THREE-SPECIES RESEARCH PROJECT SUMMARY

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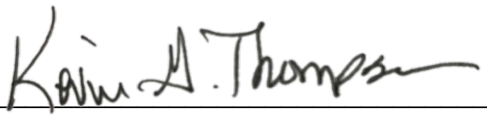
Prepared by: 

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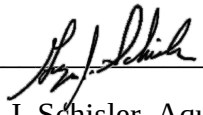
Approved by: 

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Date: February 25, 2020

The results of the research investigations contained in this report represent work of the authors and may or may not have been implemented as Colorado Parks & Wildlife policy by the Director or the Wildlife Commission.

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Three-species tributary occupancy and spawning investigations

Period Covered: March 1, 2019 to December 31, 2019

PROJECT OBJECTIVE: To determine the current distribution of Bluehead *Catostomus discobolus* and Flannelmouth *C. latipinnis* sucker and Roundtail Chub *Gila robusta* in tributary streams of Colorado's mainstem, western rivers (Green, Yampa, White, Colorado, Gunnison, Dolores, and San Juan) and to evaluate spawning ecology of the fishes and limit hybridization of the two sucker species with introduced nonnative suckers.

RESEARCH PRIORITY:

Identify the current distribution of three-species fishes in tributary streams of western Colorado Rivers.

OBJECTIVES:

I) Identify occupancy of tributaries in relation to historically occupied sites and randomly selected sites where occupancy was historically unknown.

II) Correlate occupancy and detectability to stream and survey characteristics.

INTRODUCTION

Bluehead Sucker *Catostomus discobolus* (BHS), Flannelmouth Sucker *C. latipinnis* (FMS), and Roundtail Chub *Gila robusta* (RTC) are collectively known as the "three-species" under conservation regimes shared by numerous western States that contain habitats occupied by these fishes. All three are native to the Colorado River basin, and FMS and RTC are endemic to the basin while BHS also occupy portions of the Snake and Bear River drainages to the north of the Colorado River Basin. All three species have experienced significant population reductions, and have been extirpated from many habitats.

Much of the available data and literature on the distribution of the three-species focuses on large rivers and major tributaries – habitat that is indeed important to a substantial component of these species' populations. However, small tributaries and even intermittent waters can be important for all three species annually, and some populations are even restricted to small tributaries. Therefore, to truly understand the three-species distribution in Colorado and to spatially and temporally implement conservation efforts with the most effect, it is necessary to identify the distribution and changes in the distribution of these fishes in these tributary habitats. Associating general physical parameters with presence and detectability can identify conditions under which these species persist, and can inform future sampling programs.

METHODS

This study estimated Bluehead Sucker, Flannelmouth Sucker, and Roundtail Chub occupancy of tributary streams throughout their range in Western Colorado. Randomly selected, spatially balanced waters as well as historically occupied locations were sampled using standardized methods to determine overall occupancy as well as change in occupancy over time at historically occupied locations. Occupancy and detection probabilities were modeled along with covariates such as stream gradient and conductivity to inform future sampling programs. Detailed methodology may be found in the publication referenced below.

RESULTS AND DISCUSSION

This Research Priority is complete. A technical report including this project was completed and published in September 2019.

- Thompson, K. G., and Z. E. Hooley-Underwood. 2019. Present Distribution of Three Colorado River Basin Native Non-game Fishes, and Their Use of Tributary Streams. Colorado Parks and Wildlife Technical Publication 52.

In brief, Bluehead Sucker were estimated to occupy 62.6% of historic sites and 23.1% of randomly selected sites within suitable parameter ranges. Flannelmouth Sucker were estimated to occupy 37.1% of historic sites and were rarely found at randomly selected sites. Gradient affected Flannelmouth Sucker occupancy, with this species being much more likely to occupy sites of very gentle slope. Roundtail Chub were estimated to occupy 43.9% of species-specific historic sites when modeling on the average values of the covariates gradient and ordinal day of sampling. Like Flannelmouth Sucker, Roundtail Chub were more likely to be found occupying historic sites of low gradient. They were also more likely to be found occupying sites at sampling dates later in the calendar year. We found that surveys consisting of 2-pass electrofishing efforts over 500 feet or more of stream resulted in probabilities of detecting these species, given their presence at the site, of 0.95 or greater. Thus, 2-pass electrofishing over a suitable reach of stream carries a high probability of revealing whether any three-species fishes are present. No formal occupancy analyses of the 2015 –2017 data (years with repeated visits to some sites) have yet been conducted, but occupancy by the three-species fishes was high over the 126 occasions represented, which included multiple visits per year at some sites. One or more of the study species were detected on 95% of sampling occasions in 2015, 89.5% of occasions in 2016, and 90.2% of occasions in 2017. Sampling occasions conducted during summer or fall months were likely to reveal three-species occupancy by young-of-year or juvenile fishes rather than adults, which in many tributaries are only present during spring spawning season.

ACKNOWLEDGMENTS

We thank Jenn Logan (CPW), Paul Jones (CPW), Tom Fresques (BLM), Dan Kowalski (CPW), Eric Gardunio (CPW), Kendall Bakich (CPW), Lori Martin (CPW), Jim White (CPW), Cory Noble (CPW), Ben Zimmerman (Southern Ute Tribe), Colin Larrick (Ute Mountain Ute Tribe), Jamie Ashmore (Ute Mountain Ute Tribe) as well as Summer Stevens (CPW), Nick Salinas (CPW), and numerous additional seasonal technicians for sampling assistance. Grant Wilcox

(CPW), Andrew Treble (CPW), Kristin Broms (CSU Coop), Ryan Fitzpatrick (CPW), and Jon Runge (CPW) provided advice and assistance with sampling design, data analysis, and statistics.

RESEARCH PRIORITY:

Evaluate the feasibility of a picket weir to exclude non-native suckers from a spawning tributary thereby decreasing hybridization of suckers.

OBJECTIVES:

- **I)** Install and operate a picket weir and fish trap in Cottonwood Creek to exclude non-native suckers from the Creek.
- **II)** Determine degree of hybridization of larval suckers produced in Cottonwood Creek to test if weir operations resulted in less hybridized progeny.

INTRODUCTION

Causes of declines of the three-species in both mainstem and tributary habitats include habitat loss, hybridization with non-native suckers, habitat fragmentation, and predation by or competition with non-native fishes. While habitat loss and fragmentation have certainly affected the three-species in Colorado via dams, diversions and dewatering, perhaps the most pressing threat to the two sucker species and their current distribution is hybridization with non-native suckers. White *C. commersonii* (WHS) and Longnose *C. catostomus* (LGS) suckers were introduced at numerous locations in the Colorado River basin many times over the 20th century. Both species hybridize with native suckers, producing viable offspring. White Sucker have spread much farther throughout the basin, and have become much more numerous than Longnose Sucker. As a result, hybrids between both native suckers and White Sucker have become common, and in some locations prevalent, in much of Colorado's three-species habitat. For instance, nonnative and hybrid suckers out-number native suckers in the Yampa River. One of the limited number of ways available to limit hybridization of these fishes is to limit the interactions of native and non-native sucker during spawning season. We attempted to limit non-native sucker access to an intermittent creek using a picket weir and fish trap.

METHODS

Cottonwood Creek is an intermittent tributary to Roubideau Creek (itself a tributary to the Gunnison River) that is heavily used for spawning by the three-species fishes as well as non-native White and Longnose suckers. In 2015, 2016, and 2017 we installed a picket weir and fish traps in the creek to capture all immigrant and emigrant fish. We attempted to selectively pass only native fishes. Because the creek was entirely intermittent, we were certain that resident non-native suckers were not present and therefore, the only fish above our weir in the creek would be native suckers that we moved from the trap to the upstream side of the weir. We routinely sampled another tributary to Roubideau Creek, Potter Creek, which served as an unmanipulated "control." To determine the effectiveness of our non-native sucker blockade, we collected larval sucker from Cottonwood and Potter creeks after each spawning season, and used genetic analysis

to determine the proportion that were hybridized with White or Longnose suckers. Detailed methodology may be found in the publication referenced below.

RESULTS AND DISCUSSION

This Research Priority is complete. During 2019, a manuscript on native sucker use of Cottonwood Creek was published, a technical report including this project was completed and published, and a manuscript on the effect of non-native exclusion on larval genetics was submitted and accepted for publication.

- Hooley-Underwood, Z. E., S. B. Stevens, N. R. Salinas, and K. G. Thompson. 2019. An intermittent stream supports extensive spawning of large river native fishes. Transactions of the American Fisheries Society 148:426-441.

- Thompson, K. G., and Z. E. Hooley-Underwood. 2019. Present Distribution of Three Colorado River Basin Native Non-game Fishes, and Their Use of Tributary Streams. Colorado Parks and Wildlife Technical Publication 52.

- Schwemm, M. R., K. G. Thompson, E. W. Carson, M. J. Osborne, and T. F. Turner. 2020. Species composition and hybridization among native and nonnative catostomid fishes in two streams of the Gunnison River basin, Colorado. Western North American Naturalist 80(1):81-89.

A brief summary of the results follows:

In no study year were we entirely successful in controlling the spawning run. Primarily, failure of this objective was due to our inability to keep the picket weir fence clear of debris when runoff began in earnest. Thus, we were unable to demonstrate that controlling a spawning run resulted in the production of a greater proportion of genetically pure native sucker larvae. Moreover, oftentimes the genetic results of larval fish identification didn't meet our expectations that the larval fish population would generally reflect the adult spawning fish population composition. This was especially so in Cottonwood Creek where more species were encountered. Despite our efforts to remove White Sucker, a majority of the larvae were identified as White Sucker or hybrids in 2016. We were more successful at maintaining closure in 2017, but White Sucker and hybrid larvae were still plentiful in samples. In Potter Creek, very high proportions of both the spawning sucker population and the resulting larval sucker population were dominated by native species, a situation which appears to be a general phenomenon within the Roubideau Creek tributary network. Those parts of the basin closest to the Gunnison River are most prone to host non-native suckers and their hybrids, while stream segments further away from the Gunnison River are found to be used predominantly by native suckers.

ACKNOWLEDGMENTS
We would like to thank Summer Stevens, Nick Salinas, and many other seasonal technicians for assistance with operating the weir and collecting larval samples. Numerous biologists from CPW, BLM, and USFS as well as local volunteers rendered help on the weir during periods of heavy migration and debris flow. Colleagues at the University of New Mexico and Pisces Molecular (Boulder, CO) analyzed larval and adult genetics.

RESEARCH PRIORITY:
Identify tributary fidelity rates and spawning movement patterns in three-species fishes as well as non-native suckers in the Roubideau Creek drainage.

OBJECTIVES:

- **I)** Determine annual spawning tributary fidelity of PIT-tagged three-species fishes.
- **II)** Compare the extent of tributary use between native and non-native suckers via longitudinal larval sucker sampling and genetic identification.

INTRODUCTION
Information is relatively sparse on whether individual Bluehead and Flannelmouth suckers tend to select specific tributaries and locations for spawning repeatedly or if they stray among tributaries. If they do exhibit high rates of spawning tributary fidelity, efforts to limit hybridization in tributaries such as described in the previous research priority are more likely to result in decreased hybridization in the basin over the long term. In this scenario, a higher proportion of natives are likely to return to controlled tributaries as genetically pure fish recruit to the spawning population following control measures, even if hybridization continues to increase in uncontrolled portions of a basin. Alternatively, if fish stray from tributary to tributary among years, we would expect to see a long term increase in hybridized fish in a controlled tributary, reflecting the basin wide continued increase in hybridization incidence. Therefore, in conjunction with testing the feasibility of spawning run control measures (see previous Research Priority), we deemed it important to simultaneously evaluate tributary fidelity among the three-species fishes. Additionally, in light of the differences in both adult and larval species composition observed between Cottonwood and Potter creeks discussed in the previous Research Priority (i.e. a much higher proportion of White Sucker and their hybrids in Cottonwood Creek than in Potter Creek), we hypothesized that there may be differences in the overall mileage of spawning tributaries that native and non-native suckers use. If this is so, then there may be a natural level of protection against hybridization in some tributaries, wherein pure native sucker recruits are produced in tributary reaches farther from mainstem rivers. We tested this hypothesis by examining the species composition of larval sucker collected at discrete locations between the mouth and headwaters of Roubideau Creek.

METHODS

Since 2014, CPW and partners have been PIT-tagging three-species fishes in the Lower Gunnison Basin. Many of those have been tagged in the Roubideau Creek drainage. In 2015 we installed a PIT-tag detecting, passive interrogation array (PIA) at the mouth of Roubideau Creek. The PIA has been operated continuously since 2015. In 2016, we began deploying portable, submersible PIT-Tag readers (SPRs) in various locations in Roubideau Creek and its tributaries. We have used redetections of PIT-tagged fish on the PIA and SPRs to determine fidelity to the Roubideau drainage as a whole (via PIA detections), and to specific tributaries within the drainage (via SPR detections). We have estimated short term fidelity rates as simply the proportion of fish detected in a given year that return in the following year.

To determine differences in the upstream extent of tributary use between native and non-native sucker, we collected larval fish from multiple locations differing in distance from Roubideau Creek's confluence with the Gunnison River from 2017 through 2019. Larvae morphologically identified as suckers were sent to a genetics lab and analyzed for species or hybrid identification. We then compared the proportion of native versus non-native or hybrid sucker among the difference stream segments sampled.

More detailed methodology on both components of this Research Priority (through 2018 sampling) can be found in the publication referenced below.

Fidelity estimates for 2019 were formulated following the complete methodology described in the technical report, and the longitudinal larval samples collected in 2019 will also be a future component of the final Research Priority addressed in this progress report.

RESULTS AND DISCUSSION

This Research Priority is partially complete. A technical report including this project was completed and published in 2019.

- Thompson, K. G., and Z. E. Hooley-Underwood. 2019. Present Distribution of Three Colorado River Basin Native Non-game Fishes, and Their Use of Tributary Streams. Colorado Parks and Wildlife Technical Publication 52.

During 2019, additional data was collected relating to both objectives. The following is a brief summary of the published results of this Research Priority, as well as an update on the results of the 2019 data collection:

In 2016 -2019, we observed high rates of tributary fidelity to the Roubideau Creek drainage as reported in the technical report. For PIT-tagged native suckers detected entering Roubideau Creek during the spawning period in any given year, 69 to 78% of those fish (not adjusted for annual mortality) were detected again the following year during the spawning period (Figure 1). For Roundtail chub, rates ranged from 75 to 81%. Non-native suckers and hybrids were tagged in low numbers, and after 2016 recaptured tagged fish were culled, but fidelity rates were still observed as high as 72%. Additionally, fidelity to specific tributaries within the Roubideau

Creek drainage appeared high, with the majority of the fish detected in different tributaries having been originally tagged in those same tributaries.

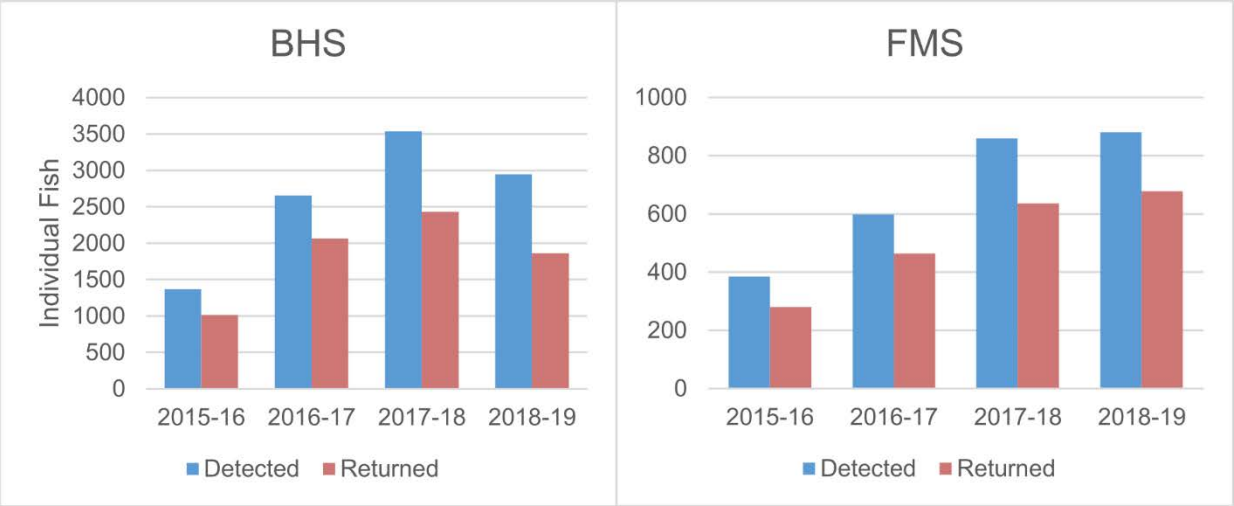

Figure 1. One-year fidelity rates for PIT-tagged BHS and FMS based on Roubideau Creek PIA detections. Bar-pairs represent the number of individual fish detected in $year_i$ (Blue) and redetected in $year_{i+1}$ (Red). The difference between the pairs represents an estimate of the number of individuals that did not return.

The longitudinal larval sucker sampling in 2018 indicated that larval species composition ranged from 100% non-native or hybridized near the mouth to 70% native at 8.6 miles and 100% native at 23.9 miles from the mouth.

The 2019 results from the fidelity study showed similar trends as those presented in the technical report for earlier years, but an extremely elevated snowpack and the resulting heavy runoff interfered with our ability to detect fish. There is some indication from the PIA data that our detection efficiency may have suffered during the higher flows that occurred in Roubideau Creek. The SPRs were deployed as in the previous year, with the exception that the Upper Roubideau Creek SPR was placed in Potter Creek instead, and two additional SPRs were placed in Cottonwood Creek at the same location as in 2017. For much of the peak runoff season, we were unable to access all SPRs due to un-wadeable water conditions. Therefore, readers were inoperable for up to a month as batteries went un-changed for the period. As a result of both this SPR down time and the potentially decreased PIA efficiency, as well as some unknown level of mortality of tagged fish coupled with minimal new tagging in 2018, we detected fewer fish in the drainage in total than in previous years. Nonetheless, fidelity to Roubideau Creek was still high. Of the fish detected on the PIA in 2018, 63.2% and 77.0% of BHS and FMS, respectively, were redetected in 2019 (Figure 1). For RTC, 72.1% of 2018 fish returned in 2019. When averaged across all years of data, the annual return rates were 70.9% (SE = 3.2), 75.4% (SE = 1.2), and 76.3% (SE = 1.9) for BHS, FMS, and RTC, respectively.

In 2019, we also estimated the mean fidelity rate for each species across all years of sampling. For this estimate, we plotted all individuals redetected in $year_{i+1}$ as a function of the number

detected in $year_i$ (Figure 2). To see if fish are likely to return multiple years in a row, we followed groups of fish across time. For instance, we used all fish detected in 2015 ($year_i$) coupled with the number of those redetected in 2016 ($year_{i+1}$) as a data point, as we had in the previous analysis, but then also used those particular 2016 ($year_{i+1}$) redetections coupled with the number of those redetected again for the third time in 2017 ($year_{i+2}$) as a data point. We applied this scheme to all years of data for each species, and fit linear regressions. In the case of non-native suckers (including non-native hybrids), we plotted the data points that were affected by our culling efforts at the 2017 Cottonwood Creek weir separately from points unaffected by those removals and fit regressions for both data sets. The slopes of these regressions approximate

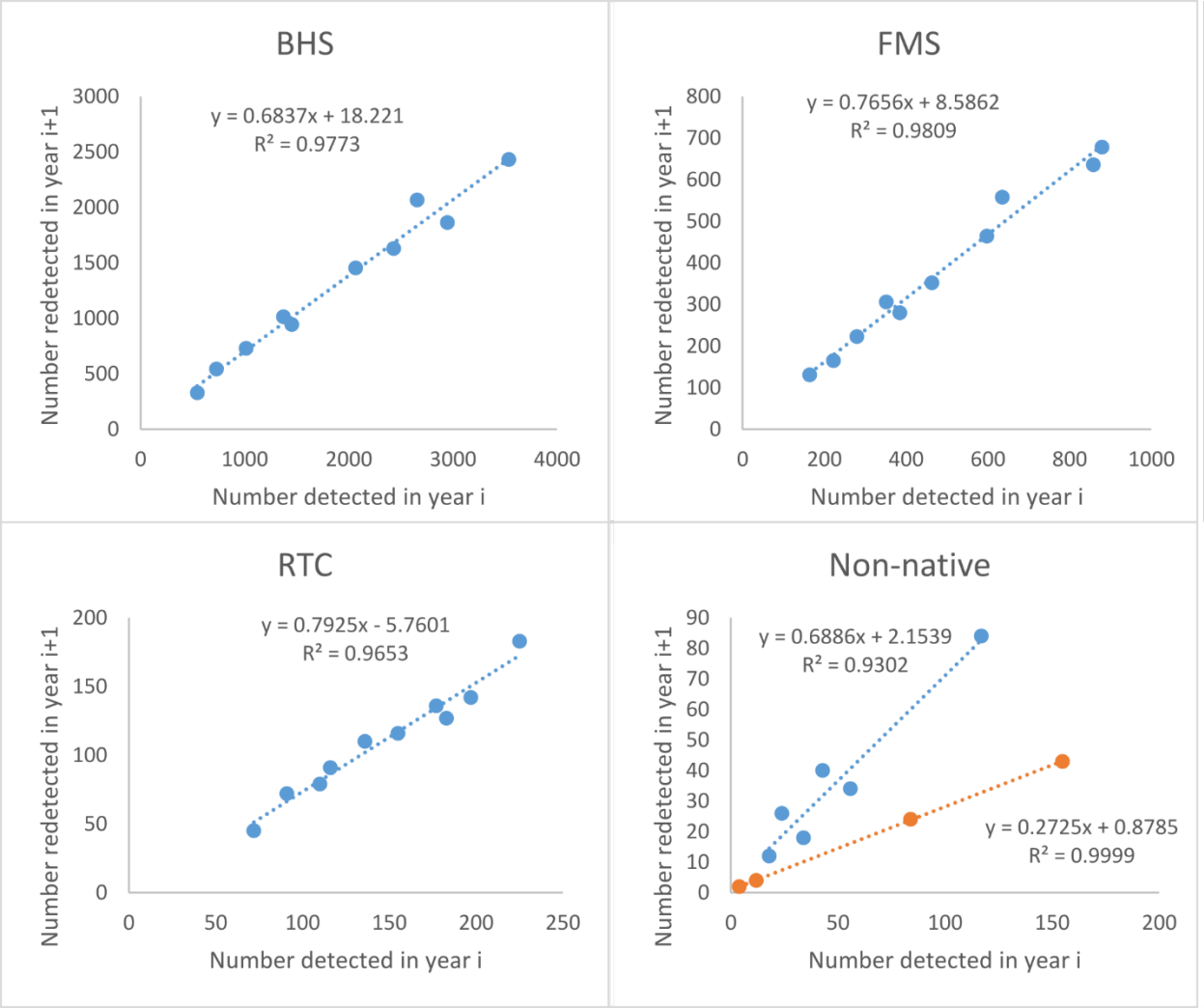

Figure 2. Redetections of PIT-tagged BHS, FMS, RTC, and Non-native suckers (WHS, LGS, and hybrids combined) as a function of the total number of individuals detected in the previous year for all years of data (2015 detections-2019 redetections). Included are multiple year redetections (i.e. Number of redetections of $year_i$ fish in $year_{i+2}$ as a function of the total number of $year_i$ fish detected in $year_{i+1}$). Slopes approximate the overall annual return rate for each species. For non-natives, we plotted data that was affected by our culling efforts of PIT-tagged fish on the Cottonwood Creek Weir in 2017 (orange) independently of the unaffected data (blue).

the average annual return rates and deviations from the average annual rates presented above indicate that groups of fish that return in multiple consecutive years exhibit return rates that vary from the 1-year average. For BHS, FMS, and RTC, the regressions fit the data very well.

Analyzed this way, annual fidelity rates are surprisingly steady (as indicated by the large r^2 values) and are actually somewhat depressed compared to our single year rates for BHS, but slightly elevated for RTC and FMS. This may suggest that FMS and RTC that return at least once to the stream are more likely to return in subsequent years. For non-native suckers, when considering only the data unaffected by 2017 culling, a regression with a slope of 0.69 fit the data well ($r^2 = 0.93$). This indicates that non-native suckers also have high rates of tributary fidelity. The annual rate estimate associated with our culling effort was 27.3% ($r^2 = 0.999$) indicating that even with non-native removal occurring only at Cottonwood Creek we were still able to greatly decrease the number of non-native fish returning to the Roubideau drainage as a whole (it is important to note that we base this estimate from only four data points). These findings offer encouragement for the upcoming resistance board weir project because removal efforts at the weir are likely to have a reducing effect on non-native suckers in following years.

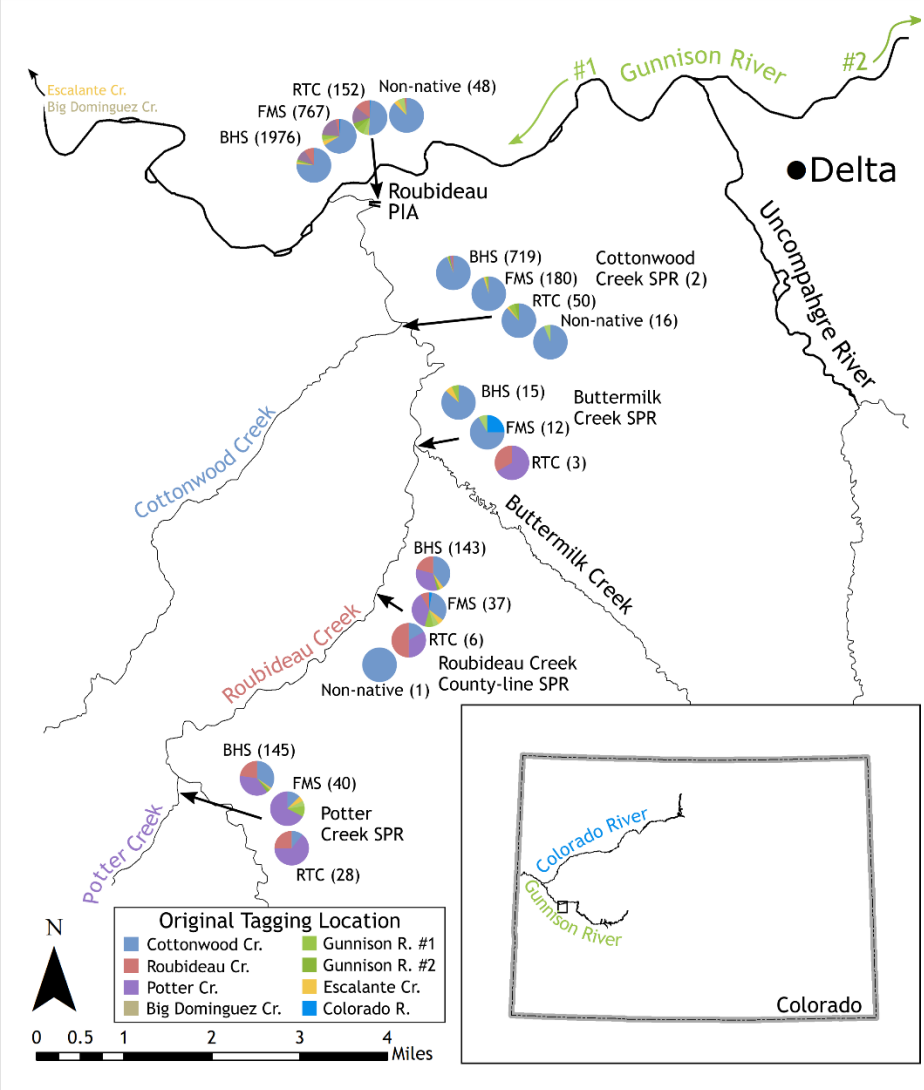

Figure 3. Original tagging locations of fish detected in 2019 at the Roubideau Creek passive interrogation array (PIA) and at five submersible PIT readers (SPR) as a proportion of all redetections (pie charts). Detection numbers (in parentheses) are limited to one occurrence of each individual tag. Data reflect tags implanted prior to 2018.

Considering specific tributary fidelity within the Roubideau Creek drainage in 2019, we saw similar trends as in previous years (as reported in the cited technical report) despite the decreased SPR function due to the high flows (Figure 3). With flows restored to Cottonwood and Potter creeks after the drought in 2018, sucker and chub used both creeks in 2019. By far, the greatest number of detections in the basin were in Cottonwood Creek. Those detections, for all three-species fishes as well as non-native suckers, were heavily weighted towards fish originally tagged in Cottonwood Creek. Potter Creek detections were much less numerous, but the majority of fishes detected there were tagged either in Potter Creek or in Roubideau Creek. The fish tagged in Roubideau Creek were likely intercepted and tagged on their way to Potter Creek in 2015. There was a fairly large number of BHS detected in Potter Creek, originally tagged in Cottonwood Creek, which does suggest that while fidelity to specific tributaries is the norm, straying does occur. The Buttermilk Creek SPR experienced the fewest interruptions in 2019, but it detected a mere 30 individuals. This corroborates our conclusions from the technical report that when water is available in upper Roubideau Creek and its tributaries, sucker and chub do not use Buttermilk Creek in substantial numbers. The Roubideau Creek – County Line SPR was likely the most jeopardized antenna this year as a significant braid formed at its location giving fish ample options to bypass the reader, and it became buried in gravel and sand. This probably decreased its read-range and prevented us from accessing it to change its battery for an extended amount of time. The majority of fish it detected were originally tagged in Roubideau and Potter Creeks, though there was a substantial constituent of fish tagged in Cottonwood Creek as well.

For the longitudinal assessment of species composition based on larval genetics, we again collected larvae from near the Roubideau Creek mouth, Cottonwood Creek, Roubideau Creek at the County Line, Potter Creek, and in upper Roubideau Creek (from Criswell Creek to Moore Creek). However, these samples have yet to be analyzed for species composition as they will serve the dual purpose of longitudinal samples as well as baseline data for the upcoming resistance board weir, non-native exclusion study (see the following Research Priority). For the future study, we will be relying upon a more robust, genotyping by sequencing approach, and will not submit samples for microsatellite analysis as in past years.

ACKNOWLEDGMENTS

All personnel mentioned in the above Research Priorities likely had a hand in the huge PIT-tagging effort behind this project. Dan Cammack (CPW), Russ Japuntich (BLM), Dan Kowalski (CPW), and many seasonal technicians helped place and maintain SPRs. We also thank the operators of the STReaMS Database as we obtained data on many of the fish we detected from that source.

RESEARCH PRIORITY:

Test a resistance board weir as means of controlling the entire Roubideau Creek spawning run, allowing for the selective exclusion and removal of non-native and hybridized suckers.

OBJECTIVES:

- **I)** Test the functionality and operability of a resistance board weir located near the mouth of Roubideau Creek.
- **II)** Evaluate the effect of the weir on the species composition of the larvae produced in the Roubideau Drainage by sampling larvae and genetically assessing their species identity.

INTRODUCTION

As stated in the Cottonwood Creek picket weir research objective, options for mitigating the threat of hybridization are increasingly limited due to the overall range-wide abundance of non-native and hybrid suckers. Protection from hybridization has been achieved for other freshwater species utilizing isolation of key habitats, mechanical removal, chemical removal, or a combination. These methods are largely impractical for BHS and FMS because both species are typically associated with large rivers where financial, resource, social, and political limitations are insurmountable. One conservation action that remains feasible and has the potential to preserve the genetic integrity of large population components involves the mechanical control of spawning runs into tributary habitats that lack resident native or non-native suckers. After determining that maintaining a picket weir in a small intermittent tributary through typical spring runoff conditions was difficult, we searched for alternative control devices. A search for a better weir design led to consideration of resistance board weirs. This design was originally conceived for use in Alaskan salmon runs, and is amenable to streams much deeper and wider than the intermittent Cottonwood Creek. It allows accumulating debris to depress the weir panels and permit debris to flow over, while still constraining upstream fish movement. Thus, it is a promising design for the mainstem Roubideau Creek.

Our objective here is to test the feasibility and effectiveness of mechanically excluding the vast majority of non-native and hybrid suckers from the Roubideau drainage during the spawning season using a resistance board weir. If effective, a decrease in the proportion of larval suckers in the drainage with non-native genetics is expected compared to sampling in previous years. Our ultimate goal is to develop a conservation strategy that can be applied to other tributaries throughout the range of BHS and FMS.

The Roubideau Creek drainage is a prime candidate for a manipulation of this nature. Roubideau Creek lacks resident populations of suckers and offers at least 25 stream-miles of spawning habitat. In 2018, approximately 12% of all fish implanted in 2014-2015 with passive integrated transponder (PIT) tags in the Gunnison River from the Colorado Highway 65 bridge, east of the city of Delta, to the confluence of Roubideau Creek were detected in the Roubideau Creek drainage. Considering this is a conservative estimate because not all PIT-tagged fish were still alive, it highlights the importance of the drainage to the Gunnison River population as a whole.

Additionally, high rates (70-80%) of tagged suckers return from one year to the next, suggesting that spawning tributary fidelity is high (as described in the previous Research Priority). Excluding non-natives and hybrids from the drainage would create a hybridization-free spawning opportunity for a significant portion of the Gunnison River native sucker population, and annual fidelity would allow for the perpetual protection of the genetic purity of the Roubideau Creek-spawning population component, even if hybrid and non-native abundance continues to grow elsewhere in the overall Gunnison River population.

METHODS

In 2019, we secured funding and began the planning process for this Research Priority. We developed a contract with a knowledgeable and experienced firm to fabricate a resistance board weir and fish trap suitably sized for Roubideau Creek. The weir will be deployed near the mouth of Roubideau Creek from about March 15 through early May in 2020 and 2021. We will handle trapped fish, passing those that are deemed pure native suckers (and RTC) upstream to continue their spawning migration while removing non-native and hybrid suckers from the population. To assess our effect on the genetic composition of the larval production in Roubideau Creek, we will collect larvae in May and June each of those two years at four sites along a longitudinal gradient within Roubideau Creek. Additionally we collected larvae in 2019 from the same sites to serve as baseline data. We will test the larvae genetically using a genotyping by sequencing approach to determine species or species admixture of individual larvae. The genetic results will allow us to gauge the success of the weir. The presence and relative abundance of pure native sucker larvae will reveal if our effort to preclude the participation of non-native and hybrid suckers was successful.

RESULTS AND DISCUSSION

This Research Priority is in the development phase. In 2019, we commissioned the construction and installation of the weir and fish trap. The components of the weir and fish trap will be delivered in late February and installed in early March 2020. We also collected larval sucker for our pre-treatment assessment of the incidence of non-native and hybridized larvae in the Roubideau Drainage. Those larvae will be analyzed in the future.

ACKNOWLEDGMENTS

We thank Dr. Elizabeth Mandeville (University of Guelph) who assisted with planning this project and will be involved heavily with the genetic assessment portion of this research. We thank Tracy Kittell and Steve Patterson for assistance with capital development, and Cramer Fish Sciences for designing and fabricating the weir.

TECHNICAL ASSISTANCE SIDE PROJECTS AND COLLABORATIONS

- **Martin/Logan** – Assist with salvage of native fish from two irrigation canals in the Grand Valley following cessation of 2019 irrigation season (Complete)
- **May/Foutz** – Mindy May requested assistance assembling evidence of the use of non-perennial waters by fish and amphibians for a work session of the Water Quality Control Division. I provided Paul Foutz with information on this topic stemming from the three-species research, which he incorporated into his presentation to WQCD. (Complete)
- **Cammack** – Requested information on three-species research that would be incorporated into a lecture given to a CSU fisheries class. Provided Dan with presentations on stream fidelity of PIT-tagged fishes and fish use of Cottonwood Creek for spawning. (Complete)
- **May** – Commented on Water Quality proposed changes for streams in western Colorado river basins. Provided evidence that proposed change of Dry Fork Piceance Creek from coldwater to warmwater status is not warranted, based on temperature data collected there during Fraser M.S. research project in the White River basin. (Complete)
- **Crockett/May –** Sought input on importance of ephemeral and intermittent waters as habitat for fish, used in assembling a CPW White Paper outlining their biological importance and in support of comments by the State of Colorado to EPA and USACOE regarding revisions to the definition of "Waters of the United States". (Complete)
- **Three-Species Rangewide Group** – Reviewed initial database populated with proposed historic habitat segments, made comments and offered rationale for eliminating streams of Strahler order 2 and less and all lentic waters as presumed historical habitat to ease editing. Edited resulting historical habitat for each species to align with historical knowledge and accounts, and expand such knowledge to allow for presumed historic habitat in areas lacking direct information. This was in preparation for populating the database with current information, which commenced in early 2020.