# Colorado River Aquatic Resource Investigations 

Dan A. Kowalski<br>Aquatic Research Scientist



Job Progress Report
Colorado Parks \& Wildlife

Aquatic Research Section

Fort Collins, Colorado

August 2017

# STATE OF COLORADO 

John W. Hickenlooper, Governor

# COLORADO DEPARTMENT OF NATURAL RESOURCES 

Bob Randall, Executive Director
COLORADO PARKS \& WILDLIFE
Bob Broscheid, Director
WILDLIFE COMMISSION

John V. Howard, Vice-Chair Marvin McDaniel<br>Michelle Zimmerman, Secretary<br>Robert W. Bray<br>Marie Haskett<br>Carrie Hauser<br>Alexander Zipp<br>Robert "Dean" Wingfield<br>James Vigil<br>Dale E. Pizel<br>Jim Spehar

Ex Officio/Non-Voting Members: Don Brown, Bob Randall and Bob Broscheid
AQUATIC RESEARCH STAFF
George J. Schisler, Aquatic Research Leader
Kelly Carlson, Aquatic Research Program Assistant
Peter Cadmus, Aquatic Research Scientist/Toxicologist, Water Pollution Studies
Eric R. Fetherman, Aquatic Research Scientist, Salmonid Disease Studies
Ryan Fitzpatrick, Aquatic Research Scientist, Eastern Plains Native Fishes
Eric E. Richer, Aquatic Research Scientist/Hydrologist, Stream Habitat Restoration
Matthew C. Kondratieff, Aquatic Research Scientist, Stream Habitat Restoration
Dan Kowalski, Aquatic Research Scientist, Stream \& River Ecology
Adam G. Hansen, Aquatic Research Scientist, Coldwater Lakes and Reservoirs
Kevin B. Rogers, Aquatic Research Scientist, Colorado Cutthroat Studies
Kevin G. Thompson, Aquatic Research Scientist, 3-Species and Boreal Toad Studies
Andrew J. Treble, Aquatic Research Scientist, Aquatic Data Management and Analysis
Brad Neuschwanger, Hatchery Manager, Fish Research Hatchery
Tracy Davis, Hatchery Technician, Fish Research Hatchery
Christopher Praamsma, Hatchery Technician, Fish Research Hatchery
Jim Guthrie, Federal Aid Coordinator
Kay Knudsen, Librarian

Prepared by:


Dan Kowalski, Aquatic Research Scientist

Approved by:


Date: $\operatorname{Ang} 30,2017$

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
Job No. 1. Salmonfly Habitat and Ecology Studies ..... 1
Job No. 2. Impacts of Whitewater Park Development on Invertebrates, Mottled Sculpin and Trout .....  8
Job No. 3. Colorado River Water Project Mitigation and Ecology Investigations ..... 25
Job No. 4: Bacterial Kidney Disease Investigations ..... 27
Job No. 5. Gunnison Tunnel Electric Fish Guidance System Evaluation ..... 30
Job No. 6. Technical Assistance ..... 31
Appendix 1. South Canal Electric Fish Barrier Final Report ..... 33

Project Title: Coldwater Stream Ecology Investigations
Period Covered: July 1, 2016 through June 30, 2017
Purpose: Improve aquatic habitat conditions and angling recreation in Colorado.
Project Objective: Investigate biological and ecological factors impacting sport fish populations in coldwater streams and rivers in Colorado.

## Job No. 1. Salmonfly Habitat and Ecology Studies

Job Objective: Investigate the habitat use, hatching ecology and limiting factors of the Salmonfly Pteronarcys californica in Colorado Rivers.

## Need

The salmonfly (Pteronarcys californica) is a large aquatic invertebrate that can reach high densities in some Colorado Rivers. They play an important ecological role as grazers in stream systems and have been documented to be extremely important to stream dwelling trout as a food resource. Nehring (1987) reported in a diet study of trout in the Colorado River that $P$. californica was the most common food item, comprising 64-75\% of the mean stomach content over the four-year study. Because of their high biomass and hatching behavior, they also play an important role in supplementing terrestrial food webs and riparian communities with stream derived nutrients (Baxter et al. 2005, Walters et al. in press). While ecologically important and found in high abundance at some sites, the salmonfly has relatively specific environmental requirements and is considered intolerant of disturbance in bioassessment protocols (Barbour et al. 1999, Fore et al. 1996, Erickson 1983).

Salmonflies are sensitive to habitat alterations in part because of their lifespan; they are one of the longest lived aquatic insects in the Neararctic (DeWalt and Stewart 1995). Salmonflies have been reported to have a three to five-year life cycle but two studies indicate it is likely to have a three or four-year life cycle in Colorado (DeWalt and Stewart 1995, Nehring 1987). These two studies also identify P. californica as one of the most synchronously emerging of all species of stoneflies with emergence at any one site lasting from 5-13 days. The synchronous emergence and hatching behavior allow it to be sampled in unique ways compared to other aquatic invertebrates. Salmonflies hatch at night by crawling from the water onto riparian vegetation and other vertical structures such as rocks, cliff faces and bridge abutments where they emerge from the nymphal exuvia which is left attached to the structure. If sites are visited soon after emergence, then the density of stoneflies emerging at a site can be estimated by completing multiple pass removal surveys of the exuvia. Nehring (2011) found a 0.95 correlation coefficient between post emergence exuvia density estimates and more traditional pre-emergent quantitative benthic sampling at 23 sites.

Previous work completed under Project F-237 indicates that the range and density of $P$. californica have declined in the Colorado River and that these declines may be associated with flow alterations (Nehring 2011). Once common in the upper Colorado River, the abundance of salmonflies has declined, especially below Windy Gap Reservoir where flow alterations associated with trans-mountain water diversions are the largest. The objective of this project is to document the distribution, density and habitat use of $P$. californica in several rivers and measure environmental variables that may be limiting factors of this species in Colorado rivers. By comparing the habitat characteristics of similar sites with differing densities of stoneflies, the optimal habitat characteristics and limiting factors will be identified. Knowledge of the preferred habitat characteristics will assist in ecological restoration of sites where P. californica have been extirpated. Once limiting habitat features are identified, the effects of flow and sediment changes on those features will be investigated. This information will benefit management and river restoration activities as well as the evaluation of re-introduction sites for $P$. californica such as those attempted on the Arkansas and upper Gunnison Rivers.

## Objectives

1. Document the distribution and density of $P$. californica at 18 sites on the Gunnison, Colorado and Rio Grande rivers.
2. Measure physical habitat variables at all 18 sites.
3. Identify the important habitat characteristics that explain their distribution and density.

## Approach

Action \#1- Develop and test population estimation techniques for salmonflies.

- Level 1 Action Category: Data collection and analysis
- Level 2 Action Strategy: Techniques development
- Level 3 Action Activity: Fish and wildlife research, survey and management techniques

Previous work under Project F-237 established that traditional sampling methods (i.e., $0.086 \mathrm{~m}^{2}$ Hess sampler) may be inadequate for accurately estimating density of salmonflies due to their patchy distribution and the large substrate they commonly occupy. Two alternative techniques that were proposed included extra large Surber type samplers and multiple pass removal estimates of the insect's exoskeleton (exuvia) post emergence. While the large Surber sampler did sample a large enough area to reduce the spatial variation of $P$. californica larvae between samples, it was time consuming to set up and use and difficult to deploy in heavy current. A $0.456 \mathrm{~m}^{2}$ Hess sampler was constructed out of $1 / 8$-inch plate steel, mimicking the design of standard Hess samplers but scaling up for the larger size. To test the new sampler, five replicate samples were taken from four sites on the Gunnison River and five sites on the Colorado River in 2016. The sites were a subset of the 18 riffle sites where exuvia density estimates have been conducted for the last eight years. All P.c. larvae in each sample will be counted, sexed and measured. All field sampling is complete for Job \#1 Action \#1. Processing of the samples is ongoing and will be reported in future Federal Aid Reports for Project F-237.

Table 1. Summary of salmonfly habitat sampling sites for Job\#1. Six sites each on three rivers were sampled over four years for exuvia density and surveyed for physical habitat characteristics.

| River | $\#$ | Site | Side | UTM NAD 83 (Zone <br> 13) |
| :---: | :---: | :--- | :--- | :---: |
| Gunnison | 1 | Orchard Boat Ramp | River Left | 247947,4295297 |
| Gunnison | 2 | Cottonwood Campground | River Left | 252129,4295940 |
| Gunnison | 3 | Goldmine | River Left | 253728,4295747 |
| Gunnison | 4 | Smith Fork | River Left | 253338,4291889 |
| Gunnison | 5 | Ute Park | River Left | 252376,4284894 |
| Gunnison | 6 | Chukar | River Left | 253421,4278775 |
| Colorado | 7 | State Bridge | River Right | 359889,4414634 |
| Colorado | 8 | Pumphouse BLM | River Left | 370827,4427300 |
| Colorado | 9 | Powers BLM | River Right | 394914,4435762 |
| Colorado | 10 | Byers Canyon | River Left | 403335,4434268 |
| Colorado | 11 | Hwy 40 Bridge | River Right | 408133,4437708 |
| Colorado | 12 | Hitching Post | River Left | 414589,4440304 |
| Rio Grande | 13 | LaGarita | River Left | 338264,4182888 |
| Rio Grande | 14 | Lower Wason 2 | River Right | 335653,4186302 |
| Rio Grande | 15 | Lower Wason 1 | River Right | 335353,4187197 |
| Rio Grande | 16 | Upper Wason 2 | River Right | 333668,4187683 |
| Rio Grande | 17 | Creede Hatchery | River Left | 332145,4187768 |
| Rio Grande | 18 | Creede Boat Ramp | River Left | 331362,4187243 |

Action \#2- Estimate the density of salmonflies at a variety of sites in the Colorado, Gunnison and Rio Grande Rivers.

- Level 1 Action Category: Data collection and analysis
- Level 2 Action Strategy: Research, survey or monitoring- fish and wildlife populations
- Level 3 Action Activity: Abundance determination

Sampling is now complete on all 18 sites on the three rivers. All sites have at least three years of data and a minimum of two years of data collected under favorable flow conditions that did not compromise the estimates. Locations and description of sites are presented in Table 1. Estimates were completed by searching 30 meter ( 98.6 ft ) sections of stream bank for $P$. californica exuvia adjacent to riffle habitat. If possible, each site was visited 2-3 times to encompass the entire emergence. If a site was visited only once, estimates were done as soon as possible after the emergence was complete (emergence usually last from 7-13 days at our study sites). Stream flow changes and weather conditions also were taken into account when planning surveys to best estimate the total emergence at each site. Three to seven people intensively searched the riparian area from one to twenty meters from the water's edge. The search area varied by site and depended on the thickness and structure of riparian vegetation. The area was extended laterally from the water's edge until no exuvia were encountered, with the exuvia at
most sites being encountered with the first 3 meters from the water. On a single sampling occasion, each area was searched two to four times with identical search areas, effort and personnel. Each exuvia on the first pass was examined to determine sex. A multiple pass removal model was used to estimate the total density of exuvia at each site (Zippin 1956). Methods were similar but not identical to previous work (Nehring 2011) and many of the sites on the Colorado and Fraser River were identical to previous work. More effort (higher number of people) was used compared to earlier studies resulting in higher capture probabilities that better met assumptions of the removal model and likely allowed unbiased estimates of exuvia with two depletion passes. Simple two pass population models were more than sufficient in the vast majority of cases. Only at very high and very low densities was there any evidence of biased estimates due to changing capture probabilities with pass (Figure 1). The two pass depletion technique worked well for these estimates and many of the issues with depletion estimates encountered in fish population estimates were not a problem due to the immobile nature of the exuvia, high capture probability, and no size selective gear (Riley and Fausch 1992, Peterson et al. 2004, Saunders et al. 2011). All sampling is now complete for Job \#1, Action \#2, analysis is ongoing and results will be presented in future Federal Aid Reports.



Figure 1. Population and capture probability estimates comparing a thee pass Huggins Closed Capture model in Program Mark (with time effects that varied capture $\hat{p}$ to a simple two pass removal model of Zippin 1956. There was some variation in the estimated capture probability at very low densities ( $<80$ exuvia per 30 m ) and very high densities ( $>6,000$ exuvia per 30 m ) indicating that the assumption of equal capture probabilities for all passes is violated with the simple two pass model. However, that bias was relatively small and population estimates of the two models were very close.

Action \#3- Measure aquatic habitat variables at salmonfly population estimate sites.

- Level 1 Action Category: Data collection and analysis
- Level 2 Action Strategy, survey or monitoring- habitat
- Level 3 Action Activity: Monitoring

Physical habitat surveys have been complete at all 18 sites. These surveys included pebble counts to characterize dominant substrate size (Potyondy and Hardy 1994) and two methods to measure substrate embeddedness. Embeddedness was visually estimated following the methods of Bain and Stevenson (1999) and was measured following the Weighted Burns Quantitative Method (Burns 1985, Sennatt et al. 2006). Physical surveys of each site were completed with survey-grade GPS equipment and a HydroSurveyor acoustic Doppler current profiler system (ADCP). The GPS and ADCP surveys were analyzed by CPW aquatic researcher and hydrologist Eric Richer. Examples of the physical habitat survey maps and bathymetric maps produced with the GPS and ADCP surveys are presented in Figures 2 and 3. The data from the physical habitat surveys will be analyzed to compile a list of variables that are hypothesized to explain differences in stonefly habitat quality. A candidate set of models will be developed to identify which variables best explain differences in stonefly density with the information theoretic approach (Burnham and Anderson 2002). Density estimates and habitat surveys are complete for 18 sites on all three major rivers in Colorado with large populations of salmonflies. The modeling exercise will identify habitat variables that explain differences in stonefly density and could explain their decline or extirpation from sites. This information can then be used to guide habitat improvement projects in the Upper Colorado River basin as well as inform water development decisions on how to protect in stream aquatic habitat. All sampling is now complete for Job \#1 Action \#3, analysis is ongoing and results will be presented in future Federal Aid Reports.


Figure 2. Survey points and bathymetry data collected with the survey-grade GPS equipment and Acoustic Doppler Current Profiler of the Pumphouse stonefly site.


Figure 3. Bathymetric map produced by the GPS and ADCP survey used to estimate physical channel characteristics of stonefly study sites

## References

Bain, M.B., and Stevenson, N.J., eds. 1999. Aquatic habitat assessment: common methods. American Fisheries Society, Bethesda, MD.

Barbour, M.T., J. Gerritsen, B.D. Snyder, and J.B. Stribling. 1999. Rapid bioassessment protocols for use in streams and wadeable rivers: periphyton, benthic macroinvertebrates and fish. Second edition. EPA 841- D-97-002. U.S. Environmental Protection Agency; Office of Water; Washington, D.C.

Baxter, C.V., K.D. Fausch, and W.C. Saunders. 2005. Tangled webs: reciprocal flows of Invertebrate prey link streams and riparian zones. Freshwater Biology. 50: 201-220.

Burnham, K. P., and D. R. Anderson. 2002. Model selection and multimodel inference: a practical information-theoretic approach, 2nd edition. Springer-Verlag, New York.

Burns, D. C., and Edwards, R. E. 1985. Embeddedness of salmonid habitat of selected streams on the Payette National Forest. USDA Forest Service, Payette National Forest, McCall, ID.

Dewalt, R. E. and K.W. Stewart. 1995. Life-histories of stoneflies (Plecoptera) in the Rio Conejos of southern Colorado. Great Basin Naturalist. 55: 1-18.

Erickson, R.C. 1983. Benthic field studies for the Windy Gap study reach, Colorado River, Colorado, fall, 1980 to fall, 1981. Prepared for The Northern Colorado Water Conservancy District, Municipal Sub-District.

Fore, L.S., J.R. Karr, and R.W. Wisseman. 1996. Assessing invertebrate responses to human activities: Evaluating alternative approaches. Journal of the North American Benthological Society 15(2):212-231.

Nehring, R.B. 1987. Stream fisheries investigations. Colorado Division of Wildlife, Federal Aid in Sportfish Restoration, Project F-51-R, Progress Report, Fort Collins.

Nehring, R.B. 2011. Colorado River aquatic resources investigations. Colorado Division of Wildlife, Federal Aid in Sportfish Restoration, Project F-237R-18, Final Report, Fort Collins.

Peterson, J.T., R.F. Thurow, and J.W. Guzevich. 2004. An evaluation of multipass electrofishing for estimating the abundance of stream-dwelling salmonids. Transactions of the American Fisheries Society 133:462-475.

Potyondy, J.P. and T. Hardy. 1994. Use of pebble counts to evaluate fine sediment increase in stream channels. Water Resources Bulletin 30:509-520.

Riley, S.C., and K.D. Fausch. 1992. Underestimation of trout population size by maximum likelihood removal estimates in small streams. North American Journal of Fisheries Management 12:768-776.

Saunders, W.C., K.D. Fausch and G.C. White. 2011. Accurate estimation of salmonid abundance in small streams using nighttime removal electrofishing: an evaluation using marked fish. North American Journal of Fisheries Management, 31:403-415.

Sennatt, K.M., N.L. Salant, C.E. Renshaw, and F.J. Magilligan. 2006. Assessment of Methods for Measuring Embeddedness: Application to Sedimentation in Flow Regulated Streams. Journal of the American Water Resources Association 42(6):1671-1682.

Walters, D.M., J.S. Wesner, R.E. Zuellig, D.A. Kowalski, M.C. Kondratieff. In Press. Holy flux: spatial and temporal variation in massive pulses of emerging insect biomass from wester U.S. rivers. Ecology 2017.

Zippin, C. 1956. The removal method of population estimation. Journal of Wildlife Management 22:82-90.

## Job No. 2. Impacts of Whitewater Park Development on Trout, Aquatic Invertebrates and Mottled Sculpin Cottus bairdi

Job Objective: Investigate the effects of whitewater parks on trout, aquatic invertebrates and mottled sculpin.

## Need

Artificial whitewater parks (WWP) are increasingly common throughout Colorado and there are concerns about how they impact fish and aquatic invertebrates (Fox 2013, Kolden 2013). Many of the rivers throughout the state with whitewater parks are also some of the best wild trout fisheries. The construction of whitewater parks involves replacing natural riffles with concrete or grouted rock grade control structures to produce hydraulic waves for recreational boating. Natural riffles serve many important physical and ecological roles in rivers. Ecologically, riffles serve as the most productive areas of a stream for periphyton and invertebrate production that form the foundation of the aquatic food web. Physically, riffles serve as grade control structures for streams and their location and frequency are main drivers of stream geomorphology. Artificial pools created below WWP waves have been found to hold a lower biomass of trout than natural pools, and have more dynamic and higher magnitude flows and velocities (Kolden 2013). Whitewater parks have also been documented to cause a suppression of fish movement that is related to fish length (Fox 2013). Concerns have been raised that whitewater parks not only impact fish habitat and fish passage but could affect some aquatic invertebrates that are primary diet items for trout (Kondratieff 2012).

In addition to sportfish concerns, native non-game fish are also common at many whitewater park sites. Mottled sculpin are a bottom dwelling fish that occupy many coldwater streams and rivers of Colorado. Their unique habitat preferences and reliance on good quality riffle and run habitat make them a good ecological indicator of stream health (Nehring 2011). Because the function of riffle and run habitat is generally impacted when stream flows are altered or instream habitat is manipulated, mottled sculpin may be impacted by habitat related changes before higher predators like trout. Sculpin could not only indicate ecological problems that will eventually affect sport fish like trout, but they serve as an important food source, especially for brown trout common in many Colorado rivers. The objective of this study is to investigate the effects of building whitewater parks on mottled sculpin, aquatic invertebrates, and trout by sampling before and after construction with control sites. Two whitewater parks were constructed in western Colorado in 2014, on the Uncompahgre River in Montrose and at the Pumphouse Recreation site on the Colorado River. Their construction provided an opportunity for the first comprehensive study of before/after impacts to fish and invertebrates. To meet the objectives of this project a before, after, control, impact (BACI) study design was used to evaluate changes in trout population, mottled sculpin density and aquatic invertebrates at these two sites.

## Objectives

1. Investigate the effects of building whitewater parks on aquatic invertebrate density and diversity at two whitewater park sites on the Colorado and Uncompahgre Rivers before and
after construction.
2. Investigate the effects of building whitewater parks on the Colorado and Uncompahgre Rivers on the density of trout and mottled sculpin before and after construction.

## Approach

Action \#1- Sample aquatic invertebrates to estimate the density and diversity above, at and below the sites of whitewater parks before and after construction.

- Level 1 Action Category: Data collection and analysis
- Level 2 Action Strategy: Research, survey or monitoring- fish and wildlife populations
- Level 3 Action Activity: Abundance determination


## Uncompahgre River

On the Uncompahgre River aquatic invertebrate samples were taken at five sites, one below the planned WWP, three within the park, and one above. Of the three sites within the WWP, one was converted from a natural riffle to a run (WWP3) while the other two remained functioning (but smaller) riffles between drop structures. The WWP on the Uncompahgre River consist of six drop structures over about 0.2 miles of river. Five replicate macroinvertebrate samples were collected at each site using a $0.086 \mathrm{~m}^{2}$ Hess sampler with a $350 \mu \mathrm{~m}$ mesh net. Samples were collected in November of 2014 (pre-construction), 2015, and 2016. Samples were collected from the same riffle with predominantly cobble substrate by disturbing the streambed to a depth of approximately 10 cm . Field samples were washed through a $350-\mu \mathrm{m}$ sieve and organisms preserved in $80 \%$ ethanol. Velocity and depth were taken at each Hess sample site to ensure samples were taken from similar riffle habitat. Macroinvertebrate samples were sorted and subsampled in the laboratory using a standard USGS 300-count protocol, except that replicates were not composited and each one underwent the protocol (Moulton et al. 2000). All organisms, except for chironomids and non-insects, were identified to genus or species. Chironomids were identified to subfamily and non-insects (e.g., oligochaetes, amphipods) were identified to class. Each replicate sample was processed separately so an average of 1,670 individual specimens were identified at each riffle site. Many more individual specimens were identified from each site compared to standard methods to ensure rare organism were sampled and to increase the power of the comparisons between riffles sites in close proximity within the same stream (Vincent and Hawkins 1996). A summary of macroinvertebrate results is presented in Figures 48. Data analysis was still ongoing at the time of this report, but invertebrate density and diversity of invertebrates declined at all sites in 2016 after generally increasing the first year post construction. Currently there are no significant trends in invertebrate density and diversity across years or between whitewater park and control sites on the Uncompahgre River. High spatial and temporal variability of aquatic invertebrates has been observed during the study on the Uncompahgre River.


Figure 4. Density and standard error of aquatic invertebrates from the Uncompahgre River in 2016.


Figure 5. Species richness of aquatic invertebrates from the Uncompahgre River in 2016


Figure 6. Density of all species of aquatic invertebrates with standard error bars on the Uncompahgre River 2014-2016.


Figure 7. Density of ephemeroptera, plecoptera, and trichoptera fauna with standard error bars on the Uncompahgre River 2014-2016.


Figure 8. Total species richness on the Uncompahgre River 2014-2016.

To monitor mottled sculpin and brown trout, three electrofishing stations were established concurrent with the invertebrate sites, one below the WWP, one within (that encompassed two invertebrate sampling riffles) and one above. Sites 1 and 3 had habitat improvement projects completed in 2007 aimed at improving fish habitat. The electrofishing stations averaged 704.3 ft (512-849) long. Attempts were made to use block nets, but they could not be kept in place due to high discharge and velocity. Natural stream features like shallow riffles were used as endpoints to best insure closure. Three pass removal electrofishing was completed at each site with a Smith Root VVP15 truck mounted electrofisher and five anodes. All fish were weighed, measured and population estimates were made with the Huggins Closed Capture model in Program Mark (Huggins 1989, White and Burnham 1999). To reduce the bias associated with the size selectivity of electrofishing, capture probabilities were modeled with fish length as a covariate similar to the approach described in Saunders et al. 2011. Four models were built for each species estimating capture probabilities by length, time, time + length, as well as a constant capture probability for all fish and all three passes. The time models allowed for different capture probabilities for the $2^{\text {nd }}$ and $3^{\text {rd }}$ passes compared to the first to address a common source of bias in electrofishing removal models. Model selection was conducted with AICc, population and parameter estimates were made by model averaging across all four models with AICc weights (Burnham and Anderson 2002). Figures 9 and 10 summarize the fish sampling data. Low capture probabilities at all sites (especially the controls) led to large variation around most of the fish population estimates. Fish are relatively low density in the Uncompahgre River compared to similar rivers and combined with high stream gradient and fast water velocities low capture probabilities lead to highly variable and imprecise population estimates. Currently no significant changes can be detected in fish populations during the study on the Uncompahgre River.


Figure 9. Brown trout population estimates from the three sampling reaches of the Uncompahgre River 2014-2016.


Figure 10. Mottled sculpin population estimates from the three sampling reaches of the Uncompahgre River 2014-2016.

## Colorado River

On the Colorado River aquatic invertebrate samples were taken at three sites, one below, one within and one above the WWP. The upper site is two riffles above the WWP site and the lower site is the next downstream riffle, all sites are with a 0.4 -mile reach. The WWP on the Colorado River consists of a single large cross channel wave structure so fewer sites were necessary. Unlike the Uncompahgre where post construction riffles remained in the WWP, at Pumphouse the middle site was converted from a run to a drop structure with pools above and below (Figure 12). Five replicate macroinvertebrate samples were collected at each site using a $0.086 \mathrm{~m}^{2}$ Hess sampler with a $350 \mu \mathrm{~m}$ mesh net. The replicate samples were collected from the same riffle with predominantly cobble substrate by disturbing the streambed to a depth of approximately 10 cm . Field samples were washed through a $350-\mu \mathrm{m}$ sieve and organisms preserved in $80 \%$ ethanol. Velocity and depth were taken at each Hess sample site to ensure samples were taken from similar riffle habitat. Macroinvertebrate samples were sorted and sub-sampled in the laboratory using a standard USGS 300-count protocol, except that replicates were not composited and each one underwent the protocol (Moulton et al. 2000). All organisms, except for chironomids and non-insects, were identified to genus or species. Chironomids were identified to subfamily and non-insects (e.g., oligochaetes, amphipods) were identified to class. Each replicate sample was processed separately so an average of 1,379 individual specimens were identified at each riffle site. A much higher number of individual specimens were identified from each site compared to standard methods, to ensure rare organism were sampled and increase robustness of the comparisons between riffles sites in close proximity in the same stream (Vinson and Hawkins 1996). A preliminary summary of macroinvertebrate results is presented in Figures 11, 13 and 14. Data analysis was ongoing at the time of this report. Overall invertebrate density, EPT density, and species richness declined at the WWP site one-year post construction but overall density and EPT density increased in 2016 back to similar levels as pre-construction. Total species richness has declined at the WWP site since construction while increasing or remaining stable at control sites.

To monitor trout and mountain whitefish populations around the WPP, mark recapture electrofishing was conducted with a 16 ft aluminum jet boat and a Smith Root 2.5GPP electrofisher. The sampling reach was $6,451 \mathrm{ft}$ long and averaged 171 ft wide and was centered on the WWP structure. The sampling reach was divided into four sub reaches to evaluate fish density with the study reach. Station 1 is from bottom of Gore Canyon to the riffle above Launch \#1, Station 2 is from the riffle above Launch \#1 to the whitewater park feature, Station 3 is from the whitewater park feature to Launch \#3, and Station 4 is from Launch \#3 to the bottom of the sampling reach. Station \#3 has significantly lower numbers of fish than the other stations at the 95\% level.

Fish population estimates were made with the Huggins Closed Capture Model in Program Mark (Huggins 1989, White and Burnham 1999). Four models were built by estimating capture probabilities by length, species, species + length, as well as a constant capture probability for all fish (but varying by time), identical to a Lincoln Petersen model (Seber 1982). Model selection was done with AICc and population and parameter estimates were made by model averaging across all four models with AICc weights (Burnham and Anderson 2002). Fish population
estimates are presented in Figures 15 and 16, density estimates in Figure 16 and catch per unit effort in Figure 18.

Trout and whitefish populations have remained relatively stable over time throughout this study and there is no evidence of large effects of the whitewater park structure on gamefish populations in the study reach at this time. However, fish densities at the sampling stations below the WWP structure were significantly lower at the $95 \%$ level than above and the sampling reach immediately below the structure had the lowest population estimate. The sampling reach immediately below the structure also had the lowest catch per unit effort of fish of any of the stations sampled. There is some indication that fish habitat below the structure is currently less suitable for trout than above but more work is necessary to evaluate this trend.

The structure does not appear to be a complete migration barrier for adult brown trout at 977$1,000 \mathrm{cfs}$. Four browns ( $371-422 \mathrm{~mm}$ ) were documented passing above the structure between the first and second passes of our 2016 estimate. No small fish, rainbows or mountain whitefish were document passing up through the structure. More work is necessary to evaluate fish passage at the structure and sampling in 2017 should help address this issue. A total of 164 rainbows, browns and whitefish of all sizes were marked with a unique fin clip and moved from above to below the structure when sampling was complete in 2016.


Figure 11. Density of all invertebrates with standard error bars at sites on the Colorado River at Pumphouse 2014-2016.


Figure 12. Before and after photos of the whitewater park feature at Pumphouse on the Colorado River. The whitewater park feature replaced a natural run with a drop structure featuring two hydraulic waves.


Figure 13. Density of EPT fauna with standard error bars at sites on the Colorado River at Pumphouse 2014-2016.


Figure 14. Total species richness at sites on the Colorado River at Pumphouse 2014-2016.


Figure 15. Fish population estimates and $95 \%$ confidence intervals before and after construction of the whitewater park structure on the Colorado River at Pumphouse.


Figure 16. Fish population estimates and $95 \%$ confidence on the Colorado River at Pumphouse for each sampling station in 2016. Station 1 is from bottom of Gore Canyon to the riffle above Launch \#1, Station 2 is from the riffle above Launch \#1 to the whitewater park feature, Station 3 is from the whitewater park feature to Launch \#3, and Station 4 is from Launch \#3 to the bottom of the sampling reach. Station \#3 has significantly lower numbers of fish than the other stations at the $95 \%$ level.


Figure 17. Fish per acre estimates and 95\% confidence intervals for the four sampling stations on the Colorado River at Pumphouse in 2016.


Figure 18. Catch per unit effort for brown trout and rainbow trout for the four sampling stations on the Colorado River at Pumphouse in 2016.

Action \#2- Develop and test population estimation techniques for mottled sculpin.

- Level 1 Action Category: Data collection and analysis
- Level 2 Action Strategy: Techniques development
- Level 3 Action Activity: Fish and wildlife research, survey and management techniques

Mottled sculpin are difficult to effectively sample for quantitative population or density
estimates. Their small size, cryptic nature and lack of a swim bladder make them less than ideal subjects for common fisheries techniques like multiple pass removal electrofishing. Because of the size of rivers in which they inhabit, accurate and precise population estimates are difficult to achieve because closure assumptions are violated and capture efficiency can be low. To test sculpin density estimation techniques on large rivers, three electrofishing stations were established on the Colorado River at the Pumphouse recreation area. Because the river averages 170.5 ft wide at this site, it was impossible to electrofish for mottled sculpin across the whole channel and smaller plots along the bank in run habitat were chosen. Site 1 was near BLM Boat Launch \#1 above the WWP, site 2 was centered on the WWP structure, and site 3 was near BLM Boat Launch \#3, below the WWP. Three pass removal electrofishing was completed at each plot with three Smith Root LR24 backpack electrofishers. To evaluate the closure assumptions of the removal model and check estimated capture probabilities, mottled sculpin were captured before each site was sampled, marked with a caudal fin clip and then released inside each plot. The electrofishing sites averaged 102 feet long and 17.6 feet wide. Twenty-five to 50 feet of stream were electrofished both upstream and downstream of each electrofishing station to look for marked fish that may have left the station, violating closure assumptions of the model. All fish were measured to the nearest mm and population estimates were made with the Huggins Closed Capture model in Program Mark (Huggins 1989, White and Burnham 1999). To reduce the bias associated with the size selectivity of electrofishing, capture probabilities were modeled with length as a covariate similar to the approach described in Saunders et al. 2011. Four population estimation models were built modeling capture probabilities by fish length, time, time + length, as well as a constant capture probability for all fish and all three passes. The time models allowed for different capture probabilities for the $2^{\text {nd }}$ and $3^{\text {rd }}$ passes compared to the first to address a common source of bias in electrofishing removal models. Model selection was done with AICc and population and parameter estimates were made by model averaging across all four models with AICc weights (Burnham and Anderson 2002).

Mottled sculpin density estimates from 2016 are presented in Table 4. Capture probabilities were average to good (0.48-0.69). Sampling in 2015 showed that measured capture probabilities were lower than the model averages estimates (Figure 19) indicating there was a violation of the closure assumption and/or individual heterogeneity in capture probabilities. These issues are well known with removal models with electrofishing but can be overcome in some instances (i.e. with salmonids) with high capture probabilities, modeling capture probabilities over time and by using length as a covariate to model capture probabilities (Riley and Fausch 1992, Saunders et al. 2011, Petersen et. al 2004). Because mottled sculpin are small, cryptic, lack a swim bladder and because we could not ensure closure, our density estimates are likely biased low. However, it does appear that the biases are relatively small and all in the same direction (low) so comparisons of relative density between these sites (all collected with same methods and equipment) should be valid. Petersen et al. (2004) states that, "at relatively high first-pass efficiencies (>35\%) and low reduction in efficiency per pass (<1.10), the removal estimates were nearly unbiased." Riley and Fausch (1992) found that the negative bias for estimates decreased as initial capture probability increased and for three-pass estimates confidence interval coverage was actually better at low population sizes because of the larger standard deviations associated with small samples.

Few marked fish were found outside of sampling reaches indicating that closure assumptions were met relatively well, especially for longer reaches. Of the four sampling reaches evaluated in 2016 (three study reaches and one extra reach) two of the four sites had no documented emigration and two sites had $8 \%$ of the marked fish found outside of the sampling reach. The two sites with documented emigration outside the study reach were 65 ft long and 100 ft long and in both cases the marked fish were found within 10 ft above the sampling reach, no marked fish were found below the stations.

The results of these investigations indicate that with suitable sampling reaches and proper protocols three pass removal estimates are appropriate to get density estimates of mottled sculpin. With sampling reach lengths of at least 110-125 ft, three passes, and moderate to good capture probabilities achieved with three backpack electrofishers, relatively accurate density estimates were made on the Colorado River. Using the Huggins closed capture model was important because it allowed capture probabilities to be modeled by fish length. This model also allowed estimates of capture probability to vary between passes which were important in both previous work and this study (Figure 19). The modeling exercises generally showed much more support in the data (lower AICc values) for models that included a length covariate and allowed different capture probabilities for the $2^{\text {nd }}$ and $3^{\text {rd }}$ pass compared to the $1^{\text {st }}$. Model averaging allowed model selection uncertainty to be included in parameter estimates but it was important to have both fish length and time varying capture probability models in the candidate set. Simple removal models (i.e. Zippin 1956) were never the top model in our analyses and always underestimated population size compared to model averaged estimates.

Table 4. 2016 Mottled Sculpin Density Estimates from the Colorado River at Pumphouse.

|  | Capture Probability (SE) |  |  | $\begin{gathered} \text { Density } \\ \text { (Fish/Acre) } \end{gathered}$ | 95\% C.I. $\pm$ |
| :---: | :---: | :---: | :---: | :---: | :---: |
|  | Pass 1 | Pass 2 | Pass 3 |  |  |
| Launch 1 | 0.479 (0.04) | 0.487 (0.07) | 0.487 (0.07) | 10,175.3 | 1,160 |
| WWP | 0.686 (0.04) | 0.663 (0.08) | 0.663 (0.08) | 4,382.4 | 221 |
| Launch 3 | 0.571 (0.05) | 0.587 (0.10) | 0.587 (0.10) | 7,671.2 | 906 |



Figure 19. "Measured" capture probability across passes in 2015 for mottled sculpin sites on the Colorado River. Measured capture probability was calculated by comparing the number of marked fish captured in a pass to the number available.


Figure 20. "Measured" capture probability compared to estimated capture probability for mottled sculpin in the Colorado River. Measured capture probability was calculated by comparing the number of marked fish captured in a pass to the number available. Estimated capture probability was from the model averaged results of the four models built in the Huggins Close Capture model in Program Mark.

Action \#3- Sample mottled sculpin density at impacted and control sites before and after construction of whitewater parks.

- Level 1 Action Category: Data collection and analysis
- Level 2 Action Strategy: Research, survey or monitoring- fish and wildlife populations
- Level 3 Action Activity: Abundance determination

Mottled sculpin were sampled from representative sites above, at and below the whitewater park structures. The sampling reaches were concurrent with the invertebrate sampling riffles in the invertebrate study (Action \#1) and were 80, 125, and 100 feet long with an average width of 17.7 ft . Three pass removal electrofishing with a concurrent mark recapture estimate was conducted to evaluate assumptions on capture probabilities between passes. Fish were measured to the nearest millimeter and density estimates were made for each site with the Huggins Closed Capture model in Program Mark and are presented in Table 4 (Huggins 1989, White and Burnham 1999). Mottled sculpin densities were higher at all sites in 2016. The whitewater park site had the lowest density in 2016 (significant at $95 \%$ level) but no different from preconstruction estimates. The sites in this study show high variability from year to year and site to site in mottled sculpin densities making trends difficult to detect with the sampling protocols used. There is no evidence at this time that the WWP structure significantly affected sculpin densities at this site.


Figure 21. Mottled sculpin density estimates and $95 \%$ confidence intervals on the Colorado River at Pumphouse before and after construction of the whitewater park structure.

## References

Burnham, K. P., and D. R. Anderson. 2002. Model selection and multimodel inference: a practical information-theoretic approach, 2nd edition. Springer-Verlag, New York.

Fox, B. 2013. Eco-evaluation of whitewater parks as fish passage barriers. Master's thesis. Colorado State University, Fort Collins, Colorado.

Huggins, R. M. 1989. On the statistical analysis of capture-recapture experiments. Biometrika 76:133-140.

Kolden, E. 2013. Modeling in a three-dimensional world: whitewater park hydraulics and their impact on aquatic habitat in Colorado. Master's thesis. Colorado State University, Fort Collins, Colorado.

Kondratieff, M.C. 2012. Stream habitat investigations and assistance. Colorado Parks and Wildlife Progress Report, Federal Aid in Sportfish Restoration F-161-R18n Progress Report, Fort Collins.

Moulton, S.R., II, Carter, J.L., Grotheer, S.A., Cuffney, T.F. \& Short, T.M. 2000. Methods of analysis by the U. S. Geological Survey national water quality laboratory: processing, taxonomy, and quality control of benthic macroinvertebrate samples. Open-File Report 00-212, U.S. Geological Survey, Washington D.C.

Nehring, R.B. 2011. Colorado River aquatic resources investigations. Colorado Division of Wildlife, Federal Aid in Sportfish Restoration, Project F-237R-18, Final Report, Fort Collins.

Peterson, J.T., R.F. Thurow and J.W. Guzevich. 2004. An evaluation of multipass electrofishing for estimating the abundance of stream-dwelling salmonids. Transactions of the American Fisheries Society 133:2, 462-475.

Riley, S. C., and K. D. Fausch. 1992. Underestimation of trout population size by maximum likelihood removal estimates in small streams. North American Journal of Fisheries Management 12:768-776.

Saunders W.C., K.D. Fausch, and G.C. White. 2011. Accurate estimation of salmonid abundance in small streams using nighttime removal electrofishing: an evaluation using marked fish. North American Journal of Fisheries Management 31:403-415.

Seber, G. A. 1982. The estimation of animal abundance and related parameters, Second edition. Charles Griffin and Company, Ltd, London.

Vinson, M.R. and C.P. Hawkins. 1996. Effects of sampling area and subsampling procedure on
comparisons of taxa richness among streams. Journal of the North American Benthological Society 15(3): 392-399.

White, G. C., and K. P. Burnham. 1999. Program MARK: survival estimation from populations of marked animals. Bird Study 46(Supplement): 120-139.

Zippin, C. 1956. The removal method of population estimation. Journal of Wildlife Management 22:82-90.

## Job No. 3. Colorado River Water Project Mitigation and Ecology Investigations

Job Objective: Investigate the ecological impacts of stream flow alterations on aquatic invertebrates and fish of the Colorado River and assist in the planning and evaluation of mitigation efforts to address those impacts.

## Need

Trans-basin and local water use divert approximately $67 \%$ of the flow of the upper Colorado River and future projects will deplete flows further. Previous work under Project F-237 identified ecological impacts of streamflow reductions and a main stem reservoir (Windy Gap) on the invertebrates and fish of the river. Native mottled sculpin, once common are now rare or extirpated immediately below the reservoir. The health of the invertebrate community declined after the construction of Windy Gap; there has been a $38 \%$ reduction in the diversity of aquatic invertebrates from 1980 to 2011 and 19 species of mayflies, 4 species of stoneflies and 8 species of caddisflies had been extirpated from the sampling site below Windy Gap (Erickson 1983, Nehring 2011). Previous work under F237 Kowalski (2014) included mottled sculpin sampling above and below WGR (as well as other impoundments of the upper Colorado River) corroborated patterns of sculpin distributions and established that sculpin have been functionally extirpated from the Colorado River below WGR. Once common in this reach, sculpin are now absent for many miles downstream of WGR but become increasingly common as tributaries increase streamflows as depletions are offset by reservoirs releases to satisfy downstream senior water rights.

Increased trans-basin water diversions are planned and there are ongoing efforts to implement mitigation measures to reduce the impact of the new projects. A large component of the mitigation plan is constructing a bypass around the reservoir. This would reconnect the river and address various impacts of a large main channel impoundment but would not reduce the impacts of water withdrawals from the system. The planned bypass channel offers a unique opportunity to evaluate the effects reconnecting the river around the reservoir as well as investigate if mitigation measures can offset the impacts of large water diversions on the ecology of the river. The need for this project is to assist stakeholder groups in planning mitigation efforts and then to evaluate those efforts (if they are completed). This need is evident in the "Key Habitats" designation of riparian/wetlands systems and West Slope rivers identified in Colorado State Wildlife Action Plan. The need is also highlighted by the description of the important salmonid
sport fisheries in the Colorado River Basin Aquatic Management Plans as well as the designation of the Colorado River under the Gold Medal program.

## Objectives

1. Assist CPW staff as needed in planning of mitigation efforts.
2. Continue monitoring invertebrate and fish populations of the upper Colorado and Fraser Rivers.
3. Evaluate the effectiveness of mitigation measures in restoring and improving the ecological function of the Colorado River in Middle Park (if they are completed).

## Approach

Action \#1- Provide technical assistance as needed to stakeholders in the Upper Colorado cooperative effort.

- Level 1 Action Category: Technical assistance
- Level 2 Action Strategy: Technical assistance
- Level 3 Action Activity: With individuals or groups involved in resource management decision making

Provide technical assistance as needed to stakeholders in the Upper Colorado cooperative effort.
Coordination is continuing among project stakeholders including CPW personnel, the Upper Colorado River Learning by Doing Management Committee, Windy Gap Technical Assistance Committee (TAC), Trout Unlimited, and private landowners downstream of Windy Gap. The two most relevant efforts to this research are the bypass channel planning and construction being handled mostly by the TAC and the planned stream habitat improvement that CPW will be heavily involved with. Coordination with all of the stakeholders will continue under project F237 and increase as projects move from the planning stage to implementation.

Action \#2- Sample aquatic invertebrates and fish above and below Windy Gap Reservoir to collect baseline data before mitigation projects occur.

- Level 1 Action Category: Data collection and analysis
- Level 2 Action Strategy: Research, survey or monitoring- fish and wildlife populations
- Level 3 Action Activity: Baseline inventory

A large amount of baseline data has been collected previously under Project F-237. If mitigation measures are decided on and implementation appears eminent, routine sampling will continue at historic sites. The exact sampling protocols and sampling sites will depend on the specifics of mitigation measures and will be defined in cooperation with other researchers. Currently it appears that the largest mitigation measure, a bypass channel around Windy Gap Reservoir could be constructed as early as 2018. Invertebrate and fish sampling is planned to resume in 2017 to collect pre-construction data above and below Windy Gap.

## References

Erickson, R.C. 1983. Benthic field studies for the Windy Gap study reach, Colorado River, Colorado, fall, 1980 to fall, 1981. Prepared for The Northern Colorado Water Conservancy District, Municipal Sub-District.

Kowalski, D.A. 2014. Colorado River aquatic resources investigations. Colorado Division of Wildlife, Federal Aid in Sportfish Restoration, Project F-237-R21, Progress Report, Fort Collins.

Nehring, R.B. 2011. Colorado River aquatic resources investigations. Colorado Division of Wildlife, Federal Aid in Sportfish Restoration, Project F-237R-18, Final Report, Fort Collins.

## Job No. 4. Bacterial Kidney Disease Investigations

Job Objective: Investigate the distribution and prevalence of Renibacterium salmoninarum, the causative agent of Bacterial Kidney Disease in Colorado’s sport fisheries.

## Need

Renibacterium salmoninarum is an important fish pathogen in Colorado that can have large economic and management impacts. Hatcheries in Colorado were free from the pathogen for almost 19 years but recently several cases have been found at both state and federal hatcheries. $R$. salmoninarum is classified as a prohibitive pathogen in the state of Colorado and there are restrictions throughout the west on transportation of this bacteria. Hatcheries that are positive for this disease are often depopulated to remove the pathogen of extensive management of broodstock units must occur to manage around the disease. While BKD is known to have a large impact on fish in hatcheries, its prevalence and effects on wild trout fisheries and stocked sport fisheries has not been thoroughly examined. The objective of this study is to investigate the distribution and prevalence of $R$. salmoninarum in Colorado's wild trout fisheries and stocked sport fisheries. This need is apparent in the "Key Habitats" designation of riparian/wetlands systems and West Slope rivers identified in Colorado State Wildlife Action Plan. The need for this project is also implied in the descriptions of the important salmonid sport fisheries in the Gunnison Basin Aquatic Management Plans and Colorado River Basin Aquatic Management Plan.

## Objectives

1. Survey a stratified random sample of wild trout streams in all major river basins in Colorado to determine the distribution and prevalence of $R$. salmoninarum.
2. Survey sport fisheries recently stocked with fish from hatcheries that tested positive for $R$. salmoninarum to determine if stocking has affected the prevalence and distribution.

## Approach

Action \#1- Survey wild trout streams throughout Colorado for the presence of R. salmoninarum.

- Level 1 Action Category: Data collection and analysis
- Level 2 Action Strategy: Research, survey or monitoring- fish and wildlife populations
- Level 3 Action Activity: Monitoring

Trout streams were stratified by river basin and size and randomly selected to be sampled for $R$. salmoninarum. Third to fifth order wild trout streams under CPW management categories 302, 303, 405, and 406 were eligible for selection. Ten waters in each major river basin were randomly selected, reviewed by the area fish biologist for inclusion in the study giving 99 total waters. Figure 22 shows the geographic location of selected waters. Streams were sampled to determine species composition and disease samples were taken from up to 60 individuals of the dominant salmonid species with preference to brook trout and brown trout. Fish were combined into five fish lots by species and age class to reduce processing time. Samples were sent to the CPW Aquatic Animal Health Laboratory in Brush, CO and the USFWS Bozeman Fish Technology Center in Bozeman, MT. Samples will be tested for R. salmoninarum with four tests; quantitative PCR or real-time PCR, nested PCR, DFAT (direct fluorescent antibody test), and ELISA (enzyme-linked immunosorbent assay).

As of July 2017 a total of 21 wild trout waters have been sampled (Figure 23). Sample processing is ongoing but partial results have been received for five wild trout waters. One water has tested positive for R. salmoninarum; the Fraser River in Grand County. Sixty brook trout were sampled from the Fraser and one 5 fish lot out of 12 tested positive by qPCR and was confirmed with nPCR. Table 5 contains a summary of all positive test results from the wild trout survey.

Action \#2- Survey stocked sport fisheries throughout Colorado for the presence of $R$. salmoninarum.

- Level 1 Action Category: Data collection and analysis
- Level 2 Action Strategy: Research, survey or monitoring- fish and wildlife populations
- Level 3 Action Activity: Monitoring

Sport fisheries that have been stocked by hatcheries that recently came down positive for $R$. salmoninarum were matched with nearby waters with similar management strategies and species composition to compare if recently stocked waters have different prevalence of the bacteria than other fisheries. Figure 22 shows the geographic location of selected waters, there were 91 total waters included in this study. Other waters around the state outside of the stocked fish survey have also been sampled by crews during this project. These include waters that have specific management needs relating to BKD, waters around positive hatcheries (e.g. upper Poudre River, Quartz Creek), and waters with observed fish health issues. Lakes, reservoirs, and streams were sampled to determine species composition and disease samples were taken from up to 60 individuals of the dominant salmonid species and up to 60 of the dominant warmwater game fish if present. Ultimately the number and type of samples taken were determined by the number of fish present with routine sampling techniques on a water by water basis. Fish were combined into five fish lots by species and age class to reduce processing time. Samples were sent to the CPW Aquatic Animal Health Laboratory in Brush, CO and the USFWS Bozeman Fish

Technology Center in Bozeman, MT. Samples will be tested for $R$. salmoninarum with four tests; quantitative PCR (real-time PCR), nested PCR, DFAT, direct fluorescent antibody test (DFAT), and enzyme-linked immunosorbent assay (ELISA).

As of July 2017 a total of 67 stocked trout waters have been sampled (Figure 23). Sample processing is ongoing but partial results have been received for 28 waters and four have tested positive for R. salmoninarum; Cherry Creek Reservoir, Eagle Lake, Windsor Reservoir and Gross Reservoir. At Cherry Creek 21 of 30 walleye tested positive with a qPCR screening test and 11 of those were confirmed positive by nested PCR. Out of 30 gizzard shad tested from Cherry Creek, 13 tested positive by qPCR and five were confirmed positive by nPCR. At Eagle lake Sixty brook trout were sampled and two five-fish lots out of 12 tested positive by qPCR and were confirmed with nPCR. One of these lots also tested positive by DFAT. At Windsor Reservoir 27 white suckers, 17 yellow perch and 16 gizzard shad were sampled and one five fish lot of gizzard shad tested positive by qPCR and was confirmed by nPCR. At Gross Reservoir 29 brown trout, 29 rainbows, one brook trout and one lake trout were sampled. One eight-fish lot of rainbows tested positive by both qPCR and nPCR. Table 5 contains a summary of all positive test from the stocked waters survey.

Table 5. Summary of waters that tested positive for $R$. salmoninarum with both real-time or quantitative PCR (qPCR) and nested PCR (nPCR) in 2016.

| Water | Water <br> Code | Study | Species | qPCR | nPCR | DFAT |
| :--- | :---: | :--- | :--- | :---: | :---: | :---: |
| Cherry Creek Reservoir | 52580 | Stocked Waters | Gizzard Shad | POS | POS | NA |
|  |  |  | Walleye | POS | POS | NA |
| Eagle Lake | 66363 | Stocked Waters | Brook Trout | POS | POS | POS |
| Windsor Reservoir | 53645 | Stocked Waters | White Sucker | NEG | NA | NA |
|  |  |  | Yellow Perch | NEG | NA | NA |
| Fraser River |  |  | Gizzard Shad | POS | POS | NA |
| Gross Reservoir | 20355 | Wild Trout | Brook Trout | POS | POS | NA |
|  | 55043 | Stocked Waters | Brown Trout | NEG | NA | NA |
|  |  |  | Rainbow Trout | POS | POS | NA |



Figure 22. Map of selected stream and lake sampling sites for the wild fish BKD survey, stocked waters BKD survey, and the Upper Poudre River BKD survey.


Figure 23. Map of BKD sampling sites completed in 2016.

Job No. 5. Gunnison Tunnel Electric Fish Guidance System Evaluation

Job Objective: Evaluate the effectiveness of an electric fish guidance system on the South Canal of the Gunnison River

## Need

The Gunnison Tunnel diverts an average of 360,600 acre feet of water annually from the Gunnison River, a Gold Medal trout fishery, and fish loss in the canal has been an ongoing concern. The construction of several hydropower plants on the canal was expected to increase mortality of entrained fish so an electric fish guidance system was installed at the diversion structure in 2012. The fish guidance system was a novel design and this type of system had not been tried in the orientation it was applied on the Gunnison. Fish entrainment in irrigation canals is a large and generally unquantified problem across the west and fish guidance technology is more commonly being applied to address the problem. The need for this specific project was to provide a rigorous evaluation of the effectiveness of this type of system. This need is apparent in the "Key Habitats" designation of riparian/wetlands systems and West Slope Rivers identified in Colorado State Wildlife Action Plan as well as the descriptions of the important salmonid sport fisheries and Gold Medal designation in the Gunnison Basin Aquatic Management Plans.

## Objectives

1. Monitor entrainment of fish in the South Canal.
2. Evaluate the effectiveness of the electric guidance system by marking fish in the Gunnison River and sampling in the South Canal.

## Approach

Job \#5 is complete and the final report appears in Appendix 1. And is available at: http://cpw.state.co.us/Documents/Research/Aquatic/pdf/Publications/South-Canal-Fish-Barrier-Final-Report.pdf.

Results have been presented at the 2016 Western Division of American Fisheries Society meeting in Missoula Montana (Kowalski and Gardunio 2016). The electric barrier appears to meet its objective and successfully exclude larger fish from the study reach, but not smaller age 0 , age 1 , or age 2 trout. The entrainment, growth and survival of smaller fish maintains a stable population of fish in the canal, but fewer entrained mature fish is likely a benefit to the fish population of the Gunnison River. The low rate of exclusion of smaller fish is likely due to high approach velocities at the fish barrier during high stream flows.

## References

Kowalski, D.A. and E. Gardunio. 2016. Evaluation of an electric fish barrier on the South Canal of the Gunnison River, Colorado. 2016 Annual Meeting of the Western Division of the American Fisheries Society. Missoula, Montana. May 24, 2016.

## Job No. 6. Technical Assistance

Job Objective: Provide information and assistance to aquatic biologists, aquatic researchers and managers in a variety of coldwater ecology applications.

## Need

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

## Objectives

1. Provide technical assistance to biologists, managers, researchers, and other stakeholders as needed.

## Approach

Action \#1- Provide technical assistance as needed.

- Level 1 Action Category: Technical assistance
- Level 2 Action Strategy: Technical assistance
- Level 3 Action Activity: With individuals or groups involved in resource management decision making

Technical assistance is provided as necessary and requested by biologist and other stakeholders. Current technical assistance projects include developing more effective and efficient methods of trout fry estimation using distance sampling and spatial mark recapture models and identifying a new method to simplify using length covariates to improve trout population estimation.

# Appendix 1. Evaluation of an Electric Fish Barrier on the South Canal, an Irrigation Ditch on the Lower Gunnison River, Colorado 

Dan Kowalski<br>Colorado Parks and Wildlife Aquatic Research Section<br>Montrose, CO 81401


#### Abstract

An electric fish barrier was installed on the east portal of South Canal to reduce fish entrainment associated with the construction of two hydropower plants in 2012. The objective of this study was to monitor fish entrainment and evaluate the effectiveness of the barrier. Three groups of fish were tagged and released upstream of the barrier; fish from the canal, wild Gunnison River fish, and hatchery reared fingerlings. Mark recapture boat electrofishing was completed and population estimates were made with the Huggins Closed Capture model using fish length to model capture probabilities. The study reach contained 2,994 $\pm 1,043$ fish ( $>150 \mathrm{~mm}$ ) in October 2011, 1,764 $\pm 279$ in October 2013, 1,224 $\pm$ 239 in July 2014 and $1,900 \pm 379$ in October 2014. Fish population estimates have declined after the electric barrier, significantly at the $95 \%$ level for brown trout but not for rainbows. A total of 288 tagged fish less than 300 mm and four fish greater than 300 mm were recovered below the barrier, representing $1.3 \%$ of all tagged fish. The electric barrier appears to meet its objective and successfully exclude larger fish from the study reach, but not smaller age 0 , age 1 , or age 2 trout. The entrainment, growth and survival of smaller fish maintains a stable population of fish in the canal, but fewer entrained mature fish is likely a benefit to the fish population of the Gunnison River. Further study is needed to evaluate if smaller adult trout can be successfully excluded by the electric barrier with operational modifications.


There are over 105,000 irrigation structures on rivers and streams across Colorado, most in fish bearing waters. Fish entrainment in irrigation canals is known to be a large problem in the western U.S. (Carlson and Rahel 2007) and the loss of fish in irrigation canals has been shown to be a population sink for trout in Wyoming (Roberts and Rahel 2008). The impact of fish lost to irrigation canals on fish populations in Colorado is unquantified. The South Canal is an irrigation ditch in southwest Colorado that diverts an average of 360,600 acre feet of water each year, about 857 cfs average daily flow March-November, from the Gunnison River for agriculture (Bureau of Reclamation 2012). The river contains a Gold Medal trout fishery despite documented entrainment of fish for many years in the canal. The construction of a hydropower plant was expected to increase mortality of entrained fish so an electric fish barrier was installed at the diversion structure in 2012. From the diversion structure and barrier, the canal travels through a 5.7 -mile-long tunnel before egressing approximately 0.5 miles above the power house (Figure 1). There is a total of 7.7 miles of earthen canal that contains the majority of fish that are entrained from the Gunnison River. The canal diverts water from March through November each year with the amount of water depending on water supply and irrigation demand. During winter months the canal is generally shut off with only a very small amount of flow as a result of
accretions and seepage. About twice a month it is partially opened to run approximately 100 cfs through the canal for 24-48 hours to fill a drinking water supply reservoir. Because of low and intermittent flows in the canal, fish survival over winter was generally thought to be low but variable year to year depending on frequency of freezing temperatures. However, in the winter of 2012-2013, a constant flow of $20-25$ cfs was run all winter long to keep water supply reservoirs full during construction of the hydropower plant. This resulted in what appeared to be a much larger number of fish in the canal in spring of 2013 due to increased survival of entrained fish.

The study reach for this project was downstream of the concrete drop below the West Portal (just below the first powerhouse) and was 0.72 miles, ending at the $2^{\text {nd }}$ concrete drop structure (Figure 2, UTM NAD83 258703, 4262335). The canal averaged 46.1 feet wide with 20-25 cfs in March 2013 and 70.2 feet wide at 540 cfs in October 2013. The study reach represents $9.4 \%$ of the total earthen portion of the South Canal but is suspected of containing the highest density of entrained fish due to its proximity to the West Portal. While fish routinely pass through the high velocity concrete portions of the canal, the majority of fish reside in the lower gradient earthen portion of the canal.


Figure 1. Area map of the Gunnison Tunnel and South Canal (Bureau of Reclamation 2012). The study site is between the West Portal and Drop 1.

The fish barrier was constructed in 2012 and was operational before the 2013 irrigation season. It consists of a series of vertically suspended electrodes across the east portal of the Gunnison Tunnel (Figure 3). The waterway at the barrier is 74 ft wide, 16 ft deep, and has water velocities between $0.2-0.7 \mathrm{~m} / \mathrm{s}(0.66-2.3 \mathrm{fps})$ and conductivity of $180 \mu \mathrm{~s} / \mathrm{cm}$. The system is powered by three 1.5 KVA Smith Root pulsators with a max power output of 4.5 kW and is designed to operate with a frequency of 2 Hz , pulse width of 0.005 s and a field strength of $1 \mathrm{v} / \mathrm{inch}(0.4 \mathrm{v} / \mathrm{cm})$. The barrier was designed to exclude "brood stock" rainbow and brown trout but target size was not specified (Smith Root 2011). The barrier is believed to have operated continuously as planned throughout the entire 2013-2014 irrigation seasons. Communication has been lost for brief time periods (i.e. 6 out of over 6,000 hours of operation in 2013) but operation of the barrier was thought to be unaffected and it is assumed that is has functioned continuously during irrigation season the last two years (J. Heneghan, personal communication).

The purpose of this study was to estimate fish populations in the South Canal before and after the barrier and investigate the entrainment of fish from the Gunnison River. To accomplish this, fish population estimates were compared before and after the barrier was built over different seasons and across years while tagged fish were used to document any movement across the barrier.


Figure 2. Fish sampling site on the South Canal. The sampling reach was 0.72 miles long (3,802 feet) and was between the first and second concrete drop structures below the West Portal.

## METHODS

South Canal was sampled with mark-recapture electrofishing (October 2011, October 2013, July 2014 and October 2014) and multiple pass removal (March 2013) to estimate fish populations of adult and juvenile trout. The study reach for all three occasions was the same but differing methods were used in the spring sampling because of the different habitat and flows when water is not being diverted (20-25 cfs vs. 500-900 cfs).

On March 29, 2013, the canal consisted of two distinct habitat types, consisting of the concrete stilling basin just below the first drop and the earthen portion of the canal below. The density of fish was much higher in the stilling basin and the physical habitat dictated that different sampling methods be used in the two locations. The reach was stratified by habitat types and two sampling reaches were chosen. The entire stilling basin was sampled with 50 ft . bag seine that was 6 ft . deep with $1 / 8 \mathrm{in}$. mesh. Multiple seine hauls were made through the stilling basin so a depletion population estimate could be made (Zippin 1956, White et. al 1982). Fish were held in a live pen and then measured for total length to the nearest millimeter. Capture probability was high (estimated to be 0.74 for rainbows and 0.79 for browns) and model assumptions of closure appeared to have been met due to the isolated and simple structure of the stilling basin. The high capture probability and lack of evidence of size selectivity of the seine is expected to help meet assumptions of the removal model and there was no evidence in the data to indicate an unacceptable amount of bias. The portion of the canal below the stilling basin consisted of shallow, slow moving channel that was 46.1 ft . wide $3,528 \mathrm{ft}$. long. A sampling reach was randomly chosen in this portion of the study reach that was $1,000 \mathrm{ft}$ long and block nets were used to ensure closure. Five Smith Root LR24 backpack electrofishers were used to complete a two pass removal population estimate. Fish were held in a live pen and then measured to nearest millimeter and weighed to the nearest gram, and then returned to the canal. After the March estimate, 876 fish were removed from the canal in an effort to depopulate the study reach before the barrier's first season of the operation. One hundred and twenty-five fish from the stilling basin were tagged with coded wire tags (CWT) and adipose fin clips and transported by aerated fish truck to the Gunnison River in East Portal. They were stocked at the boat ramp approximated 0.7 miles above the East Portal and the barrier.

Because electrofishing removal estimates are known to be biased low due to size selectivity and individual capture heterogeneity, we took several approaches to reduce this bias recommended by Riley and Fausch (1992) and Saunders et al. (2011). First efforts were made to use sufficient effort for high capture probabilities. Second, capture probabilities were modeled by fish species and length to account for heterogeneity. The data was analyzed in Program Mark with the Huggins Closed Capture Model (White and Burnham 1999, Huggins 1989). To reduce the bias associated with the size selectivity of electrofishing, capture probabilities were modeled with length as a covariate similar to the approach described in Saunders et al. 2011. Four models were built by estimating capture probabilities by length, species, species + length, as well as a constant capture probability for all fish. Model selection was done with AICc and population and parameter estimates were made by model averaging across all four models with AICc weights (Burnham and Anderson 2002). To estimate the total trout in the study reach in March 2013, the two pass removal estimate was expanded for the length of canal that contained similar
habitat and added to the estimate for the stilling basin. The confidence intervals were calculated by summing the variances of each estimate (Delta Method) and multiplying by 1.96.

Four groups of fish were tagged and released in East Portal upstream from the Gunnison Tunnel to challenge the barrier. One hundred and twenty-five fish (59 brown trout and 66 rainbow trout) from the March 2013 sampling of the stilling basin were moved from below the barrier to above and received both coded wire tags and adipose fin clips. Mean length of the tagged fish was 241 mm for brown trout (range $165-310 \mathrm{~mm}$ ) and 232 mm for rainbows ( $180-392 \mathrm{~mm}$ ). Wild fish were captured by boat electrofishing on June 17 and 19, 2013, in the Gunnison River above the barrier and tagged with both coded wire tags and adipose clips. A total of 1,265 fish (653 rainbow trout and 612 brown trout) were tagged, the mean length of brown trout was 281 mm ( $103-737 \mathrm{~mm}$ ) and $336 \mathrm{~mm}(82-547 \mathrm{~mm}$ ) for rainbows. Fingerling rainbow trout from the Rifle Falls Fish Hatchery were also tagged and released into the Gunnison River in East Portal above the barrier. A total of 19,800 fish with a mean length of 68 mm were tagged with coded wire tags on June 24-26, 2013 and stocked into the Gunnison River 0.7 m above the barrier on July 26. Due to the results of the first study season, the focus in 2014 was on tagging larger fish and 1,841 wild fish from the Gunnison River above the barrier were tagged with 32 mm half duplex PIT tags. The mean length was $396 \mathrm{~mm}(200-545 \mathrm{~mm})$ and an estimated $21.7 \%$ of the fish larger than 200 mm in the Gunnison River above the barrier were tagged. A total of 23,031 trout from 68 mm to 737 mm were tagged in the 2013-2014 and released in the Gunnison River above the barrier.

Mark recapture population estimates in the study reach were conducted in October 2011, October 2013, July 2014 and October 2014 with a 14 ft aluminum jet boat with Smith Root 2.5 GPP electrofisher. The study reach, equipment and methods for all occasions were the same. Fish were measured to the nearest millimeter and all fish on the recapture pass were weighed to the nearest gram. All captured fish were examined for fin clips and checked for coded wire tags with a Norwest Marine Technology T-Wand Detector and for PIT tags with an Oregon RFID handheld reader. On the marking pass all fish greater than 150 mm were marked with a caudal fin punch and held in a live pen to ensure recovery. Fish were returned by boat throughout the study reach to ensure redistribution in the population. The recapture pass was completed 72 hours after the marking pass and generally accepted methods were followed for mark recapture studies (Curry et al. 2009). The interval between capture events was chosen to maximize redistribution of marked fish throughout the population but to attempt to meet demographic and geographic closure assumptions of the model. The first power plant served as an upstream migration barrier further ensured geographic closure; block nets downstream were not feasible to the high volume of water in the canal (600-900 cfs). Model assumptions appear to have met well as marked fish were not observed to be encountered in any temporal or spatial pattern in the canal. Capture probabilities were good and the catch per unit effort of fish was similar between the passes.

A stationary PIT tag antenna was constructed above the penstock of the power plant but below the barrier in the spring of 2014. The objective was to differentiate fish deterred by the barrier and turbine mortality as well as increase detection of tagged fish. The antenna was operational for less than two months as the extreme velocities of the water ( 900 cfs in a 10.5 ft . wide
concrete channel) made it impossible to keep in place. No tags other than test tags were detected by the antenna.

Fish population estimates were made with the Huggins Closed Capture Model in Program Mark (Huggins 1989, White and Burnham 1999). Four models were built by estimating capture probabilities by length, species, species + length, as well as a constant capture probability for all fish, identical to a Lincoln Petersen model (Seber 1982). Model selection was done with AICc and population and parameter estimates were made by model averaging across all four models with AICc weights (Burnham and Anderson 2002).


Figure 3. The electric fish barrier on the east portal of the South Canal.

## RESULTS

The results of the population estimates are summarized in Table 1 and Figure 4. Length frequency histograms from the fall 2013 sampling are presented in Figures 5-7. Model selection results from are summarized in Appendix A, Tables 2-6.

The population modeling exercise in Program Mark provided good results and estimates appeared accurate (all years) and relatively precise (except for October 2011). The expected bias of population estimates should be low due to model assumptions being met, and the ability to model the size selectivity of electrofishing with fish length covariates. The top population model for the October 2011 data contained terms that varied capture probability by length and time while the second ranked model that contained terms for species, length and time was $2.40 \Delta \mathrm{AICc}$ units behind. Models with a term for fish length contained $0.98 \%$ of the model weights. Capture probabilities were lower during this survey ( 0.10 ) compared to subsequent surveys due to the
higher flows and lower total number of fish captured.
In March 2013, the top population model for the canal and the stilling basin had a single capture probability for all fish regardless of species or length while the second ranked model contained a term for species. These two models are essentially identical to the simple Zippin two pass removal model and had $73.2 \%$ of the model weight (Zippin 1956). Although it has been shown that electrofishing surveys generally have a size related bias, this effect was not seen in these data because of how few fish were in the canal outside of the stilling basin and there was little variation in fish size compared to the fall surveys. Because of the low density of fish, moderate capture probabilities and similar sized fish, the data from the canal were too sparse to support more detailed models. There was no evidence of size selectivity in the stilling basin with the small mesh seine.

The top population model for the October 2013 data contained terms that varied capture probability by length, species and time while the second ranked model that contained terms for length and time. These two models accounted for $100 \%$ of the model weights and had much higher support than a simple Lincoln-Petersen (19-27 $\Delta \mathrm{AICc}$ units behind). Capture probabilities were high ( 0.33 ) due to the lower flow conditions than 2011. Model selection uncertainty was taken into account in all surveys by model averaging across all four models with model weights to get parameter estimates and population estimates.

The significant increase (95\% level) of total fish in the canal from April 2013 to October 2013 is evidence for fish successfully running the barrier and surviving the turbines. After the March estimate, when 876 fish were removed from the canal, the population estimate increased by 1,057 fish by October. The total number of estimated fish was significantly greater (at the 95\% level) in October than in April and was not significantly different than in the October 2011, before the barrier. The size structure and species composition of the fish in the 2013 also provide evidence of fish entrainment, specifically for brown trout (Figures 5, 6 and 7).

Table 1. Fish Population Estimates and 95\% Confidence Intervals from the South Canal 20112014. These estimates are for age 1 fish and older, the stocked CWT tagged rainbows are excluded from the rainbow trout estimates.

| Date | Species | \# Caught | Population Estimate in Study Reach |
| :---: | :---: | :---: | :---: |
| October 2011 | Brown Trout | 415 | 2,359 $\pm 981$ |
|  | Rainbow Trout | 108 | $634 \pm 354$ |
| March 2013 | Brown Trout | 683 | $924 \pm 52$ |
|  | Rainbow Trout | 495 | $659 \pm 46$ |
| October 2013 | Brown Trout | 573 | 1,035 $\pm 150$ |
|  | Rainbow Trout | 277 | $728 \pm 235$ |
|  | Stocked CWT Rainbow | 246 | 1,486 $\pm 768$ |
|  | CWT, Adipose Clipped Brown | 2 | NA |
|  | CWT, Adipose Clipped Rainbow | 0 | NA |
| July 2014 | Brown Trout | 225 | $586 \pm 52$ |
|  | Rainbow Trout | 132 | $638 \pm 469$ |
|  | Stocked CWT Rainbow | 25 | NA |
|  | CWT, Adipose Clipped Brown | 0 | NA |
|  | CWT, Adipose Clipped Rainbow | 0 | NA |
| Oct 2014 | Brown Trout | 305 | $964 \pm 258$ |
|  | Rainbow Trout | 277 | $936 \pm 278$ |
|  | Stocked CWT Rainbow | 15 | NA |
|  | CWT, Adipose Clipped Brown | 0 | NA |
|  | CWT, Adipose Clipped Rainbow | 4 | NA |



Figure 4. Estimated total number of trout age 1 and older and $95 \%$ confidence intervals in the South Canal study reach. After the March 2013 estimate, 876 fish were removed from the canal study reach and the barrier was operational at the start of the irrigation season in April 2013. There are about 1,094 fewer fish in the study reach since the barrier was installed but the decline is not significant at the $95 \%$ level, mostly because of the low capture probability and corresponding high uncertainty around the October 2011 estimate.


Figure 5. Length frequency histogram of trout captured in the South Canal in October 2013. A total of 246 coded wire tagged rainbows were captured that had been stocked upstream of the guidance system (plus 10 recaptures). They had a mean length of 163 mm (123-204). Two other coded wire tagged fish were captured, a 310 mm brown and 337 mm brown (the 310 mm fish was also recaptured). No tag loss was observed, all of the larger fish were double marked and no fish were observed with an adipose clip but without a CWT.


Figure 6. Length frequency histogram of brown trout captured in March and October 2013.


Figure 7. Length frequency histogram of rainbow trout captured in March and October 2013.
The top population models for the July and October 2014 data contained terms that varied capture probability by length, species and time. The top two models that included length accounted for $100 \%$ of the model weights. Capture probabilities were good in July (0.11-0.32) and (0.17-0.20) October. Model selection uncertainty was taken into account in all surveys by model averaging across all four models with model weights to get parameter estimates and population estimates.

The population modeling exercise for all the mark recapture data indicated that modeling capture probabilities by length was important under these conditions which agrees with previous work on the topic (Saunders et. al 2011). Using a simple Lincoln-Petersen model under these conditions could underestimate population size by overestimating the capture probability for small fish, even when using a length cutoff designed to exclude age 0 fish. Figure 8 shows an example of the estimated capture probability by length and Figure 9 shows a comparison of population estimates with and without the length covariate.

In October 2011, there were an estimated 2,994 $\pm 1,043$ fish in the South Canal study reach. In the spring of 2013 there were an estimated $1,583 \pm 70$ in the study reach, $89 \%$ in the stilling basin. Eight hundred and seventy-six of these fish were removed from the study reach leaving an estimated 707 fish when the irrigation flows first began in the spring of 2013. In October 2013 the estimated population had increased to $1,764 \pm 279$ trout. The population estimate of total fish in the study reach decreased from October 2011 to 2013 but that difference was not significant at the $95 \%$ level, likely due to the uncertainty around the 2011 estimate caused by lower capture probability likely due to higher flows. Subsequent sampling occasions had much higher capture probabilities generally in the $20 \%$ range ( $0.11-0.33$ ). In 2014 the study reach contained $1,224 \pm 239$ fish in July and $1,900 \pm 379$ in October.

In 2013, a total of 248 coded wire tagged fish from 123 mm to 337 mm were documented passing through the barrier, mostly smaller stocked rainbow trout ( $\mathrm{n}=246$ mean length 163 mm in October). Only two larger wild brown trout were confirmed passing the barrier (310 and 337 mm ). Of the tagged fish that were documented to have run the barrier in 2013, the stocked rainbows represent $1.24 \%$ of the fish marked in East Portal and the wild brown trout were $0.3 \%$. Overall, $1.17 \%$ of all the tagged fish in East Portal were captured in the study reach in 2013. In the 0.72 -mile study reach there was an estimated $1,486 \pm 768$ coded wire tagged rainbows or $7.5 \%$ of the tagged fish in East Portal.

These results do not represent a direct estimate of entrainment rates as only $9.4 \%$ of the total length of the canal was sampled at a single time interval. Rather, the results represent the number of entrained fish in the study area that were detected. It should be interpreted as a minimum number of fish that navigated the barrier because fish would have to pass the through the guidance system, travel the 5.7 -mile-long tunnel, avoid entrainment in two small lateral canals, survive passage through the hydropower turbines and remain in the first 0.72 miles of the 7.7mile canal to be detected. If the density of fish in the study reach is representative of the rest of the canal, then an estimated 15,809 coded wire tagged rainbow fingerlings would have been entrained or $79.8 \%$ of those fish and $3.2 \%$ of the larger marked wild brown trout in 2013. This is most likely an over estimate of entrained fish because the study reach could have a higher density of fish than the other reaches of the canal, but it demonstrates the same trend of high entrainment rates for small fish and relatively low rates for larger fish and can be interpreted as a potential maximum number. While estimating robust entrainment rates with the barrier is not possible in this study, the true rates are likely between these minimum and maximum values; 8$80 \%$ for small rainbow trout and 0.3-3.2\% for larger brown trout.

In 2014, a total of 44 tagged fish were encountered, 40 of the hatchery rainbows (mean length 326 mm at the time of capture). Four CWT and fin clipped wild rainbow trout (296-398 mm) were found. It is unknown exactly when or what size all the tagged fish in 2014 passed the barrier because fish lived and grew in the canal throughout the study. The 2013 data give the best idea of size of fish that ran the barrier because they were in the canal for a maximum of seven months. The large number of CWT tagged rainbows could have passed the barrier as small as 68 mm and then survived to be captured at a larger size.

By the end of the study 288 small or medium sized fish had been documented passing the barrier. Only four fish $>300 \mathrm{~mm}$, and no fish $>400 \mathrm{~mm}$ were documented passing the guidance system. Only $1.3 \%$ of all tagged fish were recovered in the canal study reach in two years. While turbine mortality and fish excluded from the study reach by the trash racks on the penstock cannot be differentiated from fish excluded by the barrier, very few large fish have been observed passing these barriers. The number of large fish ( $>350 \mathrm{~mm}$ ) in the study is not significantly different after the barrier (Figure 10) even though there is no evidence that fish of that size are passing through in great numbers. Large numbers of smaller fish have been shown to run the barrier as evidenced by both the number of marked fish and the stable trout population in the canal even after the barrier was in use. The lack of a decline in fish populations in the canal after the barrier is likely related to higher than expected survival and growth of small fish entrained in the canal.

In July 2014, 17\% of the fish captured during the population estimate (37\% greater than 350 mm ) had been handled the previous October by the presence of a healed caudal punch scar. This indicates that there is fair to good over winter survival in the canal. Growth of fish that live in the study reach is also relatively high; coded wire tagged rainbows grew an average of 6.4 inches from age 1 to age 2 . With the good annual survival and growth rates, the large numbers of smaller fish that pass the barrier maintain a relatively stable population of fish in the study reach, even though large fish do appear to be excluded from the canal by the electric barrier.


Figure 8. Estimated capture probability by length and 95\% confidence interval (dashed lines) for trout in the South Canal in October 2014.


Figure 9. Brown trout population estimates from the Huggins Closed Capture model in Program Mark comparing models with a fish length covariate to a standard Lincoln-Petersen. The estimates that used length to model capture probabilities were on aver 23\% higher (6-41\%) than the Lincoln-Petersen. Models containing length as a covariate had between $98-100 \%$ of the model weight across all mark recapture sampling occasions.


Figure 10. Estimated total number of trout greater than 350 mm in the South Canal study reach in the October sampling periods. While very few (4) fish greater than 300 mm have been
documented passing the barrier and turbines, growth and survival of smaller entrained fish supports a stable number of larger fish in the study reach.

## DISCUSSION AND MANAGEMENT RECOMMENDATIONS

The South Canal contained approximately 1,094 fewer fish in October 2014 after the barrier, than in October 2011. While the total fish estimates in the canal have declined since the barrier was installed, there is not a significant difference at the $95 \%$ level mostly due to the low capture probability ( $0.8-0.12$ ) and corresponding high uncertainty around the October 2011 estimate. The number of brown trout only is significantly lower at the $95 \%$ level in 2014 (two years after the barrier was installed) while the number of rainbow trout has remained relatively stable (Figure 11).


Figure 11. Population estimates of rainbow and brown trout and 95\% confidence intervals for the South Canal Study reach 2011-2014. There were signifcantly fewer brown trout in 2014, two years after the installation of barrier. Rainbow trout numbers have raimained relatively stable.

Of the 23,031 tagged fish, only $1.3 \%$ were recovered in the canal study reach in two years. At the end of the study, 288 small or medium sized fish had been documented passing the barrier. Four fish greater than 300 mm and no fish greater than 400 mm were documented passing the barrier. This size selectivity is expected with electrically based barriers and electrofishing is known to be highly size selective as well (Saunders et. al 2011). It is also likely that turbine mortality is higher on larger fish, further selecting for smaller fish to make it into the study reach. The growth and survival of fish in the canal is higher than expected as evidenced by the high proportion of recaptured fish from October 2013 to July 2014. The practice of running 100 cfs into the canal twice a month in the winter and relatively mild recent winters apparently allowed
for good fish survival during the winter of 2013-2014.
Fish in the Gunnison River are successfully passing the electric barrier and surviving the turbines, but mostly smaller fish. Their growth and survival in the canal maintains a stable fish population that is lower than before the barrier, significantly (at the $95 \%$ level) for brown trout only. The difference between species is likely due to two factors; larger size of age 0 brown trout and potential spawning of rainbow trout in the study reach. Because brown trout emerge about $8-10$ weeks earlier than rainbow trout they are larger during their first summer. Because the barrier is size selective, brown trout fry are expected to be entrained at a lower rate than rainbows. The canal is first filled with water around April $1^{\text {st }}$ of each year, just before rainbow trout spawn. Large numbers of age 0 rainbow trout were observed in the canal in July 2014 (they were smaller than the 150 mm size cut off used in the fish population estimates). It is unknown if they were entrained fish from the Gunnison River or were spawned in the canal, both are likely. Brown trout spawn in October in the Gunnison River and flows are generally shut off in the canal around October 31. Water flow is then stagnant or 100 cfs (twice a month for 24 hours) in the canal in winter. There is very little spawning habitat for brown trout in the canal and it is variable and poor quality compared to rainbow trout, which spawn at higher flows that are stable or increasing. A combination of higher entrainment rates and better potential spawning success in the canal likely leads to higher number of small rainbow trout in the canal.

If the barrier is successfully excluding many of the fish greater than 300 mm and most of the fish greater than 400 mm , then it is excluding approximately $15 \%$ of the trout greater than 150 mm and $26-71 \%$ of sexually mature fish based on 2013 data on the Gunnison River. Low numbers of age 2 fish in the Gunnison are sexually mature (mostly males) while most age 3 fish are mature (E. Gardunio, Colorado Parks and Wildlife, unpublished data). So while the barrier is generally meeting its stated objective, it’s not protecting all of the sexually mature fish. Excluding higher proportion of small trout from downstream passage is likely to be difficult and will be dependent on several factors including the voltage gradient of the barrier and the approach velocities of the water at the barrier. Excluding a larger proportion of adult fish than is currently occurring is a more reasonable expectation for the East Portal barrier with some operational changes.

As approach velocities increase above 2.5 fps the probability of excluding small salmonids with an electric barrier decreases (Demko et al. 1994, Pugh et al. 1970). The approach velocities at the South Canal barrier varied between 0.7 and 2.3 fps in October 2011 when the tunnel was flowing about 730 cfs and the river below the tunnel was about 580 cfs. Under those flows, better deterrence of small trout should be possible with operational adjustments, but more work is needed to determine approach velocities at various flows. The field strength of the South Canal barrier is currently about $1 \mathrm{v} /$ inch or $0.4 \mathrm{v} / \mathrm{cm}$ (Smith Root 2011) which is relatively conservative compared to other barriers designs. Most other downstream oriented barriers are graduate-field fish barriers (GFFB) where several rows of electrodes produce increasing voltage gradients between 0.2-1.2 v/cm while other designs have utilized voltage gradients as high as 3.0 v/cm (Raymond 1956, Burger et al. 2015). The GFFB technology appears more effective in deterring downstream movement of fish but was not applied at the South Canal due to site specific conditions. Diverting downstream moving fish is one of the more difficult applications of electric fish barriers (Burger et al. 2012). Achieving complete deterrence of all fish is unlikely
in scenarios like East Portal. The objective there should be to reduce the amount of entrainment much as feasible within the constraints of the system. More work is necessary to determine if increasing the voltage gradient, or other operational changes at the East Portal barrier could improve performance on smaller fish.

The electric fish barrier on the South Canal of the Gunnison River appears to effectively exclude large fish from the south canal, resulting in fewer entrained fish from the river. Fish populations in the South canal, while lower than before the barrier, appear stable due to the number of entrained smaller fish, potential spawning of rainbow trout and better than expected growth and survival of fish in the canal. The electric barrier on the South Canal should continue to be operated whenever feasible during the irrigation season and future study is needed to examine if operational changes of the current barrier can increase the probability of excluding more adult fish.

## REFERENCES

Bureau of Reclamation. 2012. Final Environmental Assessment South Canal Hydropower Project. Western Colorado Area Office Upper Colorado Region, U.S. Department of the Interior.

Burnham, K. P., and D. R. Anderson. 2002. Model selection and multimodel inference: a practical information-theoretic approach, 2nd edition. Springer-Verlag, New York.

Burger, C.V, J.W. Parkin, M. O’Farrell and A. Murphy. 2015. Barrier technology helps deter fish at hydro facilities. Hydo Review 34(5).

Burger, C.V, J.W. Parkin, M. O’Farrell, A. Murphy, J. Zelgis. 2012. Non-lethal electric guidance barriers
for fish and marine mammal deterrence: a review for hydropower and other applications. HydroVision Brazil, September 26, 2012.

Carlson, A.J. and F.J. Rahel. 2007. A basinwide perspective on entrainment of fish in irrigation canals, Transactions of the American Fisheries Society, 136:1335-1343.

Curry, R.A., R.M. Hughes, M.E. McMaster, and D.J. Zaft. 2009. Coldwater fish in rivers. Pages 139-158 in S.A. Bonar, W.A. Hubert, and D.W. Willis, editors. Standard methods for sampling North American freshwater fishes. American Fisheries Society, Bethesda, Maryland.

Demko, D.B, S.P. Cramer, D. Neeley and E.S. Van Dyke. 1994. Evaluation of a sound and electrical fish guidance system at the Wilkins Slough diversion operated by Reclamation District 108. Annual Report S.P. Cramer \& Associates Inc., Gresham OR.

Huggins, R. M. 1989. On the statistical analysis of capture-recapture experiments. Biometrika

76:133-140.
Pugh, J.R., G.E. Monan and J.R. Smith. 1970. Effect of water velocity on the fish-guiding efficiency of and electrical guiding system. Fishery Bulletin 68 (2):307-324.

Roberts, J.J. and F.J. Rahel. 2008. Irrigation Canals as Sink Habitat for Trout and Other Fishes in a Wyoming Drainage. Transactions of the American Fisheries Society 137:951-961.

Riley, S. C., and K. D. Fausch. 1992. Underestimation of trout population size by maximum likelihood removal estimates in small streams. North American Journal of Fisheries Management 12:768-776.

Saunders W.C., K.D. Fausch, and G.C. White. 2011. Accurate estimation of salmonid abundance in small streams using nighttime removal electrofishing: an evaluation using marked fish. North American Journal of Fisheries Management 31:403-415.

Seber, G. A. 1982. The estimation of animal abundance and related parameters, Second edition. Charles Griffin and Company, Ltd, London.

Smith Root. 2011. Electric Fish Barrier Design, Hardware Supply, Commissioning and Training Gunnison Tunnel, CO. 16 pp.

Raymond, H.L. 1956. Effect of pulse frequency and duration in guiding salmon fingerlings by electricity. U.S. Fish and Wildlife Service Research Report 43, Washington D.C.

White, G. C., D. R. Anderson, K. P. Burnham, and D. L. Otis. 1982. Capture-recapture and removal methods for sampling closed populations. Los Alamos National Laboratory, Report LA-8787-NERP, Los Alamos, New Mexico.

White, G. C., and K. P. Burnham. 1999. Program MARK: survival estimation from populations of marked animals. Bird Study 46(Supplement): 120-139.

Zippin, C. 1956. The removal method of population estimation. Journal of Wildlife Management 22:82-90.

Appendix A. Model Selection Results for Population Estimation Models

Table 2. Model Selection Results for the Mark Recapture Electrofishing in October 2011. Population estimates and capture probabilities were calculated by model averaging across all four models using model weights. The "Time" and "Time+Species" models are identical to the standard Lincoln Petersen model.

| Model | AICc | Number of <br> Parameters | Delta <br> AICc | AICc <br> Weights | Model <br> Likelihood |
| :--- | :---: | :---: | :---: | :---: | :---: |
| Time+Length | 893.0038 | 3 | 0 | 0.75 | 1.00 |
| Time+Species+Length | 895.4048 | 5 | 2.40 | 0.23 | 0.30 |
| Time | 900.3091 | 2 | 7.31 | 0.02 | 0.03 |
| Time+Species | 902.7106 | 4 | 9.71 | 0.01 | 0.01 |

Table 3. Model Selection results for the Two Pass Removal Electrofishing in March 2013. Population estimates and capture probabilities were calculated by model averaging across all four models using model weights.

| Model | AICc | Number of <br> Parameters | Delta <br> AICc | AICc <br> Weights | Model <br> Likelihood |
| :---: | :---: | :---: | :---: | :---: | :---: |
| Constant p | 117.40 | 1 | 0 | 0.528 | 1.00 |
| Species | 119.30 | 2 | 1.90 | 0.204 | 0.39 |
| Length | 119.39 | 2 | 2.00 | 0.195 | 0.37 |
| Length+Species | 121.34 | 3 | 3.94 | 0.073 | 0.14 |

Table 4. Model Selection Results for the Mark Recapture Electrofishing in October 2013. Population estimates and capture probabilities were calculated by model averaging across all four models using model weights. The "Time" and "Time+Species" models are identical to the standard Lincoln Petersen model.

| Model | AICc | Number of <br> Parameters | Delta <br> AICc | AICc <br> Weights | Model <br> Likelihood |
| :--- | :---: | :---: | :---: | :---: | :---: |
| Time+Species+Length | 1760.461 | 5 | 0 | 0.77 | 1.00 |
| Time+Length | 1762.837 | 3 | 2.38 | 0.23 | 0.30 |
| Time+Species | 1779.036 | 4 | 18.58 | 0.00 | 0.00 |
| Time | 1787.185 | 2 | 26.72 | 0.00 | 0.00 |

Table 5. Model Selection Results for the Mark Recapture Electrofishing in July 2014. Population estimates and capture probabilities were calculated by model averaging across all four models using model weights. The "Time" and "Time+Species" models are identical to the standard Lincoln Petersen model.

| Model | AICc | Number of <br> Parameters | Delta <br> AICc | AICc <br> Weights | Model <br> Likelihood |
| :--- | :---: | :---: | :---: | :---: | :---: |
| Time+Species+Length | 663.08 | 5 | 0 | 0.802 | 1 |
| Time+Length | 665.88 | 3 | 2.80 | 0.198 | 0.2469 |
| Time+Species | 685.91 | 4 | 22.83 | 0.000 | 0 |
| Time | 693.13 | 2 | 30.05 | 0.000 | 0 |

Table 6. Model Selection Results for the Mark Recapture Electrofishing in October 2014. Population estimates and capture probabilities were calculated by model averaging across all four models using model weights. The "Time" and "Time+Species" models are identical to the standard Lincoln Petersen model.

| Model | AICc | Number of <br> Parameters | Delta <br> AICc | AICc <br> Weights | Model <br> Likelihood |
| :--- | :---: | :---: | :---: | :---: | :---: |
| Time+Length | 1106.96 | 3 | 0 | 0.876 | 1.000 |
| Time+Species+Length | 1110.87 | 5 | 3.9097 | 0.124 | 0.142 |
| Time | 1122.71 | 2 | 15.7451 | 0.000 | 0.000 |
| Time+Species | 1126.72 | 4 | 19.7574 | 0.000 | 0.000 |

