Colorado River Aquatic Resources Investigations
Federal Aid Project F-237R-18

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

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Project Title: Colorado River Aquatic Resources Investigations

Project Objective: To document the relative distribution and abundance of the mottled sculpin *Cottus bairdi* and the aquatic invertebrate fauna of the Colorado River in Middle Park, Colorado in 2010 and compare the results with historical data and records compiled over the past 25-40 years, prior to the construction and operation of Windy Gap Dam in 1983.

Job No. 1 Colorado River Aquatic Invertebrate Investigations

Job Objective: Document the relative abundance and distribution of the aquatic invertebrate fauna of the upper Colorado River between the confluence with the Blue River and Windy Gap Dam west of Granby, Colorado.

INTRODUCTION

History - In the early 1980s, the Northern Colorado Water Conservancy District (NCWCD) was granted a permit to build Windy Gap Dam (WGD), the fifth of six mainstem impoundments constructed on major streams within upper Colorado River basin in Grand County during the 20thcentury. WGD created a shallow lake approximately 40 hectares (113 acres) in surface area just downstream of the confluence of the Colorado and Fraser rivers, west of Granby, Colorado. The initial purpose of WGD was to capture and pump up to 56,300 acre-feet (AF) of water annually from the upper Colorado River basin into Granby Reservoir and deliver it to the Front Range via the United States Bureau of Reclamation's (USBOR) Colorado-Big Thompson Project (C-BT) Project. Collected primarily from the Fraser River Basin, the water is pumped into Lake Granby, pumped again into Shadow Mountain Reservoir, whereupon it flows via gravity into Grand Lake for delivery to the East Slope via the Alva B. Adams Tunnel.

The infrastructure that comprises the C-BT Project was built between 1938 and 1957. Three other man-made impoundments that are part of the C-BT project on the West Slope include Granby, Shadow Mountain and Willow Creek reservoirs. Granby and Shadow Mountain dams collect and impound water from the North Fork of the Colorado River, Arapaho Creek and other tributary streams flowing out of the west side of Rocky Mountain National Park (RMNP). Willow Creek Reservoir (WCR) collects, impounds, stores and diverts water from Willow Creek through a canal for storage in Lake Granby and subsequent pumping into Shadow Mountain Reservoir.

Williams Fork Reservoir (WFR), the fifth impoundment in the upper Colorado River Basin in Grand County, owned and operated by the Denver Water Department (DWD), captures and stores water from the Williams Fork River Basin for later release to service senior water rights downstream in western Colorado. Wolford Mountain Reservoir impounds water flowing down Muddy Creek north of Kremmling, Colorado. In addition, the DWD captures water from numerous headwater streams that are tributary to the Fraser and Williams Fork rivers via an elaborate system of canals, delivering it to the Moffat Tunnel east of Winter Park, Colorado, for transmountain diversion to the East Slope.

According to the NCWCD web-page, the C-BT Project is the largest transmountain water diversion project in Colorado, providing supplemental water to 30 cities and towns, helping to irrigate approximately 693,000 acres of farmland in northeastern Colorado. Annually, the project delivers 213,000 acre feet (AF) of water to northeastern Colorado for agricultural, municipal and industrial uses. On the East Slope, the water is used to generate electricity as it falls almost half a mile through five power plants on its way to Colorado's Front Range for storage in Carter Lake, Horsetooth Reservoir and Boulder Reservoir. The water is subsequently released as needed to supplement native water supplies in the South Platte River basin.

During the 1980s, two studies were undertaken that developed baseline information on the fish and aquatic invertebrate fauna of the upper Colorado River from WGD downstream to the confluence with the Blue River near Kremmling, Colorado. The first study, an intensive, year-long sampling effort from September 1980 to September 1981, provided detailed, in-depth documentation of the species composition, diversity and relative abundance of the aquatic invertebrate fauna at seven sampling sites in this reach of the river prior to WGD construction (Figure 1). That study was funded by the NCWCD and conducted by Dr. Robert Erickson. Study results are contained in a report titled Benthic Field Studies for the Windy Gap Study Reach, Colorado River, Colorado – Fall 1980 to Fall 1981 (Erickson 1983). In this study, the density (numbers/m^2), relative abundance and distribution of the large stonefly larvae *Pteronarcys californica* was well documented throughout the 48 km (29.5-mile) reach of the upper Colorado River. Moreover, the Erickson study documented the distribution and relative abundance of many dozens of species of aquatic insects belonging to five major insect orders

(Ephemeroptera, Plecoptera, Trichoptera, Diptera and Coleoptera) as well as other aquatic invertebrate fauna.

The second study, an eight-year research project (Colorado River Aquatic Invertebrate Investigations) was initiated by the Colorado Division of Wildlife (CDOW) in April 1982 (Nehring 1983). The overall study objective was to develop baseline information on the various aspects of the riverine ecosystem for effective evaluation of the long-term effects of water withdrawals on the fish and aquatic invertebrate biota of the Colorado River in Middle Park. *Pteronarcys californica*, known to anglers as the willowfly, salmonfly or giant stonefly, was the primary aquatic invertebrate species chosen for study because of its importance as a food item for wild rainbow and brown trout in the river. The results from the first six years of this investigation are summarized in a 1987 report (Nehring 1987).

At the end of the 20th century, well over half of the average annual water yield of the upper Colorado River Basin (at the confluence of the Colorado and Fraser rivers near Granby) was being diverted through transmountain diversions to the East Slope. Now, two additional diversion projects are being proposed to transfer substantially more of the basin yield to Colorado's Front Range. The Windy Gap Firming Project as proposed by the NCWCD would divert additional water to the East Slope via the C-BT Project. The Moffat Tunnel Enhancement Project, as proposed by the DWD, would divert an additional 15,000 AF of water from the headwaters of the Fraser and Williams Fork rivers.

As these two water-yield enhancement proposals have moved through the process of public comment and review, environmental impact statements (EIS), permit application and approval, serious concerns about numerous potential impacts to the aquatic ecosystem of the upper Colorado River basin have been raised by many interested parties. The Colorado Division of Wildlife (CDOW) is the lead agency legally mandated by the state of Colorado to evaluate the validity and adequacy of any proposed mitigation measures set forth by the project proponents. As a result, researchers and managers of the Aquatic Program section of the CDOW were assigned various tasks to review historical data available from previous studies within the upper Colorado River basin as well as collect and analyze additional field data to address these concerns on behalf of the people of Colorado. The data collected and summarized in this report are (in part) the result of that assignment.

LITERATURE REVIEW

Freshwater resources worldwide have been negatively impacted from over appropriation and flow alteration due to centuries of human modification (Robarts and Wetzel 2000, Vörösmarty et al. 2000, Wohl 2006, Bates et al. 2008). The declining quality of water and these aquatic ecosystems is elucidated by documented extirpations and extinctions of aquatic organisms (Stein et al. 2000, Fochetti and Tierno De Figueroa 2006, IUCN 2008). In regulated rivers throughout the world, biodiversity is markedly decreased in areas directly below dams, whereas total invertebrate biomass may increase (Vinson 2001, Ward 1998). In a long-term analysis of macroinvertebrate assemblages below Flaming Gorge Dam in Wyoming, Vinson (2001) reported that nearly 40 years after the dam was completed, 23 insect taxa were extirpated from the riverine reaches directly below the dam. Sampling much further downstream revealed most of these 23 taxa were still present. The naturally complex habitats of the upper Colorado River host relatively diverse communities of aquatic fauna. Anthropogenic effects, particularly through river regulation, can change these natural flow regimes, leading to changes in native aquatic plants and animals that are important in natural ecosystem processes (Vinson 2001, Strange et al. 1999, Ward 1998). Long-term biomonitoring of aquatic insect communities may provide insight into how human induced alterations are affecting community structure, ecosystem functions and water quality.

Aquatic insects are a vital component of freshwater ecosystems. They process large amounts of organic material in the stream and transfer that energy to animals in higher trophic levels such as fish, crayfish, frogs, bats, birds, and spiders, making them a critical link connecting the aquatic and riparian communities (Baxter et al. 2005). Aquatic insect larvae occur in both lotic (running water) and lentic (standing water) habitats. Emerging from the water as larvae or adults, they exist in the terrestrial environment for a few hours to a few weeks. Reproduction is the primary function of the adult life stage (Hynes 1976, Brittain 1982). Many species in temperate regions have a one-year life cycle with much of that time spent feeding and growing as larvae in the stream.

Larvae can be divided into four functional feeding groups (FFGs), shredders, collectors, scrapers, and predators, based on morphology, food ingested, and behavior (Merritt et al. 2008, Cummins 1973). All FFGs have critical roles in nutrient cycling and the food chain. Shredders consume Coarse Particulate Organic Matter (CPOM, >1mm), collectors filter Fine Particulate Organic Matter (FPOM, <1mm) out of the water column, scrapers shear algae from the benthic substrate and predators consume living animal tissue (Cummins 1974, Vannote et al. 1980).

Although most larvae are generally placed in only one FFG, many show a degree of omnivory, i.e., shredders will incidentally consume small amounts of living animal tissue (Cummins and Klug 1979). A large portion of the energy in streams comes from the riparian area as CPOM and forms the base of the food web between the stream and the adjacent riparian areas (Cummins and Klug 1979, Vannote et al. 1980).

During aquatic larval stages, downstream drift is the major direction of movement, both unintentionally when disturbed and as a natural behavior usually at night to seek optimal habitat, avoid predators and competitors, and to avoid pollutants (Williams and Hynes 1976). Williams and Hynes (1976) reported that downstream drift of benthic fauna was responsible for 44.4 percent of recolonization. Several investigations have found the downstream drift is the most important pathway for colonization of downstream areas by aquatic insects (Vinson 2001, Townsend and Hildrew 1976, Williams and Hynes 1976). Upstream flight of adult aquatic insects accounted for only 18.2 percent of recolonization in one study (Hershey et al. 1993). The short life span and fragile nature of adult mayflies can limit their dispersal over long distances (Brittain 1982). Adult male stoneflies are generally weak fliers and some are wingless or have only vestigial wings (Hynes 1976). Downstream drift increases dramatically during the night time hours, increasing exponentially beginning at dusk (Anderson 1966, Elliott 1965, Hynes 1972, Müller 1963). Nocturnal drift also is higher during the dark phases of the moon (Anderson 1966; Müller 1963). Impoundments such as WGD interrupt this process, significantly compromising the biotic integrity and proper functioning of the stream ecosystem.

Mayflies (Ephemeroptera), stoneflies (Plecoptera) and caddisflies (Trichoptera), collectively referred to as the EPT taxa, are often the dominant groups in coldwater streams in mountainous regions of western North America. EPT species are the preferred groups for biological monitoring of freshwater ecosystems. Their long aquatic larval life stage, widespread distributions, predictable response to disturbance, high population densities, wide range in sensitivity to disturbances, and relatively low cost of quantitative methods to collect data make them reliable biological indicators for assessing ecosystem health (Brittain 1982, Hynes 1976, Lenat and Penrose 1996, Wallace et al. 1996, Barbour et al, 1999). Johnson et al. (1993) list eight attributes of an ideal indicator species; 1) easily identifiable, 2) cosmopolitan distribution, 3) numerical abundance, 4) low genetic and ecological variability, 5) large body size, 6) limited mobility and long life history, 7) ecological characteristics well known, and 8) suitable for use in laboratory studies. *Pteronarcys californica* (see below) has many of these attributes of an ideal indicator species.

The construction of man-made water impoundments may be responsible for eliminating or reducing local aquatic insect populations through dewatering, reductions in time and duration of flushing flows, and physical river bed disturbance. Additionally, they may alter natural temperatures and flow regimes which are considered to be the most important factors in regulating community structure (Brittain 1982, Ward 1976b, Hynes 1976, Steel and Lange 2007, Vinson 2001, Ward 1998). Dams pose a unique set of barriers and ecological conditions that may inhibit upstream larval migration, eliminate downstream drift, as well as reduce water quantity and quality required for aquatic insect survival. These changes in habitat combined with the difficulty that insects have recolonizing upstream habitat can contribute to the marked decline in species diversity usually seen at sites immediately below a dam.

Temperature can vary daily, monthly, seasonally and on an annual scale (Steel and Lange 2007). The life histories of many species of aquatic insects are often synchronized to a specific set of abiotic factors like day length and heating degree days (Hynes 1976, Brittain 1982, Wiggins 1996). Increased water temperature below dams may cause insect larvae to mature more quickly either through increased degree days or by increasing primary production (Gregory et al. 2000, Rockwell and Newell 2009, Hynes 1976), disrupting adult emergence patterns and successful larval recruitment (Rockwell and Newell 2009, Bunn and Arthington 2002).

Flow variability is another important factor affecting the life histories of aquatic insects. Snow-fed streams, like the Colorado River, experience dramatic changes from low to peak flows (Rockwell & Newell 2009, Poff et al. 1997). Peak flows inundate lateral margins of the stream bed, which may cue adult emergence and mating, providing more suitable habitat for oviposition and larval recruitment (Vinson 2001). Regular flushing flows are critically important for maintaining habitat quality by scouring out fine sediment and creating interstitial spaces under and around cobbles where larvae feed and find shelter. Many species of aquatic insects require this microhabitat type compared to embedded cobble or fine particle substrates (Brusven & Prather 1974, Bunn and Arthington 2002).

Natural flow regimes can act as cues in the timing of events such as egg hatch for aquatic insects (Brusven & Prather 1974, Bunn and Arthington 2002). Once mating and egg deposition in the stream has occurred, *Pc* eggs undergo a diapause phase that can last nine months to a year before hatching (Townsend and Pritchard 2000). During the diapause, little or no embryonic development occurs. Alterations in streamflow after oviposition can have a detrimental effect on long-term persistence and survival by desiccating developing eggs on dewatered, lateral margins of river banks. Analysis of long-term changes in the aquatic invertebrate community structure can elucidate both past and present factors contributing to the contemporary assemblage and point to bottlenecks or disconnects in ecosystem function (Lenat and Penrose 1996).

Pteronarcys californica is an excellent organism for biomonitoring. Large and easily identifiable, it occurs in many large streams and rivers in the western United States (Rockwell and Newell 2009, Kauwe et al. 2004). The larvae are restricted to swift, mountain streams among high gradient riffles dominated by cobble and boulder substrates. While this species usually occurs at elevations between 1,905 and 2,500 m (6,250 and 8,200 feet) (Rockwell and Newell 2009, Kauwe et al. 2004, Surdick and Gaufin 1978), in Colorado they can be found at elevations ≤1,524 m (5,000 feet) in the Gunnison River Gorge, and > 2,744 m (9,000 feet) in the Rio Grande basin near Creede. Larvae feed on coarse organic material and can reach high densities in areas with high concentrations of organic matter such as growth of filamentous green algae like *Cladophora*, leaf packs and woody debri. This species has relatively narrow environmental requirements and is given a very low tolerance level by western standards in rapid bioassessment (Barbour et al. 1999, Erickson 1983, Surdick and Gaufin 1978). It has a four to five year larval life cycle and undergoes incomplete metamorphosis (Nehring 1987, DeWalt and Stewart 1995, Townsend and Pritchard 1998, 2000). *Pc* larvae emerge and molt in late spring in a highly synchronous event with average adult survival spanning one week (DeWalt and Stewart 1995).

Pteronarcys californica larvae link the aquatic and terrestrial ecosystems as an important food item for fish, frogs, and crayfish. The adults are an important energy source for trout, birds, bats and spiders (Baxter et al. 2005). Late instar larvae and adults can exceed weights of 1g and lengths of 50 mm (Rockwell and Newell 2009). When present in high densities, *Pc* larvae can constitute a large portion of the total benthic biomass (Erickson 1983; Nehring 1983, 1984, 1985, 1986, 1987). In a diet study of wild rainbow and brown trout on the Colorado River in Middle Park, Grand County, Nehring (1987) reported that *P. californica* was consistently the single most important food item found in trout stomachs, comprising 64 to 75 percent of the mean stomach content over the course of a four-year study from 1980 through 1983. Among 72 stomach samples examined, the volumetric contribution of *Pc* larvae ranged from 42 to 95 percent of the gut content among individual fish. Erickson (1983) and Nehring (1987) agree that this one species of stonefly was a critically important component of the diet of wild trout in the upper Colorado River in Grand County.

BENTHIC SAMPLING METHODS

Sampling sites – During the 2010 field season, our intention was to collect benthic invertebrates at the same riffle areas that were sampled during the 1980-1981 study using the same sampling protocol (Erickson 1983). A map of the sampling sites for both the 1980-1981 and 2010 studies can be seen in Figure 1 in the Appendix. In the original study, the Colorado River was divided into three reaches. Sampling sites WG11, WG 12 and WG 13 were in Reach One, bounded by WGD and the confluence with the Williams Fork River. Sampling sites WG 21 and WG22 were in Reach Two, bounded upstream by the confluence with the Williams Fork River and downstream by the confluence with Troublesome Creek. The sampling sites in Reach three were WG31 and WG32, bounded upstream and downstream by Troublesome Creek and the Colorado Highway 9 Bridge near the confluence with the Blue River. This reach of river is negatively impacted by large inputs of sediment from Troublesome Creek.

Dr. Robert Erickson was hired for a day to visit the river in early April 2010, to insure that (if possible) the 2010 samples would be collected from the same seven riffle areas that were sampled during his 1980-1981 study. At five of the seven original sites we were able to accomplish that goal. The two sites where this was not possible were WG11 and WG32. The original WG11 site was located just 100 m downstream from the superstructure of Windy Gap Dam (WGD). That site was not sampled because the channel was completely modified during and after construction of WGD and is now extremely wide, very shallow, and matches the width of the entire spillway of WGD. Therefore it made no sense to sample at that site and we did not collect any sample for WG11 in the spring of 2010. However, in the fall of 2010, we added a station 1.45 km (0.9 miles) downstream of WGD, at a steep riffle area approximately 100 m upstream of the Grand County Road 57/578 Bridge, referred to as the “New WG11” or “Hitching Post Bridge” at times in this report.

Similarly, it made no sense to sample at the original WG32 site in the 1980-81 study. That riffle area was completely buried in sediment. Therefore, the 2010 – new WG32 sample site is approximately 3 km upstream from the original site. It is located at the first visible riffle area upstream at the point where the Colorado River and U.S. Highway 40 are in closest proximity to each other in the reach of the river downstream of the confluence with Troublesome Creek. In the interest of continuity and comparability of data between the 1980-1981 Erickson study and the 2010 collections, we used the same sample site identification system set forth in the earlier study (Table 1). However, in comparisons of invertebrate data from the 1980-1981 study and the 2010 study, we did not include any data from the June 30-July 1, 1981 (summer) collections.

Only data from the spring and fall collections were compared between the two studies. In the sampling matrix shown in Table 1, the year 2009 is also included. However, in the 2009 study, the only invertebrate species targeted was *Pteronarcys californica.* Data from the 2009 study was summarized and reported previously (Nehring et al. 2010).

Table 1. Benthic sampling site locations (stream and designation) and sampling periods (month and year) between the 1980-1981 (Erickson 1983) study compared with samples collected by the Colorado Division of Wildlife from 2009 through 2011.

Stream Name	**Site Descriptor**	**Sampling Period(s) – month/year**						
		Sept 1980	**May 1981**	**July 1981**	**Sept 1981**	**2009**	**Spring 2010**	**Fall 2010**
Fraser River	Solvista	no	no	no	no	no	no	**Yes**
Fraser River	Kaibab Park	no	no	no	no	**Yes**	**Yes**	**Yes**
Willow Creek	Above Willow Crk Rsvr	no	no	no	no	no	no	**Yes**
Colorado River	Old WG11	**Yes**	**Yes**	**Yes**	**Yes**	no	no	no
Colorado River	New WG11	no	no	no	no	**Yes**	no	**Yes**
Colorado River	WG12	**Yes**	**Yes**	**Yes**	**Yes**	no	**Yes**	**Yes**
Colorado River	Highway 40 Bridge	no	no	no	no	**Yes**	no	**Yes**
Colorado River	WG13	**Yes**	**Yes**	**Yes**	**Yes**	no	**Yes**	**Yes**
Colorado River	WG21	**Yes**	**Yes**	**Yes**	**Yes**	no	**Yes**	**Yes**
Colorado River	Skylark Ranch[a]	no	no	no	no	no	**Yes**[a]	no
Colorado River	WG22	**Yes**	**Yes**	**Yes**	**Yes**	no	**Yes**	**Yes**
Colorado River	WG31	no	**Yes**	**Yes**	**Yes**	no	**Yes**	**Yes**
Colorado River	New WG32	no	no	no	no	no	**Yes**	**Yes**
Colorado River	Old WG32	**Yes**	**Yes**	**Yes**	**Yes**	no	no	no
Gunnison River	Ute Park	no	no	no	no	no	**Yes**	no
Colorado River	Pumphouse	no	no	no	no	no	**Yes**	**Yes**

Note: **[a]:** the site at Skylark Ranch was sampled in early April 2011.

In order to provide a broader perspective and assessment of the status of the aquatic invertebrate community, eight additional sites were sampled. Four of these are shown in Figure 1 as yellow dots. Six were in the Colorado River Basin, three upstream and four downstream from the WGD (Table 1) and one on the Gunnison River. Two of the upstream sites were on the Fraser River, one approximately 100 m downstream of the U.S. Highway 40 Bridge across the river at the east end of Granby, and the other approximately 3 km upstream of the Highway 40 Bridge and 3 km downstream of the Fraser River Canyon on the Grand Elk/Solvista Ranch property. The third upstream site was on Willow Creek, approximately 10 km upstream of WCR and 1 km upstream of the confluence with Cabin Creek. These sites were added to provide an assessment of the aquatic invertebrate fauna on two tributary streams and three sites not affected by impoundments. Also within the basin, three sample sites on the Colorado River downstream of WGD were added to provide a better longitudinal perspective in that reach of the study area. The Highway 40 Bridge site (approximately 3 km east of Hot Sulphur Springs) is located between WG12 and WG13, well upstream of the confluence with the Williams Fork River. The Skylark Ranch site was added in April 2011 as an additional site in Reach Two, between WG 21 and WG22, upstream of the confluence with Troublesome Creek. The Pumphouse sample site is located downstream of the mouth of Gore Canyon, near the Bureau of Land Management (BLM) Pumphouse Boat Launch. This site is well downstream of the Blue River confluence. Blue River water releases from Green Mountain Reservoir during the spring to fall season (to service senior water calls downstream) essentially restores a more natural hydrograph to the Colorado River by providing flushing flows on an annual basis. In average and above average water years those flushing flows can be very high and sustained for several weeks.

Finally, the seventh site was on the Gunnison River, within the Black Canyon of the Gunnison River Gorge (GRG) downstream of Black Canyon National Park, and downstream of the largest reservoir system in Colorado. Since the width of river running through the GRG is only 10-20% greater than that of the Colorado River at site WG11, this study site was added as an "external" control, for comparative purposes. The average width of the Gunnison River at the invertebrate sampling site is only 6% wider than that of the Colorado River at WG21. It has a managed flow regime downstream of the largest capacity reservoir system in Colorado.

Approximately 33% (over 300,000 AF/yr) of the average annual water yield of the Gunnison Basin at Blue Mesa Dam is diverted through the Gunnison Tunnel to the Uncompahgre River Basin for agricultural, municipal and industrial purposes. The Curecanti (Aspinall) Unit on the Gunnison River is a series of dams that are part of the Colorado River Storage Project. It consists of three reservoirs, Blue Mesa, Morrow Point, and Crystal Dams. Blue Mesa Dam has a total storage capacity of slightly less than 1 million AF of water, the approximate average annual water yield of the Gunnison River basin upstream of Blue Mesa. The Blue Mesa and Morrow Point units operate as peak-power hydroelectric generation facilities. Each unit requires a volume of 5,000 ft^3/s under full load. Bypass flows of 150 ft^3/s occur when the turbines are not operating. Crystal Dam captures the water flowing from the two upstream hydropower units, and stabilizes the flows released to the river in the Gunnison Gorge.

Sampling Protocols - Three replicate samples were collected at riffles with visually similar depth, substrate, and velocity at each sampling location. For each replicate, a 0.25m^2 rectangular-shaped sampling frame was placed on the stream bed and a trailing, cone-shaped benthic drift net (150 micron mesh) over 2 m in length was set up immediately downstream and held in place with steel rods driven into the substrate. Wings attached to the sides of the cone-shaped net extended 1 m upstream, directing the flow and dislodged macro-invertebrates into the net (Figure 2 in the Appendix). The length and total surface area of the benthic drift net was great enough to allow water velocities up to 2-3 m/s to pass through the cone without any resistance or backwash, thereby reducing the possibility of loss of dislodged invertebrates being able to bypass the throat of the net. All rocks and cobbles ≥ 50 mm in diameter were individually scrubbed with a vegetable brush and invertebrates washed into the net. The rocks were placed back into the stream outside of the sampling rectangle. Once cleared of rock and cobble, the remaining substrate within the frame was stirred by foot and hand action to a depth of about 10-12 cm to dislodge macroinvertebrates. Replicates were generally collected within 3-4 m of each other at each sampling site. The trailing net was rinsed with stream water to concentrate the sample at the end of the cone. The cone was then inverted and washed into a 20 L plastic pail. The sample was then drained, transferred by hand into 1.9 L (1/2 gallon) plastic jars, and preserved in 70% ethanol (ETOH). Three separate benthic drift nets were used at each sampling location. The nets were thoroughly rinsed, cleaned and examined for any material clinging to the net to prevent cross-contamination between stations.

Preserved samples were transported to a lab at Colorado State University for processing by Division of Wildlife invertebrate taxonomists. Specimens were sorted, identified to the lowest practical taxonomic level, counted, and entered into a database. All aquatic insect and invertebrate taxa data reported were standardized to 1 m^2 for comparative purposes between sample sites, seasons and years (Appendix Tables A1 through A12).

During the spring of 2010, nine sites (27 replicates) were sampled. All of the spring 2010 replicates were completely sorted and sampled (100%). During the late-summer of 2010, benthic collections were made at 12 locations (36 replicates). Six of those replicates were sampled at 50%, 30 at 100%, and the April 2011 (Skylark Ranch) replicates were sampled at 50%. A reference collection was prepared and deposited at the C.P. Gillette Museum of Arthropod Diversity at Colorado State University. Taxonomic name changes that occurred between 1980-1981 and 2010 required an alteration of identifications from1980-1981 data in a few instances in order to allow better comparison with the 2010 data. One example was moving all *Alloperla* sp. from the 1980-81 data to family Chloroperlidae in 2010 because in 1980-1981 the genus *Alloperla* was comprised of several subgenera that are currently recognized as separate genera.

***Pteronarcys californica* Sampling Protocols** - Among all replicate benthic samples collected during the 2010 study, total body length (mm) was determined for every *Pc* larva at every site. This afforded the development of a length-frequency distribution for all *Pc* larvae collected in the spring and fall. In addition, determination of sex for each *Pc* larva was also made. This was possible using techniques originally developed by Branham and Hathaway (1975) and described by Nehring (1987). Each *Pc* larva was examined under a binocular stereozoom microscope for the presence (or absence) of a vaginal "scar" on the ventral posterior edge of the 7th abdominal segment. Presence of the vaginal scar indicated the larva was a female. Absence of the scar indicated the larva was a male. This afforded determination of sex among *Pc* larvae down to 4-5 mm total body length (not including the length of the cerci or the antennae). Male *Pc* larvae in their 3rd and 4th years of life develop a curved shield or hood on the dorsal surface of the terminal abdominal segment that covers the male genitalia. During the study in the 1980s, numerous benthic samples were collected during the spring, early summer, late summer and fall seasons to determine the length frequencies distributions of the pre-emergent male and female *Pc* larvae (in April) and compare with the post-emergence length-frequency distributions in July, a month after emergence (Nehring 1983,1984,1985,1986, 1987). This sampling protocol afforded an accurate assessment of the minimum and maximum body length for each sex for pre-emergent *Pc* larvae. It also affords visual representation of growth increments and supports the conclusion that the life cycle of *Pteronarcys californica* in the Colorado River is four years post-hatching (see Figures 9 and 10 in the Appendix of the report).

In addition, during May and June 2010, we physically collected and counted the cast exoskeletons of *Pc* larvae at two – 30.5m (100 feet) sections of riffle areas along one bank of the Colorado River at all seven WG benthic sample collection sites, at the BLM Pumphouse sampling area, at three sites on the Fraser River upstream of WGD, and on the Gunnison River in the same area as the benthic samples were collected. We were able to determine the statistical correlation between the last instar, pre-emergent *Pc* larval density with *Pc* adult density by 1) comparing the number of pre-emergent *Pc* larvae at the benthic sites sampled in April 2010 (standardized to $1m^2$) with 2) the number of cast exoskeletons of *Pc* larvae collected and counted at the same riffle areas in June 2010. The *Pc* adult density was standardized to individuals/m^2 by dividing the total exoskeletons counted by the estimated stream reach cross sectional area (m^2) at those sites.

This component of the study required almost four weeks to complete as *Pc* larval emergence to the adult stage typically begins in the downstream reaches, where the water is warmer, and then moves upstream to the highest elevation reaches as the water warms. Monitoring of the water temperatures at each sampling site demonstrated that *Pc* larval emergence to the adult life stage in both the Colorado and Gunnison rivers was triggered when the water temperature at a given location reached 10˚ C. The 2010 snowpack in the Colorado River and Gunnison River basins was well above average, and the spring was cold and wet. *Pc* larval emergence began at the BLM Pumphouse Launch site May 28 and proceeded slowly upstream. Emergence of *Pc* larvae at the sample sites on the Fraser River was not complete until June 21st.

RESULTS AND DISCUSSION

Aquatic Macroinvertebrate Results - The results of the spring 2010 benthic invertebrate samples are summarized in Tables A.1 through A.6 in the Appendix of the report. Results of the fall 2010 benthic invertebrate samples are summarized in Tables A.7 through A.12 in the Appendix. The data are all standardized and reported as numbers (per taxa) per m^2.

Benthic samples were collected from 14 stations in the Upper Colorado River basin between April 2010 and April 2011. Ten stations were from the Colorado River (five were the same locations as 1980-1981 stations, one from Willow Creek, two from the Fraser River, and one from the Gunnison River. Unless otherwise noted, this summary refers to the seven WG stations taken in spring and fall 2010. Table 1 above contains a matrix of the sample sites and sampling periods.

Sixty-nine macroinvertebrate taxa were identified from the Colorado River at seven WG stations in 2010 (Table 2). The insect taxa were primarily represented by 67 species in five orders. The station closest to Windy Gap dam (WG11) displayed the lowest diversity of total taxa and the middle station (WG21) displayed the highest, with a general increase in diversity at sites further downstream from the WGD (Table 2).

Mayflies (Ephemeroptera), stoneflies (Plecoptera), and caddisflies (Trichoptera), known collectively as the EPT taxa, are considered sensitive to changes in habitat and are widely accepted as a means to measure stream health (Gaufin and Tarzwell 1952, Barbour et al. 1999, Lenat and Penrose 1996) and are the focus of this brief summary. Eight EPT taxa that were common in the lower WG stations were rare or absent at the upper WG stations including the mayflies *Drunella grandis*, *Epeorus* sp., and *Rhithrogena* sp., the stoneflies *Isogenoides* sp., *Pteronarcella badia, Pteronarcys californica,* and *Cultus aestivalis,* and the caddisfly *Psychomyia flavida*. There were five EPT taxa unique but not common, in the upper WG stations including the stoneflies *Paracapnia angulata* and *Prostoia besametsa* and the caddisflies

Amiocentrus sp., *Agraylea* sp., and *Psychoglypha* sp. Simulids, Isopods, and segmented worms, organisms considered more tolerant to environmental perturbations, were more abundant in samples from the upper stations.

Table 2. Number of taxa from each station in spring and fall 2010 individually and combined and distance in river miles downstream from Windy Gap Dam. The data presented here are summarized from Appendix Tables A.1 through A.12

69 taxa total	WG11*	WG12	WG13	WG21	WG 22	WG31	WG32
Mayflies	10	11	9	15	11	10	13
Stoneflies	6	6	10	9	9	9	9
Caddisflies	9	14	10	10	11	13	9
Beetles	5	4	4	3	4	3	3
Dipterans	5	7	5	5	6	7	5
Non-Insects	11	11	6	12	10	8	8
Other Insects	0	1	0	1	1	0	1
Species per station	46	54	44	55	52	50	48
Distance↓WGD (km)	1.5	3.2	16.1	28.2	39.5	41.1	47.6

***** = fall sample only

The stations upstream of WGD (Willow Creek and Fraser River) exhibited the greatest diversity of macroinvertebrate fauna, harboring 29 taxa not found in stations downstream from WGD in 2010. Willow Creek had the highest number of taxa with 53. Nine were unique to that station. See Appendix Tables A.1 through A.12 for details.

Table 3. Numbers of species in each major group of aquatic macroinvertebrates from Windy Gap sampling stations in 2010 and 1980-1981. The data presented here are summarized from Appendix Tables A.1 through A.12

WG Stations	Total	Mayflies	Stoneflies	Caddisflies	Beetles	Diptera	Other insects	Non-insects
1980-1981	95	35	14	21	7	26	2-4	11
2010	69	16	10	13	7	8*	2	13
% change ± since 80-81	-38%	-54%	-40%	-62%	0%	-69%	0	+18%
All stations 2010	98	21	17	20	10	12	2	16

*****= Probably an underestimate as true flies were identified with more resolution in 1980-1981 than in 2010.

Although 2010 data exhibited greater diversity with increasing distance downstream, there was an approximate 38% loss of total diversity from 1980-1981 to 2010 (Table 3). The three most sensitive insect groups (EPT) experienced losses from 1980-1981 to 2010 including

19 mayfly, four stonefly, and eight caddisfly species. As sensitive species like *Pteronarcys californica*, *Pternarcella badia*, and *Drunella grandis* were eliminated below Windy Gap Dam, density of more tolerant taxa like the mayflies *Ephemerella dorothea infrequens*, some *Baetis* sp. and the caddisfly *Hydropsyche* sp. increased. This pattern agrees with Vinson (2001), Ward and Short (1978), and Ward (1976a) who showed aquatic macroinvertebrate diversity decreased while density increased below dams in large and small streams.

Lower numbers of stonefly species and individuals were apparent in the upper stations but appeared to slightly recover further downstream (Figure 3). Since 1980-1981 however, the percentages of stoneflies have declined by 40% with the greatest losses at stations nearest WGD. During the four sampling periods from September 1980 to late-August 1981 (prior to the dam construction), stoneflies were often the dominant order and the greatest contributor to total invertebrate biomass (Erickson 1983).

Pteronarcys californica (the giant stonefly) - The stonefly most noticeably absent from the upper stations in the benthic samples for both spring and fall 2010, was the salmonfly or giant stonefly, *Pteronarcys californica (Pc)*. Erickson (1983) provided a detailed, longitudinal assessment of biomass and density of *Pc* larvae in the 1980-1981 study, highlighting the critical ecological role of this species and its importance for measuring stream health. Examination of the relative abundance of *Pc* nymphs in Appendix Tables A.2 (spring 2010) and A.8 (fall 2010) reveals that this species was present in benthic samples at all sites upstream of WGD. Those sampling sites include the Fraser River at Kaibab Park (FRRKBP), Fraser River at Solvista (FRRSOL), and even in Willow Creek (WLOCRK) approximately 1 km upstream of the confluence with Cabin Creek. It is especially noteworthy that *Pc* nymphs and larvae of a closely related stonefly *Pteronarcella badia* did not occur in the benthic samples in the spring or fall 2010 at sites WG11 and WG12, the two sampling locations that are nearest WGD (see Table 2 for the approximate downstream distance). Moreover, WG12 was the one site during the 1980-1981 study that consistently had the highest densities of *Pc* larvae (Erickson 1983) as can be seen in Figures 3, 5 and 6 in the Appendix).

Percent density in m^2 of *Pteronarcys californica* (*Pc*) larvae in 2010 plotted longitudinally by station (Figure 4, Appendix tables A2 and A8) varied throughout the study reach. No *Pc* larvae were found in any of the replicate benthic samples collected at WG 11 or WG12 during either the spring or fall surveys in 2010. It was at WG12 where they were most abundant in 1980-1981. All stations below WG12 had detectable percentages of *Pc* larvae, although some of them were at extremely low levels. The only station with an increase in *Pc* larvae in 2010 compared to the 1980-1981 study was WG13 in the September 2010 collection (Figure 5). The Pumphouse station, the furthest station from WGD, had the largest percent *Pc* in 2010 and Gunnison River station had the second (Figure 4). Both of these reaches of river experience large, sustained flushing flows in all average and above average water years (see Table 6). All other stations (HWY 40 Bridge, WG21, Skylark, WG31 and WG32) had low to extremely low percentages of *Pc* in 2010. *Pc* larvae were present at all sampling sites (WLO, SOL and FRKBP) above WGD on the Fraser River and Willow Creek in the fall of 2010 (see Appendix Tables A2 and A8 for details on the data).

It is important to understand that even though stonefly larvae and in particular *Pc* larvae do not comprise a great amount of the total benthos sample (when expressed numerically or as a percentage of the total benthos), physically they are the largest aquatic insect larvae by weight and/or volume, and therefore often comprise the majority of the aquatic insect biomass. This phenomenon was most evident in the 1980-1981 study (Erickson 1983). Across all four sampling periods in that study, stonefly larvae comprised between 2.5% and 21.7% of the EPT and Diptera (D) invertebrate density at the seven WG sampling stations. However, stonefly larvae comprised an average of 42.7% of the total EPT&D biomass (g/m^2) across all sampling sites and dates. Among the five stations between WGD and the confluence with Troublesome Creek, the contribution of stonefly larvae to the EPT&D biomass ranged from a 37.2% to 67.3% and averaged 50.1% (see Table 3.5-2 on page 3.5-35 in Erickson 1983 for details).

Pc larvae often contributed more than 50% to the total wet weight biomass (g/m^2) of all aquatic invertebrate organisms in the 1980-1981 study. This was particularly true at sampling site WG12 approximately 3 km (2 miles) downstream from WGD (See Figures 3.5-48 through 3.5-55 in Erickson 1983). At WG12, *Pc* larvae comprised 34 to 37 g/m^2 wet weight biomass on three of four sampling occasions, while total wet weight biomass ranged between 45 and 50 g/m^2. (See Figures 3.5-48 through 3.5-51 in Erickson 1983 for details). In a separate study *Pc* larval densities at a collection site below the Byers Canyon Bridge west of Hot Sulphur Springs (synonymous with WG 13 in the Erickson study), Anderson et al. (1984) demonstrated that *Pc* larval densities in May 1983 (prior to adult emergence) ranged up to $1,040/m^2$ and accounted for 94% of the total dry weight invertebrate biomass. Dry weight biomass for *Pc* larvae was 35.3 g/m^2 while total dry weight invertebrate biomass was 37.4 g/m^2. Thirty years later the contribution of the giant stonefly to the total benthic invertebrate density or biomass at sampling sites WG11 and WG 12 are below detectible levels (see Figures 4,5,6,7 and 8 in the Appendix of this report).

The m^2 density of *Pc* larvae exhibit an overall loss from 1980-1981 to 2010 (Figure 5). They were completely eliminated at WG11 and WG12, the two stations closest to WG. All other stations, WG21, WG22 and WG32, saw a reduction in the number of *Pc* larvae since 1980-1981 except WG13. Not until 10 miles downstream of WGD at WG 13, was there an increase of individuals from 1980-1981 to 2010. It is important to point out that the density of all benthic macroinvertebrates in 2010 was much higher than in 1980-1981 (Average density of WG stations 1980-1981 = 4,568; average density of WG stations from 2010 = 9,917). With this in mind, the decrease in *Pc* larvae numbers is largely understated when considering numbers of individuals. The decrease in *Pc* larvae (and therefore adults) has a much larger corresponding ecological impact.

We also analyzed the percent *Pc* of all benthic density. Figure 6 compares the percent *Pc* by season for the WG stations in 1980-1981 and 2010. *Pc* percentages are reduced in both seasons at all stations in 2010 except for WG13 (0.06% in spring 2010 from 0.00% in spring 1981 and 2.35% in spring 2010 from 2.03% in spring 1980-1981). Two stations increased in *Pc* percentage in just one season, station WG22 increased from 0.04% to 0.37% in spring and WG31 from 0.00% to 0.05% at station in fall, but spring and fall combined there was an overall loss at those stations. Percentages of *Pc* at stations WG11 and WG12 are reduced to 0.00%. This is especially significant at station WG12 because historically this station had the highest percentage of all stations.

Spring and fall percentages of *Pc* larvae were averaged together by year and station to approximate the overall trend of *Pc* from 1980-1981 to 2010 (Figure 7). The populations appear to follow a similar trend from 1980-1981 to 2010, except at WG11 and WG12 where they were below detectible levels in the benthic samples. This trend clearly shows a common stressor that has negatively impacted *Pc* larvae populations throughout the entire reach of the Colorado River from WGD to the confluence with the Blue River. High quality habitat in 1980-1981 at WG11, WG12, and WG22 experienced 100% and 67% losses and at apparent marginal habitat (WG21 and WG31), small populations were nearly extirpated (Figure 8). Station WG31 was the only station to increase in *Pc* (+0.02%). WG13 had the smallest decrease in percent *Pc* (-11.03%). All other stations experienced more drastic decreases in percent of *Pc*, from -67.07% to -100.00% (Figure 8).

During May and June 2010 and again in June 2011, we collected and counted cast exoskeletons of emergent *Pc* adults at 10 locations on the Colorado River beginning 100 m upstream of the Grand County Road 57/578 Bridge (also known as the Hitching Post Bridge) downstream to the BLM Boat Launch recreation site downstream of Gore Canyon, and at two or three locations on the Fraser River upstream of WGD. For comparative purposes, we also collected and counted exoskeletons of emergent *Pc* adults at two sites on the Gunnison River in June 2010, and at 15 sites throughout a 30 km reach of the Rio Grande in SW Colorado during June 2011. At each location, cast exoskeletons were collected and counted along two - 30.5 meter (100 foot) reaches of bank at the riffle areas where the benthic invertebrate samples were collected in April 2010. Results of this portion of the study are summarized in Table 4. The data are arranged in an upstream to downstream manner. Moderate numbers of *Pc* exoskeletons were seen at two of three sampling sites on the Fraser River upstream of WGD. It is noteworthy that among the sampling sites where the substrate type was considered “prime” (cobble and boulder substrates), the fewest *Pc* exoskeletons were found at the two sites (new WG11 and WG12) which are in closest proximity to WGD. Moreover, it was at site WG12 where the density of *Pc* larvae was consistently the highest among all seven sampling sites during the 1980-1981 study (Erickson 1983).

The numbers of exoskeletons of emergent *Pc* adults counted at all sampling sites on the Colorado River in 2011 stayed approximately the same or increased substantially compared to 2010 except for the first 3 sample sites downstream of WGD (new WG11, WG12 and Hwy 40 Bridge. At these three sites the numbers of *Pc* exoskeletons declined more than 90% in 2011 compared to 2010. In contrast, the number of *Pc* exoskeletons counted on the Fraser River in June 2011 increased by 355% and1,270% (compared to June 2010) at the Kaibab Park and Solvista sites, respectively. (See Table 4 and Figure 14 in the Appendix for details.)

There were only three sampling sites where the substrate type was considered poor, Erickson’s WG32, the new WG32 and WG31. All three sites are downstream of the Troublesome Creek confluence and receive large amounts of sand and silt input due to the highly erosive nature of the drainage. Not surprisingly, very few *Pc* exoskeletons were present at these sites (Table 4).

Among the sampling areas where the substrate type was considered prime habitat for *Pc* larvae, it is noteworthy that the highest exoskeleton counts were at the BLM Pumphouse Launch area and the Gunnison River at Ute Park. In average and above average water years, both of these stream reaches experience flushing flows ranging from 1,000 to 8,000 ft^3/s. These discharges often extend over several weeks each year (see Table 6 below).

At a given sampling site, *Pc* larval emergence was always completed within three to four days after it began. *Pc* larvae begin migration out of the water at dusk, crawl onto surrounding vegetation or debris, such as rocks or wood, lock their legs onto the surface, and split down the dorsal midline beginning in the thoracic area. The adult extracts its body from the exoskeleton by pushing and flexing upward and outward. The wings emerge first (and begin drying immediately) then the head, and finally the abdomen. The entire process is usually accomplished

Table 4. Population estimates of *Pteronarcys californica* nymphal exuviae (exoskeletons) per 30.5 m or 100 feet of stream channel along one bank within prime riffle habitat at various sampling sites on the Colorado River during late May and early June 2010 and 2011 compared with sampling sites on the Gunnison River downstream of Blue Mesa, Morrow Point and Crystal dams (2010) and the Rio Grande between Creede and South Fork, Colorado in June 2011.

Date	Site Description/≈ Distance in miles from Windy Gap Dam (WGD)	N	2010 Ave.	Range	2011
06/10	Fraser River on Solvista Ranch 5 mi.↑ WGD - prime habitat	2	122.5	46 – 199	1,689
06/10	Fraser River @ Granby ballpark 4 mi.↑ WGD - prime habitat	2	199	168 – 230	706
06/10	Fraser R. @ Hackstaff Ranch ≈ 1 mi. ↑ WGD - moderate habitat	2	19.5	4 – 35	-----
06/10	New WG 11 ≈ 0.9 mi. ↓ WGD - prime habitat	2	25	17 – 33	2
06/10	Erickson's WG 12 ≈ 2 mi. ↓ WGD- prime habitat	2	27.5	25 – 30	1
06/10	Hwy 40 Bridge E. of Hot Sulphur Springs ≈ 5.0 mi. ↓WGD - prime	1	280	270	26
06/10	Erickson WG 13; ↓ Byers Canyon Bridge ≈ 10.2 mi↓ WGD – prime	2	992	797 – 1,186	2,134
06/10	Erickson's WG 21≈ 18.5 mi.↓WGD –prime habitat	2	352	343 – 361	252
06/10	Erickson's WG 22≈ 21.7 mi.↓WGD – prime habitat	2	129	69 - 188	302
06/10	Erickson's WG 31 ≈ 22.7 mi.↓WGD - sediment laden poor habitat	2	1	0 – 2	187
06/10	New WG 32 ≈ 27.5 mi.↓WGD - sediment laden poor habitat	2	2	0 – 4	19
06/10	Erickson's WG 32 ≈ 28.9 mi.↓WGD –sediment laden poor habitat	2	0	0	0
06/10	Pumphouse Area in Gore Canyon ≈ 41 mi.↓WGD – prime habitat	2	5,514	4,195 - 6,833	6,225
06/10	Gunnison River Gorge in Ute Park – prime habitat	2	3,500	2,400 – 4,600	-----
06/11	Rio Grande between Creede and South Fork, CO	15	690.2	101 – 2,242	690.2

within 10-15 minutes. At all sampling sites upstream of the Blue River confluence, each exoskeleton sampling location was visited at least four times over a four to seven day period, beginning at or prior to the beginning of emergence, and continued until emergence was complete, as determined by the absence of additional exoskeletons. All exoskeletons were tabulated when collected with a hand-held counting device, collected and stored in a ziplock bag to insure that no double-counting of exoskeletons occurred at any location. At those sampling sites where larval densities were low to moderate, we separated exoskeletons by sex and were able to determine that on the first night male *Pc* larvae account for 75-100% of the emergence. The second night the male to female sex ratio usually ranged from 60:40 to 50:50. The third night the ratio was usually 25:75 to 40:60, and the forth night the female larvae accounted for 75-100% of the emergence. Peak emergence invariably occurred on the second or third evenings. The phenomenon of male *Pc* larvae emergence preceding the females by a day or two is congruent with the findings of Richards et al. 2000, on the Madison River in Montana.

Male *Pc* larvae are substantially smaller than female larvae. The results of length-frequency measurements of pre-emergent male and female larvae in April 1987 and July 1987, one month after emergence of the adults indicated that the body length of last instar female larvae ranged from 36 mm to 50 mm, while last instar male larvae ranged from approximately 33 mm to 40 mm in length (see Appendix Figures 2, 3, 4 and 5 in Nehring 1987). Standardization of densities of pre-emergent *Pc* larvae from the April 2010 benthic invertebrate samples and the *Pc* exoskeleton counts to numbers per m^2 afforded a statistical correlation to see if there was a significant relationship between the two independent measures. The correlation coefficient (r) was 0.9505 with 23 observations, a highly significant relationship.

Two doctoral research studies in the last half of the 20th century (Knight 1965, Poole 1981) suggested that *Pteronarcys californica* most likely had a three-year life cycle. However, subsequent studies of the population dynamics of the giant stonefly on the Colorado River that began in 1982 and continued into 1989 concluded that this species had a four-year life cycle after the eggs had gone through a diapause phase of 90 days or more prior to hatching (Nehring 1987). Body length measurements made on every male and female *Pc* larvae collected on the Colorado River in 2010 summarized in length-frequency distribution graphs support that conclusion (see Figures 9A, 9B, 10A and 10B in the Appendix, as well as Figures 2,3 and 4 in Nehring 1987). Our determination of a four-year life cycle is congruent with the findings of Dewalt and Stewart (1995) and Townsend and Pratt (1998).

Other stoneflies - During the 1980-1981 study, *Pteronarcella badia (Pd)* larvae were common in the collections at the first five sampling sites below WGD (WG11, WG12, WG13, WG21 and WG22) upstream of the confluence with Troublesome Creek. They were rarely collected during the 2010 field season and were not observed at all in the fall 2010 samples in Reach One between WGD and the confluence with the Williams Fork River. It is especially noteworthy that *Pd* larvae were very abundant in all three replicates collected in Willow Creek upstream of Willow Creek Reservoir in September 2010. The estimated density of *Pd* larvae at the Willow Creek site was 117/m^2 compared to only one and 3/m^2 at two of five WG sites between WGD and the confluence with Troublesome Creek (see Appendix Tables A2 and A8 for details).

Total stonefly (Plecoptera) densities in Reach One (WG11, WG12 and WG13) of the Colorado River tended to be lower in 2010, compared to the levels observed in 1980-1981 (Figure 3). In river reaches two and three, the densities were higher in 2010 compared to 1980-1981. At sample sites WG22 and WG 32 the densities in 2010 were much higher, primarily due to very high numbers of three taxa belonging to the families Chloroperlidae and Perlodidae (*Swallia* sp., *Cultus aestivalis* and *Isoperla* sp.). Detailed comparisons of the relative abundance of stonefly larvae can be reviewed in Appendix Table A2 and A8.

Mayfly larvae – Detailed estimates of density and relative abundance of the order Ephemeroptera (mayflies) can be reviewed in Appendix Tables A1 and A7. Those data are summarized graphically in Figure 11 in the Appendix. The percentage of mayfly larvae in the benthic samples in 2010 were substantially higher in Reach One of the Colorado River in 2010 compared to 1980-1981. The differences between the two sampling periods (1980-81 vs 2010) decreased in river reaches two and three (Figure 11 in the Appendix). However, the increased numerical density in 2010 was the result of a high abundance of four taxa of Ephemeroptera (*Baetis* sp., *Ephemerella dorothea infrequens, Rithrogenia* sp., and *Paraleptophlebia* sp.). Larger ephemeropteran species that were much less numerous or totally absent throughout most or all of the WG study sites in 2010 that were common and abundant in the 1980-1981 study include *Drunella grandis, Drunella doddsi,* and *Baetis tricaudatus*.

Caddisfly larvae - Detailed estimates of density and relative abundance of the order Trichoptera (caddisflies) can be reviewed in Appendix Tables A3 and A9. Those data are summarized graphically in Figure 12 in the Appendix. Tricopteran taxa that were dominant in the 2010 collections included *Brachycentrus* sp., *Glossosoma* sp., *Hydropsyche* sp., and *Lepidostoma* sp. Species that were present and/or abundant at Reach One study sites WG11 and WG12 for most or all of the 1980-1981 period but greatly diminished in number or totally absent at these two sites nearest WGD in 2010 include *Psychomia flavida,* and the large free-living taxa *Arctopsyche* sp. and *Cheumatopsyche* sp.

True flies (Diptera) larvae – The densities and relative abundance of dipteran larvae were higher at all WG sampling sites in 2010 compared to the 1980-1981 study. Two families (Simuliidae and Chironomidae) comprised more than 90% of the density for this order of insects at virtually all sampling sites for both seasons in 2010. Both of the families have numerous taxa that go through several life cycles each year and are known to thrive in altered aquatic ecosystems. Detailed estimates of density and relative abundance of the order Diptera (true flies) can be reviewed in Appendix Tables A4 and A10.

Beetle (Coleoptera) larvae – two taxa (*Zaitzevia parvula* and *Optioservus* sp.) dominated the collections for the order Coleoptera at all WG study sites in 2010. Detailed estimates of density and relative abundance of the beetle larvae can be reviewed in Appendix Tables A5 and A11.

Other Macroinvertebrate fauna - Detailed estimates of density and relative abundance of non-insect invertebrate fauna encountered during the collections of 2010 can be reviewed in Appendix Tables A6 and A12. These taxa included scuds (Amphipoda), sow bugs (Isopoda), dragonfly larvae (Odonata), mollusks (Bivalvia), zooplankton (Cladocera), leeches (Hirudinea), aquatic worms (Oligochaeta), snails (Gastropoda), nematodes (Nematoda), springtails (Collembola), and flatworms (Turbellaria).

Impact(s) of Altered Discharge on Stream Ecosystems – In this study, the greatest numbers of aquatic insect taxa were present at the study sites upstream of WGD on the Fraser River and Willow Creek (upstream of Willow Creek Reservoir). Among the study sites downstream of WGD, it is especially noteworthy that the station closest to the dam (WG11) displayed the lowest diversity of total taxa. In contrast, station WG21 in Reach Two displayed the highest diversity. This site is downstream of the confluence with the Williams Fork River, where discharge levels are greater due to water releases from Williams Fork Reservoir during the summer and early fall months, particularly during average and below average water years. However, sampling sites upstream of WGD on Willow Creek (upstream of Willow Creek Reservoir) and on the Fraser River exhibited the most diverse macroinvertebrate fauna, harboring 29 taxa not found in stations downstream from WGD in 2010. Willow Creek had the highest number of taxa with 53. Nine taxa were unique to that station. Willow Creek at this site is a free-flowing stream, unimpacted by water storage or diversions. This pattern of decreasing diversity of aquatic insect taxa downstream of impoundments is not uncommon (Vinson 2001; Ward and Short 1978; Ward 1976a,b).

A review of the stream discharge patterns for the Colorado River downstream of WGD from 1977 through 2010 (utilizing mean daily discharge data from the two stream gages below WGD and at Hot Sulphur Springs) is informative. That data is summarized in Table 5. It is evident that there has been a very large decline in the occurrence, amplitude and duration of true flushing flows since diversions of water at WGD began to occur in the mid to late 1980s. This pattern has been the most evident since the year 2000. Landowners and local fly fishermen with an intimate knowledge of the river since the early 1970s began to notice substantial declines in abundance of the adult giant stonefly *Pteronarcys californica* and larger adult mayflies like the green drake *Drunella grandis* during the past ten years.

As indicated earlier in this report, the greatest densities of *Pc* larvae and emergent adults in the upper Colorado River basin were present in the reach of river below the Blue River confluence near Kremmling, at the BLM Pumphouse boat launch access point. Similar densities of *Pc* larvae and emergent adults were found on the Gunnison River in the Gunnison River Gorge, downstream of the Aspinall or Curecanti Unit of the Colorado River Storage Project, with three mainstem impoundments with a total storage capacity > 1 million AF. Finally, densities of both *Pc* larvae (in the spring and fall benthic samples) and emergent adults (in the riparian surveys in June) were substantially higher on the Fraser River at the survey sites at Kaibab Park and Solvista compared to sampling sites WG11 and WG12, the two sites closest to WGD (see Table 4 above and Appendix Tables A2 and A8).

A summary of the discharge records for the Fraser River at Granby, the Colorado River downstream of the Blue River confluence near Kremmling (upstream of the BLM Pumphouse boat launch sampling sites) and the Gunnison River in the Gunnison River Gorge are shown in Table 6. There is a marked increase in the number of days, amplitudes and duration of flushing flows ≥ 1,000, 2,000, 3,000 and 4,000 ft^3/s for the Colorado River below Blue River confluence and in the Gunnison Gorge in comparison to that experienced by the Colorado River in Reach One, downstream of WGD. As alluded to earlier, the average width of the Gunnison River at the sampling site in the GRG is only 10-20% greater than that of the Colorado River at sampling sites WG11 and WG12. There have been only 2 days when the discharge of the Colorado River at WGD exceeded 2,000 ft^3/s for the water years 1998 through 2010. In contrast, discharges in the Gunnison River at Ute Park have exceeded 2,000 ft^3/s 182 days for the water years 1999 through 2010. The last time the Colorado River below WGD experienced discharges exceeding 3,000 or 4,000 ft^3/s was in 1997 (Table 5). There have been none since then through the 2010 water year. In contrast, the Gunnison River at Ute Park has experienced flows exceeding 3,000 and 4,000 ft^3/s for 151 days and 27 days, respectively (Table 6). Similar discharge scenarios are obvious for the Colorado River below the Blue River confluence. It is possible that the amplitude and duration of scouring flushing flows are related to the densities and relative abundance of *Pc* larvae and emergent adults at these two locations compared to sites WG11 and WG12.

Table 5. Number of days when the average daily discharge at the Windy Gap or Hot Sulphur Springs gage were equal to or exceeded various flows, for the water years from 1977 through 2010. Three dashed lines in the≥ 500 ft^3/s column indicate none were counted since in those years there were days when the average daily discharge was ≥ 1,000 ft^3/s.

Water Year	≥ 1,000 ft^3/s	≥2,000 ft^3/s	≥3,000 ft^3/s	≥4,000 ft^3/s	≥ 500 ft^3/s
1977	0	0	0	0	0
1978	7	0	0	0	---
1979	19	0	0	0	---
1980	14	0	0	0	---
1981	0	0	0	0	---
1982	0	0	0	0	---
1983	61	28	6	2	---
1984	76	53	28	7	---
1985	11	0	0	0	---
1986	53	0	0	0	---
1987	1	0	0	0	---
1988	2	0	0	0	---
1989	0	0	0	0	0
1990	0	0	0	0	0
1991	0	0	0	0	18
1992	0	0	0	0	0
1993	21	0	0	0	---
1994	0	0	0	0	12
1995	49	4	0	0	---
1996	62	17	3	0	---
1997	56	31	10	7	---
1998	4	0	0	0	---
1999	33	0	0	0	---
2000	13	1	0	0	57
2001	0	0	0	0	0
2002	0	0	0	0	0
2003	5	0	0	0	10
2004	0	0	0	0	0
2005	0	0	0	0	17
2006	0	0	0	0	7
2007	0	0	0	0	15
2008	5	0	0	0	50
2009	9	0	0	0	67
2010	25	1	0	0	46

Perhaps the most valid comparisons that can be made are for the stream discharge data for the Fraser River at Granby and the Colorado River below WGD. At first glance there does not appear to be a great deal of difference in the occurrence and amplitude of flushing flows between these two gaging stations. However, it is important to understand that the average channel width at the Kaibab Park sampling station on the Fraser River is only 10.2 m (33.4 feet) compared to 27.9 m (91.6 feet) at WG11 (Hitching Post Bridge) below WGD. Therefore, the stream power available for effective sediment transport and flushing flows at the Kaibab Park station on the Fraser River is considerably greater. It is possible that this could contribute to the somewhat higher densities and relative abundance of *Pc* larvae and emergent adults at this site relative to WG11 and WG12 below WGD (as shown in Table 4 in the text and Appendix Tables A2 and A8).

There are other complicating factors that can have serious long-term negative consequences for the proper biotic functioning and ecological integrity of stream ecosystems that result from mainstem impoundments, especially when diversions of substantial amounts of water are involved with Granby, Willow Creek and Windy Gap reservoirs. These complicating factors include 1) loss of channel connectivity, 2) elevated water temperatures, 3) fine sediment deposition and transport, and 4) rooted aquatic vegetation.

Loss of Channel Connectivity - Windy Gap Reservoir (WGR) has been a mainstem impoundment on the Colorado River at Granby since the early 1980s. Since it came on line, movement of fish species up and down the river has been interrupted. It has also interrupted downstream drift of aquatic insect larvae that is a critical function of free-flowing streams for maintenance of the biological integrity of the stream benthos community (Townsend and Hildrew 1976). Numerous adult aquatic insects often fly upstream from the point of emergence and lay their eggs. Once hatched, aquatic insect larvae drift downstream during the night. Nocturnal drift is especially heavy during the dark phases of the lunar cycle, from the last quarter through the new moon and onto the first quarter. Nocturnal drift of benthic invertebrates in streams is critically important in maintaining the greatest diversity of aquatic invertebrate fauna in stream ecosystems.

The elimination of downstream invertebrate drift from the Fraser and Colorado rivers above WGD for the past 25 years has undoubtedly played a major role in the reductions or loss of 22 species of mayfly, stonefly and caddis fly that were either not found or have declined substantially in abundance since the 1980-1981 study. In 1980-1981, the giant stonefly, *Pteronarcys californica*, was found in greatest abundance at the sampling site less than 2 miles downstream from Windy Gap. Our 2010 studies demonstrate that it is almost gone from the river between Windy Gap and Hot Sulphur Springs. Two species of large mayflies that occurred at all seven sampling sites in 1980-1981 no longer occur upstream of the confluence with the Williams Fork River near Parshall. One of those species was not found at any of the seven sampling sites on the river between Windy Gap and Kremmling. However, all three species still occur in the Fraser River upstream of Windy Gap and in Willow Creek upstream of Willow Creek Reservoir.

Table 6. Number of days when the average daily discharge at the Fraser River at Granby, the Colorado River below Kremmling and the Gunnison River at Ute Park were equal to or exceeded various flows, for the water years from 1999 through 2010. Three dashed lines in the≥ 500 ft^3/s column indicate none were counted since in those years there were days when the average daily discharge was ≥ 1,000 ft^3/s.

Water Year	≥ 1,000 ft^3/s	≥2,000 ft^3/s	≥3,000 ft^3/s	≥4,000 ft^3/s	≥ 500 ft^3/s
Fraser River at Granby above Windy Gap Reservoir					
2000	0	0	0	0	9
2001	0	0	0	0	0
2002	0	0	0	0	0
2003	5	0	0	0	23
2004	0	0	0	0	0
2005	0	0	0	0	17
2006	0	0	0	0	4
2007	0	0	0	0	19
2008	4	0	0	0	49
2009	0	0	0	0	63
2010	11	0	0	0	30
Colorado River @ Kremmling below the Blue River confluence					
2000	174	19	1	0	----
2001	54	0	0	0	----
2002	0	0	0	0	160
2003	29	4	0	0	----
2004	18	0	0	0	----
2005	68	8	0	0	----
2006	109	5	0	0	----
2007	78	22	5	1	----
2008	156	68	25	16	----
2009	181	71	45	11	----
2010	136	35	21	11	----
Gunnison River at Ute Park					
1999	107	21	0	0	----
2000	145	14	70	3	---
2001	7	1	0	0	365
2002	0	0	0	0	250
2003	5	0	0	0	88
2004	1	0	0	0	82
2005	96	0	0	0	----
2006	99	0	0	0	----
2007	112	0	0	0	----
2008	318	82	56	17	----
2009	126	55	20	6	----
2010	13	9	5	1	----

Elevated Water Temperatures – Any time water is impounded and held in a lake or a reservoir, solar radiation will heat the water. In shallow impoundments such as WGR heating is faster than in large deep lakes. In late summer, at a time when the Colorado River flows are at the seasonal low and the stream water temperatures are already high, the risks to aquatic fauna are the greatest. Water temperatures have been monitored periodically for extended periods of time at numerous locations throughout the Colorado River basin in Grand County from 1980 into the early part of the 21st century. Those records indicate that daytime water temperatures can range from 20 the 25 ˚C, particularly during the late summer months during periods of drought (Nehring and Thompson 2001, 2003).

Townsend and Pritchard (2000) conducted extensive tests of the embryonic development and hatching success of eggs of *Pteronarcys californica* in laboratory exposures over a wide range of temperatures and with eggs placed in the Crows Nest River, Alberta, Canada. The results of those studies indicated the greatest hatching success, ranging from 50% to 75%, occurred at 15.0 – 17.5 ˚C. At temperatures < 10 ˚C and ≥ 20 ˚C, hatching success declined exponentially. At an incubation temperature of 20 ˚C, the percent hatch was only 25.1% and 29% in two tests, and dropped to 4.5% and 12% after 261 days of exposure at a temperature of 22.5˚C. Similarly, for *Pc* eggs incubated at temperatures of 5.0 and 7.5 ˚C, the percent hatch was 0.9% and 6.7%, respectively. In the high temperature exposures of 20˚C and 22.5˚C, first egg hatch occurred at days 194 and 200, respectively. At the low incubation temperatures of 5.0 and 7.5 ˚C, first hatch of *Pc* eggs occurred at day 526 and 757, respectively. These results indicate that the life stage from oviposition to egg hatching for *Pteronarcys californica* has a very narrow thermal window for optimum hatching success. That is the most likely explanation why this species does not reproduce successfully in the tailrace regions immediately downstream of large, deep, bottom-release dams (Vinson 2001). It also indicates that elevated water temperatures, especially during times of drought or anthropogenic reductions in stream discharge will result in dramatic reductions in egg hatching success for *Pc* eggs (Stagliano 2011).

Fine Sediment Deposition and Transport – Sediment deposition and armoring of the Colorado River below Windy Gap Dam has been greatly exacerbated over the past 10-20 years, due to extended droughts, impoundment and storage of spring flushing flows in Willow Creek and Granby Reservoirs, and depletions from transmountain diversions through the Moffat Tunnel and Alva B. Adams Tunnel at Grand Lake. At least twice since 2001, Windy Gap Dam has been drained and untold tons of sediment has been flushed into the Colorado River in mid to late summer, long after spring flushing flows were available to transport the sediment downstream. These sediments settle out in the largest amounts in the first few miles of river below the channel for two reasons. The Colorado River channel is over width by approximately 50% due to the withdrawals of flow upstream in the Colorado and Fraser River basins and therefore the stream power (to transport sediment) has been adversely impacted. Sediment deposition clogs the interstitial spaces between and beneath the rocks and cobbles in the riffle areas, setting up a series of cascading negative impacts on the stream ecosystem. Riffle areas are critical to maintenance of the biotic integrity of the stream on a number of levels. They are the primary zones of aquatic insect production. Aquatic insects are the primary source of food for foraging fish. Sediment accumulation among the gravel, cobble and boulder substrates leads to armoring of the stream channel, dramatically reducing the carrying capacity of the stream for benthic invertebrates and aquatic insects that do not tolerate sediment accumulation. Over time, invertebrate species diversity declines and the overall biotic integrity of the stream is reduced.

Richards et al. (2000) found that the number of emergent adult *Pteronarcys californica* stoneflies were negatively correlated with the percent embeddedness of the substrate at 11 study sites over consecutive years in the Madison River in Montana (r –0.70 and P=0.02 for 1994; r=-0.62 and P=0.04 for 1995). Stagliano (2011) concluded that elevated water temperatures, loss of habitat due to sedimentation and lack of flushing flows were the overarching factors implicated in the demise of the *Pteronarcys californica* population in the Smith River in Montana.

In contrast, it is noteworthy that a stream in western Colorado that experiences massive inputs of sediment annually during the spring run-off has continued to sustain populations of both *Pteronarcys californica* and *Pteronarcella badia* for more 35 years (1977 through 2010). The North Fork of the Gunnison River downstream of Paonia Reservoir right in the town of Somerset, Colorado (a center of major coal-mining activity) harbors a good population of both species (Nehring, unpublished data). Paonia Reservoir is substantially larger than WGR and it has received and trapped so much sediment over the past half century from West Muddy Creek, that much of the storage capacity of the reservoir has been lost. The question arises, *How can these two species of stoneflies continue to exist and apparently thrive in such an unfavorable environment*? The obvious difference is that all of the water flowing into Paonia Reservoir is stored only temporarily, for delivery downstream during the summer months for agricultural and municipal needs. However, during the spring snow melt and run-off, the reservoir fills and spills, and no water is diverted out of the stream channel until it has flowed past Somerset. Therefore, the stream power of the North Fork of the Gunnison River is not impacted by water storage and/or out of channel diversions during the period when the greatest amounts of sediment are mobilized and carried downstream. The North Fork of the Gunnison River is “exercised” annually by the normal spring snowmelt and run-off from the Grand Mesa, the Raggeds

Mountains, and the West Elk Wilderness area. Once the run-off is over, the continuing chronic input of sediment from West Muddy Creek settles out in the upstream end of Paonia Reservoir, and the sediment content of the outflow is low enough that these two species of stoneflies continue to exist. The same situation applies (in part) for the Colorado River in the Gore Canyon west of Kremmling, where an incredibly dense population of *Pteronarcys californica* thrives in the face of considerable sediment input. Yet, like the North Fork of the Gunnison River, the sediment input from Muddy Creek and other smaller erosive tributary streams remains mobilized by high levels of flushing flows on an annual basis.

Rooted Aquatic Vegetation – Over the past 10-20 years there has been an amazing expansion of aquatic vegetation mats in the Colorado River. That problem has been especially problematic between WGR and the confluence with the Williams Fork River at Parshall, Colorado. Due to the shallow nature of WGR, rooted aquatic vegetation quickly invaded and colonized all parts of the lake soon after construction was completed. Rich fertile soils, warm water temperatures and extensive sunlight provided optimum growing conditions for rooted aquatic macrophytes in WGR. These aquatic plants fragment throughout the summer months, flow out of WGR and settle out in the slower moving parts of the river. Once established they continue to grow in size and number over the decades, slowing the flow of water, trapping fine sediments and particulate organics that provide a nutrient base and substrate for growth and expansion of the vegetation mat. These can be seen everywhere on the river between WGR and the confluence with the Blue River. The problem is greatest between WGR and the confluence with the Williams Fork River. These vegetation mats also choke out the interstitial spaces in the cobble-strewn habitats throughout the river that are critical microhabitats for aquatic invertebrates, especially those species of aquatic insects that require sediment-free, cobble-rubble substrates.

Vegetation mats become increasingly difficult to dislodge or remove when true flushing flows have become all but a thing of the past as flow depletions have increased, particularly during the spring flush period. As an example, the Gunnison River in western Colorado, downstream of Black Canyon National Park (BCNP) is only about 10%-20% wider than the average channel width of the Colorado River in Reach One below WGR. The Gunnison River Gorge receives large sediment inputs during the summer months from side canyon flash floods that result from summer thunderstorms. These large inputs of sediment led to development of huge sediment laden vegetation mats during periods of extended drought. However, these vegetation mats and massive amounts of sediment have been largely eliminated from long sections of the Gunnison River Gorge by flushing flows $\geq$ 3,000 ft^3/s that are sustained for 2-4 weeks in many years, with peak flows ranging from 6,000 – 8,000 ft^3/s. The Gunnison River Gorge, only 10%-20% wider than the Colorado River below WGR rarely experiences flows below 300 cfs, due to minimum flow agreements. It is noteworthy that the Gunnison River downstream of BCNP has outstanding populations of the giant stonefly and sculpins, both of which coexist with perhaps the best stream trout sport fishery in Colorado. Predatory brown trout ≥ 500 mm in length occur throughout the Gunnison River and coexist in sympatry with sculpins and *Pteronarcys californica.*

CONCLUSIONS

Dave Stagliano (2011), an aquatic ecologist in Montana, published an article in the May-June 2011 issue of Montana Outdoors titled *Searching for Salmonflies: Scouring River Records for Data on Montana's Biggest Trout Bugs.* The article summarized the results of a year-long study to determine whether or not the numbers and populations of the salmonfly *Pteronarcys californica* in streams across Montana were in decline. The results were mixed and varied from stream to stream. Some populations seemed to be stable, some decreasing, some extirpated from long reaches of some rivers, and rarely, one was thought to be increasing. For those streams in Montana where populations of the salmonfly were in decline or extirpated, Stagliano concluded that elevated water temperatures and loss of habitat due to sedimentation and lack of flushing flows were the overarching factors implicated. The Smith River, a famous trout stream in SW Montana, historically had a large salmonfly hatch up into the early 1990s. However, between 1993 (when a large emergence occurred) and 1997, the population went into rapid decline. The salmonfly hatches on this river have been almost non-existent since 1997 according to one river guide who "*has been quantitatively sampling salmonfly larvae populations*" since 1991, according to Stagliano. In his article Stagliano states that his own *"anecdotal observations of the river's increased silt and water temperatures support his findings"*. The Smith River, approximately 160 km (100 miles) in length from the headwaters to the confluence with the Missouri River, is heavily diverted for irrigation and flows decline rapidly as the run-off from snowmelt decreases and diversions increase, rendering the river too wide and shallow for float fishing from a raft after mid-June in most years.

After studying the *Pteronarcys californica* population in the upper Colorado River in Middle Park throughout most of the 1980s, Nehring (1987) concluded (in part) the following: " *If Pteronarcys californica is to succumb to man-induced environmental stresses (that rudimentary populations of salmonids might withstand) it would probably be in the arena of pesticide* *pollution, heavy metal pollution such as cadmium (Colburn 1985), or loss of interstitial spaces among the boulder-cobble substrates to excessive silt accumulation. Loss of this critical portion of microhabitat through siltation probably poses the greatest threat to survival of this species in the upper Colorado River. The Windy Gap Project is just one more incremental assault on riverine aquatic habitats in the upper Colorado River basin that have been going on in Colorado for more than half a century. And it certainly will not be the last. … According to Ward (1984) the annual flow of the Colorado River was reduced 91% with the construction of Lake Granby. Without the annual spring run-off, silt rapidly choked and forever altered the stream habitat and benthic fauna of the Colorado River below Lake Granby. … Thus, if Pteronarcys californica disappears from the Colorado River it will probably be over a very long period of time due to the slow incremental accumulation of silt and loss of microhabitats, and not due to thermal and/or dissolved oxygen stress."*

Twenty-five years later it appears that prognostication has largely come to pass. The numbers of *Pteronarcella badia*, a closely related species that was common throughout the Colorado River from Windy Gap to the confluence with the Blue River in the early 1980s, have dwindled to barely detectible levels, particularly in Reach One. Likewise, the large mayfly species such as *Drunella grandis* and *Drunella doddsi*, also common in the same reach in the early 1980s are now gone. Among the caddisflies, *Arctopsyche* sp., *Cheumatopsyche* sp., and *Psychomyia flavida* were common in all collections at all seasons at site WG11 in the 1980-1981 study. They were rarely observed in 2010.

It is our conclusion that chronic sedimentation and clogging of the interstitial spaces in the cobble-rubble dominated riffles areas of the upper Colorado River below WGD is the overarching problem that has increasingly compromised the biotic integrity and proper function of the river over the past 25 years. The proposed firming projects at Windy Gap and the Moffat Tunnel are only going to further exacerbate this situation. Storage, impoundment and diversion of the waters of almost all creeks and major streams in Grand County have created and untenable situation that will soon be impossible to correct unless a massive investment and commitment of time, manpower and capital is forthcoming on a number of fronts.

The title of Job Number Two within this study (Colorado River Aquatic Resources Investigations) is Colorado River Mottled Sculpin Population Studies. After the conclusions section of Job Number Two, recommendations will be set forth for future study and approaches for continued monitoring of the upper Colorado River aquatic ecosystem in Grand County to help insure that this very important resource does not continue to degrade further. Given an adequate level of funding and a sustained investment of time and manpower, it should be possible to restore the Fraser and Colorado rivers to some semblance of a healthy coldwater stream ecosystem.

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Appendix For Job 1

Tables

and

Figures

Table A1. Estimated densities (numbers/m^2) of mayfly larvae at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the spring (April-May) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	Invertebrate Classification (Taxon - Genus/species/subspecies)	ORDER – Ephemeroptera – Mayflies													
		WLO CRK	FRR SOL	FRR KBP	COL NEW WG11	COL WG12	COL Hwy40 BR	COL WG13	COL WG21	COL SKY LARK[a]	COL WG22	COL WG31	COL NEW WG32	COL PUMP	GUNN
Ameletidae	*Ameletus* sp.	nss	nss	0	nss	0	nss	0	0	0	0	0	0	0	0
Baetidae	*Acentrella insignificans*	nss	nss	0	nss	0	nss	0	0	0	0	0	0	0	0
Baetidae	Baetidae*	nss	nss	103	nss	2,613	nss	755	472	nss	451	185	788	2,507	784
Baetidae	*Baetis bicaudatus*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Baetidae	*Baetis flavistriga*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Baetidae	*Baetis* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Baetidae	*Baetis tricaudatus*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Baetidae	*Diphetor hageni*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Baetidae	*Fallceon quilleri*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Baetidae	*Plauditis* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Caenidae	*Brachycercus* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Caenidae	*Caenis* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Ephemerellidae	*Attenella margarita*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Ephemerellidae	*Drunella doddsii*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Ephemerellidae	*Drunella grandis*	nss	nss	0	nss	0	nss	0	9	nss	9	1	7	4	0
Ephemerellidae	*Drunella* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Ephemerellidae	*Ephemerella dorothea infrequens*	nss	nss	475	nss	2,109	nss	1,169	856	nss	2,347	640	331	87	596
Ephemerellidae	*Ephemerella excrucians*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Ephemerellidae	*Ephemerella* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Ephemerellidae	Ephemerellidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Ephemerellidae	*Serratella micheneri*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Ephemerellidae	*Serratella* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Ephemerellidae	*Timpanoga hecuba hecuba*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Heptageniidae	*Cinygmula* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Heptageniidae	*Ecdyonurus simplicioides*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Heptageniidae	*Epeorus albertae*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Heptageniidae	*Epeorus* sp.	nss	nss	1	nss	68	nss	8	13	nss	29	0	5	43	0
Heptageniidae	*Heptagenia elegantula*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Heptageniidae	*Heptagenia solitaria*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Heptageniidae	*Heptagenia* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Heptageniidae	*Rhithrogena hageni*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Heptageniidae	*Rhithrogena* sp.	nss	nss	0	nss	9	nss	41	96	nss	891	477	937	67	145

Notes: nss = no spring sample; [a] = spring sample collected in Early April 2011, *= all baetid taxa included in this row.

Table A1 (continued). Estimated densities (numbers/m^2) of mayfly larvae at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the spring (April-May) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	Invertebrate Classification (Taxon - Genus/species/subspecies)	ORDER – Ephemeroptera – Mayflies													
		WLO CRK	FRR SOL	FRR KBP	COL NEW WG11	COL WG12	COL Hwy40 BR	COL WG13	COL WG21	COL SKY LARK[a]	COL WG22	COL WG31	COL NEW WG32	COL PUMP	GUNN
Leptohyphidae	*Tricorythodes explicatus*	nss	nss	0	nss	0	nss	0	1	nss	4	4	31	0	0
Leptophlebiidae	*Choroterpes albiannulata*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Leptophlebiidae	*Leptophlebia* sp.	nss	nss	0	nss	0	nss	0	1	nss	0	0	0	0	0
Leptophlebiidae	*Paraleptophlebia bicornuta*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Leptophlebiidae	*Paraleptophlebia debilis*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Leptophlebiidae	*Paraleptophlebia heteronea*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Leptophlebiidae	*Paraleptophlebia memorialis*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Leptophlebiidae	*Paraleptophlebia packii*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Leptophlebiidae	*Paraleptophlebia* sp.	nss	nss	57	nss	133	nss	96	276	nss	109	32	167	17	137
Siphlonuridae	*Siphlonurus occidentalis*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0

Notes: nss = no spring sample; [a] = spring sample collected in Early April 2011

Table A2. Estimated densities (numbers/m^2) of stonefly larvae at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the spring (April-May) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	Invertebrate Classification (Taxon - Genus/species/subspecies)	ORDER – Plecoptera – Stoneflies													
		WLO CRK	FRR SOL	FRR KBP	COL NEW WG11	COL WG12	COL Hwy40 BR	COL WG13	COL WG21	COL SKY LARK[a]	COL WG22	COL WG31	COL NEW WG32	COL PUMP	GUNN
Capniidae	*Capnia* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Capniidae	*Paracapnia angulata*	nss	nss	4	nss	0	nss	0	0	nss	0	0	0	0	0
Chloroperlidae	*Alloperla* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Chloroperlidae	Chloroperlidae	nss	nss	39	nss	0	nss	0	197	nss	148	0	123	0	0
Chloroperlidae	*Haploperla brevis*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Chloroperlidae	*Suwallia* sp.	nss	nss	37	nss	0	nss	0	0	nss	81	5	1,757	0	0
Chloroperlidae	*Sweltsa* sp.	nss	nss	120	nss	15	nss	32	33	nss	255	19	5	9	15
Chloroperlidae	*Triznaka* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	49	0	0	0
Nemouridae	*Prostoia besametsa*	nss	nss	0	nss	0	nss	1	0	nss	0	0	0	0	0
Nemouridae	*Zapada cinctipes*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Perlidae	*Claassenia sabulosa*	nss	nss	5	nss	93	nss	1	4	nss	225	23	29	0	7
Perlidae	*Hesperoperla pacifica*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	279
Perlidae	Perlidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Perlodidae	*Cultus aestivalis*	nss	nss	69	nss	0	nss	8	29	nss	393	81	555	0	0
Perlodidae	*Diura knowltoni*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Perlodidae	*Isogenoides* sp.	nss	nss	51	nss	21	nss	41	33	nss	13	15	23	17	92
Perlodidae	*Isoperla pinta*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Perlodidae	*Isoperla quinquepunctata*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Perlodidae	*Isoperla* sp.	nss	nss	3	nss	87	nss	272	40	nss	423	153	1,095	227	1,525
Perlodidae	Perlodidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Perlodidae	*Skwala americana*	nss	nss	25	nss	1	nss	44	11	nss	13	11	25	60	168
Pteronarcyidae	*Pteronarcella badia*	nss	nss	0	nss	0	nss	1	0	nss	1	0	4	3	0
Pteronarcyidae	*Pteronarcys californica*	nss	nss	7	nss	0	nss	4	4	nss	44	0	5	363	449
Taeniopterygidae	*Taenionema* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Taeniopterygidae	Taeniopterygidae	nss	nss	0	nss	0	nss	1	0	nss	0	0	0	0	0
Plecoptera	Plecoptera	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Notes: nss = no spring sample; [a] = spring sample collected in Early April 2011

Table A3. Estimated densities (numbers/m^2) of caddisfly larvae at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the spring (April-May) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	Invertebrate Classification (Taxon - Genus/species/subspecies)	ORDER – Trichoptera – Caddisflies													
		WLO CRK	FRR SOL	FRR KBP	COL NEW WG11	COL WG12	COL Hwy40 BR	COL WG13	COL WG21	COL SKY LARK[a]	COL WG22	COL WG31	COL NEW WG32	COL PUMP	GUNN
Brachycentridae	*Amiocentrus* sp.	nss	nss	0	nss	1	nss	0	0	nss	0	0	0	0	0
Brachycentridae	*Brachycentrus americanus*	nss	nss	0	nss	783	nss	49	0	nss	0	65	0	175	69
Brachycentridae	*Brachycentrus occidentalis*	nss	nss	0	nss	0	nss	0	0	nss	0	3	0	0	0
Brachycentridae	*Brachycentrus* sp.	nss	nss	49	nss	0	nss	0	263	nss	379	0	155	0	0
Brachycentridae	*Micrasema* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Glossosomatidae	*Agapetus* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Glossosomatidae	*Glossosoma* sp.	nss	nss	0	nss	0	nss	0	208	nss	896	220	155	1	415
Glossosomatidae	Glossosomatidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Glossosomatidae	*Protoptila* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Helicopsychidae	*Helicopsyche* sp.	nss	nss	0	nss	28	nss	209	0	nss	3	13	0	0	0
Hydropsychidae	*Arctopsyche* sp.	nss	nss	185	nss	43	nss	0	19	nss	5	0	9	0	116
Hydropsychidae	*Cheumatopsyche* sp.	nss	nss	85	nss	4	nss	1	44	nss	48	1	65	3	1
Hydropsychidae	*Hydropsyche* sp.	nss	nss	187	nss	263	nss	21	232	nss	364	91	335	395	1,629
Hydropsychidae	Hydropsychidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Hydroptilidae	*Agraylea* sp.	nss	nss	0	nss	3	nss	0	0	nss	0	0	0	0	0
Hydroptilidae	*Allotrichia* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Hydroptilidae	*Hydroptila* sp.	nss	nss	0	nss	1	nss	0	0	nss	0	0	0	0	4
Hydroptilidae	*Hydroptilidae* sp.	nss	nss	0	nss	3	nss	0	0	nss	0	0	0	0	0
Hydroptilidae	*Leucotrichia* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Hydroptilidae	*Ochrotrichia* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Hydroptilidae	*Stactobiella* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Lepidostomatidae	*Lepidostoma* sp.	nss	nss	95	nss	436	nss	132	532	nss	2,129	151	351	31	0
Leptoceridae	*Ceraclea* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Leptoceridae	*Oecetis* sp.	nss	nss	3	nss	5	nss	104	19	nss	1	28	1	0	0
Limnephilidae	*Clistoronia* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	1	0	0	0
Limnephilidae	*Psychoglypha* sp.	nss	nss	0	nss	1	nss	0	0	nss	0	0	0	0	0
Philopotamidae	*Wormaldia* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Polycentropodidae	*Polycentropus* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	3	0	0	0
Psychomyidae	*Psychomyia flavida*	nss	nss	0	nss	0	nss	0	0	nss	0	7	0	0	0
Rhyacophilidae	*Rhyacophila* sp.	nss	nss	4	nss	4	nss	3	7	nss	57	1	0	19	240
Unoidae	*Oligophlebodes* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0

Notes: nss = no spring sample; **[a] =** spring sample collected in Early April 2011

Table A4. Estimated densities (numbers/m^2) of true fly larvae at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the spring (April-May) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	**Invertebrate Classification (Taxon - Genus/species/subspecies)**	**ORDER – Diptera – True flies**													
		WLO CRK	**FRR SOL**	**FRR KBP**	**COL NEW WG11**	**COL WG12**	**COL Hwy40 BR**	**COL WG13**	**COL WG21**	**COL SKY LARK[a]**	**COL WG22**	**COL WG31**	**COL NEW WG32**	**COL PUMP**	**GUNN**
Athericidae	*Atherix pachypus*	nss	nss	1	nss	0	nss	1	0	nss	0	0	0	73	172
Ceratopogonidae	Ceratopogonidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Ceratopogonidae	*Palpomyia* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Chironomidae	Chironomidae	nss	nss	1,341	nss	3,324	nss	3,255	2,171	nss	1,228	1,535	537	2,040	18,517
Culicidae	*Aedes* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Empididae	Empididae sp.	nss	nss	5	nss	32	nss	1	5	nss	19	8	8	8	91
Empididae	*Wiedemannia* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Heleidae	Heleidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Muscidae	*Limnophora* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Psychodidae	*Chelifera* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Psychodidae	*Hemerodromia* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Psychodidae	*Pericoma* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Psychodidae	Psychodidae	nss	nss	1	nss	7	nss	0	9	nss	33	0	0	0	0
Simuliidae	Simuliidae	nss	nss	3,212	nss	2,647	nss	143	51	nss	112	5	76	80	3
Tabanidae	*Chrysops* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Tipulidae	*Antocha* sp.	nss	nss	0	nss	17	nss	0	0	nss	0	0	0	3	263
Tipulidae	*Dicranota* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Tipulidae	*Hexatoma* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Tipulidae	*Tipula* sp.	nss	nss	0	nss	12	nss	0	0	nss	0	0	0	1	0
Tipulidae	Tipulidae	nss	nss	0	nss	0	nss	0	0	nss	0	5	0	0	123
Blephariceridae	*Agathon* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Dipterasp.	Diptera	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Tanyderidae	*Protanyderus* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0

Notes: nss = no spring sample; [a] = spring sample collected in Early April 2011

Table A5. Estimated densities (numbers/m^2) of aquatic beetle larvae at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the spring (April-May) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	Invertebrate Classification (Taxon - Genus/species/subspecies)	ORDER – Coleoptera – Aquatic Beetles													
		WLO CRK	FRR SOL	FRR KBP	COL NEW WG11	COL WG12	COL Hwy40 BR	COL WG13	COL WG21	COL SKY LARK[a]	COL WG22	COL WG31	COL NEW WG32	COL PUMP	GUNN
Carabidae	Carabidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Dytiscidae	Dytiscidae	nss	nss	0	nss	3	nss	3	0	nss	0	0	0	9	0
Dytiscidae	*Hydroporus* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Dytiscidae	*Liodessus* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	55	0	0	0
Elmidae	*Cleptelmis addenda*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Elmidae	Elmidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Elmidae	*Narpus concolor*	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Elmidae	*Optioservus* sp.	nss	nss	217	nss	415	nss	11	140	nss	548	377	1,412	636	616
Elmidae	*Zaitzevia parvula*	nss	nss	84	nss	0	nss	0	41	nss	459	0	95	0	0
Hydrophilidae	*Helophorus* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Hydrophilidae	Hydrophilidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Staphylinidae	Staphylinidae	nss	nss	0	nss	0	nss	0	0	nss	0	7	0	0	0

Notes: nss = no spring sample; [a] = spring sample collected in Early April 2011

Table A6. Estimated densities (numbers/m^2) non-insect aquatic invertebrates at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the spring (April-May) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	**Invertebrate Classification (Taxon - Genus/species/subspecies)**	**Non-Insect Aquatic Invertebrate Taxa**													
		WLO CRK	**FRR SOL**	**FRR KBP**	**COL NEW WG11**	**COL WG12**	**COL Hwy40 BR**	**COL WG13**	**COL WG21**	**COL SKY LARK[a]**	**COL WG22**	**COL WG31**	**COL NEW WG32**	**COL PUMP**	**GUNN**
Amphipoda	Amphipoda	nss	nss	0	nss	0	nss	0	3	nss	1	0	0	0	0
Gammaraidae	*Gammarus lacustris*	nss	nss	0	nss	0	nss	0	0	nss	0	3	0	0	0
Bivlavia	Bivlavia	nss	nss	0	nss	35	nss	24	0	nss	0	0	0	0	0
Bosminidae	Bosmina	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Eubranchipoda	Eubranchipoda	nss	nss	0	nss	8	nss	0	0	nss	0	0	0	0	0
Ancylidae	*Ferrissia* sp.	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	1	0
Corbiculidae	Corbiculidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Gastropoda	Gastropoda	nss	nss	0	nss	0	nss	0	11	nss	20	0	0	0	0
Lymnaeidae	Lymnaeidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Physidae	Physidae	nss	nss	0	nss	0	nss	0	0	nss	0	3	0	1	8
Planorbidae	Planorbidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Sphaeridae	Sphaeridae	nss	nss	0	nss	0	nss	0	0	nss	0	1	0	0	0
Hirudinea	Hirudinea	nss	nss	0	nss	52	nss	0	0	nss	0	17	0	0	0
Hydracarina	Hydracarina	nss	nss	0	nss	8	nss	0	0	nss	0	0	0	36	0
Asellidae	*Caecidota* sp.	nss	nss	0	nss	425	nss	8	0	nss	0	0	0	0	0
Hyalellidae	*Hyalella azteca*	nss	nss	0	nss	292	nss	0	0	nss	0	0	0	0	0
Asellidae	*Lirceus* sp.	nss	nss	4	nss	0	nss	0	1	nss	1	0	0	0	0
Nematoda	Nematoda	nss	nss	0	nss	5	nss	17	0	nss	0	1	0	8	0
Oligochaeta	Oligochaeta	nss	nss	52	nss	1,313	nss	32	41	nss	157	1,411	33	7	409
Turbellaria	Turbellaria	nss	nss	0	nss	0	nss	0	104	nss	1	0	0	0	0
Collembola	Collembola	nss	nss	0	nss	0	nss	0	0	nss	0	0	0	0	0
Gomphidae	Gomphidae	nss	nss	0	nss	0	nss	0	0	nss	0	0	1	0	0

Notes: nss = no spring sample; [a] = spring sample collected in Early April 2011 * = uncommonly found insect groups.

Table A7. Estimated densities (numbers/m^2) of mayfly larvae at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the fall (August-September) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	**Invertebrate Classification (Taxon - Genus/species/subspecies)**	**ORDER – Ephemeroptera – Mayflies**													
		WLO CRK	**FRR SOL**	**FRR KBP**	**COL NEW WG11**	**COL WG12**	**COL Hwy40 BR**	**COL WG13**	**COL WG21**	**COL SKY LARK**	**COL WG22**	**COL WG31**	**COL NEW WG32**	**COL PUMP**	**GUNN**
Ameletidae	*Ameletus* sp.	39	4	1	0	1	0	0	1	0	0	0	0	0	nss
Baetidae	*Acentrella insignificans*	105	4	5	8	0	29	0	72	0	163	73	177	7	nss
Baetidae	Baetidae	20	516	131	0	195	621	0	20	0	57	20	45	60	nss
Baetidae	*Baetis bicaudatus*	0	51	15	0	0	3	0	20	0	5	5	5	0	nss
Baetidae	*Baetis flavistriga*	0	0	0	1	0	11	0	0	0	0	0	0	0	nss
Baetidae	*Baetis* sp.	53	3993	3557	3943	4149	3475	2265	1017	152	701	341	1109	660	nss
Baetidae	*Baetis tricaudatus*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Baetidae	*Diphetor hageni*	0	24	25	9	371	27	12	292	1	45	23	185	1	nss
Baetidae	*Fallceon quilleri*	0	0	0	0	0	3	0	0	0	0	0	0	0	nss
Baetidae	*Plauditis* sp.	0	0	0	0	8	5	0	3	0	0	0	13	0	nss
Caenidae	*Brachycercus* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Caenidae	*Caenis* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Ephemerellidae	*Attenella margarita*	4	0	0	5	1	3	3	19	0	5	0	93	3	nss
Ephemerellidae	*Drunella doddsii*	173	0	0	0	0	0	0	0	0	0	0	0	0	nss
Ephemerellidae	*Drunella grandis*	425	1	0	0	0	3	0	65	23	79	12	56	0	nss
Ephemerellidae	*Drunella* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Ephemerellidae	*Ephemerella dorothea infrequens*	812	16	11	23	71	1104	56	453	1233	125	95	784	17	nss
Ephemerellidae	*Ephemerella excrucians*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Ephemerellidae	*Ephemerella* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Ephemerellidae	Ephemerellidae	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Ephemerellidae	*Serratella micheneri*	0	0	1	5	31	11	4	24	0	11	1	35	3	nss
Ephemerellidae	*Serratella* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Ephemerellidae	*Timpanoga hecuba hecuba*	5	0	0	0	0	0	0	0	0	0	0	0	0	nss

Notes: nss = no fall sample.

Table A7 (continued). Estimated densities (numbers/m^2) of mayfly larvae at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the fall (August-September) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	**Invertebrate Classification (Taxon - Genus/species/subspecies)**	**ORDER – Ephemeroptera – Mayflies**													
		WLO CRK	**FRR SOL**	**FRR KBP**	**COL NEW WG11**	**COL WG12**	**COL Hwy40 BR**	**COL WG13**	**COL WG21**	**COL SKY LARK**	**COL WG22**	**COL WG31**	**COL NEW WG32**	**COL PUMP**	**GUNN**
Heptageniidae	*Cinygmula* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Heptageniidae	*Ecdyonurus simplicioides*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Heptageniidae	*Epeorus albertae*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Heptageniidae	*Epeorus* sp.	0	0	1	0	0	91	0	68	401	5	0	0	33	nss
Heptageniidae	*Heptagenia elegantula*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Heptageniidae	*Heptagenia solitaria*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Heptageniidae	*Heptagenia* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Heptageniidae	*Rhithrogena hageni*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Heptageniidae	*Rhithrogena* sp.	1588	0	0	1	9	48	31	219	488	695	388	1559	3	nss
Leptohyphidae	*Tricorythodes explicates*	0	15	47	196	132	56	1	97	4	27	31	415	240	nss
Leptophlebiidae	*Choroterpes albiannulata*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Leptophlebiidae	*Leptophlebia* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Leptophlebiidae	*Paraleptophlebia bicornuta*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Leptophlebiidae	*Paraleptophlebia debilis*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Leptophlebiidae	*Paraleptophlebia heteronea*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Leptophlebiidae	*Paraleptophlebia memorialis*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Leptophlebiidae	*Paraleptophlebia packii*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Leptophlebiidae	*Paraleptophlebia* sp.	155	72	109	40	117	259	56	363	563	159	15	755	5	nss
Siphlonuridae	*Siphlonurus occidentalis*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss

Notes: nss = no fall sample.

Table A8. Estimated densities (numbers/m^2) of stonefly larvae at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the fall (August-September) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	**Invertebrate Classification (Taxon - Genus/species/subspecies)**	**ORDER – Plecoptera – Stoneflies**													
		WLO CRK	**FRR SOL**	**FRR KBP**	**COL NEW WG11**	**COL WG12**	**COL Hwy40 BR**	**COL WG13**	**COL WG21**	**COL SKY LARK**	**COL WG22**	**COL WG31**	**COL NEW WG32**	**COL PUMP**	**GUNN**
Capniidae	*Capnia* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Capniidae	*Paracapnia angulata*	0	0	0	1	0	0	0	0	28	0	0	0	0	nss
Chloroperlidae	*Alloperla* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Chloroperlidae	Chloroperlidae	88	24	44	4	1	37	3	4	49	11	25	33	0	nss
Chloroperlidae	*Haploperla brevis*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Chloroperlidae	*Suwallia* sp.	0	0	5	0	0	0	0	0	0	0	0	0	0	nss
Chloroperlidae	*Sweltsa* sp.	372	24	11	0	0	0	0	0	0	0	0	0	0	nss
Chloroperlidae	*Triznaka* sp.	0	0	0	1	0	0	0	0	0	0	0	0	0	nss
Nemouridae	*Prostoia besametsa*	0	0	0	0	0	0	0	0	3	0	0	0	0	nss
Nemouridae	*Zapada cinctipes*	3	0	0	0	0	0	0	1	0	0	0	0	0	nss
Perlidae	*Claassenia sabulosa*	36	79	107	131	81	461	155	11	59	195	324	212	0	nss
Perlidae	*Hesperoperla pacifica*	4	0	0	0	0	0	0	0	0	0	0	0	0	nss
Perlidae	Perlidae	0	0	7	0	0	8	0	0	0	0	0	0	0	nss
Perlodidae	*Cultus aestivalis*	1	5	33	1	1	40	7	3	27	52	21	168	11	nss
Perlodidae	*Diura knowltoni*	4	0	0	0	0	0	0	0	0	0	0	0	0	nss
Perlodidae	*Isogenoides* sp.	0	0	0	0	0	3	0	0	0	0	1	48	0	nss
Perlodidae	*Isoperla pinta*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Perlodidae	*Isoperla quinquepunctata*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Perlodidae	*Isoperla* sp.	16	0	1	1	0	16	1	76	112	24	11	35	0	nss
Perlodidae	Perlodidae	45	13	11	4	7	69	15	147	0	48	80	45	1	nss
Perlodidae	*Skwala Americana*	44	57	31	23	4	16	0	7	19	7	21	80	0	nss
Pteronarcyidae	*Pteronarcella badia*	117	0	0	0	0	0	0	1	0	3	0	29	0	nss
Pteronarcyidae	*Pteronarcys californica*	1	55	3	0	0	3	239	12	32	116	5	21	253	nss
Taeniopterygidae	*Taenionema* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Taeniopterygidae	Taeniopterygidae	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Plecoptera	Plecoptera	3	4	5	0	3	21	1	1	0	11	3	11	0	nss

Notes: nss = no fall sample.

Table A9. Estimated densities (numbers/m^2) of caddisfly larvae at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the fall (August-September) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	Invertebrate Classification (Taxon - Genus/species/subspecies)	ORDER – Trichoptera – Caddisflies													
		WLO CRK	FRR SOL	FRR KBP	COL NEW WG11	COL WG12	COL Hwy40 BR	COL WG13	COL WG21	COL SKY LARK	COL WG22	COL WG31	COL NEW WG32	COL PUMP	GUNN
Brachycentridae	*Amiocentrus* sp.	1	0	0	0	0	0	0	0	0	0	0	0	0	nss
Brachycentridae	*Brachycentrus americanus*	1111	552	1048	0	79	304	109	697	361	237	89	277	3477	nss
Brachycentridae	*Brachycentrus occidentalis*	356	101	31	0	3	3	0	1	0	0	0	0	0	nss
Brachycentridae	*Brachycentrus* sp.	0	0	0	172	0	0	0	0	0	0	0	0	0	nss
Brachycentridae	*Micrasema* sp.	24	0	0	0	0	0	0	0	0	0	0	0	0	nss
Glossosomatidae	*Agapetus* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Glossosomatidae	*Glossosoma* sp.	163	1636	303	19	48	1456	2593	313	1271	3173	3597	456	5	nss
Glossosomatidae	Glossosomatidae	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Glossosomatidae	*Protoptila* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Helicopsychidae	*Helicopsyche* sp.	0	0	0	0	1	59	11	0	65	4	3	0	0	nss
Hydropsychidae	*Arctopsyche* sp.	289	803	164	4	143	24	3	96	15	36	11	7	0	nss
Hydropsychidae	*Cheumatopsyche* sp.	0	1012	269	17	1	760	116	12	9	179	29	584	15	nss
Hydropsychidae	*Hydropsyche* sp.	28	2003	681	2311	1205	6120	1401	1003	521	1232	487	1203	412	nss
Hydropsychidae	Hydropsychidae	15	47	31	0	0	107	3	0	0	12	1	43	95	nss
Hydroptilidae	*Agraylea* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Hydroptilidae	*Allotrichia* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Hydroptilidae	*Hydroptila* sp.	0	0	0	0	3	3	0	3	0	0	0	0	149	nss
Hydroptilidae	*Hydroptilidae* sp.	0	3	4	0	23	3	0	0	0	3	0	13	0	nss
Hydroptilidae	*Leucotrichia* sp.	0	0	0	0	0	5	9	0	0	0	0	0	0	nss
Hydroptilidae	*Ochrotrichia* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Hydroptilidae	*Stactobiella* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Lepidostomatidae	*Lepidostoma* sp.	1504	475	195	163	227	765	279	2591	2811	1545	1264	3331	121	nss
Leptoceridae	*Ceraclea* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Leptoceridae	*Oecetis* sp.	0	0	11	15	59	99	4	100	19	5	15	53	4	nss
Limnephilidae	*Clistoronia* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Limnephilidae	*Psychoglypha* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Philopotamidae	*Wormaldia* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Polycentropodidae	*Polycentropus* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Psychomyidae	*Psychomyia flavida*	0	0	3	4	0	29	0	0	55	179	81	8	9	nss
Rhyacophilidae	*Rhyacophila* sp.	20	0	13	1	4	29	12	15	21	0	0	0	8	nss
Unoidae	*Oligophlebodes* sp.	919	0	0	0	0	0	0	0	0	0	0	0	0	nss

Notes: nss = no fall sample.

Table A10. Estimated densities (numbers/m^2) of true fly larvae at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the fall (August-September) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	Invertebrate Classification (Taxon - Genus/species/subspecies)	ORDER – Diptera – True flies													
		WLO CRK	FRR SOL	FRR KBP	COL NEW WG11	COL WG12	COL Hwy40 BR	COL WG13	COL WG21	COL SKY LARK	COL WG22	COL WG31	COL NEW WG32	COL PUMP	GUNN
Athericidae	*Atherix pachypus*	12	187	3	31	19	3	471	16	17	20	4	9	123	nss
Ceratopogonidae	Ceratopogonidae	17	0	1	0	0	3	0	0	0	0	1	0	4	nss
Ceratopogonidae	*Palpomyia* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Chironomidae	Chironomidae	1204	1795	1067	1757	2027	1837	431	1817	2148	1536	2049	3044	12293	nss
Culicidae	*Aedes* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Empididae	Empididae sp.	0	16	23	13	19	83	1	4	32	21	61	16	61	nss
Empididae	*Wiedemannia* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Heleidae	Heleidae	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Muscidae	*Limnophora* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Psychodidae	*Chelifera* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Psychodidae	*Hemerodromia* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Psychodidae	*Pericoma* sp.	379	0	0	0	0	3	0	0	0	0	0	0	0	nss
Psychodidae	Psychodidae	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Simuliidae	Simuliidae	68	1733	220	1197	689	643	156	244	71	268	68	461	144	nss
Tabanidae	*Chrysops* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Tipulidae	*Antocha* sp.	8	0	5	11	23	72	35	137	107	16	3	0	15	nss
Tipulidae	*Dicranota* sp.	31	0	0	0	0	0	0	0	0	0	0	0	0	nss
Tipulidae	*Hexatoma* sp.	49	16	4	0	0	0	0	0	0	0	3	0	0	nss
Tipulidae	*Tipula* sp.	0	0	0	0	0	3	0	0	0	0	0	0	3	nss
Tipulidae	Tipulidae	0	4	7	0	5	3	0	0	0	0	0	4	0	nss
Blephariceridae	*Agathon* sp.	27	0	0	0	0	0	0	0	0	0	0	0	0	nss
Diptera	Diptera	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Tanyderidae	*Protanyderus* sp.	3	0	0	0	0	0	0	0	0	0	0	0	0	nss

Notes: nss = no fall sample.

Table A11. Estimated densities (numbers/m^2) of aquatic beetle larvae at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the fall (August-September) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	Invertebrate Classification (Taxon - Genus/species/subspecies)	ORDER – Coleoptera – Aquatic Beetles													
		WLO CRK	FRR SOL	FRR KBP	COL NEW WG11	COL WG12	COL Hwy40 BR	COL WG13	COL WG21	COL SKY LARK	COL WG22	COL WG31	COL NEW WG32	COL PUMP	GUNN
Carabidae	Carabidae	0	0	0	0	0	0	0	0	0	0	0	0	1	nss
Dytiscidae	Dytiscidae	0	3	0	1	0	0	0	0	0	0	0	0	1	nss
Dytiscidae	*Hydroporus* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Dytiscidae	*Liodessus* sp.	0	0	0	3	4	3	7	35	17	21	8	1	0	nss
Elmidae	*Cleptelmis addenda*	49	0	0	0	0	0	0	0	0	0	0	0	0	nss
Elmidae	Elmidae	9	387	457	0	13	45	19	0	20	80	131	405	477	nss
Elmidae	*Narpus concolor*	31	0	0	0	0	0	0	0	0	0	0	0	0	nss
Elmidae	*Optioservus* sp.	761	3513	2711	687	709	456	900	505	339	1211	1300	3799	1204	nss
Elmidae	*Zaitzevia parvula*	1940	544	863	155	195	229	455	167	60	551	355	900	27	nss
Hydrophilidae	*Helophorus* sp.	0	0	0	0	0	0	0	0	0	1	0	0	0	nss
Hydrophilidae	Hydrophilidae	0	0	0	1	0	0	0	0	0	0	0	0	0	nss
Staphylinidae	Staphylinidae	0	0	0	0	0	0	0	0	0	0	0	0	0	nss

Notes: nss = no fall sample.

Table A12. Estimated densities (numbers/m^2) of non-insect aquatic invertebrates at various sampling sites on Willow Creek, the Fraser River and Colorado River in Middle Park, Grand County, and the Gunnison River in Ute Park (Black Canyon of the Gunnison River Gorge) on the Montrose-Delta County line, during the fall (August-September) 2010. Three substrate samples (0.25 m^2) were taken at each sample site for a total sample area of 0.75 m^2 for each season. All data reported are standardized to 1 m^2.

Family	Invertebrate Classification (Taxon - Genus/species/subspecies)	Non-Insect Aquatic Invertebrate Taxa													
		WLO CRK	FRR SOL	FRR KBP	COL NEW WG11	COL WG12	COL Hwy40 BR	COL WG13	COL WG21	COL SKY LARK	COL WG22	COL WG31	COL NEW WG32	COL PUMP	GUNN
Amphipoda	Amphipoda	0	0	0	1	0	0	0	0	0	0	0	0	0	nss
Gammaraidae	*Gammarus lacustris*	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Bivlavia	Bivlavia	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Bosminidae	Bosmina	3	0	4	3	0	5	0	0	9	0	0	0	20	nss
Eubranchipoda	Eubranchipoda	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Ancylidae	*Ferrissia* sp.	0	0	0	0	0	0	0	0	0	0	0	12	235	nss
Corbiculidae	Corbiculidae	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Gastropoda	Gastropoda	0	0	1	0	1	0	0	0	0	0	0	0	7	nss
Lymnaeidae	Lymnaeidae	0	0	0	0	1	0	0	0	0	0	0	0	117	nss
Physidae	Physidae	0	0	3	4	4	72	0	89	35	64	9	1	153	nss
Planorbidae	Planorbidae	3	1	3	3	16	13	0	40	1	3	3	4	1	nss
Sphaeridae	Sphaeridae	0	31	11	5	61	64	1	553	229	31	13	4	8	nss
Hirudinea	Hirudinea	1	1	0	11	21	43	0	224	7	0	3	4	0	nss
Hydracarina	Hydracarina	199	111	32	12	75	101	27	115	37	148	47	209	1773	nss
Asellidae	*Caecidota* sp.	0	0	0	16	35	0	0	1	0	0	0	0	0	nss
Hyalellidae	*Hyalella azteca*	0	0	0	0	17	11	4	9	1	3	0	0	0	nss
Asellidae	*Lirceus* sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	nss
Nematoda	Nematoda	75	263	111	41	72	48	23	75	12	33	28	88	193	nss
Oligochaeta	Oligochaeta	0	333	97	336	1337	677	231	189	149	260	67	240	14120	nss
Turbellaria	Turbellaria	0	0	0	3	0	0	0	0	0	0	0	0	0	nss
Gomphidae*	Gomphidae	0	0	0	0	0	0	0	0	0	0	0	5	3	nss
Collembola*	Collembola	0	0	1	0	1	0	0	4	0	5	0	0	0	nss

Notes: nss = no fall sample. * = uncommonly found insect groups.

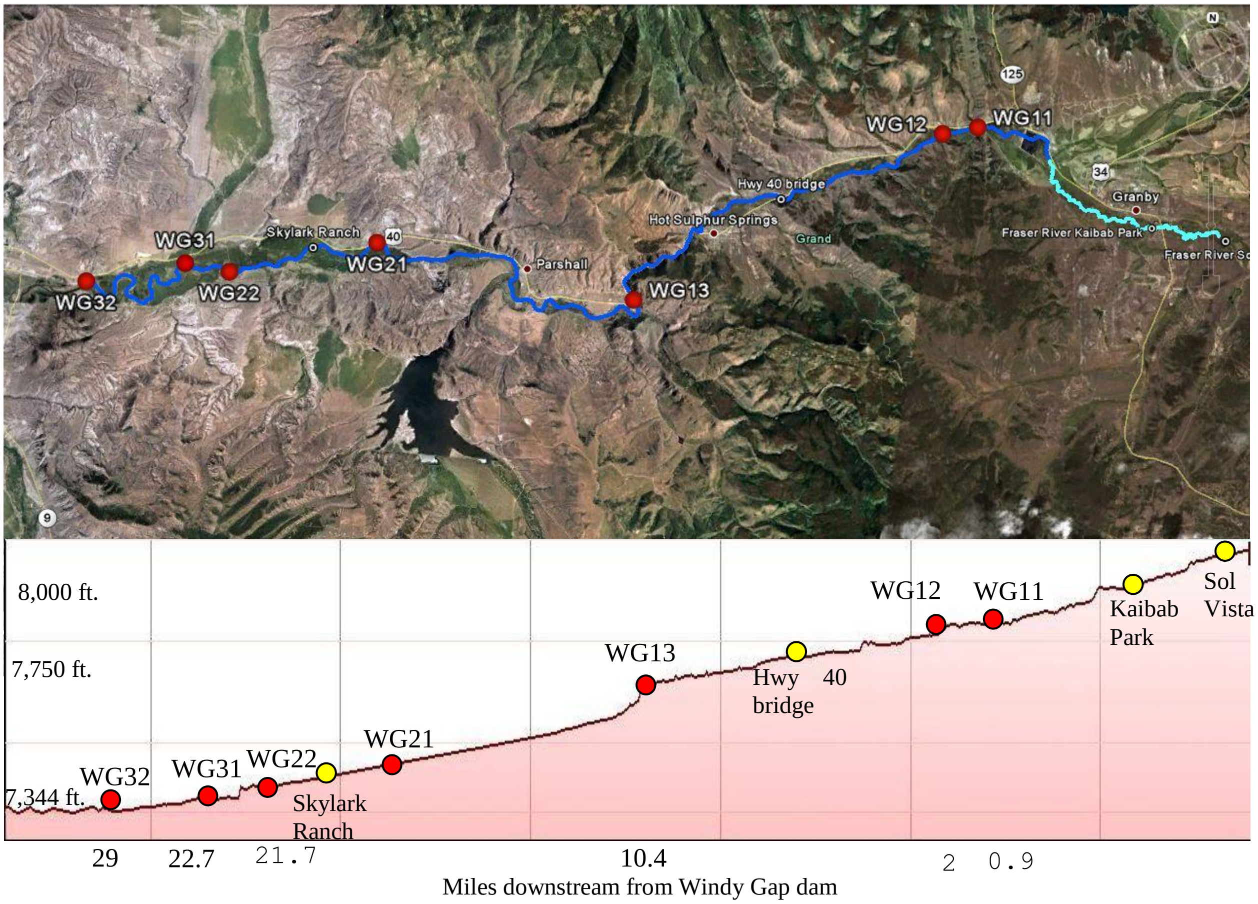


Figure 1: Map of the Windy Gap study area. Red dots are locations of Erickson 1983 original stations, dots are location of additional stations added in 2010. Willow Creek, Pumphouse and Gunnison River stations are not shown. Blue line is the Colorado River, turquoise line is the Fraser River.

Figure 2: 0.25m² benthic sampling net used at all Windy Gap study sites.

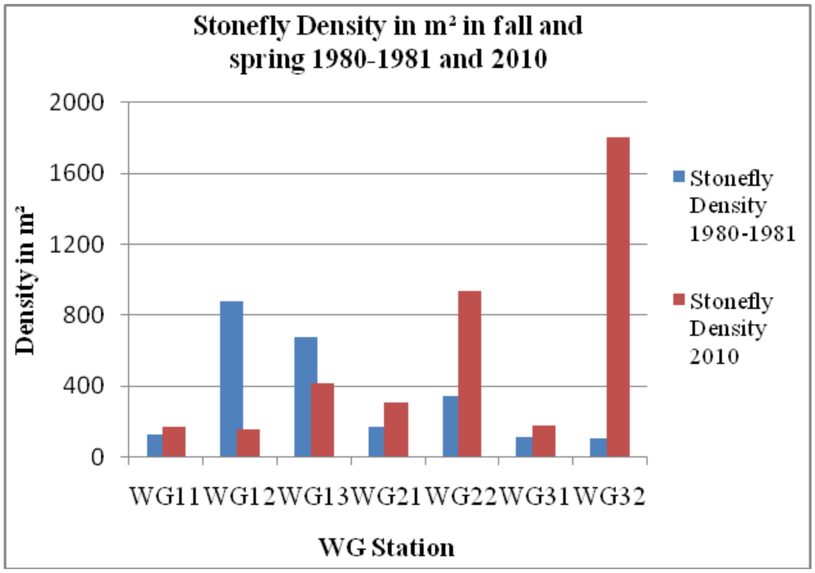


Figure 3: Density of stoneflies (individual / m^2) of the Colorado River at WG sites in spring and fall 2010 and 1980-1981.

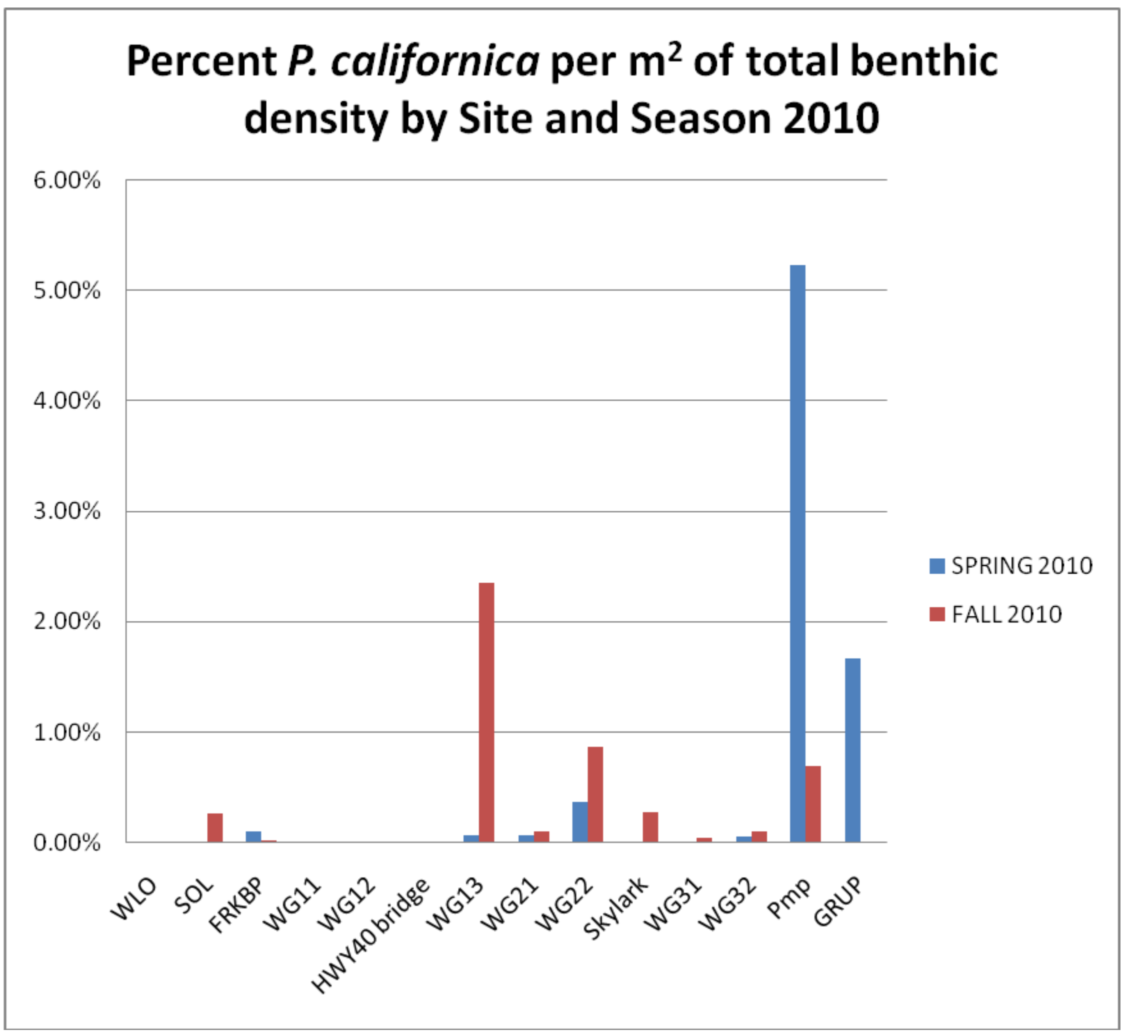


Figure 4: Percent *Pteronarcys californica* in spring and fall 2010. All replicates per season averaged. No spring sample at WLO, SOL, WG11, HWY40bridge and Skylark, no fall sample from GRUP.

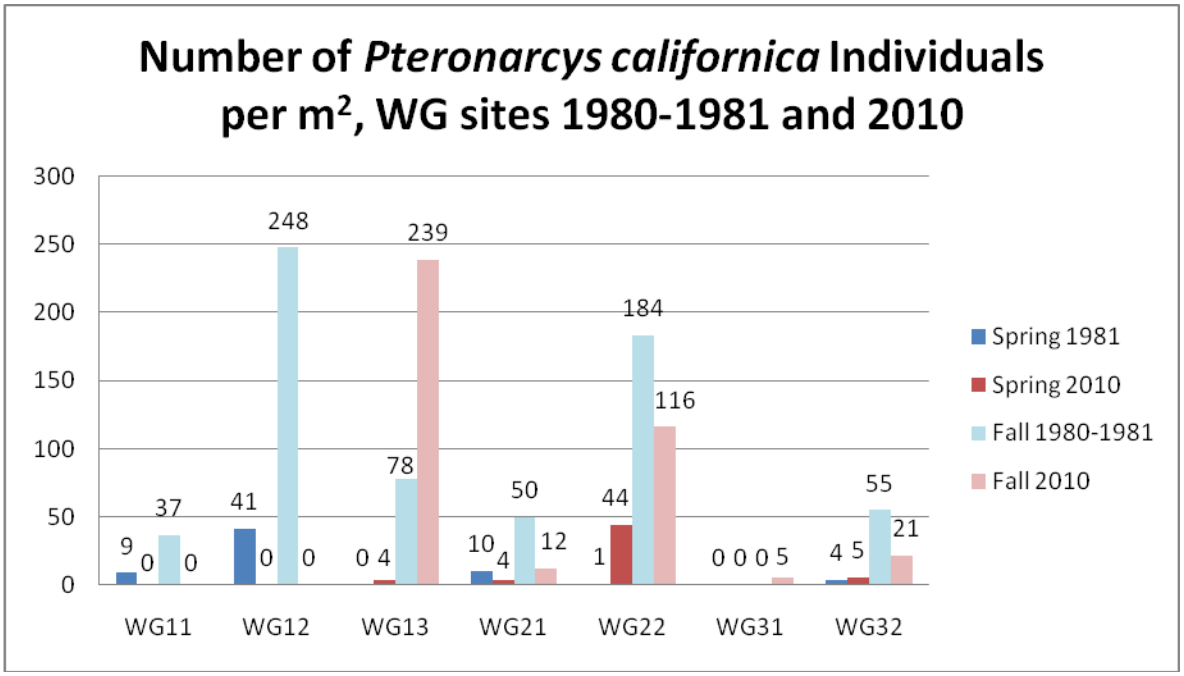


Figure 5: Number of individual *Pteronarcys californica* collected, transformed into m^2 per WG station, 1980-1981 and 2010.

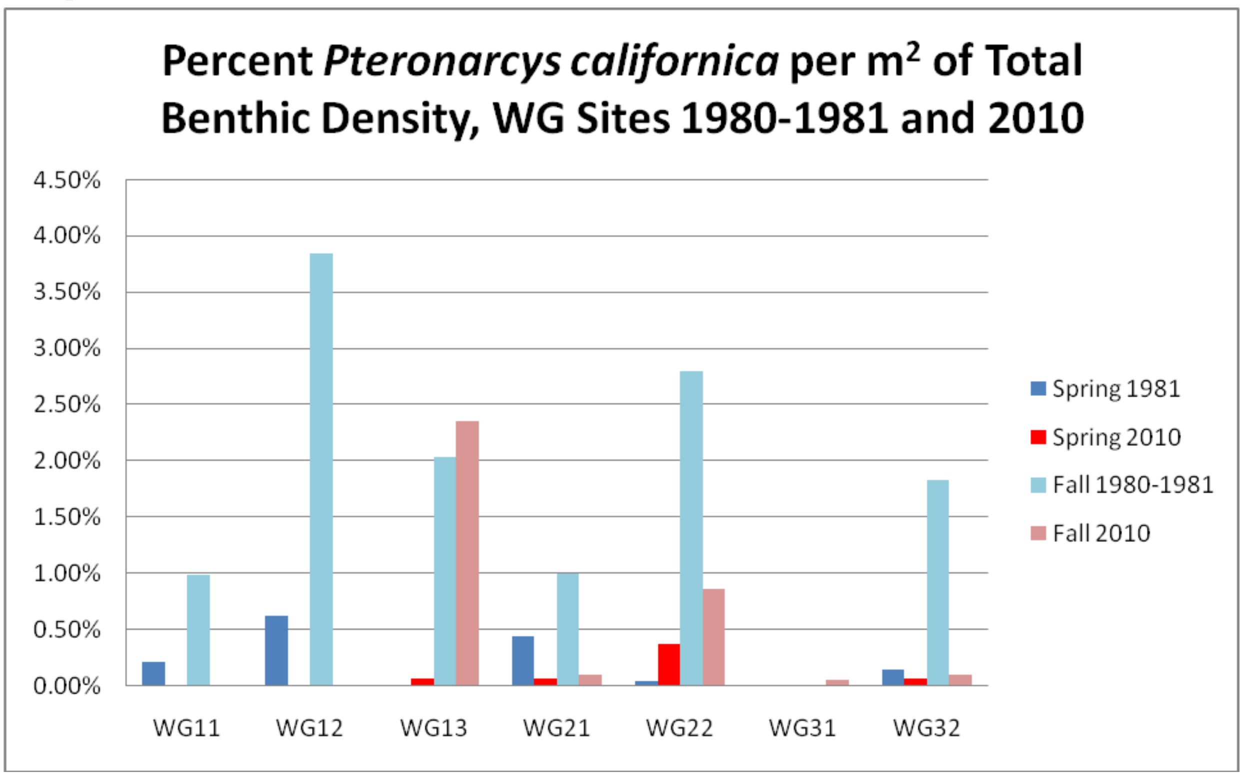


Figure 6: Percent *Pteronarcys californica* by station, season and year. No spring sample at WG11 in 2010.

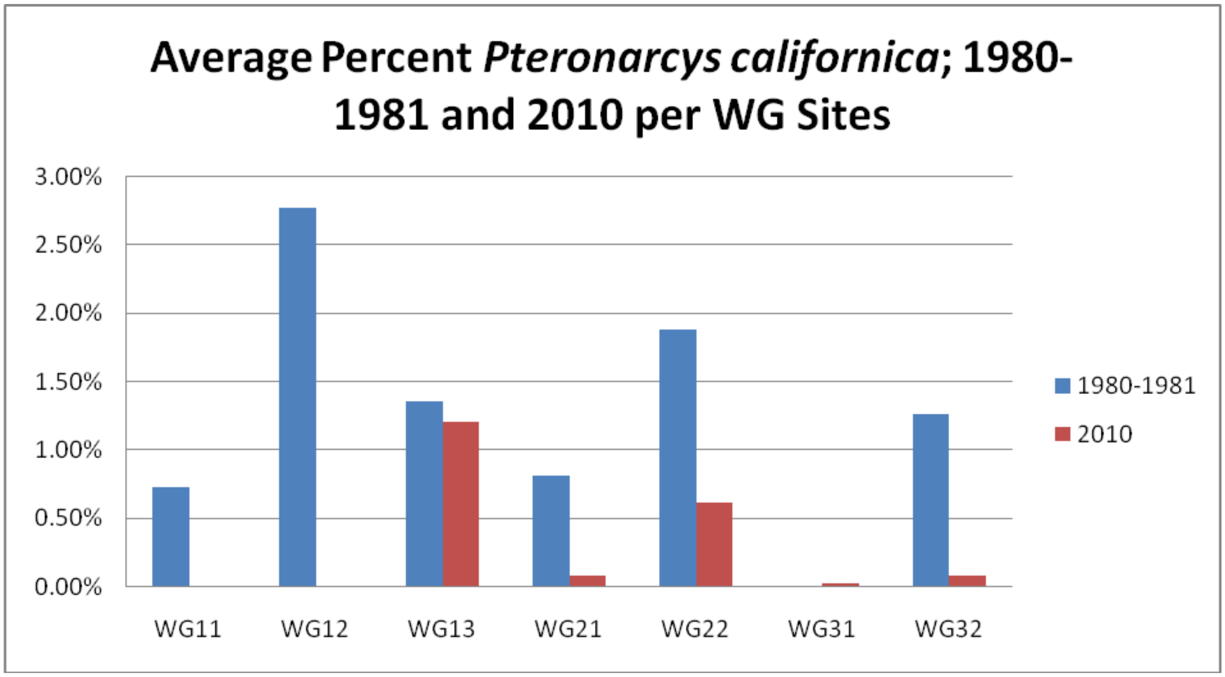


Figure 7: Percent *Pteronarcys californica* for 1980-1981 and 2010 averaged for all sampling periods per WG Station.

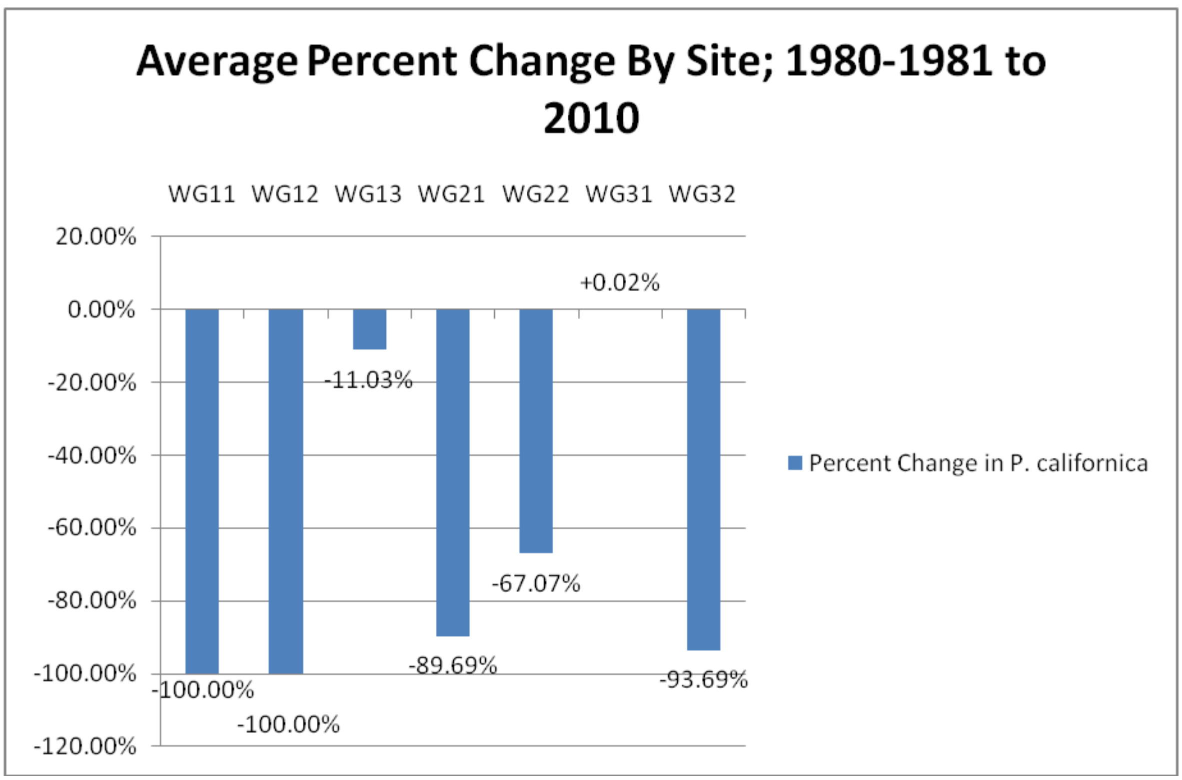
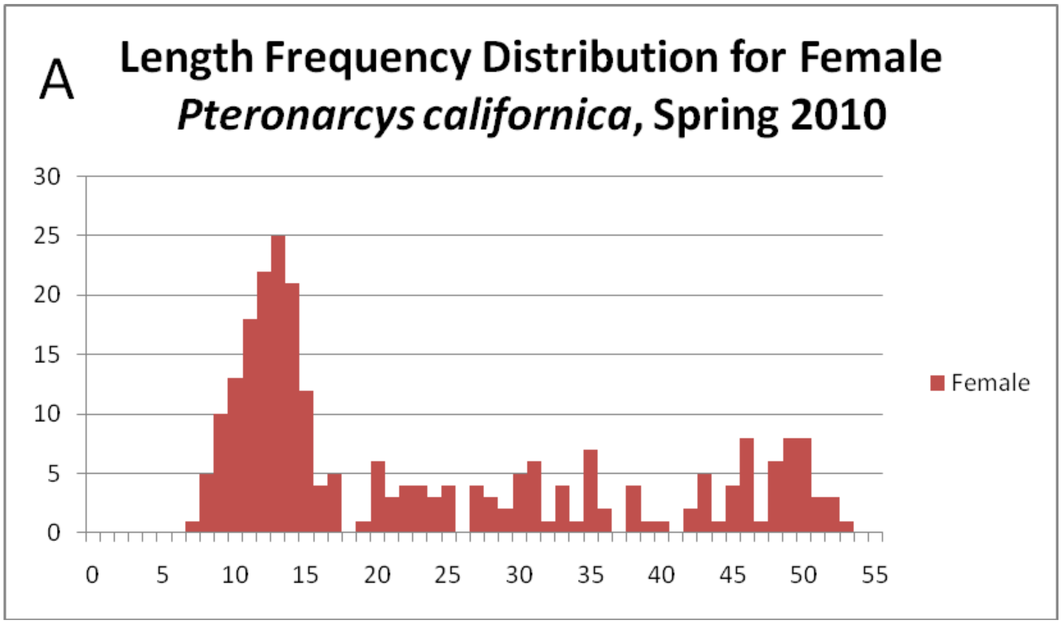


Figure 8: Average percent change of *Pteronarcys californica* by station with all sampling periods combined ((2010 average - 1980-1981 average) / 1980-1981 average).



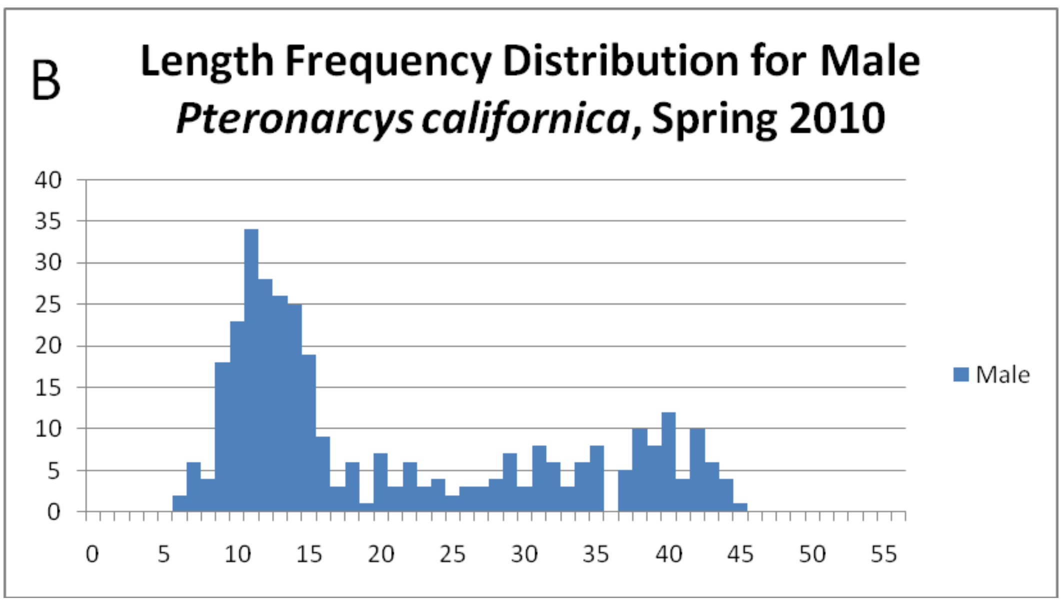


Figure 9: Length frequency distribution for female (A) and male (B) *Pteronarcys californica* from spring 2010.

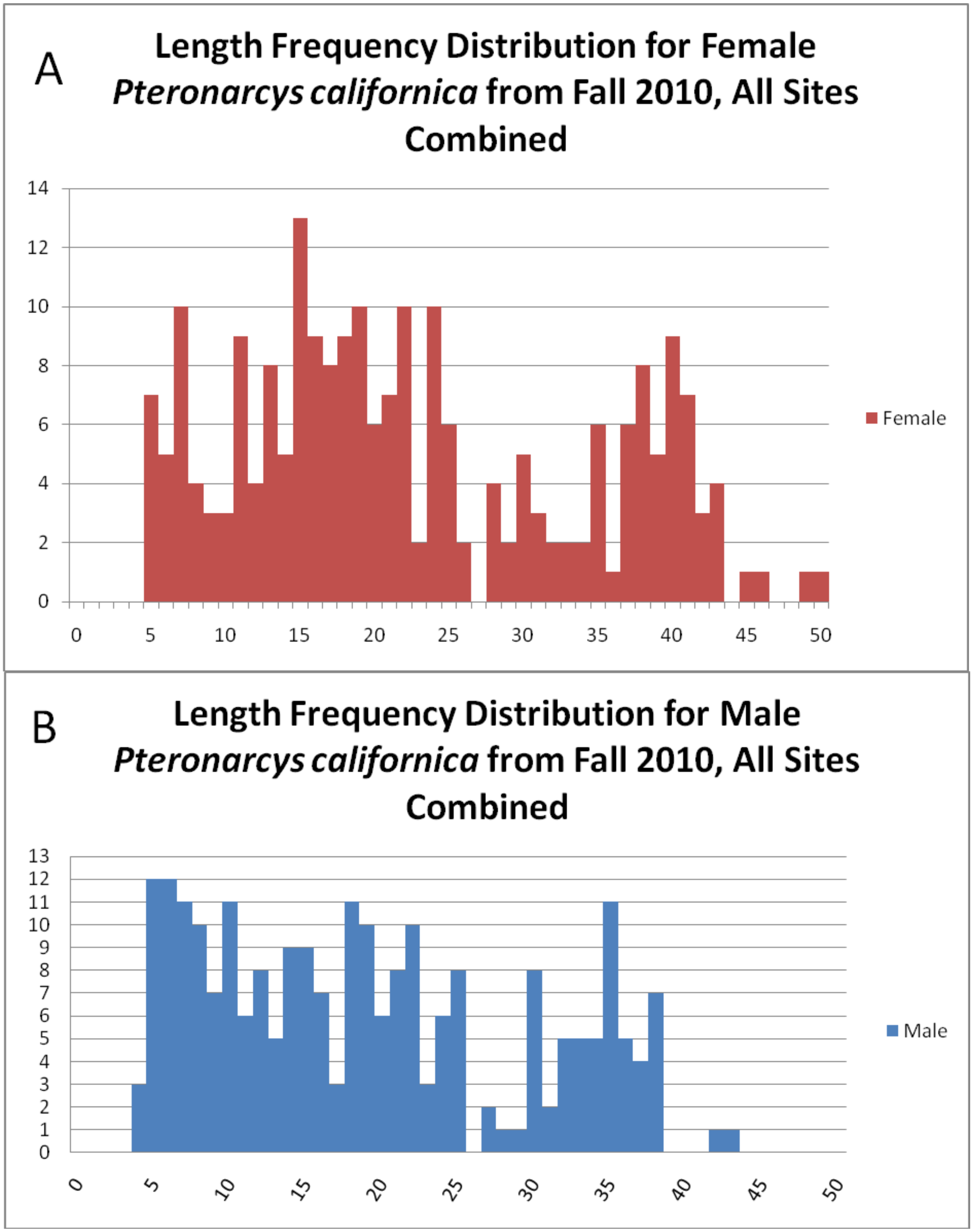


Figure 10: Length frequency distribution for female (A) and male (B) *Pteronarcys californica* from fall 2010.

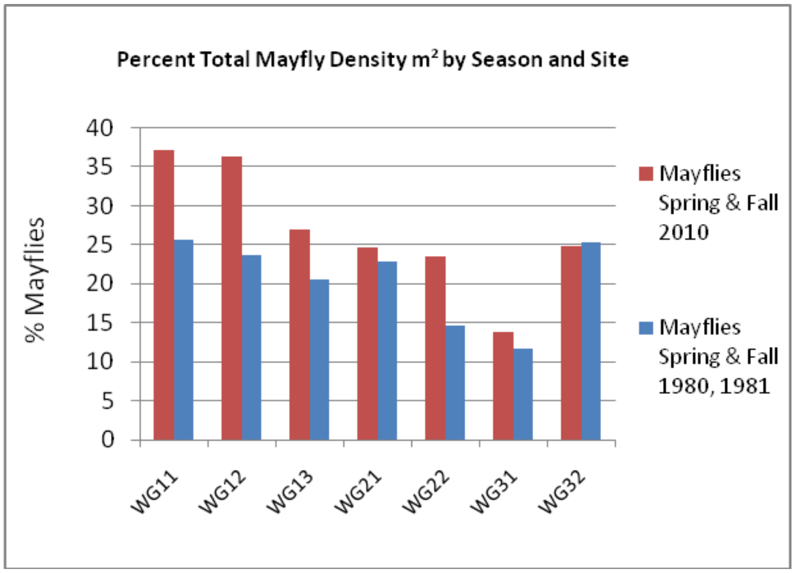


Figure 11. Percent density of mayflies of the Colorado River at WG sites in spring and fall 2010 and 1980-1981.

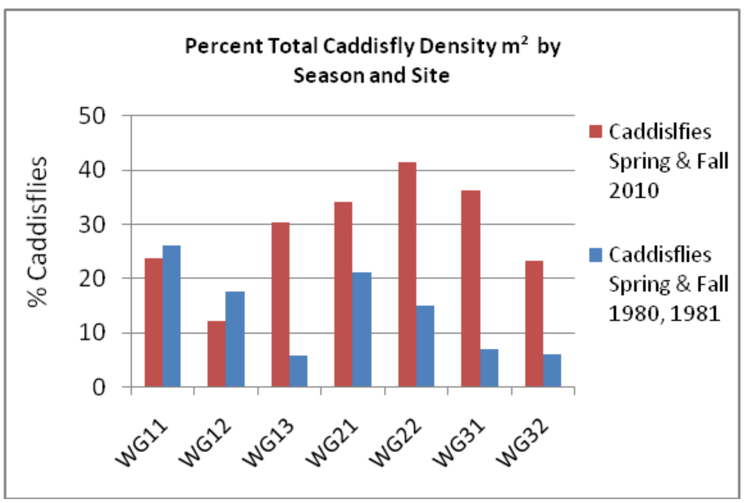


Figure 12: Percent density of caddisflies of the Colorado River at WG sites in spring and fall 2010 and 1980-1981.

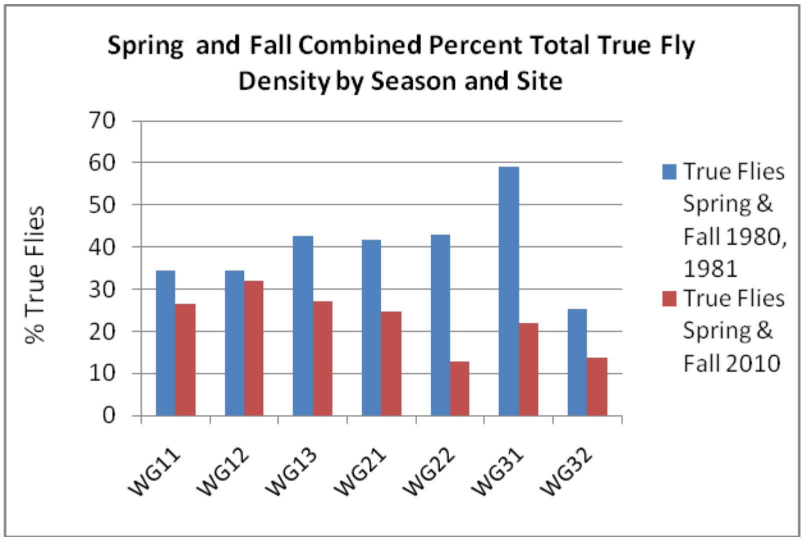
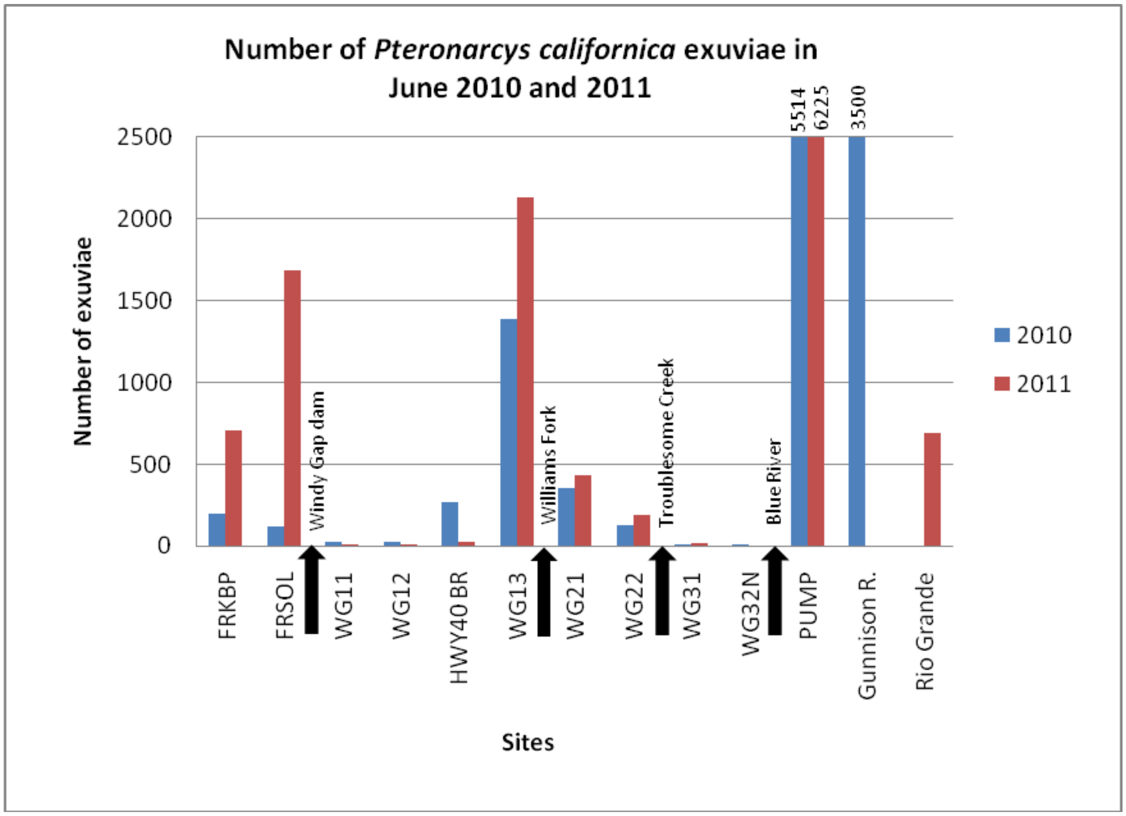


Figure 13: Percent density of true flies of the Colorado River at WG sites in spring and fall 2010 and 1980-1981.



NOTE: All sites are on the Colorado River except FRKBP and FRSOL on the Fraser River upstream of Windy Gap dam, Gunnison River in Black Canyon of the Gunnison and Rio Grande near South Fork.

Figure 14: Number of Pteronarcys californica exuviae from sample stations in June 2010 and 2011. Black arrows indicate the location of Windy Gap Dam and points where major tributaries meet the Colorado River. Pumphouse and Gunnison River station actual numbers are labeled on top of the bars for easier reading. Data for the Gunnison River and the Rio Grande are included for comparative purposes. For more detailed information on the data summarized in Figure 14, see Table 4 in the text of Job No. 1.

Job No. 2. Colorado River Mottled Sculpin Population Studies

Job Objective: Document the relative abundance and distribution of the mottled sculpin *Cottus bairdi* in the upper Colorado River basin from the upstream end of Gore Canyon to the upstream end of the Fraser River Canyon near Tabernash, Colorado, including Willow Creek and the Fraser, Colorado and Williams Fork rivers, upstream and downstream of the mainstem impoundments (Granby, Windy Gap, Williams Fork and Willow Creek reservoirs).

INTRODUCTION

The 5th edition (1991) of the American Fisheries Society (AFS) Special Publication 20 lists 39 different genera for the family Cottidae, commonly known as the sculpins that occur in the United States and Canada. The same publication lists 25 different species belonging to the genus *Cottus* in this family, making it one of the most widely dispersed species occurring in freshwater habitats in North America. Numerous species and subspecies belonging to the genus *Cottus* commonly occur in streams and in some lakes throughout the intermountain and coastal regions of western North America. In Colorado, sculpins commonly occur in streams of all sizes at elevations ranging from 1,524 m to near 3,048 m (5,000 - 10,000 feet). The Gunnison River basin upstream of the Smith Fork of the Gunnison River is only major drainage in Colorado where sculpins do not occur. Extreme water velocities and cascading waterfalls with vertical drops > 15 m in the Black Canyon of the Gunnison have blocked upstream migration by this bottom-dwelling, benthic species.

Historically, two species of sculpin have been considered native to western Colorado, the Paiute sculpin - *Cottus beldingi* and the mottled sculpin - *Cottus bairdi*. Until recently, the mottled sculpin was long considered to be the most common and widely distributed compared to the Pauite sculpin. Recent research efforts using DNA-based genetic markers on sculpins from 20 separate locations across western Colorado seem to suggest that both species occur across wide geographic areas in the Colorado River Basin (Shiozawa et al. 2010). However, the taxonomy and exact distributions of the two species remains to be thoroughly elucidated. For our purposes, the sculpin occurring in the upper Colorado River basin in Middle Park (Grand County) is the native fish of interest, whether the populations under study are the mottled sculpin, the Paiute sculpin, or mixed populations of both species. A systematic review of fishery survey data collected over the past two decades suggested that sculpins rarely occur in significant numbers in the Colorado River between Windy Gap Dam and Kremmling or in the reaches of tributary streams (such as Willow Creek and the Williams Fork River) downstream of the mainstem impoundments on these drainages (Nehring et al. 2010). The purpose of this study is to assess whether or not this is the case, and if so, discern what might be limiting their distribution. An in-depth review of the published, peer-reviewed literature was undertaken in conjunction with field investigations during 2010 to guide this investigation (see the **Literature Cited** section for a list of publications and documents reviewed). All peer-reviewed literature actually cited in the text of this report is highlighted by an asterisk (*) in the **Literature Cited** section.

Interspecific Interactions **–** Some have suggested that the high density and biomass of brown trout (*Salmo trutta*) in the Colorado River below Windy Gap Reservoir is a plausible explanation for the lack of sculpins in this reach of the river, allegedly due to predation and/or interspecific competition. However, there is ample empirical evidence from many streams in Colorado that demonstrate these two species are capable of maintaining high densities and biomass while occurring in sympatry. The Gunnison River downstream of the confluence with the Smith Fork (SF) of the Gunnison River in Delta County is probably the best direct comparison with the Colorado River in Middle Park. The Gunnison River upstream of the SF confluence has three mainstem reservoirs with a storage capacity >1 million acre-feet (AF). During most of the 1980s, adult brown trout biomass within this 6 km reach averaged 52 kg/ha and ranged up to more than 104 kg/ha. Total trout biomass averaged 146 kg/ha, ranging as high as 251 kg/ha with more than 2,000 brown and rainbow trout in the 6 km reach ≥ 400 mm total length (Nehring 1988). Nonetheless, mottled sculpin have been thriving in sympatry with these high densities of potential competitors capable of predation for 30 years.

Other investigators have demonstrated that sculpins and brown trout occur in sympatry without any apparent adverse impacts on either species, despite the fact that brown trout are an introduced, non-native predator. In Valley Creek, Minnesota, the density of the slimy sculpin (*Cottus cognatus*) ranges between 10,000 and 60,000/ha. The slimy sculpin in Valley Creek remained the most abundant fish in the stream for more than three decades, despite the fact that it existed in sympatry with brook trout (*Salvelinus fontinalis*), rainbow trout (*Oncorhynchus mykiss*) and brown trout (Reutz et al. 2002, 2003; Petrosky and Waters 1975). Moreover, average densities of non-native brown trout in Valley Creek were as high as 7,000/ha (Waters 1999).

Similarly, mean density and biomass of slimy sculpins (*C. cognatus*) in South Branch Creek, Minnesota were 8,000/ha and 63.7 kg/ha, respectively. In this stream, brown trout density existing in sympatry with the slimy sculpin ranged between 500 and 3,000/ha while biomass estimates ranged from 61 to 205 kg/ha among eight study sections over a three-year period (Newman and Waters 1989). Likewise, mottled sculpins were the numerically dominant species in three separate streams including Prickly Pear Creek, Wolf Creek and the West Gallatin River in southwestern Montana (Bailey 1952). It is noteworthy that this was true despite the fact that these streams varied greatly in width and discharge and that the sculpins occurred in sympatry with brown trout, rainbow trout, brook trout, cutthroat trout, rainbow-cutthroat hybrids, mountain whitefish (*Prosopium williamsoni*), longnose sucker (*Catostomus catostomus*), white sucker (*Catostomus commersoni*) and longnose dace (*Rhinichthys cataractae*).

The White River in Arkansas is impounded by a series of peak-power hydroelectric dams that created tailrace habitats that have been stocked with four species of non-native salmonids, including brown, brook, rainbow and cutthroat trout. These non-native salmonids were managed with restrictive size and bag limits to produce and maintain a high-quality sport fishery that has produced world record brown trout (Hudy and Rider 1989). These tailrace habitats experience wild fluctuations in flow (1 m^3/s, 129 m^3/s and 258 m^3/s) that result from the number of turbines in use for hydropower generation as well as high releases (1,416 m^3/s) for flood control (Quinn and Kwak 2000). Despite these perturbations in stream discharge and potential competition for food and living space, the Ozark sculpin (*Cottus hypselurus*) a native species in the White River thrives in sympatry with these non-native salmonids (Quinn and Kwak 2000; Pender and Kwak 2002). Densities of the Ozark sculpin ranged from approximately 16,600 to 34,000/ha across three study reaches. Similarly, sculpin biomass was estimated at 68.1, 86.2 and 89.1 kg/ha among the three reaches (Pender and Kwak 2002). Total trout biomass in the study reaches ranged from 103 to 252 kg/ha) and trout density ranged from 300 and 2,700/ha over the two-year study period.

In summary, investigators worldwide have been conducting studies for more than half a century, attempting to evaluate interspecific interactions between sculpins and salmonids, assessing predation risks and effects between these taxa (Dineen 1951; Bailey 1952; Straskraba et al. 1966; Brocksen et al. 1968; Petrosky and Waters 1975; Moyle 1977; Newman and Waters 1984, 1989; Gabler and Amundsen 1999; Pender and Kwak 2002; Ruetz et al. 2003). The results have been mixed. Some studies have purportedly demonstrated that sculpins decreased densities of benthic prey, thereby reducing the number of drifting prey available to drift-feeding trout (Brocksen et al. 1968) while other studies suggest the brown trout were the superior competitor (Reutz et al. 2003). However, Moyle (1977) concluded after review of the literature available at the time that the scientific evidence supporting competition between sculpins and trout was weak. Review of the literature in the preceding paragraphs, much of it published subsequent to the Moyle (1977) review, indicates that this still the case. Sculpins lack a swim bladder and are poor swimmers. They are benthic feeders that are cryptic by nature, whose preferred habitat is among the interstitial spaces between and beneath cobble-boulder substrates. Compared to most other fishes with which they coexist, sculpins are largely inactive, sedentary and non-migratory. All of these factors allow them to avoid predation and coexist with larger, potential predators such as brown trout or others salmonids (Brown 1999; Polivka 2007).

Growth Rates – Sculpins belonging to the genus *Cottus* rarely exceed 40 mm TL at age I and the maximum terminal length rarely exceeds 140 mm. Petrosky and Waters (1975) reported young-of-the-year (YOY) slimy sculpins averaged almost 40 mm TL by mid-September in Valley Creek Minnesota. A gravid-five year old female was only 111 mm TL in the same stream. In the West Gallatin River in Montana, YOY mottled sculpins averaged 29 mm TL on October 28, 1950 (Bailey 1952). In the same study the maximum length among 112 male and female sculpins that were aged using otoliths was 119 mm TL and 94 mm TL, respectively. During the late summer and fall of 2010, more than 1,000 mottled sculpins were measured individually measured at 20 sampling sites across 10 different streams in Colorado. Only three sculpins exceeded 130 mm TL and the largest was 140 mm TL and weighed 42.2 g (wet weight).

Life Expectancy - Among those studies where age was determined for species of *Cottus*, the maximum age attained was generally five years, with relatively few fish reaching that age. Bailey (1952) reported five years as the maximum age observed among mottled sculpins in the West Gallatin River in southwestern Montana. The same was true among slimy sculpins in Valley Creek, Minnesota (Petrosky and Waters 1975), the Utah sculpin - *C. bairdi semiscaber* (Zarbock 1952) and the river sculpin *C. hangiongensis* in Japan (Goto 1984). However, Goto (1987b, 1989b) reported river sculpins belonging to the species *C. hangiongensis* can attain a maximum age of seven to nine years in the Daitobetsu River, in southern Hokkaido, Japan.

Spawning and Reproduction – Onset of spawning activity is variable among *Cottus* species and appears to be largely dependent on water temperature. Spawning among slimy sculpins in Valley Creek, Minnesota occurs in late April with fry emergence commencing in June (Petrosky and Waters 1975). Bailey (1952) reported that mottled sculpin spawned in the West Gallatin River, Montana throughout the month of June, while egg hatching was observed the first three weeks of July, indicating that the incubation period ranged from 21-28 days. Afternoon water temperatures ranged from 7.7 – 17.2 ˚C during the spawning-egg incubation period. Mottled sculpin eggs hatched successfully when incubated in hatchery water at temperatures of 10 – 12.8 ˚C. Sac-fry averaged 7.1 mm TL at day one after hatching, and ranged in size from 5.8 – 8.1mm TL. At this temperature range, 14 days were required for total absorption of the yolk sac. Two-week old fry averaged 9.5 mm TL, ranging in size from 9.0 – 9.9 mm TL. Mottled sculpin fry ranging in size from 10-12 mm TL have been observed in the Gunnison River downstream of the confluence with the Smith Fork of the Gunnison River during the first two weeks of August on numerous sampling occasions over the past two decades.

Among the genus *Cottus*, the males (on average for a given age) are larger than the females. The males are nest builders that become sexually active earlier than the females. They court females through mating displays at the entrance to the nest. Larger males are polygynous, spawning with multiple females (Goto 1987a). Females tend to spawn with larger males and attach the egg clutch to the ceiling of the nest cavity, where the clutch is fertilized by the attendant male. Males guard and defend the nest from potential predators, at times fanning the incubating eggs, cleaning the clutches of dead or fungused eggs, and guarding the sac fry after hatching until the fry have absorbed the yolk sac and dispersed from the nest (Goto 1989a). These reproductive behavioural traits are consistent among numerous congeneric sculpin species, including but not limited to *C. bairdi* (Downhower and Brown 1980; Brown 1982), *C. gobio* (Marconato and Rasotto 1983; Marconato and Bisazza 1988) and *C. cognatus* (Mousseau et al. 1987) and even among marine sculpins as well (Westin 1969). In riverine habitats within the temperate climatic zone, the egg incubation period is temperature dependent but usually lasts from 21 to 28 days, with another 10 to 14 days from hatching to dispersal of the fry from the nest (Bailey 1952; Brown 1981; Downhower and Brown 1980; Goto 1982, 1983, 1984).

Few studies have been done to evaluate the importance of paternal care and protection for the incubating eggs, successful embryonic development and hatching. However, those studies that have been done suggest that this behavioural characteristic is of critical importance. Goto (1989a) evaluated the survival rate to hatching among 11 nests containing fertilized eggs of the river sculpin *C. amblystomopsis* where the guarding male was removed from five nests compared to six nests where the male was left undisturbed through the hatching process. Among the undisturbed nests, hatching success was 30, 50, 70, 80, 90 and 90%. Time to hatching among these nests ranged from eight to 14 days. In contrast, hatching success was zero among all nests that were left unguarded. Egg survivorship among all 11 nests remained at 95 to 99% for the first 48 hours after removal of the males. However, survivorship of developing eggs and embryos among the five unguarded nests dropped to zero at four, six, eight, 10 and 12 days post removal of the males. These findings strongly suggest that paternal parental care of the nest is of critical importance for successful natural reproduction and the long-term survival of a sculpin population. Moreover, fluctuations in stream discharge, and in particular large depletions in discharge during the three to four week egg incubation period, are likely to have deleterious consequences for population viability (Adams and Schmetterling 2007).

Food Habits – The aquatic invertebrate fauna consumed by most species of sculpins belonging to the genus *Cottus* is quite similar despite the large number of species and very wide range of habitats and climatic zones where they occur. Dipteran larvae from the families Chironomidae and Simuliidae are usually the dominant food items found in stomach samples of young-of-the-year (YOY) sculpins. This holds true for *Cottus cognatus* in Valley Creek, Minnesota (Petrosky and Waters 1975), *C. bairdi* in the Gallatin River and Prickly Pear Creek in Montana (Bailey 1952), cottids in New York (Koster 1937), *C. gobio* in Sweden (Andreasson 1971), and *C. hypselurus* in the White River in Arkansas (Pender and Kwak 2002).

Microhabitat Requirements – Sculpin species belonging to the genus *Cottus* occur in both lentic (Hanson et al 1992) and lotic environments (Zarbock 1952; Bailey 1952; Goto 1983; Petrosky and Waters 1975). Many cottid fishes live exclusively in freshwater; however some species live part or all of their life in marine habitats (Westin 1969; Goto 1983). Life histories of cottid species can vary greatly even among species existing in close proximity to each other. At least seven separate cottid species (including six belonging to the genus *Cottus*) occur on the islands of Japan that have wildly divergent life histories, including catadromous, amphidromous, lacustrine-landlocked and fluvial-landlocked (Goto 1990).

In Colorado, mottled and Pauite sculpins occur primarily in streams, but can be found in lakes and reservoirs receiving inflow from streams where they are endemic. For purposes of this study, our focus is upon those populations living primarily in stream environments. While they can occur in moderate numbers in pool habitats, juvenile and adult sculpins in Colorado are generally found in the greatest densities in rocky, cobble and boulder strewn riffle and run sections of streams. By the fall, YOY sculpins (≤ 40 mm TL) tend to be concentrated at the stream margins, living cryptically among and beneath leaves or other detritus, or burrowed into shallow sand or mud substrates during daylight hours. Adult sculpins are more concentrated in the deeper portions of the channel, preferring the interstitial spaces beneath cobble and boulder substrates ranging in diameter from 150 – 400 mm and can occur in very high densities where these substrates are not imbedded in fine sediments, mud or small gravels. Surface water velocities can exceed 1 m/s; however, preferred nose velocities are mostly likely near zero.

Thermal Tolerance and Preferences – Studies conducted on the range of thermal tolerances and preferences for any species of sculpin are rare. One was completed in the 1970s on the freshwater sculpin (*Cottus cognatus gracilis*) by Otto and O'Hara-Rice (1977). These investigators reported that the upper lethal temperature was approximately 26.5 ˚C. However, the critical thermal tolerances were variable and dependent upon the acclimation temperature. Incipient lethal temperatures ranged from 18.5 to 23.5 ˚C. Avoidance temperatures ranged from 15.2˚C for sculpins acclimated at 5˚C and 21.5˚C for fish acclimated at 15˚C. Preferred temperatures for 5˚C and 15˚C-acclimated fish were 9˚C and 12˚C, respectively, and the final temperature preferendum was about 10˚C. Symons et al. (1976) in another study reported the mean and median preferred temperatures for the slimy sculpin as 13.1 and 13.3˚C. These investigators reported upper lethal temperatures in the range of 25-26 ˚C. Across their range in western North America, it is well known that the mottled sculpin and Paiute sculpins occur in sympatry with many salmonid species whose temperature tolerances, preferences and incipient lethal levels are in a range very similar to those for *C. cognatus gracilis* (Coutant 1977). Ferguson (1958) reports the final thermal preferendum for the mottled sculpin as 16.5 ˚C.

Life History and Distribution **-** Sculpins (family Cottidae) are among the most widely distributed species of fish capable of occupying a great range of aquatic and marine environments in the Northern Hemisphere. They occur throughout Scandanavia, Europe and Asia. In North America *Cottus* species occur as far south as the Carolinas on the Atlantic coast and northward into Canada, throughout much of interior North America, including the Great Lakes all the way to the Arctic Circle, in the intermountain west from New Mexico and Arizona into the Canadian Rockies and on the Pacific coast from California to Alaska. In North America, *Cottus* species occurring in streams tend to lead a very sedentary existence, and are often very territorial. Petty and Grossman (2007) demonstrated that adult mottled sculpin in a 1-km study reach of Shope Fork, a 4th order tributary of Coweeta Creek, NC are highly territorial, and extremely sedentary, with a median movement distance of only 1.42 m, while more than 80% of all movements were less than 3 m linear distance. Mean home-range size varied inversely with size and age. Among juvenile mottled sculpin (≤ 48 mm standard length-SL) mean home range size varied from 1.6 to 10.9 m^2 over the three-year study. The mean home-range size among small adults (49-64 mm SL) varied from 0.8 to 8.7 m^2 over the same time period. Mean home-range size was the smallest among large adults and varied less over the course of the study (1.7 m^2 to 2.8 m^2). They found that small and larger adults occupied territories that were deeper and that had higher current velocities and higher amounts of cobble and boulder than areas used by juveniles. Territories of adults were more likely to be located in areas of stable substrate that never went dry. All life stages (juvenile, small adult and large adult) selected home ranges and habitat patches dominated by cobble and boulders and areas where the mean column water velocities were ≥ 38 cm/s. The mean column water velocity for large adults (≥ 65 mm SL) was

48 cm/s. All three life stages of mottled sculpins in Shope Fork avoided habitats dominated by gravel, sand, silt and debris (Petty and Grossman 2007). Other investigators have shown that sculpins in montane streams move greater distances, in some cases ≥ 200 m in just a few weeks time (Schmetterling and Adams 2004; Natsumeda 2007). Natsumeda (2007) found that higher numbers of Japanese fluvial sculpin *Cottus pollux* moved farther in high-flow years compared to a low-flow year, but that a weir < 0.5 m high blocked all upstream movement of these fish.

Sculpins as Indicator Species – Traditional fisheries management in coldwater stream ecosystems historically has focused on salmonids as the umbrella species to conserve and protect all aquatic vertebrates (Adams and Schmetterling 2007). Often times the salmonids are introduced or non-native species that may have different spatial and temporal microhabitat requirements that may not coincide with those of the native fish assemblage. Adams and Schmetterling (2007) point out that sculpins are well suited for use as a bioindicator species for several reasons. First, sculpins are a sedentary benthic fish, and poor swimmers that do not have a large home range. This makes studying them easier. Second, man-made barriers or in-channel obstructions that can be negotiated by sculpins will almost certainly be passable to almost all other fish species; but the converse is not true (Schmetterling and Adams 2004; Natsumeda 2007). Third, sculpins are very sensitive to metal contaminants (Maret and MacCoy 2002). Brinkman and Woodling (2005) reported that mottled sculpins are so sensitive to zinc that they are not adequately protected by existing federal water quality standards. Fourth, sculpins are a nest-building species that require five to six weeks from spawning to fry dispersal from the nest. In most freshwater streams spawning occurs between spring and mid-summer, at a time in the intermountain west when stream flows are near the peak discharge during snow melt and then declining. Sudden changes in streamflow during the egg incubation period could result in nest abandonment or drying out of the nest. Sculpin egg clutches incubate on the underside of rocks up in the water column as opposed to salmonid eggs that incubate in gravels on the stream bottom. This makes incubating eggs of sculpins more sensitive to alterations or reductions in stream discharge than those of salmonids.

STUDY DESIGN

The job objective for this study is to document the relative abundance and distribution of the mottled sculpin *Cottus bairdi* throughout the upper Colorado River basin. The lower terminus of the study area was downstream of Gore Canyon near Radium, Colorado. The upstream terminus was on the Fraser River near the confluence with St. Louis Creek near Fraser, Colorado. The upstream electrofishing station on the Colorado River was northwest of Grand Lake, Colorado near the Winding River Ranch. Other streams and stream reaches within the study area included Arapaho, Beaver, Little Muddy and Troublesome creeks, the Colorado and Williams Fork rivers, upstream and downstream of the mainstem impoundments (Granby, Windy Gap, and Williams Fork reservoirs) as well as Willow Creek upstream and downstream of Willow Creek Reservoir). Bank electrofishing surveys were conducted at 26 separate locations in the upper Colorado River basin in Grand County. Results of three additional bank electrofishing surveys from other westslope streams downstream of major mainstem impoundments were included for comparison purposes with fish population data from the survey sites in Grand County. These streams were the Dolores River below McPhee Dam, the Gunnison River below Crystal Dam and the Yampa River below Stagecoach Dam.

We used the two pass removal method (Seber and LeCren 1967) to estimate the population size at 28 of 29 study sites. At the study site on the Dolores River, four electrofishing passes were needed to deplete the sculpin population to the level required to obtain a satisfactory level of precision and accuracy for estimating population size. At most sampling sites, length and weight information was taken on up to 100 individual fish of all fish species collected with the exception of speckled dace (*Rhinichthys osculus*). In most cases, at most sampling sites, all fish collected were identified to the species level. In a few instances, it was not possible to definitely identify small members (< 70 mm TL) of the sucker (Catostomidae) or minnow (Cyprinidae) families to the species level. The body conformation and dorso-ventrally flattened head of sculpins made it easy to identify members of the genus *Cottus* down to the minimum sizes encountered (< 20 mm TL). An ultra-light weighing scale made it possible to determine the wet weight of sculpins down to a minimum detection limit of 0.1 g with a precision of ±0.1 g. Larger fish (≥ 150 mm TL) were weighed on larger platform balances with a precision level of approximately ±10 g. At all sites we attempted to gather enough length-weight data pairs to generate a length-weight regression for estimation of biomass for each species. When the sample size was inadequate a standard weight equation was used to estimate biomass. All data sets were run through the Division of Wildlife software program JAKOMATIC to determine population estimates with 95% confidence limits, density estimates (numbers/ha) and estimate biomass (kg/ha).

Study section length was measured along one bank to the nearest meter. The average width was determined to the nearest 0.1 m by measuring the cross-sectional width at a minimum of 10 transects between the upper and lower terminus at each study site. Study section length varied with stream size and the relative abundance of the target species, ranging from 61 m to 335 m. All study reaches included riffle areas where the predominant substrate types included cobble, rubble and/or small boulders, the substrate type preferred by sculpins in streams. The stream names and stream reaches, including study reach length, width, area, GPS coordinates and sampling dates are shown in Table A.1 in the Appendix of this report. Fish population density and biomass estimates for brown, rainbow and brook trout and sculpins at the 29 stream reaches sampled are summarized in Table A.2 in the Appendix of this report.

RESULTS AND DISCUSSION

Fish Population Density and Biomass Estimates –Brown trout densities (numbers/ha) across the 26 sampling stations within the Colorado River basin ranged from 79 to 3,333. Brown trout biomass estimates (kg/ha) at those same sites ranged from one to 236. Rainbow trout were present at 19 of 26 sampling sites. Brook trout were present at eight of 26 sample sites. At those sites where two or three salmonid species were present, total trout biomass across the 26 sampling locations within the Colorado River basin ranged from nine to 312 kg/ha.

Sculpins were collected at 18 of 26 sampling reaches within the Colorado River basin. Among the sites where sculpin were present, densities ranged from one to 21,252/ha. At those same sites sculpin biomass ranged from <0.01 to 70 kg/ha. Sculpins were abundant at 12 of 14 sampling stations located on streams that were either upstream of mainstem impoundments or tributary streams unaffected by impoundments (Table 1). The only two streams in this category where sculpins were not collected were at the downstream terminus of Little Muddy Creek and Troublesome Creek at the confluence with the Colorado River. In both cases, the effects of heavy sedimentation and lack of any cobble-rubble habitats rendered those stream reaches unsuitable for sustaining mottled sculpins. At the 14 sampling sites, sculpin densities averaged 4,228/ha, ranging from 0 to 21,252/ha (see Table 1 and Appendix Table A.2 for details).

Smaller creeks that are tributary to the Colorado and Fraser rivers sampled in 2010 included Arapaho, Beaver, Little Muddy, Troublesome and Willow creeks. YOY sculpins were collected from four of the five creeks at five of seven sites sampled. The only two sampling sites where no sculpins were collected were on the Troublesome and Little Muddy creeks at the confluence with the Colorado River. Both sampling sites were inundated with sand, silt and clay sediments and were largely devoid of any cobble-rubble substrates that are the preferred substrate type for sculpins.

At the 11 sampling stations downstream of mainstem impoundments in the Colorado River basin, sculpins were collected from only five stream reaches. Only one sculpin was collected from four sampling stations on the Colorado River between Windy Gap Dam (WGD) and the confluence with the Blue River at Kremmling. That fish measured 138 mm TL and was captured in a steep riffle approximately 50 m upstream of Grand County Road 57/578 (Hitching Post) Bridge, 1.5 km downstream of WGD. The combined reach length sampled at those four sites was 0.976 km (0.61 miles or 3,200 feet). Excluding the sampling site approximately 0.1 km downstream of Willow Creek Reservoir (WCR), average sculpin densities were only 21/ha, ranging from zero to 167/ha (Table 1). The estimated density of sculpins at the site immediately below WCR was extremely high (15,003) and inflated due to a low capture probability (p=0.11) on the first electrofishing pass, resulting in a very imprecise estimate with a 95% confidence limit (CL) of ±25,746. At the sampling site on Willow Creek at the C Lazy U Ranch upstream of WCR the density estimate was higher (21,252/ha) but much more precise with a 95% CL of ±3,531, owing to a much higher capture probability (p=0.39).

Table 1 Average densities (N/Ha) for trout and sculpins upstream and downstream of mainstem impoundments (Granby, Williams Fork, Willow Creek-WC and Windy Gap reservoirs) versus streams downstream of mainstem impoundments outside of the Colorado River drainage.

Category	Sample Size (N)	Trout Density (N/Ha)		Sculpin Density (N/Ha)		Trout %	Sculpin %
		Ave	Range	Ave.	Range		
Above Impoundments Av.	13	1,842	192 – 9,585	4,228	0 – 21,252	30	70
Below Impoundments Av.	11	1,020	626 – 1,742	1,611[a]	0 – 15,003[a]	39	61
BelowImpoundments Av.[b]	10	1,023	626 – 1,742	272	0 – 2,524	79	21
Colorado R. @ Radium	1	1,335	--------	7,194	-------	16	84
Dolores R. ↓ McPhee Dam	1	95	--------	6,453	-------	2	99
Gunnison R. ↓ Curecanti Dams	1	2,070	--------	4,348	-------	32	68
Yampa R.↓ Stagecoach Dam	1	194	--------	968	-------	18	82

Notes: [a] Includes reach on Willow Creek 100 meters below Willow Creek Reservoir (WCR).
[b]: Below mainstem impoundments without the Willow Creek site 100 meters below WCR.

We suspect that the sculpins captured at the Willow Creek site immediately below WCR had spilled or been flushed out of WCR during releases from the dam, for two reasons. First, these sculpins were mostly larger, older fish based on the range of sizes captured. Only 2.1% (8 of 376) of the sculpins collected were ≤ 60 mm TL, indicating that the 98% of the sculpin collected were ≥ two years of age. It is noteworthy that water releases from WCR for 18 consecutive days during May and June of 2008 averaged 334 ft^3/s, ranging from 161 to 857 ft^3/s. Similarly, in 2009 water releases from WCR averaged 172 ft^3/s for 15 days, ranging from 100 to 225 ft^3/s. Sculpins do live in reservoirs, but are weak swimmers and when accidentally entrained in fast moving water, they would be subject to flushing over an overflow outlet or through a discharge valve. In 2010, water releases into Willow Creek were a rare occurrence, never exceeded 98 ft^3/s and never lasted for more than one to two days. June is the month that male sculpins would most likely be seeking out an acceptable nesting site for spawning. July and early August would be the period of egg incubation and hatching. During June, July and August of 2010, the mean monthly discharges from WCR were 15.1, 19.8 and 7.3 ft^3/s and ranged between one and 7 ft^3/s for 23 days during June. Due to the average width of the channel (10.7 m) and low discharge, very low spawning success would be expected. Not surprisingly, only 0.5% (2 of 376) sculpins measured during the survey on September 15, 2010 were ≤ 34 mm TL, the maximum size to be expected for YOY mottled sculpins in September. Second, if the very high density of sculpins observed at this site immediately below WCR was representative of natural reproduction and a healthy, self-sustaining population, we would expect to capture sculpins in the creek downstream as well. However, none were captured or observed at the survey site approximately 2 km downstream from WCR.

The sculpin population size and age structure was much different at the sampling reach on Willow Creek approximately 3 km above WCR at the C Lazy U Ranch. Forty-seven of 159 sculpins (29.6%) measured were ≤ 34 mm TL, the maximum expected size for YOY sculpins. These fish averaged 27.8mm TL, ranging from 20 to 34 mm TL. At this site the average channel width was 7.3 m and mean monthly discharges for June, July and August were 328, 55 and 27 ft^3/s. At the upstream sampling site on Willow Creek approximately 1.5 km above confluence with Cabin Creek, YOY sculpins were smaller, averaging 20.5 mm TL and ranging from 17-24 mm TL. Age-one sculpins at this site averaged 44 mm TL, ranging in size from 34 – 51 mm TL.

Two sampling surveys were completed on the Williams Fork River upstream of Williams Fork Reservoir (WFR) during 2010. The upstream site was at the confluence with Ute Creek approximately 16 km upstream of WFR, and 1 km upstream of the Keyser Creek confluence. Estimated sculpin density was 1,964/ha at this site, comprising 82.5% of the fish density. At the lower site above the first bridge crossing 0.2 km upstream of WFR, estimated sculpin density was 5,404/ha, comprising 81.5% of the fish density. (See Appendix Tables A.1 and A.2 for details).

No sampling was conducted on the Williams Fork River downstream of WFR in the fall of 2010. However, at least 16 electrofishing surveys have been completed at three separate sampling locations on this reach between May 2001 and November 2006. Sculpins were collected on only five of the 16 surveys, and four of the five collections were at the sampling site immediately below WFR at the USGS gaging station, less than 50 m below the dam outlet. The total number of sculpins collected during each of those five surveys ranged from one to four, never comprising more than 0.9% of the total number of fish collected, a sharp contrast with the surveys upstream of WFR where sculpins comprised more than 80% of the total fish density.

Four separate reaches of the Fraser River upstream of WGR were surveyed by electrofishing in the fall of 2009 and 2010. Estimated sculpin numbers ranged from 64% to 85% of the total trout and sculpin density, averaging 2,673/ha and ranging from 1,651 to 4407/ha. See Tables A.1 and A.2 in the Appendix for detailed information on site location, density and biomass estimates.

On the Colorado River at Winding River Resort, upstream of Shadow Mountain and Granby reservoirs, the estimated densities of brown trout and sculpin were 1,630 and 2,912/ha. Similarly, estimated densities of sculpin and brown trout on the Colorado River between Shadow Mountain and Granby reservoirs were 2,524 and 98/ha, respectively. However, at the sampling site at the Grand County Road 627 Bridge, approximately 3 km downstream of the Granby Dam outlet, estimated brown trout density was 1,678/ha while estimated sculpin density was 167/ha. At the Horn Ranch sampling station, approximately 0.2 km upstream of the confluence with the Fraser River above WGR, the estimated densities of trout and sculpins were 974/ha and 24/ha, respectively.

On the Colorado River upstream of WGR, the size and age structure of the sculpin population varied greatly between the four sampling sites. At the Horn Ranch, five of the six sculpins captured ranged in size from 107 to 140 mm TL, suggesting those fish were ≥ age four and most likely immigrants from the Fraser River some 200 m downstream. There were no YOY sculpins collected at this sampling station. Similarly, 33 of the 44 sculpins captured at the Grand County Road 627 Bridge sampling site were ≥ 100 mm TL and 43 of 44 were ≥ 72 m TL, suggesting that virtually all of the sculpins were age three or older, and 75% (those ≥ 100 mm TL) were probably at least age four or older. Only one sculpin at this site was of a size (25 mm TL) suggesting that it was a young-of-the-year. A review of the discharge records for the Colorado River below Granby Dam indicate that mean monthly discharge levels have never approached 100 ft^3/s for the months of May through September from 2001 through 2010, ranging from a low of 15.6 ft^3/s in September 2001 to a high of 84.5 ft^3/s in May 2008. The lack of natural reproduction by sculpins is not surprising, given the lack of flushing flows for more than a decade, an over width channel, chronically low water velocities, and the interstitial spaces beneath cobble-rubble substrates clogged with sediment or covered with beds of rooted aquatic macrophytes, High velocities riffles dominated by sediment-free, cobble-rubble substrates are the preferred microhabitats that male sculpins seek out for nesting sites during the late-spring, early summer months (June to mid-August).

Sculpins were captured in significantly greater numbers on the reach of the Colorado River between Granby Reservoir and Shadow Mountain Dam outlet. A total of 307 sculpin were captured, including 15 YOY sculpins, ranging in size from 22 to 25 mm TL. Fifty-nine of 115 sculpins measured were≥ 100 mm TL and most likely ≥ age four. Only nine of 100 sculpins measured that were greater than age one ranged in size from 74-88 mm TL. These fish were probably the 3-year old cohort (2008 year class). Ninety-one of 100 were ≥ 92 mm TL, and probably ≥ age four (age classes 2006 and 2005) in a reach of stream 91.5 m in length and dominated by sediment-free, large cobble and small boulder substrates. Given that the sculpin population in this reach is dominated by fish likely to be age four and older and that most sculpins are sexually mature at age two and the substrate and microhabitat are ideal for sculpin nesting sites, the apparent lack of reproductive success is somewhat enigmatic. A thorough review of the scientific literature (see the Introduction section in this report) indicated that the time from spawning to hatching for many species of sculpins in temperate climatic zones in the northern hemisphere is four to six weeks. Given that 1) the nests must be guarded by mature male sculpins from spawning to hatching, 2) nesting site selection will be dependent on stream discharge, and 3) nests will be vulnerable to large fluctuations in flow, particularly dramatic reductions in discharge, it is possible that spawning success in this reach could be closely tied to variations in flow between June (presumed period of spawning) and mid-August (presumed period of hatching and dispersal of fry from the nests.

Table 2 Summary of discharge records for various time periods for estimated spawning, egg incubation and hatching periods of sculpins in the Colorado River basin in Grand County versus estimated sculpin year class size (n/ha) for various years from 2006 through 2010.

Sculpin Age(yr) & (n/ha)	**Year**	**Discharge (ft^3/s) records for various time periods**						
		Average June Discharges			**July Discharge**		**8/1-15 Discharges**	
		Mo. Mean	**6/15-30 Av.**	**Range 6/15/-30**	**Mo. Mean**	**7 Day Min./(% Δ)**	**15 D Mean**	**7 Day Min./(% Δ)**
Colorado River between Shadow Mountain Dam and Lake Granby								
0+ (75)	2010	1,579	913	50-1,566	50	50/ -95%	41	40
1+ (5)	2009	1,292	1,466	905-2,080	224	50/ -97%	56	40
2+ (86)	2008	768	730	482-1,234	72	50/ -93%	102	67
3+ (707)[a]	2007	655	678	257-1,401	79	50/ -93%	40	40
4+ (610)[a]	2006	439	50	50	50	50/ 0%	40	40
Fraser River above Windy Gap Reservoir at Kaibab Park in Granby								
0+ (292)	2010	931	597	331-849	143	83/ -86%	98	79
Willow Creek @ C Lazy U Ranch above Willow Creek Reservoir								
0+ (4,142)	2010	328	164	88-283	55	37/ -77%	35	29
Williams Fork River 0.2 km upstream of Williams Fork Reservoir								
0+ (155)	2010	625	342	168-540	61	23/ -93%	103	78
1+ (1,174)	2009	669	612	474-814	194	109/ -82%	84	71

[a]: These estimates of year class strength are probably fairly accurate based on excellent separations of year classes based on length-frequency distributions; however, the size of these year classes may be due in part to immigration out of Shadow Mountain Reservoir.

A review of discharge records from Shadow Mountain Dam from 2006 through 2010 suggest that between year variations in flow for the critical period from June through mid-August may well explain the explain the apparent low reproductive success in 2008, 2009 and 2010 compared to 2006 and 2007 (Table 2). A similar analysis for sculpin year-class strength for 2009 and/or 2010 versus discharge patterns from mid-June through mid-August for sampling sites on the Fraser and Williams Fork rivers upstream of mainstem impoundments and Willow Creek upstream of WCR reveal similar results. These data sets imply that the greater the percent decrease in discharge between June 15-30 (estimated spawning period) compared to the seven-day minimum discharge during July (estimated egg incubation period) or the first 15 days in August (fry hatching/yolk sac absorption period) the greater the depression of YOY sculpin year-class strength. This sort of a negative relationship between high discharge levels and fluctuations in stream discharge during the early life stages (incubating eggs, intra-gravel, post-hatch alevin, and two to four week old fry emergence) among salmonids is well documented in Colorado (Nehring and Anderson 1993). Likewise, spawning and nesting success among stream-dwelling sculpins are also highly vulnerable to discharge fluctuations (Adams and Schmetterling 2007).

A total of 453 YOY sculpin (based on length-frequency distribution) were collected at nine of 14 sampling sites upstream of mainstem impoundments in the Colorado River basin in 2010. In contrast, a total of 18 YOY sculpins were collected at all sampling sites combined downstream of the five mainstem impoundments, Granby, Shadow Mountain, Willow Creek, Williams Fork, and Windy Gap reservoirs. Fourteen of those 18 YOY sculpins were collected downstream of Shadow Mountain Dam, the one stream reach that is largely not impacted by sedimentation, and usually experiences substantial flushing flows during the month of June.

CONCLUSIONS

Transmountain diversions and mainstem water storage reservoirs have been used to supply the needs of agriculture and development of the infrastructure required to support an expanding population base in Colorado for a century or more. This has been especially true for the Colorado River basin. Lying just across the Continental Divide west of the Front Range population centers of Fort Collins, Greeley, Boulder and the greater Denver Metropolitan area, the headwater streams in the basin have been diverted through direct transmountain diversions such as the Grand Ditch in the northern part of Grand County and the Moffat Tunnel, which collects and transports water from almost every stream tributary to the Williams Fork and Fraser rivers in the southeast portion of the county. Five mainstem dams (Granby, Shadow Mountain, Williams Fork, Willow Creek and Windy Gap) all collect, store, divert and release water to satisfy a variety of water rights, agricultural, municipal and industrial needs and purposes. Windy Gap was the last mainstem dam on the upper Colorado River in Grand County, completed in the early 1980s.

While there have been attempts to address the environmental costs and impacts accruing to the upper Colorado River as new projects and diversions came on line during the last half of the 20th century, it is only within the last decade that the total cumulative impacts have begun to be noticed and serious attempts made to address the issue(s). There are at least six significant issues that need to be taken into account when considering the overall health of the Colorado River downstream of Windy Gap Dam through Middle Park to the confluence with the Blue River near Kremmling. These are 1) restoration of channel connectivity, 2) channel reconfiguration, stream power and flushing flows, 3) sediment deposition and transport, 4) water temperature, 5) encroachment of rooted aquatic vegetation, and 6) whirling disease.

While the six issues listed above are all important, we have ranked them (from highest to lowest) in what we consider a descending order of priority. The problems of channel armoring and chronic sedimentation and clogging of the interstitial spaces in the cobble-rubble dominated riffles areas has been ongoing in the upper Colorado River basin for more than half a century. These proposed firming projects at Windy Gap and the Moffat Tunnel are only going to further exacerbate this situation. Two things must be done if there is to truly be any hope of enhancement of aquatic ecosystem in the upper Colorado River in the future. A bypass channel around Windy Gap Dam and a major investment in stream channel reconfiguration for the Colorado River below WGD are both equally important and the only way true enhancement has any possibility of success. Either one without the other will have virtually no chance of succeeding.

Restoration of Channel Connectivity - Since WGR came on line, movement of all fish species up and down the river has been impeded. Downstream drift of aquatic insect larvae has been interrupted. A bypass channel around WGR would re-establish channel connectivity and allow both of these important biological functions to be restored. It would allow free movement of spawning trout and other species of fish in both directions and open the river below WGD for recolonization by aquatic insect species and other invertebrates through downstream drift.

Our 2010 study of 26 sites in the basin has conclusively demonstrated that a native fish, the mottled sculpin, has been extirpated from the Colorado River below WGR all the way to the Blue River confluence. A bypass channel around WGR would open the river below the lake to the possibility of re-colonization by sculpins from the Fraser River upstream. It is noteworthy that the densities of mottled sculpins are very high in the Colorado, Fraser, and Williams Fork rivers, as well as Arapaho and Willow creeks upstream of the mainstem impoundments. Sculpin density is also high in the Colorado River downstream in and below Gore Canyon, after the inflow of water from the Blue River essentially restores the "health" and flows of the river in a manner that largely mimics the natural hydrograph. Likewise, sculpins occur in significant densities with multiple year classes present in non-impounded tributary streams such as Beaver and Troublesome creeks. Given these distributions, it is not likely that the factor(s) responsible for extirpation of the species downstream of WGR are associated with water quality parameters as they exist in these other locations. It is a function of flow depletions beginning in the late-spring and early-summer months during run-off period and the loss and impairment of physical habitat due to lack of sustained flushing flows.

Similarly, the giant stonefly *Pteronarcys californica* still exists in low to moderate densities in the Fraser River upstream of WGD, along with other aquatic insect species that have been significantly reduced or extirpated below WGD. Reconnection of the river channel with a bypass around WGD would open the stream of recolonization by aquatic insects of many species through downstream drift.

***Channel Reconfiguration, Stream Power and Flushing Flows* –** The combined effects of 1) collection and diversion of water in the Fraser River Basin by the DWD through the Moffat Tunnel, and 2) impoundment, pumping, storage and export of water from the Colorado River basin to the East Slope via the Grand Ditch and the Alva B. Adams Tunnel at Grand Lake have resulted in significant, cumulative degradation of the stream ecosystems in Grand County. These more serious initial impacts began on the Colorado River downstream of Granby Dam mid-way through the 20th century, due to the lack of flushing flows and the associated decrease in stream power. The severe reductions in the frequency, magnitude and duration of high flushing flows below WGD since its construction in the 1980s, has severely reduced the stream power in the Colorado River downstream of Windy Gap. The combined, cumulative effects of the proposed DWD Moffat Tunnel Enhancement Project and the NCWCD Windy Gap Firming Project will greatly exacerbate this situation unless the Colorado River stream channel width is significantly reduced. That is the only way to increase stream power in the face of larger and continuing water withdrawal from the basin.

Periodic flushing flows of sufficient magnitude and duration are critically important for maintaining the biotic integrity of stream ecosystems. Project proponents have proposed at various times that a discharge in the range of 450 to 600 ft^3/s for less than 3 days is an adequate flushing flow for the Colorado River below WGD. However, a true flushing flow requires a discharge substantially greater than 1,000 ft^3/s for several weeks, as occurred in 2010 and 2011. Flows of lower magnitude and shorter duration will not result in the deep cleaning of the cobble-boulder substrates in the riffles, chutes or pools.

Fine Sediment Deposition and Transport – The combined impacts of extended droughts, impoundment, storage of spring flushing flows in Willow Creek, Williams Fork and Granby reservoirs, interception and pumping of water out of WGR, and depletions as a result of transmountain diversions through the Moffat and Alva B. Adams tunnels have greatly exacerbated sediment deposition in the Colorado River below Granby and Windy Gap dams. Sediment deposition can be ameliorated and even prevented by adequate flushing flows of sufficient periodicity, magnitude and duration. However, all the benefits of a true flushing flow are negated when accumulated sediments in a reservoir are mobilized, entrained and then flushed downstream during the low-flow period in late summer and fall. At least twice since 2001, WGR has been drained and large volumes of sediment flushed into the Colorado River in mid to late summer, long after spring flushing flows were available to transport the sediment downstream. The first occurrence was during the severe drought during 2001-2002 when WGR was drained in mid-summer to eliminate evaporation losses. The second occurrence was in late summer 2010, when the lake was drained to dredge sediments out of the lake that had accumulated in front of the penstock intakes for the pumps in WGR. On both occasions, accumulated sediments in the reservoir were mobilized, entrained and subsequently settled out in the first few miles of river below the dam.

Sediments are most efficiently flushed and transported out of the receiving stream at the time of entrainment. When this does not happen, deposited sediments harden and require greater stream power for a longer duration to be mobilized and transported. Due to the aforementioned problems resulting from these transbasin diversions over the past half century, the Colorado River channel is approximately 50% overwidth. Stream power (to transport sediment) has been adversely impacted. Sediment deposits choke the interstitial spaces among the gravel, cobble and boulder substrates in the riffle areas.

Riffle areas are critical to maintenance of the biotic integrity of the stream on at least four levels. First, riffles are the primary zones of aquatic insect production. Second, healthy aquatic insect populations are the primary food source for foraging fish. Third, riffle habitats are the primary spawning zone for rainbow trout, brown trout, and sculpins. Gravels and cobbles imbedded with sediments are an impediment to spawning success of trout and sculpins. Large cobble, rubble and small boulder substrates in the steep gradient riffles that are sediment-free and un-imbedded are the prime habitats necessary for maintaining healthy populations of sculpin, the giant stonefly *Pteronarcys californica* and other aquatic insects. Fourth, free-stone cobble substrates at the margins of the channel, even in pools are critically important microhabitats for young-of-the-year (YOY) rainbow and brown trout as well as sculpins and dace. When those areas are clogged with sediment build-up, the carrying capacity of the stream for successful rearing of YOY trout and sculpins is greatly degraded. A bypass channel around WGR should eliminate the need for inadvertent or intentional flushing of sediments during the low-flow summer periods, adding a significant layer of protection for the river below the WGR at the times when it is most vulnerable to stress. When WGR needs to be drawn down to eliminate evaporation losses (which happened at least once in the past decade) sediment flushing would not occur since all the water would be flowing through the bypass channel around WGR once the lake pool was drained and the outlet works closed off.

Water Temperatures – Any time water is impounded, solar radiation will heat the water. Impounded water always heats faster in shallow reservoirs such as WGR. Water heating in the Colorado River in Grand County will be most problematic for coldwater species such as trout and sculpins in late summer, at a time when the river flows are at the seasonal low and the stream water temperatures are already high. Periodic monitoring of water temperatures at various locations in the Colorado River basin carried out by the CDOW since the early 1980s has shown that daytime water temperatures can range from 20 to 25 ˚ C during the summer months, particularly in below average water years (Nehring and Thompson 2001, 2003). The combined effects of elevated water temperatures and severely reduced flow can act in a negatively synergistic manner, posing serious risks to the aquatic fauna. A bypass channel around WGR eliminates the problem of additional heating of the impounded water during the late-summer low flow period prior to its release to the river.

Rooted Aquatic Vegetation – Growth and expansion of aquatic vegetation mats in the Colorado River since the construction of WGR has been extensive. It has been especially problematic between WGR and the confluence with the Williams Fork River at Parshall, Colorado. Rooted aquatic vegetation now essentially has invaded all parts of WGR. Many aquatic plants seed and populate other habitats through fragmentation. Fragments float out of WGR, settling out in the slower moving areas of the river. Once established, they continue growing in size and number, slowing the flow of water, trapping fine sediments and particulate organics that (in turn) provide a nutrient base for growth and expansion of the vegetation mat. Vegetation mats choke out the interstitial spaces in the cobble-strewn margins of the river that are critical microhabitat rearing areas for YOY trout, sculpins, dace, and other fishes. Vegetation mats can be seen everywhere on the river between WGR and the confluence with the Blue River. Downstream of WGR, the problem is greatest between the dam and the confluence with the Williams Fork River. However, after more than a half century of impoundment of the Colorado River at Granby Dam, and no flushing flows at all except during rare spilling events, the choking of the river channel with sediment and aquatic vegetation mats is most severe from Granby Dam to the confluence with the Fraser River.

Vegetation mats become increasingly difficult to dislodge when true flushing flows have become all but a thing of the past as flow depletions have increased, particularly during the spring flush period. The Gunnison River in western Colorado, downstream of Black Canyon National Park (BCNP) is an outstanding example of a healthy river corridor. Above the confluence with the Smith Fork of the Gunnison, it is only about 10%-20% wider than the average channel width of the Colorado River below WGR. The Gunnison River Gorge (GRG) receives large, annual sediment inputs during the summer months from side canyon flash floods that result from summer thunderstorms. These large inputs of sediment led to development of huge sediment laden vegetation mats during periods of extended drought, such as occurred in 2001 and 2002. However, these vegetation mats and massive amounts of sediment have been largely eliminated from long sections of the GRG by flushing flows ≥ 3,000 ft^3/s that are sustained for 2-4 weeks in many years, with peak discharge levels as high as 6,000 – 8,000 ft^3/s. The GRG rarely experiences flows below 300 ft^3/s, due to minimum flow agreements. The only time between October 1991 and September 2010 that flows in the GRG dropped below 300 ft^3/s, was from November 2002 until early May 2003 at the end of the extended, severe drought of 2001-2002 when daily discharge levels ranged from 248 – 313 ft^3/s. Mean monthly flows in the GRG have exceeded 1,000 ft^3/s for 85 of 240 months between October 1991 and September 2010. The GRG downstream of BCNP has outstanding populations of the giant stonefly and sculpins, both of which coexist with perhaps the best stream trout sport fishery in Colorado. A bypass channel around WGR would eliminate the seeding of the Colorado River by fragmentation of rooted aquatic vegetation flowing out of the lake. Over time, with true flushing flows (as will occur this year), the rooted aquatic vegetation mats could be dramatically reduced in size and number.

Whirling Disease - The Whirling Disease (WD) issue with WGR being a serious point source of infection is the one issue that has largely resolved naturally over the past decade. Much has been learned through intensive, worldwide research efforts beginning in the mid-1990s about the way this parasitic disease of trout and salmon operates in lake and stream ecosystems. What has been learned that applies directly to WD in WGR is that the aquatic worm population in the lake during the 1990s was complex of at least six different types of tubificid worms. Only one of those types of worms was the worm host for the *Myxobolus cerebralis (Mc)*, the parasite that can cause WD. However, in the 1990s the worm that was susceptible to infection by the *Mc* parasite was one of the dominant worm types in WGR, especially in the main channel of water flowing into and through WGR. The high densities of those worms in the main channel of water flowing into WGR resulted in very high levels of the floating TAM spores that were infectious to trout flowing out the lake and into the Colorado River (Nehring et al. 2003). Most parasites have a deleterious effect on the host, in this case the lineage 3 *Tubifex tubifex.* When the infection is serious enough, the lineage 3worm is reproductively impaired (Shirakashi and El-Matbouli 2009). However, the other species or strains of worms in the lake (*T. tubifex* lineages 1, 5 and 6 and at least two other species) do not become infected by the parasite, and therefore continue to reproduce successfully, while the heavily infected lineage 3 worms do not. Over time (apparently about a decade-from 1991 to 2001) the once-dominant lineage 3 worm which comprised about 75% of the *T. tubifex* (*Tt*) population in 1998 decreased in abundance such that by 2005 it accounted for only 5% of the *Tt* population, while and the non-infectible lineage 1, 5 and 6 worms made up 95% of the population (R. B. Nehring unpublished data). Numerous laboratory exposures of all 4 lineages of *Tt* worms have repeatedly shown that 250 *Tt* worms can produce 2,000 to 10,000 progeny in 6 months. Given that sort of a reproductive advantage over the once-dominant lineage 3 *Tt* worms, at some point in time a shift could occur in the composition of the *Tt* worm population structure favoring the non-infectible lineage 1, 5 and 6 worms. This sort of shift in the *Tt* worm population structure has been observed in other aquatic ecosystems where the *Mc* parasite has been enzootic for a decade or longer, including the Gunnison River below the BCNP (Nehring, unpublished data). These worms have been proven (through CDOW research efforts) to function as biological controls of the fish-produced myxospores that infect the worms, thereby further reducing the rate of exposure of the now increasingly rare lineage 3 worms in WGR.

Evidence from research efforts across Colorado indicate that the WGR population is unlikely to shift back toward dominance by lineage 3 strain worms.. Nonetheless, the WD problem in the Colorado River downstream of WGR as well as in the Fraser River upstream of

WGR still exists. A bypass channel around WGR serves to reduce the exposure of the Colorado River downstream of WGR to WD in three significant ways. First, the worm-infective myxospores of the WD parasite that are produced in brown, brook and rainbow trout in the Fraser River upstream of WGR get mobilized from the sediments into the water column during spring run-off (May and June) and are deposited in WGR when the water velocity slows to almost zero in the lake. Those myxospores are ingested by aquatic worms, some of which are still lineage 3 *Tt* worms. Those worms will produce the fish-infective TAM spores beginning in July and would be released downstream into the river (Nehring et al. 2003). However, with WGR managed and operated as a no-spill impoundment, especially during the low flow, non-pumping periods (usually July through March), all TAMs spores produced in WGR stay impounded in the lake. Second, the water bypassed around the lake through a bypass channel is largely devoid of myxospores because they are not so easily mobilized during low flow periods and seasons. Third, since the aquatic worm population in WGR is comprised of more than 95% strains and species that are not susceptible to the *Mc* parasite, they truly function as biological filters by deactivating and destroying the myxospores in the lake, thereby significantly reducing the level of exposure to the trout populations below the lake, compared to a system operating without the bypass.

A bypass channel around WGR is important in numerous ways for restoration of functionality of the Colorado River between WGR and the confluence with the Blue River in Middle Park. Over time, with true flushing flows the biotic integrity of the river could be restored. However, true functionality and biotic integrity cannot be restored without re-establishment of the connectivity of the river.

RECOMMENDATIONS

Indicators of Biotic Integrity - In the event that the proposed Windy Gap Firming Project and the Moffat Tunnel Enhancement Project become a reality, one of the mitigation requirements will be a much more concerted effort on a number of fronts to try and return the upper Colorado River to some semblance of a more properly functioning ecosystem. One of the proposals to help insure that becomes a reality is a process called "***Learning by Doing***". A part of that process will entail regular monitoring the aquatic ecosystem and observation of physical, chemical and biological indicators for evidence of restoration and hopefully enhancement of the affected aquatic ecosystems. This will require (among other things) selecting species of aquatic insects and fish to function as biological indicators of ecosystem health.

Aquatic insects that have been adversely impacted by WGD and flow depletions, as shown in this report, can best serve in that capacity. EPT taxa that should be considered as indicator species are the following; among the stoneflies, *Pteronarcys californica, Pteronarcella badia* and an *Isogenoides* species. Among the mayflies such species as *Drunella grandis*, *Drunella doddsii,* and *Tricoythodes explicates* should be considered. Among the caddisflies such taxa as *Ochrotrichia, Protoptila, Arctopsyche, Cheumatopsyche* and *Psychomyia flavida* should be considered.

The mottled sculpin is the best species of fish to use as a biological indicator of ecosystem health for a multiplicity of reasons. First, it is a native species that once occurred throughout the upper Colorado River basin, and still does in most of the creeks and major tributary streams upstream of the mainstem impoundments on Willow Creek, as well as the Colorado, Fraser and Williams Fork rivers. Therefore, continued monitoring of this species in the unimpounded headwater reaches will quickly give an indication of microhabitat degradation in those areas. Second, sculpins are a benthic dwelling species that are much less mobile, quite sedentary. Their prime microhabitat is among the cobble-boulder strewn riffle areas that are prone to sedimentation and clogging when stream function has been compromised due to excessive flow depletions. They have been extirpated from the mainstem of the Colorado River from WGD downstream to the confluence with the Blue River. Restoration of a sculpin species in this reach of river would be the hallmark of success and *prima facie* evidence that “enhancement of the river” has truly happened. Third, because sculpins are largely a sedentary species, with a very limited home range, monitoring of their population levels and relative abundance can be much more easily accomplished with less time and manpower than is necessary to monitor trout populations. Fourth, the sculpin spawning period runs from late spring through mid-summer, spanning a period of 6-8 weeks that probably begins in mid-June and ends in early to mid-August. Male sculpins attract females into a nest beneath cobble-boulder substrates in riffle areas, guard and clean incubating eggs until they hatch. During this 6-8 week period, the nests are vulnerable to dewatering and destruction by desiccation. Failure of sculpin reproduction will be most likely the result of severe fluctuations and depletions of stream flow between spawning and hatching. In contrast, successful reproduction will once again be *prima facie* evidence that “enhancement of the river” has truly happened. Fifth**,** sculpins exist in sympatry with all salmonid species (cutthroat, brook, rainbow and brown trout, grayling and mountain whitefish) in streams all across western Colorado and the Intermountain West. Mottled and Paiute sculpins are native species that are easy to study. Sixth, their thermal tolerances are very similar to the stream-dwelling salmonids that they coexist with throughout the west. Seventh**,** they are a nongame species and are not exploited by anglers, making them a better target species for assessment of impacts due to water diversion and man-made manipulation of stream flows over short reaches of stream. For all of those reasons they are an excellent indicator species for assessing the functionality and biotic integrity of mountain stream ecosystems.

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Appendix for Job 2

Tables

Table A.1. Summer and fall 2010 electrofishing station descriptions, length, width, area, GPS coordinates and sampling dates.

Stream Name	Station Location & Description	Length (m)	Width (m)	Area (ha)	X & Y Coordinates (NAD 83)		Date
Arapaho Creek	Below Monarch Lake	97.6	11.5	0.1121	13T 436112	4440537	10/20/2010
Beaver Creek	Beaver Creek SWA	61	4	0.0224	13T403755	4433733	9/16/2010
Colorado River	Winding River Resort	91.5	11.4	0.1043	13T427656	4458874	9/15/2010
Colorado River	Double A Barns below Shadow Mtn Rsvr	91.5	20.4	0.1867	13T428710	4450559	9/9/2010
Colorado River	County Rd. 627 Rd Bridge below L. Granby	106.7	27.5	0.2934	13T424169	4442110	10/19/2010
Colorado River	Jack Horn Ranch	182.9	15.8	0.289	13T417391	4439633	10/20/2010
Colorado River	Hitchin Post	304.9	27.9	0.8507	13T414734	4440532	9/14/2010
Colorado River	Red Barns	335.4	29.2	0.9794	13T412699	4439618	9/9/2010
Colorado River	Lone Buck SWA	243.9	29.9	0.7293	13T402733	4433676	9/9/2010
Colorado River	BLM above the Ritschards Ranch	91.5	34.1	0.312	13T394848	4435729	10/19/2010
Colorado River	BLM Pumphouse	61	6.4	0.039	13S370978	4427501	9/22/2010
Fraser River	Behind Safeway	190.5	6.4	0.1219	13S430424	4422619	9/7/2010
Fraser River	At Elkdale in Fraser River Canyon	192.1	18.5	0.3554	13T424704	4432658	9/11/2009
Fraser River	Solvista	122	21.9	0.2672	13T424497	4435696	9/2/2010
Fraser River	Kaibab Park	198.2	10.2	0.2022	13T420741	4437199	9/2/2010
Little Muddy Creek	200 m North Grand County 361 Bridge	34.8	3.67	0.0128	13T402933	4432456	9/17/2010
Troublesome Creek	1.5 mi above E. Fork confluence	88.4	3.14	0.0278	13T389344	4448510	10/22/2010
Troublesome Creek	Below Hwy 40 Bridge Colo R. confluence	122	9.1	0.111	13T388710	4434818	9/21/2010

Table A.1 (Continued) Summer and fall 2010 electrofishing station descriptions, length, width, area, GPS coordinates and sampling dates.

Stream Name	Station Location & Description	Length (m)	Width (m)	Area (ha)	X & Y Coordinates (NAD 83)		Date
Williams Fork River	Above Bridge Crossing Reservoir	104	8.7	0.0905	13S399324	4428292	9/8/2010
Williams Fork River	Ute Creek confluence	182.9	13.1	0.2396	13S406704	4415663	9/21/2010
Williams Fork River	0.1 km below Williams Fork Reservoir	182.9	9.5	0.1729	13T397183	4432363	9/12/2001
Williams Fork River	Below DOW Irrigation Diversion headgate	266.8	12.8	0.3415	13T398369	4434068	9/12/2001
Willow Creek	1 mi above Cabin Creek confluence	122	7.7	0.0939	13T410299	4452711	9/7/2010
Willow Creek	CLazyU Ranch	91.5	7.3	0.0668	13T415666	4446516	9/10/2010
Willow Creek	Upper site below Willow Creek Reservoir	122	10.7	0.1305	13T420008	4444396	9/15/2010
Willow Creek	2 miles below Willow Creek Reservoir	106.7	7.5	0.08	13T421231	4443103	9/16/2010
Gunnison River	Above North Fork	122	4.9	0.0598	13S253612	4296301	11/9/2010
Yampa River	Above Sarvis Creek	152.4	13.2	0.2011	13T346451	4462224	11/4/2010
Dolores River	Lower Metaska Day Use Area	61	19.4	0.1183	12S713113	4161698	11/11/2010

Table A.2. Fish population density and biomass for 2010 in the Colorado River basin and at three reference sampling sites, outside the basin for comparative purposes.

Stream Name	General Sample Location Description	Brown Trout		Rainbow		Brook Trout		Mottled Sculpin		Smallest
		N/ha	Kg/ha	N/ha	Kg/ha	N/ha	Kg/ha	N/ha	Kg/ha	(mm TL)
Arapaho Creek	Below Monarch Lake	1,386	236	2,754	72	288	4	3,455	24	35
Beaver Creek	Beaver Creek SWA	204	1	0	0	9,381	84	3,666	16	33
Colorado River	Winding River Resort	1,630	47					2,912	9	32
Colorado River	Double A Barns below Shadow Mtn Rsvr	98	19	7,918				2,524	36	22
Colorado River	Grand County 627 Rd Bridge below L. Granby	1,678	89	29	11			167	3	25
Colorado River	Jack Horn Ranch	971	87	3	1			24	1	52
Colorado River	Hitchin Post	1,031	170	176	30			1	0	134
Colorado River	Red Barns	773	139	97	21			0	0	
Colorado River	Lone Buck SWA	532	111	94	10			0	0	
Colorado River	BLM above the Ritschards Ranch	789	31	6	0			0	0	
Colorado River	BLM Pumphouse	1,335	75					7,194	38	39
Fraser River	Behind Safeway	206	54	41	8	712	52	4,407	45	46
Fraser River	at Elkdale in Fraser River Canyon	654	116	265	18			1,651	17	57(a)
Fraser River	Solvista	286	56	11	0	8	0	1,797	17	27
Fraser River	Kaibab Park	589	73	79	11			2,838	20	25
Little Muddy Creek	200 m North Grand County 361 Bridge	236	6					0	0	
Troublesome Creek	1.5 mi above E. Fork confluence	3,333	109			1,090	21	4,477	25	28
Troublesome Creek	Below Hwy 40 Bridge at Colo R. confluence	502	63					0	0	

Table A.2 (continued). Fish population density and biomass for 2010 in the Colorado River basin and at three reference sampling sites, outside the basin for comparative purposes.

Stream Name	General Sample Location Description	Brown Trout		Rainbow		Brook Trout		Mottled Sculpin		Smallest
		N/ha	Kg/ha	N/ha	Kg/ha	N/ha	Kg/ha	N/ha	Kg/ha	(mm TL)
Williams Fork River	Above Bridge Crossing Reservoir	924	48	171	34	213	2	5,404	20	31
Williams Fork River	Ute Creek confluence	79	7	79	2	258	12	1,964	15	45
Williams Fork River	0.1 km below Williams Fork Reservoir	638	179	215	63			0	0	
Williams Fork River	Below DOW Irrigation Diversion headgate	1,197	191	164	57			0	0	
Williams Fork River	At raft launchgate station	4,258	274	65	25			0	0	
Willow Creek	1 mi above Cabin Creek confluence	160	26			32	0.5	6,370	19	17
Willow Creek	CLazyU Ranch	135	70	75	56			21,252	70	23
Willow Creek	Uppersite below Willow Creek Reservoir	978	16	8	0			15,003	122	40
Willow Creek	2 miles below Willow Creek Reservoir	1,730	187	12	1			0	0	
Gunnison River	Above North Fork	2,070	26					4,348	70	64
Yampa River	Above Sarvis Creek	24	9	170	66	19	1	968	13	47
Dolores River	Lower Metaska Day Use Area	86	13	9	5			6,453	69	49