community of species. The alpha diversity of Stream A can be compared to Stream B's to determine their beta diversity, or similarity. The less similar the streams, the higher their beta diversity. The lower similarity between sites, or the higher the number of species in a given region, the higher the gamma diversity.

The climate-induced shift in habitat characteristics and temperatures have caused a loss of specialized species that contribute to both the beta diversity and gamma diversity of alpine stream macroinvertebrate communities in mountainous regions (Jacobsen et al., 2012). The rapid rate of climate change and the mountaintop limit of these species make them especially difficult to conserve as they are pushed out of their range. It may be too late to conserve these species, however, scientists might be able to use them to relate to the impacts of anthropogenic climate change and habitat loss on the biodiversity loss in other ecosystems. This review uses alpine streams as a case study for the reduction of gamma diversity in ecosystems across the world by highlighting the patterns that are causing the homogenization of these vulnerable communities.

Climate Change and Alpine Streams

The most influential factors in the composition and flow of alpine streams are the characteristics of the hydrological inputs that supply them (Hotaling et al., 2017). Each stream is fed by groundwater, glacier melt, and snow and rain runoff. The characteristics and proportion of

these contributions vary greatly across the world due to differences in climate, temperature, and substrate (Hotaling et al., 2017). The seasonal variation of inputs such as high glacier and snowpack melt contributions in the spring and purely groundwater flow in the late summer can also diversify a stream. This mosaic of inputs throughout the year generates heterogeneous stream characteristics and biotic communities, even between those that are within the same mountain range (Brown et al., 2003, 2007).

The recent and anticipated increase in global temperatures will especially impact precipitation levels and glacier reserves (Hall & Fagre, 2003; Jacobsen et al., 2012). Snowfall and accumulation are predicted to decrease leaving some streams with higher seasonal variation of runoff (Hauer et al., 1997; Siebers et al., 2020). In glacier-fed streams, there will be an initial increase in meltwater contributions followed by a long-term reduction due to loss of reserves (Khamis et al., 2014). The reduced contributions from glaciers and snowmelt due to higher temperatures will also significantly alter flow rates and channel features in alpine streams (Brown et al., 2007). Climate changes may result in longer summer drying periods that will increase stream intermittence which has negative consequences on aquatic communities, such as dry stream beds and limited flow (Datry et al., 2014; Siebers et al., 2020). The significant reduction in glacier melt and precipitation contributions will leave the majority of flow to groundwater, ultimately reducing flow and homogenizing stream characteristics across a region.

Alpine Aquatic Macroinvertebrates

The small footprint of alpine streams has a disproportionately high amount of biodiversity in comparison to other habitats (Hotaling et al., 2017). Aquatic macroinvertebrates are one of the most studied inhabitants of these streams and include insect larvae, gastropods, annelids, crustaceans, and others that dwell in the water. These creatures are commonly used as

water quality indicators because of their ranging sensitivity to pollutants (Khamis et al., 2014; López-López & Sedeño-Díaz, 2015). Additionally, macroinvertebrates are sensitive to flow rates, temperature, and sediment loads (Brown et al., 2007; Finn et al., 2013). Therefore, the impact of increasing temperatures in alpine streams will have a significant impact on alpine aquatic macroinvertebrate community diversity and specialized species' distributions.

Distribution of Macroinvertebrate Diversity

The diversity of macroinvertebrates in alpine streams typically follows an altitudinal gradient. Water temperatures decrease as the elevation and proximity to glaciers or snowpacks increase, resulting in harsh terrestrial and aquatic conditions (Brown et al., 2007; Finn & Poff, 2005). Specialized species with limited ranges and dispersal periods commonly inhabit these extreme headwaters. As the stream flows down the mountain, stream alpha diversity increases as more species can withstand water temperatures and easily disperse (Brighenti et al., 2019; Brown et al., 2007). Conversely, beta diversity is highest at the top of stream reaches where specialists thrive and dispersal rates are low (Finn & Poff, 2005; Hotaling et al., 2017).

Finn and Poff (2005) compared four physically similar streams in the Rocky Mountains and sampled the macroinvertebrate communities across five different altitudinal positions along each stream. They found that alpine stream communities had higher beta diversity than streams at lower altitudes (Finn & Poff, 2005). They attribute the dissimilarity of alpine streams to the harsh conditions, cold temperature, and insularity of these environments that inhibit the dispersal of flying, adult macroinvertebrates to other streams.

As temperatures rise, generalists and other species that live below the alpine region are expected to expand their ranges and disperse upstream. Additionally, cold-adapted species' ranges will shrink due to the mountaintop limit of their habitat and inability to move to colder

water (Hotaling et al., 2017). The combined effects of habitat restrictions and the homogenization of hydrological inputs due to the loss of glacier and snowpack reserves threaten the specialized species and beta diversity of alpine streams.

Influence of Hydrological Inputs on Macroinvertebrate Diversity

The diversity of hydrological inputs (groundwater, glacier melt, and snow or rain run-off) into alpine streams heavily impacts alpine communities. The macroinvertebrates are substantially influenced by the characteristics of the stream inputs that determine the sediment load, flow, and temperature of the water. As contributions from glaciers and snowpack decrease and groundwater becomes the major contributor to alpine streams in a region, the stream characteristics will homogenize and the beta diversity of streams will decrease (Brighenti et al., 2019; Finn et al., 2013; Finn & Poff, 2005). Additionally, flow rates are predicted to change through an initial increase in flow when glaciers melt followed by a long-standing reduction when glaciers diminish (Khamis et al., 2014).

Many studies have analyzed the impacts of receding glaciers and precipitation changes on alpine macroinvertebrate diversity (Brighenti et al., 2019; Brown et al., 2007; Finn et al., 2013; Giersch et al., 2017; Jacobsen et al., 2012; Muhlfeld et al., 2011; Siebers et al., 2020). The reduction of flow combined with increasing temperatures increase alpha diversity in a stream and lowers beta diversity between streams (Brown et al., 2007; Siebers et al., 2020). The increase of alpha diversity is likely due to warmer temperatures and lower amounts of suspended sediments from runoff, which allows generalists to move upstream to compete with specialized species (Brown et al., 2007). Unfortunately, the populations of species that have adapted to cold and harsh conditions are reduced as their long-standing niches diminish and communities from downstream invade, reducing beta diversity.

Two specialist species are listed as “threatened” under the *Endangered Species Act*: the glacier stonefly (*Zapada glacier*) and the meltwater stonefly (*Lednia tumana*). These species have been extensively researched and are endemic to glacier-fed alpine streams in Glacier National Park (Giersch et al., 2015, 2017; Muhlfeld et al., 2011). They are extremely sensitive to the loss of glaciers in this region and have already lost close to 80% of their historic range (Muhlfeld et al., 2011). These specialists likely represent a range of cold-adapted alpine species that are being heavily impacted by losses in ideal ecological niches due to increased temperatures and the reduction of cold water contributions (Giersch et al., 2017). The number of species that are threatened in alpine regions is unknown and their extinction will reduce gamma diversity. Filling this knowledge gap by identifying endemic species in alpine regions and monitoring their populations can provide useful information for how the homogenization of communities will impact the dynamics of the stream and diversity of a region.

The loss of cold-adapted species due to range restrictions and the homogenization of hydrological inputs will decrease the beta diversity between streams in alpine zones (Finn et al., 2013). Gamma diversity will also be lessened by the impacts of climate change on these pristine communities (Brown et al., 2007; Hotaling et al., 2017; Jacobsen et al., 2012). Historically, the beta diversity of two alpine streams on the same mountain could be high and significantly contribute to the gamma diversity and taxonomic richness of an area (Jacobsen et al., 2012). Regional biodiversity improves ecosystem resilience by allowing for the adaptability of species to disturbance (Singh et al., 2017). As alpine communities begin to homogenize, they will lose endemic species that heavily contribute to regional diversity and will be more vulnerable to further environmental impacts (Khamis et al., 2014).

Connections to Regional Biodiversity Loss in Other Ecosystems

Many ecosystems around the world are being impacted by anthropogenic pollution, habitat loss, invasive species introductions, and climate change (Singh et al., 2017). The insularity of alpine streams makes climate change the primary threat to the diversity of aquatic macroinvertebrate communities. The untouched nature of these ecosystems allows us to use alpine communities as a somewhat controlled case study for regional biodiversity loss in ecosystems around the world.

The altitudinal gradients seen in mountain ecosystems are analogous to the latitudinal gradients extending from the equator (Hauer et al., 1997). In alpine zones, elevational limits prevent species from dispersing to cooler temperatures, unlike in other habitats where animals and plants can migrate away from the equator to escape higher temperatures and potentially adapt over time. Additionally, anthropogenic land use has fragmented ecosystems and destroyed heterogeneous habitats that support diverse biotic communities (Singh et al., 2017). Therefore, the reduction of gamma diversity in alpine zones from range restrictions and the homogenization of ecological niches relates to the loss of diversity in ecosystems around the world.

The grasslands of North America have experienced extreme habitat fragmentation due to the human-induced conversion to cropland along with woody and urban expansion (Augustine et al., 2019). Historically, a multitude of bird species thrived in the heterogeneous landscapes of grasslands that were a result of periodic fire and grazing species that could move across extensive areas without barriers. Monoculture crops, removal of native grazers, fire suppression, and habitat loss have homogenized grasslands (Augustine et al., 2019; Brennan & Kuvlesky Jr., 2005). Therefore, grassland bird species have experienced significant declines as their unique niches and breeding habitats are infringed upon. These factors align with those that influence

diversity loss in alpine aquatic macroinvertebrate communities most: range restrictions and homogenization of ecological niches.

Additionally, tropical rainforests are the most biodiverse terrestrial ecosystems on the planet and contain more than half of the known species on Earth (Dirzo & Raven, 2003). Rain forests are full of specialist and endemic species that require specific conditions to survive, similar to alpine macroinvertebrates. The productivity and biodiversity are so high in tropical rainforests that some insect species may be endemic to a single tree. The Amazon Rainforest in South America has experienced significant declines in biodiversity due to deforestation for agriculture, logging, mining, and cattle ranching (Dirzo & Raven, 2003). Studies have shown that, in these areas, diversity declines along a gradient of increasing human disturbances (Solar et al., 2015). Not only are some ecological niches being homogenized or completely removed, but there has also been a significant reduction in the range of native forests that were historically used by plant and animal species (Foley et al., 2007). Again, the two most influential factors in alpine macroinvertebrate diversity can be seen in reducing biodiversity in tropical rainforests. This trend in biodiversity loss is present across ecosystems that are both isolated from and in proximity to human development which indicates this could apply to many regions.

Conclusion

A frequent saying in biology is, “common species are rare and rare species are common.” Gamma diversity is heavily influenced by the prevalence of rare species that are typically specialists in an ecosystem (Jacobsen et al., 2012; Solar et al., 2015). As specialists are lost to range restrictions and the homogenization of ecological niches, we can expect reductions in regional diversity across the globe.

Anthropogenic climate change and rapid habitat loss negatively impact the biodiversity of ecosystems around the world, reducing their resilience and adaptability. The traditionally high beta diversity of alpine aquatic macroinvertebrate communities decreases as heterogeneous hydrological inputs and viable habitats for cold-adapted specialist species are lost. The loss of these rare and endemic species could give insights into the umbrella factors that influence regional biodiversity loss. The patterns seen in alpine streams and other regions show that habitat fragmentation and homogenization are the biggest contributors to regional biodiversity loss. When creating conservation, restoration, or reserve designs, the emphasis on the preservation of large, connected areas with heterogeneous ecological landscapes is necessary to protect the specialist species reliant on them.

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CHAPTER 2. GRANT PROPOSAL

The Effects of Receding Glaciers on Alpine Aquatic Macroinvertebrate Diversity in Glacier National Park

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Section 1. Abstract

Full deglaciation of Glacier National Park (GNP) is expected to occur within the next few decades. Rapid glacier loss threatens the specialist macroinvertebrate communities dwelling in high alpine streams that heavily contribute to regional diversity. Previous studies conclude that as glaciers recede, water temperatures increase, stream characteristics homogenize, and specialists are outcompeted by generalists moving upstream. The isolated impact of reduced glacial runoff on aquatic macroinvertebrate diversity in GNP is unknown. I plan to isolate the impacts of glacier meltwater on these communities by sampling in early September when the annual snowpack has melted. I will classify the glacial influence in 16 sites across 4 glacier-fed streams using a glaciality index that incorporates the physicochemical characteristics of each stream segment. I will identify and evaluate the macroinvertebrate community diversity among and within streams with varying glacial influence to assess the impact of glacier loss on alpha and beta diversity. My analysis will provide crucial insights into the response of alpine aquatic macroinvertebrate community diversity as glacier-fed stream characteristics change and become less heterogeneous in GNP.

Section 2. Objectives, Anticipated Value, Literature Review, Hypotheses

Objectives

This study will investigate the impacts of receding glaciers on the diversity of alpine aquatic macroinvertebrate communities. The effect of increasing temperatures and reduced flows on the distribution of macroinvertebrate diversity in the upper segments of glacier-fed alpine streams in Glacier National Park (GNP) is unknown. Evaluating shifts in diversity indices across streams with varying glacial influence will provide information on the loss of specialist species that heavily contribute to regional biodiversity as global temperatures rise.

Anticipated Value

This investigation will provide key insights into the future of alpine macroinvertebrate diversity as climate change causes global temperatures to rise and glaciers to recede. Glacier-fed streams are only survivable by specialist macroinvertebrate species because they are cold, harsh environments with heavy suspended sediment loads. Isolating the influence of glacier meltwater from snowmelt will provide a focused perspective of how much glaciers influence stream diversity and how stream communities with high glacial influence will respond to deglaciation. Rare and endemic species heavily contribute to gamma diversity and are being threatened by glacier loss and the upslope movement of generalist species (Hauer et al., 1997). In this study, I will classify the aquatic macroinvertebrate community diversity within and among streams with varying levels of glacial influences in GNP. Ultimately, this analysis will help predict how future glacier loss will impact regional biodiversity and alter mountain stream ecosystems.

Literature Review

Anthropogenic climate change has caused global temperatures to rapidly increase over the last century and mountainous regions at mid-latitudes are warming twice as fast as other parts

of the world (Hall & Fagre, 2003; Pederson et al., 2010; Pörtner et al., 2021). As a result, the glaciers of GNP have significantly declined and full deglaciation is expected within the next few decades (Hall & Fagre, 2003). Typically, glacier-fed alpine streams harbor some of the most pristine and biodiverse ecosystems on Earth because of their insularity and limited human influence (Hotaling et al., 2017). Each glacier holds a unique set of sediment and water characteristics that increase environmental heterogeneity and diversify headwater stream characteristics. Biotic communities can vary significantly between nearby alpine streams because of distinct conditions and harsh environments that lead to genetic isolation (Finn & Poff, 2005). However, the reduction of glacial inputs into these streams will increase temperatures while reducing the flow and suspended sediment loads that influence the composition of macroinvertebrate communities. As alpine streams in GNP shift to warmer and less turbid water, abundances of unique and cold-adapted specialists will decrease, reducing regional biodiversity (Hauer et al., 1997; Khamis et al., 2014). Biodiversity is crucial to improving a community's resilience to change by giving it a multitude of avenues to adapt (Singh et al., 2017). Therefore, analyzing macroinvertebrate biodiversity within and among alpine streams that have varying glacial influences will evaluate the impact of climate change on the biodiversity of the streams in GNP.

The most influential factors in the composition and flow of alpine streams are the characteristics of the hydrological inputs that supply them (Hotaling et al., 2017). A unique mix of groundwater, glacier melt, and precipitation runoff feeds each stream. Glacier meltwater has distinct characteristics determined by the suspended sediments within it and the substrate the glacier rests on (Hall & Fagre, 2003). Streams that have high glacial influences are typically very cold with stable flows and high volumes of suspended sediments (Finn et al., 2013). As glaciers

shrink and temperatures rise, the flow will rely on snowpack runoff and groundwater, which will homogenize stream characteristics across a region.

Aquatic macroinvertebrates are one of the most studied inhabitants of alpine streams and include insect larvae, gastropods, annelids, and others that dwell in the water. They are used to indicate water quality because of their variable sensitivity to pollutants (Khamis et al., 2014; López-López & Sedeño-Díaz, 2015). Additionally, aquatic invertebrates are sensitive to stream flow rates, temperature, and sediment loads (Brown et al., 2007). Therefore, increasing temperatures will significantly impact the diversity of alpine aquatic macroinvertebrate communities and specialized species' distributions.

The unique and harsh characteristics at the headwaters of glacier-fed streams lead to small, distinct communities that can vary significantly among streams. Water temperatures increase as the elevation and proximity to glaciers decreases, allowing more species to survive as conditions become less restricting (Brown et al., 2007; Finn & Poff, 2005). Finn and Poff (2005) found streams closest to runoff source were more physically and biologically different from each other than streams at lower altitudes (farther from the source). They attribute the dissimilarity of high altitude, glacier-fed alpine streams to the harsh conditions, cold temperature, and insularity of these environments that inhibit the dispersal of flying, adult macroinvertebrates to other streams.

The high environmental and biological heterogeneity of glacier-fed streams are threatened by the loss of glaciers and the homogenization of hydrological inputs. Macroinvertebrates from downstream are likely to invade cold-adapted species' ranges while specialists are pushed to the brink of extinction (Finn & Poff, 2005; Khamis et al., 2014). Losing

these species will undoubtedly reduce regional biodiversity as alpine streams become more similar (Hauer et al., 1997).

Biodiversity can be quantified by measuring alpha, beta, and gamma diversity. Alpha diversity (α) is the diversity of a specific stream within a region, while beta diversity (β) refers to the comparison of diversity among sites. Both alpha and beta diversity are used to calculate regional biodiversity, or gamma diversity (γ) (Babu, 2016). The lower similarity among sites, or the higher the number of species in a region, the higher the gamma diversity. Traditionally, the alpha diversity of glacier-fed streams is lowest near the meltwater source and increases as proximity decreases. Additionally, the communities closest to the source of different glaciers have a high beta diversity (low similarity) because of the unique conditions and insularity of each outfall. Therefore, losing glaciers threatens gamma diversity as specialist species are lost to stream characteristic homogenization, flow reduction, and warmer temperatures.

Few studies have isolated the impact of glacier input from snowmelt by sampling strictly during August and September when flow is restricted to glacier meltwater and groundwater (Brown et al., 2007; Finn et al., 2013). The type of flow can then be distinguished using a glaciality index to characterize stream conditions (Finn et al., 2013). A stream with high glaciality (high glacial influence) has low temperatures, high suspended sediment loads, low substrate stability, and high meltwater contributions. Evaluating the isolated impact of glacier meltwater will provide useful insight into how important glaciers are to regional biodiversity and the future of alpine stream aquatic macroinvertebrate communities. As glaciers are lost from GNP in the next thirty years, it is important to understand how physicochemical characteristics of headwater streams will change and impact stream ecosystems and food webs.

Hypotheses

To understand and isolate the impacts of receding glaciers on alpine stream macroinvertebrate community diversity in Glacier National Park I will test three hypotheses:

1. The alpha diversity of a stream with low glaciality will be higher than streams with high glaciality.
2. The beta diversity among streams with high glaciality will be higher than the beta diversity among streams with low glaciality.
3. The alpha diversity of a stream will increase as the proximity to the glacier outfall point decreases.

Section 3. Methods

Study Sites

I will conduct this study in four catchment streams of Glacier National Park, Montana, USA that are fed by the Vulture, Rainbow, Carter, and Agassiz Glaciers (Figure 1 and 2). Each stream will have four sampling sites that are within 5 km of the source. If possible, the altitude of each site will correspond to the altitude of sites under the other glaciers to remove the confounding variable of elevation. I will measure these factors in September to ensure meltwater conditions are from glaciers and long-term snowpack after annual snowpacks have already melted in the summer. I selected these basins because they have relatively steep gradients of glacial influence.

Classification of Glacial Influence

Following Ilg and Castella (2006) and Finn et al. (2013), I will classify each of the 16 sites as “high,” “mid,” or “low” glacial influence based on four physicochemical factors: Water temperature, substrate stability, suspended sediment concentrations (SSC), and conductivity. I

will measure water temperatures and conductivity using a YSI 5563-10 Probe. I will also collect water samples from each site to determine SSC using the standardized method, ATSM D3977 - 97, in which samples are weighed before and after water is decanted or evaporated from the sample. Lastly, I will visually determine the substrate stability using the Pfankuch Index (PI) method (Collier, 1992).

Combining these data will allow me to cluster the sites hierarchically by relativizing the variables from 0 to 1 and using Ward's linkage method with Euclidean distance in pc-ord (Finn et al., 2013). I aim to get similar results of clusters seen in Finn and Poff (2013) that group each site into a glaciality index with high-glaciality being glacier run-off dominated and low-glaciality being groundwater dominated. High glaciality sites will have low temperatures, high SSC, high PI, and high percent meltwater (derived from conductivity measurements).

Aquatic Macroinvertebrate Sampling

At each site, I will hold D-frame kicknets (150 micron) directly downstream from my boots and kick up rocks and substrate for 30 seconds in each riffle microhabitat at the site (Finn & Poff, 2005). Samples from each site will be filtered through a 50 μM sieve and preserved in 70% ethanol in the field for further identification by Aquatic Biology Associates to identify them to the highest possible taxonomic resolution.

Statistical Analysis

For the purposes of this study, I will use Whittaker beta diversity ($\beta = \alpha/\gamma$). Each site will have two beta diversity indices: beta diversity along sites in the same stream (β_1) and beta diversity among sites with the same glaciality measure (β_2). For β_1 , I will calculate the Shannon's diversity of each sampling site (α) and divide it by the Shannon's diversity of the entire stream (γ_1). For β_2 , I will use the same α but divide it by the Shannon's diversity of all

the streams within a glaciality group (γ^2). I will test for significant differences between values using an ANOVA test and post-hoc Tukey tests. I will also compare the alpha diversity (α) of each site to other points along the same stream and among its glaciality group using an ANOVA test and post-hoc Tukey tests. Differences of alpha observed along a stream will indicate whether proximity to the glacier impacts α . Lastly, differences of alpha among streams in different glaciality groups will indicate whether glacial influences impact α .

Negative Impacts

There will be minimal negative impacts of this study. I will only collect macroinvertebrates once to ensure there are minimal impacts to the functionality of sampling sites and the survival of macroinvertebrates that are not collected. I will be sure to only disturb the stream's substrate in the areas where I collect with the kick net and will avoid walking in the stream when it is unnecessary.

Project Timeline

Date	Activity	Deliverable
September 1st - September 14th	Collect physicochemical stream characteristics and macroinvertebrate specimens.	Data from each sampling point. Macroinvertebrate samples from each sampling point in 70% EtOH.
October 1st	Send macroinvertebrate samples to Aquatic Biology Associates (ABA) for identification.	Billing information and order confirmation from ABA.
October 2nd - October 10th	Input, tidy, and clean data from physicochemical stream characteristics and classify glaciality of each sampling site.	Tidy data, figures, and analyses of glaciality index of each sampling site.
October 21st - November 1st	Analyze macroinvertebrate specimen identification results from ABA.	Finished data analysis and figures for the results section.
December 1st	Draft, edit, and complete report.	Final Report.

Section 4. Budget

Budget

Item	Justification	Cost/Unit	Quantity	Total
Aquatic Net, 12" Dia. Bag, D Shape	To collect macroinvertebrate samples in the field	\$77	1	\$77
YSI 5563-10 Probe with Conductivity and Temperature Sensor	To collect temperature and electrical conductivity from sampling sites.	\$1,285	1	\$1,285
Aquatic Biology Associates	High resolution taxonomic macroinvertebrate identification.	\$250	16	\$3,750
Plane ticket	To get to Flathead County Airport (FCA), Montana and back to Denver	\$289	1	\$289
Rental car	To get from hotel to sites in GNP	\$87/day	14	\$1,224
Hotel stay	Housing while collecting data in GNP	\$113/night	13	\$1,827
Total Resource Expenditures				\$8,452

Appendix:



Figure 1. Map of Glacier National Park. Location of study sites indicated in red box (see Figure 2 for close up).



Figure 2. Locations of the four glaciers included in this study.

Section 5. Qualifications of Researcher

Mary Strecker

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EDUCATION

Master of Science, Environmental Biology May 2022
Regis University, Denver, CO

Bachelor of Science, Biology May 2020
Rockhurst University, Kansas City, MO
Major: Organismal Biology Minor: Environmental Studies

WORK AND RESEARCH EXPERIENCE

Laboratory Technician 2020-2021
Nectagen Inc., Kansas City, KS

- Assisted a small team with the innovative development of antibodies as biomedical research and diagnostic tools.
- Performed routine cloning in *E. coli*, including PCR amplification and DNA extraction.
- Expressed and purified proteins from microbial hosts for further testing and analysis of properties.

Student Researcher 2017-2020
Rockhurst University, Kansas City, MO; Dr. Chad Scholes

- Identified over 50 species of bacteria to expand the database of epiphytes on Midwestern tree species.
- Collected leaves and other organisms for research in the field using climbing gear to obtain samples in the mid and upper canopy of trees.
- Conducted lab procedures such as PCR, electrophoresis, pipetting, Gram staining to identify bacterial species.

Research Intern Summer 2019
Smithsonian Environmental Research Center (SERC), Edgewater, MD; Dr. Whitman Miller

- Investigated the respiration rates of organisms and variable CO₂ in the water column of the Rhode River, MD.
- Supported the Ocean Acidification Lab with current experiments and publications.
- Executed biweekly maintenance on the lab's CO₂ monitoring equipment.

Storm Water Engineering Intern Summer 2018
City Hall of Overland Park, KS; Ian Fannin-Hughes

- Aided a Water Quality Specialist in inspecting storm water discharge points for damage and pollution by recording conditions and utilizing Lucity mapping software.
- Evaluated the water quality of 15 sampling sites across Overland Park through habitat, water quality, and macro-invertebrate assessments.

ACADEMIC AWARDS

Marshall Anderson Award for Academic Achievement in Microbiology, *Rockhurst University*

Member of Alpha Sigma Nu – Jesuit Honors Society, *Rockhurst University Chapter*
NSF Funded Research Experience for Undergraduates, *Smithsonian Environmental Research Center - Ocean Acidification Lab*

Dean's List - Fall and Spring Semesters 2016-2020, *Rockhurst University*

LEADERSHIP AND COMMUNITY ENGAGEMENT

Student member of Sustainability Initiative Tripartite Committee, *Rockhurst University*
Student Ambassador, *Rockhurst University*

Active Minds Executive Board: Research Liaison, *Rockhurst University Chapter*
Volunteer with Kansas City Wildlands and Heartland Conservation Alliance

SCIENTIFIC PRESENTATIONS

Oral Presentations

Strecker, M. *One Piece of the Puzzle: Water Column Respiration Rates in the Rhode River.* Smithsonian Environmental Research Center, August 2019.

Strecker, M., Rode, O. *Overland Park Watershed Assessment-Final Report 2018.* The City of Overland Park, Public Works Department, August 2018.

Poster Presentations

Strecker, M; Rode, O, and Scholes, C. *What Bacteria are Epiphytes of Midwestern Oak Tree Species?* The National Conference for Undergraduate Research, University of Central Oklahoma, April 2018 and The Festival of Student Achievement, Rockhurst University, April 2018.

Strecker, M; Rode, O, and Scholes, C. *What Bacterial Epiphytes from Midwestern Trees Possess Nitrogen Fixing Genes?* The Missouri Academy of Science Conference, Northwest Missouri State University, April 2019 and The Festival of Student Achievement, Rockhurst University, April 2019.

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CHAPTER 3. JOURNAL MANUSCRIPT

Evaluation of Distance Sampling as a Tool for Monitoring Populations of Rare Plants (Cactaceae: *Sclerocactus glaucus*)

Abstract

Accurate population size estimates of species are crucial to develop effective management strategies and monitor threatened species, however, they are difficult to obtain. Surveying rare plant species or species that are difficult to detect with inconspicuous life stages is often time-consuming and unrealistic to sample year-to-year. Distance sampling, a plotless method typically used on mobile animals allows for imperfect detection and models detection probabilities to provide unbiased and time-efficient population estimates. The goal of this project was to assess the efficacy of distance sampling to estimate the population size of the Colorado hookless cactus (*Sclerocactus glaucus*) across its range in the Gunnison and Colorado River Basins compared to the effort required to estimate a minimum population size with sampling plots. We placed 30-meter transects 15 meters apart in two sampling sites and measured the perpendicular distance from the transect line to each cactus detected by the observer and calculated the plant cover using line-point intercept sampling at each site. We found that the required sampling effort for precise estimates can be reduced in high-density areas and that the density of *S. glaucus* differed between sites but the detectability of individuals did not. Our study suggests distance sampling would be an unbiased, faster method than previously employed plot-based methods and can be repeated yearly to closely monitor the population of the threatened Colorado hookless cactus.

Introduction

Obtaining accurate population size estimates of species is difficult but crucial to improve management strategies and to set priorities for the monitoring and recovery of threatened species (Keith, 2000). Longitudinal surveys of population size are especially important for long-lived plants when they are bound to one location, making them sensitive to changes in their environment over time. Because rare and endemic plants contribute disproportionately to regional plant diversity and are likely more threatened by environmental pressures, accurate estimates of trends in population size are necessary to manage their populations. However, the rarity of these plants often poses challenges for surveying; therefore, accurate and efficient sampling methods to monitor population sizes and trends are required to inform conservation decisions (Flesch et al., 2019).

Plot-based methods frequently used to estimate plant population sizes assume that all individuals can be detected by the surveyor. Plot-based surveys require the observer to place plots inside suitable habitat and count the number of focal plants within the plot. When the focal species is rare or when its habitat covers a large area, the sampling effort must be increased by using larger or more numerous plots (Jensen & Meilby, 2012). Additionally, the ability of the observer to achieve unbiased sampling with random plot placement may be limited by uneven terrain that results in the subjective placement of plots, thereby limiting the inferences that can be made about the size of the population (Jamali et al., 2020; Krening et al., 2021). Finally, plot-based methods may also be inaccurate when the species is small or has inconspicuous life stages (Buckland et al., 2015) because these methods may underestimate population densities by not adequately accounting for imperfect detection.

Distance sampling is often used for mobile species where detection is an issue. When animals move away from the observer, they are hard to detect, and although plants are sessile, some are rare across the landscape or have inconspicuous life stages. Distance sampling may be useful for plants that are sparsely distributed, have small seedlings, blend in with the substrate, or are hidden by surrounding vegetation, making them challenging and time-consuming to sample by traditional plot-based methods. By explicitly modeling detection probabilities, plotless sampling methods, such as distance sampling, provide a precise and time-efficient method to estimate the total abundance of species (Buckland et al., 2015). Distance sampling is typically not useful for plants that cannot be distinguished as individuals or that are so small and hidden that an observer cannot detect every individual along a transect (Buckland et al., 2015). Distance sampling allows the observer to model the zone where individuals are detected by fitting the detection probability as a function of their distance from the transect line (Buckland et al., 2015). Much larger areas can be covered in less time than plot-based methods because the same number of detections are needed to estimate abundance in both large and small regions (Buckland et al., 2015). Additionally, by making use of the assumption that not all individuals will be detected, as little as 10-30% of the individuals in an area must be detected to estimate an accurate population size (Jensen & Meilby, 2012). The unbiased nature of distance sampling achieved by explicit incorporation of the detection probability allows the observer to accurately estimate the total population size of target individuals in a more efficient way than plot-based methods.

Although less commonly used than plot-based sampling, distance sampling has been used to estimate the density and abundance of several plant species including trees (Jensen & Meilby, 2012), small herbs (Flesch et al., 2019), and cacti (Schorr, 2013). Using distance sampling to survey plants is advantageous because the observer can measure both morphological features and

exact distances to each plant's location from the transect line (Flesch et al., 2019; Schorr, 2013). Schorr (2013) used distance sampling to reliably estimate the density and abundance of Weber's saw-worts (*Saussurea weberi*) throughout its wide range in the Rocky Mountains, demonstrating that this methodology can be used to survey populations of small, rare plants.

The Colorado hookless cactus (*Sclerocactus glaucus*) is a long-lived, small perennial cactus endemic to the Gunnison and Colorado River Basins where it inhabits alluvial benches and upland desert habitats with gravelly or rocky soils (U.S. Fish and Wildlife Service, 2021). The species was federally listed as threatened under the Endangered Species Act (ESA) by the U.S. Fish and Wildlife Service in 1979 based on density and abundance data in the Element Occurrence Records from the Colorado Natural Heritage Program. These data were collected from surveys without consistent structure or time intervals (Krening et al., 2021). Updated information on the genetics, life cycle, and minimum population size of *S. glaucus* has prompted the U.S. Fish and Wildlife Service to recommend the delisting of this species under the ESA (U.S. Fish and Wildlife Service, 2021). The ability to rapidly detect changes in population size is especially important after potential delisting from the ESA to determine if any additional conservation actions are warranted after ESA protections are removed.

In 2020, the Bureau of Land Management (BLM) used plot-based surveys to estimate the minimum population size of *S. glaucus* across the southern range of the species (Krening et al., 2021). While minimum estimates in 2017 greatly surpassed the previously estimated population size of *S. glaucus* (Krening et al., 2021), the investigators were unable to infer the total population size of *S. glaucus*. Although Krening et al. (2021) used a spatially balanced random sample to select target populations, they subjectively placed rectangular sampling macroplots within each population in areas of higher cactus density. Because plot placement was not

randomized within populations for the sake of reducing survey time and cost, Krening et al. (2021) were unable to estimate the total population size of *S. glaucus*. The abundance of each macroplot was used to calculate the density of each of the habitat areas sampled, but the investigators were only able to confirm that the population contained at least as many individuals as were estimated from the macroplots. Although the minimum population size estimates showed that the population was larger than previously presumed, this sampling effort took several years and is not a repeatable way to efficiently monitor the *S. glaucus* populations because it is sensitive to the placement of macroplots and the distribution of the cactus population. Distance sampling transects could cover entire patches of suitable habitat independent of the distribution to provide an unbiased and time efficient method to estimate the total abundance of *S. glaucus* throughout its range and over time.

By empirically comparing population sizes estimated from two different sampling protocols for this species, our goal is to recommend a sampling protocol to estimate *S. glaucus* abundance including minimum sample size and effort. We compared population size estimates and the uncertainty derived from plot-based surveys (Krening et al., 2021) to abundance estimates derived from distance sampling along with the time invested in both methods. The semi-arid, high-elevation desert habitat, and sparse vegetative cover typical of *S. glaucus* habitat populations allows the cactus to be easily detected along line transects, making it an ideal candidate for distance sampling. The varying distribution of *S. glaucus* and the potential to miss seedlings or nonreproductive individuals could lead to underestimates; however, the method is faster than previously employed plot-based methods and may provide unbiased estimates of total population size. Consequently, we hypothesized that distance sampling would be an accurate and time efficient way to estimate and monitor the total population size of *S. glaucus* over time.

Methods

Study Species

Sclerocactus glaucus is a long-lived, small perennial cactus with a cylindrical body about 3-12 cm in height and 4-9 cm in diameter, with straight central spines (Ackerfield, 2015). The cactus produces lavender to red flowers during April and May and fleshy fruit develops between May and June. Each fruit produces one to several hundred seeds which are dispersed by water, gravity, and ants. The species is found between 1,370 and 2,200 meters of elevation and has a patchy, generalist distribution across upland deserts (U.S. Fish and Wildlife Service, 2021). The species has a northern and southern center of genetic diversity (Schwabe et al., 2015), and the northern center will be split into a new taxon, *Sclerocactus dawsonii* soon (U.S. Fish and Wildlife Service, 2021).

Sampling Sites

Two locations representing the two centers of genetic diversity were selected within the suitable habitat of *S. glaucus sensu lato* (Figure 1). The northern population occurs in a sagebrush community within pinyon-juniper forest stands and the southern population inhabits an alluvial bench with low grass and shrub cover. The southern population (Picnic), in Delta County, Colorado resides in cobbly and cryptogamic soil within pinyon-juniper openings where *Atriplex canescens*, *A. confertifolia*, *Sarcobatus vermiculatus*, *Artemisia nova*, and *Bouteloua gracilis* also occur. The northern population (T-Junction), in Mesa County, inhabits openings within low-density pinyon-juniper forest stands in clay soils which also support *Ephedra*, *Cercocarpus ledifolius*, and various grasses (NatureServe, 2014).

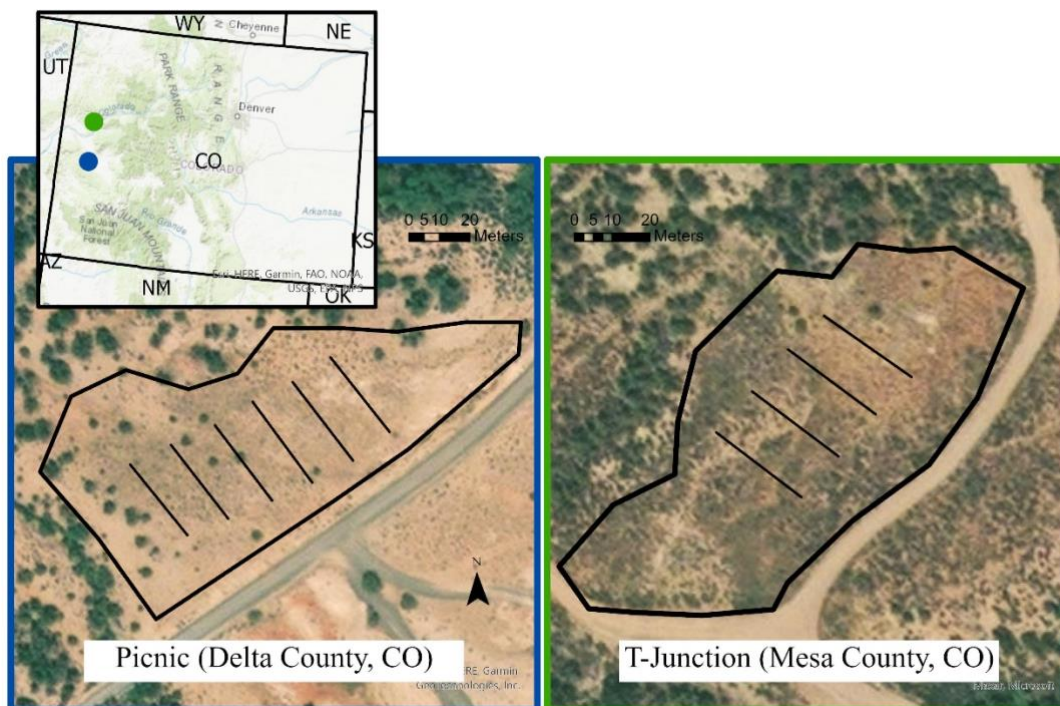


Figure 1. Map of both study sites and the orientation of transects across each area. The blue dot represents Picnic and the green dot represents T-Junction on the reference map.

Distance Sampling

In August 2021, distance sampling along parallel transects was conducted within suitable habitat that excluded areas near the road or inside dense forest stands at both sites. Six and four 30-meter line transects were systematically placed 15 meters apart, parallel to the shorter side of Picnic and T-Junction, respectively (Figure 1). The transects were placed independently of the location of *S. glaucus* individuals to satisfy the assumptions of distance sampling.

One observer walked the line transect and reported all *S. glaucus* individuals they could detect. A second observer measured the exact perpendicular distance from the line to any detected individual. The height of the cactus from the ground to the tallest point of the body and the width of the widest part of the cactus (excluding all spines and reproductive structures) were also measured using calipers. Lastly, the observer counted the number of reproductive structures including flowers or fruit.

Vegetation sampling with a line-point intercept method was used to quantify plant species and functional group diversity as well as vegetative cover at each site (Hufft et al., 2019). A pin flag was dropped every 0.25 meter along the same transects used for distance sampling. At each sampling point, observations were made of every plant species that intersected the point as well as the ground cover (including bare ground, plant litter, rock, and biological soil crust).

Data Analysis

Density and detection probability. We estimated the density and detection probability functions for all individuals. We aggregated the distance data into 25-cm bins and truncated distances to include cacti detected within six meters of the transect line. We used the *unmarked* package (Fiske & Chandler, 2011) in R version 3.6.3 (R Core Team, 2021) to estimate the population size, density, and detection probabilities of *S. glaucus* in the two sampling areas. The proportion of first hits that touched a plant across all transects at a site was used as percent cover. First, we fit a null detection function for both reproductive and vegetative groups without considering the influence of covariates using conventional distance sampling (Buckland et al., 2015). Next, we fit three models that included site as a covariate on density only, on detection only, and on both density and detection. We fit the conventional models using uniform, hazard-rate, and half-normal detection functions and covariate models with only hazard-rate detection functions. The hazard-rate detection is defined by the scale and shape of the best-fitting curve; scale is the curvature of the fit while shape determines the width of the shoulder of the curve (Clark, 2016). The best model for both the conventional and covariate models was selected by comparing Akaike information criteria for small sample sizes (Akaike, 1981) and choosing the model that had the lowest AICc value (Burnham et al., 2011).

Abundance comparisons. Previous abundance estimates were only available for the southern center of genetic diversity, where the Picnic site is located (Krening et al., 2021). Krening et al. (2021) found the minimum population size (103,086 individuals; 90% CI: 68,120 – 138,053) of *Sclerocactus glaucus* by extrapolating the density of the individuals found in a macroplot across entire habitat areas and averaging those estimates across the total habitat area using a ratio estimator (Stehman & Salzer, 2000). We used our estimated density from Picnic to calculate the abundance of *Sclerocactus glaucus* across total habitat area (16,996,891 m²) as defined by Krening et al. (2021).

Simulating the sampling effort required for precise abundance estimates. To estimate the sampling effort needed to obtain accurate and precise estimates of density and population size, we used the best fit models (Table 1) to simulate occurrences in the sampled area using the *DSsim* package in R (Marshall, 2020). From the spatial extents of both sampling sites and the distance sampling transects, we applied our estimated abundance at each site to distribute *S. glaucus* individuals uniformly across the area. The detection function for each simulation was fit using the parameters and estimated densities from our top-ranked model (Table 1). We compared the simulations for the required proportion of area needed to accurately obtain the total population size of an area by assessing where the coefficient of variation of the population size estimates stabilized with increasing effort. To calculate the minimum sampling effort needed at each site to obtain precise results using distance sampling, we simulated sampling designs where transect spacing was systematically varied from 6 to 30 meters in 2-meter increments. Each spacing unit simulation was replicated 999 times.

Results

Detection and site percent cover

We detected 184 *Sclerocactus glaucus* individuals: 159 across 6 transects (total length: 180 m) at Picnic and 25 at T-Junction across 4 transects (total length: 120 m) at T-Junction (Figure 2). The height of the detected *S. glaucus* individuals ranged from 0.25 - 11 cm (mean= 2.47 ± 1.86 cm SD) and their width ranged from 0.5 - 8 cm (mean= 3.42 ± 1.62 cm SD). The percentage of plant cover at Picnic and T-Junction respectively was 17.6% ($\pm 5.36\%$ SD) and 48.1% ($\pm 17.3\%$ SD).

Detection probability

The detectability of *S. glaucus* was not different between Picnic and T-Junction. In conventional models that excluded covariates from both the density and detection functions, the hazard-rate detection function fit the shape and scale of the distance data the best (Table 1, AICc = 595.7). With the hazard-rate detection function, the density of *S. glaucus* was different between sites in our top-ranked model but the detectability of individuals was not (Table 2, AICc = 576.8). The top-ranked model included a scale parameter of 1.16 (95% CI: 0.97 - 1.40) and a shape parameter of 2.58 (95% CI: 2.11 - 3.17, Table 1). The average detection probability across the entire half-width of the top-ranked model was 0.62 (95% CI: 0.57 - 0.68, Figure 3).

Density and abundance estimates

The density of *S. glaucus* individuals was significantly greater in the southern than the northern site ($z = -4.31$, $P < 0.001$). The estimated density of *S. glaucus* was 0.063 individuals/m² (95% CI: 0.034-0.119) at T-Junction and was 0.160 individuals/m² (95% CI: 0.130 - 0.197) at Picnic (Table 2). The estimated total population size of *Sclerocactus glaucus* at T-Junction and Picnic is 409 (95% CI: 270 – 619) and 1247 (95% CI: 1014 – 1535) individuals respectively. If

we assume the density of individuals is even across the total habitat area in Krening et al. (2021), the abundance of the southern population could be as high as 2,719,503 individuals (95% CI: 2,209,596 – 3,348,388).

Sampling effort precision

The density of individuals influences the sampling effort required to obtain precise estimates. The precision of the abundance estimates from simulations decreased as the spacing between transects increased, however, the increase in variation was much greater at the less dense T-Junction site. Placing transects 12 meters apart would be the maximum distance sampling effort because of the 6-meter half-width set in our analyses. At T-Junction, CV estimates ranged more widely (0.13 - 0.42) than at Picnic (0.06 - 0.14). Spacing transects 15 meters apart would be appropriate at either site, however similar precision could be achieved at much greater spacing intervals (effort) at Picnic (Figure 4).

Table 1. Conventional detection function models fit to estimate abundance of *Sclerocactus glaucus*. K is the number of parameters and D is the estimated density (individuals/m²) with the 95% confidence interval.

Key Function	Covariates on detection	Covariates on density	K	AICc	Δ AICc	Weight	Shape	Scale	D
Hazard-rate	-	-	1	595.7	0.00	1	2.58	1.16	0.132 (0.108-0.162)
Half-normal	-	-	1	632.0	36.31	0.00	-	-	0.107 (0.090-0.128)
Uniform	-	-	1	842.5	246.79	0.00	-	-	0.036 (0.031-0.042)

Table 2. Covariate detection function models fit to estimate abundance of *Sclerocactus glaucus*. K is the number of parameters and D is the estimated density (individuals/m²) with the 95% confidence interval. P represents Picnic and T represents T-Junction when site was used as a covariate on the parameter.

Key Function	Covariates on detection	Covariates on density	K	AICc	Δ AICc	Weight	Shape	Scale	D (95% CI)	Abundance (95% CI)
Hazard-rate	-	~ site	2	576.8	0.00	0.92	2.58	1.16	P= 0.160 (0.130- 0.197)	1247 (1014-1535)
									T= 0.063 (0.034-0.119)	409 (270-619)
Hazard-rate	~ site	~ site	3	581.8	5.04	0.07	2.56	P= 1.25	0.159 (0.128-0.198)	1242 (999-15460)
								T= 0.79	0.064 (0.030-0.139)	1003 (193-898)
Hazard-rate	~ site	-	2	587.8	11.00	0.004	2.59	P= 1.17	0.137 (0.111- 0.169)	1900 (1588-2405)
								T= 1.14		
Hazard-rate	-	-	1	595.7	18.97	0.00	2.58	1.16	0.132 (0.108-0.162)	1900 (1546-2304)

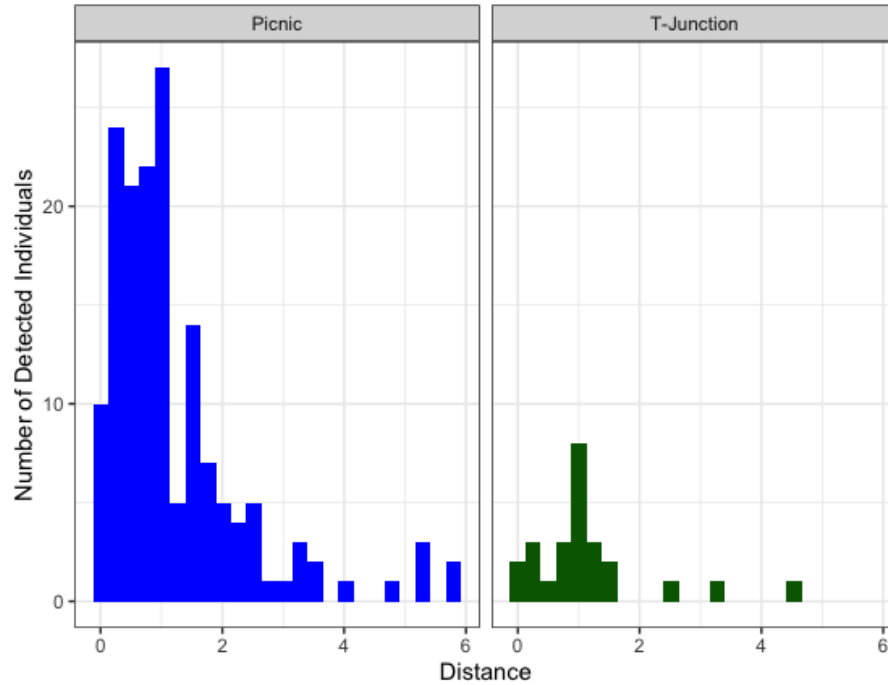


Figure 2. Frequency histogram of survey detections separated by site in 0.25 m bins and truncated at 6 meters used for modeling. Distance sampling assumes that all individuals within 0-0.5 meters of observer are detected.

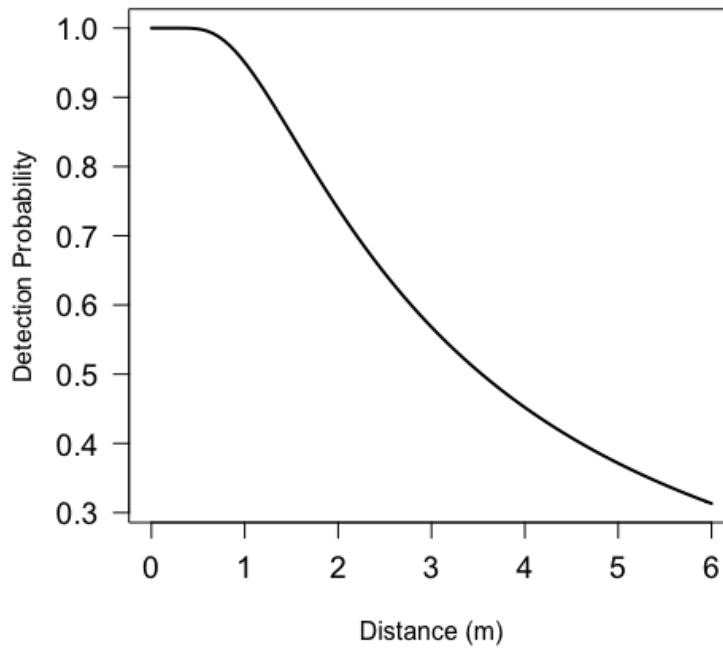


Figure 3. Detection probability of all *Sclerocactus glaucus* individuals from top-ranked model that did not include plant cover as covariate of the detection probability.

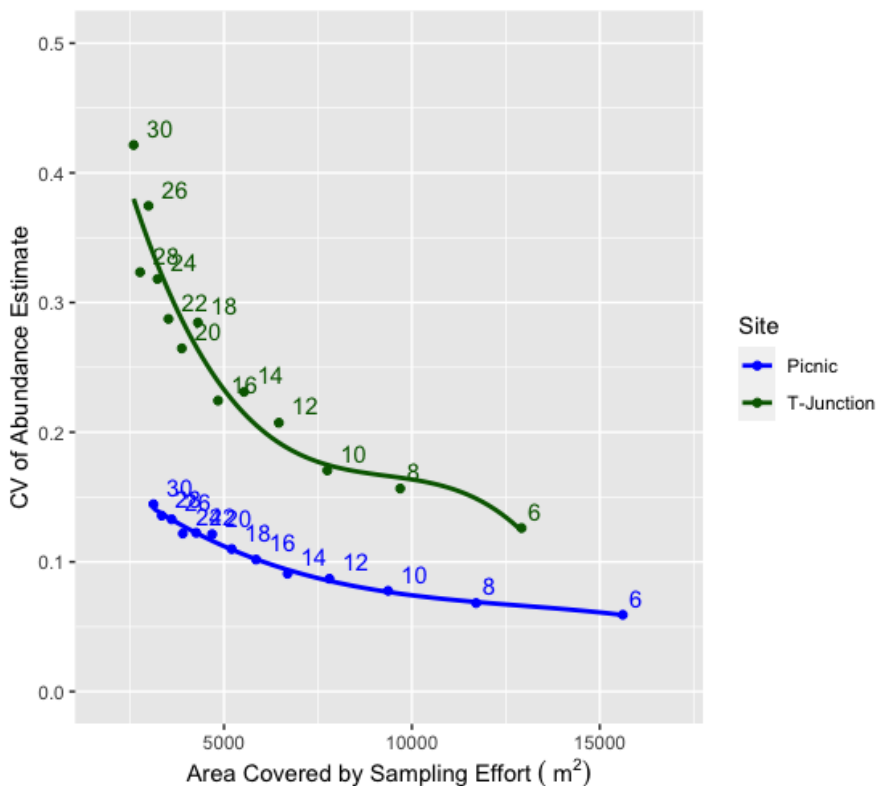


Figure 4. Scatterplot from simulations using the coefficients of variation of the abundance estimates as a function of the mean area covered by the simulated distance sampling effort. The numbers indicate the spacing between transects (in meters) in the simulation.

Discussion

In an effort to use a plotless sampling method as a time-efficient and reliable way to repeatedly monitor *Sclerocactus glaucus* populations, we found distance sampling is quicker and less biased than previously employed methods. Although our sampling effort was less time-intensive than Krening et al. (2021), our simulations suggest that the effort required to obtain accurate population size estimates could be even lower in high-density areas. Despite the fact that detectability of individuals did not differ between the two sites we surveyed, cacti density was higher at Picnic, potentially as a function of plant cover. The difference between sites did not impact the detectability of individuals yet the density was different, potentially as a function of plant cover. Lastly, our density estimates fall within the range of macroplot densities from

Krening et al. (2021) indicating the potential to use distance sampling to provide just as accurate counts in less time and ultimately estimate the total population sizes of *S. glaucus* rather than minimum estimates.

Our results suggest that distance sampling could be a much more time-efficient way to monitor *Sclerocactus glaucus* populations. Using distance sampling can reduce the sampling effort by accounting for imperfect detection and reduce bias by systematically placing transects over sections of the sampling area (Buckland et al., 2015; Flesch et al., 2019). The sampling effort conducted by Krening et al. (2021) took the course of several field seasons to complete, while our effort took approximately two field days. On average, their macroplots covered 6% of the total habitat area. For example, the Reeder Mesa site in their study was 8,408 m² and their macroplot was 900 m², only 10% of the entire habitat area. Krening et al. (2021) assumed that each macroplot contained the only cacti in the entire habitat area which led to their “minimum” population size estimates. Picnic has an area of 7,802 m² and T-Junction is 6,455 m²; our distance sampling effort covered 28% and 22% of these areas, respectively. If we were to cover the total sampling area in the Krening et al. (2021) study (275,853 m²) with a distance sampling effort similar to the one in this study (approx. 25% of the habitat area), the effort could take as little as 38 field days. This scaled effort would provide total population estimates unlike the more time-intensive and biased minimum population estimates from Krening et al. (2021).

Larger sampling efforts are required in areas where *Sclerocactus glaucus* is less dense. Although we were unable to compare precision between our study and Krening et al. (2021), we used simulation models to estimate the appropriate sampling effort needed, given the abundance values from our distance sampling model. Even in T-Junction where cacti were much sparser, spacing transects 15 meters apart gave reasonable precision estimates. Additionally, our

simulations suggest that even smaller sampling efforts are required in more dense areas to accurately and precisely estimate *S. glaucus* populations. *DSsim* has been used in other pilot distance sampling studies to gauge how different densities, population hotspots, and transect distance will influence the accuracy of estimates (Livingston et al., 2018; Witczuk & Pagacz, 2021). Both Livingston et al. (2018) and Witczuk & Pagacz (2021) found that areas with greater densities had more precise estimates and that the CV of estimates increased as effort was reduced. Our pilot study simulations proved to be a useful way to estimate the effort required to obtain accurate population estimates using distance sampling, and how sampling effort can be reduced at sites with higher density.

Despite differences in density, the detection probability of *Sclerocactus glaucus* was not different between sites. Covariates are often applied to distance sampling models and can influence detection (Flesch et al., 2019; Schorr, 2013); however, the use of only two sites limited our ability to apply covariates such as site plant cover to our models. We found that the plant cover was much higher at T-Junction, where the density and abundance of individuals was lower than at Picnic. *Sclerocactus glaucus* prefers open areas between pinyon-juniper stands in rocky or gravelly soils in desert habitats; therefore, plant cover could have played a role in density differences (U.S. Fish and Wildlife Service, 2021). Shading could be the mechanism behind lower densities; however, some studies indicate that shade can aid in seed germination, growth, and reproduction of cacti (Drezner, 2017; Godinez-Alvarez & Valiente-Banuet, 1998; Raveh et al., 1998; Valiente-Banuet & Ezcurra, 1991). *Sclerocactus glaucus* adults prefer exposed and sandy soils between pinyon-juniper stands, yet it requires precipitation or other water sources for germination, reproduction, and growth indicating the reduced density at T-Junction could be caused by competition for water with surrounding plants (U.S. Fish and Wildlife Service, 2021).

If plant cover is negatively associated with *S. glaucus* density, the invasion of grasses like *Bromus tectorum*, which was found at T-Junction, might threaten this listed cactus. Regardless of the mechanism, plant cover should be incorporated into future studies to assess how it influences *S. glaucus* density and potentially the detectability of the cactus.

The estimated density at Picnic is within the range of the macroplot-level estimates of density from Krening et al. (2021). Their study reported that the density across their 16 subjectively placed macroplots in the southern population ranged from 0.063 individuals/m² to 0.623 individuals/m² and averaged out to 0.234 individuals/m² (90% CI: 0.156, 0.312) before they extrapolated estimates to cover the size of each respective habitat area. Our estimated density of 0.160 individuals/m² across Picnic falls within their 90% confidence interval of average macroplot densities. This indicates that our estimates are within the range of densities from a much more time-intensive survey, yet distance sampling could ultimately provide a less-biased total population size across suitable habitat instead of a minimum estimate if employed across more areas.

The small sample size and lack of comparative census numbers limited the conclusions that could be made about the accuracy of our estimates. While the recommended number of detections for accurate distance sampling results is 60-80 individuals (Buckland et al., 2015), we only detected 25 individuals at T-Junction. The small sample size for individuals under a different plant cover regime may have reduced the precision and accuracy of our estimates from the top-ranked model. Next, we did not have a population size estimate from a census or previous study to which we could compare our estimates of size or precision. To avoid this issue, Flesch et al. (2019) conducted a thorough census of their sampling area prior to distance sampling to compare estimates. Our estimates were not directly comparable to Krening et al.

(2021) because their minimum population size estimates were limited to cacti inside their macroplots, leading to intentional underestimates but certainty about a minimum population size. We recommend future surveys include more sampling sites in the habitat area surveyed by Krening et al. (2021) to accurately compare density estimates and sampling effort.

Plant population size and density estimates are difficult to obtain, especially for sparsely distributed plants or those with non-random distributions. The distance sampling models and simulations we used assumed that *S. glaucus* individuals are randomly distributed across the landscape. *Sclerocactus glaucus* seed dispersal relies on ants and gravity (U.S. Fish and Wildlife Service, 2021), which restricts the distribution of the species and could cause clumped populations. There are other plotless plant sampling methods that account for clumped distributions by measuring distances between systematically placed points or transects and plants (Jamali et al., 2020). Many of these incorporate an extra layer of measurements that allows the surveyor to determine the density and distribution of individuals across an area. For example, the nearest-neighbor method requires the observer to measure the distance to the closest individual after recording the distance of the detected individual from the point or transect (Barbour et al., 1987). The accuracy and precision of different plotless methods on different plant species can be compared using simulations (Jamali et al., 2020) to determine which would require the least effort and obtain the most accurate and precise estimates. Cacti are likely to have clumped populations due to limited seed dispersal, indicating a different plotless method could be more accurate to measure population size.

In our small-scale study, we demonstrate that distance sampling is practical for estimating the population size of a small, inconspicuous plant species, such as *Sclerocactus glaucus*. With proper sampling sizes across multiple habitat areas, distance sampling could be an

accurate and time-efficient way to annually monitor the total population size of *Sclerocactus glaucus* post-delisting. Distance sampling could also be a useful method to estimate the population size of other small or rare plants that have inconspicuous life stages in areas with sparse vegetative cover, especially when conducted over large spatial scales with appropriate sample sizes (Flesch et al., 2019; Jamali et al., 2020; Schorr, 2013). Underestimates or overestimates of population size can severely impact conservation decisions, emphasizing the importance of employing methods that provide accurate estimates of the true population size (Krening et al., 2021).

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CHAPTER 4. STAKEHOLDER ANALYSIS

Protecting the Mojave Desert Tortoise and Relieving Anticipated Congestion

Using an Existing Route in St. George, Utah

Introduction

For the past 30 years, Washington County, Utah has experienced rapid population growth because of improved cooling technology and increasing job opportunities (Langston, n.d.). The economic growth in the area has been boosted by the waves of tourists who visit the scenic desert landscapes at Zion National Park, Dixie National Forest, and the Red Cliffs National Conservation Area (NCA) every year. In 2018, the Utah Department of Transportation (UDOT) applied to the U.S. Bureau of Land Management (BLM) for a right-of-way (ROW) grant to build a multi-lane, divided highway (termed the Northern Corridor) across the southern portion of the NCA (Bureau of Land Management, 2020). The project's purpose is to relieve traffic on Utah's major highway, I-15, into the city of St. George and improve east-west traffic flow. The project triggered a National Environmental Policy Act (NEPA) review which required drafting an Environmental Impact Statement (EIS) to assess significant cultural, environmental, and socioeconomic impacts the project will have. The EIS drafting process involved periods of public opinion, professional consultation, and extensive cooperation with the U.S. Fish and Wildlife Service (USFWS) and the BLM.

The proposed ROW will pass directly across the NCA, fragmenting protected desert habitat and threatening the dispersal and survival of the species that live there. The NCA is a biodiverse arid desert landscape that includes 130 miles of non-motorized trails and stunning views for tourists and local residents. The land acts as a refuge for vulnerable plant and animal

species including some listed under the Endangered Species Act (ESA), most notably the Mojave Desert tortoise (*Gopherus agassizii*). The park has creosote brush habitats with burrows and flowering plants for the critically endangered tortoise (Vaughn et al., 2020) whose density peaks near the southern portion of the park where the proposed highway would be built (Figure 1).

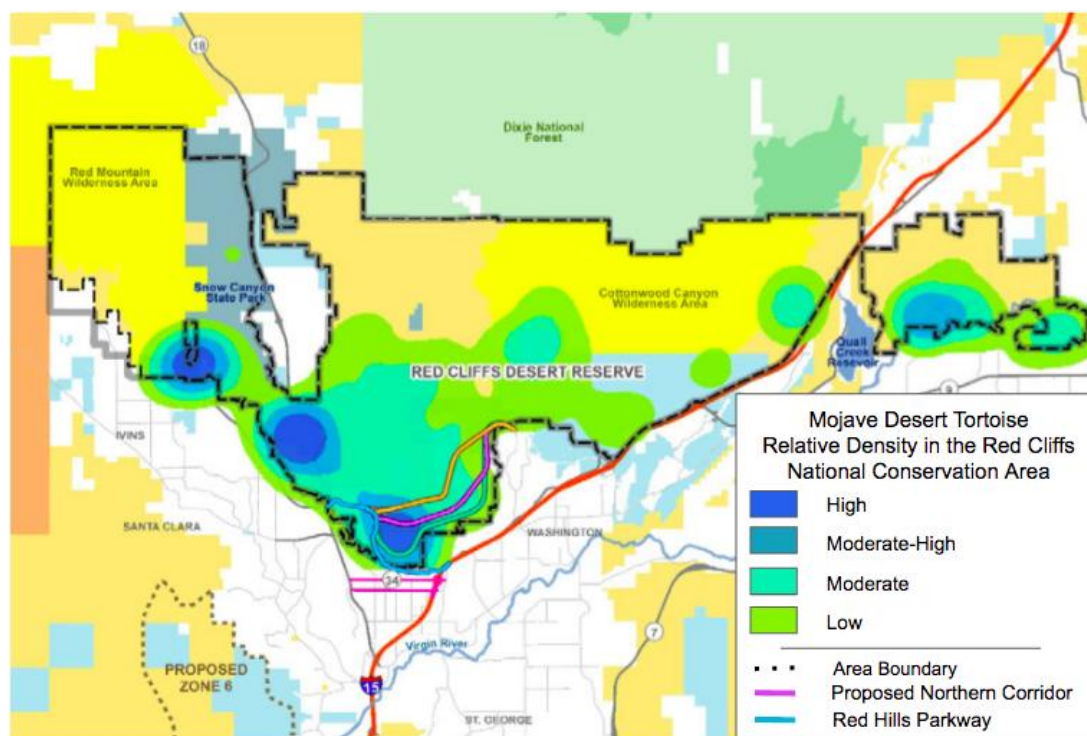


Figure 1. Red Cliffs National Conservation Area and proposed Northern Corridor right-of-way alignment. The relative density of Mojave Desert tortoises (*Gopherus agassizii*) is indicated in the legend (Bureau of Land Management, 2020).

To begin this project four major actions needed to be approved by the BLM and USFWS: the Northern Corridor ROW, the issuance of an incidental take permit for the Mojave Desert tortoise, and two amendments to the Red Cliff NCA’s Resource Management Plan (RMP). The RMP amendments would allow for the construction of a highway on the land and the addition of a 6,800 acre “Zone 6” to the NCA west of St. George that would offset the land lost to the highway (Bureau of Land Management, 2020). In the final days of the Trump Administration, the U.S. Department of the Interior (USDO I) approved the construction of the Northern Corridor.

The approval was quickly followed by litigation from seven environmental organizations against the USDOJ for suspected violations of the Omnibus Public Lands Management Act, the Land and Water Conservation Fund Act, the National Environmental Policy Act, and the National Historic Preservation Act. I propose that UDOT implement an alternative discussed in the EIS that would expand an existing road (Red Hills Parkway) below the southern border of the NCA (Figure 1) instead of significantly fragmenting untouched habitat with the approved road alignment. They could convert Red Hills Parkway to an expressway and thus avoid irreversible damage to Mojave Desert tortoise habitat while reducing congestion and improving east-west connectivity.

Background

Northern Corridor

The Utah Department of Transportation has pushed for the construction of a road across the NCA landscape since 1990 when Utah began experiencing a population boom. A new road was not considered in the 1995 Habitat Conservation Plan (HCP) for the Red Cliffs Desert Reserve nor in the RMP written in 2009 when the land was redesignated as the NCA (Conserve Southwest Utah, 2021). Local officials have repeatedly appealed the NCA's RMP but were rejected numerous times by Congress. In 2015, the HCP was up for renewal, and discussions of constructing a new road that would require the incidental take of endangered species in the park were discussed, keeping the Northern Corridor vision alive. Local elected representatives and UDOT quickly began the process of applying for a ROW that would accommodate the growing local resident and tourist populations. The proposed alignment would relieve traffic on local roads, divert traffic from I-15 north of St. George, and provide a direct east-west passage.

Red Cliffs National Conservation Area

The Red Cliffs National Conservation Area was established by Congress through the Omnibus Public Land Management Act of 2009 in which millions of acres were set aside for protection. Congress designated this 45,000-acre area for current and future generations to enjoy the scenic, wildlife, cultural, and educational resources at Red Cliffs (St. George Field Office, BLM, n.d.). Red Cliffs houses a transitional ecotone where the Colorado Plateau, Great Basin, and the Mojave Desert meet, and as such hosts a mix of wildlife and plant species adapted to survive in the unique confluence of these arid habitats. The NCA also includes part of the ancestral homelands of the Southern Paiute and Pueblo tribes (US Department of the Interior, 2012).

In 2020, two major fires burned over 12,000 acres of the NCA and destroyed habitat for the specialist and threatened species within the park (Sisson, 2021). The 2020 fire may have reduced the local population of the threatened Mojave Desert tortoise by up to 15% (Sisson, 2021). The impacts of these fires on the viable habitat of the tortoise are currently being assessed by the USFWS and were not considered when planning and approving the Northern Corridor project.

Mojave Desert tortoise

The Mojave Desert tortoise was listed as threatened under the Endangered Species Act in 1990 because rapid habitat loss, collection by humans, and upper respiratory tract disease (URTD) had reduced their populations by 50% (Jirik, n.d.). This rare tortoise is listed as *critically endangered* on the ICUN Red List of Threatened Species (Vaughn et al., 2020). Habitat fragmentation continues to threaten the tortoise's ability to find food and mates while increasing the chance for roadway collisions. Individuals range from 2 to 15 inches long, spend

up to 98% of their time in underground burrows, and can live up to 100 years (Southern Nevada District, BLM, n.d.). Harassing, collecting, or harming this species can lead to a \$50,000 fine and one year in prison. The approval of the EIS requires the USFWS to approve an incidental take permit for the project because they are needed when the project *might* result in the take or relocation of the listed tortoise, furthermore the translocation of male tortoises depresses reproduction rates (Mulder et al., 2017). So, even if the contractors lawfully relocate the tortoises found during construction, Mojave Desert tortoise populations may still decline. In 2019, surveyors encountered over 50 tortoises during the original surveys for this project, indicating a dense population surrounding the proposed road alignment (Conserve Southwest Utah, n.d.). The high density of tortoises and recent fire history in the project area has made the RCA Mojave Desert tortoise population even more vulnerable to habitat loss and fragmentation than they were when the EIS was written.

Stakeholders

Local Residents

The population of Washington County is expected to double to 355,000 residents by the year 2045 (Conserve Southwest Utah, n.d.). The residents rely on the improving economy brought by the mining industry and tourists visiting Zion National Park and Red Cliffs who stay and shop in Washington County. Over 16,000 people commented on the draft EIS; these concerns included impacts to local businesses, residential areas, the integrity of the park, and the health of the local community (Bureau of Land Management, 2020). The majority of comments expressed concerns, yet there were also supporters of the proposed new road with hopes for shorter commute times, reduced traffic on local roads, and the implementation of Zone 6 for additional NCA habitat. Reducing congestion and improving connectivity will undoubtedly help

local businesses and lower commute times for local residents. However, the integrity of the natural habitats that bring tourists to the area and pride to residents is a crucial aspect of life in Washington County. The proposed road alignment would diminish the visual aesthetics of the park that draw people in but it would enhance connectivity to reduce local commute times and improve accessibility to local businesses for residents and tourists (Bureau of Land Management, 2020).

Tourists

People from across the world travel to Washington County using I-15 to experience the distinctive and biodiverse habitats that are protected in surrounding national parks and conservation areas. Two hundred thousand tourists visit the NCA every year and over 4.5 million visit Zion National Park (Conserve Southwest Utah, n.d.). The visitors want to immerse themselves in scenic views by hiking, climbing, and biking to some of the most untouched landscapes in the Southwest. The remoteness of the NCA contributes to its aesthetic appeal, enabling tourists to experience a quiet solitude unique to the region. The preservation of these areas and their wildlife is important to tourists, however, highway congestion can degrade their experience (Sundeen, 2020). Therefore, tourists have a conflicting stake in this issue of desiring the maximum visual aesthetics and biodiversity in their visits but also wanting easy travel and convenience (Sundeen, 2020). Constructing a road that maximizes the continued integrity of natural ecosystems and minimizes traffic is crucial to pleasing tourists who travel to the area.

Utah Department of Transportation

The Utah Department of Transportation (UDOT) values and represents the interests of the residents of Utah and tourists who desire reliable travel on Utah roads. UDOT applied to construct the Northern Corridor to improve east-west connectivity and local resident satisfaction

by reducing traffic and congestion on I-15 and local roads (Bureau of Land Management, 2020). The current roads are projected to exceed their previous capacities because of rapid growth in Southwestern Utah and growing tourist pressures. UDOT desires the proposed road alignment (Figure 1) because it is one of the most cost-efficient alternatives and will divert traffic away from local St. George roads (Bureau of Land Management, 2020). Diverting traffic from the city will please both tourists and residents by allowing quick access to the main entrance of the park for tourists, thus relieving congestion on local roads for commuters. Alternatives mentioned in the EIS were not publicly examined by UDOT, and some believe that there were other viable options.

National Conservation Organizations

Conserve Southwest Utah, Conservation Lands Foundation, Center for Biological Diversity, Defenders of Wildlife, Southern Utah Wilderness Alliance, The Wilderness Society and WildEarth Guardians filed a lawsuit against the US Department of Interior in June 2021 after the approval of the highway by the Trump Administration. These organizations represent recreationalists, scientists, or tourists who value the integrity of conservation areas and the protection they bring to the species within them. These organizations and their supporters who value the area's biodiversity, fear that constructing the Northern Corridor will allow development in other protected areas, a move that would threaten endangered species across the country. Many also believe that the alternatives examined in the EIS and those eliminated from the EIS serve as viable options to prevent traffic congestion while maintaining the integrity of protected areas and survival of threatened species. Without a proper explanation of why those options were not selected by the USDO, Conserve Southwest Utah thoroughly analyzed and discussed the possible alternatives that UDOT could use to avoid construction directly across the

NCA (Conserve Southwest Utah, 2021). The alternatives included options discussed in the EIS such as expanding Red Hills Parkway (Figure 1) and boosting public transportation or improving intersections on local St. George roads.

US Department of the Interior

The US Department of the Interior under the Trump Administration approved the BLM's Record of Decision on the final EIS to improve connectivity in Utah and designate 6,800 acres (Zone 6) for species conservation south of the NCA (Bureau of Land Management, 2021). Local officials, such as US senators Mitt Romney and Mike Lee, applauded this effort as a "great win for Southern Utah" and a way to "meet community needs and manage threatened species" (Bureau of Land Management, 2021). The USDO I is led by officials appointed by the president; therefore, their decisions can be biased by politics and the agenda of the administration. The potential partisanship of the USDO I on this issue is why conservation organizations are concerned that the decision to move forward with the proposed road alignment was made too abruptly at the end of Trump's term. The Administration claims their decision sought to relieve traffic and improve the economy in Utah to promote development and small business success, yet the recent litigation against the USDO I alleges that the DOI did not adequately consider less ecologically harmful alternatives.

Solution

A solution that incorporates all of the stakeholder interests and values in this issue includes the construction of a road that will improve east-west connectivity, reduce traffic, and maintain the preservation of the NCA's ecological integrity and the threatened populations of the Mojave Desert tortoise. A pre-existing road called Red Hills Parkway (Figure 1) that travels just North of I-15 before curving around the southern end of the NCA and heading North to the point

where the proposed Northern Corridor would connect with Utah Highway 18. This alternative was listed in the EIS and classified as an “environmentally preferred alternative”, but the Record of Decision claimed the accepted road alignment through the NCA included the implementation of Zone 6, while the Red Hills Parkway alternative did not. The necessity of Zone 6 is diminished with the expansion of Red Hills Parkway because it does not damage ideal tortoise habitat already within the bounds of the NCA. I propose that they convert this road to an expressway by adding lanes on each side and efficient connections to I-15. The diversion of traffic from I-15 would be farther south than the proposed road alignment, yet east-west connectivity would still be improved. The removal of intersections and the addition of proper on- and off-ramps on the new road would further improve connectivity for commuters, tourists, and local businesses, reduce congestion, and fulfill the purpose and need for the action proposed by UDOT.

This solution would also avoid permanent damage to pristine desert habitat by only impacting previously developed land. The unnecessary incidental take of Mojave Desert tortoises would be avoided and gene flow between populations would be maintained to conserve the species. Mesh nets to avoid tortoise collisions already exist along Red Hills Parkway and could be expanded to protect tortoises from cars. The unique NCA habitats would continue attracting tourists to the non-motorized, stunning, and quiet trails in the park and supporting local businesses. The integrity of the NCA and other conservation areas around the park would be upheld by the federal government, fulfilling the interests of the conservation organizations battling the proposed alignment. Overall, the expansion of Red Hills Parkway into a faster, reliable expressway fulfills the interests of all stakeholders while conserving this protected ecosystem.

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