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MS ENVIRONMENTAL BIOLOGY CAPSTONE PROJECT

by

Tyler A. Erickson

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

> REGIS UNIVERSITY May, 2022

MS ENVIRONMENTAL BIOLOGY CAPSTONE PROJECT

by

Tyler A. Erickson

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Table of Contents

CHAPTER 1. LITERATURE REVIEW	. 1
Impacts of Trout Stocking Practices on Colorado's Alpine Ecosystems	. 1
Introduction	. 1
Cutthroat Trout	. 2
Alpine Fishless Lakes	. 3
Macroinvertebrates	. 5
Amphibians	. 7
Terrestrial Ecosystems	. 9
Conclusion and Future Management Applications	10
References	12
CHAPTER 2. GRANT PROPOSAL	21
Effects of Trout Presence on Alpine Lake Macroinvertebrate Emergence in Colorado 2	21
Section 1: Abstract	22
Section 2: Introduction	23
Section 3: Methods	26
Section 4: Budget	28
Section 5: Qualifications of Researcher	30
References	33
CHAPTER 3. JOURNAL MANUSCRIPT3	36

Post-Stocking Survival and Myxospore Evaluation of V	Whirling Disease Resistant Rainbow
Trout Strains	36
Abstract	36
Introduction	37
Methods	42
Results	46
Discussion	51
Acknowledgements	56
References	56
CHAPTER 4.	64
A Compromise for the Planned Reservoir Expansion at	t Bear Creek Lake Park to Combat
Denver's Water Crisis	64
Introduction	64
Background	65
Stakeholders	67
Recommendation	70
Conclusion	72
References	73

CHAPTER 2, LIST OF TABLES

1.	Project Budget
	CHAPTER 2, LIST OF FIGURES
1.	Map of Sample Site Locations30
	CHAPTER 3, LIST OF TABLES
2.	Rmark Model Selection Results
	CHAPTER 3, LIST OF FIGURES
2.	Parvin Lake study system42
3.	Myxospore counts4
4.	Survival rates of the four strains
5.	Survival rates across age and time5
	CHAPTER 4, LIST OF FIGURES
1.	Proposed Expansion at Bear Creek Lake Park60

CHAPTER 1. LITERATURE REVIEW

Impacts of Trout Stocking Practices on Colorado's Alpine Ecosystems

Introduction

Introductions of predatory game fishes negatively impact native fauna around the world (Eby et al., 2006). In the United States, the purpose of these introductions has been to enhance sport fishing opportunities (A. Halverson, 2010; Pister, 2001), and have led to the homogenization of fish diversity (Rahel, 2000). Of the predatory fishes introduced into the United States, trout represent the majority (M. A. Halverson, 2008). In the western United states, an estimated 95% of alpine lakes were historically fishless. By 1988, 55% of nearly 15,200 historically fishless alpine lakes had been stocked with trout. For large fishless lakes, 95% have been stocked with trout, with the remaining fishless lakes often being too small and shallow to support fish (Bahls, 1992). Trout are ultimately predators whose introductions cause trophic cascades through increased top-down food web controls in aquatic ecosystems (Eby et al., 2006). Resulting effects include changes in macroinvertebrate communities, local extirpation of amphibians, and changes in the abundances of terrestrial organisms.

Trout continue to be stocked into historically fishless alpine lakes, despite growing evidence that these practices alter alpine ecosystems. Recently, stocking practices have taken more of a conservation focus in protecting cutthroat trout in the Western United States. Cutthroat trout are an endemic polytypic species that has had broad scale declines in populations resulting primarily from introductions of non-native trout species (Allendorf & Leary, 1988; Quist & Hubert, 2004). Alpine regions serve as refuges for cutthroat trout from non-native trout, and are the sites of recovery efforts (Quist & Hubert, 2004). Understanding how trout affect food webs

in alpine lakes is essential for protecting alpine ecosystems as well as planning the recovery of native cutthroat trout species.

Colorado's alpine environment is currently in a state of management transformation that started with the discovery of greenback cutthroat trout populations in 1968, a subspecies that was once believed to be extinct (Fendt, 2019; Young & Harig, 2001). New genetic technologies are placing additional scrutiny on the purity of cutthroat trout populations in alpine lakes, spurring demand for controversial eradication efforts in the name of conservation (Havlick & Biermann, 2021). With the resurgence of the greenback cutthroat trout recovery plan, now is a great time to discuss the historical context of alpine watersheds in Colorado and plan the best conservation path into the future. Contrary to historical management practices, current alpine conservation management plans need to consider the trophic effects of trout introductions on both the aquatic and terrestrial riparian environments, which will likely have conflicts with other conservation and fisheries management goals. These goals should be balanced to preserve ecosystem health and species diversity when recovering and protecting cutthroat trout populations, all while providing adequate recreational fishing opportunities in Colorado.

Cutthroat Trout

Widespread introductions of non-native trout species, in conjunction with watershed disturbances from colonization, have led to the decline of cutthroat trout populations in Colorado (Fendt, 2019). Brook trout, rainbow trout, and brown trout have all been stocked in Colorado in the late 1800's and early 1900's to enhance cold water fisheries by increasing the abundance of novel fish species (A. Halverson, 2010; Wiltzius, 1985). All three species of non-native trout outcompete cutthroat trout when in sympatry. Generally, brown trout are thought to physically dominate cutthroat trout while rainbow trout typically obtain and maintain more ideal

feeding lies (Seiler & Keeley, 2007; Wang & White, 1994). Brook trout diminish cutthroat trout reproductive recruitment from competitive advantages in juvenile brook trout that have comparative size advantages, presumably from spawning earlier in the fall (McGrath & Lewis Jr, 2007; Peterson et al., 2004). In all, cutthroat trout populations have drastically declined since the early 1900's, so much so with greenback cutthroat trout that they were declared extinct in 1937 (Young & Harig, 2001).

Recovery efforts began in 1968 when a biologist named Robert Behnke discovered a population of greenback cutthroat trout surviving in Como creek in the South Platte River watershed (Young & Harig, 2001). The greenback cutthroat trout was listed on the Endangered Species Act of 1972 and the recovery plan set forth the goal of obtaining 20 large viable populations of greenbacks as the qualification for delisting (Young & Harig, 2001). The recovery was going fine until genetic studies conducted first in 2008, and later confirmed in 2012, by Metcalf et al. found that the recovery efforts misidentified Colorado river cutthroat trout as greenback cutthroat trout, which had been stocked across the continental divide but not documented. With many of the suitable fishless lakes having been stocked with the wrong cutthroat trout species, conservation efforts now look to remaining fishless alpine lakes and removal of impure and non-native fish for the subsequent recovery of true greenback cutthroat trout lineage in the South Platte watershed (Havlick & Biermann, 2021).

Alpine Fishless Lakes

Alpine watersheds are typically unproductive environments due to the colder temperatures constraining metabolic processes (Kraemer et al., 2017). Temperature, stream habitat, and energy influxes vary from headwaters to the mouth of the streams, causing natural shifts in macroinvertebrate assemblages with elevation and stream order (Doretto et al., 2020;

Vannote et al., 1980). Alpine lakes respond similarly to changes in temperature with changes in elevation, but also provide different habitats that hold slightly different invertebrate communities (Monaghan et al., 2005). Regional diversity of macroinvertebrate communities in alpine watersheds is a product of high amounts of species turnover between different lakes and streams (Monaghan et al., 2005). This site variation in macroinvertebrates is due to cold water alpine specialist species (Brown et al., 2007). Furthermore, activity levels and emergence of macroinvertebrates vary seasonally with peak hatches occuring in the spring and a drop off in mid to late summer (Nakano & Murakami, 2001). These hatches provide seasonal prey items for terrestrial predators.

Fishless lakes have long been the norm in alpine ecosystems and are home to diverse populations of specialist macroinvertebrates and amphibians. Typical alpine lakes have a variety of macroinvertebrate taxa including mayflies (Ephemeroptera), caddisflies (Trichoptera), and midges (Diptera) with lesser biomass contributions from stoneflies (Plecoptera), scuds (Amphioda), damselflies (Odonata), and more (Monaghan et al., 2005). In addition, four species of amphibians rely on alpine lakes in Colorado for breeding and to harbor young larvae through the summer (Corn et al., 2005). These amphibian populations are experiencing declines due to a variety of factors. The boreal toad, in particular, is a species that uses the shallows of alpine lentic ecosystems to breed between April and July, and is listed as an endangered species in Colorado (Muths & Corn, 2000). The fishless habitats that these amphibian and specialized macroinvertebrate species require are declining. In Colorado, only 24% of alpine lakes remained in a fishless condition in 1988, of which only 3% of total lakes were large fishless lakes (Bahls, 1992).

Macroinvertebrates

Introductions of trout into mountain lakes have similar and drastic effects on macroinvertebrate communities (Eby et al., 2006; Knapp et al., 2001; Pope et al., 2009; Tiberti et al., 2014; Toro et al., 2020). In Europe, non-native brook trout removal from alpine lakes has been the target of management operations to restore ecosystem communities in historically fishless lakes. Studies of brook trout removal suggest that trout selectively feed on larger macroinvertebrates. In the Western Italian Alps, brook trout presence had major impacts on littoral macroinvertebrates, where the presence of fish led to the local extinctions of many vulnerable species and entire feeding guilds of macroinvertebrates (swimmers and clingers) (Tiberti et al., 2019). Similarly, eradication of brook trout from an alpine lake on the Iberian Peninsula in Spain increased the richness of macroinvertebrate communities from 13 taxa to 27 taxa, resulting from increases in swimmer habit invertebrates (Toro et al., 2020). Reduction of certain macroinvertebrate species will shift community compositions and thus change the dynamics and health of alpine aquatic ecosystems.

Findings from studies in Europe are supported and consistent with studies from mountain ranges in the United States (Knapp et al., 2001; Pope et al., 2009; Schilling et al., 2009). In the Sierra Nevada Mountains, Knapp et al. (2001) found that trout presence significantly lowered the abundance of five out of the six clinger/swimmer mayfly taxa (Ephemeroptera) and four out of the five caddisfly taxa (Trichoptera), while increasing the abundance of worms (Oligochaeta), water mites (Acari), and mosquito larva (Diptera), with neutral effects on other burrowing taxa. In the Klamath-Siskiyou Mountains, Pope et al. (2009) found that lakes where trout were removed had higher amounts of hatching insect biomass resulting from increases in caddisflies, mayflies, and damselflies, but decreased midge hatches compared to lakes with trout. In addition,

insect predators were more likely to be found in the shallows than in deep water in lakes with trout compared to lakes without trout. Trout predation consistently targets larger bodied taxa from swimmer and clinger macroinvertebrate feeding guilds in alpine lakes at different geographical areas. This generally leads to shifts in invertebrate communities away from insect families like mayflies and caddisflies and towards burrowing macroinvertebrate taxa such as midges and aquatic worms.

Relatively few studies have examined the effects of cutthroat trout introductions on macroinvertebrate communities, but these limited studies suggest that the effects are consistent with the findings from trout introduction in alpine lakes in other areas. In Colorado, one of the first studies to look at the effects of trout on alpine aquatic ecosystems examined macroinvertebrate communities following the removal of brook trout and the subsequent introduction of cutthroat trout two years later in Emmaline Lake (Walters & Vincent, 1973). The authors found considerable similarities and few differences between the predatory habits of these two trout species. During the short amount of time that the lake was fishless, midge abundance and emergence increased and even a species of caddis returned to the lake at the end of the second year. Similarly, in the Uinta Mountains in Northeastern Utah, both cutthroat trout and brook trout selectively feed on and reduce the abundance of large invertebrates (mayflies, caddisflies, damselflies, and scuds) as well as three smaller chironomid midge taxa, while increasing the abundance of small daphnia, aquatic worms, and cyclopoid copepods (Carlisle & Hawkins, 1998). There appears to be no drastic differences in the predator influences of cutthroat trout vs brook trout. In cutthroat trout ranges, trout predation has similar effects on macroinvertebrate communities to other areas, reducing the number of insects that live to adulthood and targeting larger bodied insects.

Amphibians

Around the world, amphibians are declining at astonishing rates and extinction rates are currently about 200 times higher than the estimated background extinction rates (Carey, 1993; Collins, 2010). These declines are attributed to commercial use, contamination, changes in land use, infection, and introduced species (Collins, 2010). In the Rocky Mountains, declines are occurring at a higher rate in the southern regions into Colorado (Corn et al, 2005). Particularly, northern leopard frogs have already gone extinct in Rocky Mountain National park and remaining populations of boreal toads (*Bufo borealis*), wood frogs (*Rana sylvatica*), tiger salamanders (*Ambystoma tigrinum*), and boreal chorus frogs (*Pseudacris maculata*) are all rare and declining (Corn et al., 2005). Introductions of trout are putting additional stress on these already vulnerable amphibian species by adding competition and predation.

The negative effects of trout predation on frogs has been heavily documented. Introduced trout predators decrease the populations of amphibians through predation, increased injuries, induced changes in behavior/habitat, and competition (Collins, 2010). Even more, the small number of lakes that do remain fishless are often too shallow to sustain viable populations of amphibians through the winter and periods of drought (Bahls, 1992; Pilliod & Peterson, 2001). These effects are exemplified in the Sierra Nevada Mountain range in California, which has been heavily studied and documents drastic declines in yellow-legged frogs (*Rana muscosa*) following trout introductions. Trout were stocked in the early 1900s to enhance recreational fishing opportunities in the park, but these practices had detrimental effects on the amphibian populations due to tadpole predation (Bradford, 1989; Knapp et al., 2001, 2007; Vredenburg, 2004). The findings from these studies are supported by other studies from other areas (Bosch et

al., 2019; Lynne et al., 2007; Pilliod & Peterson, 2001; Tiberti et al., 2014), and suggest that trout negatively affect amphibian species, often leading to the local extirpation of amphibians.

Salamander larvae can also fall victim to trout predation, despite typically being larger than frog tadpoles (Hoffman et al., 2004; Pilliod & Peterson, 2001). Trout replace salamanders as the apex predators in alpine lakes. This dynamic changes the behaviors of larval salamanders, causing them to be more nocturnal hunters and restrict their movement to the shallows (Hoffman et al., 2004). The only species of salamanders that inhabit mountain lakes in Colorado is the tiger salamander. Tiger salamanders are top predators in the environments that they inhabit, but occupy a lower trophic level than adult trout (Boeckman & Whiteman, 2017). Introduced trout also consume similar but broader insect communities than do tiger salamanders, and thus the two species compete for food resources when in sympatry (Olenick et al., 1981). This competition has, in part, led to declines in the populations of tiger salamanders in the Rocky Mountains (Spear et al., 2006).

In addition, trout impact amphibian populations through non-consumptive ways. True toads have bufotoxins that make them unpalatable, but trout can still impact the health of tadpoles and eggs from the elevated stress and taste testing (Grasso et al., 2010; Lanier et al., 2017). In particular, Lanier et al. (2017) experimentally explored the non-consumptive effects of greenback cutthroat trout presence on the survival of boreal toad tadpoles in Colorado. They found that both captive breed and wild tadpole survival decreased by 25% and 13% respectively when exposed to trout due to higher stress and injuries from trout taste testing. The amount of taste testing stayed constant throughout the study suggesting that the trout didn't learn to associate boreal toad tadpoles with being unpalatable or were simply sampling out of aggression. Currently, attempts to replicate these results in natural settings through observational studies

have not been successful (Crockett et al., 2021; Lynne et al., 2007). Larger lakes, even when in competition with trout, still provide better habitat and survival rates for boreal toads than shallower fishless lakes that contain populations infected with a fungal pathogen.

Terrestrial Ecosystems

Growing knowledge of aquatic ecosystems suggests that there are considerable connections between aquatic and terrestrial environments (Baxter et al., 2004; Soininen et al., 2015). For one thing, both macroinvertebrates and amphibians' adult lives take place in riparian ecosystems surrounding lakes and rivers. Introductions of trout significantly reduces the emergence of aquatic insects and thus reduces the amount of prey and nutrients available to terrestrial organisms (Epanchin et al., 2010; Pope et al., 2009; Walters & Vincent, 1973). In addition, terrestrial invertebrates make up a substantial portion of trout diets in alpine lakes, especially later in the summer and fall when there is reduced aquatic invertebrate activity (Nakano & Murakami, 2001; Walters & Vincent, 1973). These influxes of prey and nutrients are important to many riparian organisms such as birds and spiders (Baxter et al., 2005).

Reductions in the emergence of aquatic insects from trout predation can affect bird populations around alpine lakes. Studies of bird consumption of aquatic insects suggest that insectivorous birds that are more heavily reliant on aquatic insects obtain approximately 25% of their diet from aquatic insects and spend more time in riparian habitats than do other insectivorous birds (Epanchin et al., 2010; Murakami & Nakano, 2002; Nakano & Murakami, 2001). Epanchin et al. (2010) looked at the indirect effects of introduced trout on the presence of Rosy-Finches through competition for the consumption of mayflies in the Sierra Nevada mountain range in California. They found that Rosy-Finches had significantly lower abundances at lakes containing fish due to reductions in available adult mayflies. The timing of the peaks of

these spring aquatic hatches aligns with when many birds like Rosy-Finches are reproducing and feeding their young (Epanchin et al., 2010; Nakano & Murakami, 2001). Therefore, aquatic hatches could be an important seasonal prey item for reproducing birds. Trout induced reductions in aquatic invertebrates could be indirectly affecting the reproduction and survival of terrestrial bird species.

Like birds, riparian arachnids are heavily dependent on aquatic insects, yet there are seemingly few studies on spiders around aquatic ecosystems in conjunction with introductions of non-native fish predators. In New Zealand, Collier et al. (2002) found that riparian spiders were heavily reliant on aquatic insects, with 66% of web-building spiders diets and 55% of free-living spiders diets coming from aquatic sources. Another study conducted in Japan sought to experimentally test the effects of introducing non-native rainbow trout on aquatic insects and terrestrial spider communities in a small mountain stream (Baxter et al., 2004). Out of the three treatments (net covered, rainbow trout, and rainbow trout plus net covered streams), all three treatments had significantly lower terrestrial spider abundance compared to the normal stream (control). The introduced rainbow trout treatment had significantly lower emergent biomass when compared to the control, suggesting that trout interactions in the stream alter aquatic food webs. This reduction in emergent biomass resulted in a 65% decrease in spider abundance in the riparian environment. Trout introductions inevitably change these ecosystem dynamics and functions, affecting organisms that reside in both aquatic and terrestrial environments.

Conclusion and Future Management Applications

Trout introductions into historically fishless lakes alter the state of these alpine ecosystems. Introduced trout decrease the richness and abundance of macroinvertebrates, threatening to eliminate specialized taxa and homogenize and decrease regional diversity.

Amphibian species that reside in lakes with trout face increased predation and competition for food resources, adding to other factors leading to the declines in these amphibian populations. Changes in macroinvertebrates and amphibians indirectly affect terrestrial environments, reducing riparian spider and bird abundances. Importantly, peak macroinvertebrate emergences span a timeframe in the spring that aligns with the breeding seasons of many alpine birds and amphibian species. Reductions in macroinvertebrate emergent biomass, from trout predation, could have long term effects on terrestrial organism populations. Given the extent to which alpine lakes have been transformed into trout fisheries, few lakes remain fishless for species that are intolerant to trout predation and competition.

Future selection of lakes to remain fishless needs to be systematic and balanced with the conservation of cutthroat trout. Given the negative impacts that trout introductions can have on alpine aquatic and terrestrial environments, conservation management should plan to leave some lakes and rivers fishless. Currently, there is a need for more deep fishless lakes as they can better harbor amphibian populations and a greater diversity of macroinvertebrate communities (Pilliod & Peterson, 2001). In addition, the conservation of cutthroat trout depends on stocking trout into deep fishless alpine lakes. These conflicting conservation agendas will require the removal of non-native trout from some lakes to be left fishless or stocked with cutthroat trout. Cutthroat trout translocations should be primarily directed towards lakes and rivers where fish have been removed that might have already lost their specialized macroinvertebrate communities. Finding solutions for the recovery of cutthroat trout and sustaining aquatic alpine ecosystem health is no easy task and will require thoughtful planning that takes a more holistic ecosystem approach.

Brook trout and other non-native trout species have been targeted for removal from alpine lakes to protect native diversity in Europe, California, and more places around the world.

From these locations, alpine lakes appear to be quite resilient (Pope et al., 2009; Tiberti et al., 2014), and can completely recover in 10 to 20 years following the removal of fish (Knapp et al., 2001). Removal of trout will likely not be supported by the public (Harig et al., 2000). Trout fishing is a prominent hobby in Colorado, and it provides large contributions to the economy (Loomis & Ng, 2012). Public approval is unfortunately an important factor in these management considerations. Recreational fishing opportunities will need to be, for a large part, maintained. In the meantime, precaution should be taken with stocking trout into fishless lakes and should take into account presence of amphibians and overall habitat complexity. The historic fishless condition of these lakes and the ecosystem changes that trout induce warrants further consideration. Managers should, moving forward, take a more holistic approach that balances the constraining effects of trout predation with the urgent recovery needs of greenback cutthroat trout and other cutthroat trout subspecies in Colorado. The haphazard introductions of predator fish with little concern for native ecosystems is a largely recognized mistake of our nation's past. A mistake that as a society, we should attempt not to repeat in current times.

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CHAPTER 2. GRANT PROPOSAL

Effects of Trout Presence on Alpine Lake Macroinvertebrate Emergence in Colorado

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October 30, 2021

Section 1: Abstract

Trout have been extensively introduced across the United States into many watersheds including historically fishless alpine lakes, mainly for fishing purposes. Trout are ultimately predators that exhibit top-down control in aquatic ecosystems. Evaluating the impacts of trout introductions on aquatic and terrestrial ecosystems will aid fisheries managers in making decisions regarding seemingly competing agendas: enhancing recreational fishing opportunities and protecting native environments. Aquatic macroinvertebrate communities are a perfect focus group for evaluating the ecosystem interactions of trout as they are the predominant prey source for trout and have connections to larger ecological functions in both aquatic and terrestrial environments. Currently, there have been very few studies looking at the effects of trout predation on alpine macroinvertebrate communities in Colorado, which will be the basis of this study. I propose to test whether trout presence reduces macroinvertebrate emergence and whether trout predation affects the temporal variation in emergent biomass and diversity. Sampling will start in May and go until the end of August. Three sets of geographically paired alpine lakes, one fishless and one cutthroat trout lake, will be sampled simultaneously each month with randomly placed emergence traps. Results from this study will shed light on the impacts of trout introduction into historically fishless lakes, specifically changes in adult macroinvertebrate emergent biomass and diversity. Macroinvertebrate emergence represents a unique connection between aquatic and terrestrial environments, aiding managers in understanding and protecting the ecological health of alpine aquatic and terrestrial ecosystems. This study fits into the broader field of invasion ecology and would have implications for the general management of trout fisheries worldwide.

Section 2: Introduction

Objective:

This study will assess changes in macroinvertebrate emergent biomass in alpine lakes with and without trout. The conclusions from this study will add to the limited research on trout introduction impacts on alpine lake ecosystems in Colorado. This knowledge will inform management of both the aquatic and terrestrial ecosystem effects of trout introductions into historically fishless lakes, which is particularly important for identifying appropriate recovery efforts of cutthroat trout subspecies in this state.

Literature Review:

Fish Introductions

Introductions of fish species such as trout have been pervasive across the United States since the early 1900s (Eby et al., 2006; Halverson, 2008). Historically, 95% of alpine lakes in the United States were fishless, but introductions of trout for recreational fishing have decreased the number of fishless lakes to 40% (Bahls, 1992). The remaining fishless lakes are typically too shallow to sustain trout populations. In Colorado, only 3% of the large alpine lakes in the state remain in a historically fishless state as trout were stocked into 76% of all alpine lakes in the state by 1988 (Bahls, 1992). Trout in these lakes include both non-native trout species like brook trout and native cutthroat trout subspecies, all of which experienced declines in abundances and range sizes over the past century (Allendorf & Leary, 1988; Wiltzius, 1985). For native cutthroat trout subspecies, alpine lakes and rivers now serve as refuges due to the isolated nature of these watersheds, separating cutthroat trout populations from detrimental interactions with non-native trout species (Young & Harig, 2001).

Macroinvertebrate Impacts

Introductions of trout have caused trophic cascades in alpine lakes. Trout predation targets larger bodied macroinvertebrate taxa, resulting in decreases in the abundances of Ephemeroptera (mayflies), Trichoptera (caddisflies), and predatory taxa such as Coleoptera, Megaloptera, and Odonata (Knapp et al., 2001; Pope et al., 2009). Over time, entire macroinvertebrate species can become locally extinct in lakes containing trout, decreasing macroinvertebrate diversity on local and regional scales (Tiberti et al., 2014; Toro et al., 2020). In the Klamath-Siskiyou Mountains of California, these subsurface feeding interactions decreased the emergence of larger bodied macroinvertebrates including mayflies and caddisflies, while increasing the emergence of smaller Diptera (midge) species (Pope et al., 2009). Studies on fishless lakes are generally widespread but incomplete with the majority of literature coming from mountain ranges in California and Europe.

Connections to Terrestrial Environments

The cascading effects of trout introductions on macroinvertebrate communities can percolate into terrestrial ecosystems. While the majority of the lifespan of macroinvertebrates takes place subsurface, the adult lifespan takes place in surrounding riparian ecosystems (Baxter et al., 2005). Macroinvertebrate emergence from alpine lakes varies seasonally with a general peak in the early summer from larger macroinvertebrate hatches and consistent yearlong hatches of midges (Nakano & Murakami, 2001; Salvarina et al., 2017). This emergence represents major fluxes of nutrients between aquatic and terrestrial ecosystems, and comprise significant portions of terrestrial organisms diets including various species of birds, bats, and spiders (Collier et al., 2002; Epanchin et al., 2010; Nakano & Murakami, 2001). A reduction in earlier life stages of

macroinvertebrate would reduce macroinvertebrate emergence and have cascading effects on organisms in terrestrial environments.

Colorado Studies

Very few studies have examined macroinvertebrate communities in alpine regions in Colorado, especially in conjunction with fish introduction. Walters and Vincent (1973) examined the effects of trout on macroinvertebrate communities in Emerald Lake, where brook trout were removed and replaced with cutthroat trout a few years later. During the fishless period, midge emergence and abundance increased suggesting that there could be large ecological differences between lakes containing trout and lakes that don't. The only other study to look at this topic in the state was conducted by Detmer and Lewis in 2019. They found that macroinvertebrate food web functions are resilient to trout introductions, despite species compositional changes, as smaller macroinvertebrates increased and filled the niches of macroinvertebrate species that were targeted by trout predation. A question that arises from this study is, do these new communities of macroinvertebrates also fulfill the terrestrial ecosystem roles of historically fishless lake macroinvertebrate communities that were/are seasonal food sources for riparian inhabiting organisms? Colorado needs a current alpine lake macroinvertebrate study to assess the impacts of trout introductions on aquatic and, indirectly, riparian ecosystems.

Anticipated Value:

This study will determine to what extent trout introductions change macroinvertebrate emergence across the summer season in Colorado. These findings will provide managers information on how to best allocate resources to conserve alpine ecosystems and maintain trout recreational fishing opportunities. This knowledge is particularly pertinent to current cutthroat trout recovery efforts, which focus on establishing more pure genetic cutthroat trout populations

in alpine watersheds to isolate these populations from the detrimental interactions with nonnative trout species (Havlick & Biermann, 2021). In a broader sense, this study will add to the literature on the ecological interactions and implications of trout introductions. As trout are a prominent and important game fish, knowing about the trophic interaction of these species will help to ensure their introductions don't come at the expense of native ecosystems.

Questions (*Q*) and Hypotheses (*H*):

Q1: Does macroinvertebrate emergent biomass and composition differ between different trout conditions (present vs fishless)?

H1: Trout will consume larger macroinvertebrates, reducing mayfly and caddisfly emergence.

Lakes with trout will have lower macroinvertebrate emergent biomass compared to fishless lakes. Trout presence will be correlated with decreased abundance of caddis and mayfly emergence, but increased midge emergence.

Q2: Are there seasonal differences in emergence patterns between lakes with or without trout?

H2: The loss of caddis and mayfly emergence will decrease peak summertime emergence. In all lakes, emergence will spike in the early summer around early June, but the peak will be greater in fishless lakes while lakes with trout will have less variation in emergence.

Section 3: Methods

Study Sites:

Study sites are restricted to the alpine lakes in the Colorado Rocky Mountains (Figure 1).

Lakes are paired, one fishless and one cutthroat trout lake, by geographical similarity to control for spatial variation in macroinvertebrate communities. From Colorado Parks and Wildlife databases, a total of three lake pairs were identified for this study: (1) Silver Dollar Lake (trout)

and Square Tops Lake (fishless), (2) Lost Lake (trout) and Wheeler Lake (fishless), and (3) James Peak Lake (trout) and Little Echo Lake (fishless).

Data Collection:

I will sample macroinvertebrate emergence using devices developed by Pope et al. (2009). The devices are conically shaped polyester No-See-um meshing with a wire framework attached to an inflatable bicycle tube. Four emergence traps will be deployed to each lake, positioned around the lake using a random number generator for compass oriented location. Each trap will be staged for 40 hours in ~1 m depth of water, and secured to the lake bottom and shoreline with tent stake anchors. Macroinvertebrate samples will be obtained throughout the 40 hour timeframe in the morning, evening, and following morning at each lake, once a month. Macroinvertebrate emergent biomass samples will be preserved in 70% ethanol and brought back to the lab for weighing and grouping based on their orders. Habitat surveys will consist of visual estimates of 30 transects around the lake at distances 50 cm, 100cm, 250cm, and 500cm from the bank, looking at presence of aquatic vegetation, woody debris, and silty substrate. Water temperature, elevation, and lake area will be measured using temperature probes and ArcGIS software.

Data Analysis:

Data will be compared using an analysis of variance (ANOVA) in R statistical software (R Core Team, 2021). To test for differences in macroinvertebrate communities between troutcontaining and fishless lakes, an ANOVA will be run to compare the totality of emergent biomass and diversity between lake treatments. To look at the seasonal variation in macroinvertebrate emergence, the lake pairs will be further grouped by month. Habitat characteristics will be added as interaction terms to be kept constant. Multiple models will be fit

using the linear models function in the program R, and the Akaike Information Criterion (AIC) will be used to determine the best model. Post hoc determination of significant differences will be made with adjusted p-values from a Tukey HSD test. All the data will be visualized with a principal components ordination analysis.

Negative Impacts:

Impacts from the study to alpine communities should be minimal. Data collection will involve minor habitat disturbances that will be less than that on an angler visit. We will mitigate these disturbances by using leave-no-trace guidelines. Macroinvertebrate samples will be obtained and removed from the study sites, but this will be a low proportion of the total emergent biomass, less than 1%, given the size of the emergence traps and the total lake area.

Schedule:

The study will begin the first week of May 2022 and go until the end of August 2022. Prior to the study, my field assistant will be taught the sampling procedures during the last week of April. One lake pair will be simultaneously sampled per week, divided up between myself and a trained research assistant. Each lake pair will be sampled once a month for a total of four samples per lake. One week will be taken off per month for recovery and lab examinations of macroinvertebrate taxa, biomass weighting, and data entry. Following data collection, the month of September will be spent writing and submitting the final paper.

Section 4: Budget

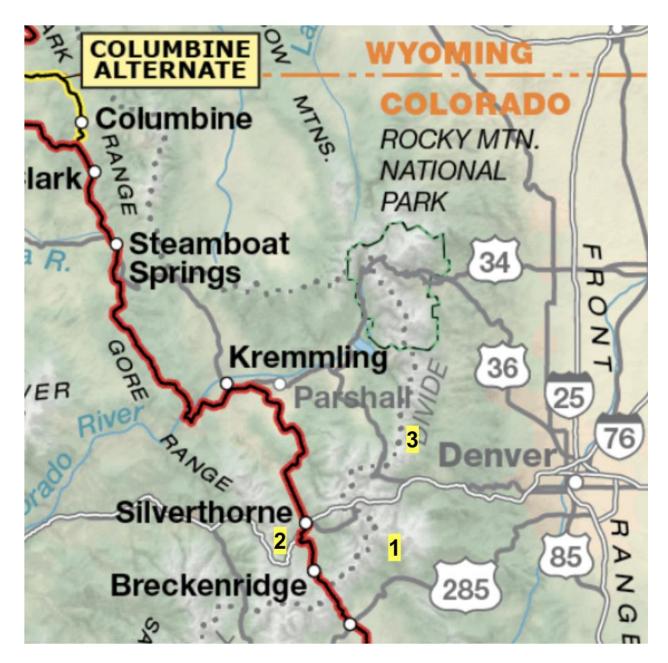
Item	Cost (unit)	Quantity	Total	Justification
Field Assistant Stipend	\$1000 per month	1 assistant x 4 months	\$4000	To assist with data collection and analysis. Include physical labor of trekking gear up to the alpine lakes.

Coleman 10-in Steel Tent Pegs (4 stakes)	\$7	4	\$28	Attachment points for emergence traps
Zebco Omniflex 20 lb Monofilament Fishing	\$3	1	\$3	Anchor line for emergence traps
Hand Held Bicycle Pump	\$27	2	\$54	Inflate bicycle tires on emergence traps
27 in Bicycle Tubes	\$7	8	\$56	Flotation for the emergence traps
No-See-Um Meshing	101 inch role x \$5/ft	32feet	\$160	Netting for the emergence trap
Mechanics Wire	\$1	4	\$4	Structural support for the netting
Traceable Waterproof Remote Probe Thermometer	\$43	2	\$86	For gathering water temperature in the habitat survey.
Food Budget	\$8 per meal	240 meals	\$1920	Meals in the field
Gas	\$0.56 per mile	1320	\$740	Travel to study sites
MSR IsoPro Fuel	\$6	8	\$48	Cook food
Aspirator	\$9	2	\$18	Collect insects from the netting
70% ethanol, gallon	\$45	1	\$45	Preserve captured insects

Total Proposal Request = \$7117

Appendix

^{*}Additional gear and tools for data analysis will be provided by Regis University. *



<u>Figure 1:</u> The distribution of lake paired sites in the Rocky Mountains of Colorado as distinguished by yellow highlighted numbers 1-3: (1) Silver Dollar Lake (cutthroat trout) and Square Tops Lake (fishless), (2) Lost Lake (cutthroat trout) and Wheeler Lake (fishless), and (3) James Peak Lake (cutthroat trout) and Little Echo Lake (fishless).

Tyler A. Erickson

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EDUCATION

Masters: Environmental Biology

Regis University- Denver, CO (expected graduation May 2022)

Double Majors: Biology and Environmental Studies; Cumulative GPA: 3.87

Pacific Lutheran University- Tacoma, WA (BA- May 2020)

- Dean's list Pacific Lutheran University (2017, 2018, 2019, 2020)
- Chi Alpha Sigma inductee (2020)
- Magna Cum Laude Graduate (May 2020)
- Studied abroad in New Zealand (January 2018)

PROFESSIONAL/WORK EXPERIENCE

Bear Creek Golf Course (August 2020- September 2021)

 Communicated with and served customers' needs while maintaining the cleanliness of the carts, members clubs, garage, and golf range. Special experience included stocking rainbow trout into the main lake, driving small vehicles, and course/facilities management.

Wildlands Restoration Volunteers - Volunteer (Summer 2019)

 Participated in various volunteer restoration activities through this non-profit organization aimed at managing invasive species, suppressing fires, and planting native trees, shrubs, herbs, and grasses.

Trout Unlimited- Volunteer (July 2019)

 Backpacked greenback cutthroat trout in the Herman and Dry Gulch areas of the Colorado high country as part of Trout Unlimited's efforts to reintroduce this native trout subspecies into creeks within its historical range. We removed beaver dams at Rock creek in another effort to prepare a section of river for greenback stocking.

Blue Valley Ranch- Volunteer (July 2019)

 Assisted in the marking, measurement, and stocking of rainbow trout into three lakes on the Blue Valley Ranch in Summit County, CO.

Colorado Addicted Trail Builders- Volunteer (June 2019)

 Helped create new trails with a non-profit organization using pickaxes, rock bars, and shovels to move dirt, rocks, and boulders into optimal/precise positions in the Poudre Canyon.

Good Shepherd Orphanage in Ghana (July 2017)

I served on a mission trip designed to help the orphanage obtain better food security, establish a
sustainable water and vegetable supply, and reduce the spread of malaria by designing and implementing
mosquito net barriers for the windows of the dorms. I tutored basic math at the local school and
organized soccer drills with the orphans.

LEADERSHIP/ACHIEVEMENTS

NCAA Div. III Athlete (Men's Soccer 2016-2020)

o Trained and competed for 16+ hours a week while juggling a full course load. Part of the leadership and camaraderie that enabled us as a team to win three Northwest Conference titles.

Eagle Scout Award (January 2016)

 Earned the rank of Eagle Scout through countless hours of dedication, leadership, and service with the Boy Scouts of America: troop 537. Extensive outdoor experience in many environments and weather conditions for extended amounts of time.

UNDERGRADUATE RESEARCH EXPERIENCE

Environmental Studies Capstone Project - Pacific Lutheran University (Fall 2019-Spring 2020)

- "Greenback Cutthroat Trout Case Study: Shifting human values from mining and exotic fish culture to native species conservation from 1850s to present in Colorado"
 - Examined the history of conservation practices and human values with regard to greenback cutthroat trout from an interdisciplinary perspective as my capstone project for Environmental Studies.

Conservation Genetics Biology Capstone Project - Pacific Lutheran University (Fall 2019)

- "Hybridization in Westslope Cutthroat Trout: Implications for Conservation"
 - Explored the threat of hybridization between Westslope Cutthroat Trout and rainbow trout, culminating in a synthesis paper and a biology department capstone presentation.

<u>Plant Diversity and Distribution Course Field and Lab Work- Pacific Lutheran University</u> (Spring 2020)

 Field and laboratory research included identification of plants in different seasons. Additional work looked at the cultural use and value of yarrow, a native medicinal herb.

Environmental Methods 350- Pacific Lutheran University (Fall 2018-Spring 2019)

 Combined disciplines in humanities and the sciences to assess the health of the Clover/Chambers Creek watershed: specifically utilizing environmental history, indigenous studies, geology, chemistry, and biology. Featured macroinvertebrate sampling, water quality testing, stakeholders analysis, and ample write ups and presentations.

Evolution Course Lab Work- Pacific Lutheran University (Fall 2019)

 Studied the population dynamics of the reticulate sculpin in 10 rivers in Washington to test the Chehalis refugium hypothesis using morphological measurements and genetic analysis, utilizing excel and gel electrophoresis.

Natural History of Vertebrates Course Field and Lab Work- Pacific Lutheran University (Spring 2020)

 Sampled many different habitats using various techniques to identify vertebrates found in Washington state and visualize the morphological changes that make up the evolutionary history of all vertebrates.
 Experience with identification of preserved specimens of fish, mammals, and reptiles using dichotomous keys. Formulated two natural history reviews for the Pacific giant salamander and the rubber boa.

GRADUATE RESEARCH EXPERIENCE

Externship: Colorado Parks and Wildlife- Regis University (Upcoming, Spring semester 2022)

Statistical analysis of mark and recapture fisheries data from Parvin Lake, Co looking at the survival rates of different hatchery strains of rainbow trout.

Grazing Advanced Ecology Lab - Regis University (Fall 2021)

- "The Impacts of Bison and Prairie Dogs on Managed Semi-Arid Mixed-Grass Prairie Plant Communities"
 - Field quadrat sampling of grassland plant communities, soil, and scat using GPS located random points to access black-tailed prairie dog and bison grazing impacts on mixed-grass prairie in Colorado. Data was compiled and analyzed using R statistical software.

Asian Elephant Behavioral Analysis: Denver Zoo- Regis University (Fall 2021)

- "Male Asian Elephant Nighttime Resting Behavior and Sociality"
 - Coded 18 elephant hours of nighttime elephant behavior on Zoomonitor from the bachelor herd at the Denver Zoo.

Macroinvertebrates Advanced Ecology Lab-Regis University (Fall 2021)

Sampled benthic macroinvertebrates and fish with D-nets and seines using standardized protocols at Coal creek in Colorado, including extensive habitat surveys.

SKILLS

- R statistical software
- Macroinvertebrate, fish, and wildlife identification
- Plant identification
- NEPA process and document writing
- Grant proposal writing
- Time management and organization

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CHAPTER 3. JOURNAL MANUSCRIPT

Post-Stocking Survival and Myxospore Evaluation of Whirling Disease Resistant

Rainbow Trout Strains

Abstract

The introduction and ensuing spread of whirling disease in the United States in the late 1950s caused drastic declines in Rainbow Trout populations. Currently, the German Rainbow (GR) is a strain known to have high whirling disease resistance but has an extensive history as a domesticated food production fish. Prior research suggests that crosses of GR with susceptible wild Colorado River Rainbows (CRR) create offspring with moderate whirling disease resistance and survival rates in lotic environments, but analysis on lentic environments with larger predators has been limited. We evaluated the survival rates and infection severities of fingerling Rainbow Trout stocked into a lentic environment from four different strains with theoretically varying resistances to Mxyobolus cerebralis: GR, Harrison Rainbow (HAR), and two crossed strains between GR and HAR. CPW researchers conducted a mark-and-recapture study of these four strains over seven years and three age-classes in Parvin Lake. Using Seber dead-recoveries models, we found that HAR and the F1 crossed strain (HXH(50:50)) had comparably high survival rates followed sequentially by the F2 cross (HXH(75:25)) and GR. Whirling disease resistance, as estimated from the severity and probability of infection, generally increased with increasing GR strain genetic background. To reduce spore loads and increasing resistance in managed Rainbow Trout populations, we suggest stocking GR crossed strains like HXH(50:50) that have respectable whirling disease resistance and high survival rates, enabling wild

recruitment with increased cost-efficiency of stocking. Future studies should assess more GR crosses across a greater suite of aquatic habitats.

Introduction

Rainbow Trout Oncorhynchus mykiss, a salmonid native to the west coast of North American, is an extremely important and influential sport fish worldwide (A. Halverson, 2010; MacCrimmon, 1971). Since first being cultured on the McCloud River in California in 1874, Rainbow Trout have been introduced to every continent except Antarctica to enhance fisheries stocks (MacCrimmon, 1971). In the United States alone, Rainbow Trout have been frequently introduced into highly managed put-and-take fisheries in nearly every state and have successfully established populations in 41 states, with only six of those states being within their historical native range (MacCrimmon, 1971; Rahel, 2000). A considerable amount of time and money is expended enhancing and sustaining cold water fisheries in the United States, where management of Rainbow Trout populations accounts for upwards of 40% of the total weight of fish stocked across all freshwater environments. Of these Rainbow Trout, 60% are stocked into western states (M. A. Halverson, 2008). Not only are Rainbow Trout a significant sport and food fish, they also are heavily used in research as a model organism and therefore contribute to fisheries and ichthyological knowledge and conservation more broadly (Thorgaard et al., 2003). Continued research on Rainbow Trout provides insight into how to efficiently and optimally manage these important fish populations for future generations in the face of adverse and changing environmental conditions.

A major challenge for populations of Rainbow Trout is the myxozoan pathogen *Myxobolus cerebralis*, the cause of salmonid whirling disease. When a salmonid comes into contact with the triactinomyxon (TAM) stage of the parasite, the spores attach to and enter the

fish's body, subsequently spreading through the peripheral and central nervous system and congregating in the cartilage (Sarker et al., 2015). With high enough spore concentrations, M. cerebralis infection leads to neurological difficulties and skeletal deformities in the host fish. Younger salmonids with more cartilage relative to bone are at greater risk of whirling disease abnormalities and related death, which has constrained wild salmonid recruitment (EL-Matbouli et al., 1995). Rainbow Trout are one of the most susceptible species to whirling disease, followed closely by Eastern Brook Trout Salvelinus fontinalis and various subspecies of Cutthroat Trout Oncorhynchus clarkii (Vincent, 2002). The spread and development of whirling disease has severely depleted populations of Rainbow Trout in the United States (Nehring & Thompson, 2001). Loss of self-sustaining Rainbow Trout populations is particularly acute in Colorado where populations were reduced to 10% of historical levels following the outbreaks of whirling disease in 1987 (Schisler et al., 2000; Schisler & Fetherman, 2009). Whirling disease remains a prominent issue for salmonids such as the Rainbow Trout, and with recent outbreaks of M. cerebralis in Alberta, Canada and the first occurrence of whirling disease in natural populations in the Southern Appalachian Mountains (James et al., 2021; Ksepka et al., 2020), finding solutions is paramount.

There have been two main courses of action for addressing whirling disease, both focusing on one of the two hosts for *M. cerebralis*. *M. cerebralis* has a two-stage life cycle dependent on two families of hosts: salmonid fishes and tubificid oligochaete worms (R. P. Hedrick & El-Matbouli, 2002). One of the promising avenues for combating whirling disease is with finding whirling disease resistant strains in both of these host families. Currently, two main strains of Rainbow Trout are known to possess whirling disease resistance: the German Rainbow (also known as Hofer Rainbows, referred to as GR hereafter) and the Harrison Lake Rainbows

(referred to as HAR hereafter). Despite originating from Rainbow Trout populations in the United States, GRs possess substantially greater whirling disease resistance than HARs due to their subsequent lengthy domestication as a food fish in Germany, where whirling disease is endemic (R. Hedrick et al., 2003; M. P. Miller & Vincent, 2008). Whirling disease resistance is heritable in Rainbow Trout varieties created from cross strain breeding with GR (Fetherman et al., 2011; Schisler et al., 2006), opening up possibilities for fisheries specific, strategic strain combinations. With the complexity of addressing *M. cerebralis* at the environmental scale (removing spores and/or tubificid habitat), efforts to regain historical Rainbow Trout populations and enhance fisheries fall heavily on identifying whirling disease resistant strains of Rainbow Trout and *Tubifex tubifex* that can supplement wild populations.

Different strains of Rainbow Trout are known to vary in survival rates in natural settings and even have habitat specific interactions (Ayles & Baker, 1983; Brauhn & Kincaid, 1982). One of the determining factors for the survival of strains of Rainbow Trout is the degree of domestication. There is concern related to stocking domesticated fish because their genetic characteristics developed during human captivity may not translate to survival in natural settings and, even more concerningly, could dilute the genetics of locally adapted wild populations (Allendorf et al., 2001; M. P. Miller & Vincent, 2008). Domesticated strains are associated with diminished behavioral responses to the threat of predation and increased growth rates, which can have variable effects on survival in natural settings (Johnsson et al., 1996; Vandersteen et al., 2012). GRs have been bred in hatcheries for more than 100 years and are highly domesticated (R. Hedrick et al., 2003). Observations suggest that GRs are very surface oriented which would presumably lead to increased risk of predation and decreased survival rates (Schisler & Fetherman, 2009). On the other hand, HARs are a wild strain from Montana that is well adapted

to lake habitats and tend to prefer cruising and foraging in deep water (Schisler & Fetherman, 2009). Crosses between these two strains could theoretically maximize the benefits of both strains individually to create a whirling disease resistant "wild" strain. Ongoing research from biologists at Colorado Parks and Wildlife (CPW) on survival and physiological performance of whirling disease resistant strains has shed light on the possibility of using pure GRs for brood stock (Avila et al., 2018; Fetherman et al., 2011). In particular, Avila et al. (2018) found comparable survival rates between pure GRs and F1 crosses of GRs and wild Colorado River Rainbow Trout (referred to as CRR hereafter) in both small tributary streams and laboratory experiments in which Brown Trout were used as predators. Despite this compelling evidence, little is known about how survival of stocked fingerlings of whirling disease resistant strains compare to wild strains of Rainbow Trout in lentic environments.

The goal of this study is to assess how survival rates differ among five strains of Rainbow Trout when stocked into a natural lake setting as fingerlings. We hypothesize that survival rates of fingerling Rainbow Trout will vary between the different strains due to inherent variation in the levels of domestication and resistance to whirling disease. In other words, "wild" strains that have a higher resistance to whirling disease will likely have greater survival than domestic strains in natural settings. Therefore, we expect fish with higher ratios of genetic background from GR Rainbows will have decreased survival rates, probability of *M. cerebralis* infection, and infection severity due to the highly domesticated and whirling disease resistant nature of this strain. Due to the overall size and age of the fingerling Rainbow Trout stocked into lentic environments, whirling disease is not likely to be a major source of mortality for these stocked fish (Ryce et al., 2005). Given this information, we predict that HARs will have the highest survival rates due to their wild ancestry in lake environments and moderate whirling disease

resistance. Identifying a Rainbow Trout strain that blends the benefits of whirling disease resistance with wild genetics for maximal survival will assist in fisheries management in lakes, providing optimum recreational fishing opportunities with efficient economic returns to creel.

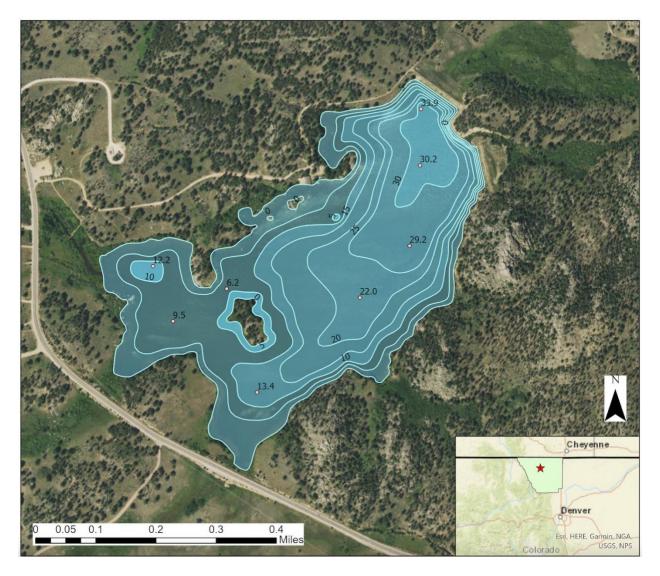


Figure 1: Bathymetric map of the Parvin Lake study system in Larimer County, Colorado. Numbers represent depth in feet.

Methods

Study system

Assessment of the survival of different strains of Rainbow Trout took place at Parvin Lake in the Northern Part of Rocky Mountains in Larimer County, Colorado, about 45 miles Northwest of Fort Collins (Klein, 1983). Parvin Lake is situated in Parvin Lake State Wildlife Area at 8,130ft in elevation, and is one of several lakes and State Wildlife Areas in the area known as Red Feather Lakes. Parvin Lake was created in 1927 from the damming of Lone Pine Creek and has since served as a fisheries research station for CPW (Buscemi, 1961). It is a small shallow mountain lake, about 62 acres in area, with a large littoral zone containing abundant aquatic vegetation and invertebrates (Fig. 1). Riparian habitats are primarily dominated by willows (Salix sp.) and mixed conifer forests. Several stocked species of fish are present in this lake and include notable populations of predatory fish, including Brown Trout (Salmo trutta), Tiger Muskie (Esox masquinongy x Esox lucius), and Splake (Salvilinus namaycush x Salvelinus fontinalis). Fishing pressure is monitored with check-in stations at access points from the parking lot, and gear is restricted to artificial flies and lures only with a bag limit of 2-fish. M. cerebralis was inadvertently introduced to the system in 1989 from Rainbow Trout that were stocked from an infected hatchery. The presence of large predatory populations of both fish and birds (e.g., Osprey, Great Blue Heron), along with the presence of *M. cerebralis*, makes Parvin Lake an appropriate study site to thoroughly test the survival and recruitment of different strains of whirling disease resistant Rainbow Trout.

Fish Rearing and Marking

CPW staff stocked five strains of Rainbow Trout that were reared and marked at the Rifle Falls Fish Hatchery from 2007-2009, including the highly resistant but domesticated GR and

moderately resistant wild HAR. The other two strains introduced are the result of cross of the strain of GR and HAR (referred to as HXH(50:50) hereafter) and a backcrosses of HXH(50:50) with GR (referred to as HXH(75:25) hereafter). These cross strains are inferred to have intermediate degrees of whirling-disease resistance and domestication (Schisler et al., 2006). The hatchery reared fry for approximately two months and then uniquely batch marked them with coded wire tags (Northwest Marine Technology, Shaw Island, Washington) using a Mark IV CWT injector to insert the tag into the snout of each trout to distinguish individual strains. Coded wire tag retention is estimated to be upwards of 90% based on previous studies (Munro et al., 2003). Hatchery personnel monitored tagged fish for 24 hours to ensure tag retention, and mortalities or fish with tag losses were replaced with freshly tagged fish. At the same time, personnel measured, recorded, and compiled the weight (grams) and fork length (mm) of each individual fish as an average for each strain of Rainbow Trout in each year.

Stocking and Electrofishing

For recapture analysis, CPW biologists stocked a total of 2800, 2050, and 1005 fingerling fish per strain into the inlet of Parvin Lake in 2007, 2008, and 2009, respectively. Timing of stocking varied slightly between years with fish being stocked on August 14 in 2007, July 31 in 2008, and August 12 in 2009. CPW researchers sampled Parvin Lake using boat electrofishing. Sampling took place four times a year in the months of April, June, August, and October starting in late August of 2007 and continuing until October of 2013. Electrofishing was conducted at night using a 17 ft Waterman Welding aluminum electrofishing boat with floodlights and a single anode boom, operating with a Coffelt Manufacturing VVP-15, powered by a Honda 5500 watt generator. The pulse DC current had voltage set at 350 to 400 volts, 40% pulse width, and 60 pulses per second. Each survey consisted of systematic collection consisting of a single pass

around the entire perimeter of the lake, maintaining a ≤10 meter distance from the shoreline. Two dip-netters collected fish from the bow of the boat, with the objective of sampling 30 individual marked rainbow trout from each age-class during each sampling event. Fish remained in livewells with flow-through fresh water until processing. Collectors sorted Rainbow Trout using a Northwest Marine Technologies handheld coded wire tag detector and retained fish with tags. Researchers weighed, measured, and removed the heads of Rainbow Trout for latter strain and year class identification and myxospore analysis.

Laboratory Analysis

Field samples were frozen for later tag extraction and *M. cerebralis* myxospore enumeration. Tags were excised from individual heads by dissection with a scalpel, and a coded wire tag detector was used to identify the location of the tag in the tissue. Researchers cleaned tags with 100% ethanol immediately after extraction and viewed them under a dissecting microscope to read tag numbers, which were then used to identify the strain of Rainbow Trout and the year of stocking for each fish collected. Cranial tissue was then delivered to the CPW Aquatic Animal Health Laboratory (AAHL) in Brush, Colorado for myxospore enumeration. Myxospores counts were ascertained using the pepsin-trypsin digest method (Markiw & Wolf, 1974b, 1974a). Following an enzymatic digest of the fish skulls, the solution was inserted into a centrifuge to separate the myxospores from other dissolved skeletal material. The concentrated myxospores solution was allocated to a gridded dish and grids were randomly selected for myxospore counts. AAHL staff counted the subsamples of myxospores under 400x magnification and extrapolated the counts to estimate the total myxospore loads of individual fish heads.

Statistical Analysis

To quantify the differences in whirling disease severity between the different Rainbow Trout strains in this system, we fit two different generalized linear models (GLM) in R (R Core Team, 2021). First, we used a binomial distributed GLM to evaluate the presence and absence of infection as a function of the four strains. Next, to compare the infection severity of individual fish from each strain, we compiled the myxospore counts from infected fish for each strain. We then fit a GLM of myxospore counts as a function of the four strains with a Poisson distribution to account for deviations from a normal distribution typical of count-based data. Both GLMs were assessed for overdispersion and were appropriately adjusted with quasi-likelihood if needed.

To statistically assess the difference in survival rates between strains of Rainbow Trout, we performed dead recovery models using Seber parameterization formulated in the package *Rmark* in R statistical software (Laake, 2013; R Core Team, 2021). Dead recovery models were used to account for the fact that all recaptured fish were dispatched for laboratory myxospore analysis and batch identification. We formatted capture histories by year, with the individual monthly captures summed together, to meet data entry requirements for models in MARK to use maximum-likelihood estimates for survival and capture probability (recovery rates). Multiple models were fit with logit link functions where survival varied as a function of the strains, intercept-only, time, age, and combinations of strain with time and age. All these models were fit with different groupings of the four strains to ascertain model level difference between the survival rates of the strains. Here, survival estimates are the probability of an individual surviving between time intervals, whereas recovery rates are estimates of an individual being caught at each time. Recovery rates were held at a constant rate for all four strains to reduce the

chances of overparameterization. We selected our best model using Akaike information criterion corrected for small sample sizes (AIC_c), with an Δ AIC_c cutoff of three. We extracted parameter estimates and their associated confidence intervals from the best model and compared effect sizes and model support to assess survival rates between the Rainbow Trout strains.

Results

Myxospore counts from captured fish varied substantially and significantly by strain (Figure 2). The probability of HAR being infected with M. cerebralis was roughly 38.5% (95% CI: 30.4% - 47.0%). Compared to HAR, the probability of HXH(50:50) infection was 30.3% lower (95% CI: 2.2% - 40.4%), while the probability of HXH(75:25) infections is 46.2% lower (95% CI: 39.6% - 48.8%). The GR strain effectively had zero probability of infection as there were no individuals recovered from this strain that possessed myxospores. The infection severity of captured Rainbow Trout was significantly lower for HXH(50:50) compared to HAR (p-value= 0.0275, from a TukeyHSD test), but was insignificant between HXH(75:25) and HAR and between HXH(75:25) and HXH(50:50) (p-values= 0.4847 and 0.9998, from a TukeyHSD test). On average, infected Rainbow Trout from the HAR, HXH(50:50), and HXH(75:25) strains had 88,441 (95% CI: 61,030-123,035), 28,336 (95% CI: 10,527-63,593), and 28,837 (95% CI: 1,707-132,047) myxospores per individual, respectively (Figure 2).

We found considerable differences between the survival rates of the five different strains. From the model of survival rates varying by strain alone, yearly estimated survival rates for GR was 0.24 (95% CI: 0.15-0.34), HAR was 0.48 (95% CI: 0.43-0.54), HXH(50:50) was 0.52 (95% CI: 0.46-0.57), and HXH(75:25) was 0.40 (95% CI: 0.32-0.49) (Figure 3). On average, the HXH(50:50) strain had the highest survival rate but had considerable overlap of 95% confidence intervals with the HAR and HXH(75:25) strains, with 66.1% and 28.2% of the 95% confidence

interval of HXH(50:50) overlapping with the confidence intervals of HAR and HXH(75:25) respectively. The GR strain had the lowest survival rate which was significantly different from the HAR and HXH(50:50) strains, but had a slight (12.8%) overlap of 95% confidence intervals with the HXH(75:25) strain (Figure 3). These survival rates were time and age dependent. Our strain survival analysis found the best model was one where survival rates were allowed to vary between only GR, HXH(75:25), and the other strains (HAR and HXH(50:50) combined), but this model's support was only modestly greater than a model with survival rates varying between all the strains (ΔAICc: 0.948, Table 1). From this best model, survival rates were predicted to decrease across all the strains, reaching nearly zero around age 3 (Figure 4). Recovery rates were estimated to be very low for this study at 0.0246 (95% CI: 0.0227-0.0267).

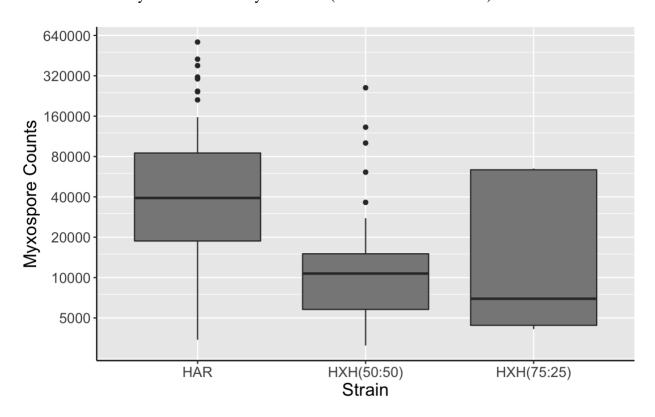


Figure 2: Boxplots display the severity of infection, based on myxospore counts, for the three strains of Rainbow Trout on the log scale. The GR strain is not displayed in this graph as none of the individual fish that were captured were infected.

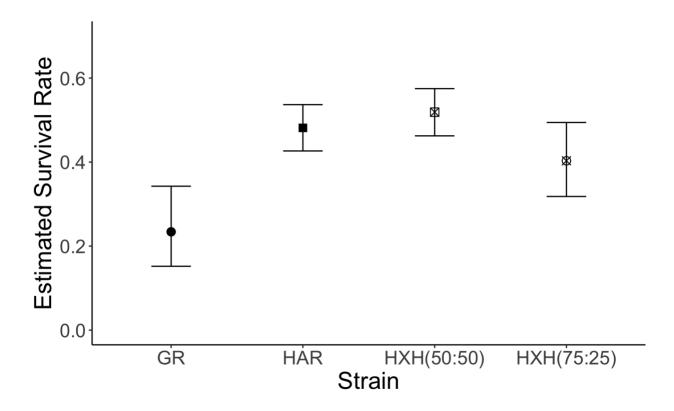
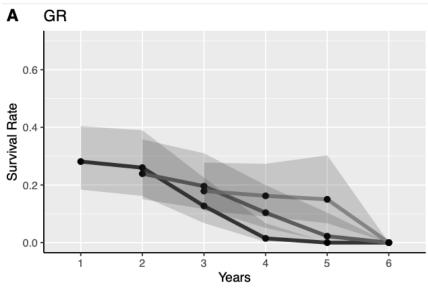
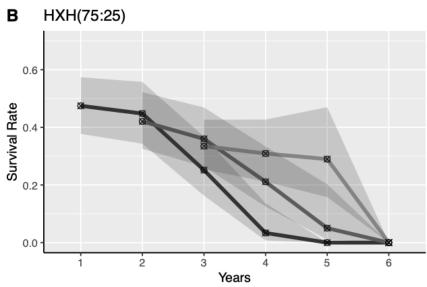


Figure 3: Compiled yearly estimated survival rates for the four strains of Rainbow Trout in Parvin Lake. Error bars represent 95% confidence intervals. Closed points represent pure strains while open points represent crosses and back-crosses of the two pure strains.

Table 1: Model selection results comparing survival rates varying with different strain groupings and additional age and time predictors. Model subsection notation for strain grouping variables in the survival rates designates the strains allowed to vary, with commas dividing groups.

Model	K	AICc	DeltaAICc	Weight
$S(\sim strain(GR,HXH(75:25),Other) + age + time)r(\sim 1)$	14	6747.1	0.000	0.434
$S(\sim strain(All) + age + time)r(\sim 1)$	15	6748.1	0.948	0.270
$S(\sim strain(GR, HXH(50:50), Other) + age + time)r(\sim 1)$	14	6750.5	3.424	0.078
$S(\sim strain(GR, HXH(75:25), Other) + age)r(\sim 1)$	9	6751.0	3.883	0.062
$S(\sim strain(All) + age)r(\sim 1)$	10	6751.7	4.595	0.044
$S(\sim strain(GR + HXH(75:25), Other) + age + time)r(\sim 1)$	13	6752.5	5.374	0.030
$S(\sim strain(GR,Other) + age + time)r(\sim 1)$	13	6752.5	5.374	0.030
$S(\sim strain(HAR,HXH(50:50),Other) + age + time)r(\sim 1)$	14	6753.7	6.614	0.016
$S(\sim strain(HAR,GR,Other) + age + time)r(\sim 1)$	14	6754.5	7.362	0.011
$S(\sim strain(GR, HXH(50:50), Other) + age)r(\sim 1)$	9	6754.8	7.665	0.009
$S(\sim strain(HAR,HXH(50:50),Other) + age)r(\sim 1)$	9	6756.7	9.613	0.004
$S(\sim strain(GR, HXH(75:25), Other) + time)r(\sim 1)$	9	6757.1	10.028	0.003
$S(\sim strain(GR+HXH(75:25),Other) + age)r(\sim 1)$	8	6757.5	10.362	0.002
$S(\sim strain(GR,Other) + age)r(\sim 1)$	8	6757.5	10.362	0.002
$S(\sim strain(All) + time)r(\sim 1)$	10	6758.5	11.372	0.001
$S(\sim strain(HAR,GR,Other) + age)r(\sim 1)$	9	6759.5	12.357	0.001
$S(\sim strain(GR, HXH(50:50), Other) + time)r(\sim 1)$	9	6760.2	13.101	0.001
$S(\sim strain(GR+HXH(75:25),Other) + time)r(\sim 1)$	8	6761.1	13.932	0.000
$S(\sim strain(GR,Other) + time)r(\sim 1)$	8	6761.1	13.932	0.000
$S(\sim strain(HAR,GR,Other) + time)r(\sim 1)$	9	6763.0	15.901	0.000
$S(\sim train(HAR,HXH(50:50),Other) + time)r(\sim 1)$	9	6764.3	17.217	0.000
$S(\sim time + age)r(\sim 1)$	12	6774.2	27.052	0.000
$S(\sim strain(HAR,Other) + age + time)r(\sim 1)$	13	6774.7	27.530	0.000
$S(\sim age)r(\sim 1)$	7	6778.8	31.647	0.000
$S(\sim strain(HAR,Other) + age)r(\sim 1)$	8	6779.4	32.262	0.000
$S(\sim time)r(\sim 1)$	7	6781.6	34.524	0.000
$S(\sim train(HAR,Other) + time)r(\sim 1)$	8	6782.1	34.956	0.000
$S(\sim strain(All))r(\sim 1)$	5	6782.5	35.378	0.000
$S(\sim strain(GR,HXH(50:50),Other))r(\sim 1)$	4	6782.5	35.405	0.000
$S(\sim strain(GR+HXH(75:25),Other))r(\sim 1)$	3	6782.8	35.695	0.000
$S(\sim strain(GR,Other))r(\sim 1)$	3	6782.8	35.695	0.000
$S(\sim strain(HAR,GR,Other))r(\sim 1)$	4	6784.8	37.637	0.000
$S(\sim strain(HAR,HXH(50:50),Other))r(\sim 1)$	4	6786.4	39.245	0.000
$S(\sim 1)r(\sim 1)$	2	6796.1	48.974	0.000
$S(\sim strain(HAR,Other))r(\sim 1)$	3	6797.8	50.699	0.000
$S(_{\text{strain}(GR,HXH(75:25),Other)})_{r}(1)$	4	6807.1	59.957	0.000





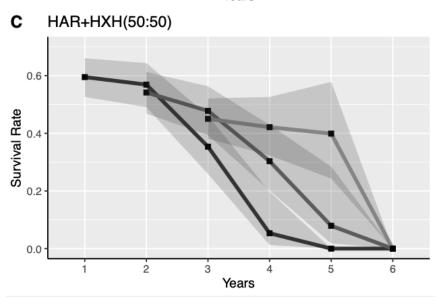


Figure 4: Estimated survival rates between sampling years for each strain of Rainbow Trout extracted from our Rmark best model (Table 1). HAR and HXH(50:50) strains were combined in our best model, so they are visualized together as one grouping in this figure. Shaded regions represent the 95% confidence intervals for the survival estimates. Each set of lines represents one of the three age-classes of fish stocked into Parvin Lake, with dark, dark-gray, and gray shades representing the 2007, 2008, 2009 cohorts, respectively.

Discussion

The goal of this study was to quantify the survival rates and *M. cerebralis* infection severities of different strains of Rainbow Trout to aid fisheries management decisions. We document pronounced differences in the survival and myxospore counts of different strains of whirling disease resistant Rainbow Trout in a natural lake setting. As we predicted, the GR strain had the lowest survival rates of the four strains evaluated in this study, with an average survival rate of 24%, but had zero cases of M. cerebralis infection. Contrary to what we expected, the first generation cross of GR and HAR produced the strain with the highest average survival rates, but this effect lacked support in our overall best model and had considerable overlap of 95% confidence intervals. Myxospore analysis of the four strains revealed, as predicted, decreases in the probability of M. cerebralis infection with increasing genetic inputs for the GR strain, but the HXH(75:25) strain had high variability in infection severity contributing to insignificant comparative results to the HAR and HXH(50:50) strains. This could be partially due to the low sample size of HXH(75:25) infected individuals (n=5) compared to the HXH(50:50) (n=29) and HAR (n=50) strains, but other studies by CPW have documented similar variability in the infection severity of these crossed populations, and therefore the variability likely reflects real trends (Schisler & Fetherman, 2011).

The average lower survival rates of the HAR strain compared to the HXH(50:50) observed in our study may be explained by numerous factors. For example, the modestly higher survival rates of the crosses may indicate hybrid vigor, but this attribute among trout strains is

inconsistent in the literature. Hybrid strain populations of rainbow trout and other salmonids have largely been suggested to result in outbreeding depression (L. M. Miller et al., 2004; Negus, 1999; Ostberg et al., 2004; Tymchuk et al., 2007), but F1 hybrids of rainbow trout with Yellowstone Cutthroat Trout have been suggested to confer fitness advantages (Campbell et al., 2002). While our finding of higher F1 cross average survival rates was not significant compared to HAR, the F1 cross survival was significantly greater than the GR strain, suggesting a relatively higher fitness. Additionally, these differences may reflect higher whirling disease related mortality in the HAR strain. Myxospore counts from the recapture Rainbow Trout reveal some extreme cases of *M. cerebralis* infection, with counts ranging from 0 to 571,305 spores per fish. Survival of salmonids infected with M. cerebralis is dependent on the severity of infection (myxospore concentration) as well as the age and size of the fish (MacConnell & Vincent, 2002; Ryce et al., 2005). The higher probability and severity of infection in the HAR strain may have contributed to a greater proportion of whirling disease related mortalities compared to the HXH(50:50) strain. Lastly, Negus (1999) found intermediate wariness of hybrid fry between the parental steelhead and Kamloops Rainbow Trout. These intermediate behaviors may have simply best suited the habitat at Parvin Lake, blending the deep-water specialties of the HAR strain with the more surface-orientation and lower flight response of the GR strain.

Combating whirling disease in salmonid populations is currently a balancing act of two desired traits: whirling disease resistance and moderate survival rates. Domestication is well known to have adverse effects on survival of salmonids (Johnsson et al., 1996; Johnsson & Abrahams, 1991; Skaala et al., 2019), but these effects are not always as pronounced and can be dependent on the environments of study (Vandersteen et al., 2012). While our findings of the reduced survival rates of the domesticated GR Rainbow Trout is not very surprising, studies by

Colorado Park and Wildlife biologists found comparable swimming abilities and survival rates between GR and F1 hybrids of GR crossed with the Colorado River Rainbow Trout (GR x CRR) (Avila et al., 2018; Fetherman et al., 2011). We attribute these differences in findings to differences in the habitat, variation in wild strains used for the crosses, and the scale of predation. Notably, GR Rainbows may survive better in environments where predation isn't the main source of mortality. Additionally, one of the limitations of these studies was the inability to assess the survival and genetic contributions to multiple generations. Even though domestication would theoretically continue to negatively affect future populations and be selected against, Fetherman et al. (2014) showed that the first introductions of GR x CRR to the Upper Colorado River contributed to 80% of fry populations of future generations based on genetic surveys, despite low survival rates. With whirling disease predominantly impacting younger salmonids (<6 months age) (Ryce et al., 2005), mortality of future progeny may dictate the need for whirling disease resistance over wild genetic heritage. Depending on the management objectives, stocking strains with low survival rates but high whirling disease resistance may be desirable.

Our study quantified survival of four strains of Rainbow Trout, two parental strains and two crossed strains, over one generation in one shallow mountain lake. Future studies should expand to look at additional strategic crosses with GR Rainbows and assess survival rate and myxospore counts in different lentic and lotic habitats. Whirling disease is known to have habitat specific interactions which causes spore loads to vary over spatial and temporal scales (Halliday, 1976; Hiner & Moffitt, 2002). Particularly, the presence of the second host of *M. cerebralis* (*Tubifex tubifex*) and increasing water temperature are positively correlated with infection severity (Hiner & Moffitt, 2002). Minute differences between lakes in elevation and substrate could impact *M. cerebralis* presence and abundance, with this potential variation not captured in

this study. Furthermore, Schisler et al., (2006) found whirling disease resistance remained high in crosses between GR and CRR strains at intermediate degrees between the parental strains. Our study showed similar trends with the probability and severity of infection used as a gauge of whirling disease resistance with a different wild parental strain (HAR). Follow up studies should add additional crosses with other wild strains that are known to have higher survival rates as well as desired traits and particular habitat adaptations for the intended stocking area. One potential wild population to investigate is the Snake River Cutthroat Trout *Oncorhynchus clarkii behnkei*. Recent studies have shed light on the increased survival rates and overall success of the Snake River Cutthroat Trout and their hybrids (R. P. Hedrick et al., 1999; Sipher & Bergersen, 2005; Thompson et al., 1999). Combined with the lower *M. cerebralis* infections of these cutthroat trout compared to rainbow trout (R. P. Hedrick et al., 1999; Sipher & Bergersen, 2005; Thompson et al., 1999), the Snake River Cutthroat Trout is a promising wild strain for resistant strain crosses.

M. cerebralis infection and subsequent whirling disease have dramatically reduced populations of Rainbow Trout in the United States (Nehring & Thompson, 2001). Controlling the supply of myxospores and TAMs from suitable hosts is a promising avenue for minimizing M. cerebralis populations and thus minimizing occurrences of whirling disease (Wagner, 2002). Our HXH(50:50) crosses had comparable survival rates to the parental wild strain (HAR) but with nearly two thirds the probability of infection and four times lower infection severity. Therefore, the use of F1 hybrids as fingerling plants into lakes where whirling disease is enzootic may be effective. Stocking HXH(50:50) into lakes has several advantages. First, the high survival rates would ensure efficient returns to creel, maximizing management and financial efforts (Cassinelli & Meyer, 2018). Secondly, the presence and death of these fish would release

fewer myxospores to the environment (due to their whirling disease resistance), decreasing the spore burdens of future populations of all salmonids in the watershed. Lastly, reproduction of the HXH(50:50) strain with Rainbow Trout present in the system may pass on the whirling disease resistance to the next generation and help natural recruitment and sustainability of these highly managed fisheries (Fetherman et al., 2014). In certain situations, though, when the priority is whirling disease resistance, the tradeoffs in survival rates of HXH(75:25) strains and even the GR strain for the higher and nearly complete whirling disease resistance would be justified and appropriate.

Our study provides clear evidence that crosses of the GR strain have higher survival rates close to or matching the wild HAR strain and have moderately high whirling disease resistance. Both the HXH(50:50) and HXH(75:25) strains had higher survival rates than the domesticated GR strain, but lower whirling disease resistance. Therefore, the choice of which of these strains to stock should be context and management specific. Given the significance of Rainbow Trout as a sport fish, using these whirling disease resistant strains can help to recover, maintain, and even buffer the impacts of whirling disease on wild populations of Rainbow trout. Additionally, combating whirling disease is a problem facing many salmonids other than just Rainbow Trout, including Brook Trout, various subspecies of Cutthroat Trout, and Kokanee Salmon Oncorhynchus nerka (Vincent, 2002). Where other salmonids are sympatric with Rainbow Trout, managing the Rainbow Trout populations for whirling disease resistance could mutually benefit future generations of both species by reducing the spread of whirling disease, decreasing spore burdens and related infection severity, and therefore minimizing whirling disease related mortality. It is important, though, to weigh the costs and benefits of stocking domesticated and genetically limited fish like Rainbow Trout from the GR strain, especially when dealing with

other wild and native salmonid populations. Research should continue to investigate other crosses between GR and other wild populations to try and best blend locally adapted wild genotypes with whirling disease resistance for put-and-take fisheries.

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CHAPTER 4.

A Compromise for the Planned Reservoir Expansion at Bear Creek Lake Park to

Combat Denver's Water Crisis

Introduction

The use and storage of water has sparked controversial and emotional debates dating back to the environmental movements of the 1970s (Biswas, 2012; Davitt, 2011; Tyler, 2017). Water storage is particularly prevalent in western states, such as Colorado, where the highly variable and semi-arid nature of the climates in this region result in seasons with limited water supplies and severe droughts (Davitt, 2011; Kang & Ramírez, 2007). With populations expanding in Denver, Colorado, and climate change a looming threat, the water in the South Platte watershed is becoming an increasingly scarce commodity. To increase water security in the South Platte watershed, Colorado Water Conservation Board (CWCB) in collaboration with the United States Army Corps of Engineers (USACE) have proposed an increase in water allocation to Bear Creek Reservoir from 2,000 acre feet to 22,000 acre feet (AF)(City of Lakewood, 2022). Unfortunately for local residents and the 652,389 annual park visitors, this plan of action would inundate much of the park's trails and adjacent wildlife habitat, forever changing the park's services (Hutchison, 2017). There is not a comprehensive solution that can satisfy all stakeholders for this issue. To maintain much of the recreational value of Bear Creek Lake Park (BCLP) and add additional water storage and security for Denver and downstream agricultural communities, BCLP should be expanded to 12,000 AF accompanied by deliberate and timely habitat enhancement and trail construction. In addition, CWCB should follow through and expand their goal outlined in the Colorado Water Plan of increasing community-level water

conservation by establishing water-use limits and imposing fines that match the scale of Colorado's climate challenges. Combining the increase in water allocation with more efficient water-use should address the water demand of future population increases in the greater Denver area.

Background

Denver's Water Crisis and Population Increases

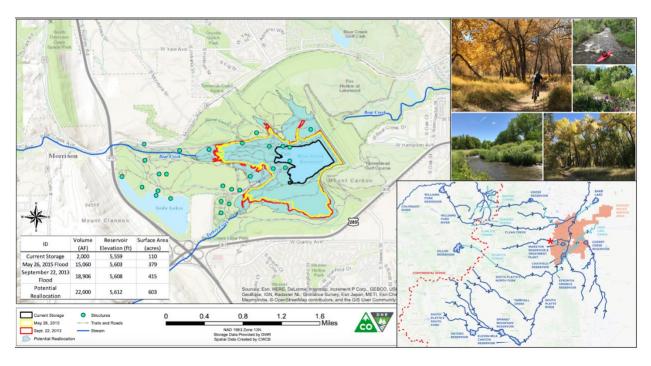
In Colorado, populations are growing across the front range from Fort Collins to Colorado Springs (Minor et al., 2021; Tyler, 2017). The city of Denver is estimated as of 2019 to hold nearly 727,211 residents, which is up from the 2010 census that estimated the population at 600,158 residents (US Census Bureau, 2021). With populations in these areas continuing to grow, Colorado's natural resources, most notably water, are coming under increasing pressure. Recent climate scenarios for the South Platte watershed, in the worst-case climate change scenario, predict decreases in surface runoff of 20%, groundwater recharge of 35%, and groundwater discharge of 9% (Aliyari et al., 2021). In 2015, the CWCB created the Colorado Water Plan with several goals including increasing total water storage to 400,000 AF and increasing sustainable community water-use practices (CWCB 2022). To meet these water storage goals, CWCB is investigating options to expand current reservoirs in the Denver region including the Tri-Lakes in the South Platte Watershed: Chatfield, Cherry Creek, and Bear Creek Reservoirs (Chatfield Storage Reallocation Project, 2021; USACE, n.d.). Out of the three main reservoirs that provide water to the South Platte River flowing through Denver, Bear Creek Reservoir is an obvious choice for expansion as the dam itself is already capable of holding the additional water and the terrain is ideal to maximize storage with less lake surface area, minimizing evaporative losses.

Bear Creek Lake State Park

Originally, USACE created Bear Creek Reservoir in 1968 in Jefferson County for flood control, but the lake and park has since gained popularity as a hotspot for recreational activities. Between 2011 and 2020, BCLP has steadily increased in popularity, gaining 245,295 visitors in that timeline. In 2020, there were an estimated 652,389 park visitations which City of Lakewood supervisors believe is still an underestimate despite the fact that the estimate includes recorded vehicular visitations and an additional percentage added for walk/ride-in access to the park from alongside Morrison Road and Fox Hollow Golf Course (Katie Gill, pers. comm., March 14, 2022). These visitors enjoy the plethora of recreational activities in the area including horseback riding, fishing, biking, hiking, birding, and more, all of which are central to the popularity and significance of this state park in the greater Denver metro area (City of Lakewood, 2022). CWCB and USACE Proposal

The CWCB, with USACE approval, proposed expanding water allocation at Bear Creek Reservoir. The reservoir itself is supplied by two fairly small creeks, Turkey Creek and Bear Creek, whose headwaters are situated in the Mount Evans Wilderness Area. Outflow from the reservoir supplies the lower Bear Creek, which is a tributary of the South Platte River that flows through Denver, CO (Grundman et al., 2012). Current water storage is at 2,000 AF, with the proposal set to increase storage to 22,000 AF (CWCB, 2021). This additional water will cover 615 acres of land and eliminate much of the park's terrestrial habitats, recreational trails, roads, parking lots, and picnic shelters (Figure 1). Water rights to the increased water supplies would be allocated to the city of Brighton (6,600AF), Evergreen (100AF), Berthoud (3,000 AF), Dacono Municipality (3,000), Foothills Parks and Recreation (65AF), Hidden Valley Water District (50AF), and the environment (4,550AF) (CWCB, 2021). The proposal has come under scrutiny

from local communities concerned with losing large parts of the park. Numerous activities in the park will be forever changed or eliminated. Some of the main draws to the park are the scenic network of trails around the lake and rivers. Additionally, recreationalist and local communities are concerned about the potential seasonal fluctuation in reservoir water levels, which is generally lacking in aesthetic and ecological appeal (Hutchison, 2017). Collectively, affected communities have formed the "Save Bear Creek Lake Park" campaign in an attempt to stop the proposed reservoir expansion.



<u>Figure 1:</u> Map of BCLP, with trails and major structures, comparing current 2,000 AF water levels to both the floods in 2015 and 2013 and the USACE and CWCB proposed expansion (Hutchison, 2017). On the right are pictures of recreational sites that would be lost and a map of the South Platte Watershed supplying the Denver Metropolitan Area with water (Beaty, 2020; Save Bear Creek Lake Park, 2021). The Red Star denotes the location of BCLP.

Stakeholders

USACE, CWCB, and the City of Lakewood

These three government organizations are working together to access water allocation at Bear Creek Reservoir and are the main advocates for the expansion. CWCB is in charge of

monitoring and managing the water resources in the state of Colorado. Following the creation of the Colorado Water Plan, CWCB explored the option of expanding Bear Creek reservoir (CWCB, 2022). The USACE owns most of the land in BCLP and is the regulatory authority over the reservoir including any changes made to the water discharge regimen (City of Lakewood, 2022). Under section 404 of the Clean Water Act, USACE is the regulatory authority for making jurisdictional determinations for waters of the United States (USACE, 2007). Together, these two organizations are allotted \$2,500,000 from the state of Colorado to conduct the study of the Bear Creek Expansion and to write the Environmental Impact Statement starting in 2015 (City of Lakewood, 2022). The first public scoping meeting was held on October 14, 2021. The City of Lakewood, which owns the Soda Lakes and oversees the recreational side of BCLP, supports the proposed expansion despite the potential short-term loss of park visitations.

Golf Courses

Jefferson County uses substantial amounts of water (>5,000 AF) for irrigation of the 23 golf courses in the county (Ivahnenko, 2009). There are three golf courses in the vicinity of the park that obtain substantial amounts of water from Bear Creek: Foothills, Homestead, and Bear Creek Golf Courses. People associated with these golf courses value the economic and recreational benefits of golf, which requires golf courses with watered and maintained grass. Maintenance of these golf courses uses substantial amounts of water throughout the growing season and would benefit from greater water security in the region, but it is unclear if these courses would receive any additional water allocation from the reservoir.

Recreationalists and Local communities

The loudest voices come from area residents who oppose the change for many reasons, including the desire to maintain recreational opportunities and preserve the beauty and sanctity

of the park. In particular, Katie Gill founded Save Bear Creek Lake Park to try and raise awareness of and opposition to the reservoir expansion (Save Bear Creek Lake Park, 2021). The flooding of the lake would submerge many trails and terrestrial habitats that Katie believes are "the heart and lungs of the park" (Nicholson, 2021). Losing these sections would mean losing the value of the park to these stakeholders. Cyclists, birders, horseback riders, and hikers all enjoy the trails in the park, principally the trails by Turkey and Bear Creek that go through lush cottonwood forests. These semi-natural riparian habitats are home to diverse populations of mammals, birds, reptiles, and amphibians and hold cultural and sentimental value to local communities (Grundman et al., 2012). These groups value nature and the high-quality, local recreational opportunities that are accessible close to home. The ideal solution for this broad stakeholder depends on the individuals' vested interest in the park, but general preference is to avoid the reservoir expansion. It is important to note that these stakeholders do value water security either directly or indirectly and how it relates to quality of life.

Fishermen and boaters represent a unique subset of recreationalist that could benefit from the expansion in the long term. Logically, aquatic activities would expand at the park. Boats would have more space to spread out on the lake and explore but would temporarily lose access to boat ramps and parking lots during the expansion phase. Additionally, the seasonal water extraction from Bear Creek Reservoir is expected to cause larger vertical water fluctuation (Hutchison, 2017). In times of low precipitation, barren soil would be exposed all around the lake which would compromise some of the aesthetics of the lake and terrestrial environments (Save Bear Creek Lake Park, 2021). Furthermore, fish would have additional cover and space to spread out into, but the Lake overall would be able to hold greater numbers of fish in the future. Increased lake cover would come at the expense of decreased river habitat, which would be a

concern for fishermen (particularly fly fishermen) who prefer fishing in the creek for the wild brown trout populations. Overall, many regular fishermen of the park would likely resent the loss of known fishing locations and the inconsistent water levels with compromised aesthetics, a view they share with boaters, other recreationalist, and residents in the area.

Recommendation

There is no one solution to satisfy all the stakeholders, meaning a compromise is needed. Even though most climate scenarios predict increases or no change in average future precipitation in the South Platte watershed, worst case scenarios predict decreases that would have drastic impacts on agriculture and metropolitan areas like Denver (Aliyari et al., 2021). Colorado's hydrology has drastic fluctuations in precipitation and flow, which makes water storage essential (Kang & Ramírez, 2007). To best accommodate the views and values of all the current and future stakeholders, we should expand the reservoir with a few contingencies. To start, Bear Creek Reservoir should be expanded to only 12000 AF as this water level retains several trails, structures, and stream riparian habitat while still adding 10,000 AF to the lake as a whole to meet some of CWCB's water storage goals. Some notable locations saved by this restricted increase are the Turtle Pond Fishing Area, Muskrat Meadows picnic and parking area, 3.79 miles of trails, and 224 acres of terrestrial habitat (City of Lakewood, n.d.). Many of these locations would be lost or compromised with any further increases in water allocation.

While an increase of 10,000 AF is substantial (500% increase), it is still half of the water area that CWCB proposed. The 20,000 AF goal set by CWCB appears to be the maximum amount of water that BCLP could hold without risking the original intent of the reservoir for flood mitigation (CWCB, 2021). Of the 20,000 AF proposed, only 16,850 AF of the expansion was broken down as to who would have the rights to that water (CWCB, 2021). It is not clear

exactly why the CWCB allocated the water to different cities in the proportions that they did, but they do state that these decisions were made secondarily to the proposed 20,000 AF increase and upon asking the stakeholders essentially how much water they would want. Both the amount of water allocated in water rights and the 20,000 AF proposed expansion are likely overestimates design to sway public opinion to higher water allocation compromise. The CWCB openly stated in the Colorado Water Plan that the goal of obtaining 500,000 AF storage across Colorado was an overshot of the 400,000 AF that they say they needed for water security (CWCB 2022). Reducing the proposal to 10,000 AF seemingly would be a matter of reworking the numbers on water right allocation. This increase coupled with better water-use practices should be adequate to meet future demands in low water years/seasons while also maintaining some of the recreational value of BCLP.

To mitigate for habitat and recreational losses, the park should develop more aesthetically appealing paths around the new inundation zone with revegetation and reseeding. Native seed collections should be conducted in the current riparian habitat and stored for later enhancement of on bank environments. Some of these plantings should begin as soon as the decision to fill the lake is made to ensure a smooth transition to the newest water level. For instance, planting shrubs and larger trees like cottonwoods around the future water line. Additionally, park picnic tables and shelters in the flood zones need to be removed and incorporated into the landscape at higher elevations outside of the inundation zone. To help maintain reservoir fishing quality, additional fish should be stocked each year into the reservoir during the time needed to fill the lake through collaborations with Colorado Parks and Wildlife to keep up with the increasing water volume. Finally, BCLP should continue to fulfill the educational purposes in the park concerning wildlife and water conservation. Bear Creek

Reservoir has several outreach programs, which benefit from these natural areas. Placing educational signs and plaques around the park to explain the history of the park and the water issues facing Denver with future population growth would achieve many of BCLP education goals and benefit all involved parties. By taking these steps, BCLP will be able to reach a more natural, aesthetically pleasing, recreationally satisfying state in the least amount of time and continue to have positive impacts on communities in the area.

Increasing water storage levels at Bear Creek Reservoir would only provide water security for low water years and seasons, but water usage would continue to be limited by inputs of water from precipitation. Therefore, CWCB in collaboration with Denver Water should reach out to homeowner associations and local businesses to start developing better landscaping measures for water conservation and better water allocation, as discussed in the Colorado Water Plan in 2015. Of particular interest would be working with the local golf courses to update irrigation practices and ensure the best possible management of water resources considering the abundance and importance of golf courses in Jefferson County. Finally, the city of Lakewood and greater Denver Metropolitan Area should explore water budgets and fines for exceedances, particularly for low water times of the year such as late summer and fall. Only restrictions in water-use combined with water storage for droughts will provide Colorado with the water security needed to sustain a strong economy and high quality of life.

Conclusion

To ensure a more rapid transition of recreational and natural areas while maintaining healthy flows for the lower Bear Creek River and greater Denver South Platte river, USACE should stick to its plan to achieve the 12,000 AF volume in five years or more depending on the climatic conditions in those years. At this water level, some valuable recreational areas would be

preserved, and additional restoration should eventually satisfy the recreationalists that love this park, while still providing water security for drought years. To the degree possible, this proposed solution would address the majority of the concerns of major stakeholders for this environmental issue. This particular stakeholder analysis focused solely on the merits of a reservoir expansion at Bear Creek reservoir and not the additional expansion proposed around the state such as at Chatfield reservoir and Gross reservoir. The cumulative impacts of all these expansions on the local recreational activities may deserve further consideration. Additionally, this paper abstracted the views of the major stakeholders, but there is undoubtedly some variation between individuals' opinions and views within these broad stakeholder categories. Overall, expansion of Bear Creek Reservoir has not and will continue to not be a popular choice among the stakeholders that use and treasure this park, but mitigating for some of the impacts would help expedite the transition to a future with different quality recreational opportunities and increase water storage for times of need in the greater Denver Metropolitan Area.

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