

# THREE-SPECIES RESEARCH PROJECT SUMMARY

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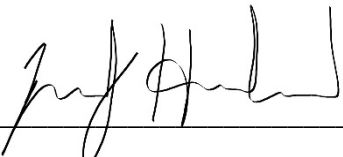
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## **Three-species tributary occupancy and spawning investigations**

Period Covered: March 1, 2020 to October 31, 2020

**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:**

Identify precise distribution of the three-species in several streams and drainages in the Colorado, Dolores, and Gunnison basins.

### **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. Over the past decade, we have sampled extensively to model the fishes' distribution in Colorado, the results of which are available in our 2019 Technical Report. In 2020, we attempted to sample certain locations that refined our knowledge of distribution in certain stream reaches or stream networks.

## **METHODS**

We consulted the three-species database created in winter 2019/2020 for areas where data was missing or indeterminate. We selected locations reachable via day-trips due to COVID-19 travel restrictions, which limited our efforts to the Colorado, Dolores, and Gunnison River basins. We conducted presence/absence surveys with backpack electrofishers. We did one or two pass surveys. If no three-species were captured or observed on the first pass, we did not do a second pass. Occasionally, time limitations precluded a second pass even when presence was established.

## **RESULTS AND DISCUSSION**

For our range-wide sampling and modeling results, please see the following technical report.

- 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 2020, we sampled 20 waters or reaches that have no sampling records in ADAMAS. We found BHS at 11 sites, FMS at 6 sites, and RTC at 5 sites. Only two locations (the Uncompahgre River and Tongue Creek (both Gunnison River tributaries) had all three species. Based on fish size, reproductive condition, and stream connectivity, we tried to identify whether fish collected represented resident or transient (spawning fish or their offspring from mainstem waters) populations, or both. We identified new probable and resident and transient population streams. At several sites (e.g. Roubideau and Naturita creeks) the presence of fishes extended the confirmed upstream distributions of the fishes in the stream. Several surveys (e.g. Uncompahgre River, Tongue and Whitewater creeks) confirmed that previously un-sampled tributaries support at least some spawning and rearing use. These surveys also highlighted several notable populations. First, BHS and FMS were collected in Muddy Creek directly above Paonia Reservoir. This section of Muddy Creek had not been sampled since the creek and Paonia Reservoir were chemically treated for Northern Pike removal in 2012. BHS were found in abundance, but several FMS were found as well. This indicated that the two sucker species persisted in upstream creek reaches and have colonized the lower section of creek. BHS are known to persist in stream reaches upstream of the reservoir, but the FMS caught represent the only ones caught anywhere above the reservoir since the rotenone treatment.

Another notable finding was the presence of small, sexually mature BHS in the Dry Fork of Escalante Creek. This creek is dry the majority of the year, and has a potential passage barrier near its confluence with Escalante Creek. However a reach with possible perennially-wetted pools was found, and BHS did exist in those reaches, even when the rest of the stream was fully dry. Because the fish were small and sexually mature, we suspect that a resident population exists in this isolated section of the stream.

The last notable population we found was in Plateau Creek, which flows into McPhee Reservoir. We sampled Plateau Creek on Lone Mesa State Park, and on BLM lands downstream of the state park. In both locations fish were abundant in disconnected pools. No surface flow existed at the

time of the surveys. Satellite imagery indicates that annually, the stream runs nearly dry aside from pools and beaver ponds. It is interesting that so many fish apparently persist in this intermittent stream with no downstream population source. On the lower BLM section (which had not been previously sampled), we found numerous BHS and WHS, and only a few hybrids. On Lone Mesa State Park, we found one WXB and numerous WXF hybrids. In fact, nearly all suckers were WXF with only a couple pure FMS and WHS present. This population may be a nearly fully introgressed population. In 2007, CPW identified a similarly hybridized population of WXF on the state park, but also found RTC. No BHS were present in 2007. Any pure three-species fishes remaining in this stream are very isolated aboriginal fishes that may have conservation value.

### **ACKNOWLEDGMENTS**

We thank technicians Chase Garvey, Gwen Harris, and Christian Prince, who assisted with or conducted all of the sampling.



## TABLES

Table 1. Surveys conducted to identify three-species presence/absence in streams or stream reaches not previously sampled in Colorado. Stream Condition indicates whether water was present at the time of the survey (Pools = disconnected pools but no active flow). The species columns indicate whether a species was present (+) or absent (-). Population type indicates a suspected resident (R) population, transient (T) component of a downstream population, combination of the two (B), or unknown (U).

Water	Drainage	Water code	Survey Date	Stream Condition	BHS	FMS	RTC	Nonnative/hybrid suckers	Other fish	Population type
Plateau Creek	CR	21559	6/29/20	Wet	-	+	-	-	+	U
Plateau Creek	DO	48531	6/24/20	Pools	+	+	-	+	+	R
Plateau Creek	DO	48531	7/14/20	Pools	-	+	-	+	+	R
Naturita Creek	DO	41804	7/21/20	Wet	+	-	-	-	+	R
Beaver Creek	DO	38201	6/24/20	Pools	-	-	-	-	-	N/A
Lee Creek	GU	41070	4/7/20	Wet	-	-	-	-	+	N/A
Muddy Creek	GU	41741	4/7/20	Wet	+	+	-	+	+	U
Uncompahgre River	GU	46905	4/21/20	Wet	+	+	+	+	+	B
Whitewater Creek	GU	22765	4/28/20	Wet	-	-	+	-	+	T
Whitewater Creek	GU	22765	4/28/20	Wet	-	-	+	-	+	T
Dry Fork Escalante Creek	GU	49432	5/13/20	Wet	+	-	-	-	+	R
Dry Fork Escalante Creek	GU	49432	5/13/20	Wet	+	-	-	-	+	R
Tongue Creek	GU	43656	5/15/20	Wet	+	+	-	+	+	T
Tongue Creek	GU	43656	5/15/20	Wet	+	-	-	+	+	U
Roubideau Creek	GU	42717	5/20/20	Wet	+	-	+	-	-	T
Minnesota Creek	GU	41688	5/27/20	Wet	+	-	-	+	+	R
Kannah Creek	GU	20800	7/9/2020	Pools	-	-	-	-	+	N/A
Dry Fork Escalante Creek	GU	49432	7/15/20	Wet	+	-	-	-	+	R
Cottonwood Creek	GU	39699	7/16/20	Dry	-	-	-	-	-	N/A
Hubbard Creek	GU	40751	5/26/20	Wet	-	-	-	+	+	N/A
Hubbard Creek	GU	40751	5/26/20	Wet	+	-	-	+	+	U

Table 1 continued.

Water	Drainage	Water code	Survey Date	Stream Condition	BHS	FMS	RTC	Nonnative/hybrid suckers	Other fish	Population type
Billy Creek	GU	38415	6/25/20	Wet	-	-	-	-	-	N/A
Tongue Creek	GU	43656	4/30/20	Wet	+	+	+	+	+	T
Tongue Creek	GU	43656	5/6/20	Wet	+	+	+	+	+	T

## **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:**

Determine annual spawning tributary fidelity of PIT-tagged three-species fishes.

### **INTRODUCTION**

Information is relatively sparse on whether individual BHS and FMS 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 following 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 following Research Priority), we deemed it important to simultaneously evaluate tributary fidelity among the three-species fishes.

### **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, and 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. More detailed methodology on this Research Priority (through 2018 sampling) can be found in the publication referenced in the results and discussion section below.

### **RESULTS AND DISCUSSION**

This Research Priority is partially complete. During 2019, a technical report including this project was completed and published and can be referenced for detailed methodology and results through 2018.

- 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 2020 additional PIT tag data was collected. The following is a brief summary of the published results of this Research Priority, as well as an update on the results of the 2020 data collection:

In 2016 -2018, we observed high rates of tributary fidelity to the Roubideau Creek drainage as reported in Thompson and Hooley-Underwood (2019). For PIT-tagged native suckers detected entering Roubideau Creek during the spawning period in any given year, 69 - 78% of those fish (not adjusted for any annual mortality) were detected again the following year during the spawning period (Figure 1). For Roundtail Chub, rates ranged from 75 - 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.

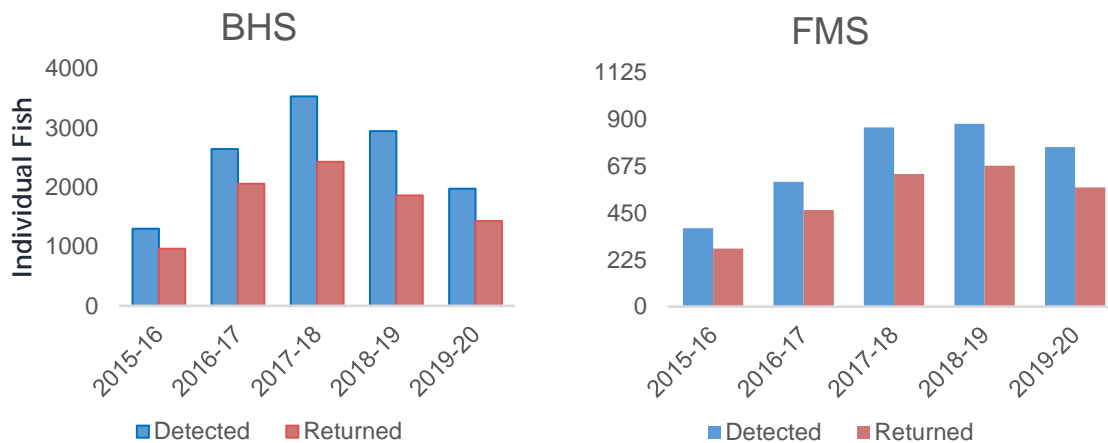


Figure 1. One-year fidelity rates for PIT-tagged BHS and FMS based on Roubideau Creek PIA detections. Bar-pairs represent the number of individual fish detected in year<sub>i</sub> (Blue) and redetected in year<sub>i+1</sub> (Red). The difference between the pairs represents an estimate of the number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

The 2019 results from the fidelity study (see our 2019 progress report) 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. Nonetheless, fidelity to Roubideau Creek was still high. Of the fish detected on the PIA in 2018, 63.2% of BHS and 77.0% of FMS were redetected in 2019 (Figure 1). For RTC, 72.1% of 2018 fish returned in 2019.

In 2020, the drainage received an average amount of snow, but dry soil conditions resulting from an extremely dry fall in 2019 led to a diminished runoff. The duration and magnitude of runoff was well below normal. Cottonwood Creek was usable by fish for less than two weeks –

insufficient time to allow for spawning and maturation of embryos. Potter Creek was useable for a longer period but certainly flows were lower than is typical.

The PIA appeared to be fully functional during the spawning season (March 14 to July 1) in 2020. Of the PIT-tagged three-species fishes and non-native (and hybridized) suckers detected in 2019 on the PIA, redetection rates in 2020 were 72.4% for BHS, 74.7% for FMS, 72.1% for RTC, and 53.1% for other suckers. These rates remain very similar to rates measured in previous years. When averaged across all years of data, the annual return rates were 71.3% (SE = 2.7) for BHS, 75.5% (SE = 0.8) for FMS, and 75.4% (SE = 1.93) for RTC (Figure 1). We have noted an annual drop in individual fish (BHS, FMS, RTC, and non-native/hybridized suckers) detections on the PIA (2020: 2,307 fish; 2019: 2,943 fish; 2018: 4,073), which is almost certainly due to senescence of tagged fish following our last major tagging effort in 2017.

In 2019 and 2020, 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<sub>i+1</sub> as a function of the number detected in year<sub>i</sub>. 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<sub>i</sub>) coupled with the number of those redetected in 2016 (year<sub>i+1</sub>) as a data point, as we had in the previous analysis, but then also used those particular 2016 redetections of 2015 detections (i.e. year<sub>i</sub> & year<sub>i+1</sub>) coupled with the number of those redetected again for the third time in 2017 (year<sub>i+2</sub>) 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 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. A data analysis error was made in 2019, wherein several fish were counted as returned fish from multi-year groups they did not belong in. However these errors were limited and did not noticeably alter the results. The errors have been corrected for this report. For BHS, FMS, and RTC, the regressions including all years fit the data very well (Figure 2). Analyzed this way with the inclusion of 2020 detections, annual fidelity rates remained 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 0.78 fit the data very well ( $r^2 = 0.99$ ). This indicates that non-native suckers also have high rates of tributary fidelity. The annual rate estimate associated with our culling effort was 38.3% ( $r^2 = 0.66$ ) 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 off of only six data points however). 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.

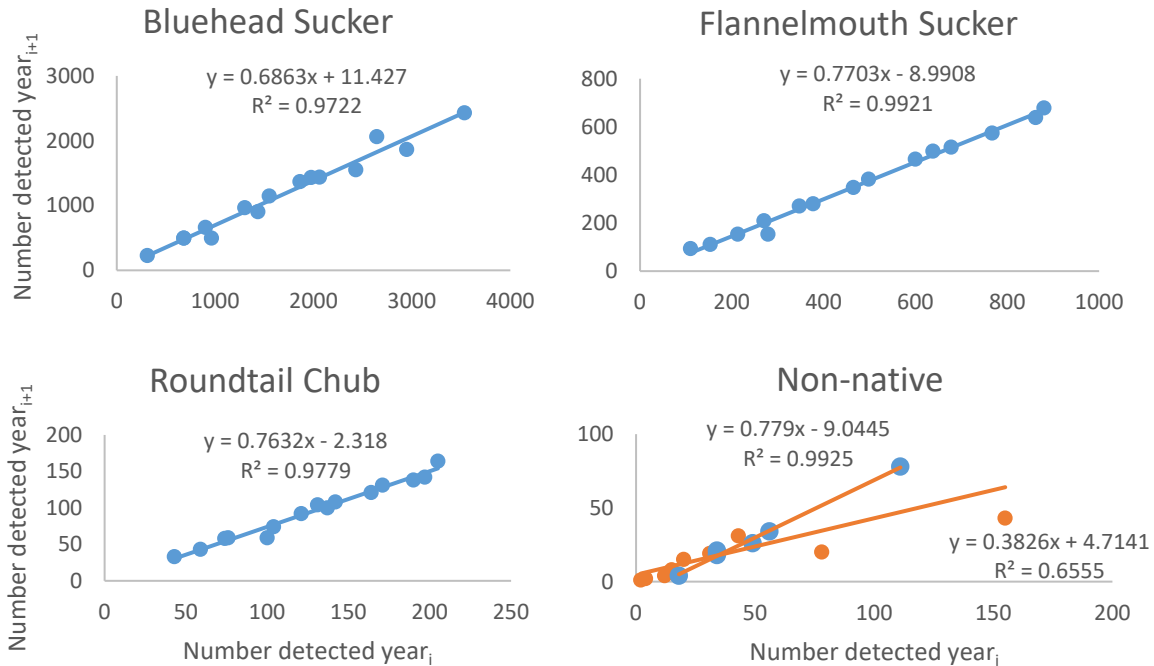


Figure 2. Redetections of PIT-tagged BHS, FMS, RTC, and Non-native suckers (WHS, LGS, and hybrids combined) as a function of the total number of individuals detected in the previous year for all years of data (2015 detections-2019 redetections). Included are multiple year redetections (i.e. Number of redetections of year<sub>i</sub> fish in year<sub>i+2</sub> as a function of the total number of year<sub>i</sub> fish detected in year<sub>i+1</sub>). 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).

Considering specific tributary fidelity within the Roubideau Creek drainage in 2020, we redetected fewer fish than in previous years in general. This was largely due to the low water year which resulted in shorter (and later) windows of accessible flow in the smaller streams throughout the drainage. Even Roubideau Creek upstream of Buttermilk Creek had flows low enough that access was hindered during a substantial portion of the spawning period. We deployed SPRs and detected fish at the same locations as in previous years, except that we did not deploy an SPR at the county-line site on Roubideau Creek, but instead deployed one in Roubideau Creek above Potter Creek (at nearly the same location as in 2018; see the technical report). As in 2018 when flows were exceptionally low, the tributary that resulted in the most individual fish detected this year was Buttermilk Creek, which corroborates our past conclusion that Buttermilk Creek is only used when alternatives are scarce (Figure 3). The fish detected in Buttermilk Creek were mostly BHS, with only a handful of FMS and RTC. Nearly 70% of all individuals were originally tagged in Cottonwood Creek, which is unsurprising considering Buttermilk Creek is the next tributary that fish encounter when Cottonwood Creek is low or dry. In Cottonwood Creek itself, we only detected 31 fish total, but again, fish had about 10 days to access the creek this year, and spawning was likely already underway throughout the drainage by

the time Cottonwood Creek began flowing. The majority of the fish that were detected in Cottonwood Creek were originally tagged in Cottonwood Creek as in past years. In Potter Creek and in Roubideau Creek above Potter Creek, most of the detected fish were originally tagged in either Roubideau or Potter creeks. Due to the limited volume of data this year, it is difficult to draw strong fidelity conclusions based on SPR detections, but there is nothing that counters our conclusion from past years that fish tend to return to the stream in which they were originally tagged when they can.

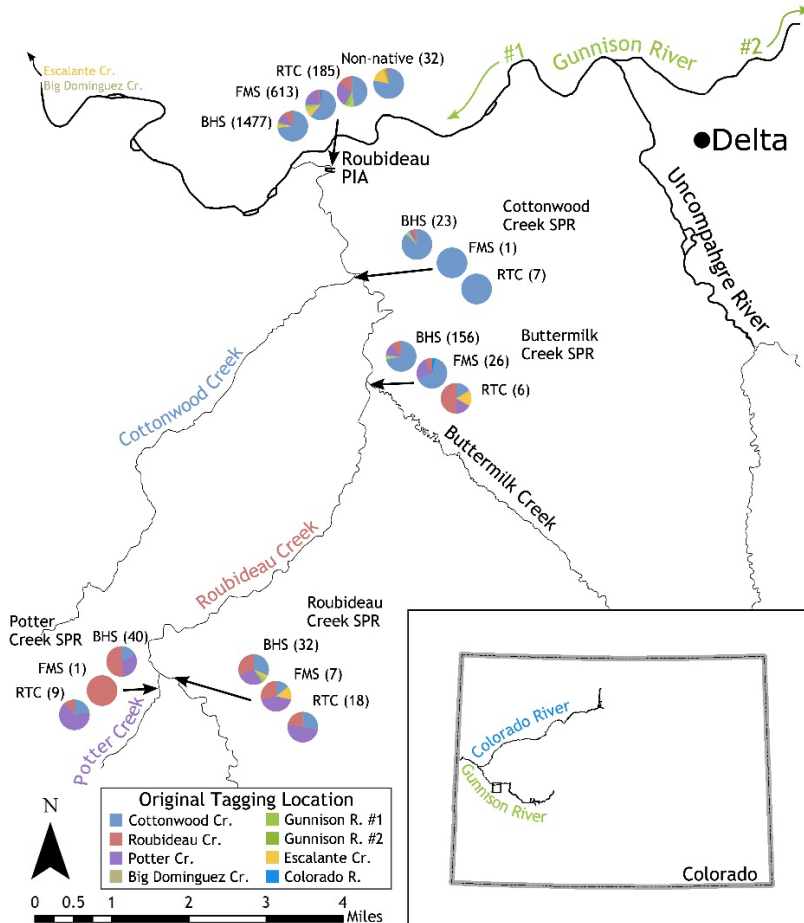


Figure 3. Original tagging locations of fish detected in 2020 at the Roubideau Creek passive interrogation array (PIA) and at four submersible PIT readers (SPR) as a proportion of all redetections (pie charts). Detection numbers (in parentheses) are limited to one occurrence of each individual tag. Data reflect tags implanted prior to 2020.

### ACKNOWLEDGMENTS

All personnel mentioned in the above Research Priorities 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.
- III) Compare the extent of tributary use between native and non-native suckers via longitudinal larval sucker sampling and genetic identification.

## **INTRODUCTION**

Options for mitigating the threat of hybridization to BHS and FMS are increasingly limited due to the overall range-wide abundance of non-native and hybrid suckers. Protection from hybridization has been attempted and sometimes 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 non-native or non-native hybrid adult suckers. Having determined that maintaining a picket weir in a small intermittent tributary through typical spring runoff conditions was very 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 so that debris can flow over the weir, 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 efficacy 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 (RBW). If effective, a decrease in the proportion of larval suckers in the drainage with non-native genetic influence 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 significant resident populations of adult suckers, especially above the confluence of Buttermilk Creek, and offers more than 25 stream-miles of spawning habitat, with more available in tributaries of Roubideau Creek. 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.

Additionally, in light of the differences in both adult and larval species composition observed between Cottonwood and Potter creeks discussed in the Research Priority of investigating a picket weir as a control device (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 seek to continue evaluating this hypothesis by examining the species composition of larval sucker collected at discrete locations between the mouth and headwaters of Roubideau Creek.

## **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 RBW and fish trap suitably sized for Roubideau Creek. We planned to deploy the RBW near the mouth of Roubideau Creek from early March through the middle of May in 2020 and 2021. All trapped fish were handled, and those that were deemed pure native suckers (and RTC) were passed upstream to continue their spawning migration while non-native and hybrid suckers were removed from the population. To assess our effect on the genetic composition of the larval production in Roubideau Creek, we collected larvae in May and June (and will do so each year of the project) at three sites along a longitudinal gradient within Roubideau Creek. We collected larvae in 2019 from the same sites to serve as baseline data. We plan to test the larvae genetically using a genotyping by sequencing approach to determine species or species admixture of individual larvae. The genetic work will be conducted by a graduate research assistant at the University of Guelph, and 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.

To determine differences in the upstream extent of tributary use between native and non-native sucker, we continue to collect larval fish from three locations differing in distance from Roubideau Creek's confluence with the Gunnison River, adding to data collected in 2017 and 2018 under prior research efforts. Larvae deemed to be of the genus *Catostomus* were sent to the University of Guelph to be analyzed for species or hybrid identification. Baseline data, in particular will shed light on longitudinal differences in the genetic composition of larval samples

and help us to understand differing life history behavior among native and non-native or hybrid sucker populations accessing Roubideau Creek for spawning purposes.

## RESULTS AND DISCUSSION

We deployed the weir and trap, under the direction of Cramer Fish Sciences biologist Jesse Anderson, on March 3-4, 2020. The same location will be used in future years, as the substrate rail across Roubideau Creek is a semi-permanent installation and would be very difficult to extract and move. After installation, we used a Smith-Root barge electrofisher to check for the presence of suckers in the vicinity of the weir on March 5. We documented 19 WHS, of which 14 exceeded 280 mm (range 282-358), which would likely be mature adults. This was a surprising result, because no PIT tags had yet been detected on the PIA downstream of our weir site, indicating that no immigration movements from the Gunnison River were yet underway.

Concluding that these fish may have spent the winter in Roubideau Creek, we expended additional effort over four separate days to electrofish Roubideau Creek from the RBW site to above the confluence of Cottonwood Creek, a reach of about 4.5 km. It is 4.1 km from our PIA site to the Cottonwood Creek confluence, and an additional 5.1 km from the mouth of Cottonwood Creek to the mouth of Buttermilk Creek, which provides nearly all the wintertime flow in lower Roubideau Creek. Thus, we fished about half of the available stream below Buttermilk Creek, the portion deemed potentially hospitable to winter occupation. These efforts were conducted from March 10 – 18, and in combination with initial efforts resulted in the removal of 473 WHS and WHS hybrids. Portions of the reach were covered two times. Thus, we believe that we significantly reduced the population of WHS in Roubideau Creek upstream of our weir site prior to the beginning of spawning immigration. Moreover, the stream appeared to host fewer WHS in the portion above Cottonwood Creek, leaving us hopeful that most overwintering fish were in the lower 5 km of Roubideau Creek (including that portion downstream of our RBW). Such efforts will likely be necessary in the remaining years of the study to enhance the odds of success of our exclusion efforts.

The weir was in place and operational from March 4 through March 31. On April 1, in response to the declared COVID-19 pandemic, the weir was removed. The first fish trapped entered on March 15, a date similar to the first tagged fish detections on the PIA in previous years. During the period of operation, 2,470 fish were processed through the weir. Of those, 2,395 were suckers and 84.5 % of suckers were deemed to be pure native suckers (Table 1). Two Brown Trout were captured, but not displayed in Table 1. All non-native and non-native hybrid fish were removed from the population.

Table 1. Numbers of fish captured from March 15 through March 31 at the Roubideau Creek resistance board weir and trap. BHS = Bluehead Sucker, BXG = Bluehead and Longnose sucker hybrid, FMS = Flannelmouth Sucker, LGS = Longnose Sucker, WHS = White Sucker, WXB = White and Bluehead sucker hybrid, WXF = White and Flannelmouth Sucker hybrid, and RTC = Roundtail Chub.

BHS	BXG	FMS	FXB	LGS	WHS	WXB	WXF	RTC
1,689	1	335	94	1	40	47	188	73

The numbers of fish captured at the weir through the first two weeks of immigration were less than expected, based on PIA detections in previous years. When the weir was removed, an explanation became apparent. A number of screws used to hold pvc pickets on to the bulkhead frames that enclose the functional weir had sheared, allowing conduit clamps to loosen or fall off altogether, which in turn allowed excessive flex in the pvc pickets. We suspected that fish were able to bypass the trap entry by forcing their way through these loosened pvc pickets. Substantiating the hypothesis that immigration numbers were low, data downloaded from the PIA after RBW operations ceased showed that 897 unique PIT tags had been detected through March 31, whereas only 268 of those were handled through the RBW and trap. The PIA is just downstream of the RBW location, and it is exceedingly unlikely that many of those fish were occupying the short reach of water between the two facilities.

The combination of poor trapping success and the early shutdown of the weir as a response to COVID-19 has resulted in a re-structuring of the project timelines. We intend to proceed, but will count 2020 as a second baseline year. Although there was a manipulation attempted, it was not successful in forcing all immigrants through the trap, and the trap was disabled with several weeks of heavy immigration still pending. Thus, we believe that efforts in 2020 would have had little effect on the proportion of pure native sucker larvae produced in Roubideau Creek, although it will be interesting to see whether the data indicate a weak effect.

Prior longitudinal larval sucker sampling in 2018 indicated that larval species composition ranged from 100% non-native or hybridized near the mouth to 70% and 100% native at 8.6 and 23.9 miles upstream from the mouth. Larvae collected in 2019 and 2020 were submitted to the University of Guelph in September 2020, and processing of those samples is underway.

## **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 as the major advisor of the graduate research assistant. We thank Tracy Kittell and Steve Patterson for assistance with capital development, and Cramer Fish Sciences for designing and fabricating the weir. We thank Jesse Anderson of Cramer Fish Sciences for assistance and training in installation and maintenance of the resistance board weir.

## TECHNICAL ASSISTANCE AND COLLABORATIONS

- **Three-Species Rangewide Group** – Kevin Thompson and Zack Hooley-Underwood participated on behalf of Colorado in populating the three-species rangewide database through several scheduled workshops and continuing efforts through November 2020. Zack Hooley-Underwood queried State data for all pertinent data and manipulated into a format that made populating the rangewide database efficient for all CPW biologists and external agency collaborators in Colorado.
- **Graduate Committee** – Kevin Thompson is serving on the graduate committee for Reece Samuelson, M.S. candidate at Western Colorado University under Dr. Derek Houston. Also assisted with sampling in the Dolores River basin to implant additional PIT tags in support of this graduate study.
- **Stream Reclamation Treatment** – Kevin Thompson assisted biologists Stev Vigil and Carrie Tucker with rotenone reclamation of Sand Creek, Great Sand Dunes National Preserve.
- **Razorback Sucker Observational Paper** – Zack Hooley Underwood and co-authors Kevin Thompson and Kevin Bestgen (Colorado State University) prepared a manuscript detailing Razorback Sucker detections and captures in Roubideau and Cottonwood Creeks. The intent of the manuscript is to provide Razorback Sucker managers with observational evidence that intermittent streams may be more frequently used by the species than the literature indicates, thereby helping them tailor their conservation strategies in the region. As of February 2021, the manuscript was accepted by the North American Journal of Fisheries Management, pending revision.