Projecting climatic suitability for a high-elevation amphibian in the Cascades range: a case study of the Cascade frog (*Rana cascadae)*

by

Dylan Kubina

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Dylan Kubina

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by

John Withey, Ph. D.
Member of the Faculty

Date

Abstract

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Amphibians in the Cascades Region are declining. In montane wetlands of the Cascades Range, several species of amphibians have been decreasing in number since the late 1800s primarily due to the stocking of Eastern brook trout (*Salvelinus fontinalis*) that took place in originally fishless lakes. The various trout species introduced to these lakes often prey on the different life stages of the mountain-dwelling amphibians. The present study focuses on the threat of anthropogenic climate change for the Cascade frog (*Rana cascadae*). Cascade frogs occur in high mountain lakes from northern California through Oregon, and northeast Washington. To examine the effects of climate change, I took 1,600 records from a variety of databases to represent known occurrences of Cascade frogs. I used bioclimatic features from Worldclim.org as inputs into the species distribution modeling (SDM) software *MaxEnt* to project the current probability of occurrence for Cascade frogs. In addition, I used four Representative Concentration Pathways (RCPs) from Worldclim.org to project the suitability of where Cascade frogs may occur. *MaxEnt* predicts high probabilities of occurrence of Cascade frogs in Mount Rainier National Park, North Cascades National Park, Olympic National Park, and as far south as Lassen Volcanic National Park and the Mount Shasta region. MaxEnt showed that future projections of Cascade frogs dwindling across their Cascades Range distribution, with increases in suitability moving toward northern California in RCPs 4.5 and 6.0. The most dramatic decreases in Cascade frog suitability was in RCP 8.5; all the Cascade frogs' hot spots showed decreases in suitability, with new suitability appearing in northeast Washington.

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Chapter 1: Introduction and Literature Review

Introduction

Amphibian declines are a global phenomenon. Amphibians are more threatened than taxa such as birds or mammals (Stuart et al. 2004) (Table 2). Montane wetlands (also referred to as high mountain lakes, or mountain wetlands) are particularly affected as climate change is reducing habitat for amphibians in these regions (Lee et al., 2014). Warming climate has led to habitat drying and reduced hydroperiods, which is believed to have promoted the decline of many amphibian species (Carey & Alexander, 2003; Ryan et al., 2014, Lee et al., 2014).

In the Cascade Mountain Range of Western North America, climate change co-incident with the introduction of novel predatory fishes in the 1800s, namely several species of trout, has also led to a decrease in amphibians (Bahls, 1992; Drake & Naiman, 2000; Knapp, Corn, and Schindler, 2001; Pilliod & Peterson, 2001; Pister, 200; Ryan et al., 2014). Precipitation, snowpack, and evaporation are the most important factors reflecting the warming climate in the montane wetlands. Once fishless lakes are now facing many disturbances, however this study will focus specifically on the Cascade frog (*Rana cascadae)* which occupies the Cascades Range of the western U.S. The present study aims to examine both abiotic and biotic factors in relation to how climate change may be influencing the habitats of the decreasing Cascade frog.

Montane wetlands are among the most sensitive ecosystems to climate change (Lee et al., 2014). They include lakes found in high elevations with surrounding sub-alpine to alpine regions. The regions offer habitat to a large range of species and are part of crucial hydrologic and geochemical processes (Lee et al., 2014). Humans use the land more frequently in lowland wetlands, which causes those areas to sustain more impacts. Relatively untouched by human development, montane wetlands areas are more affected by climate change impacts. Montane wetland areas have a diverse vegetation and fauna, which make them unique and unblemished in relation to many other parts of the world (Ryan et al., 2014; Lee et al., 2014). Unfortunately the introduction of trout to has affected many of these ecosystems. State fisheries managers estimated 95% of roughly 16,000 large mountain lakes in the western U.S. were naturally fishless, but now almost all the lakes contain fish (Bahls, 1992; Drake and Naiman, 2000, Knapp et al., 2001). Now, 60% of smaller ponds and lakes contain fish (Ryan et al., 2014). Around 60% of the total number of lakes and 95% of deeper lakes (>3 m) and larger (>2 ha) lakes contain trout. There is little research on base stocking programs (Bahls, 1992).

Climate change typically affects the mountainous ecosystem most severely because of the unique abiotic factors such as high elevation and permanent snow. Wetlands themselves are important globally because they are critical for both natural communities and human habitat. Since wetlands are so vulnerable to the impacts of climate change, focused research is necessary to address why species such as the Cascade frog are in decline (Lee et al., 2014).

The Cascade frog is an endemic flagship species of the Cascades, a volcanic mountain range of the Western US. Cascade frogs also have an isolated population system in the Olympic Mountains. Looking at frog populations in montane wetlands of the Cascades Range over the last 100 years has indicated significant decline in most mountain-dwelling frogs since the late 1800s (Bahls, 1991; Knapp et al., 2001; Pilliod and Peterson, 2001; Pister, 2001; Ryan et al., 2014). Although the Cascade frog is not federally listed, climate change coupled with other abiotic and biotic influences could lead to serious future declines for the Cascade frog. The Cascade frog is listed as near threatened (NT) by the IUCN (IUCN, 2004; NatureServe, 2017).

Abiotic features are crucial when discussing an amphibian's population status in high mountain lakes. Climate change influences abiotic features such as precipitation and temperature most directly, leading to earlier snowmelt at more rapid rates. Increased warming in all seasons leads to earlier snowmelt, which in turn, leads to loss of mountain snowpack and increased evapotranspiration and increased soil-moisture in late summer (Lee et al., 2014). Duration and severity of cold weather of both the surroundings and water temperature are important abiotic factors affecting Cascades high mountain lakes. Lastly, hydrological (i.e. groundwater recharge, surface water storage, and filtration) and geochemical cycles (nutrient cycling, carbon sequestration, sediment and contaminant transport) exist that greatly affect populations of the Cascade frog. Montane wetland ecosystems tend to be understudied, so looking in greater detail can help provide links to biotic features and climate change (Ryan et al., 2014; Lee et al., 2014).

Biotic Factors

Biotic factors affect Cascade frogs through processes such as nutrient cycling and food webs (Ryan et al., 2014). Climate change is proposed to influence biotic factors as well such as warmer temperatures year-round leading to earlier and faster wetland drawdown rates. This causes water surface levels to go down and therefore take away habitat from species in addition to amphibians (Ryan et al., 2014; Lee et al., 2014). Biotic factors that heavily influence Cascade frogs include vegetation, the amphibian chytrid fungus, and introduced trout (Bahls, 1992; Knapp et al., 2001; Pister, 2001; Ouellet et al., 2005; De León, Vredenburg, and Piovia-Scott, 2017). Vegetation provides habitat and a food source for Cascade frogs (Ryan et al., 2014). At the lower elevations of the Cascade frog range, well-developed coniferous forest dominates the habitat (US EPA, 2017). Cascade frogs in Crater Lake tend to reside along the shoreline areas of ponds, tarns and lakes, where they may conceal themselves under woody debris surrounded by moss and other wetland plants (Farner and Kezer, 1953).

Prominent among biotic factors thought to affect Cascade frogs is the fungal disease, chytridiomycosis, that prominently affect amphibians globally (Ouellet et al., 2005; De León, Vredenburg, and Piovia-Scott, 2017). This fungus has profound effects on amphibian populations globally as it is thought responsible for the extirpation of up to 200 species of amphibians. The chytrid fungus (*Batrachochytrium dendrobatidis* or *Bd*) that causes chytridiomycosis has dramatically affected amphibians in Central America, but it may be the biggest threat to montane populations of amphibians globally. Studies reveal that montane-dwelling amphibians like the Cascade frog such as the two species of Mountain yellow-legged frogs (*Rana muscosa* and *Rana sierra*) are species prominently affected by the amphibian chytrid fungus (Ouellet et al. 2005). In northern California, chytrid fungus has been speculated to be the cause of large declines in distribution of the Cascade frog. Laboratory experiments show that post-metamorphic Cascade frogs are susceptible to *Bd* (Garcia et al., 2006; Piovia-Scott et al., 2015). The most dramatic declines of Cascade frogs in northern California are thought to be because of *Bd* (Fellers et. al., 2008; Pope et al., 2014; Piovia-Scott et al., 2015). Although *Bd* is widespread throughout the range (Adams et al.. 2010; Piovia-Scott et al., 2011), declines have not been observed everywhere (Pear et al., 2009) suggesting that there is variation in the effect of *Bd* on populations of this host amphibian (Piovia-Scott et al., 2015).

Aquatic taxa are also important biotic elements of the Cascade frog landscape. Introduced trout represent an especially high risk to frogs due to predator abilities. Trout presence trout has led to trophic cascades as well as drops in occupancy levels (Bahls, 1992; Knapp et al., 2001; Pister, 2001). Trophic cascades occur due to consequences of novel predator introduction. In this case, the trout eat the Cascade frog's eggs, thus reducing the number of Cascade frogs. The balance of the food web changes due to larger, native predators that would have typically fed on Cascade frog life stages having less to feed from, so they also experience declines in population. Strong feedbacks take place between fish, herbivores, and algae (Drake & Naiman, 2000). These feedbacks are related to the addition of top predators that alter lake food chains through trophic cascades. Perturbations also take place in food-chain structure and primary production (Carpenter and Kitchell, 1993; Drake & Naiman, 2000).

Climate Change Influences

The Cascades are seeing climate change from human influences. Anthropogenic climate change appears to be eliminating crucial habitat for amphibians that occupy these mountain lakes. In fact, the changing climate is thought to place amphibians; in this case, the Cascade frog, at risk of being extirpated (Ryan et al., 2014). It has been predicted that by the 2080s, (Lee et al. 2015) climate change will cause a reduction in wetland habitat availability for many species, namely amphibians (Lee et al., 2014). Climate change effects can be seen impacting amphibian populations already in many sections of the western U.S. (Carey and Alexander, 2003; Ryan et al., 2014; Lee et al., 2014). In the Cascades Region, many amphibians could be losing habitat crucial for breeding and overwintering (Ryan et al., 2014).

The first section of this thesis will investigate existing peer-reviewed literature to ascertain what information exists for amphibians in the western US. After I describe abiotic and biotic factors in some detail, I will discuss my modeling results and explore projections of how areas of climatic suitability may be changing for Cascade frogs. I will discuss reasons for decline, future suggestions, and future implications for the Cascade frog and mountain-dwelling amphibians in general.

Literature Review: Abiotic and Biotic Factors in High Mountain Wetlands that Affect Amphibians of the Western United States

Introduction

Amphibians in the high-elevation lakes of the Western United States (US) are dwindling now more than ever due to climate change and the introduction of trout during the 1800s. Climate change affects abiotic and biotic factors (i.e. vegetation), potentially leaving amphibians with reduced suitable habitat. Evidence increasingly suggests that climate change leads to a decline in amphibians, however, more research is needed to demonstrate that some aspect of climate change is the causal factor. Some data have revealed indirect effects of climate change, for example, on the earlier initiation of breeding activities (Carey & Alexander, 2003). Since amphibian population decline reflecting climate change has received relatively little attention since the turn of this century, this thesis focuses on examining how different climate change scenarios may influence Cascade frog distribution, and consider that in context of trout introduction and other abiotic factors (Carey & Alexander, 2003; Linder et al., 2003).

Understanding historical accounts from previously published works is crucial to observe why certain events are taking place. This literature review covers a history of the trout introduction of the 1800s, which have been introduced to many montane lakes in the western United States. After I discuss this historical background, I will discuss important factors likely to affect Cascade frogs, including the impending changes in the climate, and other abiotic as well as biotic factors. I will use past research to describe six different types of montane wetland amphibian investigations: 1) experiments based on habitat segregation, 2) paleolimnological observations**,** 3) removal restoration techniques, 4) trout removal experiments, and 5) amphibian and trout abundance findings, and 6) low elevation/artificial ponds

Trout Introduction History

A variety of fish-stocking clubs brought fish into the montane wetlands throughout the 1800s and 1900s. The US Forest Service, which was established in 1905, stocked trout for recreational purposes. While they stocked trout for sport, other local fish and hunting clubs introduced trout illegally for resource purposes for years and continue to do so to this day (Bahls, 1992; Pister, 2001; Ryan et al., 2014). Drake and Naiman (2000) discovered from personal observation that fish in the Mount Rainier National Park (MORA) had been stocked illegally.

The elevated lakes within the mountains of the western U.S. are landscapes dating from the end of the Pleistocene epoch. Recession of glaciers, which took place over

10,000 years ago during this epoch, created many lakes impassible to entry by fishes because of barriers. The fishless lake's biota was fragile due to set up of nutrient cycling occurring in the lakes. Lakes that are fishless contain important habitat, refuge, and routes of recolonization for plants, invertebrates, and vertebrates (Drake & Naiman, 2000). Other semi-aquatic species, that could negotiate those barriers, such as amphibians, prospered here thousands of years ago in lake environments devoid of fish (Knapp et al. 2001).

European settlers facilitated the introduction of trout into the lakes for sport fishing (Bahls, 1992; Drake & Naiman, 2000; Knapp et al., 2001; Pilliod & Peterson, 2001 and Pister, 2001). Due to technological advances, the U.S. Forest Service could introduce trout at more rapid rates (Bahls, 1992). In the 1940s, aircraft became a common method of transporting the trout into the lakes. A variety of trout species have been stocked in western lakes over the years (Table 1). In 1964, stocking came to a halt in Wilderness lakes that had been stocked prior to their Wilderness designation due to federal guidelines (Bahls, 1992). Fish stocking ceased in 1963 in MORA because the Leopold Report (Leopold et al., 1963), which tried to urge against the trout introductions in naturally fishless lakes solely for the sake of recreation (Drake & Naiman, 2000). Leopold was more concerned for the conservation of the parks rather than the prospect of sports and resource management gain. Again, in the 1970s and 1980s, fish stocking was stopped in most national parks in the Western U.S. (Knapp et al., 2001). However, the U.S. Forest Service continued stocking fish in other unprotected areas in the montane West (Landres et al., 2001; Knapp et al., 2001). Roughly 5% of about 16,000 mountain lakes in the Western US were estimated to contain fish, but now almost all the lakes in these montane areas contain some species of trout (Bahls, 1992; Drake & Naiman, 2000). Historical introductions occurred in a near vacuum of scientific knowledge on the ecological impacts of trout in montane still-water systems. Native Americans may have had knowledge about the biota found in these high mountain regions, but this information was not transmitted to European settlers, who planted trout for the sake of sport and resources. When trout additions first started, European settlers were interested in using the land available to them to their benefit (Pister, 2001). Amphibians might have fared better had the trout species died off over time. Instead, entire trophic systems underwent changes. Fish continued to survive after stocking ceased (Knapp et al. 2001). The management of trout continues to be a controversial issue because many people desire the ability to fish recreationally in the high mountain lakes, while others realize how harmful these non-native predators are to the ecosystem and how amphibian occupancy levels are depressed because of fish-altered trophic cascades (Bahls, 1992; Knapp et al. Schindler, 2001; Pister, 2001).

Table 1. Trout Occupying the Montane Wetlands of Western North America

Standard English Name (aka Common Name)	Scientific Name	Use in High Lakes Stocking
Cutthroat Trout	*Oncorhynchus clarki*	Infrequent
Rainbow Trout	*Oncorhynchus mykiss*	Widespread & Frequent
Kokanee Salmon	*Oncorhynchua nerka*	Selected Larger Lakes
Eastern Brook Trout	*Salvelinus confluentus*	Widespread & Frequent

Table 2. Amphibians Occupying the Montane Wetlands of Western North America

Standard English Name (aka Common Name)	Scientific Name	Distribution
Long-toed Salamander	*Ambystoma macrodactylum*	CA, ID
Northwestern Salamander	*Ambystoma gracile*	BC, CA, OR, WA
Western Toad	*Bufo boreas*	AK, BC, CA, CO, ID, MN, NV, OR, UT, WA, WY
Yosemite Toad	*Bufo canorus*	CA
Southern Treefrog	*Pseudacris hypochondriaca*	BJ, CA
Northwestern Treefrog	*Pseudacris pacifica*	BC, OR, WA
Pacific Treefrog	*Pseudacris regilla*	CA, OR
Cascade Frog	*Rana cascadae*	CA, OR, WA
Columbia Spotted Frog	*Rana luteiventris*	AK, BC, CA, ID, MN, NV, OR, UT, WA, WY
Southern Yellow-legged Frog	*Rana muscosa*	CA
Sierran Yellow-legged Frog	*Rana sierrae*	CA, NV
Rough-skinned Newt	*Taricha granulosa*	AK, BC, CA, OR, WA

Province or State Abbreviations are: Alaska (AK), British Colombia (BC), Baja California (BJ), California (CA), Colorado (CO), Idaho (ID), Montana (MN), Nevada (NV), Oregon (OR), Utah (UT), Washington (WA), and Wyoming (WY).

The Cascade Frog Biology

The Cascade frog is a high mountain frog, which live for around 5 years and can survive under 30 feet of snow. Mottled yellow, tan, brown-olive skin with black spots on the yellow areas allows the amphibian to stay relatively hidden in the temporary pools where it hides (Farner & Kezer, 1953; Nussbaum et al., 1983; NatureServe, 2017). The frog breeds in spring-summer, from March to August, directly after ice and snow melt (Stebbins, 1985; NatureServe, 2017). Most eggs are laid out over a few days in a pond and each female lays around 300-500 eggs, most often in aggregations. Tadpoles metamorphose into frogs about 2-3 months after the female laid their eggs. It is speculated that frogs breed after their third hibernation (Nussbaum et al., 1983; NatureServe, 2017). Overwintering takes place in late fall. Cascade frogs are found congregated in spring-fed ponds and perennial streams (Garwood, 2009; Pope et al., 2014). The frogs were found overwintering in deep, loose silt at the bottom of a pond (Briggs, 1987; Pope et al., 2014).

Factors Relating to Decline of Cascade Frogs

Factors influencing declines in amphibians globally are diverse (Stuart et al. 2004). Factors thought to influence the decline of Cascade frogs represent a subset, most prominently introduced trout, solar UVB radiation, fungal pathogens, and loss of meadow habitat from fire suppression (Hayes and Jennings 1986, Fellers and Drost 1993, Blaustein et al. 1994, Fite et al. 1998, Adams et al. 2001). Declines in Lassen Volcanic National Park, which are particularly severe, most likely reflect a combination of local factors, including (1) presence of non-native predatory fishes that have restricted available habitat and limited dispersal of frogs, (2) gradual loss of open meadows and associated aquatic habitats, and (3) loss of breeding habitat due to drought (Fellers and Drost 1993); and 4) based on more recent data, the amphibian chytrid fungus (Piovia-Scott et al. 2015). Climate change is an abiotic shift resulting from changes in biotic influences from humans. Climate change influences both abiotic and biotic factors, which in turn affects amphibian habitat. Moreover, this region in Northern California may be more strongly influenced by climate change than areas further North in its geographic range (i.e., Oregon and especially northern Washington). Cascade frogs have declined greatly near Lassen Volcanic National Park at the southern end of the range in northern California (Fellers and Drost, 1993). Jennings and Hayes (1994) and Fellers and Drost (1993) estimated that this species is extirpated from about 99 percent of its southernmost population clusters (Mt. Lassen and surroundings) and 50 percent of the total historical distribution in California. Cascade frogs in their northern range of Oregon and Washington appear to face fewer/less severe threats posed by climate change and habitat loss. Declines in Oregon were cited by Nussbaum et al. (1983) and Blaustein and Wake (1990), but other data found in the literature do not suggest low occupancy rates (Brown 1997, unpublished data cited by Pearl and Adams 2005). In Oregon, it has been estimated that a 22% decline in Cascade frog populations has occurred over the past hundred years (Fite et al. 1998). Cascade frog declines may be related to the sensitivity of their eggs to increased levels of ultraviolet-B radiation resulting from ozone depletion (Blaustein et al. 1994). However, this hypothesis is controversial because the abiotic spectral characteristics of natural waters likely shields eggs from most detrimental physiological effects of UVB (Palen et al. 2002).

Water quality, physical characteristics and nutrient concentrations are among the most important factors scientists have identified that influence amphibian habitats (Ryan et al., 2014). These abiotic factors as also interact with biotic factors such as vegetation and fauna. Abiotic features that appear to be important include precipitation and temperature, whereas important biotic features of influence include vegetation that the Cascade frog thrives off and fungal diseases. After all, the Cascade frog hides under vegetation and needs it in the lakes to support their nutrient cycling. The frog lives in sphagnum bogs and lay their eggs in submerged vegetation (Briggs, 1987). They tend to eat algae, detritus, and plant tissue from the vegetation (NatureServe, 2017). Fungal diseases, which directly affect amphibian health, have spread globally (Piovia-Scott, 2015). Changes due to climate change and introduction of fish are influencing changes in the ecosystem that do not just impact the Cascade frog, but other species as well.

Various political and conservation entities have given a status to Cascade frogs reflecting the level of threats and the overall risk to the species. For example, both the States of California and Oregon list it as a Species of Special Concern, a non-legal designation indicating that listing of the species may be justified at some point. The species has no such designation in the State of Washington. Further, the US Fish and Wildlife Service consider the Cascade frog a Species of Concern, a designation that carries a meaning like the state-level Species of Special Concern. Based on the IUCN Red List (2004), the Overall Threat Impact to the Cascade frog has a near threatened (NT) ranking. Collectively, all designations for the Cascade frog indicate a level of concern that, though not severe, bears watching.

Abiotic Factors Affecting Amphibians

People tend to overlook montane wetland ecosystem's abiotic factors in literature regarding amphibians (Ryan et al., 2014; Lee et al., 2014). For example, these factors can provide powerful insight on habitat changes and loss of biodiversity (Ryan et al., 2014). Temperature and precipitation are two of the most important abiotic features that are expected to change the most because of climate change. High elevation coupled with warming temperatures are factors that scientists and ecologists take into consideration when performing montane research. This pattern can lead to substantial changes in amphibian habitat due to the rapid and earlier recession of snow (Carey and Alexander, 2003; Ryan et al., 2014; and Lee et al., 2014). Climate change also influences water storage, nutrient cycling and carbon sequestration. These processes are not unique to the high mountain wetlands, but they do negatively affect these sensitive, yet unique landscapes. Water storage experiences changes related to earlier snowmelt. Snowpack has declined by greater than 50 percent in some regions over the last half century. An altered hydrology system means there will be shifts in nutrient transfer, which could throw off the entire ecosystem. Also, soil inundation can take place, resulting in different rates of carbon sequestration and release (Ryan et al., 2014; Lee, 2014). Ryan et al. (2014) used object-based remote sensing is used to categorize wetlands and to classify the distribution of wetlands within different regions. This method was seen in Ryan et al. (2014) study with the use of Variable Infiltration Capacity (VIC), which is a coarse-scale model projection of change in how likely it is for drying to occur in intermediate hydroperiods wetlands in Washington state (Hamlet et al., 2005; Ryan et al. and 2014; Lee et al., 2014). There are different types of ponds within the montane wetlands, including: 1) hydrologic intermediate pools that are places that hold water for most years, but may sometimes dry up, 2) perennial ponds, which are usually inundated by water, but with fluctuations in volume, which allows for a broader range of species, and 3) permanent ponds, which always have water in them and provide habitat for the broadest number of species (Ryan et al., 2014; Lee et al., 2014). These ponds all contain varying species of amphibians. Perennial ponds are more biodiverse and contain permanent obligate or facultative aquatic amphibian adult life stages. The Cascade frog can be observed in either intermediate or perennial ponds. Intermediate ponds are also at a high risk to climate change because they are vulnerable to drying up and experience a hydroperiods anywhere from around 2-5 years. The shorter the hydroperiods, the more at risk the pond.

The physical characteristics that make up most high mountain lakes have changed significantly over the years. Drake and Naiman (2000) quantified a lake's environmental conditions using dissimilarity in diatom composition and relative abundance. Dissimilarities in diatom assemblages can help elucidate past biological details of lakes or ponds, and trajectories of change. Dissimilarity analysis can provide an index of disturbance and identify baseline conditions (Drake & Naiman, 2000). Dissimilarity analysis of diatoms is based on findings from sediment samples. Sediments revealed that for any particular lake, organic matter was consistent within each core, but an increase in organic matter was evident in recent years. This could reflect in lake productivity or organic terrestrial input. Lastly, invertebrate's assemblages in lakes varied, but variation did not appear to reflect fish introduction.

Observing the connections between abiotic and climate change factors is fundamental to understanding the Cascade frog's response to climate change in montane wetlands because the two are inextricably intertwined. Elevation, temperature, physical characteristics of the lakes, water quality, water storage and carbon sequestration were all factors researchers considered when analyzing why amphibian populations globally and regionally are declining. Emerging diseases, UV radiation and synergistic interactions between these factors and others have been shown to contribute to amphibian declines (Vredenburg, 2014). Gradients in predation are also associated with the aforementioned hydrologic characteristics and codetermine the distribution of species in varying habitat types (Welborn et al., 1996; Snodgrass et al., 2000).

Biotic Factors Affecting Amphibians

Biotic factors can be as important as abiotic factors when assessing amphibian occupancy in the montane wetlands. Many types of biotic factors that can influence the population status of amphibians; this literature review focuses on vegetative features, animal species that are part of the ecosystem, interactions with predators (including trout), and fungal disturbances. Vegetation, aquatic and terrestrial species are the main categories of biotic factors that influence montane amphibians.

Cascade frogs occur in wet mountain meadows, mossy bogs, and in patchy coniferous forests. They tend to lay their eggs in shallow water or among submerged vegetation (Briggs, 1987). The frog adults eat mostly invertebrates, algae, detritus, plant tissue and small organisms. Egg masses can be found at the surface of shallow water where emergent vegetation is present. They have also been found in free-floating in lakes (Garwood et al., 2009; Pope et al., 2014)

Cascade frogs share the ponds with many other animal species. The predation of fish can exclude many native species from viable habitat. Cascade frogs often encounter other amphibians such as Long-toed salamanders, Western toads, Northwestern salamander, mosquito larvae, caddis flies and mayfly larvae. The effects of climate change and trout introduction may also lead to loss of biodiversity and loss of environment. Habitat diversity is crucial when maintaining high levels of biodiversity. Climate change can limit the levels of biodiversity in montane wetlands, so montane wetlands are at risk now more than ever to losing vertebrate and invertebrate species (Chesson, 2000; Whittaker, 2001; Ryan et al., 2014)

Different species interact with each other in diverse ways, with predation being a leading cause to changes in the amount of habitat biodiversity (Knapp et al., 2001). Gradients in predation determine the patterns of amphibian life cycles (Welborn et al., 1996; Snodgrass et al., 2000, Ryan et al., 2014). Ryan et al. (2014) explains how the presence of large salamanders and some dragonfly larvae, both predators, can limit larvae production of smaller amphibians. This will result in less breeding and rearing in the habitats that the predators occupy (Hoffman et al., 2003; Ryan et al., 2014).

The trout introduction of the 1800s led to altered trophic patterns, manifest as trophic cascades, leading to changes in primary productivity, food chain structure, and processes, such as nutrient cycling (Knapp et al., 2001; Kats and Ferrer, 2003, Ryan et al., 2014). Trophic cascades arise when a novel predator is introduced, in this case the trout, and that disproportionally consume the early life stages of the amphibians, thus reduced the likelihood the enough individuals will survive to adulthood to reproduce. Such a shift reduces the resource base for larger, natural, predators of the amphibians, which may decline as well if they are unable to switch to alternate prey or emigrate. However, alteration of trophic pathways is not the only consequence of trout introduction: interactions between fishes, herbivores and algae, which are strong, can also be altered (Drake & Naiman, 2000). The predation on the Cascade frog by trout results in changes in the entire biological structure of montane wetlands. Intense predation by introduced trout has led to alterations in the food web and in nutrient dynamics (Ryan et al., 2014). Pond-breeding amphibians as well as invertebrates have adapted to fluctuating environments. Over time, amphibians have enhanced their natural resilience. Amphibians have large clutch sizes buffer populations per location. This means they persist through poor years and rebound as soon as conditions improve (Ryan et al., 2014).

Emerging fungal diseases represent a relatively novel factor influencing amphibian populations. For example, the amphibian chytrid fungus (*Batrachochytrium dendrobatidis*), the causal agent of the fungal disease, chytridiomycosis, has devastated amphibians in the Central American tropics, including several montane forms (Berger et al., 1998). This fungus may be the biggest threat to the world's montane amphibians. In California, the two species of Mountain Yellow-legged frog appear to be naturally more susceptible to the chytrid fungus than many other frogs (Ouellet et al., 2005). Temperate zones, such as the Pacific Northwest (PNW), must also deal with chytridiomycosis. The Mountain Yellow-legged frogs in California are two of several hundred amphibians globally that have been affected by this disease. The fungus attacks keratinized areas of a frog's body. The fungus does not severely affect tadpoles because they are only heavily keratinized on their jaw sheaths and tooth rows. In contrast, post-metamophic frogs have keratin-rich skin and suffer worse infections (De León, Vredenburg, and Piovia-Scott, 2017).

Varied biodiversity, trophic processes, amphibian life cycle patterns and fungi make up biotic features experienced in the high mountain lakes. These influence abiotic factors and vice versa. Changes in temporal pulses of peak water may strongly influence several biotic features. This affects local pond metabolism and primary productivity, the structure of plant communities and the patterns of wildlife connectivity (Mitsch & Gosselink, 2007; Lee et al., 2014).

Climate Change Factors Affecting Amphibians

Climate change may be negatively impacting montane wetlands (Ryan et al., 2014; Lee et al., 2014). To understand these impacts, the historical backdrop to montane wetland formation must be understood. Montane wetlands reflect repeated glaciation during the Pleistocene era (defined as the time that began 1.8 million years ago and lasted until about 11,700 years ago). When the glaciers receded and the most recent ice age occurred, most lakes and ponds we see today were formed. These lakes, mostly devoid of fish (Knapp et al., 2001), provided habitat for both aquatic and terrestrial species (Lee et al., 2014).

Climate change may be affecting the way the different amphibians breed and survive. Snow, a vital part of the hydrologic cycle in the western US, appears to be one of the pivotal abiotic factors being affected by climate change. In montane wetlands, amphibians may be negatively impacted by the reduced snowpack and more rapid snowmelt earlier in the year. Anthropogenic climate change has resulted in warming air temperatures and decreased precipitation as snow (more as rain), which melts available snowpack more rapidly, and increases plant evapotranspiration and soil moisture stress in the later summer months (IPCC, 2007; Ryan et al.,2014; Lee et al., 2014). High mountain lakes may be drying up earlier or, in some cases, permanently due to this warming weather pattern. Water will be at lower levels in the wetlands, which can increase amphibian mortality from drought. Earlier seasonal rates of wetland drawdown and increased drought frequency in the summer months especially have been shown to place amphibians at risk (Carey & Alexander, 2003; Ryan et al., 2014; Lee et al., 2014).

The undesirable effects of climate change are anticipated to be felt strongly across the Western U.S., including the PNW, with higher temperatures and much more severe summer droughts (IPCC, 2007; Ryan et al., 2014; Lee et al., 2014). Until recently, most studies conducted in the Western U.S. have tended to focus on the effects of trout introduction that began in the 1800s, however, few studies have focused on climate change as the main factor in amphibian declines. Carey & Alexander (2003) focused on the rapid warming that took place during the latter half of the 20th century. Temperatures now rise at about 0.5°C with an increase in the severity of weather patterns across the world (Easterling et al., 2000; Carey & Alexander, 2003). This is a fact that must not be taken lightly, especially for the sake of declining amphibians.

There are many factors that can lead to climate change in the montane wetlands, pollution being a main factor. Greenhouse Gases (GHG) are gas that trap heat in the atmosphere: carbon dioxide, methane, nitrous oxide, and fluorinated gases such as hydrofluorocarbons. Changes in GHG emissions and aerosols can lead to alterations in the energy balance of the climate system, therefore leading to potential climate change

(ICPP, 2007; US EPA, 2015). Besides GHG emissions, motorization of transport has created congestion and pollution in large cities, which leads to climate change (IPCC, 2007). Ryan et al. (2014) illustrate the significance of using simulations to predict hydrologic changes, which can be used to demonstrate interactions with pollution. Increased levels of air pollution and pesticides were also highlighted in Vredenburg's (2004) discussion about the endangered mountain yellow-legged frogs. Wetland dynamics and hydrologic features can be forecasted over broad geographic regions using predictive modeling programs, such as object-based remote sensing. The predictions have emphasized not only the impact that trout introduction has had on wetland ecosystems, but the clear negative impacts felt in relation to climate change (Halabisky et al., 2011; Ryan et al., 2014). Coupling these climate-predictive models to amphibian occupancy patterns, it become possible to identify vulnerable areas and meta-population or landscape-scale resilience of wetlands and amphibians.

Amphibian Studies in Montane Wetlands Performed in the Western U.S.

Habitat Segregation

Hoffman et al. (2003) performed intensive research on both the Long-toed salamander and the Northwestern salamander within MORA addressing what influenced their distributions, and was meaningful in demonstrating now two species can partition their habitat. Physical characteristics, water quality and nutrient concentrations of the 27 sites and 22 ponds and lakes were measured the Northwestern salamander tended to occupy larger, deeper lakes at somewhat lower elevations that contained coarse woody debris, whereas the Long-toed salamander tended to be found in smaller, shallower water lakes and ponds at higher elevations in areas with substantial emergent and aquatic vegetation habitat in the surrounding area. This habitat segregation reflects the life history and interspecific interactions of each species. The most valuable take-away from this habitat segregation-based study, is that larvae from montane populations develop much more slowly than populations from lower elevations. The effects of climate change might cause the populations to develop at even slower rates in higher elevations. It does not help that higher elevations are more vulnerable to the changes of earlier snowmelt and overall warmer temperatures. The amphibians dwelling in MORA lakes and ponds may not have the time to adapt to the rapid changes that are taking place in higher elevations, thus leading to declines in distribution (Hoffman et al., 2003).

Paleolimnological Observations

Paleolimnological studies analyze indicators in lake sediments, such as diatoms or invertebrates to draw conclusions about, or make comparisons to, a lake's current state. Drake and Naiman (2000) indicate that fish introductions in MORA have led to many detrimental effects revolving around amphibians and climate change. These paleolimnological studies were conducted 20-30 years after fish removal had taken place. Dissimilarity analyses of diatoms were used to compare the current physical conditions of the lakes to how they looked before the trout were removed. Eight lakes were examined; diatoms were used as the indicator because they are very sensitive and can reflect both damage and recovery in lakes, as scientific experiments and analyses have demonstrated (Drake & Naiman, 2000). The article stressed just how bad sport fish are to lakes that previously never had any fish in them. The dissimilarities showed that the introduction of trout led to many state changes in lakes. There were dramatic changes in diatom communities, but the researchers also observed that there were several sustained state changes which took place hundreds of years before European settlement. Although these lake systems have a great deal of capacity to adapt to changes in the climate, the resiliency is still limited. At some point in time the system loses the ability to absorb change and will shift into another condition. Ecological conditions in stocked lakes may have passed the threshold of change, which exceeds the bounds of resiliency (Drake & Naiman, 2000).

Removal Restoration Techniques

Drake and Naiman (2000) also conducted a removal experiment to see if lakes could return to predisturbance conditions. Previous studies lacked sediment records. In 1996 and 1997, sediments cores were collected in the deepest sections of the eight lakes. Half the lakes had previously introduced fish; the other half had never had fish in them. The researchers chose to observe seven lakes; three lakes occupied by trout, two that were previously stocked, but that now do not have any trout in them due to removal and two that were never stocked. An additional 'restored' lake that was not one of the original four lakes was added in two years after the start of the study; this lake once had fishes, but then they were removed. Sediment cores provided a 480-year-old record from the eight lakes based on the 210-lead isotope (^{210}Pb). A general model of dissimilarity (SDC), which was calculated by comparing each assemblage in the 480-year old core samples to lake-specific baseline values (Drake & Naiman, 2000).

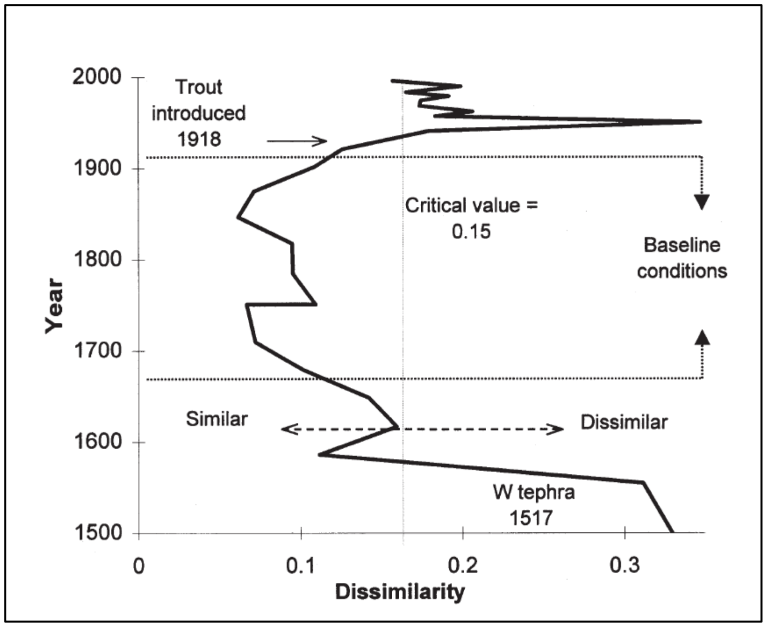

Figure 1. General Model of Dissimilarity (SCD) Time Series. The model is general and not based on real data. SDC analysis is typically based on comparing proportions of species in the different strata. As time increases, assemblages become more dissimilar. When an SCD comparing two different assemlages surpassed 0.15, the two assemblages are considered different (Drake and Naiman, 2000).

With proper baseline conditions, meaning diatom floras were relatively stable between 315 and 90 years before being present in all lakes, then the lake is at a critical value of 0.15 generated in the general model of dissimiliarty (SCD) time series. As time increases, assembledges become more dissimilar. When two assemblages exceed this critical value of 0.15, the two assemblages are considered different. Several lakes in MORA are undergoing fish removal programs. Since 1973, fishes have either disappeared naturally or have been removed by angling (Drake & Naiman, 2000). The two lakes that were known to once contain fishes and now that currently have do not contain any fishes (Eunice and Owyhigh) contained diatoms, which did not return to, nor show a trend toward pre-disturbance conditions. The explanation for why these lakes are not showing a trend toward making their way back to pre-disturbance conditions is because recovery may take longer than 20-30 years since fish were removed from the lakes. Also, ecological conditions in stocked lakes might have experienced a threshold such that the bounds of resiliency (i.e., returning to pre-disturbance conditions) might have been exceeded, this means the lakes will not return spontaneously. Lastly, other disturbances such as lakeshore vegetation may have confounded the fish effect and affected diatoms as well (Drake & Naiman, 2000).

The Drake & Naiman (2000) experiment suffered from high variabilities in density and composition of preserved invertebrates in the MORA lakes, so this variable has the potential to mask identifying changes due to fish introduction. This experiment focused on general dissimilarities and compositions of diatoms of the lakes to determine variation as well as the effects of fish introduction with relation the removal restoration projects, but the experiment did not address amphibians. Perhaps extending the study to include both abiotic characteristics as well as biotic characteristics of this ecosystem could aid in explaining why amphibians are declining.

Trout Removal Experiment

A study performed by Vredenburg (2004) focused on removal of previously stocked trout lakes in the High Sierras of California. Although fish have been disappearing or have been removed through angling since 1973 in MORA, no researchers have performed a trout removal experiment of this magnitude (Drake & Naiman, 2000; Vredenburg, 2004). Vredenburg (2004) was aware of other potential factors, such as diseases; UV radiation and climate change and increased levels of air pollution and pesticide use; and that these could have synergistic effects on frogs in montane wetlands. But Vredenburg (2004) also realized that many more studies have been done on these factors without the consideration of the predation effects of introduced trout. Studies that took place before Vredenburg's trout removal experiment considered mostly the role local factors play. The Mountain yellow-legged frogs of the High Sierras had been declining dramatically over the last couple of decades (Vredenburg, 2004). Addressing the effects of introduced trout in the High Sierras was important because the High Sierras were a focal area during the trout introduction interval beginning in the late 1800s.

The experiment was lengthy (8 years), which reflects the amount of time needed to see the response in a before-and-after-impact design. Additionally, it reflects the large number of water bodies utilized (n = 81). The study site was the Sixty Lakes Basin in Kings Canyon National Park, California, which was chosen largely because Mountain yellow-legged frogs were still relatively abundant in several fishless lakes in this basin. Mountain yellow frogs were visual encounter surveyed, and gill nets were used to determine whether fish were present in each specific lake that was chosen. To obtain a baseline, the average number of frogs and tadpoles per 10 m of shoreline in lakes with and without introduced trout were first compared (Vredenburg, 2004). Next, a trout removal experiment was conducted in a serial fashion in a series of lakes to see if trout limit both the size and distribution of the mountain yellow-legged frog. Interestingly, Mountain yellow-legged frogs and tadpoles began to increase in every lake in which fish removal had occurred; reference (or control lakes) that either already had no fish or from which fish were not removed showed essentially no change over time. Vredenburg (2004) concluded that with trout removal, frog populations could recover if source populations existed nearby from which colonization could occur. This study was particularly telling because a study of this type has only been performed once previously, but not to this magnitude (Knapp et al., 2001; Vredenburg, 2004).

In a study conducted in North Cascades National Park (NOCA), Tyler and colleagues (1998a) focused on the Long-toed salamander and the physical properties of 45 studied lakes. The researchers sampled seventeen lakes two or more times in a year, 8 lakes once a year over 2 or more years, and 20 lakes once. A snorkeling method was used to survey for salamanders, which was beneficial because Long-toed salamander sometimes like to hide out under or in the substrate. Kjeldahl-N (TKN) concentration (a method for quantitative determination of organic nitrogen) was an important factor because in lakes without fish, crustacean zooplankton (a focal food of Long-toed salamanders) was positively related to TKN concentration. The more zooplankton found in the lakes (fish-containing or fishless), the greater the number of long-toed salamanders. When TKN was low(<0.045mg/L), no significant difference existed in the abundance of larval Long-toed salamanders in lakes with and without fish, but when TKN was higher (>0.045mg/L), larvae densities of this salamander were higher in the lakes where trout were not reproducing (Tyler et al., 1998a). The important part here is that one must not only look at the abundance levels to get the entire picture of what is going on here, but the natural abiotic and biotic factors and processes as well (Tyler et al., 1998a; Hayes & Jennings, 1986). This NOCA study only focused on one species, where other species could be important to fully understand the observed patterns. TKN was the only nutrient measured, but other nutrients might have refined understanding the pattern. It might make for a more thorough experiment if Tyler et al. (1998a) observed factors other nutrients besides TKN, or perhaps factors related to climate change taking place in NOCA.

Amphibian and Trout Abundance Findings

Though it does not address Cascade frogs, the following research is important because it focuses on the trout introduction in relation to amphibians in high lakes outside the Cascade Mountains. Although several amphibian trout introduction relationship studies have been done on small numbers of bodies of water, Pilliod & Peterson (2001) conducted a study that focused on an entire mountain range in the Salmon River Mountains of Idaho. Columbia spotted frogs and Long-toed salamanders were the studied species. The Salmon River Mountains of Idaho, which are part of the Rocky Mountains, have a climate with long winters (up to 8 months) with heavy snowfall (Pilliod & Peterson, 2001). Understanding the climate and elevational footprint of an area is essential to understanding its seasonal pattern of suitability for amphibians. Surveys were done for both amphibians and fishes at 101 sites. Three different types of trout (*O. clarki, O. mykiss, and O. m. aguabonita*) were recorded at 43 lakes; 42.6% of all sites surveyed. Surveyed showed that fish-occupied sites had fewer amphibians. Lack of pre-stocking data provides uncertainty as to whether low numbers already existed in the lakes that had been stocked with fish later. This study suggests that amphibians in fish-occupied lakes will continue to decline in number (Pilliod & Peterson, 2000).

Low Elevation/Artificial Ponds

Another study examined Long-toed salamanders and Northwestern salamander relative abundances (Tyler et al., 1998b), both species prevalent in montane wetlands.

Salamanders were observed in a controlled lab setting using artificial ponds with and without fish (Tyler et al., 1998b). Long-toed salamander larvae were collected from low elevation ponds in Benton County, Oregon in 1996. Rainbow trout (*O. mykiss*) obtained from Oregon State University were used in the experimental (i.e., fish) treatments and control ponds were chosen randomly. The researchers measured 20 larvae snout-vent lengths (SVL) and selected physical characteristics, such as water temperature and substrate abundance, which were obtained daily. Both salamander species fared better in the ponds without trout than in the ponds, which contained trout. These conclusions are based on larval growths of both Long-toed and Northwestern salamanders. Both species showed longer SVLs in control ponds than in treatment ponds after the experiments. Although this experiment facilitated measurement that was easy to control and was not done in a natural amphibian habitat setting, the study did lead to findings that show reduced foraging opportunities associated with refuge use. This refuge use could lead to avoided encounters with trout or competition between larvae and trout for a limited food resource. This then could result in limited growth of larval Long-toed and Northwestern salamanders. The actual mechanism responsible for reduction in growth of these larval salamanders in the presence of trout was unable to be determined in this experiment (Tyler et al., 1998b).

Conclusion

Gaps in the Research

Only the sparsest of scientific information exists on amphibian populations before the introduction of trout in the 1800s. This condition requires that we take indirect approaches, such as the work of Drake and Naiman (2000), to infer the condition of fishless lakes and match that with similar condition in current amphibian-occupied fishless lakes.

Tyler and colleagues (1998b) indicated that many studies on eastern Ambystomatid larvae exist. Most East Coast findings regarding declining amphibians have been like West Coast findings (Petranka, 1983; Semlitsch 1988). What could this mean for different species of amphibians? Focusing on salamanders may help inform what is happening on different species of amphibians in major amphibian taxa (i.e. particular frog or salamander families). Considerable information exists regarding trout of other fish introductions in relation to amphibians, but few of those data have integrated the effects of climate change. However, many aspects of climate change are uncertain. However, we do know that many amphibians, because of their water-dependent life histories, are likely to be vulnerable. We do know that air temperatures are rising and are expected to continue to rise. We anticipate that rising air temperatures will accelerate snow melt and increase rates of evaporation, which may accelerate the seasonal shrinkage or drying of aquatic habitats.   How fast changes in hydrology will affect montane wetlands amphibian populations needs greater region-specific focus.

Amphibian Survey in an Undocumented Montane Wetland

My research included a preliminary amphibian survey I was a part of in the summer of 2016. The survey took place in the Enchantments Basin of the Wenatchee

Okanogan/Wenatchee National Forest. This forest deals with disturbances such as forest fires and landslide. No amphibian surveys had been previously done in the region. While there, I teamed up with the US Forest Services, specifically Shannon Claeson of the Wenatchee Station. On our first trip was from June 20-24; we observed Columbia spotted frogs tadpoles, juveniles, and adults, and Long-toed salamander larvae. I went back twice after that, once from July 11-15, and again in from August 15-19 and observed the same species; larvae, juvenile, and adult Columbia Spotted frogs, and larvae of Long-toed salamanders. The larvae and tadpoles could be found hiding under rocks on the edges of the ponds or sometimes swimming near the center of the ponds. The juvenile adult frogs were often seen on the edges near rocks or hiding under vegetation. The amphibians typically hid under the rocks near the shores of the lakes and ponds.

Because the core Enchantments Basin sits near the anticipated elevation limit of amphibian abundance, it seemed crucial to explore the area because of lack of documentation of frogs found prior in the area. The amphibians were spotted in the Snow Lakes at about 1524 m (5000 ft) elevation. There is permanent snow only in the highest areas (i.e. 2133 m (7000) ft elevation) in the Upper Enchantments Core Lakes (there were roughly 15 core lakes with many various unnamed ponds and tarns). This area provided an opportunity to assess how abiotic factors versus climate change factors, such as earlier and more intense snow melt runoff, may affect amphibian abundance. If no amphibians are present in areas from which the snow disappears late, it could have more to do with this region not having sufficient seasonal breadth for amphibians to survive in. Without previous knowledge of amphibians in this area, interpreting my findings is difficult. There is not any information which exists regarding whether amphibians were abundant in the Enchantments or any of the Cascade montane wetlands for that matter. With climate change advancing rapidly, understanding how these near-pristine landscapes are changing is crucial.

Chapter 2: Article Manuscript

Abstract

Geographic patterns of a high-elevation amphibian

in the Cascades range a case study of the

Cascade frog (*Rana cascadae)*

Dylan Kubina

The present study focuses on the threat of anthropogenic climate change in the montane wetlands and modeled its potential effect on Cascade frog (*Rana cascadae*) distribution. I first gathered all available verifiable records (~1,600) to estimate the current suitability of Cascade frogs. I then ran a predictive Species Distribution Modeling (SDM) software, *MaxEnt* to estimate Cascade frog suitability under different climate change scenarios. I obtained 19 bioclimatic features for modeling from Worldclim.org, and used these inputs to estimate Cascade frogs' suitability under four future climate scenarios (Year averages for 2050 and 2070). *MaxEnt* output revealed that habitat highly suitable for Cascade frog was more frequent in Mount Rainier National Park, Olympic National Park, and as far south as Lassen Volcanic National Park. Overall, my training and testing datasets appeared similar and both datasets were significantly different from random data based on the Specificity versus Sensitivity output, indicating that a non-trivial model. Using Representative Concentration Pathway (RCP) climate change scenarios, MaxEnt showed that future projections reveal Cascade frogs dwindling across their geographic range, with overall decreases in suitability in Mount Rainier and the Olympics. The most dramatic overall decrease in suitable Cascade frog habitat was in RCP 8.0 (2070). RCP 2.6 was the most similar scenario to the current suitability, while RCP 4.5 and 6.0 displayed growing

suitability in northern California with RCP 8.0 showing the sparsest suitability of Cascade frogs overall.

Introduction

Montane wetlands are keystone ecosystems in the Cascades Mountains. Over the last 150 years, two significant changes have affected these montane wetlands and their occupant amphibians. The first was when fish were introduced to previously fishless lakes, which began in the 1800s. The second is anthropogenic climate change, which though not recognized until recently, promises to dwarf all previous disturbances to montane wetlands (Carey & Alexander, 2003, Ryan et al., 2014, Lee et al., 2014). Because these two factors (climate change and introduced fish) interact, amphibians in montane wetlands are at particular risk. This is unsettling because these wetlands also provide important ecological services for both the assemblage of species living in them as well as human society (Ryan et al., 2014; Lee et al., 2014). Importantly, these montane wetlands provide food to aquatic and terrestrial species, and influence local hydrologic processes (i.e. groundwater recharge, surface water storage, and filtration), and geochemical cycles (Lee et al., 2014). They also tend to be understudied despite their potentially high sensitivity to climate change. There are few data available for montane wetlands relative to other ecosystem types. Montane wetlands may be experiencing declines in amphibian populations because of anthropogenic climate change and the introduction of trout in the 1800s. This thesis focuses on climate change factors affecting montane amphibians because amphibians in montane wetlands may be particularly sensitive to climate change, and research in montane wetlands that explores habitat changes as a function of climate change is sparse (Burkett and Kusler, 2000, Ryan et al., 2014, Lee et al., 2014).

Temperature and precipitation are the two most important abiotic climatic factors that affect amphibians. Abiotic factors are understudied in montane landscapes (Ryan et al., 2014, Lee et al., 2014). High elevations may experience disproportionately warmer air temperatures than lower elevation locations. Hence, water storage, nutrient cycling and carbon sequestration processes, may be differentially affected in montane landscapes (Carey & Alexander, 2003, Ryan et al., 2014, Lee et al., 2014). If so, montane amphibians may be losing their habitat disproportionately faster than elsewhere.

Climate change influences changes in biotic factors, which in turn affect Cascade frogs. Vegetation which makes up the frog's habitat and food sources, animal species that meet the Cascade frog, and fungi leading to diseases leading to the extirpation of over 200 amphibian species are the largest factors, which influence Cascade frogs. Trout did not exist in most high mountain lakes of the Cascades before the 1800s (Bahls, 1992). Trout, a potential predator of amphibians, now occupy almost all the lakes found in Cascade frog geographic range (Bahls, 1992). Fungi, specifically the amphibian chytrid fungi that causes chytridiomycosis, not only affects species such as the High Sierras Mountain yellow-legged frog as well as the Cascade frog in the PNW (DeLeón, Vredenburg, and Piovia-Scott, 2017) may be the biggest threat to the world's entire montane amphibian population (Ouellet et al., 2005).

Climate change influences both abiotic and biotic factors in the montane wetlands. Mountain lakes are vulnerable to rapid environmental changes stemming from climate change due to their high elevations which lead to permanent snow, which is melting earlier on in the year Ryan et al., 2014). Snow is an increasingly important part of the hydrologic cycle in the Western U.S. Montane wetlands are sensitive to hydrologic drivers that determine the rate and balance of water inflow and seasonal fluctuations in pond levels (Ryan et al., 2014; Lee et al., 2014). In vulnerable montane wetlands, amphibians are likely to be negatively impacted by the more rapid snowmelt resulting in a reduced later season snowpack because of warming temperatures (IPCC, 2007, Ryan et al., 2014, Lee et al., 2014). Increased warming results from human disturbances that add greenhouse gases (GHGs), leading to greater warming and shifts from less precipitation as snow and more precipitation as rain, which both lead reduced snow packs, increased evapotranspiration and soil moisture stress in the later summer months (IPCC, 2007, Ryan et al., 2014, Lee et al., 2014). Areas in the montane wetlands will be drier due to warmer weather and wetland water levels will be lower, so more amphibians may beat the risk of successful metamorphosing (Ryan et al., 2014, Lee et al., 2014). Earlier rates of wetland drawdown and increased frequency of summer drought is already occurring, both of which appear to be detrimental to amphibians. These negative symptoms of climate change are expected to worsen in the Western U.S., that is reflecting even higher summer temperatures and much more severe droughts (IPCC, 2007, Ryan et al., 2014, Lee et al., 2014).

As a PNW montane endemic, the Cascade frog represents the ideal species for observing the declining patterns among amphibian populations in the Cascades Region. This species lives in sphagnum bogs and lay their eggs in submerged vegetation (Briggs, 1987). They tend to eat algae, detritus, and plant tissue from the vegetation (NatureServe, 2017). The frog was once more abundant, but due to the fish stocking of the late 1800s (Bahls, 1992) and potential abiotic and biotic features as well as anthropogenic climate change, the species has suffered declines in habitat distribution

(Knapp et al., 2001, Ryan et al., 2014). The Cascade frog was historically especially abundant in Lassen Volcanic National Park, California, which is located at the Southern end of its range. Fellers & Drost (1993) conducted research in Lassen Volcanic National Park to find very few frogs when compared to the much larger number of frogs found in the past. Additional data demonstrates that a similar pattern is taking place in breeding grounds in various montane wetlands in the western U.S. (Ryan et al., 2014, Lee et al., 2014).

Extensive research has been done on the trout stocking in high mountain lakes of the western U.S., however only recently have a few articles focused on anthropogenic climate change as a real threat to the amphibians of the Cascades (Cary & Alexander, 2003; Ryan et al., 2014; Lee et al., 2014). If populations continue to decline, the Cascade frog could be put on the endangered species list. The goal of this research is to illustrate that climate change may influence habitat suitability in the Cascades montane landscapes, which may decrease Cascade frog occupancy. First, I will describe my methods used to obtain Cascade frog data. Next, I will show where Cascade frogs occurred geographically using MaxEnt species distribution modeling. Then I will also use MaxEnt to make predictions about Cascade frog suitability under different climate change scenarios. I will close by providing suggestions of how to refine the model, how to test predictions, and what actions should be taken in the future.

Materials and Methods

Geographic limits of distribution:

The Cascade frog range begins in northern California in Lassen Peak National park and runs through the Cascades of Oregon, north into Washington (Figure 2). A population also exists in the Olympic National Park of Washington (which is not a part of the Cascade Range). Populations in the Olympic Mountains of Washington and the Trinity Alps, Mount Shashta, and Mount Lassen are disjunct from the primary distribution along the main Cascades Range (Pearl and Adams, 2005; NatureServe, 2017). Lassen Volcanic National Park contains smaller numbers of Cascade frogs with warmer year-round temperatures than Oregon and Washington.

Figure 2. Range of Cascade Frogs (IUCN, 2004)

Elevational limits of distribution:

The Cascade frog lives at elevations of around 1524 m (5000 ft). Historical locations at low elevations in Washington suggest that the Cascade frog may have once been more broadly distributed (Leonard et al., 1993). Historical elevation range extended from around 400 to 2,500 meters (1312 ft- 8202 ft) (NatureServe, 2017).

Habitat Types of the Cascade Frog

The Cascade frog lives in sphagnum bogs and lay their eggs in submerged vegetation (Briggs, 1987). The frogs do not live out their days where they were bred, but rather near edges of ponds and streams. Overwintering takes place in late fall. Cascade frogs are found congregated in spring-fed ponds and perennial streams (Garwood, 2009; Pope et al., 2014). The frogs were found overwintering in deep, loose silt at the bottom of a pond (Briggs, 1987; Pope et al., 2014).

Preliminary Research: The Enchantments Basin

I surveyed the Enchantments Lakes Basin in the summer of 2016 to allow me to get a sense of montane wetland habitats. The survey, conducted in conjunction with the U.S. Forest Services and WDFW, is of special interest as the Enchantments sit on an anticipated elevation boundary of amphibian occupancy. This means that if amphibians exist here they are strongly influenced seasonal by abiotic factors. I traveled to the Enchantments Basin on three separate occasions from the dates June 20-24, July 11-15, and August 15-19, 2016. No previous amphibian surveys had been conducted in the Enchantments prior to this study. To reach the high lakes, a trek to around 1,524 m (5,000 ft) elevation was necessary to see where the first high-elevation amphibians occurred. Surveys were conducted upwards on to around 2,133 m (7,000 ft), where permanent snow and near-freezing water was apparent. Our surveys in the Enchantments documented amphibians using standard visual encounter techniques along shorelines and streams using a dip net. I measured amphibians trapped in nets and released them back into the habitat. I documented the species by keying them out per features such as snout to vent length and physical features including spot size, color, and eye placement. Tadpoles were measured as well. I recorded notes on dominant vegetation type, animals present, water quality based on substrate, temperature of water, and the size of lakes. I used visual encounters to observe vegetation animal species, water quality and lakes over a meter. I used a digital thermometer to measure water temperature in degrees Celsius. Although there were not any amphibians onwards of 7,000 ft in the Upper Enchantments lakes, there were amphibians in the 5,000 ft zone, which contained Upper and Lower Snow Lakes. There was also an intermediate pond between Lower and Upper Snow Lake, which I called Little Upper Snow Lake. The amphibians I did find, all in the Snow Lakes complex, around 5000 ft elevation, were Columbia spotted frogs, Pacific tree frogs, and Long-toed salamander larvae.

Data Collection

I assembled Cascade frog data from several sources. These included the Vertnet database, and databases obtained from the Mount Rainier National Park headquarters, the Olympic National Park headquarters, and USGS. Vertnet, a National Science Foundation-funded vertebrate database, had the most extensive database, but it was entirely based on verifiable museum specimens. Remaining databases included a mix of survey data (the majority) and specimen-verifiable information[1]. Most location records in the databases were georeferenced, but points lacking georeferenced data or for which the geo-referencing data were too vague were not used in the analysis dataset. Over 5,000 georeferenced records existed from all four databases combined, but this included duplicates. I removed duplicates for the analysis dataset. I kept latitude, longitude pairs and dates when the specimens were collected or documented for the GIS portion of the results[2]. I converted all georeferenced locations in UTM zone 10N to WGS84 latitude, longitude coordinates. Once converted, the latitude, longitude pairs depicted a spatial distribution of where the frogs occurred (Figure 2). I elected to keep the points that were missing dates if they were georeferenced on my maps, I left the Central Valley of California on my maps to show lands outside of the Cascade frog range.

There were some points that I was skeptical of due to the location on the map and other parameters suggesting they were possible misidentified frogs and their locations were in areas unlikely to be occupied by Cascade frogs, so I deleted these points. Details of these points are provided in Appendix 2.

Data Analysis

I used the screened georeferenced data points from the databases as inputs for MaxEnt use. I used parallel georeferenced 19 bioclimatic features from Worldclim.org (an ESRI supported website) as inputs as well. Worldclim.org derives variables from monthly temperature and rainfall values to generate more biologically meaningful factors. Bioclimatic variable across georeferenced locations represent interpolations of observed data over the period 1960-1990. Worldclim.org is a climate data site the Environmental Systems Research Institute (ESRI) uses, which one uses for ecological modeling and GIS. ESRI develops ArcGIS software, which applies the science of using maps to reveal deeper insight into data. Worldclim.org uses a set of global climate layers (gridded climate data) with a spatial resolution of about one km^2. The data are 30 arc-seconds resolution downloaded in 30 × 30 degree footprints. I chose to include the rasters for the 19 of the bioclimatic factors from Worldclim.org (Table 3).

Table 3. Bioclimatic Factors from (Worldclim.org, 2017)

Code	Bioclimatic Feature
BIO1	Annual Mean Temperature
BIO2	Mean Diurnal Range (Mean of monthly (max temp - min temp))
BIO3	Isothermality (BIO2/BIO7) (* 100)
BIO4	Temperature Seasonality (standard deviation *100)
BIO5	Max Temperature of Warmest Month
BIO6	Min Temperature of Coldest Month
BIO7	Temperature Annual Range (BIO5-BIO6)
BIO8	Mean Temperature of Wettest Quarter
BIO9	Mean Temperature of Driest Quarter
BIO10	Mean Temperature of Warmest Quarter
BIO11	Mean Temperature of Coldest Quarter
BIO12	Annual Precipitation
BIO13	Precipitation of Wettest Month
BIO14	Precipitation of Driest Month
BIO15	Precipitation Seasonality (Coefficient of Variation)
BIO16	Precipitation of Wettest Quarter
BIO17	Precipitation of Driest Quarter
BIO18	Precipitation of Warmest Quarter
BIO19	Precipitation of Coldest Quarter

I used Bioclimatic factors for environmental inputs in MaxEnt analysis. The future data were drawn from $IPCC_5$ climate projections from global climate models

(GCMs) for four representative concentration pathways (RCPs) or scenarios (Table 4). These are the most recent GCM climate projections that researchers used in the Fifth Assessment IPCC report (IPCC, 2014). I used Worldclim.org 1.4 downscaled and calibrated (also known as bias corrected) using its software as my baseline 'current' climate. I used CCSM4 GCM, also known as CC for short for the year 2070. These files are in 30 seconds' resolution. The time periods for 2070 represent, respectively, averages over 2061-2080.

Table 4. AR5 Global Warming Increase (°C) Projections for 2081-2100

Scenario	**Mean and *likely Range***
RCP 2.6	1.0 (0.4 to 1.6)
RCP 4.5	1.4 (0.9 to 2.0)
RCP 6.0	1.3 (0.8 to 1.8)
RCP 8.5	2.0 (1.4 to 2.6)

Across all RCPs for 2081-2100, global mean temperature is projected to rise by 0.3 to 4.8°C (ICPP, 2015).

Chapter 3. Results

Current Spatial Distribution

The current distribution of the Cascade frog is based on the collective georeferenced points from the database (Figure 3). Current spatial distribution shows Cascade frogs spanning the Cascades Range and the Olympics. The more dots that are clustered together, the more known occurrences are present in that region. There are high occurrences of Cascade frogs in the Olympics, NOCA, MORA, the Cascades that run through Oregon, and Lassen Peak.

MaxEnt Current Suitability Projection

The current suitability of Cascade frogs is shown with a scale bar, which explains areas of green representing little to no Cascade frog occurrences and areas of red representing areas of high suitability of Cascade frogs.

Model Performance

Omission and Predicted Area for 0 and Sensitivity vs. Specificity were generated with the given parameters: random seed, random test percentage of 25%, one replicate, and subsample replicated run type. These graphs explain the current suitability projection. In both graphs, area under the curve's (AUCs) for the Training and Test data appear to be correct because 1) One they are very close to each other, meaning the test data agree that the training data produced a good model; and 2) both the training and testing data sets are substantially different from the random data set, meaning the model carries significant weight (meaning) (Figures 5 and 6). The area under the curve in this case is closer to 1, which indicates a better model performance and predicted probability of occurrence using sample size (Young, Carter, and Evangelista, 2011).

Jackknife Analysis

Jackknife analysis compares the training data in isolation to the training gain with all the variables included. Jackknife is represented in terms of regularized training gain for 0 (Figure 7). The jackknife analysis shows that precipitation for the driest quarter (BIO17), precipitation for the warmest quarter (BIO18), mean temperature of the driest quarter (BIO9), and mean temperature of the warmest quarter (BIO10) had the most influence on the projections for current suitability (Figure 4). The Jackknife shows the training gain of each variable had the model been run in isolation.

MaxEnt 2070 Projections

The future scenarios, which consider RCP 2.6, 4.5, 6.0, and 8.5 for the year 2070 show where suitable areas where Cascade frogs might exist based off bioclimatic features as inputs. Projections of Cascade frogs' suitability for Scenario 2.6 shows high suitability in the Olympics and MORA. There are slight decreases in suitability when compared to Current Suitability (Figure 4) in these hot spots (Figure 8). Scenario 4.5 areas of high suitability in the Olympics, MORA, Oregon, and new high suitability in northeast Washington and Lassen Peak (Figure 9). When compared to Figure 4.5, there is new suitability in northeast Washington and northern California with decreases in the Olympics and MORA. Scenario 6.0 shows areas of high suitability in the Olympics, MORA, and Lassen Peak (Figure 10). When compared to Figure 4, there is a decreased suitability in the Olympics and MORA. The high suitability in northern California is a newly suitable area. Lastly, the Scenario 8.5 depicts low suitability in MORA, the Olympics, Oregon, and California, with high suitability in northeast Washington (Figure 11).

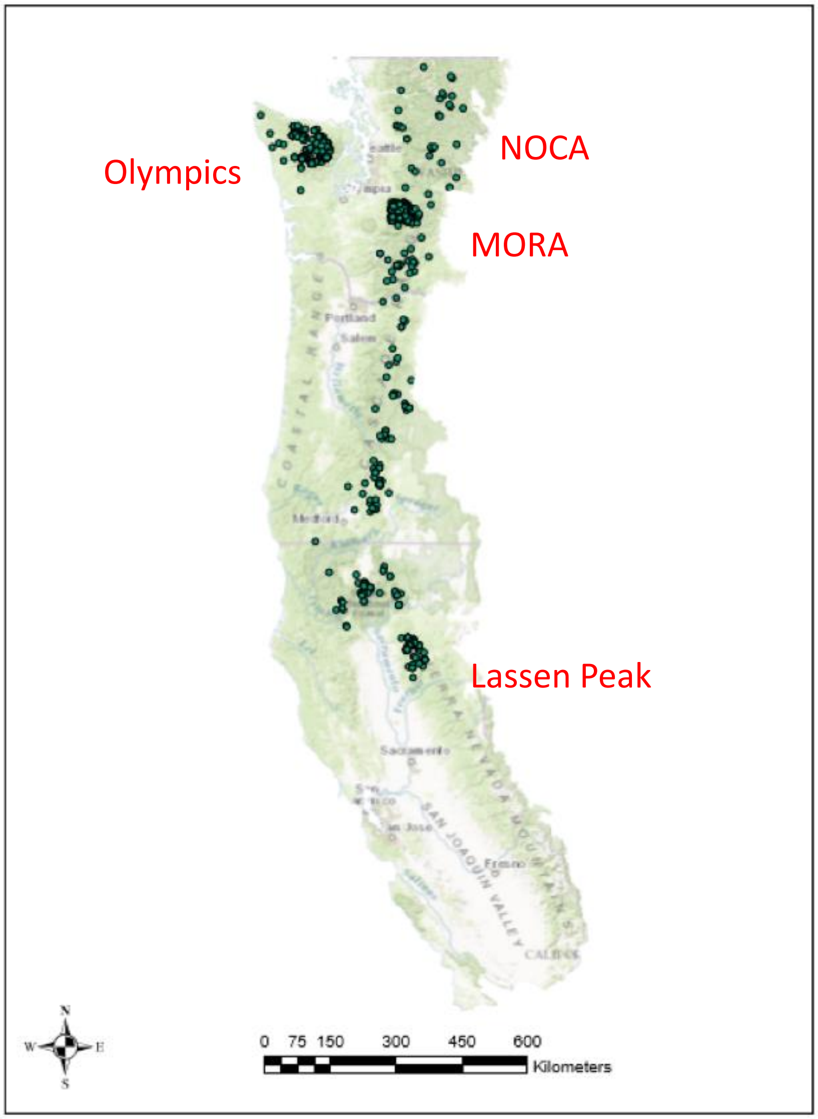

Figure 3. Cascade Frog Occurrences in the Cascades Region

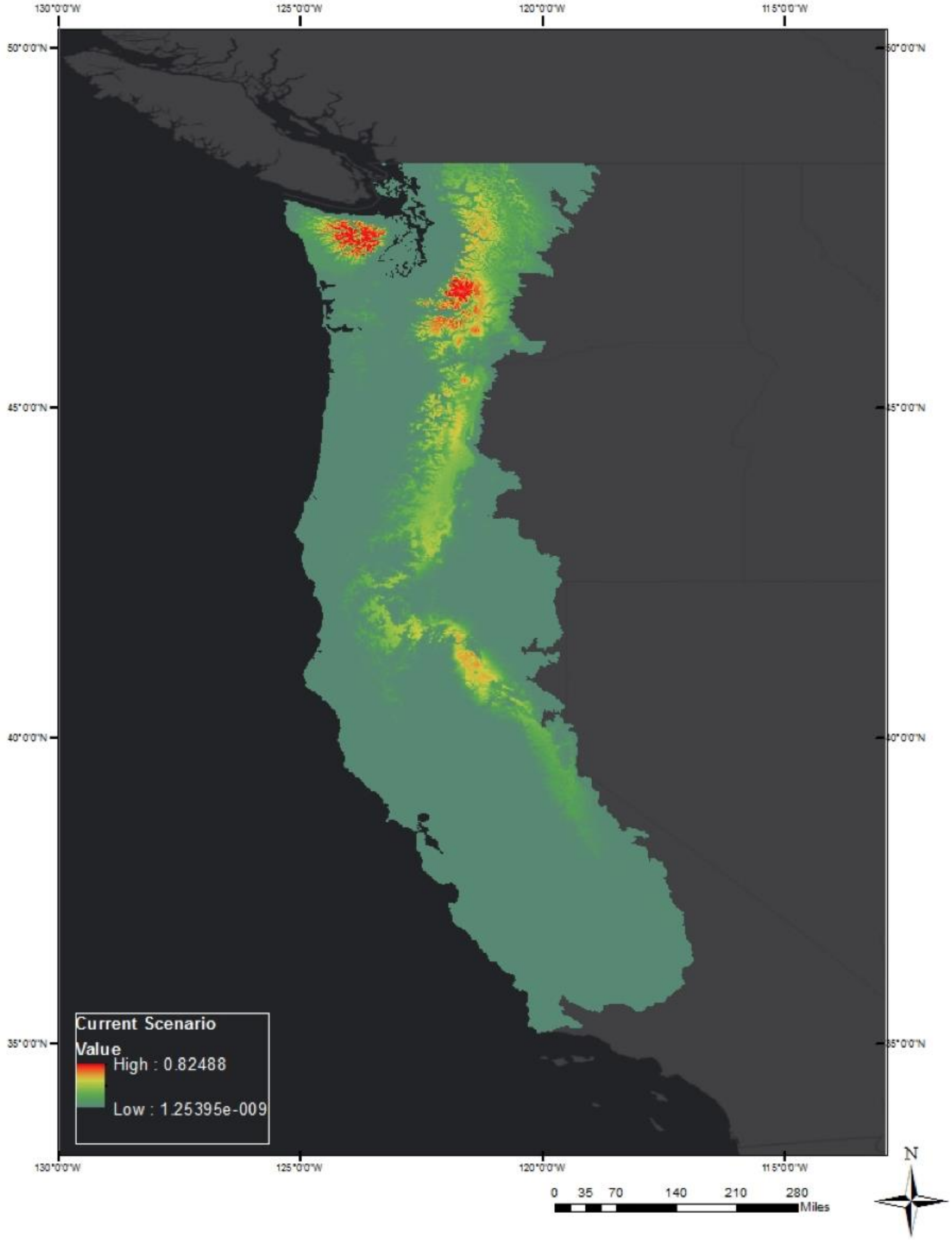

Figure 4. Current Suitability for Cascade Frogs

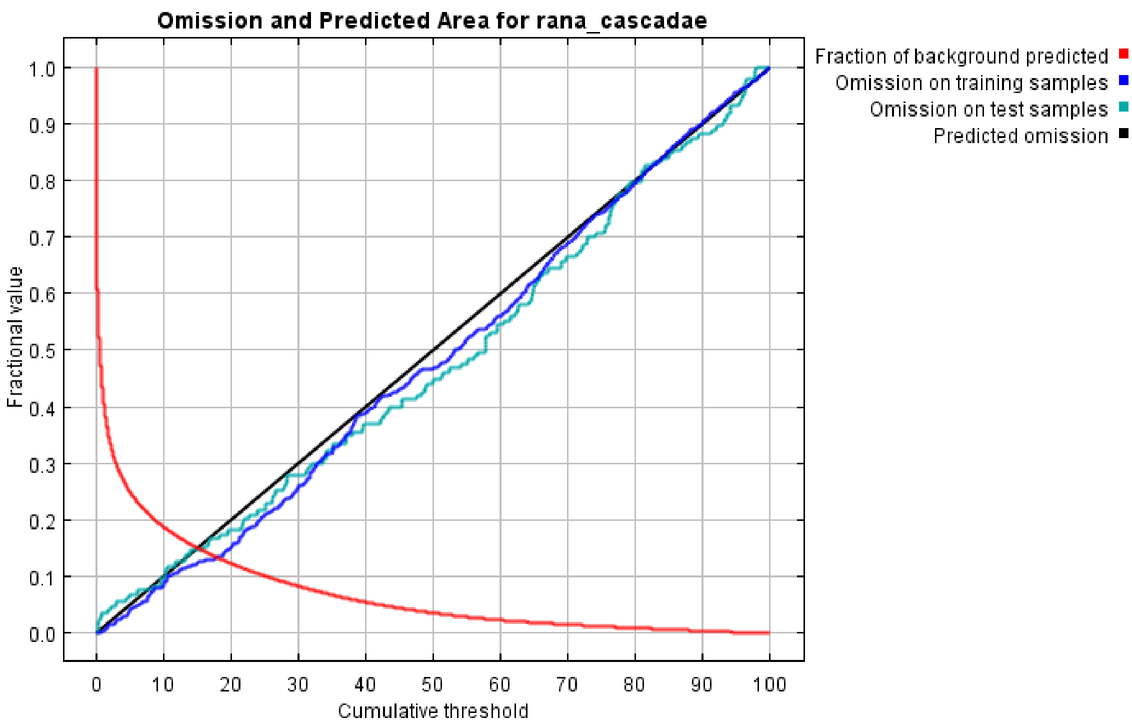

Figure 5. Omission and Predicted Area Omission and Predicted Area. for 0 for a random seed, random test percentage of 25%, one replicate, and subsample replicated run type. The output format is logistic with a 10% threshold percentage; this is for the Current Suitability Projection.

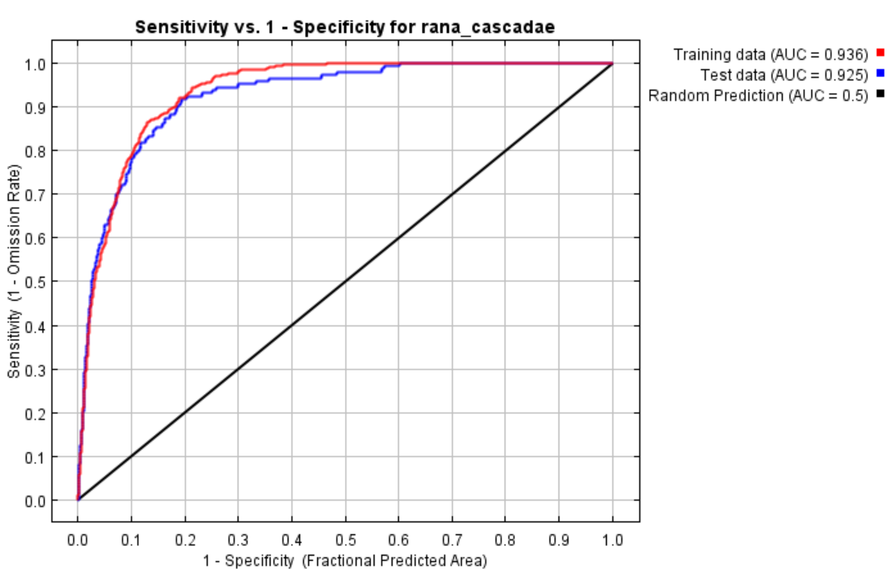

Figure 6. Sensitivity vs. Specificity. Training and testing datasets for a random seed, random test percentage of 25%, one replicate, and subsample replicated run type. The output format is logistic. The training data has an Area under the curve (AUC) of 0.936, an AUC of 0.925 for test data and an AUC of 0.5 for random prediction.

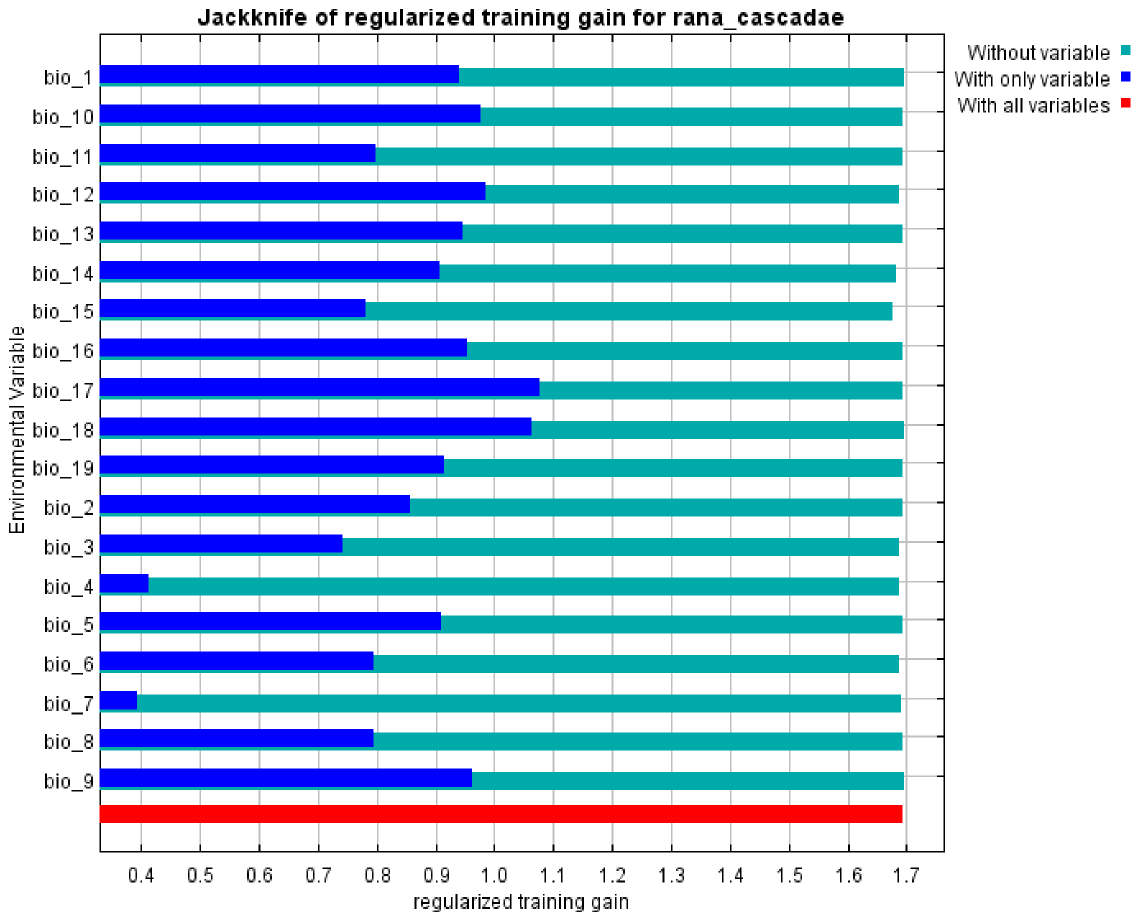

Figure 7. Jackknife of regularized training gain. The Jackknife shows the training gain of each variable and the influence said variable had when MaxEnt was run in isolation, and compares the training data ran in isolation to the training gain including all the variables. This is for Current Suitability.

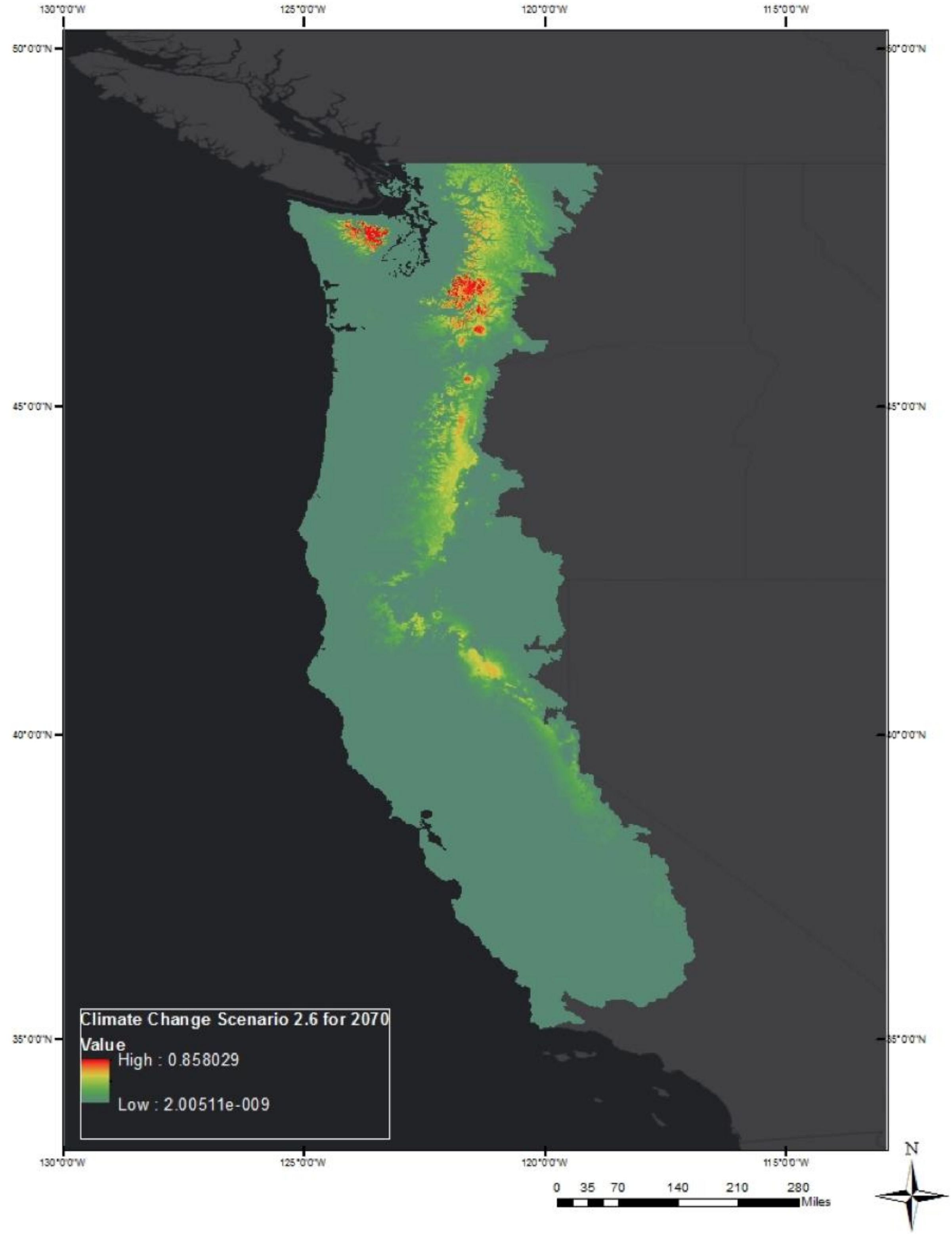

Figure 8. Scenario 2.6 for Year 2070

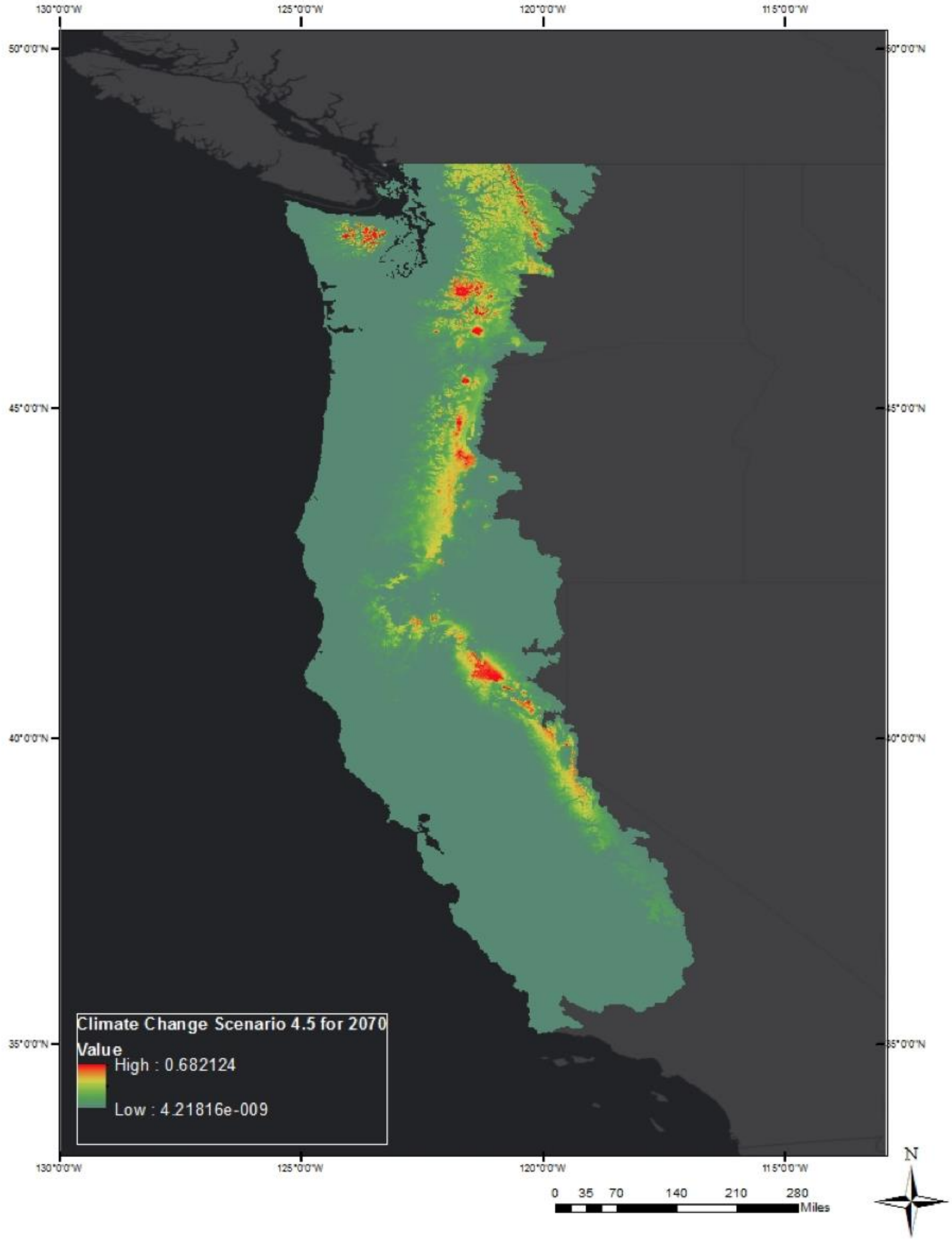

Figure 9. Scenario 4.5 for Year 2070

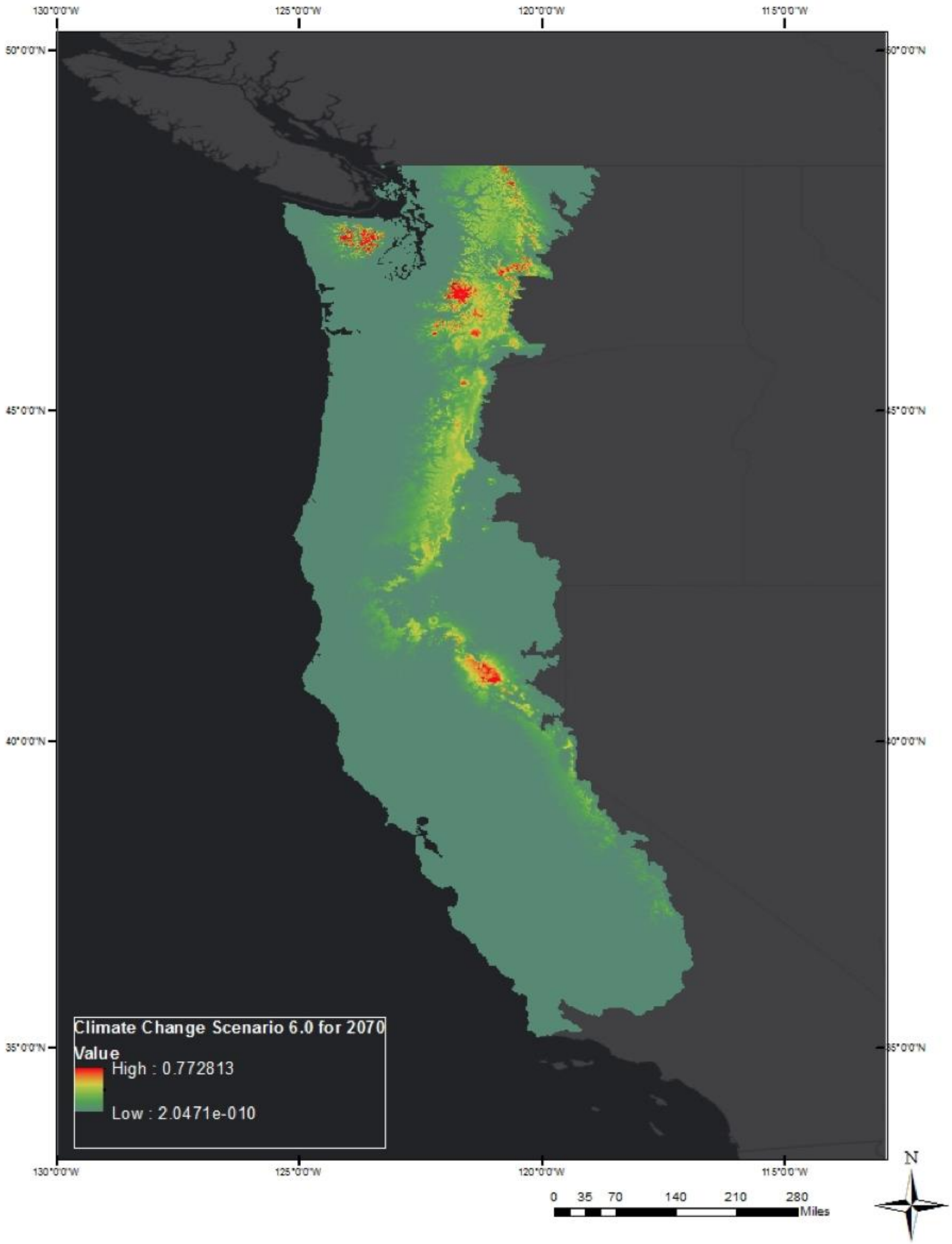

Figure 10. Scenario 6.0 for Year 2070

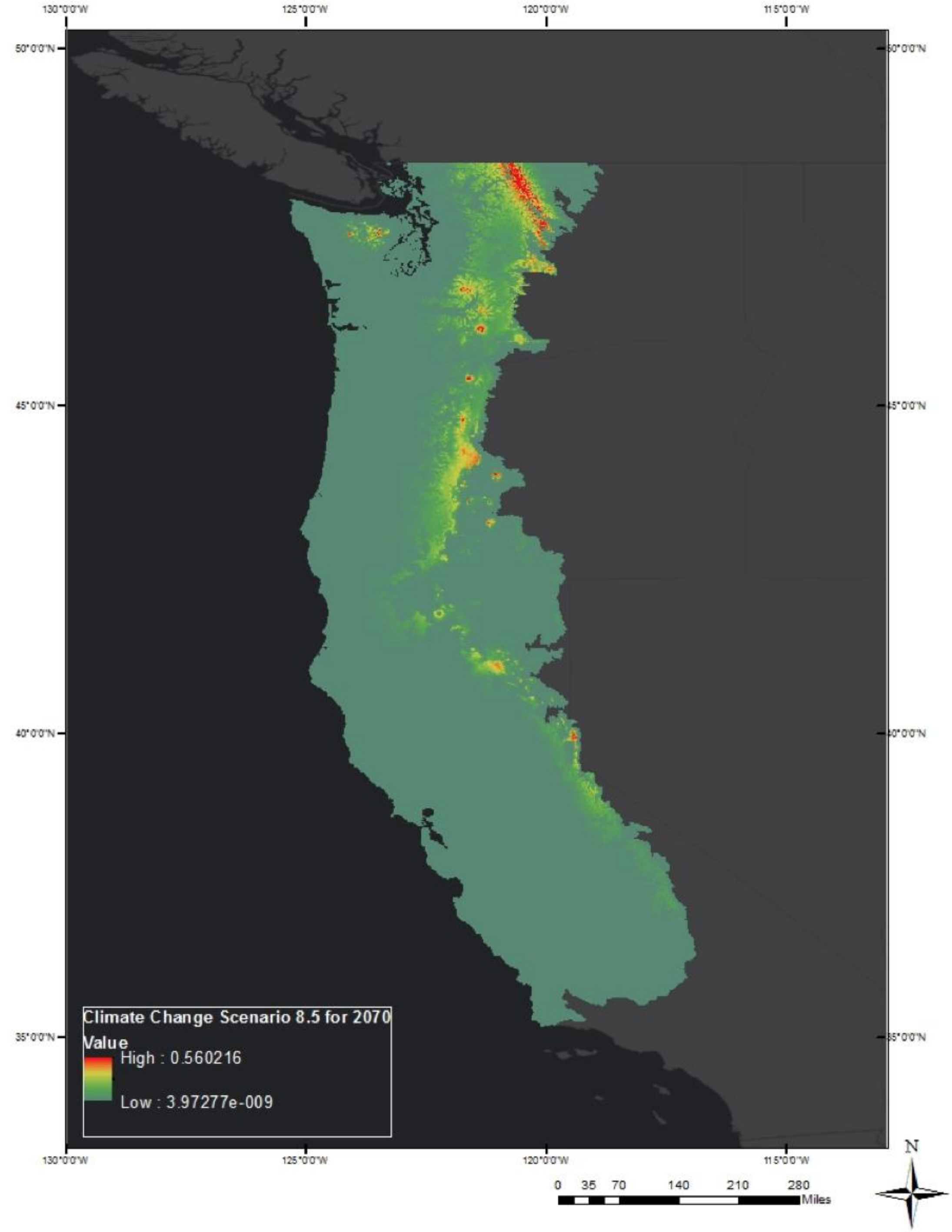

Figure 11. Scenario 8.5 for Year 2070

Chapter 4. Discussion and Conclusion

Specific changes in temperature and rainfall values from the associated RCP climate change scenarios led to overall decreases in suitability as well as increases in newly suitable areas of Cascade frogs. These monthly temperatures and rainfall factors were projected to change based on future climate change patterns. These changes influence an apparent decrease in amphibian populations from recent literature. My findings support these results by demonstrating most of the future raster outputs result in dwindling suitability for the Cascade frog. In each scenario, precipitation was reduced in the spring and summer months and overall temperatures were warmer overall (Table 4). The bioclimatic variables: BIO9 (Mean temperature of driest quarter), BIO10 (Mean temperature of warmest quarter), BIO12 (Annual precipitation), BIO17 (Precipitation of driest quarter) and BIO18 (Precipitation of warmest quarter) (Table 3) are the bioclimatic factors that had the most influence (Figure 7) on the current suitability projection (Figure 4). Without data on population trends from past years, I cannot conclude that Cascade frogs are declining. The purpose of this discussion is to fill some of the gaps from the literature review with my findings. I will do this by going more in depth with how my findings complement the literature. I then describe what my MaxEnt findings mean. Lastly, I will provide suggestions about what I might model differently, what additions I would make to the study, and lastly suggestions on what actions should take place.

Reasons for Decline

The Cascade frog's Red List Category and Criteria is listed as Near Threatened (IUCN, 2004), meaning there should be cause for concern that the frog may end up on the

Endangered Species List. Declining patterns of Cascade frogs are seen in northern California from RCP 2.6 and 8.5 (Figures 8 and 11). The MaxEnt generated maps, except for RCP 4.5 and 6.0 (Figures 9 and 10) support the past literature by recognizing that there was once more suitable habitat in these regions compared to the Current Suitability (Figure 4).

Ryan and colleagues (2014) interpreted fish introduction and climate change to be the two largest factors resulting in amphibian decline in montane environments. Fish introductions which begin in the late 1800s, was not addressed in my study, but was certainly recognized as a factor as to why amphibians in the western U.S. are dwindling. My study was similar Ryan's in assessing future climate scenarios. I used MaxEnt while Ryan et al. study utilized Variable Infiltration Capacity (VIC) as a form of wetland projection modeling software. Ryan and colleagues (2014) also demonstrated how the current climate change trajectory is already affecting summer water availability, resulting in summers being warmer and having less precipitation.

The spring months when Cascade frogs breed is crucial because with the warmer weather in winter, ice melts sooner, which triggers amphibians, like the Cascade frogs, to breed. The warmer spring months and overall less amount of precipitation projected from the bioclimatic features and climate change models suggests a pattern that Cascade frogs overwintering and breeding patterns are being changed. This will in turn affect their life cycles. Different areas in the Cascades include montane wetlands, and they are sensitive to snowpack volume, snow runoff, direct precipitation and evapotranspiration.

MaxEnt Explanation

Projections of Cascade frog locations were determined from the environmental inputs, (i.e. bioclimatic features). I collected data from four different databases and had to convert different projections, which could result in inadvertent errors. Comparing the current spatial suitability (Figure 4) to Climate Change Scenario 2.6 for 2070 (Figure 8) shows that Cascade frogs will result in lower suitability in both MORA and the Olympics. RCP 2.6 differed slightly from the Current Suitability projection, but was still overall similar. Scenario 4.5 shows lower suitability in MORA and the Olympics, with new higher suitability in northeast Washington, Oregon, and northern California. (Figure 9). Scenario 6.0 showed decreases in suitability in MORA and the Olympics, but a newly suitable area became evident slightly in northeast Washington and in northern California as well (Figure 10). Lastly, Scenario 8.5 shows the overall greatest decrease in suitability throughout the Cascade frogs' Range in the Cascades. There is also new suitability in northeast Washington (Figure 11). This may be because the global temperature is expected to rise the most for this scenario (Table 4). The area of higher bioclimatic suitability by 2070 in northern California under RCP 4.5 (Figure 9) is characterized by BIO14 (more precipitation in the driest month) and BIO18 (more precipitation in the warmest quarter). It would make sense for northern California and northeastern Washington to show higher suitability for RCP 4.5, 6.0, and 8.5 because California and northeastern Washington have very hot and dry climates in the summer. More precipitation in the summer (when it is the warmest and driest) may cause for this spikes in suitability for the Cascade frog. It would be beneficial to implement a habitat mask into MaxEnt to take into account where Cascade frogs actually exist based on habitat features. Cascade frogs will most likely not have higher suitability in northeast

Washington and northern California in the future because these places will have droughts and experience the impacts of climate change much more than more temperate regions such as mid-western/western Washington and Oregon.

Potential Changes to the Analysis

In the future, I would expand my study to include waterbody data with and without fish descriptors (Ryan et al. 2014). This may allow for an independent opportunity to evaluate the interaction of fish presence and climate change may affect Cascade frog occupancy in wetlands. I would also like to use object-based remote sensing; VIC simulations of historical wetland dynamics may enable identifying trajectories of change for different type of wetlands that can be integrated into the modeling. For future study, I would like to incorporate other environmental features into MaxEnt, such as distance from the water, snow cover, elevation, and slope are all potentially important factors when considering the Cascade frog's life history. I would also like to run MaxEnt multiple times for different parameters and different percent thresholds. This would change the Jackknife and Sensitivity analyses and assist me in identifying the best models. A habitat mask would also be an important addition to this MaxEnt analysis in order to project where Cascade frogs might occur in the future.

Future Ecological Implications for Wetland Biota

The Cascade frog, once abundant in the Southern Cascade Range and Klamath Mountains of California, has become extremely rare in the Southern Cascades. However, up to very recently, the species is still abundant in the Klamath Mountains (Pope et al., 2014). Many potential reasons exist for the decline of Cascade frogs, I explored climate change using MaxEnt to predict how Cascade frog habitat might change in the future. The future projections, especially the 2070 projections, do not look good for the species because highly suitable habitat will essentially vanish from the northern California where it is currently abundant (specifically in the Klamath region of Oregon and Washington). Climate change factors, such as decreased precipitation in summer months, earlier breeding times due to earlier onset of snow melting and warmer temperatures may be causing the frogs to decline as well as lose their habitat. Cascade frogs are adapted to living at elevation in snowy regions. My trip to the Enchantments Basins allowed me to take a closer look at the habitat where Cascade frogs may live. During my three trips (once in June, July and August) allowed me to see just how fast the snow melted; whether those habitats are ultimately colonized by Cascade or Columbia spotted frogs as seasonal snow melts earlier remains to be seen. The research done over the last 100 years shows that the indicator amphibian species are declining in number. Actions should be taken to ensure more amphibians appear on the endangered species list. The focus of my thesis was to examine current Cascade frog suitability and to project where their habitat might in the future year 2050. Time still exists to protect this species as well as other amphibians living in high mountain lakes. We also cannot ignore that the climate is changing rapidly—there are earlier and faster rates of wetland drawdown, reduction in water availability and an increased frequency of complete drying (Ryan et al., 2014). Hope may still exist for other high mountain-dwelling amphibians if the proper steps are taken for conservation. I concluded that Cascade frogs probably do not exist in the Lower Lakes complex of the Enchantments Basin, although this is type of montane wetland habitat where they can be found. Targeted fish removal programs and amphibian monitoring programs should be implemented. Targeted fish removal programs may aid in revitalizing amphibian populations, while amphibian monitoring programs can better keep tabs on amphibian populations. This is not just about one frog, rather an entire ecosystem and all the species living there.

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Appendix

1. Databases included information from a group of different herpetologists and researchers showing the locations where Cascade frogs were documented, the date collected, institution, collection ID, institution code, preservation status, the age/class, sex, catalogue number, recorder name, count, occurrence marks, latitude and longitude World Geodetic System 1984 (WGS84), elevation, the museum or institute which identified the specimen, the date collected, and the nomenclatural code. The locations varied substantially and included regions from all over the Cascades and the Olympics. The Cascade frog data from the MORA headquarters, the Olympic National Park headquarters, and United States Geological Survey (USGS) contained uniform factors such as record ID, source, source contact, program, survey type, the date collected, the park and its associated code, the site name, elevation, life stage, count, Universal Transverse Mercator (UTM) zone 10N location data and notes regarding township, section, and range.

2. These points included (48.71866, -122.352255), (47.197465, -122.534167), (45.407626, -122.570384), (45.467161, -122.790306), (42.760341, -123.701202), (42.02212, -123.459195). The first point was in Bellingham, WA, right off interstate five. With further speculation, this point had an elevation of roughly 2500 feet. This appears to be relatively low elevation for Cascade frogs. W.C. Brown identified the next frog(s) documented in Pierce County, off Chambers Creek at an elevation of roughly 500 feet. The low elevation and surrounding neighborhood- from the Google maps timeline photo

from 1990 displays that this is not ideal habitat for Cascade frogs (GoogleMaps, 2017). The next point lies in Clackamas County, Oregon with an elevation of 170 feet and appears to be right in the middle of the street. The following point was in Beaverton, Tualatin Hills Park and Recreation District, Fanno Creek Park, Fanno Creek Pond, Oregon. The next frogs existed in Douglas County, Oregon in or near the Kelsay River with an elevation of 4146 feet. The point was identified in Josephine County, Bolan Lake, Oregon at an elevation of 5450 feet. I removed this point because it occurred outside of the Cascades, where Cascade frogs are normally found. The final count and sample size for the data set was roughly 1,700 points.