# **Coldwater Stream Ecology Investigations**

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2023 Progress Report

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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.

## **Table of Contents**

| Colorado River Ecology and Water Project Mitigation Investigations                   | 1  |
|--|----|
| Habitat Preferences of Pteronarcys californica and Factors Related to Range Declines | 9  |
| Sculpin Diversity, Phylogeny, and Meristics Project                                  | 16 |
| Trout Diet Habits in Upper Colorado River Basin Tailwater Fisheries                  | 21 |
| Technical Assistance   | 39 |

## COLDWATER STREAM ECOLOGY INVESTIGATIONS PROJECT SUMMARY

Period Covered: July 1, 2022 to June 30, 2023

### **PROJECT OBJECTIVE**

Improve aquatic habitat conditions and angling recreation in Colorado by investigating biological and ecological factors affecting sport fish populations in coldwater streams and rivers in Colorado.

## **RESEARCH PRIORITY**

Colorado River Ecology and Water Project Mitigation Investigations

### **OBJECTIVE**

Investigate the ecological impacts of stream flow alterations on aquatic invertebrates and fish of the Colorado River and evaluate the mitigation efforts associated with Windy Gap Firming project.

## **INTRODUCTION**

Dams are known to drastically alter the habitat of rivers and have a multitude of effects on fish and aquatic invertebrates (Ward and Stanford 1979). On the Colorado River, not only have dams altered the temperature and flow regime of the river, but trans-basin water diversions remove approximately 67% of the annual flow of the river and future projects will deplete flows further. Previous work by CPW researchers identified ecological impacts of streamflow reductions and a mainstem reservoir on the invertebrates and fish of the river (Nehring et al. 2011). The health of the invertebrate community has declined after the construction of Windy Gap Reservoir, with a 38% reduction in the diversity of aquatic invertebrates from 1980 to 2011. A total of 19 species of mayflies, four species of stoneflies, and eight species of caddisflies have been extirpated from the sampling site below Windy Gap Reservoir (Erickson 1983; Nehring et al. 2011). Historically, the Salmonfly (*Pteronarcys californica*) was common in the upper Colorado River but has become rare below Windy Gap Reservoir (USFWS 1951; Nehring et al. 2011).

In addition to impacts on the aquatic invertebrate community, Windy Gap Reservoir has altered the fish community of the upper Colorado River. Native sculpin, once common, are now rare or extirpated immediately below Windy Gap Reservoir (Dames and Moore 1977; Nehring et al. 2011). These fish currently recognized as *Cottus bairdii* are likely different species, the Colorado Sculpin *C. punctulatus* or Eagle River Sculpin *C. annae* (Young et al. 2020). Stream reaches below several dams and water projects in Middle Park have reduced density and range of sculpin (Nehring et al. 2011). The decline in sculpin distribution appears both temporally and spatially related to impoundments (Kowalski 2014). A survey in 1975-1976, before Windy Gap Reservoir construction, documented sculpin at all sampling sites (Dames and Moore 1977). In 2010, a

project investigating the distribution of sculpin in the upper Colorado River revealed that their density was 15 times higher in sites above impoundments compared to downstream sites (Nehring et al. 2011). In the main stem Colorado River between Windy Gap Reservoir and the Williams Fork, a single fish was sampled in 3,200 ft of river sampled by electrofishing. This study attributed the decline of sculpin in the upper Colorado River to habitat changes related to flow alterations, changes in sediment dynamics, and water depletions below the reservoir. Surveys in 2013, 2018, and 2019 confirmed these patterns finding sculpin common above impoundments on the upper Colorado River but rare or absent downstream (Kowalski 2014, Kowalski 2019).

The planned Windy Gap Firming Project will increase trans-basin water diversions from the upper Colorado River. There are ongoing efforts to implement mitigation measures to reduce the impact of the new projects (Northern Water Conservancy District 2011). A large component of the mitigation plan is the construction of a bypass channel around the reservoir. This will reconnect the Colorado River and address various effects of a large, main-stem impoundment but overall the firming project will exacerbate flow depletions from the system. The Colorado River Connectivity Channel (CRCC) offers a unique opportunity to evaluate the effects of reconnecting the river and investigate if mitigation measures can offset the impacts of large flow depletions on the ecology of the river.

## **METHODS**

Construction activities began on the CRCC in 2022 and the official groundbreaking occurred on August 23, 2023. Northern Water Conservancy District anticipates completion of channel construction in 2024 but the channel should be functioning and have water late in 2023. All pre-project invertebrate and sculpin sampling was completed in 2018-2021, no aquatic invertebrate samples were taken in 2022 due to construction activities. Earlier progress reports contain summary of pre-project invertebrate data (Kowalski 2019; Kowalski 2022).

Aquatic invertebrate samples were taken at six sites on the Colorado River in 2018-2021 and fish sampling occurred at four sites (Table 1, Figures 1-2). Invertebrate samples were collected by two different protocols commonly used in Colorado, the standard USGS method used by the National Water Quality Monitoring Laboratory (Moulton et al. 2000) and the MMI method used by Water Quality Control Division of the Colorado Department of Public Health and Environment (CDPHE). Samples were taken by both methods from the same natural riffles at each site.

The USGS method involved taking five replicate macroinvertebrate samples at each site using a 0.086 m2 Hess sampler with a 350  $\mu$ m mesh net. Because a known and exact area of stream bottom is sampled by the Hess sampler, true density estimates can be made. Macroinvertebrate samples were sorted and sub-sampled in the laboratory using a standard USGS 300-count protocol, except that replicates were not composited (Moulton et al. 2000). Approximately 300 individual organisms were identified from each replicate and a 15 minute search for large or rare organisms was conducted on the entire sample. All organisms, except for chironomids and non-insects, were identified to genus or species. Chironomids were identified to family and non-insects were identified to class. Each replicate sample was processed separately so that more

individual specimens were identified from each site to ensure rare organism were identified and to increase the power of the comparisons between riffle sites in close proximity (Vinson and Hawkins 1996). All taxonomic identifications followed recommendations by Moulton et al. (2000) and were completed by a single CPW invertebrate taxonomist. Recommended quality control and quality assurance procedures were followed and at least 10% of all individual identifications were verified by an independent taxonomist (Moulton et al. 2000).

The MMI is a multimetric index that is that standard regulatory method used by the state of Colorado to determine stream impairment under the Colorado Water Quality Control Act and the Federal Clean Water Act (CDPHE 2010a). Multimetric indices combine invertebrate community information with expected species composition and community metrics from reference sites. They have been shown to be an effective and cost-efficient method for invertebrate bioassessment (Hughes and Noss 1992; Barbour et al. 1995; Karr 1998). Sampling protocols followed standard methods and involved collecting a semi-quantitative kick net sample from each site (CDPHE 2010b). Approximately one square meter of stream bottom was disturbed for a timed one minute and all organisms were preserved in 80% ethanol. Sampling occurred on the same day and from the same riffles as the USGS method. Processing the MMI samples involves subsampling and identifying 300 individual organisms from the entire sample, including chironomids to species.

The Colorado MMI is made up of metrics that represent various aspects of the community structure and function and are grouped into five categories: taxa richness, composition, pollution tolerance, functional feeding groups, and habit. Combining metrics from these categories into a multi-metric index transforms invertebrate sampling data into a unit-less score that ranges from 0-100 that indicates the community health and stream condition (CDPHE 2010a).

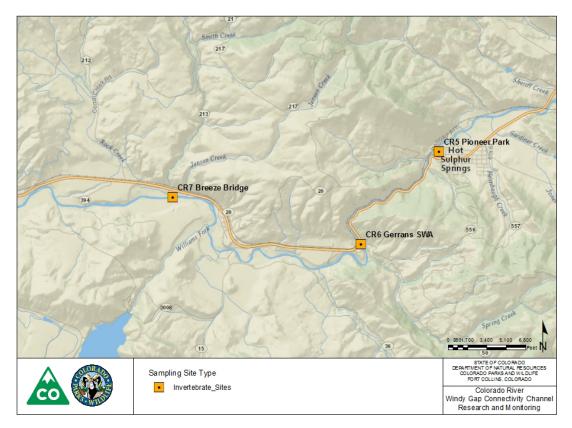
The method generates a standardized multimetric index score specifically developed for Colorado streams, the MMI. Because the area of stream bottom sampled is approximated and sampling time is restricted, the CDPHE method cannot provide true density estimates. Instead, it is an index of invertebrate community health collected by standardized methods where sites can be compared to each other as well as to reference sites of similar stream types. Because a standardize area is sampled and specific time limits, relative densities of insects can be calculated.

| Site # | Site Name                     | UTM East | UTM North |
|--------|-------------------------------|----------|-----------|
| CR1    | Fraser Confluence             | 416914   | 4439457   |
| CR2    | Hitching Post                 | 414652   | 4440330   |
| CR3    | Chimney Rock, Red Barn        | 412703   | 4439648   |
| CR5    | Pioneer Park SWA              | 405504   | 4436635   |
| CR6    | Hot Sulphur SWA, Gerrans Unit | 403440   | 4434141   |
| CR7    | Breeze Bridge                 | 398319   | 4435421   |

**Table 1.** Aquatic invertebrate sampling sites on the Colorado River 2018-2021.



**Figure 1.** Map of the upper benthic macroinvertebrate sampling sites on the Colorado River in 2020.



**Figure 2.** Map of the lower benthic macroinvertebrate sampling sites on the Colorado River in 2020.

### **RESULTS AND DISCUSSION**

Fish and aquatic invertebrate sampling results from the upper Colorado River 2018-2021 reflect the patterns presented in previous work (Nehring et al. 2011; Kowalski 2019). Generally, while healthy and diverse invertebrate communities exist upstream of the reservoir, sites downstream of Windy Gap Reservoir are less diverse, have fewer sensitive species, and are lower in density and diversity of stonefly species (Fig 3). Several sites below Windy Gap Reservoir fall below the state standard for coldwater stream impairment on some years. Fish sampling results from 2018-2021 also reflect patterns previously observed in the upper Colorado River, native Mottled Sculpin continue to be absent from sites below Windy Gap Reservoir while they are common above the reservoir and in tributary streams

Both the USGS method and CDPHE method were informative in evaluating the aquatic invertebrate community of the sampling sites and generally gave similar information on the trends between sites. The USGS method was superior for detecting rare species, fully characterizing the diversity at each site, and giving true density estimates. The CDPHE method was faster, more cost-effective, superior for identifying midges and oligochaete worms, and has the added benefit of being able to produce standard metric scores comparable to the state water quality standards and to other locations in western Colorado.

Interestingly, there has been an improvement in invertebrate community diversity at the Hitching Post site immediately below WGR in 2020 and 2021. This improvement appears to be restricted to this site, as most of the other sites downstream have generally been stable or declining in community diversity indices (Figure 3). The positive community diversity trends at the Hitching Post Bridge site were a result of an increase in EPT taxa richness driven by Plecoptera species. Four stonefly species (Chloroperlidae, *Isoperla fulva, Claassenia sabulosa*, and *Skwala americana*) were found where previously only one or two species were present 2018-2020. No *P. californica* or *Pteronarcella badia* have been sampled at this site since 2018 despite being present before Windy Gap Reservoir. Other sampling methods, such as exuvia surveys during emergence, designed to detect rare invertebrates, have confirmed that *P. californica* at this site in very low densities, whereas they are rarely found using the standard MMI sampling protocol.

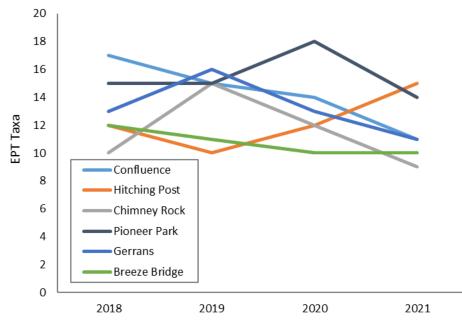


Figure 3. EPT taxa richness of invertebrate sampling sites on the Colorado River 2018-2021.



**Figure 4.** Windy Gap Reservoir in September 2020. The reservoir was drained in the fall of 2020 and 2021 leaving a remnant river channel passing through the bed of the reservoir potentially reconnecting the river for some time and allowing passage of fish and invertebrates.

The improvement of the invertebrate community at the Hitching Post site is likely related to changes in reservoir operations at Windy Gap 2019-2023. In preparation for construction of the CRCC, Windy Gap was drained each fall for during preparation and construction work (Figure 4). This has likely had some major ecological effects on the river below the reservoir. The drawdown created a more natural stream channel through the bed of the reservoir and reconnected the river above and below Windy Gap. Evidence for the temporary reconnection of the river includes documented fish movement both upstream and down through the reservoir channel during the drawdown and when the dam's auxiliary gate was open. Downstream dispersal of aquatic invertebrates was also likely during this time and may explain the increase species richness at the Hitching Post site in 2020 and 2021.

Overall, the results of benthic sampling in the Upper Colorado River 2018-2021 reflect the patterns in invertebrate community of the Colorado River presented in previous work (Nehring et al. 2011; Kowalski 2019) but with some interesting new patterns. Generally, while healthy and diverse invertebrate communities exist upstream of the reservoir and at some sites downstream, most sites below Windy Gap Reservoir are less diverse, have lower numbers of sensitive species, and are lower in the density and diversity of stonefly species. The impaired invertebrate community below Windy Gap is likely due to habitat changes in the river associated with the shallow main stem impoundment and its associated water depletions (Nehring et al. 2011; Kowalski and Richer 2020). Recent changes in reservoir operations show some promising trends and bode well for an improvement in the invertebrate community after the river is reconnect with a bypass channel.

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### **RESEARCH PRIORITY**

Habitat Preferences of the Stonefly *Pteronarcys californica* and Factors Related to Declines in Range

Coauthor: Jackson Birrell, Graduate Research Associate, University of Montana, Missoula, Montana.

### **OBJECTIVE**

Investigate the habitat use of the salmonfly *Pteronarcys californica* in Colorado rivers and explore the factors related to their decline.

## **INTRODUCTION**

Giant Salmonflies, *Pteronarcys californica*, are among the largest of all stoneflies (Insecta: Plecoptera) and are endemic to Western North America. Salmonflies frequently occur in high densities (> 400 m<sup>2</sup>) in mid-sized mountain streams and are often a major component of instream biomass (Nehring et al. 2011). They play a key role in nutrient cycling as shredders of leaf material (DeWalt and Kondratieff 2019; Vannote et al. 1980), provide an important food resource for trout populations (Nehring 1987), and transfer massive amounts of carbon to terrestrial systems during their large, synchronous emergences (Walters et al. 2014). Giant Salmonflies are sensitive to human disturbances and are used as bioindicators of river health (Barbour et al. 1999). They are also recreationally important to anglers because of the quality of the fishing during their emergence. Despite their ecological and cultural importance, reduction in the range and density of *P. californica* populations have been observed across the western United States.

Salmonfly declines have been reported in at least 10 rivers in the western U.S. They have been lost from >550 km of river in Montana, including reaches on the Madison, Smith, Big Hole and Clark Fork Rivers (Stagliano 2010). In Colorado, P. californica has been extirpated from the Arkansas River (Benzel 2016), and from several reaches of the Colorado (Nehring et al. 2011) and the upper Gunnison River (Elder and Gaufin 1973; Wiltzius 1976; Colborn 1985). In Utah, they have also been lost from the Logan River (Vinson 2011) and much of the Provo (Birrell et al. 2019) and Ogden Rivers. The factors influencing the declines of P. californica are not well understood. Changes in physical habitat, stream temperature, and oxygen levels may play a role (Anderson et al. 2019; Birrell et al. 2019; Kowalski and Richer 2020). In the Gunnison River, when temperature is not limiting, fine sediment deposition and cobble embeddedness may be driving Salmonfly range and density (Kowalski and Richer 2020). However, Giant Salmonfly disappearances in Utah do not appear to be correlated with high levels of fine sediments. Although abiotic factors, such as temperature, oxygen, and sedimentation, may play a role in *P*. californica declines, little work has been done to assess the importance of biotic interactions, such as diet and food availability. The interactions of abiotic and biotic factors likely influence the range and distribution of this stonefly, and more work is needed to explore these factors in the Gunnison and other rivers.

In the Gunnison River specifically, Salmonflies have declined in range after the completion of the Aspinall project both above Blue Mesa Reservoir and immediately below Crystal Reservoir (Elder and Gaufin 1973, Wiltzius 1976). Currently, there is a thriving population of Salmonflies approximately five miles below the lowest of the three hydroelectric dams in the lower part of the Black Canyon National Park (BCGNP) and downstream throughout the Gunnison Gorge NCA. The density and distribution of larval Salmonflies declines closer to Crystal Dam, likely due to temperature and physical habitat limitations related to the large ecological impacts of regulated flow and altered temperature regime caused by the large bottom release impoundment.

The objective of this study is to explore the abiotic (temperature and physical habitat) and biotic (diet) factors that may be influencing Salmonfly density and range and to explain their disappearance from specific rivers, specifically below regulated impoundments.

## **METHODS**

Fifteen sites on the Gunnison River were sampled from Almont to Austin, Colorado in 2022 for Salmonfly density and abiotic habitat factors (Table 2). At eight sites that contained Salmonflies, 3-10 individual larvae were collected and frozen for diet analysis to be completed by collaborators and N.C. State University. Abiotic habitat sampling involved measuring dominant particle size (D50) of riffles with a pebbled count, fine sediment with a visual grid method, cobbled embeddedness with the Bain visual method as well as a an estimate of force to move cobble particles. Temperature is being monitored with Hobo Pendant temperature logger and dissolved oxygen was monitored with a PME MiniDOT meter. Flow will be estimated using USGS gage data and discharge models. Three of these sites in BCNP were also sampled for aquatic invertebrate community structure because little invertebrate work has been done in this part of the river historically. Invertebrate community health.

## PROGRESS

Salmonfly density estimates varied by site, but generally followed expected patterns previously observed (Figure 1). There were no Salmonflies observed upstream of Blue Mesa Reservoir or immediately downstream of Crystal Reservoir. Approximately five miles below Crystal Reservoir, Salmonflies were found at Gunnison Point. Densities increased downstream with the highest densities observed in Ute Park in the Gunnison Gorge NCA. Salmonfly densities generally decline below Ute Park and they were present in low densities at Smiths Mountain and absent from Drysdales site near Austin, Colorado, approximately eight miles below the confluence with the North Fork of the Gunnison.

Sampling in 2023 focused on monitoring the emergence time of Salmonflies at all 15 sites and monitoring temperature and dissolved oxygen at select sites. Where and emergence occurred, the first and last day of emergence was observed and density estimates were made at a subset of the sites.

Habitat data is being currently being complied and analyzed and will be completed by 2024. Analysis of the diet samples is ongoing and the project is expected to be complete, including draft manuscripts, in 2024.

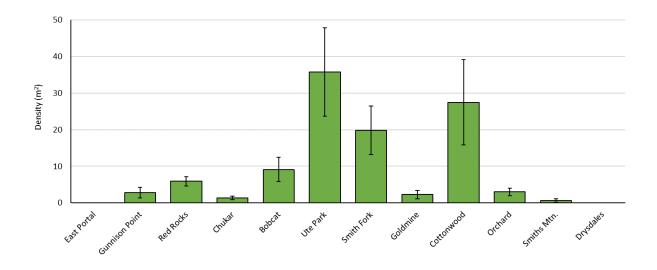
#### Aquatic Invertebrate Community Results

Preliminary analysis of the MMI sampling revealed interesting, but not unexpected results. Downstream of Crystal Reservoir the invertebrate community has low diversity but high density of select taxa (Tables 3-4). All three sites were dominated by tolerant, coldwater species like midges, blackflies, and scuds. This pattern has been reported before and is typical of invertebrate communities downstream of large bottom-release reservoirs (Ward and Stanford 1979; Vinson 2001).

The East Portal site was dominated by *Gammarus lacustris* (30.6%), and 33.4% *Baetis tricaudatus*, with only one stonefly species present *Hesperoperla pacifica*. The Gunnison Point site was dominated by oligochaete worms in the genus *Nais* (38.0%) and Chironomidae (37.1%), mostly genus Tvetenia. Two stonefly species were also present (*H. pacifica* and *P. californica*). This is farthest upstream (closest to Crystal Dam) that Salmonflies have been documented as larvae. The Red Rocks site was dominated by Simulidae (67.9%) and also had two stonefly species *H. pacifica* and *P. californica*. The densities of Salmonflies were higher at the Red Rocks site than Gunnison Point. The diversity of the invertebrate communities and the MMI scores was low at all sites but increased going downstream (Table 3).

| Site<br>Code | Site                  | UTM (NAD83, Z13) | Sampling Completed                    |
|--------------|-----------------------|------------------|---------------------------------------|
| GR1          | Almont Campground     | 338493, 4280034  | Benthic, Temp, DO, Habitat            |
| GR2          | Garlic Mikes          | 332499, 4271989  | Benthic, Habitat                      |
| GR3          | Gunnison Whitewater   | 329747, 4266462  | Benthic, Temp, Habitat                |
| GR4          | East Portal           | 269116, 4267719  | Benthic, Temp, DO, Habitat, MMI       |
| GR5          | <b>Gunnison Point</b> | 266346, 4271222  | Benthic, Diet, Temp, DO, Habitat, MMI |
| GR6          | Red Rocks             | 257277, 4275405  | Benthic, Diet, Temp, Habitat, MMI     |
| GR7          | Chukar Trail          | 253421, 4278775  | Benthic, Habitat                      |
| GR8          | Bobcat                | 251353, 4280344  | Benthic, Diet, Temp, Habitat          |
| GR9          | Ute Park              | 252376, 4284894  | Benthic, Diet, Temp, Habitat          |
| GR10         | Smith Fork            | 253338, 4291889  | Benthic, Temp, DO, Habitat            |
| GR11         | Goldmine              | 253728, 4295747  | Benthic, Diet, Temp, Habitat          |
| GR12         | Cottonwood            | 252129, 4295940  | Benthic, Diet, Temp, Habitat          |
| GR13         | Orchard Boat Ramp     | 247947, 4295297  | Benthic, Diet, Temp, DO, Habitat      |
| GR14         | Smith's Mountain      | 246534, 4295614  | Benthic, Diet, Temp, Habitat          |
| GR15         | Drysdales             | 245053, 4296502  | Benthic, Temp, DO, Habitat            |

**Table 2.** Gunnison River Salmonfly sampling sites 2022.



**Figure 5.** Salmonfly larvae density and 95% confidence intervals at Gunnison River sites below the Aspinall Unit reservoirs in 2022. No Salmonflies were sampled at Gunnison Whitewater, Garlic Mikes, Almont Campground above the dams or East Portal, or Drysdales sites below the dams.

| <b>Community Metrics</b> | East Portal | <b>Gunnison Point</b> | <b>Red Rocks</b> |
|--------------------------|-------------|-----------------------|------------------|
| Total Taxa Richness      | 20          | 26                    | 29               |
| EPT Taxa Richness        | 3           | 6                     | 8                |
| Plecoptera Richness      | 1           | 2                     | 2                |
| SDI                      | 2.62        | 2.55                  | 1.65             |
| MMI                      | 12.9        | 15.9                  | 37.0             |

**Table 3.** Community metrics and index scores for invertebrate sampling in Black Canyon of the Gunnison National Park in 2022.

| Order         | East Portal | <b>Gunnison Point</b> | <b>Red Rocks</b> |
|---------------|-------------|-----------------------|------------------|
| Nematoda      | 0           | 0                     | 0.1              |
| Oligochaeta   | 22.7        | 41.4                  | 2.8              |
| Amphipoda     | 30.6        | 0.4                   | 0.2              |
| Ephemeroptera | 33.6        | 17.5                  | 17.7             |
| Plecoptera    | 0.1         | 0.1                   | 0.4              |
| Trichoptera   | 0.0         | 0.1                   | 0.7              |
| Coleoptera    | 1.4         | 0.9                   | 2.9              |
| Diptera       | 11.6        | 39.4                  | 74.6             |
| Gastropoda    | 0           | 0.1                   | 0.1              |
| Bivalvia      | 0           | 0                     | 0.4              |

**Table 4.** Relative abundance invertebrate orders in Black Canyon of the Gunnison National Park in 2022.

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## **RESEARCH PRIORITY**

Sculpin Phylogeny, Diversity, and Morphology in Colorado

Coauthored by Michael K. Young, National Genomics Center for Wildlife and Fish Conservation, Missoula, MT. Results are summarized from Young et al. (2020) and Young et al. (2022).

## **OBJECTIVE**

Use molecular techniques to identify sculpin from Colorado to evaluate diversity within and between species, document their distribution, and to assess their phylogenetic relatedness to other lineages of sculpin. Compare morphological and meristic characters of sculpin in Colorado to identify distinctive characters and evaluate the physical differences among sculpin in Colorado.

## **INTRODUCTION**

There has long been taxonomic uncertainty about the identity of lineages of sculpins in Colorado (Woodling 1985; Moyle 2002; Kinziger et al. 2005). Sculpin are among the most difficult freshwater fishes to identify based on morphological characteristics (Jenkins and Burkhead 1994), a difficulty compounded by geographic variation in phenotypically diagnostic characters within individual species (Maughan 1978; McPhail 2007). Currently there are two recognized species of sculpin in Colorado, the Mottled Sculpin *Cottus bairdii* and the Paiute Sculpin *C. beldingii*, but the morphological characteristics of those species do not differentiate them and are not diagnostic for identification. Colorado Parks and Wildlife biologists and researchers have long suspected that sculpin in Colorado do not morphologically align with the described type specimens of Mottled Sculpin and Paiute Sculpin and recent publications have supported that hypothesis.

Gill (1862) first described a sculpin from the Colorado River basin as *Potamocottus punctulatus*, which was collected between Bridger Pass and Fort Bridger, Wyoming, likely from the Little Snake or Green River basins. Subsequently, sculpins of this lineage from the Colorado River basin were assigned a variety of generic, species, and subspecies names, and are presently recognized as Mottled Sculpin *C. bairdii*. Neely (2001) argued that *C. bairdii* should be restricted to sculpins from a portion of the Ohio River basin, and that the former members of this taxon in western North America constituted a mixed of named and unrecognized species. He proposed that those from the Colorado River basin be recognized as *C. punctulatus*, the Colorado Sculpin. Other researches have come to the same conclusions that the fish recognized as the Mottled Sculpin in Colorado (and throughout the basin) are not *C. bairdii* (McPhail 2007; Young et al. 2013, Young et al. 2022).

The second species of sculpin recognized from Colorado, *C. annae*, was originally described from individuals collected from the Eagle River near Gypsum, Colorado (Jordan 1896). With little justification, Bailey and Bond (1963) synonymized this species with the Paiute Sculpin *C. beldingii* which was originally described from Lake Tahoe, Nevada (Eigenmann and Eigenmann 1891).

The objective of this study was to use DNA barcoding and other molecular techniques to identify specimens of *Cottus* from Colorado, to evaluate divergence within and among lineages, and to assess their phylogenetic relatedness to other lineages of sculpin, especially *C. beldingii* and *C. bairdii* from near their type locations. The secondary objective was to compare lineages of sculpin in Colorado to explore any morphological or meristic difference between them.

## PROGRESS

The first phase of this project was completed in 2020 (Young et al. 2020; Young et al. 2022). The second phase of this project will began in 2022 and will continue through 2025. Phase two of the project is a cooperative study with Colorado State University and will involve exploring the morphological differences between our two sculpin species.

In the first phase of the project, Colorado Parks and Wildlife biologists and researchers sampled 262 specimens from 93 waters around the western slope of Colorado. These specimens were sent to the U.S. Forest Service National Genomics Center for Wildlife and Fish Conservation as part of a larger study of *Cottus* species across the west (Young et al. 2022).

Phylogenetic analyses based on DNA barcoding placed the Colorado specimens in two primary lineages. One lineage (referred to here as *C. punctulatus*) is currently called Mottled Sculpin *C. bairdii* but is notably divergent from that taxon. Mottled Sculpin from eastern North America was a highly supported lineage that differed substantially (mean pairwise distance, 2.1%) from a primarily western group found in the Great Basin, Colorado, and Columbia River. Pairwise distances of this size are generally indicative of differences between full species (Ward 2009). The second lineage in Colorado (referred to here as *C. annae*) was unambiguously affiliated with the *C. beldingii* species complex, particularly those in Nevada, Idaho, Utah, and Wyoming, but was divergent from *C. beldingii*. The Colorado member of the Paiute Sculpin group was found to be geographically discrete, genetically divergent, and monophyletic and is likely and unique species endemic to Colorado.

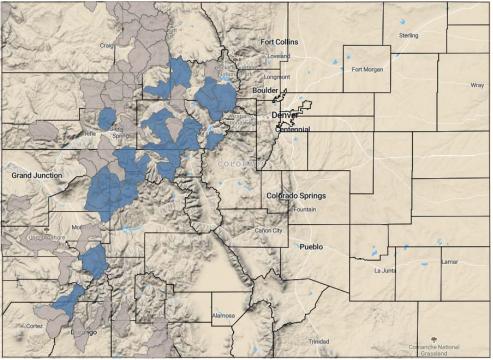
Specimens of *C. punctulatus* were more widely distributed than those from *C. annae* in Colorado (Figures 6 and 7). The fish previously referred to as Mottled Sculpin, now thought to be *C. punctulatus*, were found in every river basin in western Colorado that was a tributary to the Colorado River. In contrast, *C. annae* was not found in samples from the San Juan and Green River basins in Northern Colorado, implying that the extent of its range was the Colorado River basin above the mouth of the Dolores River. It is currently unknown if the range extends to parts of the Dolores River basin in Utah on the eastern side of the La Sal Mountains, but *C. punctulatus* was present in La Sal Creek near Paradox, Colorado.

The two sculpin lineages were found to be sympatric in the main-stem Dolores River, Dallas Creek (Gunnison River basin), the Eagle River, and the Crystal River. The co-occurrence of these taxa has been reported before; Jordan (1896) noted that *C. bairdii punctulatus* was abundant at the type location for *C. annae*. More recently, Shiozawa et al. (2010) detected both groups in samples from the Frying Pan River.

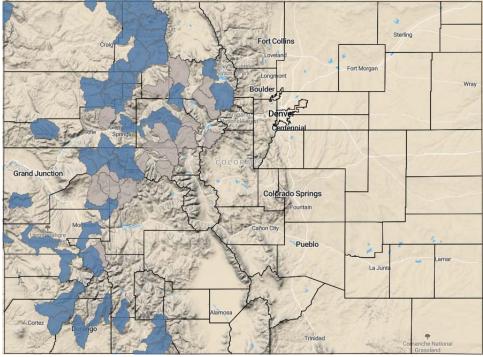
Interestingly, the distribution of *C. annae* is equivalent to that of the "green" lineage of Colorado River cutthroat trout *Oncorhynchus clarki pleuriticus* and the range *C. punctulatus* is the same as "blue" lineage of Colorado River cutthroat trout (Bestgen et al. 2019). Because these species complexes share similar ranges, their distribution implies that *C. annae* and "green" lineage cutthroats may have established in Colorado at a similar place and time, in a way that differed from *C. punctulatus* and "blue" lineage cutthroats.

Overall, these results demonstrate that there are two distinct lineages of sculpins in Colorado and they are different from their current identification. We conclude that these findings can form a basis for resurrecting the names Eagle River Sculpin *C. annae* and Colorado Sculpin *C. punctulatus* for the sculpins of Colorado, and for adding to the recognized diversity of aquatic species in the West.

The second phase of this project began in July of 2023 and field collections will continue through 2024. The project is a collaboration between CPW and Kevin Bestgen and Matthew Haworth of Larval Fish Laboratory at Colorado State University. The objective of this project is to provide a morphological description of two provisional sculpin species in Colorado. A principle outcome of the study will be an improved understanding of morphological differences between the two taxa, and whether they are useful to differentiate taxa, is required to aid effective management and conservation efforts. The project is expected to be completed by July 2026.



**Figure 6.** Distribution of *C. annae* (formerly thought to be Paiute Sculpin) in Colorado. Map created by A. Treble, CPW.



**Figure 7.** Distribution of *C. punctulatus* (formerly thought to be Mottled Sculpin) in Colorado. Map created by A. Treble, CPW.

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## **RESEARCH PRIORITY**

Diet habits of Rainbow and Brown Trout in Upper Colorado River basin recreational tailwater fisheries.

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This draft report is distributed solely for the purposes of scientific information exchange and review, results are preliminary and may change in the future. Because the report has not yet been approved for publication by the U.S. Geological Survey (USGS), it does not represent any official USGS finding or policy.

## **OBJECTIVE**

The overarching objective of this study is to understand and predict how salmonids will respond to climate change and future water storage decisions in Colorado River Basin tailwaters. The specific objective of this part of the project to document the diets of Rainbow Trout *Oncorhynchus mykiss* and Brown Trout *Salmo trutta* in six tailwater trout fisheries.

## **INTRODUCTION**

Climate change and increasing water use are altering river flow and temperature regimes around the world (Milly et al. 2005; van Vliet et al. 2013). In the western United States, increasing air temperatures (Westerling et al. 2006), changing precipitation and water runoff patterns (Mote et al. 2018; Williams et al. 2020), and increasing water demands (MacDonald 2014) are synergistically impacting river flows and temperatures (Barnett et al. 2008; Dettinger et al. 2015, Regonda et al. 2015) with resulting impacts on biological communities, ecosystem processes, and valuable goods and services for humans (Ruhi et al. 2016; Palmer and Ruhi 2019). Such changes are particularly notable throughout the Colorado River Basin (CRB), where unprecedented warm temperatures, extreme drought, and increasing demands for water are threatening naturally and economically important resources (Christensen et al. 2004; Udall and Overpeck 2017; Milly and Dunne 2020; Miller et al. 2021; Williams et al. 2022). Consequently, watershed managers in the CRB face the difficult task of sustainably balancing both human and natural ecosystem needs in a rapidly changing world.

River regulation and diversions will likely modulate the impact of changing flows and temperatures on the structure and function of ecosystems. Dams represent some of the most pervasive effects of humans on river ecosystems, and, among other things, their construction has resulted in highly altered and homogenized downstream environments. These perturbations, in

turn, cause shifts in both biological communities and ecosystem processes (Park et al. 2003; Johnson et al. 2008; Cross et al. 2011; Cross et al. 2013). Despite many negative impacts, dams often simultaneously benefit a subset of species. For instance, modified sections of river immediately downstream of dams (hereafter "tailwaters") can provide ideal conditions to support economically and recreationally important salmonid fisheries (Plummer et al. 2010). Rainbow Trout *Oncorhynchus mykiss* and Brown Trout *Salmo trutta*, two popular sport fishes, thrive in tailwaters throughout the CRB, owing to cool and clear water, stable flows and temperatures, and high production of algal and invertebrate resources. However, CRB tailwaters are not immune to ongoing changes in river flows and temperatures, and the quality of these highly valued fisheries face an uncertain future.

The overarching objective of our study is to understand and predict how salmonids will respond to climate change and future water storage decisions in CRB tailwaters. To address this, we will first utilize a drift-foraging bioenergetics modeling approach, that has previously been applied in the tailwater below Glen Canyon Dam (Dodrill et al. 2016), to estimate growth and maximum size of trout in tailwaters across the region (Figure 8A). This approach requires empirically collected estimates of water temperature, turbidity, flow, and drifting prey items, and will allow us to explicitly examine the influence of both warming and food availability on salmonid populations in a mechanistic framework. Next, we plan to model statistical relationships between rising air temperatures, changing reservoir dynamics (i.e., elevation, storage capacity, residence time), and warming tailwater temperatures using historic data. Using these relationships, we plan to simulate how a variety of climate change and water storage scenarios may influence salmonid bioenergetics via alteration to tailwater thermal regimes and prey assemblages (Figure 8B). Finally, we plan to update and improve the original foraging-bioenergetics models of Dodrill et al. 2016 using data on prey selectivity and the feeding habits of trout in CRB tailwaters (Figure 8C).

#### **METHODS**

Ten tailwaters were selected throughout Colorado based on the availability of historic reservoir and tailwater temperature data, the presence of a nearby United State Geological Survey (USGS) gaging station, and adequate fish population data (Figure 9A). Although all tailwater sites were located downstream of thermally-stratified reservoirs with hypolimnetic dam releases, the ten study sites incorporated a range a thermal and hydrological variability (Figure 9B). A graphical illustration of spatial and temporal aspects of our physicochemical and biological sampling is provided in Figure 9C.

Instantaneous discharge and reservoir attributes (e.g., storage, elevation, release) were obtained from nearby USGS gaging stations, the U.S. Bureau of Reclamation Hydromet System, Denver water, and the Upper Yampa Water Conservancy District. Water temperatures were measured every 30 minutes throughout the year (May 2022-May 2023) using HOBO® pendant temperature loggers that were anchored to the stream bed at two locations per tailwater; location one was immediately downstream of the dam (referred to "0 km") and location two was 3-10 km downstream from the dam (referred to "5 km"). Total (n = 18/tailwater) and dissolved (n = 18/tailwater) water nutrient samples were collected seasonally from the 0 km section of each tailwater and frozen until analysis. Dissolved nutrient samples were passed through 0.45  $\mu$ m

mixed cellulose esters membrane filters placed on filter housing. Seasonal estimates of primary producers were obtained at both 0 and 5 km locations using a BenthoTorch® (n = 30) to estimate total chl-*a in situ*, and by scrubbing a known area of rock (n = 30) with a wire brush to estimate Ash-Free Dry Mass (AFDM) of periphyton. Rock scrubs were preserved on ice prior to AFDM estimation. Water turbidity and dissolved oxygen were measured at both 0 km and 5 km locations on each sampling occasion using a handheld YSI® multiparameter sonde. Average channel width, depth, cross-sectional area, and median sediment size (Wolman pebble counts) were estimated at both 0 km and 5 km locations during base flows in August 2022.

Seasonal drift was sampled at both 0 km and 5 km sections of each tailwater throughout 2022-2023. Briefly, we submerged three drift nets (250  $\mu$ m mesh size) on anchored rebar posts for 5 minutes (Figure 10). The net contents were then rinsed into a 250  $\mu$ m mesh sieve and preserved in heat-sealed bags with 95% ethanol and an internal label. All drift samples were collected early to mid-day. Water velocities were measured at each net location using a handheld Marsh McBirney® velocity meter to estimate the volume of water filtered over the 5-minute drift set. On rare occasions when nets are not fully submerged, the area of net exposed was measured and factored into our estimates of volume of water filtered.

At six tailwater locations (see Figure 9A) we collected fishes via electrofishing (backpack, raft, or barge; see Table 5 and Figure 10) for diet analysis. These sampling events occurred in the fall of 2022 and the spring of 2023. All fishes were measured and weighed, and diets were obtained using non-lethal gastric lavage techniques (Stone 2004), sieved through either 63 or 250  $\mu$ m in the field, and preserved in ethanol until laboratory processing.

#### Laboratory and analysis

In the laboratory, drift samples were rinsed through stacked sieves to separate coarse (>1mm) and fine (<1mm >250µm) size classes. Invertebrates in the two size classes were removed from other organic material under a dissecting microscope. Although all coarse samples were picked in full, many fine samples contained large numbers (>500) of invertebrates and were thus subsampled using a Folsom plankton wheel. Following picking and subsampling, invertebrates were identified to the lowest practical taxonomic level, in most cases family and genus, using Merritt et al. (2008) and Smith (2001), enumerated, and measured to the nearest mm to estimate biomass (mg Dry Mass) using published length to mass regressions (Benke et al. 1999). For prohibitively large samples, the first 30 individuals of each taxon were measured, and individuals counted but not measured were assumed to have the same size distribution. All estimates of invertebrate abundance and biomass in the drift were standardized to individuals/m<sup>3</sup> or mg/m<sup>3</sup> prior to analysis. Fish diets were processed similarly to the methods described for invertebrates above; however, due to lower number of diet items they were not initially split into coarse and fine size classes. All diet prey items were estimated as a proportion of total abundance (N) or biomass (B) and averaged across individual fish at a location to examine differences in diet composition among sites and between upstream (i.e., "0 km") and downstream (5 km) locations within a site. Sample QA/QC was performed by the USGS Quality Systems Branch to ensure accurate sample processing and identification.

Estimates of total invertebrate abundance (individuals/m<sup>3</sup>), biomass (mg DM/m<sup>3</sup>), mean individual body size (mg; e.g., total biomass/total abundance), species richness (number of

unique taxa), and abundance of EPT (Orders Ephemeroptera, Plecoptera, Trichoptera; numbers/m<sup>3</sup>) in the drift were compared among study sites and between upstream and downstream locations within a site. Additionally, we explored the potential for salmonid prey selectivity by first comparing sizes of invertebrate prey items captured in the drift to sizes of prey items collected in fish diets. To do this, we estimated the average proportion of drifting and diet taxa in different length class categories (1mm size bins) regardless of identity and compared the degree of overlap between the two distributions. High overlap among drift and diet size distributions suggests little size selection, whereas deviations highlight selectivity for certain size prey items. Next, we used the linear food selection index L = r - p, where r = the relative abundance of a given taxa in the diet and p = the relative abundance of the same taxa in the environment, to examine whether fish selected prey items based on their taxonomic identity (Strauss 1979; Chipps and Garvey 2006). Values of *L* close to zero indicate no selectivity for a given prey item, whereas negative and positive values represent negative and positive selection, respectively. For these analyses we focused on a subset of sites (Taylor and Colorado River) during the fall of 2022. All analyses were conducted in R (R Core Team 2022).

#### PRELIMINARY RESULTS

We collected a total of 339 drift samples between May 2022 and May 2023. Although sample processing is ongoing, preliminary analysis suggests that total invertebrate abundance and biomass varies considerably among study sites and between upstream and downstream locations within a tailwater (Figure 11 top). Small-bodied zooplankton often comprise the bulk of total drifting invertebrate abundance and biomass directly below dams (Figure 12); however, invertebrate body size, richness, and number of EPT taxa generally increase with increasing distance downstream from dams (Figure 11 bottom).

During October 2022 and May 2023, we collected 559 fish for diet quantification (Table 5). Although preliminary (n = 19 Taylor, n = 23 Colorado), diet analysis highlights some key trends in prey use and selectivity in CRB tailwaters. First, similar to patterns observed in the drift, diets showed a general increase in diversity of prey items, including incorporating more prey from the terrestrial environment, with increasing distance downstream from dams (23 vs 27 prey items Colorado River, 17 vs 25 prey items Taylor River; Figure 13). Additionally, fish tended to consume a greater proportion of reservoir-derived prey (*Mysis*, Cladocera, Copepoda) in the more depauperate areas directly downstream of dams. These patterns were surprising and unexpected given the extremely small sizes (1-2 mm) of some zooplankton taxa. Finally, we found evidence of prey selectivity when comparing diets to drift estimates within a location, however such patterns differed by tailwater. For example, although we found lower evidence for prey selection in the Taylor River, our results demonstrate that Rainbow Trout selected for larger-bodied (8-19 mm) *Mysis* directly below Granby Reservoir on the upper Colorado River and Brown Trout selected for Isopods further downstream (Figure 13).

Currently, we are working in the laboratory to finish processing and identifying all drift and diet samples collected throughout 2022-2023. Upon completion, we will use these data to update and improve drift-foraging bioenergetic models (Figure 8) and ultimately predict the influence of temperature and food availability on current and future salmonid populations in CRB tailwaters.

### CONCLUSIONS

Changes in reservoir dynamics are already resulting in warmer tailwater temperatures in parts of the Colorado River basin. Although these changes may negatively impact salmonid populations via direct and isolated effects on individual physiology (see references in Richter and Kolmes 2005), food web dynamics and ecosystem productivity likely modify such responses (O'Gorman et al. 2016; Hocking et al. 2021; Schwartz and Warren 2022; Warren et al. 2022; Solokas et al. 2023). For example, tailwater populations that rely heavily on reservoir-derived prey and/or have a depauperate food base (Figure 15A) may be less resilient and more sensitive to future perturbations than populations that are fueled by more complex and stable food webs (Figure 15B; McCann 2007; Rooney and McCann 2012). Further, prey resources that increase in productivity with rising temperatures may buffer against the negative impacts of warming, whereas resources that decline may exacerbate responses. By providing insights into how changes in temperature and food availability interact to influence salmonid populations, our approach will provide crucial information for evaluating current dam operations, ongoing drought impacts, and mitigating the future impact of climate change on important tailwater fisheries in Colorado.

| <b>Table 5.</b> Summaries of fish collected for diet analysis in the upper Colorado River Basin in 2022-2023. RBT = Rainbow Trout, BT = |  |
|---|--|
| Brown Trout   |  |

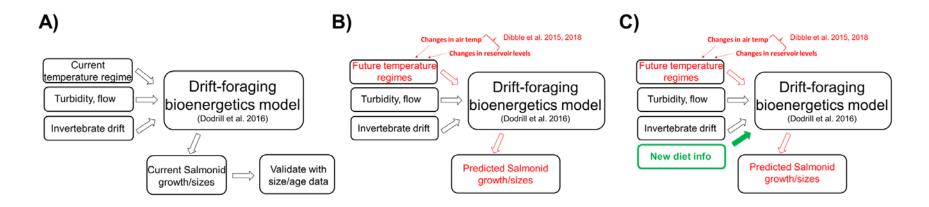
| River     | Dam            | Location   | Season      | Lat/Lon              | Species | Method          | # fish <<br>250mm | # fish ><br>250mm |
|-----------|----------------|------------|-------------|----------------------|---------|-----------------|-------------------|-------------------|
| Blue      | Green Mountain | Upstream   | Fall 2022   | 39.88167, -106.33446 | BT      | Raft e-fish     | 4                 | 10                |
| Blue      | Green Mountain | Upstream   | Fall 2022   | 39.88167, -106.33446 | RBT     | Raft e-fish     | 1                 | 3                 |
| Blue      | Green Mountain | Downstream | Fall 2022   | 39.95640, -106.35661 | BT      | Raft e-fish     | 3                 | 7                 |
| Blue      | Green Mountain | Downstream | Fall 2022   | 39.95640, -106.35661 | RBT     | Raft e-fish     | 2                 | 7                 |
| Blue      | Green Mountain | Upstream   | Spring 2023 | 39.88167, -106.33446 | BT      | Backpack e-fish | 7                 | 15                |
| Blue      | Green Mountain | Upstream   | Spring 2023 | 39.88167, -106.33446 | RBT     | Backpack e-fish | 6                 | 10                |
| Blue      | Green Mountain | Downstream | Spring 2023 | 39.95640, -106.35661 | BT      | Backpack e-fish | 5                 | 17                |
| Blue      | Green Mountain | Downstream | Spring 2023 | 39.95640, -106.35661 | RBT     | Backpack e-fish | 0                 | 4                 |
| Colorado  | Granby         | Upstream   | Fall 2022   | 40.14547, -105.86788 | BT      | Backpack e-fish | 13                | 5                 |
| Colorado  | Granby         | Upstream   | Fall 2022   | 40.14547, -105.86788 | RBT     | Backpack e-fish | 6                 | 8                 |
| Colorado  | Granby         | Downstream | Fall 2022   | 40.10697, -105.95590 | BT      | Backpack e-fish | 12                | 6                 |
| Colorado  | Granby         | Downstream | Fall 2022   | 40.10697, -105.95590 | RBT     | Backpack e-fish | 1                 | 3                 |
| Colorado  | Granby         | Upstream   | Spring 2023 | 40.14547, -105.86788 | BT      | Backpack e-fish | 10                | 11                |
| Colorado  | Granby         | Upstream   | Spring 2023 | 40.14547, -105.86788 | RBT     | Backpack e-fish | 1                 | 7                 |
| Colorado  | Granby         | Downstream | Spring 2023 | 40.10697, -105.95590 | BT      | Backpack e-fish | 7                 | 3                 |
| Colorado  | Granby         | Downstream | Spring 2023 | 40.10697, -105.95590 | RBT     | Backpack e-fish | 1                 | 0                 |
| Dolores   | McPhee         | Upstream   | Fall 2022   | 37.57711, -108.58435 | BT      | Barge e-fish    | 9                 | 10                |
| Dolores   | McPhee         | Upstream   | Fall 2022   | 37.57711, -108.58435 | RBT     | Barge e-fish    | 0                 | 3                 |
| Dolores   | McPhee         | Downstream | Fall 2022   | 37.60099, -108.61961 | BT      | Barge e-fish    | 2                 | 0                 |
| Dolores   | McPhee         | Downstream | Fall 2022   | 37.60099, -108.61961 | RBT     | Barge e-fish    | 0                 | 0                 |
| Fryingpan | Ruedi          | Upstream   | Fall 2022   | 39.36535, -106.82607 | BT      | Backpack e-fish | 6                 | 19                |
| Fryingpan | Ruedi          | Upstream   | Fall 2022   | 39.36535, -106.82607 | RBT     | Backpack e-fish | 6                 | 10                |
| Fryingpan | Ruedi          | Downstream | Fall 2022   | 39.36698, -106.84960 | BT      | Backpack e-fish | 22                | 14                |

### Table 5 cont.

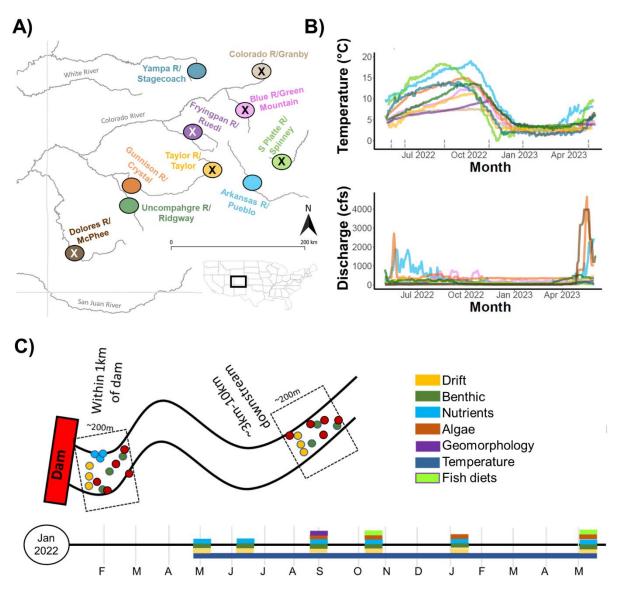
| River        | Dam         | Location   | Season      | Lat/Lon              | Species | Method          | # fish <<br>250mm | # fish ><br>250mm |
|--------------|-------------|------------|-------------|----------------------|---------|-----------------|-------------------|-------------------|
| Fryingpan    | Ruedi       | Downstream | Fall 2022   | 39.36698, -106.84960 | RBT     | Backpack e-fish | 1                 | 5                 |
| Fryingpan    | Ruedi       | Upstream   | Spring 2023 | 39.36535, -106.82607 | BT      | Backpack e-fish | 7                 | 9                 |
| Fryingpan    | Ruedi       | Upstream   | Spring 2023 | 39.36535, -106.82607 | RBT     | Backpack e-fish | 1                 | 8                 |
| Fryingpan    | Ruedi       | Downstream | Spring 2023 | 39.36698, -106.84960 | BT      | Backpack e-fish | 22                | 9                 |
| Fryingpan    | Ruedi       | Downstream | Spring 2023 | 39.36698, -106.84960 | RBT     | Backpack e-fish | 3                 | 2                 |
| South Platte | Spinney     | Upstream   | Fall 2022   | 38.97162, -105.61393 | RBT     | Backpack e-fish | 10                | 11                |
| South Platte | Spinney     | Downstream | Fall 2022   | 38.96752, -105.58115 | BT      | Backpack e-fish | 0                 | 0                 |
| South Platte | Spinney     | Downstream | Fall 2022   | 38.96752, -105.58115 | RBT     | Backpack e-fish | 0                 | 0                 |
| South Platte | Spinney     | Upstream   | Spring 2023 | 38.97162, -105.61393 | BT      | Backpack e-fish | 13                | 1                 |
| South Platte | Spinney     | Upstream   | Spring 2023 | 38.97162, -105.61393 | RBT     | Backpack e-fish | 7                 | 2                 |
| South Platte | Spinney     | Downstream | Spring 2023 | 38.96752, -105.58115 | BT      | Backpack e-fish | 5                 | 0                 |
| South Platte | Spinney     | Downstream | Spring 2023 | 38.96752, -105.58115 | RBT     | Backpack e-fish | 0                 | 0                 |
| Taylor       | Taylor Park | Upstream   | Fall 2022   | 38.81597, -106.61114 | BT      | Barge e-fish    | 5                 | 8                 |
| Taylor       | Taylor Park | Upstream   | Fall 2022   | 38.81597, -106.61114 | RBT     | Barge e-fish    | 4                 | 14                |
| Taylor       | Taylor Park | Downstream | Fall 2022   | 38.77672, -106.63350 | BT      | Barge e-fish    | 9                 | 9                 |
| Taylor       | Taylor Park | Downstream | Fall 2022   | 38.77672, -106.63350 | RBT     | Barge e-fish    | 2                 | 3                 |
| Taylor       | Taylor Park | Upstream   | Spring 2023 | 38.81597, -106.61114 | BT      | Backpack e-fish | 3                 | 11                |
| Taylor       | Taylor Park | Upstream   | Spring 2023 | 38.81597, -106.61114 | RBT     | Backpack e-fish | 1                 | 11                |
| Taylor       | Taylor Park | Downstream | Spring 2023 | 38.77672, -106.63350 | BT      | Backpack e-fish | 4                 | 6                 |
| Taylor       | Taylor Park | Downstream | Spring 2023 | 38.77672, -106.63350 | RBT     | Backpack e-fish | 0                 | 0                 |

TOTAL DIETS

521



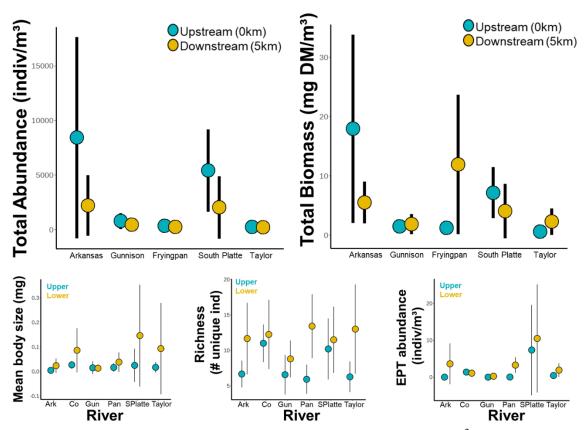
**Figure 8.** Our study relies on empirically collected estimates of water temperature, turbidity, flow, and invertebrate drift to estimate salmonid growth and sizes using a previously developed drift-foraging bioenergetics model (Dodrill et al. 2016; (A). Once this model is calibrated to current conditions, we plan to simulate a variety of water storage and tailwater warming scenarios to explore how such changes may impact future salmonid growth (B). We plan to utilize fish diet information to further inform and improve our modeling approach as the original foraging-bioenergetics model does not currently incorporate prey selectivity (C).



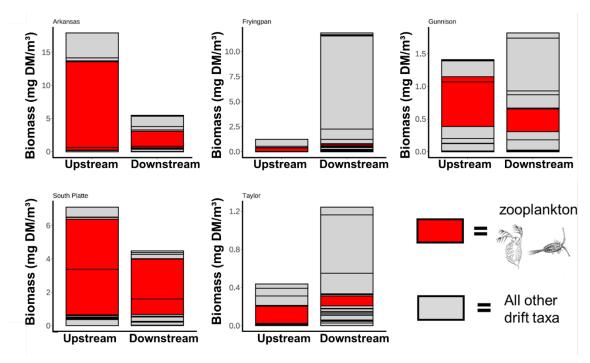
**Figure 9.** Ten tailwater sites that vary in water temperature and hydrology were selected for biological and physicochemical sampling from May 2022-May 2023 (**A and B**). On each occasion we collected samples from both upstream ("0 km" in text) and downstream ("5 km" in text) reaches that were roughly 200m in length. The dots included in this figure represent samples collected on every sampling trip. (**C**). Six tailwaters were also selected to collect fish diets (indicated by X in A) in October 2022 and May 2023.



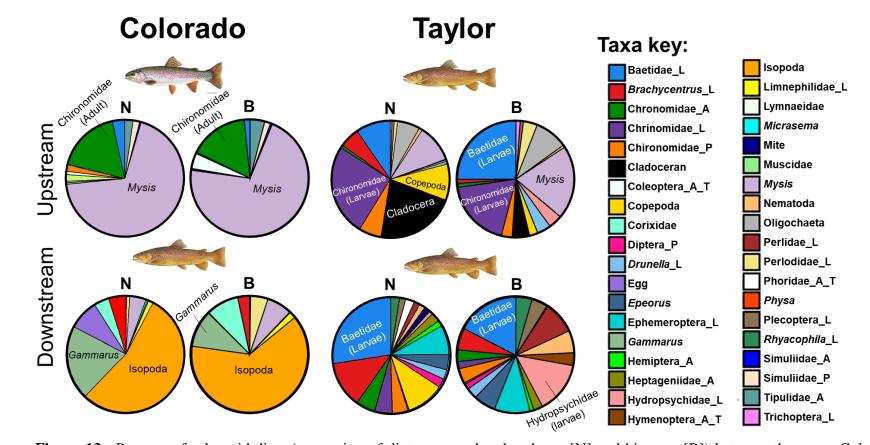
**Figure 10.** Drifting invertebrate prey items were collected using 250  $\mu$ m mesh nets deployed in the water column for five minutes at two locations (upstream 0 km, downstream 5 km) at each tailwater site (lower right). Fish were captured at similar locations in October 2022 and May 2023 using a variety of electrofishing techniques (top), and their diets were collected using non-lethal gastric lavaging (lower left) and analyzed in the laboratory.



**Figure 11.** Patterns of total drifting invertebrate abundance (numbers/m<sup>3</sup>), biomass (mg DM/m<sup>3</sup>), mean individual body size (mg; total biomass/total abundance), species richness (number of unique taxa), and abundance of EPT (Orders Ephemeroptera, Plecoptera, Trichoptera; numbers/m<sup>3</sup>) among tailwaters and between upstream and downstream locations within tailwater sites. Circles represent mean values and black bars indicate standard deviations.



**Figure 12.** Proportions of total invertebrate drift biomass comprised of zooplankton taxa (Cladocera and Copepoda; highlighted in red) among tailwater sites and between upstream and downstream locations within tailwater sites.



**Figure 13.** Patterns of salmonid diets (proportion of diet contents by abundance [N] and biomass [B]) between the upper Colorado and Taylor River and between upstream (0 km) and downstream (5 km) locations within each tailwater. For this analysis, diets were analyzed from Brown Trout in the Taylor River (n = 9 upstream, 10 downstream) and the downstream section of the Colorado River (n = 12), and Rainbow Trout (n = 11) in the upstream section of the Colorado River. L = larvae, P = pupae, A = adult, T = terrestrial.

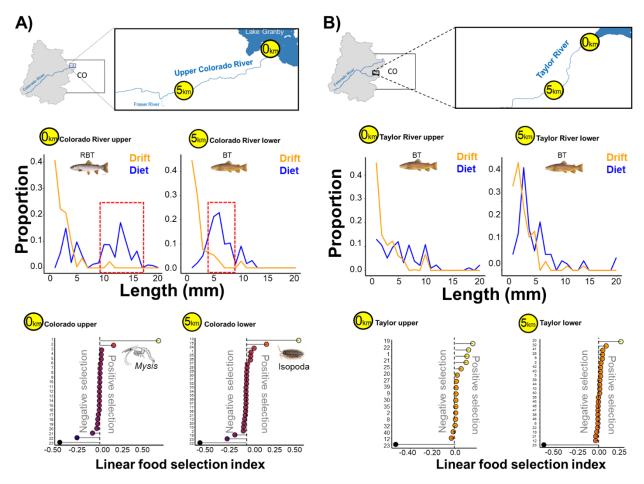
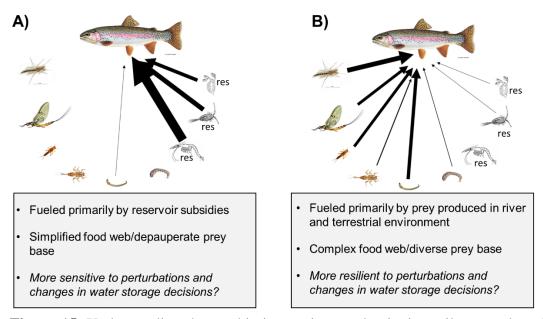


Figure 14. Preliminary data highlights the potential for salmonid prey selectivity in upper Colorado River basin tailwaters. For example, both Rainbow and Brown Trout in the upper Colorado River (A) consumed more larger-bodied Mysis (upstream) and Isopods (downstream) than what was captured in drift nets (indicated by the red dashed boxes in middle and selectivity indices on bottom). Interestingly, the degree of prey selectivity may differ by tailwater, as we found less evidence of selectivity in the Taylor River (B). Taxa in lower panel are 1) Mysis, 2) Chironomidae\_A, 3) Tipulidae\_A, 4) Simuliidae\_P, 5) Diptera\_P, 6) Coleoptera\_A, 7) Simuliidae\_A, 8) Muscomorpha 9), Chironomidae\_P, 10) Hemiptera\_T, 11) Ceratopogonidae\_P, 12) Acari, 13) Isopoda, 14) Sciaroidea, 15) Gammaridae, 16) Ceratopogonidae\_A, 17) Simuliidae\_L, 18) Planaria, 19) Cladocera, 20) Baetidae\_L, 21) Chironomidae\_L, 22) Oligochaeta, 23) Copepoda, 24) Egg, 25) Brachycentridae\_L, 26) Corixidae, 27) Perlodidae\_L, 28) Micrasema, 29) Lymnaeidae, 30) Trichoptera\_L, 31) Physa, 32) Ephemeroptera\_L, 33) 35) Hydropsychidae\_L, 36) Arachnida\_T, 34) Nematoda, Ephemerellidae\_L, 37) Chironomidae\_P, 38) Optioservus\_L, 39) Plecoptera\_L, 40) Nematocera\_A, 41) Epeorus\_L, 42), Phoridae\_A, 43) Heptageniidae\_A, 44) Perlidae\_L, 45) Hymenoptera\_A, 46) Rhyacophila\_L, 47) Heptageniidae\_L, 48) Elmidae\_L. L = larvae, P = pupae, A = adult, T = Terrestrial



**Figure 15.** Understanding the trophic interactions underpinning tailwater salmonid populations may be crucial for conserving and managing these important fisheries in the future. Tailwater populations that rely heavily on reservoir-derived prey or have a depauperate food base (**A**) may be less resilient and more sensitive to future perturbations than populations that are fueled by more complex and stable food webs. (**B**) Res = prey subsidies produced in upstream reservoirs.

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#### **RESEARCH PRIORITY**

**Technical Assistance** 

#### **OBJECTIVE**

Provide technical assistance to biologists, managers, researchers, and other internal and external stakeholders as needed in a variety of coldwater ecology applications.

#### **INTRODUCTION**

Aquatic researchers and aquatic biologist work closely to investigate and manage the aquatic resources of Colorado. The purpose is to cooperate closely with biologist and other stakeholders to disseminate results from aquatic research projects and to conduct meaningful research that addresses management needs.

Fishery managers, hatchery personnel, administrators, and CPW Field Operations personnel often need fishery ecology information or technical consulting on specific projects. Effective communication between researchers, fishery managers and other internal and external stakeholders is essential to the management coldwater stream fisheries in Colorado. Technical assistance projects are often unplanned and are addressed on an as-needed basis.

#### ACCOMPLISHMENTS

One new technical assistance project was started last year; the Cow Creek Pre-impoundment Study. The objective of this project was to collect baseline aquatic invertebrate data on Cow Creek in southwestern Colorado. A new mainstream dam is being proposed this small tributary to the Uncompahgre River between Ridgway and Montrose, Colorado and a proposed pipeline below the dam could drastically alter flows in the lower river. Three sites on Cow Creek were sampled above and below the proposed reservoir site as well as two sites on the Uncompahgre River above and below the Cow Creek confluence. This technical assistance project was submitted in 2023 through CPW Aquatic Research project selection process for evaluation as an ongoing project.

Technical assistance was also provided to whirling disease sampling efforts. A statewide sampling effort is being done to compare whirling disease in trout to historical collects from 20-30 years ago. Waters sampled included the Gunnison River, the Colorado River, Rito Hondo Creek, Cebolla Creek, Spring Creek, and the Taylor River.

Technical assistance was provided to Ben Masters and Ryan Olinger of Fin & Fur Films (<u>https://www.finandfurfilms.com/</u>). They are making a natural history film about the Colorado River and its wildlife and were interested in filming the Salmonfly emergence. Background information about the species and its emergence was provided as well as field assistance to see

and film the emergence in Little Gore Canyon and to collect larvae for filming the emergence in captivity. The feature film is expect to be leased in 2024.

## COMMUNICATION AND INFORMATION TRANSFER

Two reports were produced to summarize and disseminate information from the coldwater stream ecology research projects;

Kowalski, D. A. 2022. Coldwater stream ecology investigations project summary. Colorado Parks and Wildlife, Aquatic Wildlife Research Section. Fort Collins, Colorado.

Kowalski, D. A., R. J. Cordes, T. B. Riepe, J. D. Drennan, A. J. Treble. 2022. Prevalence and distribution of *Renibacterium salmoninarum*, causative agent of bacterial kidney disease, in wild trout fisheries in Colorado. Pages 151-157 in the Proceedings of Wild Trout Symposium XIII: Reducing the Gap between Science and Public Opinion. Available at: <a href="https://www.wildtroutsymposium.com/Proceedings\_13.pdf">https://www.wildtroutsymposium.com/Proceedings\_13.pdf</a>.

Two external presentations were contributed to for dissemination of results of aquatic ecology projects to colleagues and other fishery professionals:

Kowalski, D.A., R.J. Cordes, T.B. Riepe, J.D. Drennan, A.J. Treble. 2022. Prevalence and distribution of *Renibacterium salmoninarum*, causative agent of bacterial kidney disease, in wild trout fisheries in Colorado. Wild Trout Symposium VIII. September 29, 2022. West Yellowstone, MT.

Kowalski, D.A. and E. Gardunio. 2023. Evaluation of an electric fish barrier on the South Canal of the Gunnison River, Colorado. Electric Power Research Institute Webinar, February 14, 2023.

Kowalski, D.A. 2023. Salmonflies (*Pteronarcys californica*) of the Gunnison River. Colorado Mesa University. May 3, 2023. Grand Junction, CO.

Kowalski, D.A. and E.E. Richer. 2022. Salmonflies (*Pteronarcys californica*) of the Gunnison River. Colorado Canyons Association. November 8, 2022. Montrose, CO. https://www.youtube.com/watch?v=hEfKIywX8AY.